Optimising the Postharvest Management Of Lychee (*Litchi chinensis* Sonn.) – A Study of Mechanical Injury and Desiccation

by

Philippa Bryant

(BSc. Ag. Hons.)

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Department of Crop Sciences

Faculty of Agriculture

The University of Sydney.

Declaration

I certify that this thesis does not incorporate without acknowledgement any material previously submitted for a degree or diploma in any university, and that to the best of my knowledge and belief it does not contain any material previously published or written by another person except where due reference is made in the text.

Philippa Bryant

January, 2004.

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Summary

The major objective of the research was to improve lychee postharvest management, through a greater understanding of mechanical injury and moisture loss.

Mechanical injury is a known cause of postharvest loss in lychee, but previously published information has been limited to broad observations. In this study, the symptoms of mechanical damage in lychee were defined, including quantitative measurement of colour changes. Impact injury caused protuberance tip darkening, cracking of the pericarp and significant changes in skin colour. Compression also typically caused tip darkening, and severe loads were capable of puncture, shape distortion and skin cracking. Abrasion and vibration injuries were characterised by strong yellowing of pericarp colour, possibly due to the leakage of cell contents onto the fruit surface. Vibration also caused significant darkening and loss of colour saturation. Vibration has not previously been mentioned as an issue in lychee postharvest management, but appeared to be as important a problem as desiccation browning at the wholesale level, both in incidence and severity.

Mechanically damaged fruit consistently showed increased ethylene and carbon dioxide synthesis, and moisture loss was increased by up to 30%. Some significant changes in skin biochemistry and cuticle properties were also detected. The study of damaged tissue by SEM revealed distinctive patterns of surface tissue disruption. Open pericarp cracking was a particularly detrimental injury, causing significantly increased electrolyte leakage and rapid pathogen development.

The effects of load characteristics, such as magnitude, method of application, site, repetition and cushioning, on the extent of damage were defined. Fruit characteristics such as cultivar, gross morphology, temperature, hydration and surface wetness were shown to significantly affect damage levels. Small seed size was correlated with increased cracking susceptibility. Fruit surface wetness exacerbated vibration or abrasion damage. Turgid fruit were less susceptible to vibration and abrasion damage, but showed increased susceptibility to impact cracking.

Previously neglected aspects of desiccation browning research were studied, including cultivar and maturity effects, sites of moisture loss and the role of air currents. Cultivar effects on moisture loss were obscured by pre-harvest factors, but consistent cultivar differences were detected in desiccation browning, possibly related to skin thickness. In contrast, maturity levels over a marketable range had little effect on weight loss or browning. Moisture was lost fairly evenly over the fruit surface, but poor postharvest handling appeared to massively increase loss from the protuberance tips. Moisture loss was shown to substantially increase ethylene synthesis. The crucial role of air currents in exacerbating lychee moisture loss was emphasised, and the relationship between air speed and weight loss was defined.

The research contributed to a greater understanding of the processes of mechanical damage and moisture loss in lychee, leading to improved protocols for the postharvest management of the fruit. Improved management of mechanical damage and moisture loss will ultimately improve fruit quality and reduce postharvest losses, hence increasing returns to industry.

SUMMARISED TABLE OF CONTENTS

FULL TABLE OF CONTENTS

List of Abbreviations

a	acceleration
A	total anthocyanin
AC	air current factor
ADI	anthocyanin degradation index
ANOVA	analysis of variance
Avg	average
C	crack area
C_A	sample concentration
C_B	standard concentration
CA	controlled atmosphere
CO^2	carbon dioxide
D	displacement
D-400	dehydrated 400 g load treatment
D-800	dehydrated 800 g load treatment
DF	tissue displacement using a flat impacting head
D_F	final deformation
D_I	initial displacement
DM	dry matter
dP	vapour pressure deficit (Pa)
DR	tissue displacement using a round impacting head
D_R	deformation when weight was removed (residual)
E	energy of impact (J)
E1	experiment 1
E2	experiment 2
E3	experiment 3
E4	experiment 4
E_D	degree of elasticity,
E_M	modulus of elasticity,
e^o	saturation vapour pressure (Pa),
F	force (kg mm^{-2})

FT	core fruit temperature
G	generation of CO_2 (ml kg^{-1} hr^{-1})
GC	gas chromatograph
h	height
H-400	hydrated 400 g load treatment
H-800	hydrated 800 g load treatment
HCl	hydrochloric acid
HDPE	high density poly-ethylene
J	joule
KCl	potassium chloride
KMP	Kwai May Pink
KMR	Kwai May Red
L	length
M	mass (kg)
M1, M2…	maturity grade 1, 2…
MAP	modified atmosphere packaging
MDF	medium density fiberboard
MgCl	magnesium chloride
ML	moisture loss
NaOH	sodium hydroxide
N	sample size
O^2	oxygen
P_A	sample peak area
P_B	standard peak area
PAL	phenylanaline ammonialyase
PE	polyethylene
PG	polygalacturonase
PME	pectin methylesterase
PML	previous moisture loss factor
POD	peroxidase
PPO	polyphenol oxidase

PVC	polyvinyl chloride
r	radius
r_1	radius of a semi-spherical cap
RH	relative humidity
RL	relative level of moisture loss
RO water	reverse osmosis water
Rt	rot area
RT	room temperature
S	wind speed
SA	surface area
SA^F	contact surface area using a flat impacting head
SA^R	contact surface area using a round impacting head
SE	South East or standard error
SEM	scanning electron microscopy
SML	specific moisture loss ($kgkg^{-1}Pa^{-1}s^{-1}$)
t	time
T	temperature
TSS	total soluble solids
Ve	velocity
V	volume
VI	vibration damage index
VP_A	air vapour pressure (Pa)
VP_F	fruit vapour pressure (Pa)
VPD	vapour pressure deficit
W	weight loss
ε	impact energy per unit area

List of Figures

List of Tables

CHAPTER 1. LITERATURE REVIEW

1.1 Introduction to Lychee

1.1.1 History

Lychee (*Litchi chinensis* Sonn.) belongs to the *Sapindaceae* family, and is native to subtropical areas of southern China. Lychee trees have been cultivated for over 3500 years and the fruit have an important place in Chinese culture. Fresh lychee fruit inspired many ancient poems, and were given as gifts of great honour and as imperial offerings (Groff 1921). Some of the earliest Chinese literature refers to the perishability of the fruit and attempts to prolong postharvest life.

The fruit was introduced into Australia in the mid-1800's (Cobin 1954; Higgins 1917) by Chinese miners, but deliberate importation of improved cultivars did not occur until the 1920-30's (Greer 1990). Commercial plantings of lychee commenced in the 1970's, and the crop remains a strong minor industry, estimated at around 350 growers and 1800 hectares (Menzel *et al.* 1999).

Worldwide lychee production is centred largely in the Northern hemisphere, with growing areas including China, Thailand, Taiwan, India, the USA and Israel. Off-season Southern hemisphere production occurs in South Africa, Australia, Mauritius and Madagascar. Australian production, estimated at 3000 tonnes and $A 12-15 million value in 1996 (Menzel *et al.* 1999), is minor in the scale of worldwide production, estimated at 350,000 tonnes (Underhill *et al.* 1997). Export from Australia is primarily to South East Asia, the Pacific and Europe, and comprises around 25% of the total production (Menzel *et al.* 1999).

1.1.2 Fruit Characteristics

The lychee fruit consists of a rough reddish pericarp of 1-3 mm width (Underhill and Simons 1993) and separate translucent aril surrounding a smooth brown seed. The fragrant aril has a juicy texture, similar to a grape, with a unique sweet, but crisp flavour. The pericarp surface is covered in numerous protuberances (alternatively termed uricles (Kumcha 1998) or tubercles (Singh and Singh 1954)), resulting in a rough, spiky texture. In some cultivars, such as ‘Wai Chee’, the protuberances flatten at maturity, resulting in smoother fruit (Batten 1986). Skin colour varies somewhat between cultivars, from the light orange-pink of ‘Kwai May Pink’ (‘KMP’) to the deep dull red of ‘Tai So’ and the purple-red of ‘Souey Tung’. The ‘perfect’ lychee may vary widely in texture, size and shape due to cultivar differences, but there is one characteristic which remains constantly desirable: fresh red skin.

1.1.3 Postharvest Issues

Skin colour is one of the major characteristics used to judge the commercial quality of lychees. The retention of fresh, red fruit colour throughout the postharvest chain has been a major focus of postharvest research on lychee. Skin browning due to moisture loss is a major limitation to the retention of colour, with lychees deteriorating rapidly after harvest, often within 2 to 3 days (Huang and Scott 1985). Although desiccation browning of the skin may not affect the eating quality of the fruit, it greatly reduces the commercial value in western markets and is considered one of the major causes of postharvest loss (Snowden 1990).

Another potential cause of postharvest losses in lychee is the mechanical injury of the fruit. It has been suggested that mechanical damage may degrade the appearance of lychee fruits (Bagshaw *et al.* 1995), but this has not been the focus of any published research. In other crops mechanical damage has been shown to result in substantial postharvest losses, but the extent of the problem in the lychee industry is yet to be assessed.

1.2 Importance of Mechanical Injury and Moisture Loss

1.2.1 Economic Losses

Both mechanical injury and desiccation have long been recognised as important causes of postharvest loss in fruit and vegetable production. Mechanical damage can cause a decline in visual appeal due to symptoms such as cracking, scuffing and bruising, while moisture loss can cause wilting and browning. Desiccation has a direct effect on the value of fresh produce, through a reduction in saleable weight, while mechanical injuries may be severe enough to render the fruit unmarketable, reducing yield and profits. Less severe damage may reduce the value of the product by requiring trimming, causing downgrading, or reducing the overall visual appeal of a display.

In addition to a decline in visual appeal, mechanical injury may also result in increased moisture loss and respiration and a higher risk of pathogen invasion (Hung 1993). Increases in moisture loss and respiration triggered by mechanical injury can potentially reduce the weight of the product, and hence the market value. These processes may cause a decline in sensory quality and a reduced shelf life. Damaged fruit can also encourage the establishment of pathogens, which may spread to sound fruit, potentially causing severe losses (Wills *et al.* 1998). With these indirect types of postharvest losses it is very difficult to estimate the relative importance of mechanical damage, as many other factors influence the processes of moisture loss, respiration and pathogen growth.

The effects of mechanical injury are often latent, with symptoms appearing hours or days after stress is applied. Similarly, the loss of moisture on the farm may cause minimal change in appearance, but when added to further small losses during the marketing chain, the fruit may be rendered unmarketable. This can result in potentially unsaleable fruit unintentionally passing through the sorting process into the marketing chain. The marketing of mechanically injured or water stressed fruit may reduce the profile of the product, and negatively affect the reputation of the grower or wholesaler, risking the loss of future sales. In addition, the damaged fruit costs money to send through the postharvest chain, in cooling, packaging and transporting, but may be worthless when it reaches the buyer. The latent nature of damage makes the estimation of economic losses, both to the industry, and the individual grower, difficult to assess.

The levels and causes of postharvest loss in fruit and vegetables vary widely between produce and regions, and are notoriously difficult to estimate. In commodities sensitive to mechanical injury, such as apples, damage can occur to more than 90% of the crop during commercial handling (Timm *et al.* 1989). Average levels of postharvest loss due to mechanical injury vary widely between crops, but are often in the range of 5-30% (Ruiz Altisent 1991). Losses due to mechanical damage in the lychee industry are yet to be estimated.

Losses due to moisture stress are similarly difficult to estimate. Lychee postharvest losses in Australia due to desiccation browning have been estimated to be as high as 60% of production (Greer, personal communication. In: Underhill 1993). It is likely that the majority of losses would occur late in the postharvest chain as a result of cumulative moisture loss.

1.2.2 Fruit Quality Issues

The wide spread adoption of quality assurance procedures is indicative of the current trend in the marketplace towards very high expectations of quality. In this market environment, even slight cosmetic damage may be of significant economic importance. The grading of fruit in the lychee industry is affected by cosmetic defects such as small blemishes and skin browning. Blemishes as small as 0.25 cm^2 affect the grade, and hence the value of lychee fruit (FAO/WHO 1994). With export markets paying a premium for quality, the production of highest standard fruit can be a lucrative niche. Financial studies based on a typical lychee orchard show that a 10% increase in price improves profitability more than a 50% decrease in costs (Thew 1986), suggesting that high fruit quality is a commercially viable goal.

In the past, the development of new mechanically resistant varieties through crop breeding was attempted in some crops, such as tomatoes. The success of breeding for mechanical strength has since been overshadowed by an accompanying decline in other valuable traits, such as texture and flavour. With an increased desire for “old-fashioned” flavour, the selection of crops with good sensory qualities and the harvesting of fruit as ripe as possible have become important considerations. Premiums are paid for high quality produce, such as vine-ripened tomatoes. However, increased harvest ripeness is often associated with a higher risk of mechanical damage due to textural changes in the tissue. With the trend towards improved sensory qualities, mechanical damage continues to be a relevant area of research in the horticultural industry.

1.2.3 Mechanisation

The wide scale mechanisation of postharvest processes in recent times has resulted in increased awareness of the issue of mechanical damage. High labour costs and the economies of scale suggest that future trends in the horticultural industry will be towards greater volumes of production and further mechanisation of operations. In particular, the high labour costs of fruit harvesting make the mechanisation of harvest an economically valuable goal. The assessment of the mechanical thresholds of a crop such as lychee may provide valuable information for the future mechanisation of harvesting.

1.3 Moisture Loss

Fruit moisture loss typically reduces visual appeal, marketable value and sensory qualities. It occurs due to a water potential gradient that draws water vapour into the atmosphere. The fruit skin provides a barrier to moisture loss, which varies substantially between different fruit and vegetables. Moisture loss is encouraged by poor skin resistance to water vapour movement, air currents, warm temperature, low RH and temperature gradients between air and fruit. Water loss is a constant risk throughout the postharvest chain, with substantial losses often occurring during harvest, air-cooling and marketing. In practice, moisture loss is generally minimised by control of RH and temperature, particularly by the combined use of packaging and refrigeration.

1.3.1 Fruit Responses to Moisture Loss

1.3.1.1 Symptoms of Moisture Loss

The loss of moisture is one of the major causes of postharvest loss in the horticultural industry. Dehydration of fresh produce typically causes a reduction in weight, and a decline in appearance and textural quality. The symptoms of moisture loss vary with product characteristics, and the extent of loss. Changes in appearance due to moisture loss commonly include browning or yellowing, reduced glossiness, dry appearance and decreased intensity of colour. Textural changes may include softening, wilting, tissue shriveling and loss of crispness. In general, these symptoms appear at 4-8% weight loss (Van den Berg 1987). Some damage can be reversed by placing the product in higher RH, but prolonged water loss leads to irreversible damage (Shewfelt 1993). In lychee, desiccation browning is the most noticeable symptom of moisture loss, with the fresh red skin deteriorating to dry pale brown.

1.3.1.2 Physiological Effects of Moisture Stress

The physiological processes stimulated by moisture stress tend to result in a loss of quality and shortening of postharvest life. Moisture loss stimulates ethylene synthesis (Aharoni *et al.* 1975; Littmann 1972), which itself strongly influences fruit physiology (Kader 1985). Water stress has been shown to substantially hasten ripening in climacteric fruits, through increased ethylene synthesis, as shown in avocado, banana and pear (Littmann 1972). Ethylene also stimulates senescent processes, and accelerates deterioration, causing increased respiration, loss of cell compartmentalisation and the release of enzymes such as polygalacturonase (PG), peroxidase (POD), lipoxidase, alpha-amylase, polyphenol oxidase (PPO) and phenylalanine ammonialyase (PAL) (Kader 1985). Enzymes can also be triggered by moisture stress independently of ethylene, as shown in the stimulation of PG in cucumber by postharvest water stress. The enzyme appears to be generated both in direct response to water stress, and as a result of increased ethylene synthesis resulting from water stress (Kubo *et al.* 2000).

Moisture loss causes changes similar to senescent breakdown, including the loss of membrane integrity, leakage of cell contents, more rapid degradation of surface tissues and yellowing of colour (Ben-Yehoshua 1987; Van den Berg 1987). Moisture loss in lychee has been shown to result in increased conductivity of pericarp tissues, symptomatic of loss of membrane integrity (Chen and Hong 1992). Conductivity ranged from 50-100 μS in hydrated fruit, and increased to 100-150 μS at 10% loss of pericarp weight, and 150-250 μS at 20% loss of skin weight.

Typical side-effects of the aging process are also observed in water stressed fruit. Moisture loss tends to reduce photosynthesis in green fruit and vegetables, and reduces the ability to heal wounds (Van den Berg 1987). A combination of processes, including loss of membrane integrity and changes in cuticle structure, result in reduced ability to resist pathogen invasion. Dehydration also reduces nutritional content, with vitamins particularly affected (Van den Berg 1987). The many changes in fruit and vegetable physiology caused by moisture stress combine to result in overall loss of quality and reduced shelf life.

1.3.1.3 Lychee Desiccation Browning

It is commonly stated that browning of the skin in lychee fruit is caused by moisture loss. Browning of the pericarp is correlated with moisture loss, with both Hunter *a* values (redness) and visual colour analysis strongly related to the loss of moisture from the pericarp (Underhill and Critchley 1994). There is some conflict as to the amount of moisture loss required to cause desiccation browning. It has been suggested that browning can become apparent when as little as 2% of the pericarp moisture is lost after harvest (Underhill and Critchley 1994). Brown (1986) suggested that 3-5% weight loss from the whole lychee fruit resulted in browning, while Liang *et al.* (1998) showed that browning commenced at 7.6% weight loss, with full browning not occurring until 18% of fruit mass was lost. Similarly, Wu *et al.* (1997) (as cited in Shi *et al*. 2001) suggested that 9% weight loss was required to trigger pericarp browning. The wide range of values suggested may be due to various factors, including different methods of assessment, growing conditions, cultivar, water content at harvest, and postharvest treatment prior to measurement.

While postharvest browning of the lychee pericarp is often caused by moisture loss from the fruit, other stresses, such as pathogen and insect attack, chilling, heat stress and senescence can also lead to browning (Bagshaw *et al.* 1995; Fitzell and Coates 1995). Browning by temperature stress, rot and senescence is typically dark and water-soaked in appearance. In general, desiccation browning is distinguished by a paler, dry appearance. Desiccation browning is often initially restricted to one side of the fruit, and progressively spreads to cover the entire surface of the pericarp, eventually leaving the pericarp dry and brittle (Underhill 1993).

1.3.1.3.1 Microscopy of Lychee Desiccation Browning

The lychee pericarp consists of a thick continuous epicarp, parenchymatous mesocarp, and a thin membranous endocarp (Underhill and Critchley 1992). The mesocarp consists of parenchyma tissue containing many vacuoles, and hence the photosynthetic organelles and most of the red pigmentation of the fruit. The epicarp consists of cuticle, epidermis and a thick, heavily lignified sclerenchyma layer, which is punctuated by groups of collenchyma cells.

Microscopic examination over the course of fruit desiccation shows that browning is highly localised and initially affects only the upper epidermis (Underhill and Critchley 1995). Browning extends into the parenchymatous tissue through collenchyma intrusions of the sclerenchyma layer, and eventually encompasses the entire epicarp and part of the upper mesocarp. Browning injury is often more severe in the lenticels, stomata and collenchyma (Underhill and Critchley 1995). The fact that browning is associated with cells involved in gas exchange and moisture control provides additional evidence that moisture loss and browning are related.

Moisture stress in plant tissue typically causes shrinkage of the cell contents, eventually leading to cell plasmolysis. In rambutan, a spiky red fruit related to lychee, desiccation browning was preceded by plasmolysis of cells. Browned tissue showed severe shrinkage of cells, and some cells containing black contents, which were shown to be dead (Landrigan *et al.* 1994). Shrivelling of epidermal cells, and cracks in the wax layer in rambutan due to water stress were observed by SEM (Landrigan *et al.* 1996b). Transmission electron microscopy of lychee pericarp showed that moisture loss from the tissue resulted in cytoplasmic dehydration, contraction and coagulation, and disintegration of cell structure (Chen and Hong 1992). Moisture stress in lychee is also thought to exacerbate the formation of microcracking in the pericarp (Underhill and Simons 1993).

1.3.1.3.2 Biochemistry of Lychee Desiccation Browning

Physical changes in cell structure resulting from moisture stress, such as cell plasmolysis and disruption of cytoplasmic contents, are thought to cause the tissue browning typical of dehydration. Enzymes and substrates can be decompartmentalised by these processes, triggering enzymatic browning reactions.

A substantial volume of research has been carried out to elucidate the biochemical processes underlying lychee desiccation browning. It was originally hypothesised that anthocyanins, the pigments conferring the characteristic red colour of lychee, were broken down by the enzyme PPO to result in lychee browning (Akamine 1960). Subsequent research has suggested that additional processes are involved. Some research suggests that other enzymes may be involved in the browning process (Underhill and Critchley 1995; Jiang and Fu 1999; Zhang *et al.* 2001). Other studies have suggested that the pericarp pH is important in browning, as reduced pH results in improved red colouration (Underhill and Critchley 1994). Underhill and Critchley (1993) suggest that anthocyanin is converted to a colourless form under high pH conditions, and that this allows independent browning in the epicarp to become more visible. It appears that PPO mediated breakdown of anthocyanins is one mechanism of browning (Jiang 2000), but that other processes may also contribute.

1.3.1.3.2.1 Anthocyanins

The red colouration so desired in lychees is primarily conferred by the presence of anthocyanins (Lee and Wicker 1991a). Anthocyanins are a common group of plant pigments responsible for most red, purple and blue colouration in plants. These pigments are water-soluble and are situated within vacuoles in the mesocarp and epicarp of the lychee skin (Underhill and Critchley 1994). At maturity the concentration of anthocyanins is most dense in the upper mesocarp. Anthocyanins can be degraded by enzymes such as PPO and POD (Markakis 1982), a process that may contribute, at least to some extent, to lychee pericarp browning.

It has been widely assumed that PPO-mediated degradation of anthocyanin, and the associated formation of oxidised by-products is the major cause of pericarp browning (Akamine 1960; Lin *et al.* 1988; Tongdee 1998). The observation that PPO activity increases after harvest and anthocyanin levels decrease (Lin *et al.* 1988) has been used to support this hypothesis. Underhill and Critchley (1993) showed that anthocyanin degradation in lychees was correlated with pericarp moisture loss and the development of browning. Complete browning was observed in the fruit after 48 hours (at 25^{o}C and 60% RH), when around 27% of the total anthocyanins in the pericarp had been degraded (Underhill and Critchley 1993). Under slightly more humid conditions (27-30^{o}C and 73-79% RH) Lin *et al.* (1988) recorded a much slower decline in anthocyanin concentration, with a 10% reduction observed over seven days storage.

However, methods of extraction may have created false high readings in some previous research on lychee anthocyanins. Song *et al.* (1997) found that lychee anthocyanins were soluble in water and alcohol, but once tissue had browned, pigments were soluble only in organic solvents. Brown pigments are therefore soluble in the acidic methanol extract commonly used for anthocyanin measurement. The use of 0.1M HCl, in which the browned pigments were insoluble, revealed much stronger loss of anthocyanin content as browning progressed. More than half of total anthocyanin was lost as fruit browned (Zhang *et al.* 2001). Furthermore, the use of fresh weight measurements in the sampling of pericarp tissue would be likely to obscure the strength of this trend. The massive loss of moisture from the pericarp during desiccation browning would tend to concentrate the remaining anthocyanins when measured on a fresh weight basis. It is therefore likely that the actual loss of anthocyanins is much higher than previously suggested.

It appears that enzyme mediated anthocyanin degradation is not the only mechanism of browning. Red pericarp colour can be temporarily restored in desiccated brown fruit by the application of acid dips (Underhill and Critchley 1994). This effect is due to the direct influence of pH on anthocyanin colour. The ratio between the flavylium cation and colourless carbinol forms of the anthocyanin molecule varies with pH, resulting in red colour at low pH, with gradual loss of colour as pH increases (Brouillard 1982). Solutions of pH 4 or less are required to allow the expression of anthocyanin colour. An increase in pH in the pericarp occurs during postharvest desiccation, which may be representative of pH changes within the vacuoles. Underhill and Critchley (1994) observed an increase in pericarp pH from 4.15 to 4.52 over 48 hours under ambient conditions (25^{o}C and 60% RH), which would be sufficient to cause some anthocyanin decolourisation. In addition to the direct effect of pH on anthocyanin structure, increasing pH has a secondary effect of reducing the stability of the pigment, hence increasing the rate of anthocyanin degradation (Pang *et al.* 2001). It was theorised that the loss of anthocyanin colour due to pH change allowed greater expression of independent browning (Underhill and Critchley 1993). The return of redness in acid dipped fruit is thought to result from anthocyanins being changed into a coloured form and masking the epicarp browning.

1.3.1.3.2.2 Polyphenol Oxidase (PPO)

For many years PPO has been associated with the degradation of anthocyanin and other phenolic compounds in a diverse range of fruit species (Mathew and Parpia 1971). It has been widely assumed that PPO breaks down anthocyanins in lychees and that this is the mechanism of pericarp browning (Akamine 1960). It is also possible that PPO causes browning through the oxidation of other phenolic compounds. The level of total phenolics in lychee pericarp is correlated with the extent of browning, suggesting that the polymerization of phenols contributes largely to browning (Jiang and Fu 1999). PPO causes hydroxylation of monohydroxyphenols to *o*-dihydroxyphenols and dehyrogenation of *o*-dihydroxyphenols to *o*-quinones (Mayer and Harel 1979). The *o*-quinones generated by this process are polymerised by a non-enzymatic reaction, and it is this process which results in the formation of dark coloured by-products and hence browning (Mayer and Harel 1979).

Moisture loss and high pH increase the activity of PPO, and these conditions have also been shown to encourage pericarp browning. Lychee PPO shows a peak in activity at pH 7, and is most stable at pH 7.4 (Jiang *et al.* 1997b). Activity is reduced at lower pH levels, and no activity was observed below pH 4.2 (Jiang *et al.* 1997b). The acidic pH of the lychee pericarp would tend to inhibit PPO activity. However, the increase in pH from 4.15 to 4.52 during desiccation (Underhill and Critchley 1994) would be likely to stimulate PPO activity. Many factors are known to initiate PPO activity (Mayer and Harel 1979), but moisture loss is considered the most important in lychee (Akamine 1960). Jiang and Fu (1999) showed that PPO levels increase under conditions of lower RH.

There is some conflict in the literature regarding the activity of PPO after harvest. Lin *et al.* (1988) showed a rapid increase in PPO activity after harvest, with a peak at 48 hours (at approximately 28^{o}C and 76% RH). In contrast, Zauberman *et al*. (1991) found no significant change in PPO in the first 48 hours (at 22^{o}C and 85% RH). Jiang and Fu (1999) found that storage RH affected the changes in PPO after harvest. Over three days, PPO levels significantly declined in fruit stored at lower humidity (60-80%). At high humidity levels (90-95%) the total amount of PPO was lower, and did not significantly change over the three day storage period. In addition, Underhill and Critchley (1993) showed that under dry ambient conditions (25^{o}C and 60% RH) PPO activity decreased significantly once fruit were harvested, with the rate of oxygen consumption more than halved in the first 24 hours. Conflict in measurements of PPO activity after harvest may relate to differences in initial water content, growing conditions or cultivar. PPO activity in ‘Nuomici’, a cultivar susceptible to browning, was higher than that in ‘Guiwei’, a more resistant cultivar (Chen *et al.* 2001). The activity of PPO in ‘Nuomici’ decreased during storage, while in ‘Guiwei’ PPO activity changed little during browning, suggesting that the role of PPO may vary substantially between cultivars.

The localisation of PPO activity within the pericarp provides further evidence towards a role in desiccation browning. Underhill and Critchley (1995) used tissue blots to show that PPO activity was concentrated in the epicarp and upper mesocarp. Minor activity was also shown to occur in the vascular tissue and the endocarp. Strong activity appeared on the apices of protuberances, the area which first shows browning (Underhill and Critchley 1995). The concentration of PPO activity in areas affected by desiccation browning supports the hypothesis of PPO-mediated degradation.

Jiang (2000) showed that PPO was capable of rapidly degrading anthocyanin in solution when a lychee phenolic extract was added. Without the phenolic extract, PPO activity was minimal. While conditions in solution are obviously very different to within the fruit, these results add some further evidence to the occurrence of PPO-mediated anthocyanin degradation, despite the low affinity of lychee PPO for anthocyanins.

1.3.1.3.2.3 Peroxidase (POD)

POD is an enzyme shown to cause browning in peaches (Flurkey and Jen 1978), mangoes (Zauberman *et al*. 1988) and walnuts (Piffaut and Metche 1991). The enzyme is involved in tissue breakdown, and is measured as an index of senescence. In the presence of hydrogen peroxide, POD acts on *o*-dihyroxyphenols, causing similar browning to PPO (Kahn 1985) and making it difficult to differentiate between the action of these two enzymes. The role of POD in desiccation browning of lychees is not clear, with some sources concluding it is not important (Zauberman *et al*. 1991), while others suggest that it may be significant (Underhill and Critchley 1995; Jiang and Fu 1999). POD activity increases early in storage, in the period of rapid pericarp browning (Lin *et al*. 1988; Jiang and Fu 1999), but declines after extended storage (Huang *et al*. 1990). Low RH conditions, which encourage browning, also result in higher levels of POD (Jiang and Fu 1999). While total levels of POD are lower under high humidity, the rate of increase appears to be similar regardless of humidity (Jiang and Fu 1999). It is possible that the different conclusions drawn regarding the role of POD may relate to cultivar differences. In the cultivar ‘KMP’, levels of POD in the epicarp were shown to be much higher than in ‘Tai So’, suggesting that substantial cultivar differences may exist in the role of this enzyme (Underhill and Critchley 1995).

1.3.1.3.2.4 Anthocyanase

An enzyme previously detected only in fungi, anthocyanase has been recently detected in lychee skin (Zhang *et al.* 2001). The enzyme shows a strong capacity to degrade anthocyanin, which is utilised commercially to reduce the redness of products such as juice and wine (Huang 1955). The degradation is thought to result in the formation of anthocyanidin, which is inherently unstable. Zhang *et al.* (2001) suggested that the anthocyanase-mediated conversion of anthocyanin to anthcyanidin may contribute to the rapid degradation of the pigment in lychee. The instability of anthcyanidin would encourage further degradation of the molecule.

1.3.1.3.2.5 The Role of Other Chemicals

The role of ethylene in lychee browning is uncertain. Jiang *et al.* (1986) observed that ethylene enhanced PPO and POD activity, and could thereby promote browning. The production of ethylene in lychee prior to harvest is negligible (Akamine and Goo 1973), and postharvest production varies. Tongdee *et al.* (1982) measured low rates of postharvest ethyene production, while Underhill and Critchley (1993) detected high levels immediately after harvest, which quickly declined. This increase in ethylene immediately after harvest may have been due to a wounding response, due to harvest or other injury.

It has been suggested that ascorbic acid may prevent pericarp browning through an antioxidant effect. In solution, it was shown that the degradation of anthocyanin by PPO occurs only after all ascorbic acid is consumed (Jiang 2000). The initial levels of ascorbic acid in the pericarp would therefore be expected to influence the development of browning.

1.3.1.3.2.6 An Overview of Browning Biochemistry

It seems likely that desiccation browning occurs due to simultaneous processes of increasing pH and enzyme activation. The slight shift in pH observed during desiccation reduces the stability of the anthocyanin molecule, and increases conversion to a colourless form. The degradation of anthocyanins and other phenolics occurs due to the activity of enzymes such as PPO and anthocyanase. POD may also play a role in the degradation of pericarp phenolics. The combination of reduced expression of red colour, and enzymatic degradation of pigments results in the rapid desiccation browning observed in lychee.

1.3.1.4 Lychee Microcracking

Microcracks are tiny cracks, around 20 to 100 μm in width, which occur on the surface of the lychee pericarp (Underhill and Simons 1993). The sclerenchyma layer in the pericarp is disrupted by groups of small collenchyma cells, which appear as fine white lines on the pericarp surface. With dehydration, these lines can develop into microcracks. The collenchyma cells appear to be a natural point of weakness in the skin and may facilitate moisture loss from the pericarp. Underhill and Simons (1993) considered the cracks to be the result of desiccation rather than the initial cause. However, it is likely that microcracks promote browning by facilitating further desiccation of the pericarp tissue. Microcracks would also be likely to encourage tissue browning through increased cellular disruption, and the exposure of underlying tissues to the air. While not yet proven to be directly involved in desiccation browning, it is likely that microcracks contribute to both moisture loss and browning processes.

1.3.2 Factors Affecting Moisture Loss

The loss of moisture from fresh produce is determined by the conditions of storage and the produce characteristics. Temperature, RH and air currents are the major variables influencing the pull for moisture from the atmosphere. The ability of the fruit to resist moisture loss is affected by gross morphology and the structure of surface tissues, which can be influenced by factors such as mechanical damage, maturity and pre-harvest conditions.

1.3.2.1 Storage Factors Affecting Moisture Loss

RH and temperature are the major storage factors influencing the rate of moisture loss. The very high water content of fruit and vegetables results in an almost constant pull for moisture from the atmosphere. The difference in water vapour pressure between the fruit tissue and the surrounding air drives the movement of water vapour from the fruit. This difference in vapour pressure is termed the vapour pressure deficit, or VPD (Scheer 1994). A higher VPD shows an increasing pull for moisture from the atmosphere, resulting in greater moisture loss. Low air RH is associated with an increased VPD, and hence greater rate of moisture loss. The VPD increases in a linear fashion as RH declines. For example, while 100% RH at 20^{o}C gives a VPD of zero, 90% RH results in 234 Pa, and 80% RH gives 468 Pa. Control of RH is most commonly achieved by the use of packaging, to maintain a high RH atmosphere in the air surrounding the fruit.

Both air temperature and the difference between air and fruit temperature affect the VPD. Air temperature strongly affects the VPD, with warmer storage resulting in faster moisture loss. At 70% RH, the VPD increases exponentially with temperature, from 183 Pa at 0°C to 701 Pa at 20°C, and 2213 Pa at 40°C. The VPD, and hence moisture loss, doubles from 0 to 10°C. A difference in temperature between fruit and air also affects the VPD. A warm fruit in a cool environment loses substantially more moisture than a cool fruit. For example, at 0°C and 70% RH, a 0°C fruit has a VPD of 183 Pa, while a 30°C fruit is subject to 508 Pa. Cooling of the fruit in air therefore unavoidably causes moisture loss. The main methods of reducing this effect are to either use hydrocooling, or reduce cooling time by fan-forced air-cooling.

Increasing air velocity generally raises the rate of moisture loss from fresh fruit and vegetables. The effect of air currents on moisture loss ranges from negligible, as in apples, to extreme, as observed in carrots (Van Beek and Lamers 1979). Exposure to slight air currents (0.05-0.15 m/sec) increased moisture loss in carrots by a factor of nine. Similarly, brussel sprouts stored under 0.1 m/s air flow lost 18 times more moisture than those protected from air currents (Van den Berg 1987). Although it has been observed that protection from air currents reduces lychee desiccation (Bagshaw *et al.* 1991), the relationship between moisture loss and air speed has not been examined.

When no air movement occurs, a gradient in RH can develop near the surface of the fruit tissue. Due to the limited movement of air molecules, air very close to the surface of the fruit increases in moisture content, effectively reducing the VPD. When an air current is introduced, the development of this gradient is reduced or eliminated. The effect of air movement on moisture loss generally increases with velocity until a certain point, after which further increases in velocity have little effect. For example, increases in air velocity above 0.07 m/s do not increase moisture loss in potatoes (Lougheed and

Argue 1987). It is likely that after this point the formation of a gradient in RH at the plant surface is negligible.

Factors influencing air movement in cool storage include the characteristics of the produce and the cooling system (Lougheed and Argue 1987). The fan type and setting influence the initial pressure and volume of air currents. Air movement is further affected by the system of delivery, such as forced-air systems or whole room ventilation. The dimensions of containers, stacking methods and resistance of the product to airflow can also influence the subsequent movement of air currents through the cool room. Air movement is used to rapidly reduce field heat and to maintain low temperatures during storage, but can cause substantial moisture loss in certain commodities.

1.3.2.2 Fruit Characteristics Affecting Moisture Loss

Various characteristics of the fruit influence the loss of moisture, ranging from gross morphology to tissue ultrastructure. Fruit shape and size can strongly influence moisture loss through the SA:V ratio. Surface tissues in the skin are crucial in regulating moisture loss. The almost saturated intercellular spaces in fruit tissue are separated from the surrounding atmosphere by protective barriers. These barriers reduce the ability of water vapour to move freely between the tissue and air, thus reducing moisture loss. The ability of the skin to resist moisture loss is affected by structure and composition of the protective layers (Chambers and Possingham 1963; Denna 1970). Processes such as mechanical damage, senescence and ripening can affect surface tissue properties, and may also affect the ability of the fruit to resist moisture loss.

1.3.2.2.1 Effect of Fruit Water Content

There is a progressive decrease in the rate of postharvest moisture loss as fruit water content declines (Van den Berg 1987). This has been attributed to tissue responses to moisture stress, such as stomatal closure, and a lag effect in moisture being drawn to the surface of the tissue. In addition, the slight decrease in temperature at the fruit surface caused by evaporative cooling would reduce the water vapour deficit.

The effect of water content on moisture loss also applies to freshly harvested fruit. Kiwifruit well-hydrated at harvest showed more rapid postharvest water loss than fruit harvested while water stressed (Burdon and Clark 2001). To some extent, this negates the benefit of harvesting at peak hydration. Water content at harvest is likely to be affected by tree water supply, influenced by factors such as rainfall, irrigation, soil properties and root development. Fruit water content shows substantial diurnal variation. In lychee, water potential was around –0.5 MPa overnight, but declined to –1.2 MPa from 9am to 3pm (Olesen 2001). Time of harvest can therefore strongly influence the desiccation process. The benefits of a reduced rate of loss in water stressed fruit may be outweighed by the lower absolute level of moisture. Symptoms of water stress would be expected to occur at a similar level of water content, regardless of rates of loss.

The supply of water during growth and development can also affect postharvest moisture loss. Fruit from well-watered apple trees showed greater postharvest water loss than those from trees subjected to water stress during fruit development (Kilili *et al.* 1996). However, pre-harvest water stress in carrots tended to increase postharvest water loss. This was accompanied by increased relative solute leakage, an index of plasma membrane permeability and cellular integrity (Shibairo *et al.* 1998). These changes relate to the effect of pre-harvest water supply on fruit structure and composition.

1.3.2.2.2 Effect of Fruit Gross Morphology

An increase in SA:V ratio would tend to increase the rate of moisture loss on a percentage of weight basis, as observed in capsicums (Lownds *et al.* 1993). For this reason, smaller fruit would be expected to show a greater rate of loss, if all other variables were constant. Irregular or elongated shape would be expected to slightly increase SA:V ratio compared to a round fruit shape. While lychee protuberances would increase the SA:V ratio of the fruit, this would be counteracted by the boundary layer effect, with air movement at the fruit surface restricted.

1.3.2.2.3 Effect of Fruit Tissue Structure

The gross structure of the lychee strongly influences the loss of moisture from the fruit. Zhao *et al.* (1999) carried out research into the drying characteristics of the fruit at high temperatures (50-70^{o}C) and found that the majority of early moisture loss occurs from the pericarp. After around 2 hours of intensive drying, the pericarp reached equilibrium with the drying medium, and no further weight loss occurred from the skin. After this point, weight loss slowed substantially. It was suggested that the membrane between aril and pericarp strongly resisted moisture movement. Similar patterns of moisture loss were shown under dry ambient conditions (25^{o}C and 60% RH), with rapid moisture loss in the first 24 hours. After 48 hours the pericarp had lost around half of its original weight, and further moisture loss was minimal (Underhill and Critchley 1993). Other fruit and vegetables, such as oranges and onions show the same differential moisture loss from the outer layers. For example oranges stored at 20^{o}C and around 70% RH for 2 months lost 9.5% of peel weight, but only 2.1% of pulp mass (Ben-Yehoshua 1969).

Surface tissue structure is particularly important in the ability of fruit to resist moisture loss. The fruit cuticle consists primarily of cutin and cellulose, with waxes embedded within, and overlying the basic structure. The thickness, structure and chemical composition of the cuticle vary widely amongst different commodities, and can also differ between various stages of growth in a single species. In general, most studies have shown that thickness or volume of the cuticle does not affect the rate of transpiration (Denna 1970; Maguire *et al.* 1999a). Moisture loss is often correlated with the total amount of epicuticular wax, as observed in capsicums (Lownds *et al.* 1993). In apples, the removal of epicuticular waxes resulted in a 30-90 fold increase in water loss (Horrocks 1964). The shape and structure of the epicuticular wax layers are the major features of the cuticular membrane significantly affecting moisture loss (Chambers and Possingham 1963; Denna 1970). Complex wax shapes, forcing water vapour through a longer path, create an effective barrier to moisture loss (Pantastico 1975). Wax platelets generally lie close together, forcing moisture to move through narrow capillary channels, bordered with hydrophobic surfaces.

Many pre-harvest and postharvest factors have the potential to influence the structure and composition of epicuticular waxes, as illustrated by the variation between orchards shown in mandarin waxes (Sala 2000). In citrus, both the structure and composition of the epicuticular waxes are affected by exposure to sunlight on the tree (McDonald *et al.* 1993). Shaded fruit showed smaller wax platelets that may confer greater resistance to moisture loss. In contrast, the sun and shade sides of apples did not significantly differ in water vapour permeance, but apples from the inner canopy lost moisture more rapidly than outer canopy fruit during postharvest storage (Maguire *et al.* 1999a). Differences in wax properties may have occurred due to the influence of growing conditions, such as RH or temperature. Mechanical damage may also strongly affect moisture loss. Pre-harvest contact between apples resulted in localised 30% greater water vapour permeance at the contact site (Maguire *et al.* 1999a). It would be anticipated that wax properties could also be affected by fruit maturity, ripening processes, senescence and pathogen growth.

The lychee cuticle is continuous, but irregular in width, varying from 1-3 μm. The epidermis has few natural openings, with infrequent stomata, and some lenticels in the basal area of each protuberance (around 20 per cm^2) (Underhill and Simons 1993). The structure and composition of epicuticular waxes in lychee have not been studied. Desiccation browning first appears on the apex of the lychee protuberance (Underhill and Critchley 1995), which suggests that the localised degradation of the cuticle on the protuberance observed by Underhill and Simons (1993) may be an important pathway for moisture loss. These observations suggest a possible link between mechanical injury and moisture loss that has not been further explored.

Microcracks in the lychee pericarp surface may have an influence on moisture loss, as these cracks disrupt the continuity of the water vapour barrier. Microcracks in the apple cuticle were estimated to be 12 time more permeable to water vapour than the intact cuticle (Maguire *et al.* 1999b). Microcracks are observed on the lychee fruit prior to harvest (Underhill and Critchley 1992), with the intensity of cracking significantly increasing during postharvest storage (Underhill and Simons 1993). The extent of microcracking at harvest is affected by pre-harvest water stress. Three days of moderate water stress at the late stage of fruit development increased the extent of microcracking in lychee by 65% (Kumcha 1998).

1.3.2.2.4 Effect of Fruit Maturity

At ambient temperature, the effect of maturity on lychee postharvest water loss was slight. Very immature fruit (<1/3 red) showed slightly higher moisture loss than immature fruit (1/3-1/2 red). More mature fruit (>1/2 red) generally showed the lowest levels of moisture loss, while water loss increased slightly in fully mature fruit (Wu *et al.* 2001). On average, the total percentage weight loss in fully mature fruit was around 0.7% higher than immature fruit (>1/2 red). For example, at 72 hours, the immature fruit had lost 8.1%, while fully mature fruit lost 8.8%. This corresponded to rates of loss 20-30% greater early in storage, but after 2 days the total effect was around 7-10%. Statistical analyses were not presented with these data. The effect of maturity on desiccation browning has not been established. Sittigul *et al.* (1994) studied the effect of maturity on postharvest browning, but fruit were stored at 5^{o}C and 90-95% RH, conditions under which desiccation browning would be unlikely to occur.

1.3.3 Prevention of Moisture Loss

Factors that influence moisture loss, such as temperature, RH, air movement and fruit barrier properties, can be manipulated to reduce moisture loss. The use of refrigeration combined with impermeable packaging is the most common method of minimising postharvest moisture loss. Packaging can reduce moisture loss by maintaining a high level of RH at the fruit surface and avoiding exposure to air currents.

However, storage at high RH also carries the increased risk of rot and condensation problems. The management of moisture loss needs to be balanced with other issues such as pathogen growth and condensation, to ensure a high level of fruit quality.

There has been a great deal of success in reducing lychee desiccation browning through minimisation of moisture loss. The use of low temperatures and packaging to reduce moisture loss is an established method of reducing browning, but can be difficult in practice due to temperature fluctuations, condensation and disease development. Surface coatings have been studied, with some success. However, the benefits have not been promising enough for commercial adoption. Control of pericarp browning through other means has been attempted, including the use of controlled atmosphere storage, sulphur-based treatments, hot water and acid dips and other novel chemical treatments. The benefits of improved colour may be outweighed by disadvantages such as reduced fruit quality, health issues and environmental concerns.

1.3.3.1 Prevention of Moisture Loss During Harvest

In practice, desiccation browning in lychee is minimised by good management throughout the handling chain, commencing at harvest. The water content of the fruit on the tree can vary diurnally (Olesen 2001), and with water supply. Fruit water potential can be maximised by ensuring a ready supply of water to the tree, and by harvesting at night or early morning. After harvest, the exposure of fruit to hot dry air can cause rapid moisture loss. Bagshaw *et al.* (1991) recommend covering exposed cartons with damp towels to reduce moisture loss, and transporting fruit to the packing shed as soon as possible after harvest.

Substantial benefit has been shown in rehydrating fruit after harvest by soaking in water. Immersion in water for 1 hour immediately after harvest changed fruit water potential from roughly –1.1 MPa to –0.1 MPa (Olesen 2001). This is approximately equivalent to a 6% increase in weight, and would be expected to significantly delay the onset of desiccation browning. The increase in weight also provides immediate benefit to crop value. The ability to rehydrate declined when fruit were stored in air prior to soaking. A 15 to 30 minute delay between harvest and soaking resulted in around a 4% gain, while 2% gain in weight occurred when fruit were soaked after an hour stored in air. Ensuring a high level of water content prior to handling gives a greater margin for permissible moisture loss.

1.3.3.2 Management of Temperature and Relative Humidity

The loss of moisture from fresh produce can be minimised by storage at low temperature and high RH. In practice, there are numerous limitations to the maintenance of low temperature and high RH, including problems such as excessive cost, disease development, chilling injury, temperature fluctuation and condensation. To develop an optimal postharvest handling system, a balance must be struck between minimising moisture loss and reducing the risk of disease and condensation, without excessive expense.

1.3.3.2.1 Pre-cooling to Reduce Moisture Loss

Temperature control is a key method of reducing postharvest moisture loss. Management of fruit temperature begins with pre-cooling, an initial cooling stage to remove field heat prior to packing. Pre-cooling can be carried out as fruit arrive from the field, or after other packing shed operations such as grading and sorting. Benefits of pre-cooling can be reduced if fruit subsequently increase in temperature on the packing line. Some postharvest operations include cooling of the entire packing shed to low temperatures to avoid fruit warming.

Pre-cooling minimises the temperature difference between fruit and air, thereby reducing the VPD that drives moisture loss. The placement of warm fruit into a cool room can be severely desiccating. A 20^oC fruit placed into a 0^oC and 90% RH cool room shows thirty times faster moisture loss than a fruit at 0^oC. The pre-cooling process reduces the loss of moisture during cooling by rapidly removing field heat prior to storage.

Pre-cooling also reduces the risk of condensation, which is caused by temperature differences between the packaging and produce, particularly during cooling. The condensation of water vapour can result from insufficient pre-cooling, or simply from small temperature fluctuations during storage. Droplets of water form on the packaging, reducing visual appeal and increasing the chance of free water occurring on the surface of the fruit. Severe condensation can result in pooling of moisture in the base of the package. The disadvantages of condensation include a decline in visual appeal, increased risk of rots due to free water on the fruit surface, and loss of moisture from the fruit. The fresh produce in the package is the source of most condensed water vapour (Joyce and Patterson 1994). Cooling of fruit to the storage temperature or slightly lower, prior to packing reduces the risk of condensation.

Pre-cooling can be carried out either in water or in air. Hydrocooling utilises cold water dips, sprays or ice slurries to rapidly chill the fruit. These techniques remove field heat very quickly, with minimal loss of moisture. However, the water remaining on the fruit surface can be problematic, potentially increasing pathogen growth during storage. Lychee fruit cooled in water showed little benefit in colour retention, and tended to show more disease development during storage, compared with air-cooled fruit (Olesen and Wiltshire 2000). Wet fruit can also cause box collapse in materials such as fibreboard. The packaging used after hydrocooling must have high wet strength to avoid collapse. Hydrocooling is used by some lychee growers, but the disadvantages and limitations may outweigh the benefits.

Forced-air cooling drives air flow in a single direction through a pile of fruit, and has substantial benefits compared with simply placing fruit in a cool room. The speed of cooling is much more rapid, as air is forced to move between and around fruit. In a normal cool room, air will follow the path of least resistance, moving across the surface of the crate. Using a forced-air system, cold air always passes over the coldest produce first, warming as it moves through the pile. In cool rooms the outer surface of the package cools, but convection currents move warm air from the interior, onto the cold outer fruit. This process causes condensation, and slows cooling. Due to the speed of cooling, forced-air cooling can use low RH air in some cases, without causing significant water loss (Joyce and Patterson 1994). Forced-air cooling can be applied after packing, reducing the double handling of fruit. However, this requires vents in the boxes and liners, which can increase moisture loss further down the postharvest chain. For this reason, cooling by forced-air prior to packing is more beneficial. Forced-air cooling may be a viable alternative for lychee, providing the benefits of rapid cooling, without causing fruit wetness.

1.3.3.2.2 Storage Temperature

In addition to reducing moisture loss, refrigerated storage extends the shelf life of fresh produce by limiting pathogen development and slowing metabolic processes. To reduce moisture loss, the optimal storage temperature is as low as possible without causing damage to the fruit tissue. In sub-tropical fruit such as lychee, storage temperature is generally limited by chilling injury, a physiological disorder triggered by cold temperatures. In lychee, chilling injury causes watery browning of the pericarp (Bagshaw *et al.* 1995). Incidence of chilling injury depends both on the temperature and duration of exposure. Chilling injury occurs after 30 days at 7^{o}C (Tongdee *et al*. 1982) or one week at 2^{o}C, but does not occur if fruit are stored for two days at 2^{o}C (Fitzell *et al*. 1995). The optimal storage temperature therefore depends on duration. For a shorter storage time, lower temperatures can be used without inflicting chilling injury. Olesen and Wiltshire (2000) found that storage at 5^{o}C for 3 weeks resulted in better colour retention and less weight loss than either 2 or 10^{o}C storage. Queensland Department of Primary Industries recommends a storage temperature of 5 to 7^{o}C for maximum quality and shelf life (Greer and Smith 1991).

1.3.3.2.3 Control of Relative Humidity

Refrigeration is of limited value in the management of moisture loss unless accompanied by some form of RH control. Maintaining a high RH environment within a cool room is possible but involves the use of bulky and expensive equipment (Joyce and Patterson 1994). It is usually simpler and more economical to use packaging to create a high RH atmosphere surrounding the fruit.

Condensation and pathogen development are the major limitations to the use of high RH storage. Condensation tends to be a greater problem at higher RH, as smaller changes in temperature are required to trigger the process. At 98% RH, a 0.3^{o}C fall in temperature results in condensation, while at 85% RH a 3^{o}C decline is required. Temperature fluctuations are difficult to avoid, particularly during transport. Rots are also more of a problem under high RH, particularly at RH greater than 90%. Humid conditions improve the ability of pathogens to infect, grow and produce spores. There is a need to balance RH to prevent excessive moisture loss, to avoid pathogen growth and to ensure that the temperature fluctuations encountered during postharvest handling will not result in condensation.

Packaging options for fresh produce commonly include the use of fibreboard or cardboard boxes, polystyrene cartons and punnets. The most effective barriers to moisture loss are made from extruded polymer films, such as polyethylene (PE), polyvinyl chloride (PVC), polypropylene and polystyrene, which have very low rates of permeability to water vapour (Joyce and Patterson 1994). In contrast, paper based packaging can actually desiccate the fruit by drawing moisture away. For this reason, cardboard boxes are generally waxed or used with plastic liners.

Liners for fibreboard boxes can be composed of various materials, including PVC or PE, and can vary in thickness and degree of perforation. These properties affect the permeability of the liner to gases and moisture, and can be manipulated to control moisture loss, gas exchange and condensation. Similarly, drainage or ventilation holes in punnets, and properties of the plastic over-wrap can be adjusted to provide a balance between condensation and moisture loss. Inserts within the package, such as salt sachets, can be used to absorb excess moisture, but these can be prohibitively expensive.

Novel solutions to the provision of high RH without condensation have been developed. Hydrophilic surfaces cause the formation of a continuous film instead of droplets, but the benefits are largely cosmetic, as the water tends to drain away more freely, giving increased pooling (Joyce and Patterson 1994). Active packaging involves a water barrier covering a paper capillary network, with an internal layer of fabric permeable to water vapour, but not free water (Patterson *et al.* 1993). This system reduces condensation by absorbing condensed water and releasing water vapour. The product combines minimal moisture loss from the produce with good control of condensation, even under extreme temperature changes (Patterson *et al.* 1993). However, the expense involved, and the inability to visually assess the produce may limit the adoption of this system. Shrink wrap packaging is in constant contact with the fruit, eliminating the difference in temperature between fruit and packaging (Joyce and Patterson 1994). Condensation is avoided, even in widely fluctuating temperatures. Limitations to this type of packaging include fruit shape and size, cost, and consumer perception of over-packaging. Shrink wrapping would be unlikely to be practical in lychee packaging due to the irregular surface and small fruit size. By the same mechanism as shrink wrapping, cryo-vac packaging of multiple fruit in a single layer reduces condensation. This type of packaging may have some potential in lychee. Gas exchange properties of the film would need to be considered to avoid an imbalance of gases within the very small airspace.

There has been extensive experimentation into the use of various types of packaging to extend lychee shelf life. Methods used have included paper bags (Mukerjee 1957), various types of plastic liner (Campbell 1959; Kremer-Kohne and Lonsdale 1991; Underhill *et al.* 1994), moisture absorbants (Mukerjee 1957), wax emulsions (Bhullar *et al.* 1983) and aluminium foil (Macfie 1955). Although results are frequently conflicting, the use of plastic bags or liners combined with low storage temperature generally provided the best extension of shelf life. The combination of packaging in plastic with chemical disease control and cool storage has shown consistent success, with browning delayed for up to 4 weeks (Huang and Scott 1985; Scott *et al*. 1982; Wong *et al.* 1991).

In the Australian lychee industry, a plastic liner within a cardboard box is the most common form of packaging. The comparison of various types of liners has shown significant differences (Underhill *et al.* 1994). A liner developed to absorb free moisture and release vapour showed the best overall results. Low density polyethylene of 75 μm thickness retained lychee colour, but condensation was a problem. Conversely, the PY-1 crispybag showed little condensation, but resulted in the most browning. The industry standard, PY-7 crispybag showed fairly high levels of browning, and average condensation. Fruit are usually removed from the liner at the retail level, resulting in severe moisture loss at this stage of the postharvest chain.

The marketing of lychee fruit in punnets covered with plastic film has been proposed as a solution to excessive moisture loss at the retail level. Combined with cool storage, punnets give very good control of desiccation browning, but have been poorly adopted by industry due to costs and adverse wholesaler reaction (Bagshaw *et al.*

1991). Similarly, a protective case with scoop was developed for lychee storage at the retail level, but this has not been broadly adopted, despite its success (Bagshaw 1995). At retail level fruit are often sprayed with cool water and packed into cool storage overnight to reduce browning.

Fruit coatings provide an additional barrier to moisture loss by physically restricting the movement of water vapour from the fruit. Coatings used commercially on other fruits have often proven unsuitable for lychee, either due to cracking of the wax (Underhill and Simons 1993) or an alkaline pH (Tongdee 1998). Several coatings have shown some degree of success in controlling desiccation browning. A 0.1% thiabendazole dip followed by the application of a 1% chitosan coating delayed the peak in PPO activity and reduced browning in lychee (Zhang and Quantick 1997). ProLong coatings (sucrose esters at 1.5 or 2.5%) also slightly delayed the peak in PPO activity and showed some success in reducing browning in lychee fruit stored at 4^{o}C (Zhang *et al.* 1997). Polysaccharide coatings were evaluated by York (1995), and were shown to delay browning to some extent, but were not considered commercially viable. Despite some success, coatings are not commonly used in commercial operations.

1.3.3.3 Chemical Control of Desiccation Browing

Research into alternative methods of controlling lychee desiccation browning has been extensive. Sulphur based treatments can fix the red colour of the pericarp, but are generally declining in use due to health concerns. The combination of heat treatment with acid dipping shows great benefit in colour retention, and has progressed to commercial development (Kaiser 1998; Lichter *et al.* 2000).

1.3.3.3.1 Sulphur Based Treatments

Although not used in Australia, sulphur based treatments, particularly sulphur dioxide fumigation, are widely applied in many lychee growing areas world-wide. Sulphur dioxide controls browning through the inhibition of PPO (Zauberman *et al.* 1991), and by combining with anthocyanin to form a more stable complex (Markakis 1982). Sulphur treatments also have a fungicidal effect, eliminating many postharvest fungi.

Sulphur dioxide is applied by fumigating fruit in a sealed container for 20 to 30 minutes. Treatment with sulphur causes an immediate loss of redness from the fruit, resulting in a pale creamy colour. Colour returns gradually to the fruit over 24 to 48 hours, with the rate of this process depending on storage temperature (Underhill *et al*. 1992). After the colour has developed, the pigment is fixed in the fruit, and subsequent browning will not occur. One of the major problems with this technique is that the fruit colour returns as a dull orange rather than the natural bright red colour.

The return of pericarp colour after sulphur treatment can be hastened by the application of acid dips after SO_2 treatment (Zauberman *et al*. 1991; Underhill *et al*. 1992). Underhill *et al*. (1992) used 1N hydrochloric acid as a two minute dip after SO_2 treatment, and found that the acid rapidly improved skin redness. Furthermore, full pericarp colour was retained for eight weeks, despite the fact that the aril had decayed. Zauberman *et al*. (1991) found some fading of colour during storage. In addition to causing more rapid colour development, acid dipping reduced the activity of enzymes such as PPO and POD, thus limiting subsequent browning (Underhill *et al*. 1992). Reduced pH also increases the stability of anthocyanin, and could result in more anthocyanin being in the red-coloured form (Tongdee 1998). This may be an additional mechanism of increased redness in acid treated fruit.

Problems with the use of SO_2 on lychees include weight loss, residues and bleaching. The sulphur treatment can cause significant weight loss, reducing the commercial value of the fruit (Kremer-Kohne and Lonsdale 1991). In addition, *Penicillium* spp. can be a problem during storage due to the biological vacuum created on the fruit surface. Residues on the fruit can cause an undesirable aftertaste. Pulp residue levels vary widely between cultivars, with ‘Bengal’ fruit showing particularly high levels of 135 ppm after 48 hours (Underhill *et al*. 1992). There is increasing resistance to sulphur use due to health risks. The limit for residues is 10 ppm in Australia, some parts of Europe and Japan (Paull *et al*. 1995). Some countries, such as Singapore, have adopted a zero tolerance policy towards sulphur residues (Tongdee 1998).

The problems encountered with SO_2 have lead to increased experimentation with other forms of sulphur. An eight minute dip in 1% sodium bisulphite ($NaHSO_3$) with 0.5% HCl was thought to result in reduced sulphur residues, and gave good fruit quality (Jiang *et al*. 1997a). Paper sheets impregnated with sodium metabisulphite ($Na_2S_2O_5$) were also found to give good browning control (Schutte *et al.* 1991). Duvenhage (1993) showed that a dip in sodium metabisulphite followed by a HCl dip gave good results in reducing browning. Wang *et al*. (1996) recommended a ten minute dip in 10% citric acid and 0.2% sodium benzoate as a cheaper and easier alternative to SO_2 fumigation and an acid dip. While these treatments may give some benefits over SO_2, it is likely that the decreasing tolerance for sulphur residues will result in a shift away from sulphur-based treatments.

1.3.3.3.2 Manipulation of pH

Acid dips have been used to attempt to delay the onset of browning, both alone and in conjunction with other treatments such as sulphur and heat. Duvenhage *et al.* (1995) compared 8% HCl alone against combinations of acid and sulphur treatments, and found that the acid dip showed some success, but caused some browning and decline in flavour in certain cultivars. Underhill and Critchley (1994) showed that the effects of pH were reversible, with alkaline dips causing discolouration, and acid dips restoring red colour. Lychee fruit allowed to brown in ambient conditions for one week, and then dipped in acid for two minutes were restored to the redness of freshly harvested fruit (Underhill and Critchley 1994). However, these results were not stable, and further deterioration occurred with storage. To be fully effective acid dips appear to need a precursor treatment to increase pericarp permeability (Kaiser 1995).

Extreme heat treatments of short duration have been shown to have some beneficial effect on lychee pericarp colour when used in association with acid treatments, as an alternative to sulphur. It is thought that heat or sulphur treatments increase the permeability of the pericarp, thus allowing acid to reach the vacuoles (Holcroft and Mitcham 1996). Kaiser (1995) experimented with acids, detergents and heat treatments as a means of increasing pericarp permeability. Acid treatments alone were found to result in a corky brittle pericarp, and detergents required an extended dipping period to be effective. Immersion in 98^{o}C distilled water was shown to effectively break down the membrane, resulting in a pliable colourless pericarp. Following the heat treatment the fruit were immediately hydocooled in 21^{o}C water adjusted to pH 0 with HCl. A delay between the heat and acid treatments resulted in browning of the pericarp, probably due to excessive exposure to heat. In contrast, Tongdee *et al*. (1998) reported that a delay of one hour between heat treatment and the application of an acid dip resulted in reduced browning. Kaiser (1995) found that a thirty second 98^{o}C heat treatment followed by a three minute dip at pH 0 resulted in a pliable red pericarp. However, this treatment resulted in some discolouration of the aril in the pedicel region. Tongdee (1998) observed a pink staining on the aril when using acid and hot water treatment of 3 seconds at 85^{o}C. Heat treatments used to increase pericarp permeability will need to be adjusted to specific cultivars, as they vary widely in their sensitivity to heat injury (Tongdee *et al*. 1998).

Alternatives to hot water dips, including sprays and steam have proven successful. Kaiser *et al*. (1995) exposed fruit to steam at 95^{o}C for 2 seconds to damage the cuticle and solubilise cell and organelle membranes. Two seconds of steam, followed by a 4 minute dip at pH 0 and treatment with an anti-transpirant resulted in firm fruit with good taste and colour after 28 days storage at 1^{o}C. Commercial trials held in South Africa in 1995-1996 proved successful on the cultivars ‘Mauritius’ and ‘Hong Huay’ (Kaiser 1998). A hot water brushing system was developed for commercial use, with fruit sprayed with hot water while being brushed in a revolving drum, prior to treatment with 4% HCl and fungicide (Lichter *et al.* 2000). This treatment resulted in inhibition of PPO, and retained red colour for long term storage. Heat treatments may provide the additional benefit of a reduction in pathogen load, but, as with sulphur treatment, *Penicillium* spp. problems have been recorded (Lichter *et al.* 2000).

Heat and acid treatments provide a solution to the cosmetic problem of desiccation browning, by fixing the red colour of the lychee pericarp. Disadvantages of the system may include registration difficulties, due to the minor status of the lychee crop and occupational health and safety issues due to the use of acids and steam. The necessity to calibrate the system for each cultivar, and the high initial cost of equipment would be limiting, particularly for smaller scale orchards. The system also carries the potential for large scale loss due to incorrect calibration. However, fruit treated by this system would tolerate substantial moisture loss without a reduction in quality, and could therefore be stored at lower RH, reducing pathogen and condensation problems.

1.3.3.3.3 Novel Chemical Treatments

Some novel chemical treatments have shown promise in reducing desiccation browning. A dip in calcium nitrate at 1% for 5 minutes, followed by 4% HCl for 3 min gave results comparable with sulphur and acid treatments, and may be a promising technique (Duvenhage *et al.* 1995). A five minute postharvest dip in citric acid (100 mmol/L) and glutathione (10 mmol/L) reduced PPO activity by 80% compared to the control, and significantly reduced browning (Jiang and Fu 1998). Jiang and Chen (1995) tested postharvest treatments of polyamines for their effect on browning, and found that spermine was the most effective of those tested in reducing browning. Polyamines inhibit the production of ethylene, stabilise membranes and reduce peroxidation (Holcroft and Mitcham 1996). Despite some success, the difficulties involved in registering chemicals for use on minor crops may limit the adoption of these types of treatments.

1.3.3.3.4 Controlled Atmosphere Storage and Modified Atmosphere Packaging

Controlled atmosphere (CA) storage has the capacity to reduce pericarp browning, but is not commonly used in lychee storage. Kader (1994) suggests that 5%

O_2 reduced skin browning and PPO activity, with good benefits observed. Carbon dioxide at 3-5% reduced the rates of loss of ascorbic acid, acidity and soluble solids in the pulp, with moderate improvement in quality. Yuan (1997) stored lychees successfully for 40 days under an atmosphere of 3-5% O_2 and 3-5% CO_2, at 4^oC. Vilasachandran *et al.* (1997) observed no significant benefits in colour retention using CA storage, but stem end decay was reduced. In rambutan, a close relative of lychee, enhanced CO_2 at 7-12% slowed the rate of postharvest skin browning (O'Hare and Prasad 1990; Mohamed and Othman 1988).

Levels of O_2 below 1% or CO_2 above 15% have the potential to induce off-flavours in lychee and a dull grey colour in the aril, although 10-15% CO_2 may provide some benefits in control of rots (Kader 1994). Datta *et al.* (1963) reported that an atmosphere of 15.7% O_2 and 25% CO_2 at 4.4^oC retained colour, flavour and texture for 30 days, with no spoilage. In contrast, Vilasachandran *et al.* (1997) observed the development of off-flavours at 15% CO_2.

Modified atmosphere packaging (MAP) uses the gas exchange properties of packaging to manipulate the atmosphere surrounding the fruit during storage. Published results of MAP use have generally not been successful, due to rot development (Ray 1998) or skin browning after removal from storage (Lonsdale 1993). Further research is needed to develop a commercially viable MAP system.

1.3.3.4 Overview of Control of Lychee Desiccation Browning

The reduction of moisture loss by refrigeration and packaging is the major tool currently used to control postharvest browning. Sulphur-based treatments are an established method of control in many areas of lychee production worldwide, but are likely to decline due to low residue limits in many countries. A system involving the application of heat treatments followed by acid dipping has shown great benefit in retaining the red colour of the skin, and could progress to wide-scale commercial use. The heat-acid system reduces the need for high RH storage, simplifying the storage needs of the fruit. However, cost, registration difficulties and potential fruit damage may limit the adoption of this technique. Careful management of RH and temperature currently remains the optimal method of avoiding desiccation browning.

1.4 Mechanical Damage

1.4.1 Causes of Mechanical Damage

The causes of mechanical injury are numerous, and are often broadly grouped into impact, abrasion, compression and vibration damage, based on the type of force acting on the fruit (Sitkei 1986). Impact damage is characterised by a momentary application of force, which may occur when fruit is dropped, when an object drops onto fruit, or when fruit rolls into a barrier. In contrast, compression damage involves a static applied force, such as the weight of stacked fruit above another fruit. Abrasive damage results from the rubbing of fruit against each other, or against some other surface. Vibration damage occurs when fruit are subject to vibration forces, such as during transport. This type of stimulus can result in, or exacerbate impact, abrasion and compression injuries. These broad groupings define the most common causes of mechanical injuries. Other types of damage that do not fall into these categories include cuts, tears, scratches and punctures. These are severe forms of damage, generally caused by poor equipment or handling, and may contribute significantly to postharvest losses.

1.4.1.1 Impact Damage

Impact is a frequent occurrence during the harvesting, postharvest handling, packing, transportation and distribution of fruit, and has been identified as the most important cause of mechanical injury in fruits (Ruiz Altisent 1991). Impact can occur between two fruit, or against other surfaces during postharvest operations. Many impacts would be expected to occur during the transfer of fruit, for example, from tree into harvest buckets, into containers for bulk transport and during placement onto and movement along the packing line. Impact can also occur after fruit are packed, due to dropping of boxes, or tipping and sliding of cartons during transport.

In lychee, falls during harvest would generally occur onto soft surfaces, such as canvas picking bags, other fruit or the ground. However, with trees often reaching 9 to 12 metres in height (Kadam and Deshpande 1995), falls during harvest could be a potential source of injury despite the soft impact surfaces. Impacts against tree branches during harvest would also be a potential cause of damage. Impact damage would be a particular risk if fruit were tossed downwards, with a high starting velocity. Other possible causes of impact during lychee harvest may include the emptying of picking bags into crates.

Mechanical harvest creates additional opportunities for impact, between fruit and against branches and catching surfaces (Sitkei 1986). Fresh market fruit are very rarely harvested mechanically, so this is currently not an issue of concern. However, rising labour costs make this an increasingly attractive alternative, which may be of future importance.

The most important type of impact in practice is of fruit against a stationary, rigid surface (Sitkei 1986). This type of impact could occur when fruit are dropped into a storage bin or when they roll into the side wall of a packing line. Cushioning can reduce the damage caused by impact, but injuries can still result if the forces are great enough. The impact of one fruit against another stationary fruit can also be an important cause of damage. This can occur during harvesting and transfer, when fruit are falling onto other stationary fruit. It can also occur on the packing line if fruit are unable to shift their centre of gravity (for example, if wedged into a corner). While other stresses can occur, they are usually not as important in causing impact damage in general practice (Sitkei 1986).

The impact damage of fruit can occur after packing, due to dropping of boxes during handling, and the movement of fruit within boxes during transport. For boxes of apples, drops as minimal as 5 cm were capable of causing bruising damage (Schoorl 1972b). In the retail sector, further mechanical damage can occur due to rough handling of the fruit by staff or customers. Impact damage is thus a concern throughout the postharvest chain, from harvest to marketing.

1.4.1.2 Compression Damage

Compression forces result from static weight or pressure acting on a product. The long duration and static loads involved in compression often result in a creep phenomenon (Sitkei 1986). Creep occurs where the load on the fruit is constant, but the tissue continues to deform over time. Thus for compression injuries, both the duration and the magnitude of the load are important in determining the extent of injury.

The static pressure of fruit, caused by the weight of fruit above, is the major cause of compression damage (Sitkei 1986). This may occur in bulk bins, particularly if the depth of stacked fruit is high. It has been suggested that lychee are susceptible to this type of compression damage, with field container depths of less than 30 cm recommended to avoid damage (Batten and Loebel 1984). In contrast, Greer (1990) recommends a depth of 50 cm. Research has not been carried out to ascertain a threshold of compression damage for lychee.

Bridging occurs between fruit stored in bulk bins, reducing the weight resting on lower fruit. However, this arrangement can be destroyed by vibration in transit, increasing the chance of compression injury (Schoorl 1974). Compression damage caused by stacked fruit is localised to the bottom layers of the stack, as these receive the greatest load from above.

Packing fruit into cartons reduces the compression forces acting on stacked fruit, with the carton absorbing most of the load. However, box failure can result in fruit carrying the load of the box above. Box failure generally occurs due to the placement of excessive weight on the box or a decrease in the strength of the box (Schoorl 1974). The strength of boxes is greatly affected by humidity, with 20% moisture in the box resulting in a 55% loss of strength compared to 10% box moisture (Kellicut 1960; Stott 1959). These types of moisture levels occur respectively at 92% and 68% RH (Steenburg *et al.* 1963) making box failure due to changes in humidity a potential risk.

Compression can also occur during packing due to lidding pressure. When pressure is applied to one or more fruit in a container, the force is transmitted to the adjacent fruit. Closed lids can transmit pressure down through several layers of fruit (Schoorl 1974). Overfilling of boxes makes this type of compression injury more likely.

1.4.1.3 Abrasion Damage

Abrasion is generally a less important form of postharvest mechanical damage than impact or compression (Sitkei 1986), but is well recognised as a pre-harvest injury, often termed wind rub (Bagshaw *et al.* 1995). Abrasion occurs when one surface slides across another, causing friction. Although friction depends on adhesion and ploughing, it is only ploughing that affects the extent of abrasive damage. Ploughing is caused by surface irregularities, which act like chisels against the fruit surface, removing cuticle and wax layers (Schoorl 1974).

Abrasion can occur with the movement of two fruit surfaces against each other, a common type of pre-harvest injury due to windy conditions. Similarly, rubbing of the fruit against branches or leaves can also cause pre-harvest abrasion. Pre-harvest abrasion of lychees has been observed to occur when fruit rub against other fruit, leaves or twigs during the final stages of fruit development (Bagshaw *et al.* 1995).

Postharvest abrasion between fruit may occur due to vibration during transport (Hilton 1994), for example, if one fruit is stationary, while another moves slightly, causing a rub between the two. Abrasion against other surfaces, such as picking bags, storage bins or packing line components is also possible, with rough or irregular surfaces most likely to cause damage. It has been suggested that postharvest surface rub is not a problem in lychee (Lindsay and Cull 1986). Although postharvest damage due to abrasion is generally less of a concern than other types of mechanical damage, it can cause significant and economically important damage in some crops, such as pears (Mellenthin and Wang 1974).

1.4.1.4 Vibration Damage

Vibration damage occurs when fruit move around or rotate within a package due to a vibration stimulus (Hilton 1994). Damage can be caused by fruit striking against other fruit or packaging, giving an impact injury, or by rubbing of the fruit against another surface, resulting in abrasion. Vibration can also exacerbate compression injury by increasing static pressures acting on fruit (Sitkei 1986). Vibration injury may cause only one of these types of damage, or all three. For example, in the transport of kiwifruit, Lallu *et al.* (1999) found that vibration generally resulted in abrasion of the skin, with a smaller amount of compression damage and little impact injury.

Vibration injury most commonly occurs during transport, with the interaction of the road and vehicle suspension system generating vibration. The vibration caused during transport is semi-random, occurring across a large range of frequencies and with jolts and bumps in the road adding to the background vibration (Hilton 1994). The irregular nature of vibration input makes it difficult to define a threshold for vibration damage. Fruit will vibrate when the frequency of vibration reaches a certain level. If the resonance frequency of the fruit column is the same as the excitation frequency of the vehicle or road, the acceleration of the fruit can increase considerably due to resonance, and severe damage can result (Sitkei 1986). In stacked or palletised produce, the vibration can be directed up through the stack, increasing in magnitude at higher levels (Sitkei 1986). For this reason, displaced cartons and vibration injury are most common at the top of stacks. Vibration injury within a box of fruit is also localised to the top layers, as these fruit are most capable of movement.

It is likely that transportation by truck is the major source of vibration in lychee postharvest handling. Truck vibrations would be encountered during the transfer of stacked crates from orchard to packing shed, and during transportation to market. In lychees, mechanical destalking is an additional possible cause of vibration injury. Mechanical destalking often involves the deliberate agitation of the fruit to encourage stem breakage at the natural abscission point. Menzel *et al.* (2002) observed that the process can potentially damage the lychee skin surface, resulting in browning.

1.4.1.5 Other Causes of Damage

Severe mechanical injury may disrupt the integrity of the fruit skin, causing a cut, scratch or puncture. A cut occurs when a sharp edge penetrates the product, without significant crushing of the tissue, as would be the case with a sharp secateur or knife blade. Scratches are generally accompanied by tissue crushing, and result from the fruit surface dragging against a sharp point. Punctures are caused by pointed objects such as nails, stems or thorns, which penetrate the fruit surface and the tissue beneath. These severe types of damage result from poor handling or equipment, and are generally not observed as a problem in lychee, although it has been suggested that lychee are susceptible to puncture (Lindsay and Cull 1986).

In contrast, the tearing of the lychee skin due to stem pulling is a potential source of significant postharvest loss. The lychee fruit has a natural point of abscission that allows it to be harvested by holding the base of the stem and twisting the fruit (personal observation). This results in a short stem remaining attached to the fruit, the typical form of presentation used in Australian markets. Fruit may be harvested using this method, or cut from the tree in panicles using secatuers. Panicles may then be separated into individual fruit by the use of mechanical destalkers, or by hand. Hand destalking involves either twisting at the natural point of abscission, or cutting with secatuers to leave a short stem. Pulled stems result from excessive linear force (pulling on the fruit, rather than twisting), which may occur during harvest, or hand destalking (personal observation).

1.4.2 Fruit Responses to Mechanical Injury

Symptoms of mechanical damage may include tissue darkening or other changes in colour, changes in sensory qualities, cracking, splitting, shape distortion and the formation of scuff marks or corky deposits. The symptoms appearing as a result of mechanical damage, depend on both the characteristics of the product and the properties of the force inflicted.

1.4.2.1 Bruising

Bruising is a typical symptom of impact, compression and vibration damage, and may involve changes in appearance, texture and taste. Ruiz Altisent (1991) defines a bruise as a “volume of fruit tissues below the skin that is discoloured and softened”. Bruising may occur on the surface, or internally, depending on the mechanical properties of the load and the fruit. Internal blackspot of potatoes and external bruising of apples are extreme examples.

Tissue browning or darkening is often the most obvious sign of bruising. Browning results primarily from enzymatic processes triggered by cellular disruption (Ruiz Altisent 1991). However, discolouration may be exacerbated by accompanying increases in moisture loss or pathogen invasion. The cellular damage associated with bruising can result in a significant decline in sensory qualities. Mechanical injury typically causes softening of texture due to cell rupture and breakdown, and subsequent enzymatic activity (Jaensch 1996). In other cases tissue may become firm and brittle due to injury, as in mangosteeen (Ketsa and Atantee 1998). The symptoms of bruising, including changes in appearance, texture and flavour, markedly reduce the appeal of an injured fruit.

The first signs of bruising may appear within as little as four hours after injury, with full bruise development often occurring after around 24 hours (Hung 1993). However, under certain conditions it may take three days or longer for a bruise to appear (Jaensch 1996). The latent nature of this injury can make it very difficult to detect in the early stages.

There is little reference to bruising in lychee, and it is not commonly recognised as a defect. The bruising of lychee fruit is mentioned in an extension report by Bagshaw *et al.* (1991), and it is suggested that over-mature or large fruit may be more susceptible. The nature of the injury is not described in any further detail, and it is not specified whether the discolouration extended into the aril. Greer and Smith (1991) state that lychee fruit are not easily damaged or bruised. It is not specifically included in the defect chart used in packing sheds, and is not mentioned in any other literature. While bruising may occur under specific conditions, it is certainly not a common occurrence in lychee fruit. Rambutan fruit, closely related to lychee, show a skin darkening response to mechanical damage. Rambutan spinterns bent using a glass rod showed significantly greater browning, which was thought to have occurred due to enzymatic processes (Landrigan *et al.* 1996a). A similar response in lychee has not been tested.

1.4.2.2 Abrasion Damage

Abrasion may result in scuff marks or numerous small scratches. Different abrasive surfaces may affect different layers of the fruit surface. For example, abrasion against a smooth surface may affect only the cuticle, with coarser material affecting deeper layers of the skin tissue. This was shown in watermelon, where different abrasive surfaces resulted in failure at different levels of the fruit tissue (Puchalski and Brusewitz 1996). Abrasion can remove surface waxes, or the entire cuticle, and may damage underlying tissue.

Abrasion injuries can trigger a wound healing response in many fruit and vegetables. Healing may result in changes in colour and texture, for example, due to deposits of corky material (Jaensch 1996). Injury symptoms caused by abrasion may take some time to appear. For example, superficial scratches on citrus fruit may be barely visible, but can result in scar development during long term storage (Golomb *et al.* 1984).

Pre-harvest abrasion of lychees has been suggested as the cause of a variety of damage symptoms. Scattered dark blemishes observed on the fruit protuberances are thought to result from wind rub in the late stages of fruit development (Bagshaw *et al.* 1995). Personal observation of fruit on the tree suggests that the symptoms of pre-harvest abrasion may also occur as a yellowish discolouration. In addition, lychee silvering may be a symptom of abrasion, appearing as distinctive scattered silver marks on the fruit surface. It has been hypothesised that this response may occur due to rubbing, excessive heat or dehydration from dry winds (Bagshaw *et al.* 1995). A similar injury occurs in citrus, where pre-harvest wind rub against twigs or thorns in small citrus fruit can result in a thin tan to silvery scar as the fruit matures (Smoot *et al.* 1971). Postharvest abrasion injury due to the use of incorrectly adjusted destalking machines has been observed in lychee. The upper layer of skin was thought to have been rubbed off, with browning resulting overnight (Menzel *et al.* 2002). Bagshaw *et al.* (1991) also mention the possibility of brush damage as a postharvest injury.

1.4.2.3 Other Symptoms of Mechanical Injury

Mechanical damage symptoms unrelated to bruising or abrasion damage include cracks, splits, tears, punctures and distortion of shape. Cracks in the fruit are one of the most obvious signs of a severe mechanical injury, and can occur due to compression or impact. Cracks can range from microscopic splits in superficial tissues, to large gaping wounds extending into the tissues underlying the skin. Splitting is an even more severe injury, with the fruit divided into several parts. Cracking and splitting are generally a result of poor handling, and would normally render fruit unmarketable. In lychee, “splitting” refers to a pre-harvest cracking disorder unrelated to mechanical injury (Kumcha 1998). Cracking has been observed in the lychee skin during the development of a hot water brushing system (Lichter *et al.* 2000), but the damage may have related to turgor rather than mechanical injury. A delay between hot water brushing and acid dipping reduced the incidence of cracking, suggesting that the injury was caused by the acid dip. Cracking is not specifically mentioned as a postharvest issue in any of the available lychee postharvest reviews (Chen *et al.* 2001; Holcroft and Mitcham 1996; Nip 1988; Shi *et al.* 2001).

In addition to cracking, the integrity of the fruit skin may be breached by punctures, scratches, cuts and tears. Personal observation of different wound types has shown that punctures usually occur as concise injuries, with surface tissues indented. Superficial scratches due to abrasion may appear merely as lines of removed surface waxes or cuticle. Deeper scratches show jagged wound edges, while cuts are distinguished by a smooth wound edge. Tears also show rough wound edges, and are likely to show some vertical separation of the skin, with one wound edge pulled upwards. Tears are most often caused by force applied through the fruit stem. Skin tearing due to pulled stem damage is regularly observed in lychee (personal observation). These types of injuries may be associated with a bruising response, due to compression or impact forces applied to the surrounding tissue. In addition, injuries such as cracks and scratches are likely to result in some associated browning due to cell rupture.

Distortion of fruit shape is generally caused by static loads acting on a product, but may also result from impact (Sitkei 1986). Small distortions would generally have little effect on fruit appearance. However, in certain highly plastic fruit such as mandarins, distortion can be extreme and may reduce the visual appeal of the fruit (personal observation).

Lesion browning in lychee is another possible symptom of pre-harvest mechanical injury. Lesions are characterised as brown to black spots with a sharp edge, most commonly observed in the cultivar ‘Tai So’ (Bagshaw *et al.* 1995). Large lesions result in the direct loss of fruit as rejects, while fruit with small lesions may be marketed. Fruit marketed with lesions may have an increased chance of further mechanical damage, as the affected skin is brittle and will crack under slight pressure. Bagshaw *et al.* (1995) theorised that lesion browning was caused by mechanical damage or water stress during fruit development. Joubert and van Lelyveld (1975) found increased levels of PPO and POD in this type of injury and suggested that mechanical damage may be a cause, but no further evidence was given to support this hypothesis. The appearance of the lesion, as a concise area of damage affecting whole protuberances, is at odds with most observations of mechanical injury on lychees. Other pre-harvest injuries caused by mechanical stress show only scattered marks localised to the protuberance tips. One possibility is that the lesion blemish may result from wound healing, where damaged tissue is sealed off by the development of a layer of callus or suberin cells. Further research is required to confirm the causes and mechanisms of this response.

1.4.2.4 Microscopic Changes in Damaged Tissue

Bruising results when the load applied to a fruit, either through compression, impact or vibration, exceeds a certain threshold (Ruiz Altisent 1991). The application of a certain level of force causes strain energy to dissipate in the tissue, causing physical changes to cells, either through the disruption of cell contents or the rupture of cells (Sitkei 1986). The application of a small amount of force to fruit tissue causes distortion of the cell and cell wall distension (Holt and Schoorl 1977). This type of injury may disrupt the cell contents, but the cell wall remains intact. When the application of force exceeds the elastic limit of the cell wall, the cell ruptures, releasing cell contents into the intercellular space (Sitkei 1986). Under the microscope, ruptured cell walls are often observed as a symptom of damage due to injury by puncture (Spotts *et al.* 1998), impact (Underhill *et al.* 1998) and compression (Gould *et al*. 1990).

Ultrastructural changes occurring due to bruising have been observed in apples by electron microscopy. During bruise formation intensive vesiculation was observed in the damaged area within a few hours (Rodriguez *et al.* 1990). The vesicles formed either within vacuoles or in the middle lamella, between cells. Bruising is often considered a result of cell rupture, but this is not always the case. Intensive vesiculation was observed in intact cells subjected to low levels of stress, showing that rupture is not a prerequisite to bruise development. Bruising may occur due to either cell rupture, or the disruption of cell contents.

The microscopic study of compression damage in pepino fruit showed that cellular disruption was localised to the mesocarp (Gould *et al*. 1990). Exocarp cells were compacted by the injury, but were not structurally damaged. The damage to the mesocarp was characterised as partial cell collapse, the dislocation of cells from adjoining cells, and cell wall rupture. It was found that the damaged tissue showed a smaller average cell area and reduced air space due to compaction of the tissue. In tomato, deformation due to compression was observed to occur due to air being forced from the tissue, both from the loculus cavity and from intercellular spaces in the pericarp (Pereira and Calbo 2000). Pitt and Chen (1983) suggest that fluid also moves out of cells under compression, contributing to gradual tissue deformation.

Abrasion damage is characterised by injury to the skin surface, with deeper damage occurring only in severe cases. In oranges injured using sandpaper, microscopic examination of injured tissue showed the loss of the cuticle and destruction of the epidermis. Some parenchyma cells were also injured, some showing plasmolysis and disruption of plastids and mitochondria (Brown and Barmore 1981). Very mild abrasion (caused by rubbing gently with a washcloth), applied to peaches and nectarines was shown to disrupt the cuticle, compress cell walls in the epidermis, rupture cells and cause cracks on the fruit surface (Crisosto *et al.* 1993b). Underhill (1993) observed deterioration of the cuticle in lychee during development and after harvest, particularly on the protuberance tips. It was suggested that this damage was a result of physical abrasion due to wind rub, harvesting and postharvest handling.

1.4.2.5 Biochemical Responses to Mechanical Injury

Analysis of wounded tissue has shown that mechanical injury can cause significant changes to biochemical processes, prompting the development of damage symptoms. The cellular disruption and decompartmentalisation resulting from injury can cause tissue browning by bringing enzymes and substrates into contact (Ruiz Altisent 1991). Ethylene synthesis is a common wounding response, and may trigger senescent processes, such as the release of tissue softening enzymes (Kader 1985). Mechanical damage to fresh produce can result in mechanisms of resistance being activated. These responses may include the development of physical barriers such as lignin or callose and the formation of active chemicals such as phytoalexins (Lyon *et al.* 1995).

1.4.2.5.1 Ethylene Synthesis

Increased production of ethylene after wounding is a commonly observed response in many fruits, such as pear (Mencarelli and Botondi 1992), apple (Lougheed and Franklin 1974) and tomato (MacLeod *et al.* 1976). However, in some produce, such as sweet cherry (Wade and Bain 1980) and papaya (Quintana and Paull 1993), this response is not significant. In sweet potatoes, ethylene production was localised to cells located near the damaged tissue (Imaseki *et al.* 1968). However, a whole fruit response can also be observed, when ethylene diffuses throughout the intercellular spaces, as occurs in apple (Lougheed and Franklin 1974). In tomatoes, the ethylene response following impact injury was significant within an hour, and persisted for at least 13 days (MacLeod *et al.* 1976). Cooling of fruit after injury can inhibit the ethylene response, as shown in apricot (DeMartino *et al.* 2002).

Ethylene can trigger the increased activity of various enzymes, leading to tissue browning and softening (Kader 1985). In squash (*Cucurbita maxima*), ethylene production due to wounding was followed by a rapid increase in peroxidase activity in the mesocarp tissue, which did not occur when ethylene inhibitors were applied (Hyodo *et al.* 1991). However, enzyme activation through wounding can occur independently of ethylene stimulation. In cucumber, substantial changes in enzyme activity due to mechanical damage were not preceded or accompanied by increases in ethylene synthesis (Miller *et al.* 1987).

Ethylene tends to promote senescent type changes in the tissue, thus accelerating tissue degradation. Increases in ion leakage, symptomatic of loss of membrane integrity, are commonly shown as a response to mechanical injury, as observed in tomatoes (Fiore *et al.* 1992). This type of response may occur both through the initial damage to cells, and subsequent acceleration of tissue breakdown. Damaged tomatoes also show a loss of nutritional content, typical of senescence, with vitamin C levels in the pericarp and aril declining after injury (Moretti *et al.* 1998). Higher levels of ethylene also tend to cause increased respiration and greater cell decompartmentalisation (Kader 1985). In climacteric fruits ethylene also hastens ripening, leading to increased respiration and reduced shelf life. The non-climacteric nature of lychee would preclude this type of response.

1.4.2.5.2 CO_2 Production and Respiration

An immediate increase in CO_2 production observed after injury typically occurs due to the decarboxylation of malic acid spilled from damaged cells, rather than increased cellular respiration (Hung 1993). This process is illustrated by the fact that CO_2 generation increases substantially more than oxygen consumption in the hours following bruising (Pollack and Hills 1956). An initial sharp surge in CO_2 immediately following injury is symptomatic of this type of response. In cherry, impact resulted in sharply increased CO_2 production within 3 hours, with higher than normal CO_2 generation persisting for several hours (Wade and Bain 1980). The response timing would obviously vary with the tissue permeability to gas exchange. Rises in CO_2 evolution are proportional to the severity of damage in blueberries and cherries (Burton and Schulte-Pason 1985). This response may relate to the volume of tissue damaged, and hence the amount of malic acid released. The release of CO_2 due to the decarboxylation of malic acid is a commonly observed response, occurring independently of increases in cellular respiration.

A prolonged rise in CO_2 generation suggests an increase in cellular respiration due to injury. In tomatoes, a sharp rise in CO_2 generation was observed in the day following impact, but the CO_2 generation of injured fruit also remained slightly higher than the control for 13 days (MacLeod *et al.* 1976). This prolonged rise in CO_2 evolution suggests a metabolic effect, either through more rapid ripening or senescence, or the activation of defense or healing processes. Increased CO_2 evolution due to wounding is a commonly observed response, but is undetectable in some fruit, such as papaya (Quintana and Paull, 1993). Increased respiration results in the more rapid depletion of sugar and other storage products. The process can result in loss of energy reserves for the fruit, and a decline in sensory quality and nutritional value. Overall quality can be reduced and postharvest life shortened.

1.4.2.5.3 Stimulation of Enzyme Activity

In plant tissue certain enzymatic reactions are prevented by the physical separation of substrate and enzyme. For example, enzymes involved in browning are commonly located in the thylakoids of chloroplasts (Vaughn *et al.* 1988), while phenolic substrates occur in the vacuole (Macheix *et al.* 1990). Cellular disruption can result in membrane breakage, decompartmentalising enzymes and substrates, and allowing reactions such as browning and softening to take place.

Most mechanically damaged tissue will show some form of browning, commonly in the form of a bruise. Browning occurs due to oxidation of phenols into quinones, which are further converted through polymerisation. Levels of both enzymes and phenolic substrates affect the rate of the browning reaction. Browning generally occurs through the actions of enzymes such as PPO or POD, which may be released by cellular disruption, and stimulated by the ethylene response to injury.

Tissue softening is caused both by physical damage to cell structure, and by enzymes that degrade cell walls, such as PG, pectin methylesterase (PME) and cellulase. Cucumber shows substantial tissue softening in response to injury, accompanied by increases in the activity of PME and PG, as well as POD and xylanase (Miller *et al.* 1987). PME and PG act together to degrade pectin substances found in the cell wall and middle lamella. PME has little effect on the texture of the fruit, but is thought to be involved in the partial demethylation of pectin that is necessary before PG can act (Nagar 1994). Cellulase disrupts and loosens the cell wall matrix, and is involved in the softening process during ripening in some fruits, such as avocado (Nagar 1994). Tissue softening enzymes contribute to the increases in electrolyte leakage commonly observed after mechanical damage.

The activity of other enzymes may also be affected by mechanical damage, particularly those involved in lignification or other wound healing processes. For example, wounding can result in increased activity of PAL, as observed in squash (Hyodo *et al*. 1989). PAL catalyses a primary stage in the lignin biosynthesis pathway. In addition to browning, POD is involved in the lignification process. Ketsa and Atantee (1998) found that both the firmness of mangosteen pericarp and POD activity increased rapidly after impact damage. POD may have contributed to the increased firmness of tissue through lignification.

1.4.2.5.4 Wound Healing Processes

Wound healing involves the development of a protective physical barrier near the site of injury, and may reduce moisture loss and pathogen invasion in damaged tissue (Morris *et al.* 1989; Spotts *et al.* 1998). In certain fruits and vegetables, such as oranges (Ismail and Brown 1975) and potatoes (Morris *et al.* 1989), postharvest wound healing is common, but the response has not been noted in lychee.

Compounds involved in wound healing include lignin, callose, wound gums and suberin. Callose is a polysaccharide formed of glucose residues, and appears rapidly in response to mechanical stress. It may impair or eliminate cell function, but provides rigidity and possibly pathogen resistance (Hinch and Clarke 1982). Suberin is waxy, and provides a barrier to moisture loss. Lignin, a complex polymeric molecule, gives cells rigidity, and has been observed in wound healing responses in many crops, such as squash (Hyodo *et al.* 1991). PAL is the key enzyme in lignification and wound healing, catalysing a primary stage in lignin biosynthesis, and has often been used as an index of wound healing (Golomb *et al.* 1984). The role of lignin in wound healing may be less important than previously reported. The histological stain generally used to detect lignin (phloroglucinol/HCl or Pg/HCl) also stains for gum. Specific tests for wound gum show that healing of injuries in citrus involved gum deposition, rather than lignin synthesis (Stange *et al.* 1993b). The role of lignin in citrus wound healing was commonly reported prior to this study (Hung 1993).

In a single wound site, the healing process may involve numerous compounds. For example, the cell walls of mechanically injured pears showed an accumulation of callose, suberin, tannins and pectic substances, as well as gums and starch (Spotts *et al.* 1998). While the process of wound healing can vary greatly between different types of fruit and vegetable, it generally involves the formation of a barrier composed of waxy or corky cells. Rapid cell death in a wounded area is an additional mechanism related to wound healing (Ryan 1984). This hypersensitive reaction to wounding can prevent the growth of pathogens requiring living cells as hosts.

The wound healing process is generally encouraged by high RH and warm temperatures of around 25 to 30°C, as shown in oranges and potatoes (Brown 1973; Morris *et al.* 1989). Curing treatments promote wound healing through storage at high RH and warm temperature. Under curing conditions, wound gums in citrus can be deposited within 9 hours of injury (Stange *et al.* 1993b). High-density polyethylene wraps have also been shown to encourage healing, through an increase in RH (Golomb *et al.* 1984). High RH allows the undamaged cells at the edge of the wound to function normally. Under ambient RH, it was observed that desiccation of uninjured cells adjacent to the wound impaired normal cellular activity, slowing the healing process (Ismail and Brown 1975). The benefits of high temperature in wound healing are likely to relate to the speed of biochemical reactions. In pears, warm storage temperature (28^{o}C) increased the rate of wound healing, but the process occurred even at -1^{o}C (Spotts *et al.* 1998).

Wound healing responses can have significant benefits in protection against pathogens and moisture loss. Wound healing in cured potatoes has been shown to reduce subsequent moisture loss during storage (Schippers 1971). Pear wound healing was shown to protect wounds from infection, with susceptibility to decay decreasing almost linearly over 2 days at 20^{o}C, concurrent with wound healing (Spotts *et al.* 1998). However, in squash the edges of scar tissue are a frequent site of fungal growth (Hawthorne and Sutherland 1991). While the healing process reduces moisture loss, and the chance of fungal growth, the healed wound generally remains weaker than an intact epidermis.

1.4.2.5.5 Chemical Defences Against Pathogens

In addition to the physical barrier erected during wound healing, the plant uses numerous chemical defenses to inhibit fungal penetration. The specific compounds involved vary widely between plant species, but share similar mechanisms. Some of these chemical defenses are present in the plant at all times, while others form only in response to certain stimuli. Chemical defense mechanisms act against infection by pathogens, and will last for a substantial length of time. The response may be systemic, spreading out to protect other parts of the fruit, or localised, occurring only in the area subjected to stress (Ryan 1984).

Pre-formed secondary metabolites are concentrated in the outer layers of plant tissues, and are often compartmentalised in organelles or vacuoles (Prusky and Keen 1995). In general, pre-formed compounds used in defense against fungi may include substances in the cuticle which inhibit spore germination, inhibitors of enzymes such as cutinase and PG, and cutinase specific antibodies (Prusky 1998). These substances may be released in response to a stress event, such as pathogen invasion or mechanical injury.

In addition, some fruits respond to stress stimulus by the formation of antifungal compounds, such as phytoalexins, proteinase inhibitors and lytic enzymes such as chitinases and glucanases (Lyon *et al*. 1995). These substances exist in precursor forms, and are converted to active forms upon wounding or pathogen invasion. Phytoalexins have been observed in numerous fruits and vegetables, including apples, citrus, grapes, capsicums, carrot and potato (Prusky 1998). Oranges and grapefruits were shown to produce a phenolic cinnamaldehyde anti-fungal compound in the peel in response to abrasion against a steel brush (Stange *et al*. 1993a). Chitinase is capable of inhibiting pathogen growth by hydrolysis of fungal cell wall polymers, and is stimulated by wounding in many crops, including grapefruit (Porat *et al.* 1999) and pumpkin (Esaka *et al.* 1993). The combination of these numerous mechanisms of defense enhances the ability of the plant to resist fungal invasion.

The levels of phenolics within a plant commonly increase after an incident of stress, such as mechanical injury, and can reduce the ability of fungi to establish

(Macheix *et al.* 1990). In addition, chlorogenic acid is an intermediate substance partially used in the biosynthesis of lignin and suberin, so it is involved to some extent in both physical and chemical defense systems (Macheix *et al.* 1990).

1.4.2.6 Secondary Effects of Mechanical Damage

The importance of mechanical injury lies not only in the visible damage caused, but also in potentially increased susceptibility to other forms of degradation. Mechanical damage can significantly increase the risk of moisture loss and pathogen invasion. The risk of chilling injury may also increase due to wounding, as shown in citrus (Mulas *et al*. 1996). This response may occur due to more rapid general deterioration, promoted by ethylene synthesis and accelerated moisture loss. Levels of damage can be visually acceptable, but may still cause a significant decline in shelf life and quality due to these secondary factors.

1.4.2.6.1 Mechanical Damage and Moisture Loss

Mechanical injury often damages the barriers to water loss, and can thus increase the rate of moisture loss from produce (Alayunt *et al.* 1998). Impact and abrasion can damage the surface layers of the fruit, such as the cuticle, or disrupt epicuticular waxes, reducing the ability of the fruit to resist moisture loss. Increased moisture loss can occur even when damage is minimal. Superficial scratches in citrus may be barely visible, but can cause a significant increase in moisture loss under certain conditions (Golomb *et al.* 1984). Cuts and punctures can more severely affect moisture loss by breaking through the outer protective layer and exposing underlying tissues. The loss of moisture is a major cause of postharvest deterioration, and also results in the immediate loss of value through reduced weight.

1.4.2.6.2 Mechanical Damage and Pathogen Invasion

An intact cuticle and epidermis act as a significant barrier to fungal growth, and these outer layers of the fruit are the foremost defense against pathogens. When the intact rind of a fruit is compromised by a wound, the ability to defend against pathogens is substantially impaired. Damage to the fruit surface provides an entry point for pathogens into the tissue. In addition, leakage of cell contents from the injury provides a ready supply of moisture, energy and nutrients for pathogen growth. In addition to supplying nutrients for pathogen establishment, the release of cell contents from the fruit tissue can encourage the formation of droplets on the fruit surface (Wills *et al.* 1998). Leaking cell contents have a high solute concentration, and combined with high RH conditions during storage, this can result in the solution attracting water vapour due to osmotic processes. The resultant droplets on the fruit surface encourage the germination and growth of fungi. The ready supply of nutrients and free moisture in a wound promotes rapid mycelial growth and sporulation, increasing the chance of the fungus spreading between fruit.

Lychee fruit have been observed to be susceptible to blue mould rots caused by *Penicillium* spp. (Fitzell and Coates 1995). This type of rot is more common in mechanically damaged fruit, as injured or weakened tissue is generally required for the pathogen to germinate and grow. While a wound is generally required, it may be very small, and invisible to the naked eye. In apples, all that is required for penicillium mould to establish is a microscopic break in the skin, or an injured lenticel (English *et al.* 1946). Burton *et al*. (1987) studied the effect of mechanical damage on penicillium mould establishment in apples, and found that on certain cultivars impacts as minimal as 5 cm significantly increased rot development. It has been suggested that dropping a lychee carton 50 cm results in increased disease susceptibility, but this statement was not accompanied by any data (Johnson 1989).

Rhizopus stolonifer has been identified as an organism causing rots on lychees, and this species also requires a wound for infection to occur (Fitzell and Coates 1995). Wound invading pathogens such as *Rhizopus* frequently show rapid growth, and can quickly spread between fruit, resulting in severe losses during storage. A single infected fruit can have a significant economic impact, as any sign of fungal growth within a carton reduces the appeal and value of the entire carton. The economic importance of mechanical injury lies not only in the damage to an individual fruit, but in its increased susceptibility to pathogen attack, and the risk of an established pathogen spreading during storage.

1.4.3 Factors Affecting Mechanical Damage

There are many different symptoms of mechanical damage observed in fresh produce, including cracking, scuffs and bruising. The type of injury symptom that develops, and the extent of damage, depend both on the characteristics of the load and the properties of the product.

1.4.3.1 The Effects of Load Properties

The manner in which a load is applied to the fruit strongly affects the response of the tissue. Abrasion injury results in substantially different cellular damage to impact, and hence causes the development of different symptoms. Impact, compression and vibration loads can cause similar tissue damage in the form of bruising. However, the manner of application can cause differences in the characteristics of the bruise. For example, bruising in stone fruit can occur in different levels of the tissue depending on the method by which the load is applied. Damage by vibration results in bruising in the outer layers of the skin, while severe impacts can result in largely internal damage, with the fruit stone causing bruise formation (Ruiz Altisent 1991). Vibration damage often causes cell rupture just beneath the fruit skin, resulting in the formation of shallow bruises. Movement of the fruit often results in injuries occurring in a random fashion over the fruit surface.

In general, compression bruises are also typically shallow, with a smaller ratio of depth to diameter than impact bruises (Nelson and Mohsenin 1968). Under compression, the stress, or load per unit of area, applied to the fruit is lower than for the same energy input during impact. However, the strain, or deformation of tissue, is greater under compression (Ruiz Altisent 1991). Higher loading rates, typical of impact, are more likely to result in shear failure patterns in the tissue, observed as a fracture line within the tissue, at a tangent to the direction of the load. In contrast, the slower loading rates of compression show normal stress or strain failure, where damage tends to extend from the contact surface inwards (Ruiz Altisent 1991).

One of the most obvious factors affecting the extent of damage is the magnitude of the load or force acting on the fruit. Numerous factors affect the amount of stress acting on the fruit surface during impact. The most obvious factor affecting impact load is the height of the fall, as this relates directly to the amount of force acting on the fruit. In addition, the initial velocity is important in determining the force acting on the fruit, with higher velocity resulting in greater force. The size of the fruit also directly affects the load magnitude in impacts. Impact energy is directly dependent on the fruit mass, with larger fruit subjected to greater force upon impact.

In compression injury the duration and weight of the load are important factors influencing the severity of damage. The nature of the creep process makes duration particularly important, as tissue deformation can gradually increase over time under a static load (Sitkei 1986).

The magnitude of the load acting on the fruit is important in determining the damage resulting from abrasion. A linear relationship was detected between absorbed energy per unit of area and the extent of damage in the abrasion of watermelon (Puchalski and Brusewitz 1996). Chen and Squire (1970) studied the abrasive damage of oranges and found that factors affecting the amount of damage included the roughness of the sliding surface and distance dragged.

The severity of vibration injury is largely affected by the duration and the magnitude of acceleration, which is largely related to the vibration frequency. The application of vibration frequencies ranging from 2 to 30 Hz revealed that the 7.5 to 10 Hz range caused the most damage to strawberries and grapes (Fischer *et al.* 1992). During transportation, an interaction of the characteristics of truck and road determine the vibration frequency inflicted on the produce. Poor road conditions and fast truck speed enhance vibration, while well designed truck suspension dampens the vibration produced during transportation. Resonance occurs when the vibration produced during transport matches the natural vibration frequency of the product, and can result in greatly increased acceleration. Stacks of fibreboard boxes filled with fruit typically show resonance at approximately 6 Hz (Kawano and Iwamoto 1979), a frequency commonly encountered in road transport (Hilton 1994).

The degree of acceleration reached by the fruit is also influenced by produce conditions, such as position within the container and tightness of fill. Damage generally only occurs in the top layer of fruit within a package, but fast acceleration for long periods may cause damage to occur in up to three layers (O’Brien *et al.* 1969). Tight packing reduces the ability of fruit to move within the container, thus reducing vibration injury. It also raises the natural frequency of the package above that of the truck vibration, reducing the chance of resonance (Schoorl 1974).

Tearing of the tissue, typically occurring as a pulled stem, would also be expected to differ according to the severity and duration of the load. It is likely that a threshold force would be required to initially separate the tissue, and further application of force would increase the size or depth of the tear. The load direction would also be important for this type of injury, as tissue may respond differently to an upward (linear) pulling force, twisting (torsional force) or angled (shear) force, depending on the tissue structure. In lychee, torsional force applied to the stem would generally result in failure at the natural abscission zone of the stem, while a linear pull may result in tearing of the pericarp tissue.

The repeated application of loads may result in tissue failure, despite the fact that each single load is beneath the threshold of damage. This effect was observed in the compression injury of apples. It was theorised that damage did not accumulate, but that each load cycle carried an equivalent risk of tissue failure, which gradually increased the chance of damage with repeated loading (McLaughlin and Pitt 1984). For boxes of apples, a second fall from 12 inches resulted in bruising to 80% of fruit, while a single fall damaged 40% (Schoorl 1972a), but in general, damage increases only slightly with repeated loading (Hyde 1999).

The properties of the load surface, including radius of curvature, size, elasticity and cushioning, strongly influence the load properties. Surfaces of smaller size or radius result in a smaller contact surface, concentrating the force. The elasticity of the load surface is very important in determining the force acting on the fruit. For example, a fruit can be dropped twice as far onto an identical fruit as it can onto a rigid steel plate (Horsfield *et al.* 1972). When two fruit collide, the force of the impact is divided between the two, thus reducing the force acting on each individual fruit. Similarly, cushioning reduces the amount of force acting on the fruit tissue by increasing the contact area. Under impact, cushioning also increases load duration.

1.4.3.2 The Influence of Fruit Properties on Mechanical Injury

1.4.3.2.1 Cultivar Effects

Different fruit and vegetable products vary widely in their susceptibility to various forms of mechanical injury. For example, grapes are susceptible to vibration injury, but can tolerate compression, while the opposite is true of strawberries (Guillou 1964). These differences result from the wide variation in mechanical and biological properties that occur in different fruit and vegetable species.

Significant differences in the amount of damage sustained from a mechanical injury can occur between cultivars and even between individual fruits. Strong cultivar differences were observed in response to repeated impact over a range of cherry cultivars (Burton and Schulte-Pason 1987). It has been observed that the impact of two seemingly identical apples rarely results in equal damage to both, due to inherent variability in mechanical properties between fruit (Pang *et al.* 1992). Fruit characteristics such as tissue structure and biochemistry contribute to this variation. Further complexity is added by the many interacting factors affecting these properties.

1.4.3.2.2 Effect of Fruit Structure

The gross anatomy of the fruit is important in influencing the forces acting on the fruit. Fruit size, weight and shape can affect the amount of force and the contact area of the load. Differences in gross anatomy may also influence the response to mechanical damage. For example, the ratio of seed size to flesh in peaches affects the damage resulting from impact. When the stone is closer to the impact site, greater internal damage results, so fruit with a large flesh to seed ratio tend to be more resilient (Menesatti *et al.* 1999). While gross anatomical features may have some influence, the mechanical properties of fruit tissue are primarily conferred by the tissue structure.

The structural features of the fruit tissue strongly influence the response to mechanical stress. Structure affects both the amount of deformation at a given force, and the amount of deformation required to damage cell membranes. Among the most important structural differences between fruit are the characteristics of the individual cells, such as size, shape and strength, the arrangement of cells and presence of intercellular spaces.

To a large extent, the mechanical properties of the individual cells influence the strength of the tissue. Cell wall strength determines the ability of the cells to resist rupture. Large cell size is thought to generally increase bruising susceptibility (Hudson 1975). Gould *et al.* (1990) showed that in pepino fruits, the cultivar ‘Suma’, with large exocarp cells was more susceptible to damage than ‘S15/14’, a selection with smaller cells. Cell shape may also influence the response to damage. Susceptibility to concentric skin cracking in tomato was lessened in fruit with flattened epidermal cells (Cotner *et al.* 1969).

Tissue strength is influenced by the adhesion of cells and cell permeability. Bonds between cells, including pectin and cellulose-xyloglucan networks, are important in determining mechanical properties of fruit tissue (Vincken *et al.* 2003). Weak intercellular bonds may result in the separation of cells without rupture, by cell debonding (Pitt 1982). Under compression, the movement of fluid from cells into the intercellular space may allow cells to deform without rupture (Pitt 1982). The permeability of the cells may therefore influence the tissue response to compression.

Cell arrangement and geometry have been shown by mathematical simulation to have a significant effect on the type and location of tissue failure (Wenian *et al.* 1991). The volume and contents of intercellular spaces in fruit tissue vary considerably and this greatly affects mechanical properties. Pepino fruit with a greater volume of intercellular space tended to be more susceptible to impact damage (Gould *et al.* 1990). It was hypothesised that air filled spaces reduced the transfer of lateral deformation within the tissue, therefore concentrating the strain into a smaller area. In tightly packed cells, the lateral deformation would be transferred across a larger area, lessening the longitudinal strain applied to the underlying tissues. The transfer of energy through the tissue appears to be more likely in dense tissue, with minimal air-filled spaces. In tissue with low air-space volume, such as peach, impact injury tends to result in deep bruises not visible on the fruit surface. In contrast, fruit with a high volume of air-filled interstitial space, such as apples, tend to show bruising extending from the contact surface inwards. In apples, the force of impact moves inward from the contact surface until all energy is dissipated, either by cell breakage or membrane distension (Hung 1993). In the denser peach tissue, energy is more likely to be transferred through the tissue, resulting in internal damage. The effect of intercellular space on the extent of damage may vary between impact and compression. During compression, it is thought that large intercellular spaces may allow the cells to rearrange without rupture, thus reducing damage (Pitt 1982).

The skin can be particularly important in determining the mechanical properties of the tissue. Performing impact tests on tomatoes and various models, Lichtensteiger *et al.* (1988) found that the skin strongly affects the impact response when the body is a soft internal structure surrounded by a firmer, relatively thin shell. When the internal material is firmer than the skin, no skin effect is observed. The tensile strength of the skin is particularly important in determining fruit susceptibility to cracking. The thickness of the skin can be important in the response to injury. Thick-skinned fruit such as watermelons and unripe bananas tend to be resilient to stress-strain failure, but skin rupture problems can occur with these fruit (Ruiz Altisent 1991).

1.4.3.2.3 Effect of Fruit Biochemistry

Susceptibility to bruising may depend on biochemical factors such as enzyme and substrate levels, which dictate the potential of the tissue to brown. In potatoes, the availability of the substrate tyrosine was highly correlated with the bruising response (Dean *et al.* 1992). In contrast, no correlation was shown between bruising susceptibility and enzyme and substrate levels in apples (Klein 1987). It is likely that other tissue properties can override this effect. Biochemical factors may also have an effect on the extent of injury by influencing the physical properties of the tissue. For example, surface oils and waxes can reduce abrasive damage in oranges (Chen and Squire 1970).

1.4.3.3 Fruit Conditions Affecting Biochemistry and Structure

Fruit and vegetables are dynamic in their biological properties, with changes in composition, moisture content and texture occurring throughout growth, ripening and postharvest storage. Texture itself is affected by many factors during postharvest storage, including moisture content, temperature, nutrient supply and atmospheric composition. In most cases these complex interactions are difficult to define quantitatively. However, some general trends in factors affecting mechanical damage have been observed.

1.4.3.3.1 Maturity and Ripeness

The physical characteristics of both the cell walls and bonding materials vary during the ripening process, substantially altering mechanical properties (Ruiz Altisent 1991). In certain fruits the stage of ripeness or maturity can strongly influence damage sensitivity. This response may vary depending on the types of damage. Increased maturity in peaches reduced resistance to vibration and compression damage, but did not affect impact susceptibility (Vergano *et al.* 1991). In cherries, bruising increased in more mature fruit, but surface pitting, caused by cell fracture, declined (Lidster *et al.* 1980). Susceptibility to friction discolouration in pears tended to decrease with maturity (Mellenthin and Wang 1974).

A strong maturity effect on mechanical damage particularly occurs in fruit showing major textural changes during ripening, such as peaches, pears and tomatoes.

A strong increase in bruising susceptibility is shown in avocado with ripening (Baryeh 2000). Ripening may cause changes in the physical characteristics of the cell walls, and in the materials bonding these walls together. In sweet cherry, increasing resistance to surface pitting with maturity was accompanied by increased calcium in the tissue, possibly conveying greater cell wall strength (Lidster *et al.* 1980). The softening of texture tends to change failure patterns from shear towards normal stress or strain failure (Ruiz Altisent 1991). The stage of ripening can also affect moisture content, which may further influence damage susceptibility (Sitkei 1986). Major textural changes are not observed in lychee with ripening, but it has been suggested that over-mature fruit are susceptible to mechanical damage (Bagshaw *et al.* 1991).

1.4.3.3.2 Moisture Content

Moisture content is one of the most important factors affecting the mechanical properties of materials. Susceptibility to bruising typically declines when fruit are slightly water stressed, as observed in pears, potatoes and apples (Baritelle and Hyde 2001; Klein 1987). This may be explained by the fact that bruising is generally a result of cell rupture. A flaccid cell is able to tolerate greater stress before it ruptures than a turgid cell, due to a reduction in the internal pressure of the cell contents. Thus a flaccid cell will typically deform more under a load, and require greater stress to rupture than a turgid cell. Greater deformation in flaccid cells also results in effective cushioning, by increasing the contact surface of the load. Through these mechanisms, loss of moisture tends to confer greater resistance to bruising. The response would be expected to reverse where damage primarily occurs though disruption of cell contents, as flaccid cells would be more affected.

Tissue failure by skin cracking tends to be more problematic when produce is fully hydrated. Turgid potato tubers are characteristically brittle in texture and are more susceptible to shatter (Thornton *et al.* 1973). An increased risk of cracking may occur in well-hydrated fruit due to the very tight packing of turgid cells. The strong turgor of individual cells would reduce the capacity to deform, resulting in greater risk of cracking failure. In other types of fruit and vegetable, tissue may become brittle with extreme desiccation, leading to greater risk of cracking with moisture loss.

It has been observed by growers that more force is required to remove lychee fruit from the tree later in the day. This tends to increase the risk of pulled stems. It is possible that this effect is due to the influence of moisture content on mechanical properties. Substantial diurnal variation in water potential is observed in lychee (Olesen 2001). A similar response may occur in mandarins, where seasonal differences in the incidence of pulled stems were recorded. Spring harvest resulted in around 9% stem injury, while injury was negligible in the monsoon crop (Sonkar *et al.* 1999). This response may be a similar turgor effect to that observed in lychee, with greater moisture supply in the monsoon season resulting in reduced incidence of pulled stems.

1.4.3.3.3 Fruit Temperature

Temperature can have a large impact on the mechanical properties of a product, and thus on its sensitivity to damage. The effects of temperature on the mechanical properties of fruit and vegetables are complex, and can include changes in turgor pressure and elasticity. Decreasing temperature generally reduces elasticity (Sitkei 1986). Warm fruit tend to show greater deformation, in a simple stress-strain type of failure (Somner *et al.* 1960). In contrast, cold fruit tend to be firmer, and less easily compressed, but more susceptible to brittle failure, such as cracking. These are general responses to temperature occurring in many structural materials (Tetelman and McEvily 1967). In extreme cases, high temperature treatments can cause textural changes and loss of elasticity, with the material becoming brittle. In this case the susceptibility to damage can increase (Sitkei 1986).

In practice, the effects of temperature can be complex and inconsistent. Where a brittle failure occurs, as in potato shatter, more damage tends to occur at cold temperatures (Hyde 1999). Similarly, cold fruit are often more susceptible to impact damage involving cell rupture or fracture, as shown in cherries (Crisosto *et al.* 1993a). Impact damage may be reduced at cool temperatures due to reduced deformation, as in pears (Baritelle and Hyde 2001). Bruising damage due to compression is also generally lessened at low temperatures, due to reduced deformation, as observed in cherries (Patten and Patterson 1985). However, in bananas, increasing temperature results in reduced injury due to compression, but increased impact damage (Banks and Joseph 1991). Reduced compression damage at warmer temperatures may occur due to the lower viscosity of fluids. During compression, movement of water from cells can lessen tissue stress, and this process may be enhanced by reduced viscosity.

Responses to temperature are often more complex than simple increases or decreases in damage. Study of temperature effects on apple have shown conflicting results, with some papers suggesting increased damage in warm fruit (Saltveit 1984), and others the reverse (van Lancker 1979). It appears that temperatures ranging from 0 to 20^{o}C do not cause significantly different levels of damage, while warmer temperatures may cause an increase in damage (Klein 1987). Pears also show an unusual response to temperature, with higher levels of friction discolouration observed in fruit cooler than 10°C, or warmer than 20°C (Amarante *et al.* 2001). The effects of temperature on fruit mechanical properties and physiology can result in complex responses, which may conflict with the broad generalisations proposed.

In addition to influencing the mechanical properties of the tissue, fruit temperature also affects the biochemical response after impact. Temperature strongly affects the speed of biochemical reactions, and therefore influences the rate of processes such as bruising and wound healing. Cherry surface pitting disorder due to impact and apple bruising have been observed to develop more rapidly at warmer temperatures (Lidster and Tung 1980; Saltveit 1984). Similarly, in pears ethylene synthesis can be triggered by abrasion damage when fruit are stored at 20°C, but at 4°C this does not occur (Mencarelli and Botondi 1992). Humidity may also have an effect on injury development, with lower humidity observed to result in more severe injury in bananas (Santana Llado *et al.* 1998).

1.4.3.3.4 Pre-harvest Conditions

Pre-harvest factors, such as climate, soil and general crop management can strongly affect the general resistance of the fruit to damage. Pre-harvest factors can influence the biochemical and structural properties of fresh produce. For example, enzyme levels in potatoes vary substantially between locations, affecting the potential bruising response (Jaensch 1996). Stress history may predispose the cell membrane to break down under restricted conditions, resulting in a higher browning potential (Hung 1993). In general, a crop that is healthy and well managed results in fruit with improved resistance to damage.

Deficiencies in certain nutrients may result in increased susceptibility to damage. In potatoes, potassium deficiency can cause increased blackspot formation (Jaensch 1996). Mechanisms of this effect may relate any of a number of mechanical or biochemical changes due to deficiency, such as cell wall properties, substrate formation, osmotic regulation or cell size. A deficiency in calcium may also result in increased susceptibility to damage. Calcium is associated with pectic substances in the middle lamella, giving greater strength to these structural components of the cell. A lack of calcium may result in decompartmentalisation and cell rupture at lower levels of stress (Hung 1993).

Environmental conditions, such as exposure to sunlight and fruit temperature can vary substantially, even between neighbouring fruits, and can strongly influence postharvest responses (Woolf *et al.* 2000). The sides of apples exposed to the sun showed different compressive properties to the shaded sides (Abbott and Lu 1996). In contrast, pre-harvest sun exposure in apples did not significantly affect mechanical properties under impact loading (Bajema *et al.* 2000). Cherries grown on heavily cropped trees tended to be more susceptible to bruising, possibly due to reduced carbohydrate supply during growth (Spayd *et al.* 1986). Apple bruise susceptibility increased at later harvest dates, but indices of maturity were not correlated with bruising susceptibility (Klein 1987). It was thought that the mechanism behind this effect may have been increased cell turgor due to sugar accumulation.

In lychee, water stress during the early stages of fruit development can substantially affect the structure of the pericarp. Early water stress resulted in a significantly thinner mesocarp and endocarp (by 40% and 30% respectively), and reduced cell size throughout the pericarp (Kumcha 1998). These types of changes in skin structure have the potential to influence the fruit response to mechanical damage.

1.4.4 Prevention of Mechanical Injury

In order to reduce mechanical damage it is necessary to understand the susceptibility of the product, and to isolate the occurrence of mechanical stress in the postharvest chain. The ability of the fruit to tolerate various stresses, such as impact, compression and vibration, influences the optimal design of postharvest operations. Causes of injury can be determined by assessing the postharvest chain for potential mechanical stress events, identifying the source, nature and magnitude of the load. For example, for fruit susceptible to impact, a drop between levels on the packing line may be a particular concern. Identifying the events causing injury can be difficult, particularly when damage is latent. Progressive sampling generally requires prohibitively large volumes of fruit. An instrumented sphere can detect where impact events above a certain threshold occur, but access to this type of equipment is limited. In many cases, impact events can be located simply by observing the movement of the fruit during handling. Sudden stops have the potential to cause impact damage, and may result from dropping or from fruit moving at high velocity into a solid surface. Compression damage usually results from excessive stacking height, while vibration damage generally occurs during road transport, making these events relatively simple to locate.

Steps taken to minimise damage may include improved packaging, changes to packing lines, reduced depth of stacking and the use of cushioning. The estimated commercial cost of the damage needs to be weighed up against the cost of potential measures to reduce damage. In some cases, it is not economically worthwhile to reduce damage, but often inexpensive changes can yield substantial benefits.

1.4.4.1 Prevention of Injury During Harvest

The condition of the fruit at the time of harvest may profoundly affect susceptibility to mechanical injury, due to maturity effects on injury. In some crops, harvest operations can be timed to reduce susceptibility to damage (Sitkei 1986). Harvesting fruit while dry is generally recommended (Wilson *et al.* 1995). Wet fruit are slippery and more easily dropped. Surface wetness is also generally accompanied by turgor, which can increase susceptibility to injury (Baritelle and Hyde 2001).

During harvest, the use of soft canvas picking bags, or the addition of padding to hard containers will minimise damage. The addition of 2-3 cm thick foam rubber liners to plastic and metal harvesting containers significantly reduced damage to tomatoes (Fiore *et al.* 1992). Impact injury during the transfer of fruit from picking bags to bulk bins can be avoided by reducing the height from which fruit are dropped, and by lining bulk bins with padding. Maintenance of bins is crucial, as rough surfaces, protruding nails or splinters are likely to cause damage.

Bulk bin height is limited by the maximum static load tolerated by the fruit. A permissible compressive load can be converted into acceptable container height by mathematical equations (Mohsenin 1986). While shallow boxes minimise compression, they are not optimal for vibration control. The upper layer of fruit suffers the most vibration damage during transportation. In shallow containers, the upper layer makes up a greater percentage of the total contents, and for this reason shallow containers generally give higher levels of damage. For fruit susceptible to vibration injury, the optimal container height is as deep as possible without exceeding the permissible static load.

Mandarins show a similar stem pull injury to that observed in lychee. Harvest methods obviously strongly affect the incidence of this disorder. In mandarins, harvesting using secatuers reduced the number of pulled stems. Hand harvesting resulted in stem damage in up to 9% of the crop (Sonkar *et al.* 1999). The use of secatuers slightly reduced harvesting efficiency, and caused a slight increase in puncture injuries due to the longer stems.

1.4.4.2 Prevention of Damage During Handling

Careful handling of fruit throughout the postharvest chain is crucial to preventing mechanical injury. Good handling practices can be encouraged by training workers to handle fruit carefully, and paying hourly rates rather than by units of fruit processed. Spotts *et al.* (1998) detected wounding in 4.3% of fruit when workers were paid hourly rates, but 13.9% of fruit were damaged when workers were paid by the number of bins harvested. Minimal handling can reduce damage, with field packing recommended for produce very susceptible to injury (Wilson *et al.* 1995). For produce susceptible to puncture, trimmed fingernails and cotton gloves may be necessary to avoid damage (Wilson *et al.* 1995).

1.4.4.3 Avoiding Injury in Packing Lines

The risk of impact damage is the major concern in packing lines. Damage may occur due to drops, or when a rapidly moving fruit hits a solid barrier. The risk of injury is reduced by avoiding major changes in height and direction in the packing line. Operating the packing line near full capacity reduces the risk of injury, as fruit movement is more restricted (Wilson *et al.* 1995). The velocity of fruit on the packing line can also be reduced by decelerator strips or padded rollers. Water dump systems can be used to transfer fruit from bulk bins onto the packing line to avoid damage.

Inexpensive changes to packing lines, such as the addition of padding, can often significantly reduce damage levels (Brown *et al.* 1990). Rubber or foam cushioning can be applied to sharp edges and surfaces where impacts may occur. Soft padding can suffer abrasion from the movement of fruit, quickly wearing away the material and reducing its effectiveness. In some cases thin steel plates may be a preferable alternative, reducing the need for constant replacement of padding (Gan-Mor and Mizrach 1990).

1.4.4.4 Packaging Fruit to Prevent Damage

Selection of packaging is a particularly important issue for fruit susceptible to compression or vibration damage. The acceleration resulting from vibration is influenced by the dynamics of the box itself, and the arrangement of boxes into a stack (Hilton 1994). The depth of fruit within the box affects resonance properties and can thus be used mathematically to reduce the risk of resonant vibration (Bardaie and Hitam 1979). Shallow containers tend to exacerbate vibration damage due to the greater proportion of fruit in the top layer of the box. Friction properties of the container also affect abrasion and vibration type injuries.

To avoid compression injury, box depth cannot exceed the permissible stacking height. The walls of the packaging must be capable of carrying the weight of stacked loads, so that weight is not transferred onto the fruit. Box failure is a major cause of compression injury. The ability of boxes to withstand compression is affected by numerous factors, such as wetness, storage RH, produce characteristics and the inclusion of fittings within the box (Schoorl 1974). Additional stresses such as vibration increase the risk of box collapse, and subsequent compression damage.

Tightness of fill affects both compression and vibration. Fruit packed tightly are subject to compressive forces, while loosely packed fruit will be capable of greater acceleration during vibration. Tight fill packing has been suggested as a method of reducing vibration damage. A mild application of vibration is used to settle the fruit into a tighter fill, to reduce the subsequent movement of fruit during transport. This technique has given mixed results, with pears and plums showing reduced bruising, while peaches and nectarines showed no change (Mohsenin 1986).

Packaging additions can be beneficial, but are often prohibitively expensive. Additions to packaging may include liners, individual wrapping of fruit, cell or tray packs or cushioning inserts to absorb energy. Polyethylene film bags can reduce vibration damage in pears, possibly by restricting movement of the fruit within the box (Slaughter *et al.* 1998). Cell and tray packs similarly reduce damage by restricting movement, and can reduce vibration and compression damage (Lallu *et al*. 1999). Cell or tray packs must closely match fruit shape to be effective. The use of trays in tomato packaging increased impact and compression damage due to a slightly imperfect fit (Fiore *et al.* 1992). O’Brien *et al.* (1969) found that a 1 inch polyurethane pad placed on top of bulk transported peaches dampened in-transit vibration and reduced injury by 15%. However, it was suggested that the increased cost and inconvenience would be prohibitive. The increased costs of protective packaging must be off-set by a reduction in waste or higher prices. This type of cost may be particularly unappealing to the primary producer when latent forms of damage are involved, as any benefits may not appear until fruit is well into the marketing chain.

1.4.4.5 Reducing Damage During Transportation

Transport damage can be minimised by the use of well-designed suspension. Air-ride suspension reduces vibration in the 3-5 Hz range, compared to steel-spring suspension, and can be beneficial for produce sensitive to vibration (Hinsch *et al.* 1993). Stacks of boxes exceeding 1.5 m can be subject to vibration amplification in the 5-10 Hz range, increasing the extent of damage (Hilton 1994). Sliding or tipping of boxes during transport can result in extensive damage. Strapping of boxes in place will avoid shifting during transport.

1.5 Overview

The greatest challenge in lychee postharvest management is retaining the fresh red skin that characterises the fruit. Skin browning due to desiccation is one of the major causes of decline in visual appeal (Snowden 1990), and continues to be a key problem despite extensive research. Mechanical damage can also degrade fruit appearance (Bagshaw *et al.* 1995; Menzel *et al.* 2002), but the importance of this type of damage is yet to be established. Reduced visual appeal can cause a considerable loss of economic value in lychee. Degenerative processes such as increased ethylene synthesis, more rapid senescence and greater susceptibility to pathogens have been observed in fresh produce in response to both desiccation and mechanical injury. These processes can cause further loss of quality. With an increasing focus on high quality in horticultural production, the optimal postharvest management of fresh fruit remains an important goal.

1.6 General Methods and Equipment Design

1.6.1 Fruit Supply

When possible, fruit were sourced directly from farms, but in some cases fruit were purchased from Brisbane Markets. Orchards used for the supply of fruit were located near Sarina, Childers, Yandina, Nambour, Sippy Downs and Byron Bay (Fig. 1.6.1).

Figure 1.6.1 Location of orchards used for the supply of lychee fruit (SE Queensland and Northern NSW, Australia).

Fruit were harvested carefully to ensure minimal mechanical damage prior to treatment, and were selected for freedom from blemishes and uniform maturity. Moisture loss was minimised by harvesting early in the day, and by covering fruit with moist paper towel after harvest. In many cases fruit were harvested directly into water to ensure a high level of hydration. Cultivars used in general experiments included ‘Tai So’, ‘Kwai May Pink’ (‘KMP’) and ‘Wai Chee’.

1.6.2 Fruit Storage Prior to Treatment

Prior to treatment, fruit were stored covered with damp paper towel to ensure minimal moisture loss. If overnight storage was required, buckets were over-wrapped with plastic. For any longer term storage, fruit were stored at cool temperatures (usually 4 to 5^{o}C). In experiments requiring well hydrated fruit, fruit were soaked in water prior to commencement of the experiment. When the transportation of fruit to the laboratory was required, fruit were packed tightly and securely using padding to avoid mechanical damage during transport.

1.6.3 Storage After Treatment

Fruit were generally stored in a single layer in plastic containers (punnets or take-away containers), left open within large sealed plastic crates. At least one container of saturated salt per 3 containers of lychees was placed into the crate to regulate humidity. Alternatively, in some cases, saturated salts were placed into the base of the large container, and samples were suspended above the solution using small plastic stands. Measurement of RH using a small probe suggested that saturated salts were giving a good level of humidity control at published values (Table 1.6.1).

Additionally, sodium nitrate was used in one experiment (Section 5.5), estimated to give 64.8% RH at 22.8^{o}C (Ranganna 1977). In some experiments RH was not controlled, and fruit were protected from air currents using a cardboard box lid. This type of storage resulted in large variation in moisture loss due to edge effects. In addition, in some experiments fruit were stacked within punnets, and this also generated a large variability to moisture loss within samples, despite control of RH using saturated salts.

Table 1.6.1 Relative humidity levels generated by saturated salt solutions.

Salt	RH at 5^{o}C (%)	RH at 20^{o}C (%)	RH at 25^{o}C (%)
Lithium chloride	11.26 +/– 0.47	11.31 +/– 0.31	11.30 +/– 0.27
Magnesium chloride	33.60 +/– 0.28	33.07 +/– 0.18	32.78 +/– 0.16
Magnesium nitrate	58.86 +/– 0.43	54.38 +/– 0.23	52.89 +/– 0.22
Sodium bromide	63.51 +/– 0.72	59.14 +/– 0.44	57.57 +/– 0.40
Sodium chloride	75.65 +/– 0.27	75.47 +/– 0.14	75.29 +/– 0.12
Potassium chloride	87.67 +/– 0.45	85.11 +/– 0.29	84.34 +/– 0.26
Potassium sulphate	98.5 +/– 0.9	97.6 +/– 0.5	97.3 +/– 0.5

(Lide 2000)

1.6.4 Infliction of Injury

1.6.4.1 Injury Location

Where control of location was possible, injuries were applied to the cheek centre. Fruit were numbered above the cheek centre using a permanent waterproof marker, and this mark was used to keep the position of injury, colour readings and scores constant. In the case of vibration injury, the injury site could not be controlled, and thus in many experiments, 7 colour readings were taken on each fruit to give a better representation of the extent of injury. When multiple colour readings were taken, readings were taken of both cheek centres, the sides of the fruit, the base and both shoulders (Fig. 1.6.2).

1.6.4.2 Abrasion

Abrasion damage was inflicted by lightly dragging one area of the fruit for a set distance across a rough surface. Fruit were held firmly to ensure that rolling did not occur, but downward pressure was not applied. The injury was applied in a single flat steady sweep across the abrasive surface. For most experiments a bastard cut 200 mm metal file (‘Performer’) was used, with a 15 cm rub applied. In some experiments other abrasive materials were used, and these are described where appropriate. In experiments based on whole fruit response the injury was applied 10 or 15 times, spread over the fruit surface. In some other experiments, where noted, the injury was applied three times spread over a single cheek to produce a greater injury area.

Figure 1.6.2 The location of sites on the lychee fruit surface.

1.6.4.3 Impact

Impact treatments were generally applied by use of a pendulum impacting device (Fig. 1.6.3) created for research of impact injuries on potatoes by L. Jaensch, S. Tyerman and D. Edyvean (Jaensch 1993), based on the design of Skrobacki *et al*. (1989). The pendulum impacting device consisted of a 50 cm rigid arm attached to a main support strut by a smooth roller bearing. The arm could be suspended at various heights, held in place by an electromagnet to allow a smooth and consistent release. Heights of release (measured as an arc along the course of the pendulum) were 14.1, 18.1, 22.2, 25.3, 28.9, 34.7 and 40.1 cm (levels 1, 1.5, 2, 2.5, 3, 4 and 5, respectively). Fruit were held firmly in place using a cupped metal clamp. Various impacting heads and additional weights could be attached to the pendulum arm to vary the nature of the impact. The two impacting heads used in experiments were a flat head of 19.95 mm diameter, and 49.50 g mass, and a rounded head of the same diameter and 39.02 g mass. Added weights were approximately 26.5 g each.

Figure 1.6.3 Pendulum impacting device, showing pendulum arm (A), clamp (B), impacting head (C), weight attachment (D) and electromagnetic release mechanism (E).

In some early experiments impact injury was applied by dropping fruit onto a smooth flat surface (a thick glass pane), from a measured height, and catching on the first bounce. Generally a single impact was applied to each fruit, but in experiments based on whole fruit response, fruit were subjected to multiple impacts. When using the pendulum to apply multiple impacts, three impacts were applied, one each to the cheek centre, the side of the fruit (90^{o} from the cheek), and the base. In using the free-falling impact technique, fruit were dropped in random position from the same height ten times, and it was assumed that impacts were spread over the surface of the fruit.

1.6.4.3.1 Calculations for Impact Properties

1.6.4.3.1.1 Impact Energy (J)

Measurements recorded by Jaensch (1996), using the same impacting device on potatoes, were used to estimate impact properties. Using data on impact energy levels from Jaensch (1996), it was found that the effect of release height (measured as an arc length along the arm of the pendulum) on the energy of impact fitted the power equation:

$E = 0.0008 \times L^{2.3235}$ ***…Equation 1.6.1***

($R^2 = 0.9991$)

Where, E = energy of impact (J) and L = arc length (mm).

Using estimates of impact energy derived from equation 1.6.1, the velocity of the arm was calculated by the equation:

$E = 0.5 \times M \times Ve^2$ ***…Equation 1.6.2***

Where, E = energy of impact (J), M = mass (kg) and Ve = velocity (ms^{-1})

Mass was calculated as the sum weight of the pendulum arm, including the impacting head and any added weights. The velocity values resulting from equation 1.6.2 were then modeled to allow the prediction of velocity directly from arc length:

$Ve = 0.246 \times L^{1.1171}$ ***…Equation 1.6.3***

($R^2 = 0.9909$)

This allowed the velocity of each impact to be estimated simply by arc length, as pendulum mass does not affect velocity. Air resistance differences between the two heads were assumed to be negligible in the scale of calculations.

Using equation 1.6.2, the energy of impact could then be estimated based on the pendulum arm mass, and the velocity (calculated from arc length). The defined operating conditions (level of impact and attachments) could thus be used to estimate the impact energy.

1.6.4.3.1.2 Contact Surface Area

The contact surface area between the impacting head and the fruit during impact is an important impact property, affecting the level of stress (force per unit of area) applied to the fruit. The contact surface area is particularly important in comparing or collating the effects of impacting heads of different shapes. Calculation of contact surface area is dependent on a detailed knowledge of the mechanical properties of the fruit tissue, particularly Poisson's ratio, a variable that has not been estimated for lychee fruit. However, it is possible to geometrically estimate contact surface area from tissue displacement, a measurement made by Jaensch (1996) on potatoes.

Jaensch (1996) measured tissue displacement in potatoes under 5 levels of impact, using both a flat and round head of the same diameter. While the heads used were of a different diameter to those used in the current experiment, the relative difference between flat and round heads was considered more important than actual values. The displacement of lychee tissue was assumed to be similar to potato tissue for the purpose of these calculations, introducing a potential source of error. It was found that power equations gave a good model of the variation in displacement based on arc length:

$DF = 0.0711 \times L^{1.0991}$ ***…Equation 1.6.4a***

$(R^2 = 0.9954)$

$DR = 0.0861 \times L^{1.1111}$ ***…Equation 1.6.4b***

$(R^2 = 0.9981)$

Where, DF = Displacement (mm), using flat impacting head, DR = Displacement (mm), using round impacting head and L = arc length of pendulum (cm).

For the flat impacting head, the equation used to calculate contact surface area related to the properties of the semi-spherical cap of tissue displaced by the head (Fig. 1.6.4). An average fruit diameter of 40 mm and a circular contact area were assumed.

$r = (h^2 + r_1^2) / 2h$ ***…Equation 1.6.5a***

Where, h = height of the cap, and hence displacement (D), r = the radius of the fruit (assumed to be 20 mm) and r_1 = the radius of the cap, and hence the radius of the contact surface area.

Figure 1.6.4 Properties of the semi-spherical cap of tissue displaced using a round impact head, including height (h), radius of the fruit (r) and radius of the cap (r_1).

Thus by substitution;

$r_1 = (40 \times D - D^2)^{-2}$ ***…Equation 1.6.5b***

The surface area was thus calculated by;

$SA^F = \pi r_1^2$ ***…Equation 1.6.5c***

$SA^F = \pi (40 D - D^2)$ ***…Equation 1.6.5d***

(Reaching a maximum at 312.59 mm^2)

Where, SA^F = Contact surface area (mm^2) using a flat impacting head.

It was assumed that the maximum area of contact would be limited to the face of the impacting head (312.59 mm^2). While the sides of the head may also have been in contact with the tissue during high energy impacts, they were unlikely to exert significant force, being perpendicular to the direction of the force applied.

For the round impacting head, the contact surface was assumed to be in the shape of a spherical cap of the head. The area could thus be calculated by the formula for the surface area of a spherical cap;

$$SA^R = 2 \pi r h$$ ***…Equation 1.6.6a***

Where, SA^R = Contact surface area (mm^2) using a round impacting head, r = radius of the sphere (mm) (radius of the head = 9.975 mm) and h = height of the cap (mm), and hence displacement (D).

Therefore, by substitution of the radius of the head and the value of pi;

$$SA^R = 62.675 \times D$$ ***…Equation 1.6.6b***

1.6.4.3.1.3 Impact Duration

Knowledge of impact duration is useful in comparing different head shapes, as it affects the energy per unit time applied to the tissue. Data from Jaensch (1996) shows that impact duration varies significantly between flat and round heads, but does not appear to vary with impact height. The average impact duration was recorded as approximately 7.2 ms for the flat head and 8.8 ms for the round head.

1.6.4.3.1.4 Estimation of Equivalent Fall Heights

Converting impact energy into an equivalent fall height allows the data to be more easily understood and applied in practical situations.

$E = h \times M \times a$ ***…Equation 1.6.7a***

Where, E = energy of impact (J), h = fall height (m), M = mass (kg) and a = acceleration (ms^{-1}).

Thus, for any given energy of impact, where fruit mass is 25 g and acceleration is 9.8 ms^{-1}:

$h = E / (M \times a)$ ***…Equation 1.6.7b***

$h = E / (0.025 \times 9.8)$ ***…Equation 1.6.7c***

$h = E / 0.245$ ***…Equation 1.6.7d***

The equivalent fall height calculated by this equation would be assumed to be a good estimate for the flat impacting head. However, the round impact head results in a very different application of force, usually concentrated into a smaller area. Using the flat head estimates of fall height and corresponding energy per unit area measurements (kJ/mm^2), a polynomial equation was developed to predict equivalent fall height from energy per unit area.

$h = (-54.397 \times \varepsilon^2) + (210.56 \times \varepsilon) - 30.166$ ***…Equation 1.6.8***

($R^2 = 0.9987$)

Where, ε = energy per unit area (kJ/mm^2) and h = fall height (m).

This equation was used to estimate the equivalent fall height onto a flat surface for fruit injured using the rounded head.

1.6.4.4 Vibration

Vibration treatments were applied by placing fruit into a small cardboard box (Post-pak video cassette sized box) tightly strapped onto a flask shaker. Initially a Chiltern MT19 Auto Vortex Mixer (on setting 4) was used to apply the injury, with the box pressed against the test tube shaker by hand. However, using this method it was observed that the vibration frequency and amplitude varied substantially depending on the pressure applied. A Griffin flask shaker (Griffin 120W, registered design No. 896331 and 896332, Griffin and George Ltd.) and the use of straps resulted in a more consistent vibration treatment (Fig. 1.6.5). The number of fruit within the box varied between experiments, due to differences in fruit size, but generally ranged from 20-24. This level of fill allowed some restricted movement of fruit within the box, to encourage the development of a vibration injury.

Figure 1.6.5 Griffin flask shaker, box and straps used to apply vibration injury.

1.6.4.5 Compression

Compression injury was initially applied using a penetrometer (Mitutoyo Model ID-C1012MB), with a flat head of 6 mm diameter. In experiments where a uniform compression treatment was required, the head was pressed into the tissue until a defined displacement was reached (4 or 6 mm), and this was held for a specified length of time (10 to 30 seconds). In other compression experiments a small plastic cup was attached to the arm of a penetrometer to allow weights to be applied (Fig. 1.6.6). The head was rested on the fruit surface and the penetrometer was reset to zero prior to loading weights onto the fruit. Weights were balanced in the plastic cup, and displacement was measured over time. Later compression loads were applied using a specifically constructed device.

1.6.4.5.1 Compression Device

The equipment was designed to allow the application of loads to numerous fruit (Fig. 1.6.7). Sections of PVC pipe, of approximately 40 mm internal diameter, and around 10 cm length, were cut with a slit of 5 cm length, 1 cm width cut from the base. Pipe sections were attached to a MDF base, arranged to allow a clear view of each slit. Discs of MDF were cut to slide easily within the pipes, and were fitted with string on two sides to allow easy removal. The area of the disc in contact with the fruit was painted to give a smooth hard contact surface. Lead wire was cut into lengths and bound into bundles within cardboard cylinders to be used as weights.

Figure 1.6.6 Penetrometer initially used to apply compressive loads, with cup attachment for weights.

For compression treatment, a lychee was placed into each pipe, and an MDF disc was lowered onto the fruit. The height of the disc from the pipe base was measured to determine the fruit diameter prior to compression. Weights were gently lowered onto the disc, and deformation was measured using the slit. The compression device was stored on the benchtop, and was not moved throughout the course of each experiment, to prevent shifting of the discs and weights.

Figure 1.6.7 Compression device designed to apply replicable compression injuries to lychee fruit.

1.6.5 Measurement of Fruit Characteristics

1.6.5.1 Measurement of Damage

Visible signs of mechanical damage included colour changes, cracking, darkened tips and scuff marks. Colour readings were taken using a CR200 Minolta colour meter

(Osaka, Japan) calibrated prior to use each day (calibration plate No 21331182, L=97.83, a=-0.43, b=1.92). Readings were taken on the CIELAB scale. Hyperterminal was used to capture the data directly into the computer. Crack area was estimated by multiplying width by length, and a score was given for crack penetration or severity. Cracks that did not penetrate the full thickness of the pericarp were termed closed cracks, and given a score of 1. Closed cracks with a slight weeping of aril juice at one end were given a score of 1.5. Cracks clearly penetrating the pericarp, resulting in leakage of aril juice, were termed open cracks, and were given a score of 2. In some cases, severe open cracks were wide enough for the aril to be visible, and these were given a score of 3. Darkened tips were often scored for severity (1 = very mild, 2 = mild, 3 = moderate, 4 = moderate to severe, 5 = severe), with both the area and darkness of the discolouration affecting severity score. Scuff marks, rots and damaged areas were measured by two perpendicular diameters. Area was calculated using the formula for an ellipse;

$$SA = \pi \times r^1 \times r^2$$ ***…Equation 1.6.9***

Where, SA = area, and r^1 and r^2 = two perpendicular radii.

Measurements of mechanical damage were generally taken immediately before and after damage, and then after various periods of storage.

Desiccation was generally measured by the loss of weight and resultant changes in colour. Changes due to desiccation were generally measured as a time series. Weight was measured in grams to four decimal places using Sartorius A-120-S scales (Gottingen, Germany). Loss of moisture was calculated as the percentage of weight lost compared to the initial measurement. Changes in weight due to respiration were assumed to be negligible relative to moisture loss, as most experiments were carried out at warm temperatures with low RH. Colour readings were taken as described for mechanical damage. Small ink marks or numbers on the fruit were used to keep the position of the colour readings constant. Browning scores were used in some experiments, and were generally given to each cheek. Scores were based on the percentage of the fruit surface estimated to be brown, from 1 (0-10% brown) to 10 (90-100% brown). Estimates of the percentage of the surface area affected by other blemishes, such as yellowing or tip darkening, were also used in some experiments.

1.6.5.2 Interpretation of Colour Readings

Colour readings were taken in the CIELAB (L, a, b) scale and were converted to hue angle and chroma according to the formulae described by McGuire (1992);

$$C = (a^2 + b^2)^{1/2} \qquad \textbf{...Equation 1.6.10a}$$

$$h = ((\text{arctangent } (b/a)) / 6.2832) \times 360 \qquad \textbf{...Equation 1.6.10b}$$

Where, C = chroma, a = CIELAB a-value, b = CIELAB b-value and h = hue angle. The L-value is used unmanipulated as an indication of darkness, where 0 = black and 100 = white. Hue angle defines the colour of the fruit, where 0 = red-purple and 90 = yellow, and chroma gives an indication of the intensity of colour, with higher values more saturated. Thus a reduced L-value shows darkening, an increased hue value shows yellowing and decreased chroma indicates a duller colour.

1.6.5.3 Brix: Acid Ratio

Sampling size varied between experiments, but often the juice of 5 fruit was used for a single bulk measurement. A metal garlic crusher was used to extract juice from aril or pericarp tissue. Total soluble solids were measured using a refractometer calibrated at 25^{o}C, with readings corrected for temperature according to Ranganna (1977). A 10 ml sample was generally used for titration, and this was mixed with 10 ml of RO or distilled water. Samples were titrated against 1M NaOH to pH 8.2, using a pH meter. The pH meter was callibrated daily using standard buffers of pH 4 and pH 7, and included a temperature correction feature. The probe was rinsed thoroughly with distilled or RO water between each use. The volume of 1M NaOH used in titration was multiplied by 0.064 to convert the result into ml citric acid / 100 ml.

1.6.5.4 Water Potential

Water potential was measured using a pressure bomb (PMS Instrument Co., Corvallis, Oregon). Nitrogen gas was used to compress the fruit until liquid was observed bubbling from the stem. Readings were taken in atmospheres and converted to MPa.

1.6.6 Analyses

Statistical analysis was carried out using either SigmaStat 2.0 or SigmaStat 2.03, using a level of significance of $p<0.05$. For multiple comparison of data satisfying normality and equal variance, analysis was carried out by one-way ANOVA, and comparison by Tukey test. The comparison of two samples was carried out by t-test when data were normal and of equal variance. Parametric tests were used where data could not be transformed to normality. For multiple samples ANOVA on ranks and comparison by Dunn’s method were used on non-normal data, while for comparison of two samples, the Mann-Whitney rank sum test was used. In some cases, multiple comparisons were made by numerous t-tests or Mann Whitney rank sum tests. Paired t-tests (when normal and equal variance were satisfied) or signed rank tests (when transformation to normality was not possible) were used when appropriate (for example, in comparing readings taken on the same fruit before and after injury). Correlations were tested by Pearson product moment correlation when data were normal, and by Spearman rank order correlation for non-normal data.

1.6.7 Thesis Structure

Due to the large number of experiments carried out, the thesis is presented in the format of numerous separate experiments within each chapter. Chapter 2 aims to define the lychee response to postharvest mechanical injury, including visual symptoms and physiological changes. Chapter 3 studies the effect of load properties, such as magnitude, duration and repetition, on fruit condition. In Chapter 4, the effect of fruit characteristics on mechanical injury are examined, including hydration, surface wetness, temperature and cultivar. Chapter 5 explores lychee postharvest moisture loss, studying the effects of cultivar, maturity and air currents on moisture loss and subsequent browning. Due to the large number of experiments within each chapter, the methods precede the results and discussion for each individual experiment.

CHAPTER 2. LYCHEE RESPONSE TO POSTHARVEST MECHANICAL DAMAGE

Common symptoms of mechanical damage include tissue discolouration, changes in texture and sensory qualities, cracking, splitting, shape distortion and the development of scuff marks or corky deposits. Descriptions of postharvest mechanical damage to lychee fruit are rare, with no previous published research located.

Symptoms of damage in lychee are yet to be defined, but may include bruising, skin cracking, and browning. Bruising is a common symptom of injury, typically involving darkening and softening of the tissue. This type of symptom has been reported in lychee (Bagshaw *et al.* 1991), but is not described in any detail. Lychee fruit are generally reported to be fairly resistant to bruising (Greer and Smith 1991). Pericarp cracking has been observed in the development of a hot water brushing system (Lichter 2000), but this injury did not appear to result from mechanical forces. Postharvest cracking is not generally noted as a problem in lychee. Pre-harvest abrasion in lychee causes scattered dark blemishes on the fruit protuberances, and may be involved in silvering (Bagshaw *et al.* 1995). Abrasion may cause the skin browning observed by Menzel *et al.* (2002), using incorrectly adjusted destalkers.

Changes in fruit in response to mechanical damage can include gross visual symptoms, ultrastructural changes, increased enzyme activity and subsequent biochemical changes, stimulation of ethylene and CO_2 release, loss of membrane integrity, wound healing and defense mechanisms, increased moisture loss and greater rot development. Clarification of the symptoms and secondary effects of damage will assist in diagnosing problems and determining whether mechanical injury is of economic importance in lychee.

2.1 Symptoms of Impact Damage in Lychee

Bruising is the most common symptom of impact damage in fruit, and is usually assessed as an index of the extent of injury. However, bruising is not generally recognised as a problem in lychee. Greer and Smith (1991) state that lychee fruit are not easily damaged or bruised. The bruising of lychee fruit is mentioned in an extension report by Bagshaw *et al.* (1991). It is suggested that over-mature or large fruit may be more susceptible, but the nature of the injury is not described in any detail. It is not specified whether discolouration extended into the aril. No other references to impact damage in lychee have been located in the literature. While bruising may occur under specific conditions, it does not appear to be a common occurrence in lychee fruit.

Cracking is another possible symptom of impact injury. Cracks can range from microscopic splits in superficial tissues, to large gaping wounds extending into the tissues underlying the skin. Splitting is an even more severe injury, with the fruit divided into several parts after injury. Cracks and splitting would have a severe impact on marketability. Lychee splitting is commonly mentioned in the literature, but is a pre-harvest disorder, unrelated to mechanical injury. Cracking due to impact injury in lychee is not mentioned. Lichter *et al.* (2000) observed lychee cracking during hot water brushing, but the cause of the injury was not confirmed. Due to the lack of any published research on lychee impact damage, a basic study was required to define the symptoms resulting from injury.

2.1.1 Methods and Materials

‘KMP’ fruit grown in Childers were harvested carefully to ensure minimal mechanical damage prior to treatment. Impact damage was inflicted by dropping the fruit from a range of heights onto a smooth hard surface. Fruit were caught on the rebound after impact. In some cases, talc was sprinkled on the impact surface to ensure the injury site on the fruit could be clearly located. The area of damage was marked with a small dot above and below, and the talc was then washed off with water. When talc was used, the control fruit were also rinsed in water to ensure uniformity. Three randomised control trials were carried out to explore the basic effects of impact injury on the fruit.

2.1.1.1 Experiment 1

Replicate groups of 20 fruit were treated by various levels of impact including a control (0 cm), 10 cm, 50 cm and 100 cm falls. Fruit were stored at room temperature, on small plastic lids with the injured and control sites open to the air. Fruit were stored inside sealed cardboard boxes to reduce air current effects, but RH was not controlled. Damage was assessed and recorded over the three days following injury. The length and extent of penetration of cracks were noted. The number of darkened protuberance tips on each side of the fruit were counted, and the injury area (area in which distinct tip darkening occurred) was estimated by two perpendicular diameters.

2.1.1.2 Experiment 2

Replicate groups of 15 fruit were treated either by a single 50 cm impact, or as control fruit. Fruit were stored in punnets at 75% RH and RT, with saturated salts used to regulate RH. Aril appearance was checked 11 days after treatment.

2.1.1.3 Experiment 3

Ten replicate fruit were treated either as controls, or by multiple impacts, with ten 50 cm impacts applied to random sites on the fruit. Fruit were stored in open punnets at

75% RH and RT. Skin colour and browning score at the point of impact and on the opposite side of the fruit, and fruit weight were measured regularly. Measurements continued until all fruit were brown. The number of protuberances affected by yellowing (>1/3 yellow) and tip darkening were recorded before treatment and 2 days after treatment.

2.1.2 Results and Discussion

2.1.2.1 Pericarp Cracking (Experiment 1)

Cracking of the pericarp was observed in fruit treated by 100 cm impact, with 90% of fruit affected. Impacts from 10 and 50 cm did not result in any cracks, suggesting a threshold between 100 and 50 cm. Most fruit treated by 100 cm impact showed more than one crack, with 35% showing 2 cracks and 40% affected by 3 or more. Most cracks affected only the outer layers of pericarp, with the endocarp remaining intact (Fig. 2.1.1). In 20% of fruit, cracks penetrated through the endocarp, and the aril was exposed. The average length of these open cracks was 2.2 cm, while closed cracks averaged 1 cm. Cracking was not correlated with fruit weight.

2.1.2.2 Tip Darkening (Experiment 1)

Impact injuries were observed to cause darkening of the protuberance tips at the impact site (Fig. 2.1.1). This symptom was generally not obvious, and did not detract from the overall visual appeal of the fruit. Impacts as minimal as 10 cm caused a significant increase in the number of darkened tips (Mann-Whitney rank sum test, $p<0.001$), with around 10 more damaged tips than the control (Fig. 2.1.2). The 50 cm impact caused damage to a greater number of tips than the 10 cm fall (t-test, $p=0.036$), but the 100 cm impact did not cause any further significant increase. A similar

Figure 2.1.1 Symptoms of impact damage on lychee pericarp, including tip darkening (A), open cracking (B) and closed pericarp cracking (C).

trend was shown in injury area (Fig. 2.1.3), with 100 cm and 50 cm impacts resulting in significantly greater damage than the 10 cm impact (ANOVA, $p<0.001$). The area of injury was positively correlated with impact height (Spearman correlation co-efficient=0.512, $p<0.0005$).

2.1.2.3 Effect of Impact on Fruit Colour (Experiment 3)

Colour readings revealed that multiple 50 cm impacts caused significant darkening (t-test, $p=0.021$ at 99 hours) and change in hue towards yellow (t-test, $p=0.010$ at 99 hours) (Fig. 2.1.4). Impact injured fruit showed strong darkening that persisted throughout storage. A strong increase in hue was shown after injury, indicating yellowing of colour. Later in storage, a shift towards red-purple in impact-treated fruit resulted in similar hue in control and injured fruit. Impact injured fruit tended to show a reduced loss of chroma from day 2 to day 4, but otherwise followed the same pattern of chroma change as control fruit.

It is likely that biochemical changes in the fruit result in these changes in colour. Darkening and yellowing of colour due to impact may result from enzymatic browning processes similar to bruising. The late shift towards a red-purple colour could result from injury-induced anthocyanin synthesis. Impact has been observed to trigger an increase in anthocyanin production in apples, which is thought to relate to a ripening response due to wound-related ethylene release (Chalmers and Faragher 1977). Increases in anthocyanin levels of greater than 20% have been observed in lychee during postharvest storage (Lee and Wicker 1991b), illustrating a potential for postharvest synthesis. Impact may have the capacity to stimulate postharvest anthocyanin synthesis in lychee.

2.1.2.4 Effect of Impact on the Aril (Experiment 2)

The arils of injured fruit, examined 11 days after impact, showed no signs of damage due to a single 50 cm fall. No discolouration or change in texture in the aril was observed in injured fruit compared to the controls. Brown or yellowish discolouration on the internal surface of the pericarp was occasionally observed. A bruise is defined by Ruiz Altisent (1991) as “an altered volume of fruit tissues below the skin that is discoloured and softened”. Bruising is characterised by changes in colour, texture and flavour in the pulp, and these symptoms were not observed in lychee. The symptoms shown by lychee fruit in response to impact therefore cannot be described as bruising.

2.1.2.5 Effect of Multiple Impact on Moisture Loss and Browning Score (Experiment 3)

A significant difference in the pattern of moisture loss over time was shown due to impact (Fig. 2.1.5). Impact injured fruit showed more rapid loss of weight than control fruit from day 2 to day 4. This resulted in significantly different levels of moisture loss on day 4 and day 6 (t-tests, p=0.028 and 0.019, respectively). Total moisture loss was consistently 20-30% higher in impact injured fruit in the 6 days following injury. The rate of loss late in storage declined in injured fruit, resulting in similar total loss in impact and control fruit after 9 days. Increased moisture loss is a common response to mechanical injury of fruit, resulting from damage to the barriers to water loss (Alayunt *et al.* 1998). An increase in moisture loss of 20-30% would reduce the value of produce and may significantly affect shelf life.

Desiccation browning scores in impact injured fruit tended to increase concurrent with moisture loss (Fig. 2.1.6). Impact injured fruit did not show a significant increase in skin browning.

2.1.3 Conclusions

Impact damage caused significant changes in the appearance of lychee fruit, including cracking, protuberance tip darkening and changes in colour. The threshold for cracking was between 50 and 100 cm. Cracks could develop either in the upper levels of the pericarp (closed) or penetrate through to the aril (open). Where open cracks did not occur, the aril appeared to be undamaged by impact. The composite structure of lychee appears to avert aril damage, with the pericarp bearing the full brunt of the impact. The pericarp showed impact damage in the form of darkened protuberance tips, with drops as minimal as 10 cm causing tip darkening. This type of damage was not obvious, and would be unlikely to affect the visual appeal of the fruit to the consumer. Impact damage also tended to cause general darkening and yellowing of colour in the fruit. Mechanical damage may increase the rates of moisture loss and pathogen invasion in lychee, but further research is required to clarify these hypotheses. Impact has the capacity to significantly degrade lychee quality, with impacts as minor as 10 cm causing visible damage.

2.2 Symptoms of Compression Damage in Lychee

Symptoms observed due to compression of fruit and vegetables include bruising and skin cracking. Distortion of shape can also occur in more plastic commodities such as mandarin. Batten and Loebel (1984) recommend a maximum stacking depth of 30 cm for lychee to avoid damage, but the symptoms of damage are not mentioned. It has been suggested that lychee are susceptible to puncture due to long stems (Lindsay and Cull 1986), which may be exacerbated by compressive loading. Research was carried out to define and quantify the symptoms of compression injury in lychee.

2.2.1 Methods and Materials

2.2.1.1 Colour Changes Due to Compression

Fruit of cultivar ‘KMP’ were harvested in Yandina and were soaked in water for at least one hour immediately after harvest. Fruit were stored overnight at 25^{o}C in moist paper towels. In a randomised control trial, fifteen fruit were subjected to compression, and another 15 acted as controls. Compression was applied using a penetrometer with a contact surface of approximately 6 mm diameter, a displacement of 6 mm depth was held for 10 seconds. All fruit were stored at 25^{o}C and 75% RH after treatment. Colour readings were taken at the cheek centre immediately before and after treatment, and at 24 and 48 hours.

2.2.1.2 Puncture Damage Due to Compression

‘KMP’ fruit were harvested from Sippy Downs, and were stored at 20^{o}C in damp paper towel for 3 days prior to treatment. Fruit were subjected to 1 kg compression while resting on the upright stem of another fruit, within the compression device previously described (Section 1.6.4.5.1). Tissues were used to attempt to hold the fruit in place. Eight fruit were assessed for damage after 2, 4 and 6 hours. Many fruit had shifted after weight was applied, resulting in only 2-3 replicates at each time stage.

2.2.1.3 Distortion of Fruit Shape

Fruit distortion was observed in later experiments, which are referred to in the text. In these trials, a 1 kg load was applied for 2 hours using a compression device.

2.2.2 Results and Discussion

2.2.2.1 Colour Changes and Tip Darkening

Protuberance tip darkening was the major symptom observed in response to compression (Fig. 2.2.1). The damage detracted little from the overall visual appeal of the fruit. The injury was dark in colour, and was very similar in appearance to mild impact tip darkening. The aril appeared undamaged, with no discolouration.

Figure 2.2.1. Tip darkening of lychee subjected to 1 kg compression applied for 2 hours (A,B) and control fruit (C).

Compression caused rapid darkening of skin colour, with a significant effect detected immediately after injury (Mann-Whitney rank sum test $p<0.001$) (Fig. 2.2.2). Strong darkening of colour was observed in the 4 hours following injury, after which the rate of change slowed. Injured fruit were significantly darker in colour throughout storage (rank sum tests and t-tests, $p<0.003$). Changes in chroma followed a very similar pattern, with injured fruit showing rapid loss of colour saturation in the first 4 hours. The immediate change was not significant (rank sum test $p=0.074$), but from 4 to 48 hours, compression injured fruit showed significantly duller colour than controls (rank sum tests, p=0.002, 0.038 and 0.020 at 4, 24 and 48 hours, respectively). Rapid change in hue also occurred in the first 4 hours. Compression injured fruit showed a slight shift towards red-purple immediately after injury, followed by yellowing of colour. A significant effect was detected only after 48 hours (rank sum test p=0.034).

2.2.2.2 Puncture Under Compression

Compression against a fruit stem tended to result in a depression and slight purplish discolouration (Fig. 2.2.3). Puncture of the skin was observed in one fruit after 4 hours, with aril juice pooling in the wound. Neither of the fruit checked at 8 hours showed puncture, but depression in the skin and accompanying discolouration seemed more severe than at 2 and 4 hours. The puncture observed at 4 hours showed that 1 kg compression is sufficient to cause puncture in some fruit. Fruit with any room to move shifted before puncture occurred. A linear alignment of stem, fruit and load was required to cause damage. When the stem was pushed against the fruit above at a slightly oblique angle, less or no damage resulted. The results confirm that puncture can occur in lychee (Lindsay and Cull 1986), but significant loads are required to cause the injury.

Figure 2.2.3. Lychee puncture damage immediately after drying (A), and while weeping (B), and discolouration and indentation caused by compression against a stem (C).

2.2.2.3 Distortion of Fruit Shape and Skin Cracking

Fruit distortion was observed as a symptom of compression injury in certain cultivars, particularly ‘Salathiel’. The distorted and control fruit illustrated in Figure 2.2.4 were from an experiment on cultivar effects (Section 4.10). Cracking of the fruit skin was also observed under extreme loads, but was not common.

Figure 2.2.4. Mild (B) and severe (C) lychee shape distortion due to 1 kg load for 2 hours, and an undamaged control fruit (A). A closed crack is also visible (B).

2.2.3 Conclusions

Tip darkening was the major symptom of compression injury, and was very similar to impact tip darkening. Substantial changes in colour, including darkening, yellowing and loss of chroma were detected due to compression. Most colour change occurred within 4 hours of injury. Puncture of the skin was possible when pressed against a stem under compressive load. Under severe loads, cracking and distortion of shape occurred in some cultivars.

2.3 Symptoms of Abrasion and Vibration Damage in Lychee

The symptoms of pre-harvest abrasion have been described as scattered dark blemishes on the fruit protuberances, thought to develop from wind rub in the late stages of fruit development (Bagshaw *et al.* 1995). However, little mention is made of postharvest abrasion or vibration damage in lychee. Menzel *et al.* (2002) suggested that a browning injury may be inflicted by mechanical destalking, with the surface tissue of the fruit removed. The following experimental work was undertaken to quantify and further describe the symptoms resulting from postharvest vibration and abrasion damage of lychee.

2.3.1 Methods and Materials

Fruit of cultivar ‘KMP’ were carefully harvested at Childers and were transported to Brisbane. In a randomised control trial, treatments were applied to 10 fruit each:

- Abrasion was applied by fifteen 20 cm abrasions on fine grade (280) sandpaper, spread over the fruit surface;

- Vibration fruit were treated for 2 minutes, within a cardboard box held lightly against a test tube shaker (Chiltern MT19 Auto Vortex Mixer, on setting 4), with fruit re-arranged every 30 seconds;
- Control fruit were not injured.

Fruit were stored in open punnets at 75% RH and RT. Fruit weight, skin colour and browning score on each cheek of the fruit were recorded periodically during storage. Measurements continued until all fruit were brown. The number of protuberances on each fruit affected by yellowing (>1/3 of the SA yellow-brown in colour) and tip discolouration were recorded before treatment and 2 days after treatment. Arils were assessed 9 days after treatment.

2.3.2 Results and Discussion

2.3.2.1 Vibration Damage Symptoms

Injury by vibration substantially degraded the visual appeal of the fruit. Vibration damage symptoms included scattered tip discolouration and concise light brown or yellowish scuff marks (Fig. 2.3.1). Scuff marks occurred when fruit were incapable of shifting during vibration, with intense rubbing occurring in a single location. Injury by vibration tended to result in an overall dusty appearance to the fruit. Aril quality was not obviously affected by vibration injury, with no signs of discolouration or decline in texture, and the interior surface of the pericarp was unblemished.

Figure 2.3.1. Typical symptoms of abrasion (B), vibration (C) and scuff (D) damage to the lychee pericarp, and an undamaged control (A).

Vibration injured fruit showed a strong shift in hue towards yellow and significant darkening (t-test $p<0.05$) compared to the control (Fig. 2.3.2). Loss of chroma was accelerated by vibration damage, resulting in significantly duller colour after 99 hours (t-test $p=0.011$). Vibration injury resulted in a large increase in yellowed protuberances and a significant increase in the number of discoloured tips (Fig. 2.3.3). Tips injured by vibration were medium brown, rather than the almost black colour typical of impact.

Observed under a dissecting microscope, vibration injured fruit showed a superficial yellow crust on the pericarp surface (Fig. 2.3.4). The crust appeared brittle in texture, similar in appearance to confectionery honeycomb. This material could be scraped away, revealing red-coloured tissue, but the crust quickly reformed. Similarly, wetting of the fruit appeared to restore redness, but the yellow crust reappeared as fruit dried (Fig. 2.3.5). It is hypothesised that the crust may form due to the leakage and subsequent drying of cell contents. The crust often occurred as a coating on the protuberance tip, or in concise pieces that appeared to have broken away from the tip, a pattern that suggested leakage of fluid from the tip may be the major source. The same type of yellow crust was observed to form around microcracks, supporting the hypothesis that the crust forms from the contents of ruptured cells.

Figure 2.3.4 Yellow crust on the lychee pericarp surface resulting from abrasion (A) and vibration (B) damage.

Figure 2.3.5 Lychee pericarp injured by vibration while wet; original dry injury appearance (A), the same area while wet (B) and after re-drying (C).

2.3.2.2 Abrasion Damage Symptoms

A single abrasion injury against sandpaper was observed to result in injury to an average of 4 to 5 protuberance tips. The injury typically appeared as a worn back tip and highly localised light brown discolouration, and had little effect on the overall appearance of the fruit (Fig. 2.3.1). Pre-harvest abrasion injury is described as scattered dark blemishes (Bagshaw *et al.* 1995), in contrast to the light colour observed in this experiment. The colour difference may relate to the properties of the abrasive surface or type of movement involved. Repeated abrasion injury resulted in yellowing of colour, with significant effects detected at 49 and 99 hours (t-test p-values 0.016 and 0.038, respectively) (Fig. 2.3.2). Abrasion did not significantly affect darkening or loss of chroma. A greater number of discoloured tips were observed in abraded fruit compared to controls, but yellowed protuberances did not increase (Fig. 2.3.3). No changes in the aril, or the interior surface of the pericarp were observed due to injury.

2.3.2.3 Effect of Vibration and Abrasion on Browning and Weight Loss

Increased moisture loss is a common effect of mechanical damage in fruit, but was not detected in this experiment (Fig. 2.3.6). The high variation between fruits resulted in low power of analysis, hindering the detection of any significant differences. Larger sample sizes are required to confirm whether abrasion or vibration significantly affect lychee moisture loss. A significant increase in browning score compared to the control was shown in both abrasion and vibration injured fruit (Fig. 2.3.7) (t-tests $p<0.05$). Given the very small differences in moisture loss, this effect probably resulted from the direct effect of injury on colour, rather than by desiccation browning.

2.3.3 Conclusions

Vibration and abrasion damage can significantly degrade lychee appearance, but appear to cause a primarily cosmetic disorder. Lychee fruit subjected to intense vibration for as little as two minutes showed strong yellowing, which was thought to occur due to the leakage of cell contents onto the fruit surface. Significant darkening of colour and loss of chroma also occurred due to vibration. Abrasion caused a less severe injury, with light brown tip discolouration and significant yellowing observed.

2.4 Properties of the Yellow Residue Resulting from Vibration Injury

It was hypothesised that the yellow residue observed on the fruit surface after vibration damage may result from the drying and crystallisation of cell contents. The dissolved residue was compared to diluted pericarp juice to determine whether these solutions showed similar properties.

2.4.1 Methods and Materials

‘Tai So’ fruit were purchased from the Brisbane Markets, Rocklea, and were subjected to vibration for 10 minutes at setting 4 using the Griffith flask shaker. The resulting yellow residue was gently scraped away from the fruit surface using a scalpel. Residue from around 20 fruit was weighed, added to 5 ml RO water, and gently heated with stirring. The solution was filtered and the volume made up to 10 ml. Residue remaining in the filter paper was weighed after drying to allow accurate calculation of the total weight dissolved. A total of 10.3 mg of residue was dissolved. A similar, but more concentrated solution was created by soaking 20 injured fruit (for around 1 minute each, by turn) in 50 ml RO water over gentle heat and stirring. The solution was filtered to remove undissolved residue. For comparison, a solution was created from 1 ml juice squeezed from the pericarp, made up to 10ml. In an observational trial, the three solutions (and a control of RO water) were compared by measuring pH, electrical conductivity, absorbance spectra (400 to 700 nm, using a Carey 1-E UV-visible spectrophotometer manufactured by Varian, Victoria, Australia) and TSS (Brix^0).

2.4.2 Results and Discussion

The scraped and soaked solutions of residue shared similar properties to diluted pericarp juice. All contained some soluble solids, as detected by a refractometer and were slightly acidic (Table 2.4.1). The pericarp juice, diluted to 1 in 10, was more concentrated than either of the residue solutions, resulting in higher TSS, conductivity and absorbance. Pericarp juice weighed before and after air drying showed around 6.2% dry matter. The 1 ml of pericarp juice added would therefore have contained around 62 mg dry matter, 6 times more than the scraped residue (10.3 mg). TSS levels were 6 times higher in the pericarp juice solution than in the scraped residue solution, supporting the hypothesis of residue resulting from dried spilled cell contents.

Table 2.4.1 Properties of the lychee pericarp exudate solutions tested.

Parameter	RO Water	Scraped residue solution	Soaked residue solution	Pericarp juice solution
DM (%)	0	0.103	0.206*	0.622
TSS (Brix^{o})	0.0	0.1	0.2	0.6
pH	6.05	5.62	5.23	4.51
Conductivity (mV)	37.5	69	89	131
Absorbance at 530 nm	0.00	0.26	0.68	1.79 (1:4) 1.12 (1:8)

* indicates that the value was estimated, using TSS data.

The TSS of soaked residue from 20 fruit was twice that of the scraped solution. The amount of residue removed by this process is therefore estimated at 20.6 mg per 10 ml, totaling 103 mg from 20 fruit. This equates to around 5 mg of DM residue per fruit, or 0.086 ml of cell contents. Using the cross-sectional epicarp cell area of 600 μm^2 measured by Kumcha *et al.* (1996), and assuming 30 cm^2 fruit SA, this equates to the cell contents of 1-2 layers of cells, over the whole surface of the fruit. The yellow residue remained visible after soaking, suggesting that either not all residue was removed by this process, or that the process recurred after soaking (i.e. fresh cell contents oozed out of wounded tissue and dried on the skin surface). The amount of residue on the surface of the each fruit after severe vibration damage averaged at least 5 mg, but could be substantially higher.

The measured TSS showed a strong linear relationship to the DM content of each solution (R^2=1.00) (Fig. 2.4.1). Conductivity increased with concentration in a roughly linear relationship (R^2=0.93). When converted by natural logarithm, pH also showed a roughly linear relationship with DM content (R^2=0.97). Compared with the scraped and soaked solutions, the pericarp juice sample showed slightly higher pH and lower conductivity relative to concentration. These differences may have resulted from variation between layers of the pericarp. The residue would presumably be derived only from cells in the outermost layers, while the pericarp juice sample was from the entire thickness of the skin.

The pericarp juice solution showed substantially higher absorbance than soaked and scraped solutions. Even when the solution was further diluted 1:8 (equivalent to 7.75 mg DM per 10 ml), absorbance from 400 to 700 nm remained substantially higher than the more concentrated soaked and scraped solutions. Soaked and scraped solutions also showed a slightly more curved shape to the absorbance spectra than diluted pericarp juice over the 400 to 700 nm range of measurement (Fig. 2.4.2). The pigments in the scraped and soaked solutions would have been subjected to greater exposure to air, potentially resulting in greater metabolic breakdown and hence lower absorbance measurements.

2.4.3 Conclusions

A solution of the yellow residue resulting from vibration showed somewhat similar properties to diluted pericarp juice. TSS and pH were very similar, when corrected for concentration differences. Absorbance was much lower in the residue compared to pericarp juice dilutions. This may have resulted from greater metabolic breakdown in the residue. The similarity between residue and pericarp solutions supports the hypothesis that the residue results from the drying of leaked cell contents. Further research comparing the residue with extracted epicarp cell contents would be required to confirm this theory.

2.5 Scanning Electron Microscope Study of Mechanical Damage in Lychee

SEM study of damaged tissue was undertaken to observe the ultrastructural changes in lychee tissue resulting from mechanical injury.

2.5.1 Methods and Materials

'KMP' fruit grown at Byron Bay were harvested and transported to Brisbane padded with moist paper towel, and over-wrapped with plastic to ensure minimal moisture loss or damage. Fruit were stored at 4^{o}C overnight prior to treatment. In an observational trial, the fruit were subjected to mechanical damage treatments:

- Impact applied to the cheek centre from level 2, using a round head and no added weights, estimated to be equivalent in impact energy per unit area to a 67 cm fall;
- Abrasion by a single 15 cm rub against a metal file applied to the cheek centre;
- Vibration using a flask shaker on setting 4 for 10 minutes, with 15 fruit in the box, producing a moderate injury;
- Compression by maintaining 6 mm displacement for 20 seconds using a 6 mm diameter contact surface;
- Control, with no mechanical injury applied.

Samples, consisting of single injured protuberances from five fruit, were taken immediately after injury, after 5 hours and after 48 hours. Samples were rinsed in distilled water, and then placed immediately into liquid nitrogen. Additional samples of moderately and severely desiccated tissue were taken for comparison. Protuberances with silvering were also sampled, to attempt to detect signs of mechanical damage.

Samples were freeze-dried and bonded to an observation stage. The tissue edges were coated with carbon paint, and the samples were splutter coated with either gold or platinum. Samples were viewed using JEOL JSM 6300F and 6400F model field emission scanning electron microscopes, at an accelerating voltage of 5kV. Images were captured digitally. For each sample, a 20X image of the whole protuberance, and a series of slightly overlapping 50X photos were taken, mapping the entire protuberance. The protuberance tips were photographed at 100X and 300X. Areas of particular interest were studied under greater magnification.

A map of combined 50X images was created for each sample for the measurement of cracks. Cracks were recorded from the 300X and 100X protuberance tip shots, and these areas were not measured at lower magnification. The centre of the deep crevice around each protuberance was considered the boundary between protuberances. The area of the entire protuberance was estimated by averaging 16 measurements of radius, and assuming a roughly spherical shape.

The direction, location and area of all cracks were recorded. Crack area was measured by two perpendicular diameters. Where the length (L) was greater than 3 times the width (W), area was estimated by L x W. Otherwise, the formula for the area of a circle was used, the average of length and width defining the diameter. Crack direction was recorded as either radial (running roughly from tip to base), transverse (perpendicular to radial), or neither (including rounder shapes, and cracks not clearly radial or transverse). Location was defined as:

- Crevice, located within the deep crevice between protuberances;
- Base, within 0.5 mm of the crevice centre, but not within the crevice itself;
- Upper, within a 0.5 mm radius of the protuberance tip;
- Middle, not falling within base or crevice, but within a 0.5 to 1 mm radius of the tip;

- Lower, not falling within base or crevice, but greater than 1 mm from the tip.

Where base and upper areas overlapped, the area was divided evenly between the two.

2.5.2 Results and Discussion

Mechanical damage generally caused noticeable disruption of the surface tissue, with signs of damage often obvious at low magnification. Control fruit showed a generally intact surface, with varying levels of microcracking (Fig. 2.5.1). Slight tip damage, possibly from pre-harvest injury, was observed in some control fruit (Fig. 2.5.1D), but was minimal compared to the injured fruit. Abrasion caused severe damage, including substantial removal of surface tissues (Fig. 2.5.2). The wound surface showed severely injured sheared tissue, surrounded by a rugged jumble of damaged tissue. An outer flat area surrounding the wound was minimally damaged, with possibly only a single layer of cells removed. The skin surface was strongly disrupted by vibration, resulting in a rough, powdery surface appearance (Fig. 2.5.3). Accretions of removed tissue occurred primarily on the upper half of the protuberance, but were often observed over the entire protuberance (Fig. 2.5.3C). Compression injury typically appeared as mild cracking, crumbling or distortion of the tip (Fig. 2.5.4). Impact damage was similar in appearance to compression damage, although generally more severe (Fig. 2.5.5).

Greater magnification of the protuberance tip revealed slight cellular damage in most control fruit (Fig. 2.5.6A, B, C). In fruit injured by abrasion, cell-shaped holes in the tissue were often apparent, giving a honeycomb-like appearance (Fig. 2.5.6D, E). Crumbled cellular material was also visible at high magnification (Fig. 2.5.6F). Compression typically resulted in slight flattening of the tip surface, and some loss of tissue (Fig. 2.5.7A, B). Some fruit showed substantial cracking, with the tip becoming crater-like in appearance (Fig. 2.5.7C). Impact damage resulted in a similar appearance, but more often resulted in deep fissures (Fig. 2.5.7D, E, F). Magnification of vibration damage revealed tips often flattened in appearance, with a crumbly texture and small cracks (Fig. 2.5.8A, B, C).

Fruit showing signs of silvering consistently showed a slight damage to the tip, similar in appearance to vibration injury (Fig. 2.5.8D, E, F). Tips showed flattening of texture, slight crumbling of tissue, and powdery appearance. This type of injury may have resulted from mild pre-harvest rub, against branches or other fruit. The damaged areas were much smaller in scale than the areas of silvering observed, which covered around one fifth of the protuberance. While a direct link between abrasion and silvering has not been established, this study gives some further evidence towards a relationship, as suggested by Bagshaw *et al.* (1995).

Desiccated fruit did not show obvious signs of cellular damage (Fig. 2.5.9E, F). Even in severely desiccated fruit, surface texture was indistinguishable from control fruit. Vibration and desiccation can cause similar tissue browning in lychee, but appear very different under a microscope. SEM, or even a simple dissecting microscope could be used as a diagnostic tool to differentiate desiccation from vibration injury.

Surface arrangements showed a large degree of variation, even in undamaged fruit, with ornate patterns typically observed (Fig. 2.5.10). These patterns probably show the arrangement of epicuticular waxes, or the underlying cutin structures, with the complex shapes assisting in the prevention of moisture loss (Pantastico 1975). The flattened appearance of tissue in compression (Fig. 2.5.7A, B) and vibration (Fig.

2.5.11D, F) injured fruit may result from loss of wax structure, with wax arrangements being pressed into a flat layer.

Figure 2.5.1 Typical control lychee pericarp surface, showing whole protuberances (A, B, C) and magnified tips (D, E, F).

Figure 2.5.2 Lychee pericarp surface of abrasion injured fruit, showing whole protuberances (A, B, C) and the protuberance tips (D, E, F).

Figure 2.5.3 Lychee pericarp surface of fruit injured by vibration, showing whole protuberances (A, B, C) and magnified tips (D, E, F).

Figure 2.5.4 Lychee pericarp surface of compression injured fruit, showing whole protuberances (A, B, C) and magnified tips (D, E, F).

Figure 2.5.5 Lychee pericarp surface of fruit injured by impact, showing whole protuberances (A, B, C) and magnified tips (D, E, F).

Figure 2.5.6 Magnified protuberance tips of control (A, B, C) and abrasion damaged (D, E, F) lychee fruit.

Figure 2.5.7 Magnified protuberance tips of compression (A, B, C) and impact (D, E, F) injured lychee fruit.

Figure 2.5.8 Magnified protuberance tips of vibration injured lychee fruit (A, B, C) and fruit showing silvering (D, E, F).

Figure 2.5.9 Lychee pericarp protuberances showing silvering (A, B), and surface texture near tips in fruit showing silvering (C, D). Whole protuberance of moderately desiccated lychee fruit (E), and surface texture (F).

Figure 2.5.10 Lychee pericarp surface texture of control fruit, showing ornate patterns.

Figure 2.5.11 Surface texture of vibration damaged lychee pericarp.

Under high magnification, the vibration damaged surface shows chunks of tissue lifted away (Fig. 2.5.11A, E). Angular debris on the fruit surface (Fig. 2.5.11B, C) may be cellular material, or crystallised cell contents. This material is likely to contribute to the powdery texture observed in damaged fruit.

The distinctive appearance of various mechanical injuries under SEM correlated with the gross symptoms observed (Fig. 2.5.12). Compression and impact damage result in a similar symptom of dark brown tip discolouration, and also show similar tissue disruption under SEM. Cracking and crumbling of tissue was observed in the same region as tip darkening, suggesting that this physical damage triggers tissue darkening. The surface of vibration damaged tissue is scattered with damaged cellular material, conveying the powdery appearance observed as a gross symptom of damage. The confirmation of extensive cellular damage in vibration injured fruit supports the hypothesis that leaked cell contents may contribute to the yellow crust seen on the pericarp.

Figure 2.5.12 Gross symptoms of damage to the lychee pericarp caused by abrasion (B,C), impact (D), vibration (E) and desiccation (F), and an undamaged control (A).

Crack areas tended to be slightly higher in injured fruit (Fig. 2.5.13), but a significant effect was detected only due to vibration (t-test p<0.001). In abrasion damaged fruit, a significant effect was detected when the large wound area was included in crack measurements (t-test p<0.001), but not when this area was excluded. No time effects or trends were observed in any of the measured parameters (data not shown). Similarly, sampling time did not noticeably affect the sample appearance (data not shown). Cracks tended to be more likely in the upper level in impact injured fruit

(25% of cracks in the upper level), than in controls (14%). Other ratios of cracking location were fairly similar to the control (data not shown). Ratios of crack direction were also similar across all treatments, with no significant differences detected (data not shown).

2.5.3 Conclusions

Mechanical damage caused substantial visible changes in tissue ultrastructure. Control fruit generally showed intact surface tissues, with very slight damage. Ornate patterns, thought to be epicuticular waxes, were observed under high magnification. Abrasion injury was severe, with deep removal of tissue at the tip. In contrast, vibration damage was more superficial, but often covered the entire protuberance. Chunks of tissue and debris resulting from vibration were though to create the characteristic powdery appearance of the injury. Vibration significantly increased the area of cracks on the protuberance. Both vibration and compression caused some flattened areas, thought to occur through disruption of wax structure. The strong differences between desiccation and vibration browning under SEM may be used as a diagnostic tool, as desiccation caused no substantial changes in appearance. Impact and compression caused cracking and crumbling of tissue near the tip, which was probably linked with tip darkening. A possible link between silvering and mechanical injury was supported by SEM study. Affected fruit showed consistent very slight tip damage, possibly due to pre-harvest rub. The study provided a greater understanding of the mechanisms of injury in the fruit surface tissues.

2.6 Effect of Mechanical Damage on Cuticle Properties

Mechanical damage to the skin can occur due to the disruption or removal of epidermal cells and waxes from the skin surface (Schoorl 1974). This experiment sought to detect changes in tissue properties by quantitative measurements of wax and cuticle weights.

2.6.1 Methods and Materials

Mature ‘KMP’ fruit grown at Childers were harvested by hand, and transported to the Brisbane laboratory by air. The fruit were stored in moist paper towel, over-wrapped with aluminium foil, and plastic, and padded with packing foam to ensure minimal moisture loss or mechanical damage during transport. Fruit were stored at 4°C overnight in plastic bags prior to treatment. In a randomised control trial, standard mechanical damage treatments were applied to fruit:

- Light impact was applied using the pendulum from level 3, with a flat head and no added weights, with impact energy per unit area estimated to be equivalent to a 69 cm fall;
- Heavy impact was applied from level 3, using a round head and no weights, with impact energy per unit area estimated to be equivalent to a 93 cm fall;
- Abrasion was applied by two 15 cm rubs against a metal file;
- Vibration was applied by flask shaker at setting 4 for 10 minutes;
- Control fruit were not injured.

Impact and abrasion treatments were applied at three points on the fruit surface, the cheek, one side and the base of each fruit.

Measurements of wax and cuticle weight were made immediately after treatment and then after three days storage at 53% RH and 25°C. Using 20 fruit from each treatment, 3 discs per fruit of injured pericarp were cut using a cork borer (disc area of 2.42 cm^2) and were rinsed of aril juices in distilled water. The discs from each fruit were washed for five mins in 10 ml acetone, under gentle agitation. The acetone was replaced four successive times. The washings were evaporated to dryness in a pre-weighed bottle and the wax weighed. Data were expressed as weight of epicuticular waxes per unit of area.

Cuticle weight was measured by enzymatically degrading discs of pericarp to separate tissue from the cuticle (Leon and Bukovac 1978). Discs from treated fruit were placed in 5% pectinase plus 0.2% cellulase at pH 4.0 buffered with dibasic sodium phosphate-citric acid. Discs were kept in this solution for 12 days at 25°C with the solution renewed every 3 days. Underlying tissue was gently scraped, and rinsed away from the cuticle in distilled water. Each cuticle was then thoroughly rinsed, air-dried and weighed. Damage to some discs, both during storage and removal of tissue, resulted in the loss of pieces of cuticle, or shattering of the cuticle. The failure rate was fairly high, and resulted in the sample size being limited to 21 (the 21 most intact cuticles for each treatment were weighed).

2.6.2 Results and Discussion

2.6.2.1 Cuticle Weights

Unexpectedly, the flat head impact was the only treatment resulting in significantly reduced cuticle weight (t-test p=0.019). The average cuticle weight in discs treated by flat head impact was 8.5% lower than the controls (Fig. 2.6.1). Impact against a smooth hard surface would not be expected to remove tissues from the skin. It is possible that this effect may have occurred while discs were stored in the enzyme solution. Impact forces on the protuberance tips may have weakened the tissue structure. When additional forces were later applied (for example, when discs were gently shaken in solution), the already weakened tissue would be more easily lost from the disc. Impact also significantly effects weight loss from the whole fruit, thought to be due to increased moisture loss (Section 2.1.2.5*)*. The removal of surface tissue may also contribute to weight loss observed in the whole fruit. This effect may occur by a similar mechanism as observed in the isolated cuticle, with damaged tissue being lost during handling, rather than during the impact event.

The round head impact, although applying greater impact energy per unit area, had little effect on cuticle weight. The flat head results in light tip injury to many protuberances, whereas the round head results in more severe injury to fewer tips, sometimes affecting only a single protuberance. The limited number of tips damaged by the round head may prevent the detection of an effect. The mild level of damage caused by the flat head appears to be strong enough to trigger loss of cuticle weight.

Removal of part of the cuticle has been observed as a result of abrasion in other fruit, such as watermelon (Puchalski and Brusewitz 1996). In lychee, the abrasion treatment resulted in a 5.8% lower cuticle weight than controls, but a significant effect was not detected (t-test p=0.084). Lychee pericarp treated by abrasion shows a definite loss of surface tissues, revealed by the “sawn off” appearance of the protuberances, and by residual tissue seen on the abrasive surface. A greater level of replication may have detected an abrasion effect, as the power of analysis was low (0.281). In addition, only a few protuberances on each disk were damaged, hindering the detection of an effect. Vibration had no significant effect on cuticle weight. This type of injury may have a greater impact on the arrangement of surface waxes, rather than causing substantial loss of surface tissue.

Flat head impact treated cuticles increased in weight by 21% during a three day storage period after injury (t-test comparing day 0 and day 3, $p<0.001$). At this stage, the flat impact treatment was shown to significantly increase the cuticle weight compared to the control (t-test $p=0.004$), in contrast to the decrease in weight previously observed. All other mechanical damage treatments showed a small, but not significant, increase in weight, in the range of 3-6% over the three days following injury. Control fruit showed no gain in weight over the same period (-0.3% change). The increase in cuticle weight in the days following injury is possibly due to wound healing. The wound healing hypothesis is reinforced by a decline in weight loss rates from injured whole fruit over a similar period of time (Section 2.10.2).

In tissue sampled on day three, a dark substance was observed in many of the injured protuberances, usually highly localised to the injury site. This may have resulted from oxidation of phenols or wound healing processes. The dark tissue was strongly bound to the cuticle, and to the vascular system of the underlying pericarp. It was generally firmer than the surrounding tissue, but less firm than the cuticle itself. This type of symptom was most strong in abrasion injured fruit, but was seen in response to all mechanical injuries. The greater severity of the response in abrasion treated fruit does not correlate well to the observed weight increases in impact treated fruit.

However, this may again be due to the limited number of tips per disk affected by the abrasion injury.

2.6.2.2 Epicuticular Waxes

Mechanical damage did not appear to remove a significant amount of wax, with no significant difference in wax weight observed between damaged and control fruit (Fig. 2.6.2). It is likely that the major effect of mechanical damage on epicuticular waxes is the disruption of structure (Section 2.5), rather than the removal of wax.

All treatment groups showed a decline in wax weight over three days storage at room temperature. The loss of wax weight was significant in all treatments (t-tests $p<0.007$) except for the flat head impact (t-test $p=0.072$). In the three days following injury, vibration treated fruit lost 31% of wax weight, while flat head impact treated fruit lost only 10%, resulting in a significant difference between treatments (t-test $p=0.002$). It is not clear why this difference occurred. Despite the large difference in wax loss, impact and vibration treatments at day 3 were not significantly different to the control, which showed a 21% loss of wax weight over three days storage.

2.6.3 Conclusions

Cuticle weight was significantly reduced by impact with a flat head, an effect that was thought to occur during storage of tissue discs in enzyme solution. Other treatments did not significantly affect cuticle weight, possibly due to the small number of protuberances injured on each disc. An increase in cuticle weight during storage was observed in injured fruit, suggesting a possible wound healing process. The observation of a dark substance adhered to the cuticle and vascular tissue of injured fruit gave further evidence of wound healing. Mechanical injury did not appear to affect the amount of wax on the fruit surface, but is likely to affect wax structure.

2.7 Effect of Mechanical Damage on Anthocyanins, Phenolics and Flavonoids

Biochemical processes in fruits are often reflected in changes in phenolic compounds, which have a diverse range of functions. Phenolics contribute to fruit development and confer sensory properties such as colour, taste and scent. They are also involved in resistance to various stresses, with levels of phenolics commonly increasing after an incident of stress, such as mechanical injury (Macheix *et al.* 1990).

Flavonoids are a specific group of phenolic compounds, including flavonols and anthocyanins. Anthocyanins confer the red colour characteristic of the fresh lychee pericarp. Anthocyanins and phenolics tend to decline with the progression of desiccation browning in lychee, suggesting that the polymerization of phenols contributes to browning (Jiang and Fu 1999; Zhang *et al.* 2001). Tissue darkening has been observed in lychee in response to mechanical injury (Section 2.1.2.3). It has not yet been determined whether colour changes due to mechanical injury follow similar chemical pathways to desiccation browning. Impact has been observed to trigger an increase in anthocyanin production in apples, thought to occur in response to wound-induced ethylene generation (Chalmers and Faragher 1977). A shift in colour towards red purple has been observed in lychee following impact damage (Section 2.1.2.3), suggesting a possible increase in anthocyanin levels. A trial was carried out to determine the effect of mechanical damage on levels of anthocyanins, phenolics and flavonoids in the lychee pericarp.

2.7.1 Methods and Materials

Fruit of cultivar 'KMP' were harvested from Childers and were transported to Brisbane by car. In a randomised control trial, mechanical damage treatments were applied to 26-27 fruit each:

- Impact treatment was applied by dropping fruit ten times from 50 cm onto a smooth hard surface;
- Abrasion was applied by dragging fruit 15 times along 20 cm of fine grade sandpaper (280), with damage spread over the fruit surface;
- Vibration was applied for 2 minutes, holding a cardboard box lightly within a test tube shaker on setting 4, with fruit re-arranged every 30 seconds;
- Control fruit were not mechanically injured.

Fruit were stored in open punnets at 75% RH and RT (20-30^{o}C). Ten fruit in each treatment were used for measurement of changes in fruit weight, browning score and colour. Samples of pericarp tissue were taken from 16-17 fruit from each treatment after 3 days storage. The tissue was placed into liquid nitrogen in small plastic bags, and was later transferred to –80^{o}C storage.

2.7.1.1 Measurement of Anthocyanin, Phenolics, Flavonoids and ADI

Approximately 2 g of lychee pericarp was finely chopped on a cool surface, weighed and then immediately placed into 10 ml 1% HCl methanol. The sample was blended using a Polytron PTA, 1OS, 12 mm aggregate head on setting 4 for 1 minute. The blender head was rinsed using 10 ml 1% HCl methanol, and the washings were added to the sample. The solution was stored at 4^{o}C for 24 hours to allow the pigment to leach out. The solution was then filtered using No. 1 Whatman filter paper and diluted to 100 ml with 1% HCl methanol. Samples were stored for up to 7 days at 4^{o}C after filtering, as this was found to cause no significant change in readings (data not shown).

Absorbance of each solution was measured using a CARY 1E spectrophotometer. Anthocyanin levels were estimated by absorbance at 530 nm. The molar extinction co-efficient (3.43×10^4) determined by Siegelman *et al.* (1958) was used to convert absorbance to moles of anthocyanin per cm^2. Flavonoid content was expressed as absorbance at 325 nm per gram of fresh weight. The solution was diluted to 1:10 and absorbance recorded at 280 nm as a measure of phenolics. The total phenolics were calculated from a standard curve of gallic acid (**Appendix 1**), and results were expressed as gallic acid equivalents per gram of fresh weight. The anthocyanin degradation index (ADI) was measured by adjusting the pH to 4.6 and 0.9. Two 0.5 ml aliquots were taken from the original anthocyanin extract, and were diluted to 3 ml using either 0.2N KCl : 0.2N HCl (25:67) pH 0.9, or sodium acetate : 1N HCl : water (100:60:90), pH 4.6. Solutions were left to equilibrate at RT for ten minutes before the absorbance of each was measured at 530 nm. The ADI was then calculated according to Fuleki and Francis (1968);

$$\text{ADI} = \text{A (pH 0.9)} / (\text{A (pH 0.9)} - \text{A (pH 4.6)}) \qquad \textbf{\textit{…Equation 2.7.1.1}}$$

Where A = total anthocyanin.

2.7.2 Results and Discussion

Large inherent variation between samples generally prevented the detection of significant differences. Impact treated fruit showed significantly reduced levels of phenolics compared to the control (t-test p=0.045), but no other significant differences to the control were detected (Fig. 2.7.1). A decline in phenolics may occur due to tissue browning, as shown in lychee desiccation browning (Jiang and Fu 1999). Impact damage causes tissue browning in the protuberance tips, which may have resulted in the observed decline in phenolics. However, phenolics can also increase in response to stress, due to wound healing or systemic resistance responses (Macheix *et al.* 1990). These opposing changes in phenolic content could result in similar overall levels to undamaged fruit, masking biochemical changes due to injury.

2.7.3 Conclusions

Impact treated fruit showed a significant decline in phenolics compared to the control, possibly due to the induction of tissue browning reactions. The large inherent variation in biochemical properties prevented the detection of any other effects.

2.8 Effect of Mechanical Damage on Electrolyte Leakage

Tissue damage and senescence are commonly assessed by the measurement of electrolyte leakage (Studman 2001). Cell damage results in increased leakage of cell contents due to the rupture of entire cells, or loss of membrane integrity. These processes often lead to a decline in shelf life and greater susceptibility to pathogens, due to the release of nutrients and moisture onto the fruit surface. Electrolyte leakage in lychee was studied as an index of tissue damage to assess the influence of various mechanical injuries on the fruit.

2.8.1 Methods and Materials

‘Wai Chee’ fruit from Sippy Downs, in SE Queensland, were harvested into water and soaked for at least 30 minutes to ensure a reasonable level of hydration. In a randomised control trial, forty fruit were injured by each of 5 treatments:

- Impact by pendulum using a flat head with 2 added weights at level 2.5, estimated to be equivalent in impact energy to a 60 cm fall;
- Vibration for 10 minutes at setting 4, with 20 fruit (approximately 370 g) in each run, producing a moderate vibration injury;
- Abrasion against a metal file, three 15 cm rubs spread over one cheek;
- Compression of 1 kg for 2 hours;
- Control treatment.

Cracks were noted when they occurred. Twenty fruit from each treatment were each immediately placed into 50 ml RO water within a plastic cup. The cups were covered with cling wrap and stored at 20^{o}C. The remaining treated fruit were stored in damp paper towel at 20^{o}C for approximately 24 hours before being placed into cups of RO water. Fruit were removed from the water after soaking overnight, and the electrical conductivity of each solution was measured.

2.8.1.1 Analysis

Measurements made from 0-24 and 24-48 hours did not significantly differ, and were therefore combined for analysis. The number of hours spent soaking, which ranged from 23 to 37, showed no correlation with electrical conductivity. Correcting for these differences, by the use of an hourly rate (dividing the conductivity reading by the number of hours spent soaking), resulted in a significant correlation. Data were therefore not corrected for soaking time. Similarly, the weight of control fruit did not show a significant correlation with conductivity. The expression of data per unit of surface area created a significant correlation, so analysis was based on unmanipulated conductivity values.

2.8.2 Results and Discussion

Mechanical damage treatments did not cause any significant change in electrolyte leakage (Fig 2.8.1). Closed cracks had no significant effect on electrolyte leakage (compared to both control fruit, and uncracked impact-treated fruit) (Fig. 2.8.2). The number of cells ruptured by closed pericarp cracking was obviously not sufficient to cause a significant increase in electrolyte leakage. In contrast, fruit with open cracks showed a substantial increase in electrical conductivity, probably resulting from leakage of aril juice (t-test against control, $p<0.001$).

Only one fruit showed signs of a rot, and this resulted in a massive increase in electrolyte leakage. The electrical conductivity of the fruit with a rot was 113 mV, compared to an average value of 77.8, and maximum of 106 in the other 199 fruit. This fruit was removed from all analyses, but provided a basis for comparison. Even the most severe open crack did not cause as great a loss of cell integrity as pathogen growth.

Fruit weight appeared to have some effect on electrolyte leakage, with an overall positive correlation (Spearman correlation co-efficient=0.270, $p<0.0005$), suggesting that larger fruit tended to give higher electrical conductivity readings. However, this effect occurred only in mechanically injured fruit, with control fruit showing no significant correlation. This would suggest that larger fruit show a greater increase in electrolyte leakage in response to mechanical damage, when compared to small fruit. Separate analyses excluding smaller sized fruit did not reveal a significant difference in electrolyte leakage between control and injured fruit.

2.8.3 Conclusions

The levels of tissue damage resulting from standard mechanical injuries, including visible closed cracks, were not sufficient to cause a significant increase in electrolyte leakage. Open cracking, penetrating to the aril, caused a substantial increase in electrolyte leakage. However, rot development caused a far greater increase in cellular leakage than even the most severe wound. The tissue damage resulting from mechanical injury may be largely characterised by the rearrangement and disruption of the cells, rather than their actual rupture.

2.9 Carbon Dioxide and Ethylene Production in Mechanically Damaged Fruit

Increases in carbon dioxide and ethylene evolution are commonly observed in response to mechanical injury. Carbon dioxide (CO_2) can be generated due to the decarboxylation of malic acid spilled from damaged cells (Hung 1993), or stimulation of cellular respiration (MacLeod *et al.* 1976). Increased ethylene production is a commonly observed response to mechanical injury in many fruits, such as pear (Mencarelli and Botondi 1992), apple (Lougheed and Franklin 1974) and tomato (MacLeod *et al.* 1976). In some produce, such as sweet cherry (Wade and Bain 1980) and papaya (Quintana and Paull 1993), this response is not observed. Both ethylene production and increased respiration can reduce shelf life. Even at very low levels, ethylene can substantially reduce the shelf life of fresh produce (Wills *et al.* 1999). Respiration depletes sugars and other storage products, reducing the energy reserves of the fruit, and deteriorating sensory quality and nutritional value. These processes reduce overall quality and shorten the postharvest life of the fruit. A trial was carried out to determine whether mechanical damage significantly affects ethylene and CO_2 evolution in lychee.

2.9.1 Methods and Materials

Fruit of cultivar ‘KMP’ were harvested from Sippy Orchard, and were soaked in water for at least 30 minutes immediately after harvest. The fruit were dried using tissue, packed in damp paper towel and transported to the laboratory for immediate treatment. Treatments of mechanical injury and moisture loss were applied randomly:

- Impact by pendulum using the flat head with 2 additional weights at level 2.5, estimated to be equivalent to a 60 cm fall;
- Vibration for 10 minutes at setting 4, with approximately 370 g fruit in each run;

- Abrasion against a metal file, applying three 15 cm rubs spread over one cheek;
- Compression of 1 kg for 2 hours using a compression device (Section 1.6.4.5.1);
- Low humidity treatment (see Section 5.8);
- Control treatment, stored at 75% RH and 20^{o}C with no mechanical injury applied.

All fruit were stored at 20^{o}C and 75% RH after injury.

In the first experiment, 10 fruit each were subjected to the six various treatments in a randomised control trial. Half of these were immediately placed into small sealed plastic containers after injury, and were stored at 20^{o}C. The remaining fruit were placed into punnets within large plastic crates, and were stored at 20^{o}C and 75% RH for 24 hours. After this storage period, the fruit were placed into small plastic containers.

The fruit were stored singly in 100 ml “Castaway” brand plastic containers (CA-FC100), using a seal of water to ensure no gas leakage. The actual airspace of the containers was measured as 124.7 ml. A small hole in each container was sealed with tape to provide access using a syringe. Ethylene and CO_2 levels were measured in samples taken from the containers approximately 24 hours after sealing, with the exact times noted. Holes made by the syringe were taped over immediately.

Ethylene measurements were made using a gas chromatograph (GC) with flame ionisation detector (Shimadzu GC-8A) and an attached integrator unit (Shimadzu C-R6A Chromatopac). For CO_2 measurements, the same type of integrator was used with a GC using a thermal conductivity detector (Shimadzu GC-8A T.C.D.). The standard gas contained 1.8 +/- 0.1 ppm ethylene, 1.46 +/- 0.03% CO^2 and 1.53% +/- 0.03% O^2 in nitrogen.

In the first experiment, problems with the GC prevented some samples from being measured for CO_2, resulting in replication as low as 3 per treatment. Some samples were left up to 52 hours before measurement could be made. The exact time of placement into the container and time of measurement were recorded, and this allowed the measurements to be converted into hourly rates. Levels of injury and fruit weight were recorded after removal from the container. For impact damage, cracks and tip injury were recorded. For vibration, the surface area damaged was estimated, and a score was given for the severity of injury (0 to 6, where 0 = none and 6 = severe).

The experiment was repeated three days later using freshly harvested fruit from the same cultivar and orchard. In a randomised control trial, 10 fruit were subjected to each of the same mechanical damage treatments. An additional 3 fruit were treated by a slightly higher level of impact (level 3, flat head, 2 weights), estimated to be equivalent to an 81 cm fall. All fruit were placed into small containers immediately after injury, and gas was sampled after approximately 24 hours (ranging from 23.8 to 26.2 hrs). Fruit were weighed prior to treatment. In addition to observation of the extent of injury, the fruit were scored for level of pepperspot infestation (Table 2.9.1).

Table 2.9.1 Pepperspot scores assigned to lychee fruit.

Score	Level of damage	Approximate range
0	None	0 spots
1	Very mild	<50 spots
2	Mild	50-150 spots
3	Moderate	150-300 spots
4	Moderate-severe	> 300 spots and 25-50% fruit surface area
5	Severe	>50% fruit surface area

2.9.1.1 Calculations

Standard peaks were averaged from numerous measurements, with 1.46% CO_2 giving an average peak of 14845, and 1.8 ppm ethylene giving a peak of 5089. The basic formula for calculating concentration from peak (Equation 2.9.1a) was adjusted to account for atmospheric levels of CO_2 (Equation 2.9.1b). An atmospheric level of 0.03% CO_2 was assumed, and this was calculated to equate to a baseline peak of 305.

$C_A = C_B \times P_A / P_B$ ***…Equation 2.9.1a***

$C_A = C_B \times (P_A - 305) / P_B$ ***…Equation 2.9.1b***

Where C_A = sample concentration, C_B = standard concentration, P_A = sample peak area and P_B = standard peak area.

The generation of CO_2 was calculated by correcting for container volume, time and fruit weight (Equation 2.9.2).

$G = V \times C_A / t / M$ ***…Equation 2.9.2***

Where G = generation of CO_2 (ml kg^{-1} hr^{-1}), V = container volume (ml), C_A = sample concentration, t = time elapsed (hours) and M = fruit mass (kg).

Ethylene levels were similarly calculated, but without the correction for atmospheric levels. Rates of ethylene generation were expressed as $\mu L\ kg^{-1}hr^{-1}$.

2.9.2 Results and Discussion

2.9.2.1 Effect of Mechanical Injury on CO_2 Evolution

Mechanical damage was shown to significantly increase levels of CO_2 generation (Fig. 2.9.1). Compression, vibration and heavy impact significantly affected the generation of CO_2 (Analysis by t-tests, p=0.011, 0.029 and 0.009 respectively). The heavy impact, equivalent to an 81 cm fall, caused the greatest increase, with CO_2

generation more than double that of the control. This level of impact caused cracking in all three fruit treated, with 2 fruit showing open cracking. The 60 cm impact also caused substantial damage to the fruit, resulting in 40% cracking, and 15% open cracking. Despite this, the 60 cm impact did not cause a significant increase, with CO_2 evolution only 11% higher than the control. The substantial leap in CO_2 generation between 60 and 81 cm impacts suggests a damage threshold in this region.

Pericarp cracking did not appear to strongly affect CO_2 generation, with no correlation between crack area and CO_2 levels (data not shown). In contrast, the rise in CO_2 release in blueberries and cherries was proportional to the severity of damage (Burton and Schulte-Pason 1985). Analysis of lychee fruit grouped by open, closed or no cracking also showed no significant cracking effect, with no trend apparent (data not shown). Tip darkening was weakly negatively correlated with CO_2 evolution (Spearman correlation co-efficient = -0.495, p=0.0364). It is possible that less visible forms of damage (background cell disruption or rupture) trigger the increase in CO_2 generation. These types of damage may be negatively correlated with tip darkening due to the partitioning of energy between the two types of damage (Sitkei 1986). The strong effect of compression on CO_2 generation supports this hypothesis. Despite causing very little visible damage, compression of 1 kg for 2 hours resulted in a 52% increase in CO_2 evolution. The tissue damage causing increased CO_2 output may also relate to the background changes in colour observed after compression (Section 2.2.2.1), despite minimal visual signs of injury.

Vibration resulted in an average 45% increase in CO_2 generation. The severity of visible damage and surface area affected were not correlated with this response. This supports the hypothesis that increased CO_2 generation can result from cellular damage not visible to the naked eye.

In all mechanical damage treatments, the rate of CO_2 generation tended to decline slightly between 0-24 and 24-48 hours after injury (Fig 2.9.2). The low number of replicates at 24-48 hours prevented the detection of a time effect in most cases. In vibration injured fruit, a significant decline in CO_2 evolution was observed between 0-24 and 24-48 hours. The decline in CO_2 generation over time resulted in similar rates in control and damaged fruit from 24-48 hours, with no significant treatment effects detected in this period. The timing of the CO_2 response suggests that it may be primarily due to decarboxylation of malic acid spilled from damaged cells, rather than a sustained increase in respiration (Hung 1993).

2.9.2.2 Effect of Mechanical Damage on Ethylene Evolution

A significant increase in ethylene evolution was observed in response to most mechanical damage treatments (Mann-Whitney rank sum tests against the control; vibration $p<0.001$; 60 cm impact $p=0.016$; 81 cm impact $p=0.008$; compression $p=0.019$) (Fig. 2.9.3). Abrasion injured fruit did not show a significant increase in ethylene evolution compared to the control. Rates of ethylene and CO_2 evolution were strongly correlated (Spearman correlation co-efficient=0.541, $p<0.0005$).

Fruit injured by heavy impact (81 cm) showed the strongest response in ethylene evolution, with rates more than 12 times greater than the control fruit. The 60 cm impact also caused a substantial increase in ethylene release, more than 5 times higher than control. Ethylene evolution was significantly affected by open cracking, but fruit with closed cracks were not significantly different to uncracked or control fruit (Fig. 2.9.4). The release of ethylene from the aril may have contributed to the strong effect of open cracking on ethylene evolution.

Vibration treatment stimulated ethylene evolution, with rates more than 5 times greater than in control fruit. The surface area showing visible damage was positively correlated with rates of ethylene release (Spearman correlation co-efficient=0.519, p=0.0190). Fruit showing damage to 50 to 100% of the surface showed significantly higher rates of ethylene evolution than vibration treated fruit showing no damage (ANOVA p<0.001, comparison by Tukey test) (Fig. 2.9.5). Damage covering 5 to 25% of the fruit surface significantly increased ethylene evolution compared to the control. Fruit injured by vibration without any visible signs of damage did not significantly differ to the control. The vibration score for severity of damage did not show any significant correlation with ethylene evolution. Fruit with a severity score of 0 showed an average ethylene evolution of 0.028 μL kg^{-1}hr^{-1}. For scores of 1-6, average values were substantially higher, ranging from 0.110 to 0.135 μL kg^{-1}hr^{-1}, but no trend was shown over increasing severity score (data not shown). The lack of variation between severity scores suggests that even very mild injury can stimulate ethylene release.

Rates of ethylene evolution did not vary significantly between 0-24 and 24-48 hours after injury (data not shown), nor were any consistent trends shown.

The high rates of ethylene evolution shown in damaged fruit are a possible cause for concern, as the shelf life of fresh fruit tends to decline with increasing ethylene concentration. Across a wide range of non-climacteric commodities, shelf life at <0.005 μL L^{-1} ethylene averaged around 60% longer than at 0.1 μL L^{-1} (Wills *et al.*

1999). Even in mildly damaged lychees, this level of ethylene concentration could be reached in 4 hours within a sealed box, instead of the 10 hours taken by undamaged fruit. The effect of elevated ethylene on shelf life is yet to be tested in lychee. Rambutan, a fruit closely related to lychee, did not show a strong response to elevated ethylene levels (O'Hare *et al.* 1994).

2.9.2.3 Effect of Pepperspot on Ethylene and CO_2 Generation

No significant pepperspot effect on CO_2 or ethylene generation could be detected, and no trends were observed (data not shown). Pepperspot was not severe in this experiment, with almost all fruit of scores 0-3 (none to moderate). Further experimentation and greater replication would be required to confirm these results.

2.9.3 Conclusions

Mechanical damage treatments resulted in consistently higher rates of both ethylene and CO_2 evolution. Compression, vibration and impact (81 cm) significantly affected the release of CO_2, while impact (60 and 81 cm) and vibration treatments stimulated ethylene evolution. The timing of the CO_2 increase suggested that the response may have been primarily due to decarboxylation of spilled malic acid, rather than sustained respiration (Hung 1993). Higher rates of CO_2 generation in damaged fruit did not appear to be related to visible damage, such as tip darkening, cracking and vibration injury. It was theorised that this response may be related to non-visible damage, such as cell rupture. In contrast, ethylene evolution was related to visible damage, with open cracking and vibration injury SA affecting the response. The response of lychee to increased ethylene is yet to be tested, but increased levels have been shown to significantly reduce shelf life in some fresh produce (Wills *et al.* 1999).

2.10 Effect of Mechanical Injury on Weight Loss

Mechanical injury can damage the protective barriers on the fruit surface, thus increasing the rate of moisture loss. Even slight damage can have an effect, with barely visible scratches in citrus causing a significant increase in moisture loss under some storage conditions (Golomb *et al.* 1984). Increased respiration due to injury may also contribute to higher levels of weight loss in damaged fruit. Weight loss reduces the value of produce and accelerates deterioration. A trial was carried out to determine whether mechanical injury significantly affects subsequent weight loss in lychee.

2.10.1 Materials and Methods

The experiment was carried out on mature ‘Tai So’ fruit grown at Sarina. Fruit were stored in moist paper towel from harvest until treatment. In a randomised control trial, fruit were subjected to mechanical injury treatments, with each treatment applied to 60 replicate fruit:

- Impact treatment involved 3 impacts from level 2.5, using a flat head with no added weights. Each impact was estimated to be equivalent to a 51 cm fall onto a flat hard surface. Impacts were applied to the cheek centre, the side of the fruit (90^o from the cheek), and the base;
- Abrasion was applied 10 times for 15 cm using a metal file, with injuries spread over the surface of the fruit;
- Vibration fruit were treated using a flask shaker on setting 4, for 5 minutes duration;
- Control fruit were not injured.

Once treated, the fruit were stored in drying conditions (33% RH) at 20^oC. The saturated salts used in this experiment (MgCl) dissolved quickly under the humid conditions, and hence RH was higher and less rigorously controlled than desired. The weight of each fruit was measured immediately before and after injury, then at 23, 53 and 75 hours. The data were converted to % weight loss prior to analysis.

2.10.2 Results and Discussion

2.10.2.1 Vibration Injury

Vibration injury caused a slight, but significant (t-test $p<0.001$), immediate loss of weight averaging 14.3 mg, or 0.07% of fruit weight. This immediate weight loss was likely to be partially due to the loss of tissue during the vibration treatment. Moisture loss is also likely to have contributed to the loss of weight observed. The treatment was carried out within an unlined cardboard box, which is likely to have drawn water away from the fruit. This scale of weight loss is very minor compared to normal postharvest losses, and in itself would not be a concern in the management of fruit quality.

Levels of weight loss from the vibration injured fruit were slightly higher than the controls during early storage. Weight loss was significantly higher in vibration treated fruit in the period from before injury to 23 hours (Table 2.10.1). However, this period included the immediate weight loss caused during vibration injury. The removal of this

Table 2.10.1 The effect of vibration injury on lychee weight loss during storage.

Period	Average % weight loss		p-value (t-test)	% Difference due to treatment
	Vibration	**Control**		
Before-After	0.0662	0.0000	<0.001	-
Before-23 hrs	1.0112	0.8827	0.028*	14.55
After-23 hrs	0.9456	0.8827	n.s.d.	7.13
23-53 hrs	1.0097	1.0013	n.s.d.	0.85
53-75 hrs	1.3279	1.3829	n.s.d.	-3.98

N=60. * Indicates analysis on ln transformed data.

immediate effect resulted in no significant difference being detected, despite a 7% greater loss of weight in vibration injured fruit, compared to the control.

The percentage difference between vibration and control fruit shows a trend over time. In the first 23 hours after injury, the fruit showed a 7% increase in weight loss compared to the control. From 23 to 53 hours rates of weight loss in control and injured fruit were very similar, while from 53-75 hours, the vibration treated fruit actually showed reduced levels of weight loss. This trend may result from the early evaporation of ruptured cell contents, or may be due to wound healing gradually blocking moisture loss from injured sites.

2.10.2.2 Impact Damage

Impact injury resulted in unexpectedly high weight loss compared to the control fruit. It would be expected that the formation of cracks in the pericarp would result in increased moisture loss, and that cracks penetrating through to the aril would have a particularly strong effect. However, in this experiment the injury inflicted on the fruit did not result in visible cracking. Despite this, a 30% increase in weight loss was shown in impact injured fruit in the 23 hours following injury (Table 2.10.2). It is possible that the formation of microscopic cracks in the pericarp may have contributed to this increase in weight loss in impact treated fruit. A damage-induced respiration response may also have slightly increased the rate of weight loss after impact. However, the respiration effect is unlikely to have been substantial, given that severe impact (81 cm) resulted in CO_2 generation only 11 $mgkg^{-1}hr^{-1}$ above the control value (Section 2.10.2.1).

Later in storage, the difference in weight loss between impact treated and control fruit declined, with no significant difference shown after 23 hours. The effect of impact injury on weight loss appeared to decline over time, almost halving every 24 hours. This effect may be due to a wound healing response, or may result from microscopic cracks or ruptured cells drying out over time.

Table 2.10.2 The effect of impact damage on lychee weight loss during storage.

Period	Average % weight loss		p-value	% Difference due to treatment
	Impact	**Control**		
Before-After	0.0693	0.0000	<0.001**	-
After-23 hrs	1.1551	0.8827	<0.001*	30.85
23-53 hrs	1.1686	1.0013	n.s.d.	16.71
53-75 hrs	1.5062	1.3829	n.s.d.	8.91

N=60, * indicates analysis by Mann-Whitney rank sum test, ** shows analysis by t-test.

The scale of increased weight loss observed in impact treated fruit may be of significance to the postharvest management of the fruit. A 30% increase in the rate of weight loss over the 24 hour period following impact injury could mean the difference between a brown or red fruit reaching the consumer. For example, under ambient conditions (25^{o}C and 50% RH, with exposure to light air currents) for 24 hours, a 30% increase would result in an estimated 7.1% weight loss, instead of 5.5%. Impact injury could thus greatly increase the risk of desiccation browning, particularly if fruit are exposed to drying conditions in the 24 hours following impact.

2.10.2.3 Abrasion Injury

Abrasion injury, applied to ten sites on the fruit, appeared to have very little effect on weight loss during storage (Table 2.10.3). After the immediate loss of weight due to the injury treatment, the abrasion injured fruit did not show any further significant losses of weight compared to the control. It is likely that increased moisture loss does occur after abrasive injury, but that the very small area affected by each injury has prevented detection of this effect. An abrasion injury applied using a flat implement, with no downward pressure, results in injury to an average of around four protuberances. Even applying the injury ten times results in only 40 protuberances being injured. With an average of around 330 protuberances per fruit estimated on ‘KMP’ fruit, this treatment would affect only 10-15% of the protuberances. A 3% increase in rate of weight loss compared to the control, although it may not appear substantial, suggests around a 24% increase in those protuberances actually affected by the injury (assuming increased moisture loss in 40 of 330 protuberances, with the remainder the same as the control). It is therefore likely that more extensive abrasion damage would result in substantially increased weight loss.

Table 2.10.3 The effect of abrasion damage on lychee weight loss during storage.

Period	Average % weight loss		p-value	% Difference due to treatment
	Abrasion	**Control**		
Before-After	0.0439	0.0000	<0.001*	-
After-23 hrs	0.9064	0.8827	n.s.d.	2.69
23-53 hrs	1.0416	1.0013	n.s.d.	4.02
53-75 hrs	1.3748	1.3829	n.s.d.	-0.59

N=60, * indicates analysis by Mann-Whitney rank sum test,

2.10.2.4 Further Analysis

Linear equations and 2 factor polynomial curves were fitted for each individual fruit for moisture loss over time, and the resultant slopes were analysed.

Linear: $y = ax$ ***…Equation 2.10.1a***

Two factor polynomial: $y = ax^2 + bx$ ***…Equation 2.10.1b***

The linear slopes did not reveal any additional information. The impact treated fruit were shown to lose weight significantly faster than the control fruit (t-test p<0.001), but abrasion and vibration treatments resulted in no significant difference (Table 2.10.4).

Table 2.10.4 The effect of mechanical injury on lychee weight loss - analysis of linear and two-factor polynomial curves.

Treatment	Linear		Two factor polynomial	
	“a”	p-value	**“b”**	p-value
Control	**0.040**	-	**0.028**	-
Impact	**0.049**	<0.001	**0.043**	<0.001**
Abrasion	**0.042**	n.s.d.	**0.035**	n.s.d.
Vibration	**0.042**	n.s.d.	**0.032**	0.038*

N=60, * Indicates analysis of treatment against control by Mann-Whitney rank sum test, all others analysed by t-test. ** Shows square root transformation of data prior to analysis.

Analysis of variables from the two factor polynomial showed that for impact treated fruit the slope, “b” was significantly higher than the control. Vibration treated fruit were shown to have a significantly higher “b” value than the control, indicating more rapid moisture loss. This analysis confirms that vibration injury has a significant effect on weight loss during storage.

2.10.3 Conclusions

All mechanical damage treatments caused a significant, although very small, immediate loss of weight, probably due to loss of tissue and moisture during treatment. The scale of immediate weight loss was, in itself, considered insignificant in the postharvest management of the fruit.

Damage by vibration was shown to result in a significant increase in the rate of weight loss during postharvest storage. The increased rate of weight loss was thought to be predominantly due to moisture loss through wounded tissue. In abrasion injured fruit, the rate of weight loss during storage was not found to significantly differ from the control. It was hypothesised that the limited area affected by this type of injury prevented the detection of an effect.

Impact damage resulted in significantly increased weight loss. The rate of loss was on average 30% faster than control fruit in the first 23 hours. This result was considered to be potentially significant to the postharvest management of the fruit, particularly with respect to desiccation browning.

2.11 The Effects of Pericarp Cracking on Postharvest Weight Loss

Impact injury was shown to significantly increase weight loss in lychee (Section 2.10.2.2), but further experimentation was required to explore the effects of visible pericarp cracking on weight loss. Cracks sever the protective barriers on the surface of the fruit, providing a potential pathway for moisture loss. In particular, open cracks extending to the aril would be expected to greatly increase moisture loss. Cracks that do not penetrate the endocarp, a thin membrane separating pericarp from aril, may have less effect on moisture loss, as this membrane is a strong barrier to water movement in undamaged fruit (Zhao *et al.* 1999). Knowledge of the effects of pericarp cracking on moisture loss will assist in the definition of tolerable damage limits.

2.11.1 Methods and Materials

‘KMP’ fruit were harvested from Sippy Downs in SE Queensland. Fruit were harvested directly into water, and soaked for approximately one hour. They were then dried and tightly packed in damp paper towel for transport to the laboratory. In a randomised control trial, fruit were treated either by impact or as a control, with 40 replicate fruit per treatment. Impact was applied from level 2.5, using a flat head with 2 additional weights, estimated to be equivalent in impact energy to a 60 cm fall.

Storage after treatment was at 57% RH and 20^{o}C. Fruit were weighed prior to treatment, and were re-weighed after approximately 6, 24 and 48 hours storage. The time of placement into storage, and times of measurement were recorded for each fruit, to allow the calculation of precise rates of weight loss. Cracks were noted after 48 hours, and were scored as either open (penetrating through the endocarp to the aril) or closed.

Levels of pepperspot on the fruit were substantial. It has not been noted whether this cosmetic blemish affects postharvest water loss. Estimated scores were assigned to each fruit for the level of pepperspot (Table 2.11.1), to allow this effect to be tested.

Table 2.11.1 Explanation of scores for lychee pepperspot.

Score	Description
0	No pepperspot
1	1-50 spots
2	51-100 spots
3	101-300 spots
4	>300 spots and 25-50% S.A.
5	>50% S.A.

2.11.2 Results and Discussion

Pepperspot had no significant effect on postharvest weight loss (data not shown). Data were analysed by correlations and comparison of score groups (ANOVA) for all uncracked fruit, and for control fruit separately. No significant effects were detected, and no trends were apparent.

Open cracking resulted in a substantial increase in weight loss during storage (Fig. 2.11.1). Rates of moisture loss in open cracked fruit (Fig. 2.11.2) were significantly different to control fruit throughout the 48 hour storage period (Mann-Whitney rank sum tests; 0-6 hours, $p<0.001$; 6-24 hours, $p=0.007$; 24-48 hours, $p=0.002$). The difference was most extreme immediately after cracking, with open cracked fruit losing 3.6 times more weight than control fruit in the first 6 hours. This is likely to have been due to leakage of aril juice from the wound, in addition to evaporative moisture loss. Fruit with open cracks continued to show increased weight loss later in storage, with rates double that of control fruit over the 6-24 hour period, and 67% greater than control fruit at 24-48 hours. The reduction in weight loss over time is likely to be due to the reduced size of the wound. Open cracks tend to decrease in size over time (Section 4.7.2), probably due to loss of aril turgor.

Closed cracks had a minimal effect on weight loss, with rates of loss not significantly different to the control. In fruit with closed cracks, the overall increase in weight loss compared to the control was around 15%, compared with 11% in impact treated fruit without visible cracks. It can therefore be estimated that closed cracks contributed only a 4% increase in weight loss. There was no significant difference between impact treated fruit with no cracks and those with closed cracks, showing that the closed crack effect was not significant. The minimal influence of closed cracks on weight loss confirms the importance of the endocarp as a barrier to water movement (Zhao *et al.* 1999).

Fruit treated by impact, but with no visible signs of cracking initially showed a significant increase in weight loss compared to the control (0-6 hours, t-test, p=0.025). In the 0-6 hour period, rates were 15% greater than the control. Later in postharvest storage, the impact treated fruit were not significantly different to controls, with 7 and 11% increases compared to the control at 6-24 and 24-48 hours, respectively. The decreasing margin between impact and control fruit over time, observed in a previous experiment (Section 2.10.2.2), was not as strongly evident in this trial.

2.11.3 Conclusions

Further replication would be required to draw solid conclusions, but it appeared that pepperspot levels had no significant effect on postharvest weight loss. Open cracks resulted in substantial increases in weight loss, giving further impetus to the avoidance of open pericarp cracking. In contrast, closed cracks had little effect on weight loss, confirming the importance of the endocarp as a barrier to water movement. In terms of moisture loss, closed cracks would be considered tolerable. If other factors are not limiting, this may allow for increased fall height limits, giving greater flexibility to the design of postharvest operations.

2.12 Effects of Vibration Damage on Postharvest Weight Loss

Trends observed in a previous experiment suggested that vibration injury could significantly increase the postharvest rate of moisture loss (Section 2.10.2.1). However, the differences were not sufficient to provide a clear statistical difference, with analysis of polynomial curve values required to detect a significant difference to control fruit. A slightly more severe level of damage was used to attempt to confirm the effect of vibration damage on weight loss during storage.

2.12.1 Methods and Materials

‘KMP’ fruit, from Sippy Downs in SE Queensland, were harvested directly into water, and soaked for approximately one hour. They were then dried and packed tightly in damp paper towel for transport to the laboratory. In a randomised control trial, fruit were treated either by vibration or as a control, with 40 fruit per treatment. Vibration was applied at setting 4 for 10 minutes using 18 fruit (around 370 g) in each run.

Fruit were weighed before and immediately after injury, and were stored at 57% RH and 20°C. The time of placement into storage, and times of weighing were recorded for each fruit, to allow the calculation of precise time values. Fruit were re-weighed after approximately 6, 24 and 48 hours. Fruit were assessed for the extent of vibration damage at 48 hours after injury. Scores for severity (Table 2.12.1) and an estimate of percentage surface area affected were noted for each injury. An index of vibration damage was calculated as a percentage of the maximum possible level of damage:

$$VI = \Sigma\,(\text{score x SA}) / (\text{score x SA})_{max} \text{ x } 100 \qquad \textbf{\textit{…Equation 2.12.1}}$$

Where VI = vibration damage index, SA = surface area of damage (%) and $(\text{score x SA})_{max} = 600$.

Table 2.12.1 Scores for lychee vibration injury.

Score	Description
0	No vibration damage
1	Very light
2	Light
3	Light-moderate
4	Moderate
5	Moderate-severe
6	Severe

2.12.2 Results and Discussion

Vibration treatment resulted in an immediate loss of weight averaging 0.05%, which was not included in the analyses. Subsequent loss of weight during storage was consistently higher in vibration injured fruit than controls (Fig. 2.12.1). Total loss was significantly higher at the 6 hour measurement, with vibration injured fruit losing 22% more weight than controls (t-test p=0.004). Measurements made at 24 and 48 hours showed increasing variability, and were not significantly different to the control (24 hours, t-test p=0.078; 48 hours, t-test on ln transformed data, p=0.087). However, the total amount of weight lost remained around 22% greater in injured fruit after 48 hours. The results confirm that vibration damage can result in significantly increased moisture loss, particularly in the hours following injury.

Rates of moisture loss were also significantly different only in the first six hours (t-test p=0.004) (Fig. 2.12.2). During the 6-24 and 24-48 hour periods, rates of weight loss in injured fruit were not significantly different to the control. Vibration injured fruit did not show a decreasing margin compared to the control, as previously observed

(Section 2.10.2.1*)*. This trend was thought to occur due to wound healing. The inconsistency of the response may be due to the greater severity of injury in this experiment, perhaps resulting in a different rate of healing. Genetic differences may also have contributed, as the experiments were carried out on different cultivars.

Analysis of correlations between vibration damage index and weight loss showed that increasing damage resulted in greater moisture loss (6 hours, Spearman correlation co-efficient=0.289, p=0.00913) (Fig. 2.12.3). The relationship weakened over time, but remained significant at 48 hours (co-efficient=0.220, p=0.0487). However, when control fruit (almost all scoring 0) were removed from the analysis, no significant correlation was detected during storage. The percentage of the surface area injured strongly affected weight loss (Vibration treated fruit only, at 6 hours, Spearman correlation co-efficient=0.564, p<0.0005), while the score for severity of damage was not significantly correlated with weight loss.

A strong correlation was detected between vibration index and the immediate loss of weight during injury (Spearman correlation co-efficient=0.645, p<0.0005). Increased overall damage results from greater loss of tissue and/or moisture during vibration. The surface area, rather than severity of the damage, contributed more to this relationship (Spearman correlation, %SA co-efficient=0.570, p<0.0005; Severity score co-efficient=0.320, p=0.0584). The plotted polynomial relationship of vibration index against immediate weight loss was not strong (R^2 = 0.467), but showed a general trend of increasing weight loss, particularly at vibration indices greater than 50 (Fig. 2.12.4).

2.12.3 Conclusions

The effect of vibration damage on weight loss was confirmed, with the vibration treatment resulting in a 22% increase in loss above the control. Postharvest weight loss was correlated with the extent of injury, with increased surface area of damage resulting in greater loss of weight. The results provide additional incentive to avoiding vibration injury, as the damage can cause loss of value and faster desiccation browning.

2.13 Effect of Mechanical Injury on Pathogen Establishment

The intact cuticle and epidermis provide an effective barrier to fungal growth, but can be compromised by injury. Damage to the fruit surface provides an entry point for pathogens and a ready supply of moisture, energy and nutrients for growth. In certain apple cultivars, impacts as minimal as 5 cm significantly increase the risk of rot development (Burton *et al.* 1987). Research was carried out to determine the effect of various types of mechanical injury on the establishment of pathogens on lychee fruit.

2.13.1 Methods and Materials

Fruit of cultivar ‘KMP’ were harvested from an orchard at Yandina. The fruit were harvested into water, and soaked for at least 30 minutes. It was anticipated that soaking all fruit in water would make the natural pathogen load more uniform. In a randomised control trial, 60 replicate fruit were subjected to each of 4 treatments:

- Impact treatment involved the application of 3 impacts from level 1.5, using a round head and no added weights. Each impact was equivalent to a 50 cm fall in energy applied per unit area. Impacts were applied to the cheek centre, the side of the fruit (90^o from the cheek), and the base;
- Abrasion was applied by ten 15 cm abrasions against a metal file, spread over the surface of the fruit;
- Vibration treatment was applied on setting 4 for 10 minutes. The dampening effect of the raised caravan floor resulted in this being a mild vibration injury;
- Control fruit were not mechanically injured.

After injury, the fruit were stored at 75% RH at 25^oC to encourage pathogen growth.

Prior to injury, widths and length of each fruit were measured using digital calipers to give an estimate of surface area. Fruit were also given a score for pepper spot (0 = none, 1 = very mild (around 5-25 spots), 2 = mild (~26-50 spots) and 3 = mild-moderate (~51-100)) to determine whether this had an effect on pathogen establishment.

After injury, any cracks caused by impact were recorded (length and width were measured, and the extent of penetration noted). Dark spot and rot development were recorded daily for a week, with perpendicular axes of each blemish measured. Area was calculated using the formula for an ellipse. Where rots were associated with a crack, or were localised to the stem end, these features were noted. On the seventh day of storage, in addition to other measurements, the features of each mycelial growth were described (colour, density and height of mycelial growth). For each fruit, a sigmoid relationship was plotted for the total surface area of rots and dark spots (as a percentage of fruit surface) over time, and this was used to calculate the time taken for certain rot areas to develop.

2.13.2 Results and Discussion

The impact treatment clearly resulted in a substantial increase in the rate of pathogen establishment, but other treatments did not significantly differ to the control (Fig. 2.13.1). Impact treated fruit showed a significantly greater total area of rots and dark spots than the control from days 2 to 6 after treatment (t-tests; days 2-4, $p<0.001$; day 5, $p=0.002$; day 6, $p=0.018$). This effect was primarily due to fruit with open cracks. Dark spots (developing rots without mycelial growth) were consistently greater in area in impact treated fruit than in control fruit (Fig. 2.13.2) (t-tests, $p<0.003$ for all days).

Late in storage the area of mycelial growth tended to be highest in control fruit (Fig. 2.13.3). Mycelial growth was significantly higher in control fruit than in impact treated fruit on day 7 (t-test, p=0.021), but no other significant differences were detected. Comparison of the different types of mycelia observed showed no significant differences between mechanical damage treatments and the control (data not shown).

Impact treated fruit showed a greater area of stem end rot than control fruit on day 5 (t-test, p=0.010), but later in storage this trend was reversed (t-test at day 6, p=0.014; day 7, p<0.001) (Fig. 2.13.4). Vibration and abrasion treatments did not significantly differ to the control in stem end rot development.

The significant differences between impact treated fruit and controls appeared to be primarily due to pericarp cracking. The removal of those rots associated with a crack from day 2-5 analyses resulted in no significant difference between control and impact treatments (Fig. 2.13.5). On day 6 and 7, rots often overlapped, making it difficult to determine association with the crack. Open cracking had a very strong effect on rot development, increasing the development of dark spots, but reducing stem end rots and mycelial growth (Fig. 2.13.6). The area of open cracking was strongly positively correlated with dark spots throughout the 7 day period of measurement (for impact and control treatments, Spearman correlation co-efficients >0.49, p-values <0.0005). For mycelial growth initial positive correlation from day 3 to 5 (for control and impact data, Spearman correlation co-efficients >0.33, p-values <0.005) was followed by negative correlation on day 7 (Spearman correlation co-efficient= -0.28, p=0.002). Stem end rots also showed a negative correlation with open crack area late in storage, on day 6 and 7 (Spearman correlation co-efficients = -0.24, -0.35 and p=0.008, <0.0005, respectively).

In contrast, when open cracks were removed from analysis, closed crack area showed no significant correlation with any index of rot development.

Fruit with closed cracks were not significantly different to the control in the development of mycelial growth or dark spots. In impact treated fruit with closed cracks, stem end rot area was significantly higher than in control fruit on day 5 (t-test, p=0.001).

However, by day 7 this trend had reversed, with control fruit showing more stem end rot (t-test, p=0.004). Impact treated fruit without cracks showed a similar trend, with more stem end rot than the control at day 5 (t-test, p=0.028), but less by day 7 (t-test, p=0.001). It would therefore appear that impact tends to reduce susceptibility to stem end rot late in storage. The mechanism underlying this effect may relate to the activation of defensive systems in response to damage. Chemical defences against pathogens can be released in response to a stress event, such as mechanical injury (Lyon *et al.* 1995). This response has been studied in other fruit, such as oranges (Stange *et al*. 1993a), where abrasion triggered the release on anti-fungal compounds in the peel. This theorised protective mechanism, triggered by impact injury in lychee, Pepperspot significantly affected postharvest rot development (Fig. 2.13.7), with appears to be effective against stem end rot, but not against the pathogens causing dark spot rots.

Pepperspot significantly affected postharvest rot development (Fig. 2.13.7), with strong positive correlations existing between pepperspot score and total rot area from day 5-7 (Spearman correlation co-efficients = 0.199, 0.409, 0.480, respectively, and all p-values <0.009). The pattern of increasing rot with greater pepperspot was shown in mycelial growth, stem end rot and dark spots, with significant positive correlations detected between days 5 and 7.

The pepperspot response appeared to relate to specific types of mycelial growth, with significant correlation between pepperspot scores and dense low-lying mycelia coloured white and pale green (Spearman correlation co-efficients for; green, 0.203; white, 0.206; both combined, 0.217; all p-values <0.007) (Fig. 2.13.8). Possible pathogens causing these types of mycelial growth include *Penicillium* sp. and *Colletotrichum* sp. (Fitzell and Coates 1995). In contrast, feathery and fluffy white mycelial growth were unaffected by pepperspot. These rots were possibly caused by *Rhizopus* sp., *Pestalstiopsis* sp. or *Alternaria* sp. (Fitzell and Coates 1995). Pepperspot, a pre-harvest disease caused by*Colletotrichum gloeosporioides* (Cooke and Coates 2002), has previously been considered a purely cosmetic disorder, not affecting shelf life (Drew 1999). These results show that it has some potential to affect postharvest storage, through faster development of rots. Mild to moderate pepperspot can reduce shelf life by around 1 day at room temperature, with significant effects occurring even at very mild levels (Fig. 2.13.9). It is possible that pepperspot causes a slight weakening of the natural protective barriers in the skin, increasing susceptibility to postharvest rot.

However, increased rot in pepperspot-affected fruit may occur because the same intrinsic fruit characteristics influence susceptibility to both types of disease. If it is pepperspot itself that increases rot invasion, these results increase the value and importance of pepperspot control.

2.13.3 Conclusions

Pericarp cracks exposing the aril caused rapid rot development, but closed cracks had no significant effect on rot development. Abrasion and vibration treatments did not differ significantly from the control, possibly due to low levels of damage. Impact tended to reduce the development of stem end rots.

Pepperspot, previously thought to be a purely cosmetic blemish (Drew 1999), caused a substantial increase in the establishment of postharvest rots. Low dense mycelial growth, white to pale green in colour, was encouraged by pepperspot, while feathery white mycelial growth was unaffected. It was hypothesized that pepperspot may weaken the defences of the pericarp, thus increasing rot susceptibility. Further studies to confirm these results would be required before firm conclusions are drawn. In particular, studies involving a greater range of pepperspot severity and observation of the association between pepperspot and postharvest rots would be valuable.

2.14 Survey of Wholesale Fruit Quality

A survey was carried out principally to determine whether mechanical injuries were a significant issue in the lychee industry, and if so, to identify the types of damage that were of major concern.

2.14.1 Methods and Materials

Three 5 kg boxes of fruit were purchased from the Brisbane Markets, Rocklea over a 2 week period. Boxes were randomly selected, and were transported by car to the laboratory in their original packaging. Details of the cultivar, region, grade and packaging were noted (Table 2.14.1). Fruit were stored overnight at RT prior to assessment. Each fruit was scored for maturity (green skin), desiccation browning, vibration damage and darkened tips, using a scale of 1 to 5 (1=no defect, 2=very mild, 3=mild, 4=moderate, 5=severe). Scores for darkened tips were based only on injuries appearing to be due to impact (occurring in a small area and blue-black in colour), excluding tip injury due to abrasion or vibration (pale brown colour or dusty appearance). Desiccation browning was distinguished by a dry light brown colour, affecting only the tips when mild. In contrast, discolouration that was more yellow in colour and powdery in texture was judged to be vibration injury.

Rots, cracks, scuffs and discoloured areas were measured by two perpendicular diameters. Scuffs were defined as distinct severe areas of vibration injury, affecting >80% of the measured surface area. Other defects, such as pulled stems, scale and insect feeding damage were recorded. Water content was measured in 10 randomly selected fruit from each box. These fruit were weighed, before and after oven drying at 60^{o}C for 4-5 weeks.

Table 2.14.1 Characteristics of purchased lychee cartons assessed in the survey.

Box	Date	Box depth	Liner	Cultivar	Region	Grade	Water content
1	4th Dec 2001	10.6 cm	Single plastic	Unlabelled - appear to be ‘Tai So’	Near Townsville, Qld	Not labelled	73.2%
2	11th Dec 2001	10.6 cm	Single crispy	‘Tai So’	Near Cairns, Qld	Class 1	76.4%
3	13th Dec 2001	8.4 cm	2 crispy perforated	Labelled ‘KMR’ – appear to be ‘Bengal’	Near Rockhamp -ton, Qld	Class 1	75.1%

All boxes were bought while the markets were open to the public, after commercial buyers had purchased fruit. The quality of fruit would thus be likely to be below average. In addition, for survey 2, the box purchased was the last box available at the markets, and the fruit were noticeably poor in quality.

2.14.2 Results and Discussion

Before the first box was opened, the issue of insufficient labeling was raised, with cultivar and grade not recorded on the carton. Incorrect labeling was also noted on box 3, labeled as ‘Kwai May Red’, but containing fruit that appeared to be of cultivar ‘Bengal’. The correct labeling of cartons to identify the contents by quality and cultivar is a basic requirement of marketing, and is required by law under the Trade Measurement Act, 1990. This issue may require some attention in the lychee industry.

Condensation was an obvious issue in box 1, but was not observed in the other boxes. The unperforated plastic liner allowed water to pool at the bottom of the carton. Around 11% of the fruit in box 1 were resting in the condensed water. This often caused a mild, water-stained discolouration of the pericarp. Fruit resting in water showed no increase in the levels or sizes of rots. Exposure to free water would be expected to increase rot development, but this effect may have been limited by time (condensation occurring late in the postharvest chain) or temperature (cool storage slowing pathogen growth).

2.14.2.1 Stung Fruit

Insect feeding damage, caused by fruit piercing moth, is a major quality issue, with affected fruit becoming quickly inedible. The small wound caused by insect feeding inevitably leads to infection, tissue browning, aril breakdown and a characteristic fermented smell (Fig. 2.14.1). In the wholesale survey, insect damage was a significant problem in box 2, affecting 5.5% of the fruit. The stung fruit were generally in a fermented state, detracting from the overall appeal of the carton. In addition, aril juice from some stung fruit had leaked onto sound fruit, causing discolouration of the skin. The occurrence of this level of damage suggests that greater vigilance is required in sorting for stung fruit. In the other boxes, less than 1% of fruit were affected, and the fermentation was at an earlier stage. The fact that all three boxes contained at least one stung fruit suggests that most growers could improve sorting for insect damage.

In practice, insect damage is minimised by the use of fine mesh netting, as there are no pesticides currently registered for control of fruit piercing moth on lychee crops. A high standard of sorting is crucial, as some damage can occur even in netted crops. Insect damage sustained the night before harvest is very difficult to detect visually. A stung fruit will exude aril juice from the wound when gently squeezed, but labour costs would preclude this check being applied to every fruit. The preferred practice would involve storing picked fruit overnight prior to sorting, to allow the symptoms to develop. Lychee quality guidelines suggest that any level of insect damage is unacceptable, so improved sorting or control is required to solve this ongoing problem.

Figure 2.14.1 Symptoms of insect feeding damage in lychee (stung fruit).

2.14.2.2 Fruit Shape, Size and Maturity

Guidelines for lychee packing suggest that all fruit within a box should be within 20% of the average size (Greer 1990). Undersized fruit were an issue in box 1, with around 10% of fruit weighing less than 15 g, and some fruit close to 11 g. The other boxes also contained some small fruit, with 1.4% and 2.8% of fruit weighing less than 15 g in boxes 2 and 3. The average fruit weight for all boxes was 20-21 g. Fruit weighing less than 15 g decreased the uniformity of the carton, reducing visual appeal.

Additional problems with uniform appearance included long stems, attached aborted fruit and joined fruit (Fig. 2.14.2). Stems exceeding 10 mm occurred on average in 0.7% fruit. FAO/ WHO standards (1994) suggest an upper limit of 2 mm stem length extending beyond the top of the fruit, while Greer (1990) suggests a 5 mm limit. Aborted fruit greater than 10 mm remained attached in an average of 0.3% of fruit. The incidence of joined pairs varied from over 7% in box 2, to 0.8% in box 3. While these characteristics detract from uniform presentation, they do not affect fruit quality.

Figure 2.14.2 Lychee long stem defect (A), normal stem length (B) and joined fruit (C).

Fruit maturity was scored based on the amount of green colouration in the skin (Fig. 2.14.3). Some lychee cultivars retain a slightly green colour at optimal harvest maturity, but ‘Tai So’ and ‘Bengal’ are fully coloured when at optimal flavour (Menzel and Simpson 1986). Maturity levels were poor in box 1, with 16% of fruit moderately to severely green. Only half the fruit in this box were fully coloured. In box 2 maturity was also an issue, with 7% of fruit moderately to severely green. Two-thirds of the fruit were fully coloured. The ‘Bengal’ fruit in box 3 showed very good colour, with 98% completely red, and 2% only mildly green. Immature fruit have poor visual appeal and eating quality, and are likely to reduce repeat sales. Maturity was shown to be a significant issue in two of the three boxes surveyed. Immature fruit would be expected to be a greater problem at the start of the lychee season, and immediately prior to Chinese New Year, with growers seeking to capitalise on inflated prices. This is likely to have been a contributing factor, as average lychee prices were high during the week when box 1 was purchased, and declined rapidly in the following week (Table 2.14.2). A fall in prices in early December is typical, due to increased supply of fruit (Menzel *et al.* 1999).

Figure 2.14.3 Score scale (1-5) for lychee maturity, based on green colouration.

Table 2.14.2 Lychee turnover and prices at Brisbane Markets in December, 2001.

Week commencing	Turnover (tonnes) of lychee	Average price of ‘Tai So’ carton
30/11/01	8.24	$42.50
7/12/01	24.68	$26.50
14/12/01	31.35	$22.50
21/12/01	35.51	$17.50

Source: Market Information Services

2.14.2.3 Pathogen Growth

Rots were a major issue in boxes 1 and 2, with at least 16 and 18% of fruit affected, respectively. Levels of rot were much lower in box 3, with 1.6% of fruit affected. Of the rots recorded, around 20-25% showed visible mycelial growth. The mycelial growth observed was generally a very small clumped area of superficial growth, with no darkening of the surrounding tissue. It has previously been observed that these types of rots spread very slowly, and do not appear to affect eating quality. In addition, around 4% of fruit in box 2 showed a distinctive scattered pale blue discolouration possibly resulting from mycelial growth. This type of blemish was not included in the general rot totals. It did not greatly detract from fruit appearance, and did not appear to damage the tissue.

The majority of rots observed were characterised as watery, dark brown and roughly circular, with no mycelial growth (Fig. 2.14.4). From previous experiments it was observed that these symptoms were characteristic of a developing rot. These types of rots are typically caused by fungi such *as Alternaria alternata*, *Colletotrichum* sp., *Rhizopus* sp. and *Phomopsis* sp. (Fitzell and Coates 1995). The rots develop quickly, and render the fruit inedible.

Figure 2.14.4 Typical lychee rot symptoms, a watery, dark brown blemish.

The estimates of rot levels are based only on blemishes showing distinctive symptoms of pathogen growth. Areas of discolouration that were small in size (<5 mm average diameter), coloured other than dark brown (e.g. medium brown or black), or irregularly shaped (a diameter ratio of 2 :1 or greater) were recorded separately. Many of these were likely to have also been developing rots. If all dark brown spots greater than 2 mm diameter are included as rots, the rot level increases to 28 and 22% in boxes 1 and 2 respectively. These levels of rot are completely unacceptable on the day after wholesale purchase, and suggest a major problem in the postharvest handling chain. Factors which could contribute to such a high level of pathogen growth are numerous, and could include high pathogen loads in the orchard, packing fruit wet or poor sorting. Fruit kept under constant cool storage would be unlikely to develop this level of fungal growth in normal transport time frames. It is therefore likely that poor temperature control is a major factor contributing to these high rot levels.

2.14.2.4 Desiccation Browning

On average, 71% of fruit showed some signs of desiccation browning at the wholesale level. The symptoms noted were generally very mild (score 2), requiring close observation, and not detracting greatly from the overall appearance of the fruit (Fig. 2.14.5). However, it is likely that further minor water loss (2-3% of weight) would result in moderate to severe browning in these fruit (score 4 to 5). An average of 11% of fruit already showed moderate to severe desiccation browning. Browning was a major problem in boxes one and two (16-17% moderately to severely brown), but was not an issue in box 3 (0.4%). The appearance of moderate to severe browning would suggest that weight loss of 8% or greater has occurred in these fruit. The results confirm the importance of desiccation browning in postharvest losses, even at the wholesale level.

Figure 2.14.5 Scale for lychee desiccation browning (score 1-5).

Unexpectedly, levels of browning were not well correlated with the water content measured for individual fruit, or for each box. The water content in box 3 was not significantly different to the other boxes, but this carton clearly showed the lowest levels of desiccation browning (Table 2.14.3). This may simply be a cultivar effect, as ‘Bengal’ fruit tend to be more resistant to desiccation browning than other cultivars (Section 5.2.2). Boxes 1 and 2, containing the same cultivar, were significantly different in water content, but showed very similar levels of browning. It would appear that water content is not a good index of postharvest water stress in lychee fruit. It is likely that the dry matter content of the aril dominates this measurement, eclipsing even major differences in pericarp moisture content. Aril dry matter content may vary due to genetic and environmental factors, and have little effect on the pericarp moisture content that drives skin browning. Water content measurements using only the pericarp would be recommended as a better index of water stress.

Table 2.14.3 Comparison of water content and browning scores in three lychee cartons.

Box	Water content (%)		Average browning score (higher = more brown)	
1	73.2	a	2.45	a
2	76.4	b	2.34	a
3	75.1	ab	1.60	b

Different letters show a significant difference within each column. Analysis of water content by ANOVA after reciprocal transformation, p=0.005 (N=10). Analysis of browning scores using ANOVA by ranks, p<0.001 (N>215)

2.14.2.5 Blemishes

Lesion browning is a pre-harvest disorder involving necrosis of the pericarp tissue (Joubert and van Lelyveld 1975). It is observed as a concise area of brown to black discolouration with a distinct darker edge (Fig. 2.14.6A). The affected tissue is characteristically brittle, and cracks under light pressure (Bagshaw *et al*. 1995). Small areas of lesion browning are acceptable, as the disorder does not affect eating quality. However, the affected area should not exceed general guidelines for blemishes (Class 1 fruit should not have blemishes exceeding 25 mm^2 (Greer 1990)). Lesions were observed in only 2 of the 728 fruit surveyed, both in box 2. The lesions were fairly large, at 31 and 79 mm^2, and these fruit should have been removed during sorting.

Blemishes not appearing to be caused by impact, vibration, pepper spot or rot were numerous, and many exceeded the lychee guidelines (Fig. 2.14.6). In boxes 1 and 2, around 39 and 52% of fruit respectively were affected by discoloured areas. Many of the blemishes exceeded the guidelines for Class 1 fruit, with 14% of fruit in box 1 and 41% in box 2 having blemishes greater than 0.25 cm^2. A large number also exceeded the blemish limit of 0.5 cm^2 for Class 2 fruit; 10% in box 1 and 32% in box 2. In box 3, only 5% of fruit were affected by blemish, and less than 2% of fruit showed blemishes of more than 0.25 cm^2. The levels of blemish in boxes 1 and 2 suggest poor sorting and grading practices.

Typical colours of blemishes in box 1 were black-brown, with dark brown the most common. Small spots (<2 mm diameter) and larger blemishes affected a fairly even number of fruit (24 and 29%). A small number of fruit (3%) showed mottled browning. In box 2, dark brown discolouration occurring only in the protuberance bases was very common, occurring in around 25% of fruit. Solid black blemishes were more common than brown, but were generally very small in size. There was also a small incidence of speckled black (differing from pepperspot in the elongated shape of spots), which occurred in small areas on around 3% of fruit. Mottling also occurred in around 3% of fruit. Black spots and dark brown bases were the most common types of blemishes in box 3, each affecting around 2% of fruit.

Figure 2.14.6 Lesion browning (A), and other lychee blemishes of unknown cause, including dark bases (B), solid blemishes (D, E, F, H), mottled (G) and spots (C, I).

Possible causes of these blemishes include pre-harvest damage resulting from sunburn, chemical burn, mechanical injury, water stress or extreme temperatures. Blemishes could also have resulted from water or aril juice resting on the fruit surface after harvest. Unidentified blemishes are a significant issue, occurring in a high percentage of fruit, and reducing the visual appeal of the carton. To prevent these blemishes, further research is required to determine the causes of the various types of discolouration observed.

2.14.2.6 Scale and Insects

Scale were a major problem in box 1, with around 17% of fruit having one or more scale on the surface. Around 5% of fruit carried more than ten scale, and some even had greater than forty. In box 3, there were six fruit carrying a single scale, with half of these being obviously dead. Box 2 contained no scale at all. Scale obviously remains a problem in some orchards. The level of scale infestation observed in box 1 shows that control in the orchard was poor, and that sorting for scale was not sufficient.

Box 1 contained 2 small (~2 mm length) live beetles, and 7 green ants (~6 mm length), one of which was alive. A small (~4 mm long) live caterpillar was found in box 2, while box 3 contained no insects. All insects were found within the liner bags.

FAO/WHO (1994) guidelines state that containers should be free from foreign matter. The presence of scale and insects does not detract from the eating quality of the fruit, but creates a poor impression of quality control, quarantine and packing shed hygiene.

2.14.2.7 Mechanical Damage

Cracking was evident in a small number of fruit in each carton, ranging from 0.8 to 1.9%. Only one fruit in the total (728 fruit) showed an open crack, which was severe (45 x 2.5 mm in size) and occurred in the upper layer of box 2. All other cracks were superficial, ranging from 3 x 0.1 mm to 26 x 1 mm, and averaging 10 x 0.4 mm. Only 25% of the cracked fruit were located in the bottom layer of the carton, suggesting that compression within the box was not the cause. Fruit were not packed tightly enough for lid compression to be an issue, and box failure can also be eliminated as a cause, as the boxes showed no signs of collapse or damage. The cracking observed was therefore likely to have occurred prior to packing. The general resilience of lychee fruit to compression cracking suggests impact as a more likely cause. The presence of one fruit with a widely gaping crack suggests either a poor level of sorting, or a severe impact event after the sorting process. The former is more likely, given that the majority of fruit in box 2 are not affected by any symptoms of impact injury. Losses due to open cracking may be significant during sorting, but an on-farm survey would be required to assess this issue.

Dark brown tips, appearing to be caused by impact injury, were not found to be a major problem. While an average 47% of fruit showed some signs of tip injury, the symptom was almost always very mild. An average of only 1% of fruit showed a moderate tip darkening injury, and no severe symptoms were observed.

A distinct scratch was observed in only one of the 728 fruit surveyed. The injury was a similar colour to abrasion or vibration damage, occurring in a thin line, 4 x 0.2 mm. The scratch only affected the pericarp surface, and had little effect on the overall appearance of the fruit. The very low incidence and minimal impact on quality shows that scratches are not an issue of concern in lychee.

In a small number of fruit, the stem was either pulled or missing. Pulled stems showed tearing of skin tissue, and this defect occurred in an average of 0.8% of fruit. The damage was very minor, either affecting only the pericarp, or showing a slight weeping of aril juice. In most cases, the stem was also removed. An additional small number of fruit had the stem missing, but the skin was not damaged. This type of injury did not appear to significantly affect fruit quality. Damage to the stem was not considered to be a major issue in the cartons surveyed, due to the small number of fruit affected, and the minimal impact on quality.

Puncture or indentation type injuries caused by stems resting against fruit under compressive load were not observed in the survey. Shape distortion due to compression was also absent.

Vibration damage was shown to be a very common problem at wholesale level. The majority of fruit showed some signs of injury, with an average of only 23% unaffected. In box 2, all fruit showed some signs of vibration injury, with 48% moderately to severely damaged. This severely detracted from the visual appeal of the carton (Fig. 2.14.7). The other two boxes showed much lower levels of damage, with 3-5% of fruit moderately-severely affected. The measured areas of scuff were also high in box 2, with an average 9% of the fruit surface area affected. Over 13% of the fruit in box

2 showed scuff to more than a quarter of the skin surface area, while 34% showed scuff to greater than 5% of the surface area.

Vibration injury of lychee fruit has not previously been recognised as a postharvest issue, and is not mentioned in postharvest reviews (Bagshaw *et al.* 1995; Chen *et al.* 2001; Holcroft and Mitcham 1996; Nip 1998; Ray 1998; Shi et al. 2001; Snowdon 1990; Underhill *et al.* 1997). Prior to this research, symptoms of vibration damage were probably mistakenly identified as desiccation browning. While the symptoms are similar, vibration injury can be identified by a yellowish, rather than pale brown colour and a dusty or powdery surface texture (Fig. 2.14.8). The discolouration tends to be localised to the upper protuberances, but this is also the case in early desiccation browning. The survey revealed that vibration is a key issue in lychee postharvest management, having a similar impact on fruit quality as desiccation browning.

Figure 2.14.7 Poor visual appeal due to lychee vibration injury and immaturity (Box 2).

Figure 2.14.8 Scuff injury (A), severe vibration damage (B) and severe desiccation browning (C) in lychee.

The analysis of vibration injury by depth within the box gives some indication of where damage has occurred in the postharvest chain. Severe levels of damage in the upper levels of the box would suggest damage after fruit have been packed into the box. Even levels of damage throughout the box would suggest damage prior to packing. Box 1 clearly showed a higher level of damage, particularly scuffs, in the upper levels (Fig. 2.14.9), suggesting that significant damage occurred after fruit were packed into the box. This damage is likely to have occurred during road transportation to the wholesale market. The fact that some damage exists in the bottom layers shows that minor damage may also have occurred prior to packing.

The other boxes did not show higher levels of damage in the upper layers, suggesting that damage has probably occurred prior to packing. It is also possible that fruit may have been repacked at the market, particularly in box 2, which was the last box available. Damage prior to packing may have been caused during on-farm transport (for example, from field to packing shed), or mechanical destalking. The levels of damage are much higher in these boxes compared to box 1, suggesting that pre-packing vibration injury (due to on-farm transport and destalking) may be generally more severe than post-packing (due to transport).

These results have important implications in the optimal management of the fruit. It is crucial that growers are aware of the potential for vibration damage, and are able to identify postharvest processes causing vibration. The damage can then be minimised by either avoiding the practice entirely (e.g. destalking by hand, rather than machine), reducing the severity of vibration (e.g. driving more slowly from field to packing shed) or managing conditions to reduce the resultant injury. Vibration damage occurs due to the motion of fruit, so the restriction of movement by tight filling or use of padding could also reduce damage. Vibration is currently causing a significant level of damage, but this could be greatly reduced by improved awareness of the issue.

2.14.3 Conclusions

The relative importance of various postharvest issues in the survey is summarised in Table 2.14.4. The issues are scored for their effect on eating quality and visual appeal, and the overall importance of the problem in this survey.

Most of the problems observed in the survey are well recognised as issues affecting lychee quality (Menzel *et al.* 2002). The most important quality issues were those that reduced fruit eating quality, including rots, insect feeding damage and immaturity. The extent of pathogen establishment at this stage of the postharvest chain was higher than expected. The number of fruit affected, and the severe impact of rot on fruit quality, renders this the most important problem in the survey. Control of rots requires good management throughout the handling chain, with temperature control particularly important. Condensation, observed in one box in the survey, also requires

Table 2.14.4 Relative importance of various issues in lychee postharvest quality.

Issue	Impact on eating quality	Impact on visual appeal	Degree of problem
Rots	5	5	5
Insect feeding damage	5	5	4
Immaturity / green skin	2-4	4	4
Desiccation browning	1	2-5	4
Vibration damage	1	2-5	4
Incorrect or insufficient labelling	1	1	4
Condensation	1	3	3
Scale	1	2-4	3
Other blemishes	1	2-4	3
Undersized fruit	1	3	3
Insects in carton	1	3	3
Cracking	1-5	2-5	2
Lesion browning (dark edges)	1	4	2
Stem pulled or missing	1	2	2
Tip darkening (impact)	1	2	2
Scratches	1	2	2

Lack of uniformity	1	2	2

Scores: 1=none, 2=very mild, 3=mild, 4=moderate, 5=severe.

good temperature control. Insect feeding damage is an acknowledged problem in the lychee industry, and was one of the most important quality issues in this survey. Ensuring that all stung fruit are detected requires a significant input of time and effort, but the high impact of this defect on the overall appeal of a carton makes thorough sorting crucial. The marketing of immature fruit is commonly observed as growers seek to capitalise on high prices, and is another predictable quality issue. These important and persistent issues can often be easily improved, but require some shifts in attitude.

While not detracting from eating quality, cosmetic defects can substantially reduce the visual appeal of the carton. Problems such as incorrect or insufficient labeling, insects and scale were not expected, but were significant issues. While not affecting fruit eating quality, these problems give a poor impression of packing shed management. A range of minor cosmetic issues were present, including undersized fruit, mild tip darkening, long stems, lesion browning, pulled or missing stems and attached aborted fruit. These defects generally occurred in very low levels of fruit. Cosmetic blemishes of unknown cause affected a larger number of fruit, and often exceeded the guidelines for size. Further research is needed to determine the causes of some blemishes. Desiccation browning is a well-recognised cosmetic defect in lychee, and this survey confirms that it remains a key problem.

The survey was carried out principally to determine whether mechanical injuries were a significant issue in the lychee industry. Injuries thought to be caused by impact were observed, but were generally mild and affected very few fruit. A single open crack showed that severe impact injuries can occur in practice. The wholesale survey revealed the unexpected importance of vibration injury in the postharvest management of lychee. Vibration has never previously been mentioned as a cause of postharvest browning in lychee, yet it appears to be as important as desiccation browning at the wholesale level, both in incidence and severity. It seems likely that discolouration due to vibration damage has previously been identified as desiccation browning. It has a similar appearance, but can be distinguished by a powdery, yellowish discolouration. In contrast, desiccation browning typically appears as dry, light brown discolouration. The appearance of distinct scuff marks on many fruit further confirms the importance of injury caused by vibration.

It appeared that vibration damage had occurred both before and after packing. This was revealed by the difference in injury levels between fruit located at the bottom and top of the boxes. The most probable locations of damage were during transfer from field to packing shed, mechanical destalking and transportation to wholesale markets. At wholesale level, the impact of vibration damage on the visual appeal of the fruit was equal to desiccation browning. Alterations to postharvest handling to prevent vibration damage may involve destalking by hand, tighter packing and slower vehicle speed. The survey provided an overview of the problems most crucial to the postharvest management of lychee, with vibration injury surprisingly revealed as a key issue.

2.15 Overview - Lychee Response to Postharvest Mechanical Damage

2.15.1 Damage Symptoms

The symptoms of impact, compression, vibration and abrasion damage on lychee were defined and quantitatively measured. Impacts from as little as 10 cm had the capacity to cause protuberance tip darkening (Section 2.1). This symptom became more severe with greater impact height, but detracted little from the overall visual appeal of the fruit. Impact caused significant changes in skin colour, including darkening and yellowing. The interior surface of the skin also showed discolouration after severe impacts. Pericarp cracking was observed as a result of severe impacts, and could affect either the surface skin tissues (closed cracking), or penetrate through the entire pericarp to the aril (open cracking). Closed cracks were often difficult to see, and detracted little from the visual appeal of the fruit, while open cracks rendered the fruit unmarketable. Where open cracks did not occur, the aril was unaffected by impact, with no obvious symptoms of bruising. The pericarp appears to bear the main force of the load, protecting the aril from damage. It seems likely that the bruising of lychee mentioned by Bagshaw *et al.* (1991) was either pericarp discolouration, or a blemish unrelated to mechanical damage.

Compression damage typically resulted in tip darkening similar to impact injury, with the aril unaffected (Section 2.2). Significant darkening and loss of skin chroma were observed within 4 hours of injury. Later in storage, compression injured fruit also showed significant yellowing of colour. Puncture due to compression was shown to be possible, but required a linear arrangement of stem, fruit and load, and was not observed in practice (Section 2.14). This contrasts with a previous report of lychee being susceptible to puncture (Lindsay and Cull 1986). The different response may have been due to cultivar differences. Cracking due to compression was rare, but could occur under extreme loads, and shape distortion was observed in some cultivars (Section 2.2).

Vibration was capable of causing a substantial decline in visual appeal (Section 2.3). Typical symptoms of vibration included scattered light brown or yellowish tip discolouration, concise scuff marks and a dusty dry appearance. Abrasion caused similar tip discolouration, but resulted in a much milder injury. Pericarp colour showed significant yellowing due to abrasion. Vibration caused very strong yellowing of colour, along with significant darkening and loss of chroma. Both the aril and the internal pericarp surface were unaffected by vibration and abrasion damage.

Vibration damage resulted in the formation of a scattered yellow residue on the skin surface, which was thought to cause the yellowish, dusty appearance of the injured fruit. It was hypothesised that this material resulted from dried cell contents of injured tissue. Comparison of the dissolved residue with diluted pericarp juice revealed consistent TSS and pH properties (Section 2.4). Lower absorbance in the residue compared to the extracted pericarp juice may have occurred due to greater metabolic breakdown of the pigments with exposure to air.

2.15.2 Structural Changes

SEM study of mechanically injured tissue confirmed substantial disruption of the tissue surface, even at low magnification (Section 2.5). Control fruit were generally intact, although some showed slight tip damage. Undamaged fruit showed ornate and variable patterns on the tissue surface, thought to result from complex formations of epicuticular wax (Pantastico 1975) or underlying tissues. Abrasion damage was observed as the severe shearing of many layers of tissue from the protuberance tip. Vibration resulted in a rough, powdery surface due to scattered accretions of tissue and possibly dried cell contents. Compression caused mild cracking, crumbling or distortion of tissue at the tip. Impact resulted in similar symptoms to compression, although generally more severe. Desiccated fruit showed a similar appearance to control fruit under SEM. Viewing under a dissecting microscope was suggested as a simple diagnostic tool to differentiate vibration and desiccation browning, due to the obvious changes in texture in vibration damaged fruit. Samples of fruit with silvering, a cosmetic pre-harvest blemish of lychee, consistently showed a very slight flattening of the protuberance tip, which was also observed in some vibration and compression samples. This provides some further evidence of a possible link between mechanical injury and silvering, as suggested by Bagshaw *et al.* (1995), but requires further study.

Epicuticular wax weight was not affected by mechanical injury, suggesting changes in arrangement, rather than actual loss of waxes (Section 2.6). Wax loss during storage was higher after vibration than impact, suggesting that structural changes due to injury may affect subsequent loss of wax. Impact using a flat head resulted in reduced cuticle weight. This response was possibly due to cuticle weakening causing tissue loss during storage of samples in solution. Injured fruit tended to show a gain in cuticle weight during storage, suggesting a possible wound healing response. At a similar period in storage, whole fruit tend to show declining rates of weight loss, providing further evidence of wound healing. Highly localised dark tissue, tightly bound to the cuticle, was observed at many injury sites.

2.15.3 Physiological Changes

Detection of biochemical changes in damaged tissue was hampered by large inherent variability (Section 2.7). Impact injury resulted in a significant decline in phenolics compared to the control, possibly related to browning reactions.

Increased ion leakage is a typical symptom of mechanical injury (Fiore *et al.* 1992), but was not observed in lychee. Open cracking caused a significant increase in electrical conductivity, but closed cracks, vibration damage and other mechanical injuries did not strongly affect ion leakage (Section 2.8). Despite leakage of aril juice, the ion leakage in open cracked fruit was minimal compared to rot damage. The tissue damage caused by mechanical injury may result from cellular disruption and structural changes, rather than cell rupture.

As previously observed in other fruits (Lougheed and Franklin 1974; MacLeod *et al.* 1976; Wade and Bain 1980), mechanical damage was capable of increasing carbon dioxide and ethylene release in lychee. Compression, vibration and 81 cm impact significantly increased CO_2 release, but a 60 cm impact caused no significant increase (Section 2.9). The increased CO_2 generation was not related to signs of visible damage, such as cracks and vibration injury. The response appeared to be primarily due to the decarboxylation of spilled malic acid (Hung 1993), rather than a sustained increase in respiration. Impact (both 60 and 81 cm) and vibration damage caused a substantial increase in ethylene release. This response was related to the extent of injury, with open cracks causing increased ethylene evolution. Closed cracks did not strongly affect ethylene release. In vibration damaged fruit, the ethylene response was dictated by the surface area of damage, but was not strongly affected by injury severity.

Mechanical damage caused a very slight loss of weight during treatment, but this was insignificant in the scale of postharvest losses (Section 2.10). Impact damage not causing visible pericarp cracking resulted in around a 30% increase in weight loss in the 24 hours following injury. This scale of loss could increase susceptibility to desiccation browning. Open pericarp cracks caused even more substantial weight loss, but closed cracks had little effect (Section 2.11). This confirms the importance of the endocarp as a barrier to water movement (Zhao *et al.* 1999). Vibration damage resulted in a significant, but less substantial increase in weight loss (Section 2.12). Moisture loss was correlated with the surface area of vibration injury, but not with severity. Late in storage the rate of loss in injured fruit declined, resulting in total weight loss similar to the control (Section 2.10).

In general, mechanical damage did not strongly affect pathogen growth, despite previous suggestions that dropping a lychee carton 50 cm would increase disease susceptibility (Johnson 1989). Susceptibility to rots was strongly increased by open cracking, but closed cracks did not significantly affect pathogen development. Fruit injured by impact without open cracks showed reduced incidence of stem end rot, suggesting a possible defense mechanism triggered by the injury. Low levels of abrasion and vibration damage did not affect rot susceptibility.

2.15.4 Wholesale Survey

A small survey of fruit at the wholesale level provided an overview of quality issues in the lychee industry. Longstanding problems such as insect feeding damage, desiccation browning, rots and fruit immaturity were confirmed as key issues in lychee quality (Menzel *et al.* 2002). Incorrect or insufficient labeling was an unexpected problem identified in the survey. Insects and scale on the fruit were common, giving a poor impression of packing shed hygiene. Minor cosmetic issues included lack of uniformity, undersized fruit, mild tip darkening, long stems, lesion browning, missing or pulled stems and attached aborted fruit. Cosmetic blemishes of unknown cause were common, and often exceeded the class guidelines. Further research into the causes of lychee skin blemishes would be valuable in identifying problems and improving quality.

Impact damage was observed in some fruit, with one open crack, and a low level of closed cracking recorded. Tip darkening was not a major issue, and no signs of compression damage were observed.

Vibration damage has not been previously mentioned as a problem in postharvest reviews (Bagshaw *et al.* 1995; Chen *et al.* 2001; Holcroft and Mitcham 1996; Shi *et al.* 2001; Underhill *et al.* 1997), but was identified as a key issue in lychee quality. The incidence and severity of vibration injury was similar to desiccation browning. It seems likely that vibration damage has been incorrectly identified as desiccation browning in the past, given the similarity of the symptoms. Vibration damage appeared to be an issue both before and after the fruit were packaged. Packaging and handling practices may need to be adapted to avoid the current high levels of vibration damage observed in this survey.

CHAPTER 3. LOAD PROPERTIES INFLUENCING THE FRUIT RESPONSE TO MECHANICAL INJURY

Load properties strongly influence the symptoms arising from mechanical injury, and the extent of damage. Major load properties affecting the fruit response include the manner of application, magnitude, duration and repetition. Properties of the contact surface are also important in determining the fruit response to injury. The characteristics of the load surface influencing fruit injury may include radius of curvature, size, elasticity, cushioning and abrasive properties.

Symptoms of vibration, impact, compression and abrasion may vary substantially due to different types of load application. The manner of application affects the tissue damage resulting from the load. Tissue damage is also influenced by load magnitude. The factors determining load magnitude vary with the type of mechanical forces involved. In impact events, the magnitude of the load may be influenced by fall height, initial velocity and fruit size. Under compression, load weight and duration are crucial. In abrasion loads, the sliding distance and force applied are important. Vibration damage is influenced by the duration and magnitude of acceleration. Factors such as vibration frequency, position within the container, tightness of fill and packaging properties influence acceleration. Repeated application further complicates the response of fruit to damage, often resulting in a different response to a single load event.

The combination of these various load properties, and the characteristics of the fruit, determine the type and amount of damage resulting from a mechanical event.

3.1 Effect of Load Magnitude on Impact Damage

The experiment was carried out to determine the thresholds of injury for impact damage on lychees and to give some general guideline as to the level of impact that can be tolerated by the fruit. Preliminary studies on ‘KMP’ fruit, without the use of the pendulum, suggested a threshold value for cracking in the range of 50 to 100 cm (Section 2.1.2.1). The definition of an impact threshold will provide a basis for the improved design of postharvest handling operations.

3.1.1 Methods and Materials

The experiment was carried out on mature ‘Tai So’ fruit from Sarina. Fruit were picked by hand, and stored in moist paper towel at 20^oC after harvest. Impacts were applied with the pendulum impacting device, using either a round or flat impacting head, both of 19.95 mm diameter. Using the round head, two additional weights of 26.5 g each were attached, but for the flat impacting head no added weights were used. For each impacting head, impacts were applied from levels 1, 1.5, 2, 2.5, 3 and 4. Each treatment was applied to 10 fruit, in a randomised control trial. Using the round head, only 3 fruit were treated by the level 4 impact due to the excessive severity of the injury. Estimated impact energy, and impact energy per unit area for each impact type are shown in Table 3.1.1. These values were calculated as previously described (Section 1.6.4.3.1).

Fruit were stored in open plastic containers in a single layer at 20^oC and 54% RH. The saturated salt solutions used in this experiment were not at optimal saturation, and RH is likely to have been higher than desired. A single slimline paper towel was placed in each container to absorb any leaking aril juice, and to attempt to reduce RH.

Table 3.1.1 Mechanical properties of the various types of impact applied to lychee.

Head type	Impact level	Arc length (cm)	Impact Energy (J)	Energy per unit area (Jcm^{-2})	Est. equivalent fall height (cm)
Round	1	14.1	0.04	0.038	42
	1.5	18.1	0.07	0.050	62
	2	22.2	0.11	0.063	81
	2.5	25.3	0.14	0.073	95
	3	28.9	0.19	0.085	110
	4	34.7	0.29	0.105	131
Flat	1	14.1	0.03	0.021	14
	1.5	18.1	0.06	0.029	24
	2	22.2	0.09	0.037	38
	2.5	25.3	0.13	0.043	51
	3	28.9	0.17	0.054	69
	4	34.7	0.25	0.082	104

Fruit were assessed immediately before and after treatment, and at 28 and 76 hours after treatment (for round head impacts) or 30 and 102 hours after treatment (flat head impacts). In severely injured fruit, readings at 76 or 102 hours could not be taken due to the development of rots.

A single colour reading was taken at the injury site, and the number of tips showing signs of darkening on the cheek centre was recorded. A small plastic ring of 17 mm diameter was used to keep the area of measurement constant. The width and length of any cracks were recorded immediately after treatment. For some fruit, colour readings were also taken on the internal surface of the pericarp at the injury site.

3.1.2 Results and Discussion

3.1.2.1 Tip Injury and Colour Change

Using the flat impacting head, the number of dark tips tended to increase with the level of stress applied (Fig. 3.1.1). Dark tips increased concurrently with contact surface area, levelling off at impacts above 0.043 Jcm^{-2}. Tip darkening was not observed immediately, but appeared to emerge within 30 hours of injury, with no significant changes in the number of injured tips from 30 to 102 hours.

Round head impact resulted in fewer dark tips than the flat head, but all levels showed a significant increase in the number of darkened tips at 28 hours compared to the control (Mann-Whitney rank sum tests $p<0.05$) (Fig. 3.1.2). As with the flat head, the number of injured tips increased with contact area. The number of darkened tips caused by round head impact did not change significantly from 28 to 76 hours.

Using the flat impacting head, impacts of up to 0.082 Jcm^{-2} caused no significant changes in colour compared to the control (data not shown), despite observed tip darkening. In contrast, the round impact head caused immediate significant changes in skin colour from the lowest impact setting (Fig. 3.1.3). Significant instant darkening occurred after impacts equivalent to 81, 110 and 131 cm (t-test p-values=0.034, 0.001 and 0.004, respectively). Immediate significant loss of chroma occurred in all round head impacts (t-test p-values<0.009), with loss tending to increase with impact energy. No immediate changes in hue were observed. Darkening and loss of chroma were very rapid, occurring within a minute of impact. This is much faster than a typical bruising response, with bruises taking at least 4 hours to appear (Hung 1993). These changes in colour may relate to structural or biochemical changes in the skin surface tissues.

Further changes in colour occurred during storage. All round head impacts except for the lowest level (42 cm) caused significant darkening of colour after 28 and 76 hours (t-test p-values<0.008). Significantly reduced chroma persisted throughout storage in all impact treatments of 62 cm and greater (t-test p-values<0.004). Impact equivalent to 42 cm did not cause significant chroma change at 28 hours (t-test p=0.097), but was significantly different to the control after 76 hours (t-test p=0.011).

An increase in hue, or yellowing of colour, was also observed in injured fruit during storage. Significant yellowing was observed in all impact levels, except 131 cm, after 28 hours (t-test p-values<0.05) and 76 hours (t-test p-values<0.001). Low replication prevented detection of an effect at impact equivalent to 131 cm. Colour change after impact probably occurs through similar processes to bruising, with the disruption of cell contents triggering enzymatic browning.

The scale of colour change after 28 hours tended to increase with fall height (Fig. 3.1.4). Similar patterns were observed after 76 hours, with changes in hue and chroma more extreme (Fig. 3.1.5). In impacts ranging from 42 to 131 cm the relationship between equivalent fall height and the extent of darkening and loss of chroma was roughly linear. However, over the full range of impact heights there is some appearance of a sigmoid shape. Changes in hue were also close to linear, with the exception of the 131 cm fall height (including this value, $R^2 = 0.389$, without it $R^2 = 0.920$). The 131 cm result was measured using a sample size of only 3, giving a large degree of error and a possibly unrepresentative result.

Colour changes on the interior surface of the pericarp were also observed in fruit injured by impact (Fig. 3.1.6). A strong change in hue towards yellow was observed, with impacts equivalent to 42 cm and greater causing significant and strong discolouration (t-test p-values<0.001). Some significant darkening was observed, at 62 cm impact height (t-test p=0.007). A slight rise in chroma (not significant) is probably simply due to the development of colour on the pale tissue. The development of discolouration on the fruit endocarp supports the hypothesis that enzymatic browning reactions within the tissue are responsible for the changes in skin colour after impact.

3.1.2.2 Crack Thresholds

Using the round impact head, cracking first occurred at an impact force of 0.050 Jcm^{-2}, equivalent to a 62 cm fall height. Forty percent of fruit showed signs of cracking at this impact level (Table 3.1.2). At greater impact forces, a greater percentage of fruit showed cracking, crack dimensions were larger and total crack area increased. Using the flat impacting head, only the maximum impact of 0.082 Jcm^{-2} caused pericarp cracking. A flat head impact equivalent to a 69 cm fall did not cause any cracking. These results indicate that the threshold for pericarp cracking is in the vicinity of 70 cm for the fruit tested.

Table 3.1.2 Lychee pericarp cracking under various levels of impact (N=3-10).

Impact level	Head type	Impact energy (Jcm^{-2})	Equivalent fall height (cm)	% of fruit showing cracks	Avg crack values, when they occurred		Avg crack area per fruit (mm^2)
					Length (mm)	Width (mm)	
0	Round	0	0	0	-	-	0
1	Round	0.038	42	0	-	-	0
1.5	Round	0.050	62	40	3.8	0.1	0.15
2	Round	0.063	81	80	4.8	0.2	2.1
2.5	Round	0.073	95	80	13.6	0.6	10.5
3	Round	0.085	110	100	17.3	0.6	29.2
4	Round	0.105	131	100	39.7	1.3	50.9
3	Flat	0.054	69	0	-	-	0
4	Flat	0.082	104	50	30.6	1.6	26.3

The relationship between the equivalent fall height and the area of cracking was found to fit a sigmoid equation (R^2=0.99) (Fig. 3.1.7).

$$C = 53.9 / (1 + \exp (- (h - 106.6) / 8.9)) \qquad \textit{\textbf{....Equation 3.1.1}}$$

Where, C = crack area (mm^2) and h = equivalent fall height (cm).

The sigmoid relationship (Equation 3.1.1) predicts a crack area of 1 mm^2 occurring at around a 71 cm fall, doubling to 2 mm^2 at a 78 cm fall.

3.1.2.3 Rot Development

As would be expected, rot measurements made on the impact treated fruit were highly correlated with crack area (Spearman correlation co-efficient 0.939, $p<0.0005$), with higher impact levels showing increased development of rots (Fig. 3.1.8). The correlation remained strong even when the large number of fruit with both no cracks and no rots were removed from the analysis (Spearman correlation co-efficient 0.784, $p<0.0005$), confirming that greater crack area results in increased rot area. The relationship between rot area and estimated fall height showed a very strong fit to a sigmoid equation ($R^2 = 0.99$).

$$Rt = 274.1 / (1 + \exp(- (h - 100.7) / 10.7)) \qquad \textbf{\textit{....Equation 3.1.2}}$$

Where Rt = rot area (mm^2) and h = equivalent fall height (cm).

Rot area showed a hyperbolic response to increasing crack area ($R^2 = 0.98$) (Fig. 3.1.9).

$$Rt = 432 * C / (35 + C) \qquad \textbf{\textit{....Equation 3.1.3}}$$

Where Rt = rot area (mm^2) and C = crack area (mm^2).

Small cracks resulted in rot areas vastly exceeding the crack size. For example, the level 2 impact (using a round head) resulted in an average crack area of around 2 mm^2, with resultant rots totaling 54 mm^2, around 25 times greater. In contrast, at level 4 impact the rot area was around 5 times greater than crack area. These results highlight the importance of preventing even small cracks.

3.1.2.4 Differences Between the Impact Heads

Both round and flat impact heads caused tip injury, but significant changes in skin colour were detected only using the round head. Force is concentrated into a smaller area using the round head due to the reduced area of contact between fruit and head. This reduced area of contact was taken into account in calculations (Section 1.6.4.3.1), but could not fully explain the differences in fruit response. The flat head impact from level 4 exerted energy of 0.082 Jcm^{-2}, but resulted in no significant colour change, while a round head impact of 0.073 Jcm^{-2} resulted in a strong change in colour. In addition, comparison of the contact surface area and the number of injured tips showed that the round head consistently injured a lower percentage of the tips in contact with the head. On average 41% of tips in the round head contact area were injured, as compared to 66% using the flat head (Table 3.1.3).

These observations may be explained by considering the process of the impact in slow motion. The centre of the head initially comes into contact with the curved fruit surface, and from the moment of contact, energy is being dissipated into the tissue. The force behind the head continues to push it into the tissue, but as the impact proceeds, the energy is reduced. Thus the tissue at the centre of the impact is subjected to a greater intensity and longer duration of force. In the case of the rounded head, this process would be exaggerated by the fact that the head edges would take longer to come into contact with the tissue. In addition, further away from the centre of the round head, the impacting surface becomes more oblique to the direction of the force. These factors concentrate the load into an even smaller area, resulting in greater damage to a smaller number of protuberances.

Table 3.1.3 Effect of impacting head type on lychee protuberance tip injury.

Impact Level	Head Type	Contact SA (mm^2)	No. tips in contact	No. tips injured	% of tips injured

1	Round	102	11.2	4.8	43
1.5	Round	135	14.8	6.6	45
2	Round	169	18.6	6.8	37
2.5	Round	195	21.5	8.9	41
3	Round	227	24.9	11	44
4	Round	278	30.5	12	39
1	Flat	158	17.4	13	74
1.5	Flat	206	22.7	15	67
2	Flat	255	28	16	58
2.5	Flat	292	32.1	21	65
3	Flat	313	34.4	22	64
4	Flat	313	34.4	24	70

The average crack areas occurring under round and flat heads appear fairly consistent, following a sigmoid relationship (Fig. 3.1.7). However, the level 4 flat head impact, with equivalent fall height of 104 cm, caused injury to fewer fruit, but resulted in larger cracks, than the round head impacts equivalent to 81 and 95 cm (Table 3.1.2). In addition, the flat head impact equivalent to 69 cm caused no cracking, while a round head impact equivalent to 62 cm caused minor cracking to 40% of treated fruit. These differences illustrate the complexity of the biomechanics involved when using different shaped impacting heads.

3.1.3 Conclusions

The results show no definite threshold value for colour changes due to impact, but suggest that it is a gradual process, with increased fall heights causing more damage. Injuries ranged from slight tip discolouration under a minor impact, through to severe darkening covering around 1/3 of each protuberance in major falls. Significant changes in colour were not detected using the flat impacting head, while all round head impact levels caused some significant change in colour. Larger sample sizes may have revealed significant changes in colour under the flat head impacts, as injury to the tissue was visible in the form of darkened protuberance tips. However, this is largely immaterial, as tip darkening is unlikely to cause any economic loss unless severe.

In contrast, cracking injury can cause direct losses due to reduced visual appeal, and may also result in further losses due to pathogen invasion. Cracking under impact showed a threshold estimated at around 70 cm. It is possible that this value could vary substantially due to factors such as fruit condition and cultivar, but the results provide a useful ballpark figure for the tolerable drop height of lychee fruit.

3.2 Effects of Vibration Frequency and Duration on Extent of Injury

Vibration frequency and the duration of application are the key load variables influencing damage. These factors influence the acceleration of fruit, and thus the severity of damage. The effects of various vibration frequencies and duration were studied in lychee, with a view to managing these variables to minimise damage.

3.2.1 Methods and Materials

The experiment was carried out on mature ‘Tai So’ fruit harvested in Sarina. Fruit were stored in moist paper towel at 20^{o}C after harvest. Vibration treatments were applied by placing fruit into a small cardboard box tightly strapped onto a flask shaker. Treatments of vibration speed at three levels (2, 3 or 4) were combined with duration at two levels (5 or 10 minutes) in a factorial design. Control fruit were not subjected to any mechanical damage. In a randomised control trial, each treatment was applied to 2 replicates of 10 fruit. To ensure a reasonably tight fill, 22 fruit were placed into the box during each treatment. The additional fruit were re-used across treatments, but were replaced when they became severely injured. After treatment, fruit were stored in open plastic containers at 20^{o}C and 33% RH.

Measurements of damage were made immediately before and after treatment, and then at either 24 or 48 hours. Measurements taken included:

- Single colour readings at the cheek centre;
- Number of yellowish scuff marks on the whole fruit;
- An estimate of the % SA of the whole fruit affected by yellowish discolouration;
- Number of protuberances with > 1/3 yellowing on cheek centre.

A small plastic ring was used to keep the area of measurement constant for measurement of protuberance yellowing, and the total number of protuberances within the ring was recorded for each site.

3.2.2 Analysis

While measurements were made on 20 individual fruit for each treatment, analysis could be carried out only on replicate boxes, resulting in only 2 values per treatment. Factors such as tightness of fill and fruit arrangement within the box resulted in a large degree of variation between replicates. Uneven variation across samples prevented parametric analysis, and no significant treatment effects were detected by non-parametric tests. Standard error values were also large due to the small sample size and high degree of variation. Hence, no statistical analyses are provided with the data, but the general trends are discussed.

3.2.3 Results and Discussion

Vibration injury was observed both as a general yellowing of fruit colour, and as concise localised scuff marks. The indices used to estimate the extent of damage in this experiment included change in hue and protuberance yellowing at a single location, and the number of scuff marks on the whole fruit. It was found that measurements made in a single location were insufficient in detecting vibrational injury, as damage occurred randomly over the fruit surface. A single location measurement was often not representative of the condition of the entire fruit.

The results were suggestive of an increase in damage due to longer periods of vibration, but this appeared to occur only at higher levels of vibration frequency. An increase in duration from 5 to 10 minutes had little effect on the change in hue at setting

2 (Fig. 3.2.1). At higher levels of vibration frequency, the change in hue was more than three times greater under the 10 minute vibration, than the 5 minute treatment. Similar results were also reflected in estimates of the percentage of the skin area affected by yellowing (Fig. 3.2.2). The level 2 treatment showed a minimal increase in damage from 5 to 10 minutes. At level 3 and 4 the 10 minute duration again showed damage around three times greater than the 5 minute treatment. Similar trends were also shown in the number of average number of scuff marks on each fruit (Fig. 3.2.3). These results are suggestive of an interaction between the effect of vibration duration and frequency on lychee injury. Increased duration appears to have little effect at low vibration frequency, but causes an escalation of damage at higher frequency.

The doubling of vibration duration appears to cause more than a three-fold increase in injury levels. The mechanism of this duration effect may relate to the settling of fruit during a vibration treatment. Initially the fruit may roll about more within the box, eventually settling down into a rhythmic pattern of movement. Injuries caused by vibration generally occur due to repetitive forces acting on the tissue (Sitkei 1986). If the process of settling takes around 2.5 minutes, the 5 minute treatment would result in 2.5 minutes of repetitive movement, while the 10 minute vibration would result in 7.5 minutes. This could potentially explain the three-fold increase in damage caused by doubling the duration.

The change in hue occurring due to vibration was detected immediately after injury, but further change also occurred during storage (Fig. 3.2.1). It was estimated that around 2/3 of the total colour change occurred within a few minutes of injury, with the remaining 1/3 developing over the following 24 hours. Yellowing of colour during storage may result from drying of leaked cell contents on the fruit surface.

Increasing vibration frequency appeared to strongly increase damage (Figures 3.2.1, 3.2.2 and 3.2.3). Taking into account changes in the number of yellow protuberances, the percentage surface area yellowing and number of scuff marks, on average the change from level 2 to 3 caused a 312% increase in damage, while from 3 to 4 caused a 114% increase. A repeated experiment with a larger number of frequency levels, and increased replication would be needed to clarify the relationship between frequency and resultant damage.

3.2.4 Conclusions

It appeared that increases in both duration and frequency of vibration may raise the level of fruit damage. An interaction between duration and frequency was suggested, with time having a greater effect at higher frequency. The interaction of duration and frequency prevented the estimation of a simple threshold value for frequency. While trends within the data were suggestive, few solid conclusions could be drawn due to a lack of sufficient replication.

3.3 Effect of Compression Duration and Load Magnitude on Extent of Injury

Compression damage is caused by the application of a static force, generally resulting from the weight of stacked fruit above. Despite a lack of any published research, it has been implied that lychee are susceptible to compression. Batten and Loebel (1984) suggest a limit of 30 cm depth in field containers to avoid “squashed fruit”, and Greer (1990) recommends a maximum of 50 cm stacking depth. A trial was carried out to determine the threshold of compression damage in lychee. The tolerance of lychee to compression injury will influence the design of postharvest operations, particularly stacking depth.

3.3.1 Methods and Materials

‘KMP’ fruit were harvested from Byron Bay, in northern NSW, and were immediately soaked in water for at least one hour. The fruit were dried using tissue, and packed into damp paper towel over-wrapped with plastic. Fruit were transported to the laboratory by car, and were stored in damp paper towel at RT for 2 days prior to treatment. Fruit not treated on the first day were subsequently stored at 10^oC in damp paper towel. Treatments were applied over 5 days, with water potential measured in 2 to 3 fruit each day. All fruit were soaked in water for 2 hours prior to compression to attempt to increase hydration.

Treatments were applied using the compression device described in Section 1.6.4.5.1. The experiment was designed as a factorial, with various levels of weight applied (Control, 0.5 kg, 1 kg and 1.5 kg) for either 24 or 48 hours. In a randomised control trial, each factorial treatment was applied randomly to 10 fruit. The control fruit were placed in the same conditions as treated fruit, with a MDF disc resting on the fruit, but with no weight applied. A loose plastic cover was placed over the compression set-up, and shallow dishes of NaCl were placed within, to attempt to adjust RH to 75%.

Displacement of the tissue was estimated by measuring the height of the MDF disc, immediately before and after the addition and removal of weight. Symptoms of compression damage, including distortion of shape, protuberance tip darkening and cracking, were recorded after removal of the weight. The severity of distortion was scored from 0 to 5 (none, very slight, slight, moderate, severe or very severe). The number of tips at each level of severity score (0 to 5, as described for distortion) were recorded for protuberance tip injury. An index of tip injury was calculated as the sum of severity scores multiplied by number of tips affected. Cracks were measured by length and width, and the penetration (open or closed) was recorded. Fruit were re-assessed for cracks 24 hours after removal to ensure that none were overlooked.

3.3.2 Results and Discussion

Fruit water potential ranged from –0.15 to –0.4 MPa, and averaged –0.23 MPa, confirming that fruit were very well hydrated. Values did not change substantially over the 5 day period of treatment. The fruit within the device appeared to dry out very quickly, despite the use of saturated salts and plastic wraps to raise the humidity. This was possibly due to the ability of MDF to draw moisture from the fruit. In subsequent experiments, fruit were wrapped in individual plastic bags to avoid this effect.

Cracks occurred only under the 1.5 kg treatment, showing that lychee fruit can tolerate 1 kg compression for up to 48 hours without cracking. Under 1.5 kg weight, 20% of fruit showed signs of cracking, at both 24 and 48 hours. After 24 hours compression, the resultant cracks were all closed (affecting only superficial tissues), and averaged 13 x 0.7 mm. Compression using 1.5 kg for 48 hours resulted in severe open cracking (25 x 7 mm) in one fruit, and a small weeping crack in another.

Distortion of shape occurred in some fruit subjected to 1 and 1.5 kg weights, and appeared to increase both with weight and duration. After 24 hours, the 1 kg treatment did not cause any visible distortion, but at 48 hours 40% of fruit were slightly or very slightly misshapen. The 1.5 kg treatment resulted in distortion in 40% of fruit at 24 hours, again only slight or very slight. After 48 hours, the 1.5 kg treatment resulted in a total of 80% of fruit showing distortion, including moderate (20% of fruit) and severe (10%) levels of damage in some fruit. Levels of distortion were significantly increased by greater weight (comparing 1 kg and 1.5 kg at 48 hours, Mann-Whitney rank sum test, $p=0.049$) and duration of compression (1.5 kg treatment at 24 and 48 hrs, t-test on ln transformed data, $p=0.049$). In terms of distortion, 1 kg weight applied for up to 48 hours would be considered tolerable, as slight or very slight levels of distortion would be unlikely to have a commercial impact.

Tip injury tended to increase with load (Fig. 3.3.1), with 1.5 kg causing significantly greater tip injury than 500 g or 1 kg loads at 24 hours (t-tests $p<0.01$). Duration of compression showed no significant effect on tip injury, but the index of tip darkening tended to increase by around 50% when the time was doubled. Compression duration affects the extent of injury through the creep phenomenon, with fruit tissue continuing to deform under a constant load (Sitkei 1986). Tip damage generally affected only a few protuberances, and did not impact on the overall visual appeal of the fruit.

Immediate displacement significantly increased with weight, with the 1 kg and

1.5 kg weights resulting in greater deformation than 0.5 kg (t-tests, $p<0.001$) (Fig. 3.3.2). However, the 1.5 kg treatment did not significantly differ from the 1 kg load in initial displacement of tissue. The first 500 g placed on the fruit caused an average of 2.7 mm displacement, a further 500 g resulted in an additional 3.8 mm shift, while the final 500 g caused only another 1.3 mm. The high level of displacement caused by an increase from 0.5 to 1 kg could be a sign of greater tissue failure in this weight range.

The total displacement was significantly affected by mass (one-way ANOVAs at 24 and 48 hrs $p<0.001$), but not by duration. The rate of deformation tended to decrease over time (Fig. 3.3.2), explaining the lack of a significant duration effect. The decline of deformation rates over time is typical in compression (Sitkei 1986). Structural changes in the tissue that can occur easily (such as the collapse of large micro-pores) generally occur early in the compression process, and further compaction of the tissue becomes more difficult. Residual deformation in lychee was affected by compression weight, with all three treatments significantly different at 24 hours (ANOVA, $p<0.001$, comparison by Tukey test). As is typical, increasing load resulted in greater residual deformation. After 48 hours, the large variation in the 1.5 kg treatment prevented the detection of a significant difference between residual deformation caused by 1.5 and 1 kg weights.

Fruit subjected to 1 kg load for up to 48 hours show minimal loss of quality, so this could be considered a threshold of damage. Acceptable stacking depths can be calculated from the tolerable load, based on the assumption of a rhombic stacking pattern, as described by Mohsenin (1986).

$$S_P = L_\theta \times \sin\theta \qquad \text{...Equation 3.3.1}$$

Where, S_P = permissible stacking height (m), L_θ = the average length of the four axes of fruit resting above (m) and $\theta =$ the angle between a horizontal plane and the line of fruit axes (adapted from Mohsenin 1986).

The average length of the axes was calculated by;

$L_\theta = F \times d / M$ ***…Equation 3.3.2***

Where, F = total permissible compressive load (kg), d = fruit diameter (m) and M = fruit mass (kg) (adapted from Mohsenin 1986).

The stacking angle was determined by solving for the equation;

$D = 1 / (4d^3 \times \cos^2 \theta \sin \theta)$ ***…Equation 3.3.3***

Where, D = number of fruit per cubic metre (Mohsenin 1986).

The number of fruit per cubic metre was estimated based on the dimensions of a 5 kg lychee carton.

The permissible stacking depth is affected by fruit size, due to the effect on stacking angles. For example using data on 'KMP' weight : diameter relationships, 20 g fruit can be stacked to 137 cm, 25 g fruit to 116 cm and 30 g fruit to 102 cm. Stacking depths also vary slightly with the weight : diameter relationship, and therefore with cultivar. For 20 g fruit size, a 1 kg compressive load is applied at 135 cm stacking depth for 'Salathiel' and 'Wai Chee', but at 150 cm for 'Bengal' fruit. Assuming an upper limit of 500 g load was desired, the permissible stacking height would range from around 50 to 75 cm. These figures suggest that the stacking depths previously recommended (Batten and Loebel 1984; Greer 1990) have been fairly conservative. Fruit are generally packaged in cartons around 9 cm deep, and are often transported on farm in shallow crates. Deeper cartons and storage bins, which can reduce vibration injury (Sitkei

1986), may be more suitable for lychee given the fruit’s tolerance of compression and susceptibility to vibration (Section 2.14.2.7).

3.3.3 Conclusions

In general, fruit were shown to tolerate a 1 kg static load for up to 48 hours with minimal loss of quality, so this could be considered a threshold of damage. This translated into a stacking depth of at least one metre. An upper limit of 500 g compression weight would ensure a high level of fruit quality, and allows for fruit to be stacked to at least 50 cm. For shorter periods, greater loads would be tolerable. The resistance of lychee to compression injury may permit the use of deeper cartons, to reduce vibration injury. The threshold level for compression injury has been calculated based on static loads, and could vary due to vibration or jolting during transport. Other factors, such as hydration and cultivar may also affect thresholds of damage. Further research would therefore be needed to confirm the benefits of greater stacking depth in practice.

3.4 Effect of Impact Site and Method of Application on Pericarp Cracking

Pendulum devices are commonly used to apply impact injuries in experimental research, as they provide a uniform, replicable treatment (Hyde 1999). Unlike simple dropping, the pendulum eliminates the effect of fruit weight on impact energy, and allows the injury to be applied to a chosen, uniform site. However, the use of a pendulum may result in unreliable data due to differences in the load application. During impact, the contact area between impacting surface and tissue is limited to the SA of impact head. In heavy impacts, the limited SA would increase the energy per unit area applied to the tissue. The pendulum device uses a clamp opposite the impact site to hold the fruit stationary, applying a slight compressive force. The fruit is therefore under slight pressure before the impact is applied. Clamping results in a different type of loading compared to a simple drop test. Part of the load would be transferred onto the opposite side of the fruit, as it is pushed back into the clamp during impact. This load would be minimised by the large SA of the clamp, a concave surface, but may still affect the cracking response.

The use of a uniform site of impact may also create results substantially different to real impact events. During postharvest operations, fruit would be subjected to impact at random locations on the fruit surface, and these different sites may vary in their susceptibility to cracking. In most previous experiments in this thesis, single impacts have been applied to the cheek. The experiment was carried out to explore the differences between pendulum and drop impacts, and to examine the effect of impact site on cracking response.

3.4.1 Methods and Materials

Fruit of cultivar ‘Tai So’ were purchased at Brisbane Markets, and were stored wrapped over with damp paper towel at RT for 2 days prior to treatment. The experiment was of factorial design, with two application methods (pendulum and dropping) and 4 impact sites (cheek, side, base and shoulder), resulting in a total of 8 treatments. The sites, as defined in this experiment (stem, shoulder, side, cheek and base), are illustrated in Figure 3.4.1.

Figure 3.4.1 Impact sites on the lychee fruit surface.

Pendulum fruit were subjected to impact from level 4 using a flat head, with 4 added weights totaling 107 g attached. The pendulum impact was estimated to be equivalent to a 141 cm fall. Using the pendulum, site treatments were randomly applied to a total of 20 fruit each. Fruit treated by dropping were allowed to fall onto a flat hard surface (a sheet of thick glass) from a height of 141 cm, and were caught on the first bounce. The impact surface was lightly dusted with talcum powder to allow the impact site to be easily observed. The impact site could not be controlled in dropped fruit, and this resulted in slightly uneven replication, ranging from 18-21 fruit.

Resultant cracks were measured by length and width, and were recorded as either open (penetrating to the aril) or closed. The fracture pattern of each fruit was recorded by location (for example, from base to cheek) and direction in relation to the impact site (radial, oblique or neither). The nearest distance from the impact site to the fractured tissue was also recorded for each crack.

3.4.2 Results and Discussion

Dropped lychees most commonly landed either on the cheek (33% of fruit) or shoulder (33%), with fewer fruit landing on the base (18%) or side (16%). Cheeks and sides were of a similar large SA (each ~35% of total fruit), while the shoulders and bases covered much less area (~12% each). The high level of shoulder impacts, and low number of side impacts show that the impact site is not entirely random when fruit are dropped. Weighting of tissue within the fruit, or other aerodynamic properties may affect the way the fruit falls, resulting in a predominance of shoulder and cheek impacts. The ratio of impact sites would probably vary between cultivars, due to different fruit morphology. Fall height may also have a substantial effect on impact sites.

3.4.2.1 Effect of Application Method

The method of application significantly affected crack area, with the pendulum resulting in significantly increased cracking when impacts were applied to the cheek or shoulder (Mann-Whitney rank sum tests, $p=0.003$ and $p=0.038$, respectively) (Fig. 3.4.2). In impacts involving the base or side, application by pendulum and dropping gave similar results. In impacts to the cheek, the pendulum resulted in larger crack size (Mann-Whitney rank sum test $p=0.003$) (Fig. 3.4.3), but a similar percentage of fruit showed cracking under both methods (Fig. 3.4.4). Excluding fruit without cracks, the pendulum cracks after cheek impact averaged 57.8 mm^2, while dropped fruit averaged 22.9 mm^2. Larger cracks due to the pendulum method may have resulted from the light compression applied by the clamp, which could force the edges of cracks apart.

The difference between application methods in shoulder impacts resulted from a higher percentage of fruit affected by cracking due to pendulum impact (60%) compared to dropping (29%) (Fig. 3.4.4). This may have been due to impacts being located precisely on the sharp curve of the shoulder in the pendulum, giving a smaller angle of curvature, and hence greater energy per unit of area. In contrast, drop impacts would have been randomly spread over the shoulder area, giving a larger average angle of curvature.

3.4.2.2 Effect of Impact Site

Trends in crack area across different sites were similar in pendulum and dropped fruit, with cheek and side impacts showing the highest average crack area (Fig. 3.4.2). In fruit treated by dropping, only the cheek and shoulder sites were significantly different (ANOVA by ranks, p=0.032, comparison by Dunn's Method). The base impacts showed the lowest crack area in pendulum treated fruit, with both cheek and side impacts significantly different to base (Mann-Whitney rank sum tests, p=0.003 and p=0.034, respectively). The site differences were primarily caused by the percentage of fruit affected (Fig. 3.4.4), rather than larger crack size (Fig. 3.4.3). These differences may occur due to variation in cell size and arrangement in different areas of the pericarp (Kumcha 1998), resulting in differing crack susceptibility.

3.4.2.3 Crack Direction and Location

Most cracks started around 7 mm from the impact site, most commonly within the same site category as the impact. The impact site did not significantly affect the distance from site to crack (Fig. 3.4.5). Overall, the use of the pendulum significantly increased the average distance between impact site and crack (t-test, p=0.036), but within each individual site, there was no significant method effect. The pendulum had greater potential to cause distant cracks due to the different loading pattern, with fruit being impacted against two surfaces, rather than one.

Most cracks (88%) occurred radial to the impact site, even when cracking occurred some distance away. Only one crack, of the 121 recorded, occurred oblique to the impact site. The remaining 11% were neither oblique nor radial. The predominance of radial cracking may be related to complex mechanical properties of the tissue, such as the manner in which load energy moves through the tissue.

In response to a drop impact on the base of the fruit, cracks generally extended from the base up into the cheek (35% of all cracks) or side of the fruit (41%). Similar results occurred using the pendulum, with base-cheek and base-side cracks each comprising 29%. However, pendulum fruit also showed some stem cracks (29%) in response to base impact.

Dropped impacts to the cheek resulted in cheek-side (40%) or cheek-stem (24%) cracks, with cracks occasionally extending towards the base or shoulder (both 12%). Using the pendulum, cheek to side fractures accounted for 38% of cracks in fruit injured at the cheek. This level was similar to dropping, but using the pendulum the majority of these cracks extended into the opposite cheek. Similarly, cheek-shoulder fractures (10%) were at a similar level, but all extended into the opposite cheek. Fractures from cheek to stem (24%) also occurred at a similar level as in dropped fruit. No cheek to base fractures occurred using the pendulum. Secondary cracks near the stem (24%), often extending to the opposite cheek, occurred regularly using the pendulum, but were minimal (4%) in dropped fruit. These cracks were a considerable distance from the impact site (~20-30 mm), and were probably exacerbated by the different impact loading resulting from clamping the fruit in place. The additional stem cracks occurring due to the use of the pendulum contributed to the difference in crack areas between the two methods of application. However, a significant difference remained even when these cracks were removed from the analysis (Mann-Whitney rank sum test, $p<0.001$).

Impacts applied to the shoulder most commonly resulted in fracture from shoulder to cheek in both dropped (45%) and pendulum fruit (25%). Cracks starting at the cheek were also common, at 36% in dropped fruit and 25% in pendulum impacts. Cracks extending from shoulder to side also occurred, and were more common in pendulum fruit (25%) than dropped (9%). Stem cracks were observed in pendulum fruit (17%), but did not occur in dropped fruit.

In fruit dropped onto the side, fracture commonly occurred from side to cheek (36%) or from side to shoulder, often extending into the stem area (21%). Using the pendulum, side-cheek cracks were longer, extending into the opposite side of the fruit (14%). Similarly, side-shoulder cracks (21%) also extended to the stem, opposite shoulder or cheek. Cracks starting near the stem were more common in pendulum fruit (29%) than dropped (7%).

Cracking patterns using the pendulum were generally very similar to dropped fruit, showing that the pendulum gives a reasonable representation of real-life impacts. Slight differences, such as greater prevalence of stem cracks, and cracks extending further around the fruit, are likely to have occurred due to the compressive force of the clamp.

Overall, the number of cracks occurring in each fruit site was relative to the SA of the site. When all crack locations were combined into a single list, the ratios were remarkably similar to SA values. For example, 35% of crack locations were at the cheek (~36% SA) and 11% were at the base (~12% SA). The shoulders comprised 16% of crack locations, at 12% of the fruit SA, suggesting a possible slight susceptibility to cracking. Despite only 7% of the fruit SA being defined as stem, 14% of crack locations were in this area. The stem area was the area of the fruit most susceptible to cracking. This was increased by the use of the pendulum, with twice as many stem cracks occurring compared to the dropped fruit. Fruit sides appeared to be fairly resilient, at 33% SA, but only 23% of crack locations. Different resistance to cracking may relate to structural properties, or the transfer of energy through the pericarp.

3.4.3 Conclusions

The use of cheek impact applied by pendulum as a standard treatment is likely to have overestimated cracking susceptibility. On average, 47% of dropped fruit showed open cracks, while open cracks occurred in 80% of fruit hit on the cheek using the pendulum. Both the impact site and method contributed to the high level of cracking in the standard treatment. Impacts applied to the cheek showed a higher percentage of cracking than any other site. The use of the pendulum typically increased the area of cracks, rather than the percentage of fruit affected. This was probably due to the slight compressive force applied in clamping the fruit in place. Similar impacts can occur in practice, due to fruit being wedged into a corner, and struck by another moving fruit, but this is not a common type of impact. The standard pendulum treatment causes increased cracking susceptibility, an effect that must be considered in the practical application of previous experimental results.

3.5 Effect of Repeated Impact Loading

Repeated application of loads may result in tissue failure, despite the fact that each single load is beneath the threshold of damage. This effect was observed in the compression injury of apples (McLaughlin and Pitt 1984). It was hypothesised that damage did not accumulate, but that each load cycle carried an equal risk of tissue failure, which gradually increased the chance of damage with repeated loading. In any impact event deformation is partitioned into elastic (recovered) and plastic (residual) deformation. Plastic deformation occurs due to small cracks, pores and discontinuities in the tissue (Sitkei 1986), and the movement of fluid (Pitt 1982) or air from the compressed tissue (Pereira and Calbo 2000). The amount of plastic deformation is greatest in the first loading cycle, and decreases in subsequent cycles (Sitkei 1986). An equilibrium is eventually reached, where no further plastic deformation occurs. Previously damaged tissue effectively cushions the load, preventing any further plastic deformation.

A trial was carried out to determine the effect of multiple impacts on the symptoms of damage in lychee. An understanding of the effect of repeated impacts on lychee damage levels will ensure the validity of impact threshold values. If repeated loading substantially increases damage levels, the fall height tolerance may need to be reduced.

3.5.1 Methods and Materials

‘KMP’ fruit were harvested in Byron Bay, and transported to Brisbane by car in damp paper towel, over-wrapped with plastic. The fruit were placed into 5^{o}C storage for one week, and were then transported to the Sunshine Coast. Fruit were allowed to warm to room temperature prior to treatment. In a randomised control trial, fruit were treated by varying numbers of repeated impact, with each treatment of 0, 1, 2, 3, 4 or 5 impacts randomly applied to 20 fruit. Each impact was applied by pendulum from level 1.5, using a rounded head with no added weights. Each impact was estimated to be equivalent in energy per unit area, to a 50 cm fall onto a flat surface. After impact, fruit were stored at room temperature and 53% RH.

Colour readings of the cheek centre were taken before and 24 hours after treatment. Crack width, length and penetration were recorded immediately after treatment, and after 24 hours storage. The number of injured tips at each severity score (1 = light, 2 = light-moderate, 3 = moderate, 4 = moderate-severe, 5 = severe) was recorded. A tip darkening index was calculated as the sum of number of tips affected multiplied by severity score for each fruit.

3.5.2 Results and Discussion

Changes in colour after impact revealed a consistent trend (Fig. 3.5.1). The first impact caused substantial colour change, including darkening, yellowing and loss of chroma (t-tests $p<0.001$). The second impact caused further deterioration of colour, with darkening and chroma loss significantly increased (t-tests, $p=0.046$ and 0.002, respectively). However, colour changes resulting from the second impact were almost halved compared to the first impact. Subsequent impacts did not result in any further significant colour change. Similar trends were also shown in the index of tip darkening. The second impact caused a significant increase in tip darkening (t-test, $p=0.001$), while further impact events did not increase the level of damage (Fig. 3.5.1). These results show a similar pattern of resultant damage as that described by Sitkei (1986). The greatest damage occurs in the first impact, and subsequent impacts cause progressively less damage. Tissue damaged by the first impact effectively cushions the fruit from further injury.

The high degree of inherent variation in cracking prevented the detection of any significant differences between treatments. The percentage of fruit affected by cracking in each treatment ranged from 30-55%, and showed no clear increase due to repeated impact. These results are dissimilar from those observed in apples by McLaughlin and Pitt (1984), where repeated compressive loads increased the chance of tissue failure. This difference may occur due to differences between impact and compression loading, or due to inherent variation obscuring the effect. There appeared to be a trend towards increased average crack area after the third impact event (Fig. A2 in the Appendix). It is possible that subsequent impacts may cause existing cracks to enlarge. Greater replication would be needed to clarify the effect of repeated impacts on cracking.

3.5.3 Conclusions

The first impact caused the greatest colour change and tip darkening. A second impact at the same site increased the extent of damage by around 50%, and subsequent impacts did not significantly increase damage. Repeated impacts did not show a strong effect on cracking, with no significant differences detected.

3.6 Effect of Cushioning Material on Impact Damage

Cushioning material can be used on bulk bins and hard packing line components to reduce the risk of impact injury (Brown *et al.* 1990). Padding increases both the impact duration and the area of contact, thus reducing the amount of force acting on the fruit tissue, and hence the resultant damage. A trial was carried out to test the benefits of padding in the impact damage of lychee fruit.

3.6.1 Methods and Materials

‘KMP’ fruit were harvested from Yandina, and were stored at 5^oC in damp paper towel for 11 days prior to treatment. Fruit were close to room temperature at the time of injury. Impacts were applied from pendulum level 4, using a flat head and 2 additional weights. Impact energy was estimated as equivalent to a 122 cm fall onto a flat surface. Treatments involved the attachment of padding to the impact head. In a randomised control trial, replicates of 20 fruit were subjected to a single impact using:

- Impact head without any padding;
- Impact head covered with a single layer of foam (around 6 mm thick and very easily compressed);
- Impact head covered with a single thickness of lychee pericarp.

In addition, 10 fruit were treated by impact using a double layer of the same foam padding, and 10 fruit acted as controls (no impact applied). Lychee skin, used as a form of padding, was damaged by impacts, appearing to become progressively less effective. Fresh skin was separated from worn skin for analysis to compensate for this effect.

Colour readings were taken before impact, and 24 hours after. The length, width and penetration of any cracks were noted after impact, and re-measured at 24 hours (excluding double foam treatment). The number of protuberance tips showing damage was also recorded at 24 hours.

3.6.2 Results and Discussion

Darkening, yellowing and loss of chroma caused by impact were significantly reduced by the use of a double layer of foam (compared to impact without padding; L-value, t-test p=0.016; hue, Mann-Whitney rank sum test, p=0.041; chroma, rank sum test, p<0.001) (Fig. 3.6.1). Other padding treatments consistently tended to show less colour change, but were not significantly different to impact without padding. Fruit injured using double foam padding showed a greater increase in hue than the control (t-test, p=0.003), but did not show significant darkening or loss of chroma compared to uninjured fruit.

Any type of padding (including lychee skin) attached to the impact head significantly reduced the number of darkened protuberance tips (Fig. 3.6.2). All padding treatments resulted in a number of injured tips not significantly different to the control.

Fresh lychee skin and a double layer of foam significantly reduced total crack area (Mann-Whitney rank sum tests at 0 hours, p=0.005 and p=0.022, respectively) (Fig. 3.6.3), and prevented the formation of open cracks (Fig. 3.6.4). Worn lychee skin and a single layer of foam did not significantly reduce crack area. The worn lychee skin slightly reduced the total number of fruit cracked, from 80% to 60%, while a single layer of foam was of no benefit (Fig. 3.6.5). Both worn lychee skin and single layer foam appeared to give a slight reduction in the number of open cracks.

The strong cushioning properties of a double layer of foam and fresh lychee skin are also reflected in the proportion of treated fruit showing cracking (Fig. 3.6.5). The use of fresh lychee skin as padding reduced the total number of cracked fruit from 80% to 20%. In this respect, it appeared to be more successful than double layered foam, which resulted in 70% of fruit showing some cracking. Both double foam and fresh lychee skin eliminated open cracks, which occurred in 65% of fruit when no padding was used. The lychee skin had a much lower level of deformation than the foam, which was very easily compressed. Firmer padding than the type tested is likely to be of greater benefit in reducing cracking.

The increased benefit in crack reduction of the lychee skin, compared to the double foam, may result from the skin shape. The curved lychee skin placed over the flat impacting head created an air space, which may have contributed an additional cushioning effect. This air space may also have slowed down the impact, as the fruit were forced to push the lychee skin towards the impacting head for the last few millimeters of movement. In terms of colour change and tip darkening, no significant differences were found between worn and fresh lychee skin. However, fresh lychee skin seemed to reduce the incidence of fruit cracking more than worn skin, perhaps due to a decline in the air space behind the skin.

3.6.3 Conclusions

All forms of padding tested showed some benefit in reducing tip darkening. Double layer foam and fresh lychee skin were also successful in significantly reducing cracks due to impact. Other treatments also appeared to give a slight reduction in the incidence of open cracks. Double layered foam improved the colour response to impact more than any other treatment, suggesting that cellular disruption was minimised. However, fresh lychee skin appeared to give better overall control of cracking. The results suggest that the use of firm padding would give significant economic benefits in situations where there is a risk of cracking due to impact.

3.7 Effect of Carton Liners on Vibration Damage

In experiments carried out on vibration in this thesis, the standard method used to simulate vibration damage involved treatment of fruit in an unlined cardboard box. Plastic liners are generally used in the lychee industry, principally to reduce moisture loss from the fruit during transport and storage. Box liners can affect vibration damage, as shown in the transport of pears, where polyethylene liners reduced vibration damage through the restriction of fruit movement (Slaughter *et al.* 1998). As the standard technique used in this thesis differed from typical industry practice, it was desirable to ascertain whether the use of a liner has any effect on the levels of vibration damage sustained to lychee fruit.

3.7.1 Methods and Materials

Fruit of cultivar ‘Bengal’ were purchased from Brisbane Markets and were stored in damp paper towel at 4^{o}C for 4 days prior to treatment. The fruit were dry and at RT when treated. Various packaging treatments were randomly assigned to groups of 5 fruit, with treatments including a cardboard box with no liner, and three different types of liners. The liners tested included perforated PCY crispy polypropylene (the industry standard), unperforated HDPE (high density poly-ethylene) and a thick unperforated polythene. Liners were cut to 23 x 25 cm, and were heat sealed along three sides. The bags were folded over once to contain the fruit. In a randomised trial, 9 replicates of 5 fruit were treated by each method. Fruit were treated by 10 minutes vibration at setting 4, with 15 fruit per box. The additional ten fruit per box were replaced when vibration damage was severe, and bags were replaced when worn.

The level of damage was assessed by colour readings, taken at 7 sites on each fruit. Readings were taken of both cheek centres, the sides of the fruit, the base and the shoulders. Colour readings were taken both before and immediately after vibration treatment.

3.7.2 Results and Discussion

There were no significant differences between treatments, with all treatments showing similar average changes in L-value, hue, and chroma (Fig. A3 in Appendix). These results suggest that when dry fruit are subjected to vibration damage, liners do not significantly affect the extent of injury. However, liners would be expected to affect the level of injury if fruit were wet. Surface water, either due to initial fruit wetness or condensation, would be retained in the carton by plastic liners. Due to the fact that surface wetness increases injury (Section 4.5.2), liners that retain water near the fruit would probably tend to increase the level of vibration damage. In a preliminary experiment carried out on the effect of liners, fruit were treated while showing signs of condensation. Significant differences between treatments could not be detected due to high variation and limited replication, but levels of damage did seem to be higher in lined boxes (results not shown).

A high degree of variation was observed between individual replicate runs. This illustrates the difficulty in creating a truly replicable vibration treatment. Subtle changes in fruit sizes, arrangement and movement within the box appear to result in a large degree of variation in the resultant damage.

3.7.3 Conclusions

Box liners have no significant effect on the level of injury in dry fruit subjected to vibration. Thus, the absence of a box liner in the standard vibration treatment would not have significantly affected results. In practice, box liners that retain water near the fruit would be expected to increase susceptibility to vibration damage after condensation. However, it is unlikely that the risk of increased vibration damage would outweigh the benefits of liners in reducing moisture loss.

3.8 Mechanical Damage Inflicted During Destalking

Mechanical destalkers are used by some lychee growers in an effort to reduce labour costs. Fruit are harvested in panicles, and the destalker is used to separate fruit from stems. Destalkers vary in design, but the process generally involves agitation of the fruit to remove the stems. The potential for destalkers to cause fruit damage has been observed, with skin browning resulting from incorrect adjustment of the equipment (Menzel *et al.* 2002). This experiment was carried out to determine whether mechanical destalkers have the capacity to cause significant mechanical damage to lychees.

3.8.1 Materials and Methods

‘Tai So’ fruit grown in Sarina were harvested by hand in panicles and transferred to the packing shed in shallow plastic crates. They were stored in a cool room prior to placement onto the packing line. The fruit had a pulp temperature of approximately 10^{o}C, and were damp with condensation at the time of injury. The destalker, designed by KW Engineering Pty Ltd, used a series of roller brushes to remove debris and stems from the fruit.

In an observational trial, fruit were randomly sampled from the packing line, with 25 fruit taken immediately before and after mechanical destalking. Where necessary, stems were removed by hand immediately after sampling. The fruit were numbered, and colour readings were taken immediately, with the fruit surface patted dry using a tissue prior to measurement. Seven colour readings were taken on each fruit, of both cheek centres, the sides of the fruit, the base and the shoulders. Colour readings were repeated after 52 hours storage at 20^{o}C and 53% RH. Average values for each fruit were used in analysis.

For 5 fruit from each sample, the damage at each colour reading site was assessed. A small plastic ring was used to keep the area constant. The number of light brown and dark brown tips and yellowed protuberances were noted. These damage counts were analysed based on averages within each fruit. All fruit treated in the destalker were weighed and the percentage of the fruit surface showing yellowing was estimated in each fruit, to determine whether fruit weight affected the level of injury.

3.8.2 Results and Discussion

The destalker caused immediate significant changes in fruit colour, which were typical of vibration damage (Fig. 3.8.1). The average hue was 33 in control fruit, and 39 in fruit treated by destalking. This severe yellowing of colour was very obvious in visually comparing the two groups of fruit. Fruit treated by destalking also showed slight darkening and loss of chroma compared to control fruit, both of which were significant. These results confirm that vibration damage was the likely cause of skin browning observed by Menzel *et al.* (2002) due to destalking.

The treated fruit showed a slightly greater rate of yellowing over 52 hours storage (Mann-Whitney rank sum test, p=0.004). The change in hue was 0.89 in treated fruit, and –0.25 in control fruit. Loss of chroma during storage was similar in treated and control fruit (-2.34 and –2.14 respectively). Control fruit exhibited a slight darkening of colour (change in L-value of –0.26), and treated fruit lightened slightly (+0.19) during storage, but the L-values remained significantly different at 52 hours (t-test, p=0.031).

Fruit treated by destalking showed a significantly greater number of yellowed protuberances (t-test, p=0.023) (Fig. 3.8.2). It was estimated that on average, around one quarter of the skin surface area was affected by noticeable yellowing. Tip darkening was not significantly affected by mechanical destalking.

No correlation was detected between fruit weight and the level of damage sustained during destalking. However, the only 3 fruit severely affected were of a very similar weight range, from 26.2 to 27.3 g. These fruit showed yellowing to 75-80% of the surface area, while in all other fruit, the average was 16%, and the maximum 40%. It is possible these fruit may have been more severely affected due to specific design features in the equipment. However, the severe damage in these fruit may simply have resulted by chance, especially if they were from the same panicle. It appears that the destalker generally causes similar levels of damage to fruit ranging from 17.5 to 31 g.

In this experiment, fruit wetness would probably have increased the severity of vibration injury resulting from mechanical destalking (Section 4.5.2). Treating the fruit in a cool, wet condition was standard practice on this farm. Damage due to destalking may be substantially reduced by treating fruit when warm and dry.

3.8.3 Conclusions

The results confirm that vibration damage is a practical issue in the lychee industry, with a commonly used postharvest treatment shown to cause significant damage. The destalker used in this experiment subjected fruit to severe vibration injury, resulting in strong yellowing of colour, slight darkening and loss of chroma. Further simple experimentation for other cultivars is recommended, as different responses may occur. Mechanical destalking can reduce labour costs, but this needs to be weighed up against the accompanying loss of quality. It is recommended that stalks are removed by hand to maximise fruit quality.

3.9 Overview - Load Properties Influencing the Fruit Response to Mechanical Injury

Magnitude is the key load variable affecting the extent of mechanical damage, and is largely dictated by impact fall height, vibration frequency and compression load weight. For vibration and compression injuries, duration of the load is also important (Sitkei 1986).

Impacts from low height caused tip darkening and colour change, with these symptoms gradually increasing in severity with fall height (Section 3.1). For the ‘Tai So’ fruit tested, cracking first emerged at 60-70 cm fall height. The incidence of open cracks increased with subsequent rises in fall height. This threshold range for cracking gives a good estimate for the design of equipment, but could vary substantially due to factors such as cultivar and fruit condition.

Increases in both duration and frequency were shown to intensify the damage caused by vibration (Section 3.2). Longer duration had little effect at low frequency, but strongly increased damage at high frequency vibration.

Contrary to previous reports (Batten and Loebel 1984; Greer 1990), lychee fruit were fairly tolerant of compressive loads, with 1 kg weight applied for 48 hours causing minimal damage (Section 3.3). The load tolerance translated into a stacking depth of at least 1 metre. Limiting the load weight to 500 g for optimal fruit quality would allow at least a 50 cm stacking depth. The tolerance of compression in lychee would allow tighter packing, and deeper picking trays and cartons. Deeper containers may be beneficial in reducing vibration damage, as a lesser proportion of fruit would be located in the susceptible top layers (Bardaie and Hitam 1979).

Impact load properties such as method of application, location, repetition and cushioning were explored for their effects on the extent of damage. Fruit injured using the pendulum tended to show a similar incidence, but greater area of cracking than dropped fruit (Section 3.4). This may have occurred due to the slight compressive force of the clamp pushing the wound open. When applied to the cheek, impacts showed higher rates of cracking than at other locations on the fruit. The results suggest that the standard treatment used in this thesis (pendulum impact to the cheek) would slightly overestimate cracking susceptibility.

The lychee response to repeated impact was similar to the general model described by Sitkei (1986). A second impact significantly increased tip darkening and colour change, causing changes around half as great as the initial impact (Section 3.5). Subsequent impacts did not cause further deterioration in colour or tip injury. A significant difference in cracking due to repeat impacts was not detected, and no trends were observed in incidence. Although not significant, crack area seemed to increase after the third impact event, suggesting that repeat impacts may enlarge existing cracks.

Cushioning was found to be of substantial benefit in the reduction of tip darkening, colour change and cracking due to impact (Section 3.6). Thick soft foam padding resulted in minimal colour change, and significantly reduced cracking, while fresh lychee skin gave the best control of cracking. Firm foam was suggested to combine these benefits.

The effect of box liners on vibration injury was studied to ensure that the standard treatment, without a liner, was a valid representation of practice. This was confirmed, with liners not significantly affecting vibration damage (Section 3.7).

However, it was theorised that liners may exacerbate vibration damage if water were retained near the fruit.

The capacity of destalking machines to inflict vibration damage on lychee was observed (Section 3.8). Very strong yellowing, slight darkening and loss of colour saturation were shown in fruit subjected to mechanical destalking. This type of damage could easily be checked on-farm, simply by comparing fruit sampled before and after the treatment. Where substantial vibration damage is occurring, hand destalking would be recommended to retain the fresh red colour characteristic of lychee.

The research shows that mechanical damage can be lessened by reducing load magnitude, particularly impact fall heights and vibration frequency. For compression and vibration injuries, limiting the duration can also be beneficial. Practical suggestions for reducing lychee mechanical damage may include tight fill packing, deeper stacking depths and the use of cushioning where impact may be a problem.

CHAPTER 4. FRUIT PROPERTIES AFFECTING THE RESPONSE OF LYCHEE TO MECHANICAL DAMAGE

Fruit properties influencing the response to mechanical damage may include cultivar, gross anatomy, hydration, temperature and maturity. Structural, biochemical and morphological differences result in cultivar effects, which can be significant, as shown in impact damage of cherries (Burton and Schulte-Pason 1987). Gross anatomical characteristics such as fruit size, weight and shape can directly affect load properties such as magnitude and contact area. The fruit response may also be affected by gross morphology, as is the case for seed size in peaches (Menesatti *et al.* 1999). Skin properties can be important in determining the response to mechanical injury, particularly when a soft internal structure is surrounded by a firmer, relatively thin shell (Lichtensteiger *et al.* 1988). Structural features of the fruit tissue strongly influence the response to mechanical stress. Tissue structure affects both the amount of deformation resulting from a load, and the amount of deformation required to damage cells.

Fruit maturity, hydration and temperature can strongly influence fruit mechanical properties. A strong maturity effect on mechanical damage particularly occurs in fruit showing major textural changes during ripening, and is therefore unlikely to be important in lychee. Moisture content strongly affects fruit mechanical properties, with mild water stress typically reducing cell rupture, but increasing tissue deformation. The effects of temperature on the mechanical properties of fruit and vegetables are complex, and can include changes in turgor pressure and elasticity. The effects of temperature and hydration on the fruit response to injury are particularly important due to the ability to manipulate these parameters to some extent to avoid damage.

4.1 Effect of Fruit Hydration on Lychee Impact Damage

Moisture content is one of the most important factors affecting the mechanical properties of fruit and vegetables. The effect of hydration on impact damage has been studied in many crops, with increased moisture content generally increasing damage susceptibility. Water loss in apples tends to increase bruising resistance (Klein 1987. Turgid potato tubers are more susceptible to shatter, while flaccid tubers show more blackspot (Bland *et al.* 1987). The research aimed to determine the effect of fruit hydration on the response of lychee fruit to impact damage.

4.1.1 Methods and Materials

The experiment was carried out on mature ‘KMP’ fruit harvested in Yandina. Fruit were harvested into water and remained in water for at least one hour to ensure a high level of hydration prior to treatment. Fruit hydration was manipulated by storage at five different levels of RH (11, 33, 53, 75, 97%) for 3 days at 20^{o}C. Relative humidity was controlled using a few shallow dishes of the appropriate saturated salt (Table 1.6.1) in each plastic crate. The weight of each fruit was measured before and after the humidity treatment, and 6 to 8 fruit from each storage treatment were used to measure water potential after storage.

After the 3 day storage period, 15 fruit from each humidity treatment were subjected to each mechanical damage treatment, in a factorial randomised control trial:

- Impact A – from level 2, using a flat head and one added weight (26.54 g), estimated to be equivalent to a 42 cm fall;

- Impact B – from level 1.5, using a round head and two added weights (53.12 g). Impact energy per unit area was estimated to be equivalent to a 62 cm fall onto a flat surface;
- Control fruit were not mechanically damaged.

The application of treatments was spread over two days in this experiment. After injury, fruit were stored at 20°C and 53% RH. Single colour readings at the cheek centre were taken immediately before and after injury, and then after approximately 24 hours. Some 24 hour colour readings could not be taken due to equipment problems. The dimensions of any rots and cracks were recorded.

An additional 20 fruit were harvested into water one week after the original harvest date. Fruit were from the same orchard and of the same cultivar, and were of very similar maturity level to fruit used in the original experiment. These fruit were stored in water for at least one hour after harvest and were then injured by the same impact treatment as for Impact ‘A’ (flat head, level 2, one weight) in a fully hydrated state. Length, width and penetration of cracks were recorded.

Loss of moisture resulted in changes in colour, unrelated to the response of the tissue to impact. The direct effects of moisture loss on colour were removed by subtracting from each individual measurement, the average control values for the same period and humidity level. The corrected colour changes were solely due to the effect of moisture loss on response to impact injury.

4.1.2 Results and Discussion

Relative humidity treatments in the original experiment resulted in levels of weight loss ranging from 0.8 to 7.5% (Fig. 4.1.1). Water potential followed a similar trend, ranging from –1.0 to –2.2 MPa. The 33% humidity treatment did not result in as much moisture loss as expected. This may have been due to a slower equilibration process in magnesium chloride compared to the other saturated salts used.

The relationship between weight loss and water potential (Fig. 4.1.2) was close to linear ($R^2=0.899$), fitting the equation:

$$\Psi = -0.981 + L \times -0.156 \qquad \textit{…Equation 4.1.1}$$

Where, Ψ = water potential (MPa) and L = weight loss (%).

Equation 4.1.1 was used to estimate the water potential of fruit prior to treatment (at 0% weight loss). The additional fruit harvested after one week were assumed to be at this level of hydration.

4.1.2.1 Colour Change Due to Impact

Water stressed fruit showed increased colour change in response to impact (Fig. 4.1.3). Darkening and loss of chroma due to impact ‘A’ (equivalent to 42 cm) were significantly greater in fruit stored at low RH levels. A significant treatment effect on the change in hue due to impact was not detected. However, change in hue was shown to be positively correlated with individual fruit percentage moisture loss (Spearman correlation co-efficient 0.338, p=0.0043). Greater moisture loss tended to result in stronger yellowing in response to impact.

The heavier impact treatment (B) showed similar, but less extreme trends than impact ‘A’ (Fig. 4.1.4). The weaker effect, and reduced number of replicate colour readings, resulted in no significant treatment effect being detected. However, strong correlations were detected between moisture loss in individual fruit and the change in L-value (Spearman correlation co-efficient = –0.516, p<0.0005) and chroma (co-efficient = -0.518, p<0.0005).

Stronger colour changes in response to impact suggest that greater cell disruption is occurring in water stressed fruit. This may occur as a result of changes in mechanical properties of the tissue, including reduced cell elasticity and increased deformation under a given load.

4.1.2.2 Protuberance Tip Darkening

The number of dark tips resulting from impact did not tend to follow the same trends as colour changes (Fig. 4.1.5). The greatest number of dark tips due to impact ‘A’ occurred in the 11% RH treatment group, with an average of 15 tips. However, primarily due to rots obscuring the dark tips, this result was an average of only two values (14 and 16), and is thus fairly unreliable. Across other RH treatments, there appeared to be a trend of water stressed fruit showing fewer darkened tips. A significant negative correlation between the number of injured tips and individual fruit weight loss was shown for impact ‘B’ (Spearman correlation co-efficient –0.405, p=0.0023). Water stressed fruit suffered less tip injury, despite showing greater darkening and loss of chroma in response to impact.

The difference in trends between colour change and tip darkening suggest that these are somewhat independent processes. Tip darkening may result from cell rupture, occurring as a separate process to the background changes in skin colour detected by colour readings. The background colour changes may result from cell disruption, rather than rupture, or may occur at a deeper level in the tissue.

The mechanism of increased tip darkening in hydrated fruit is probably the outward pressure created by cell turgor. A turgid cell is more likely to rupture in response to a substantial impact, compared to a flaccid cell. In the turgid cell, compression of the cell contents more easily results in internal pressure exceeding the strength of the cell wall. In contrast, a flaccid cell would be able to deform without excessive internal pressure. In effect, the water-stressed cell has a greater ability to absorb energy without rupturing. However, the absorbed energy results in internal disruption of cell contents, and hence some discolouration. In addition, the greater deformation of the water-stressed tissue increases the size of the impact surface, thus effectively reducing the impact energy per unit area. The injury spreads over a larger area, resulting in general discolouration of the skin tissue, rather than the concise, severe darkening of the tips. The different behaviour of tissue in response to impact can thus be explained by changes in cell mechanical properties due to hydration.

4.1.2.3 Pericarp Cracking

A strong hydration effect on cracking was evident in fruit treated by impact ‘A’, with well-hydrated fruit showing a greatly increased susceptibility to cracking (Fig. 4.1.6). Low humidity treatments (11, 33 and 53% RH) showed no cracking, while 75 and 97% RH treatments showed cracking in 7 and 29% of fruit, respectively. Fruit at harvest water potential showed the greatest susceptibility to cracking, with 35% of fruit affected. Average crack area also increased with water potential in the range –1 to -1.3 MPa, although this was principally due to the increased number of fruit affected. A strong negative correlation between individual fruit weight loss and crack area further confirmed the relationship (Spearman correlation co-efficient –0.353, $p<0.0005$). Observation of individual weight loss and crack values revealed that fruit subjected to greater than 3% moisture loss after harvest did not crack under impact.

Only 4 fruit showed any signs of cracking in response to impact ‘B’. Cracked fruit were from 53% and 97% RH treatments, with average crack areas 0.8 and 6.9 mm^2, respectively. Due to the small number of fruit affected, analysis was not possible. However, the results suggest a similar trend of increased cracking in well-hydrated fruit.

Cracking occurs when impact force exceeds the strength of the tissue, resulting in failure (Sitkei 1986). The rupture occurs on a whole fruit scale, rather than in individual cells, but results from a similar mechanism to the tip darkening response. The turgid aril exerts constant outward pressure on the pericarp. The aril is observed to deform under impact, but the deformation appears to be mainly elastic, and hence results in no visible damage to aril cells. However, during the moment of impact, the deformation of the aril would increase the outward pressure on the pericarp. It is likely that this effect would be strongest in the region surrounding the impact site. Higher moisture content in the aril would be expected to increase outward pressure acting on the pericarp. While the aril water content does not decline as rapidly as the pericarp, changes do occur due to water loss (Zhao *et al.* 1999). In the well-hydrated fruit, aril and skin are less capable of cushioning the blow by plastic deformation. Because deformation is minimal, impact energy is more likely to transfer through the tissue and result in tissue failure away from the impact site.

Mild water stress decreases susceptibility to cracking, but increases background colour change due to impact. A similar relationship is shown in potato, where a shatter response is more common in hydrated tubers, while discolouration is typical in flaccid tubers (Bland *et al.* 1987). In potatoes, the discolouration resulting from impact is internal, due to the ability of energy to move through the tissue. Lychee discolouration is confined to the surface tissues, but the hydration response results from the same changes in cell mechanical properties.

4.1.3 Conclusions

Mild water stress increased the background colour changes resulting from impact, with darkening, loss of chroma and yellowing of colour significantly affected. However, slight water loss resulted in increased resistance to pericarp cracking and tip darkening. For an impact equivalent to a 42 cm fall, there appeared to be a threshold at 3% loss of harvested weight (approximate water potential of –1.45 MPa), above which cracking did not occur.

The effect of hydration on lychee impact cracking has strong implications for postharvest management of the fruit. A typical impact threshold of 60 to 70 cm was proposed for lychee (Section 3.1.3). However, this research shows that an impact equivalent to around a 42 cm fall can cause substantial cracking injury if fruit are well-hydrated. Avoidance of cracking is essential to the maintenance of fruit quality, so the tolerable fall height must be reduced to at least 40 cm to allow for variation in tolerance due to hydration. The design of postharvest operations must take into account the increased risk of cracking associated with turgor. More careful handling would be required if fruit were harvested in wet conditions. Deliberate treatments to increase turgor, such as soaking, would be best located after any packing line processes capable of causing impact damage.

4.2 Effect of Fruit Hydration on Cracking Susceptibility

Impact cracking of lychee was shown to be affected by hydration, with fruit of higher water potential more susceptible to cracking injury (Section 4.1.2.3). This experiment aimed to further explore the effect of mild water stress on impact cracking, with greater replication, and a range of impact levels.

4.2.1 Methods and Materials

The experiment was carried out on ‘KMP’ fruit harvested at Byron Bay. At harvest, half of all fruit were randomly placed into water and the other half into dry paper towel. The wet fruit were packed tightly in damp paper towel for transport to Brisbane, and were subsequently returned to water on arrival. Fruit remained in these storage conditions overnight at room temperature. The fruit stored in dry paper towel were mildly water stressed, and felt slightly springy compared to the very firm feel of the turgid fruit stored in water. Fruit were sampled for pressure bomb readings, but these measurements could not be carried out due to equipment problems.

Impact was applied using a round head with no added weights. In a randomised trial, 30 hydrated and dehydrated fruit were each subjected to single impacts from level 3 (equivalent to in impact energy per unit area to a 94 cm fall) and level 4 (114 cm). In addition, approximately 10 replicates were subjected to a level 2 impact (equivalent to 67 cm) and around 20 to a level 5 impact (130 cm). After injury, fruit were stored at room temperature and 53% RH. Colour readings of the cheek centre were taken prior to injury, and after 24 hours storage. The moisture stressed fruit were dipped into water and dabbed dry prior to measurement of the initial colour reading to ensure an equal surface dampness effect on colour. Crack width, length and penetration were recorded immediately after injury, and after 24 hours storage. The number and severity (0=none to 5=severe) of dark tips were also recorded after 24 hours storage. An index of tip darkening was calculated, multiplying the number of tips injured by severity score.

4.2.2 Results and Discussion

4.2.2.1 Effect of Hydration on Colour Change in Response to Impact

The effects of mild water stress on the colour change resulting from impact were inconsistent with results previously obtained (Section 4.1.2.1). In most cases hydration had little effect on colour change in response to impact (Fig. 4.2.1). Darkening after impact was not significantly affected by hydration level. Hue was significantly affected by hydration only at level 3 (equivalent to 94 cm), with water stressed fruit showing greater yellowing (Mann Whitney rank sum test, $p<0.001$). The loss of chroma after impact tended to be greater in hydrated fruit, with a significant effect occurring only at impact level 5 (130 cm) (t-test, $p<0.001$).

The greater loss of chroma in hydrated fruit was in direct contrast to previous results obtained (Section 4.1.2.1). Both experiments were carried out on the same cultivar, but fruit were from different regions. The effect of pre-harvest environmental conditions on biochemical composition or structure may have contributed to this difference. Alternatively, the higher impact levels used in the current experiment may have altered the response. Although not significant, there is some appearance of an interaction between impact level and hydration in the effect on chroma. The significantly different response between hydrated and water stressed fruit occurs only at the highest impact level, gradually narrowing to almost identical changes in chroma at level 2 (equivalent to 67 cm). In previous research (Section 4.1), impact equivalent to 42 cm resulted in greater loss of chroma in water stressed fruit.

4.2.2.2 Effect of Hydration on Impact Tip Injury

Tip darkening tended to be slightly more severe in hydrated fruit at low-moderate levels of impact (Fig. 4.2.2). Hydrated fruit showed a significantly higher index of tip darkening than water stressed fruit at impact levels 2 (t-test, p=0.014) and 4 (t-test, p=0.048). This effect was primarily due to a greater number of injured tips, which were significantly higher in hydrated fruit at levels 2 (t-test, p=0.008) and 4 (t-test, p=0.016). These results confirm those previously obtained (Section 4.1.2.2), that water stressed fruit suffered less protuberance tip injury. However, the response is not strong or consistent across the full range of impact levels.

4.2.2.3 Effect of Fruit Hydration on Cracking

Hydrated fruit consistently showed greater susceptibility to cracking, with average crack areas usually more than double those of water stressed fruit (Fig. 4.2.3). A significant hydration effect on total crack area was detected at impact levels 3 (Mann Whitney rank sum tests; 0 hours, p=0.014; 24 hours, p<0.001) and 4 (rank sum tests; 0 hours, p=0.001; 24 hours, p<0.001). Both greater incidence of cracks and larger crack size contributed to this effect. Fruit hydration had no significant effect on the size of open cracks.

The strong effect of hydration on cracking susceptibility is clearly illustrated in the percentage of fruit affected by cracking (Fig. 4.2.5). Slightly decreased moisture content can reduce the incidence of cracking by almost half. In particular, the reduction of open cracks would be very beneficial in practice, as shelf life is substantially reduced by open cracking. At impact equivalent to 114 cm, slight moisture stress decreased the incidence of open cracks from 27 to 7%.

4.2.3 Conclusions

In contrast to previous results, mildly water stressed fruit tended to show reduced loss of chroma in response to impact. The different response may have occurred due to an interaction between impact height and hydration. Tip darkening was more severe in hydrated fruit, confirming previously observed results (Section 4.1.2.2). The strong effect of hydration on cracking susceptibility was confirmed. The incidence and size of pericarp cracks were substantially decreased by mild water stress.

4.3 The Effect of Fruit Hydration on Compression Deformation Processes

Moisture content strongly affects the mechanical properties of tissue subjected to a load (Sitkei 1986). Water-stressed cells typically deform more under a given load, but require greater stress to rupture than turgid cells. This generally results in moisture loss conferring greater resistance to bruising (Baritelle and Hyde 2001; Klein 1987). However, turgid fruit may be more susceptible to shatter or cracking injury, as is the case in potato (Thornton *et al.* 1973). Research was undertaken to define the mechanical properties of lychee tissue in response to varying levels of compressive load, and to determine the effect of moisture content on these properties.

4.3.1 Methods and Materials

Fruit of cultivar 'KMP' were harvested from an orchard in Yandina. The fruit were stored for 2 days in damp paper towel at 25^{o}C prior to treatment. They were then transferred into either a low or high RH treatment. Low RH fruit were stored at 11% RH overnight, and were mildly water stressed, estimated to have lost around 5% of their weight. The high RH fruit were left in damp paper towel overnight, and were soaked in water prior to treatment. These fruit were assumed to be turgid, and were very firm compared to the slightly springy feel of the low RH fruit.

The compression was applied by constant weight for various durations. A small plastic cup was attached to the arm of a penetrometer to allow weights to be applied. The contact point, with a diameter of approximately 6 mm, was rested on the fruit surface. The penetrometer was reset to zero prior to loading weights onto the fruit. Weights were balanced in the plastic cup, and displacement was measured over time.

The factorial experimental design included two levels of hydration (hydrated and mildly water stressed), two levels of load (400 g and 800 g) and two different time periods (10 and 20 minutes), totaling 8 different treatments. In a randomised trial, 4 fruit were subjected to each treatment. Displacement was generally measured several times in the first minute, and then every minute for the remaining period. At the end of the treatment, the load was removed as quickly as possible, and the immediately recovered displacement measured.

Colour readings were taken, and the number and darkness of injured tips were recorded. However, no significant effects on colour or tip injury were detected, due to insufficient replication, so these data are not shown.

4.3.1.1 Calculations

Parameters such as the degree of elasticity and modulus of elasticity could be calculated from measured deformation.

$E_D = (D_F - D_R) / D_F$ ***…Equation 4.3.1***

$E_M = \text{stress} / \text{strain}$

$= F / (D_I / L)$ ***…Equation 4.3.2***

Where E_D = degree of elasticity, D_F = final deformation, D_R = deformation when weight was removed (residual), E_M = modulus of elasticity, F = force (kg mm^{-2}), D_I = initial displacement and L = fruit length (equations adapted from Sitkei (1986)).

The fruit length, required for some calculations, was not measured. An average length of 30.6 mm was used for all calculations. The measurement of displacement immediately after removal of the weight will be termed residual deformation (D_R), but does not include slow recovery from creep.

4.3.2 Results and Discussion

Hydration had a strong influence on the process of deformation, significantly affecting all tested parameters (D_I, D_F, D_R, (D_F-D_I), E_D and E_M) (for 400 g load, $p<0.001$ for all parameters except (D_F-D_I), $p=0.023$; for 800 g, $p<0.001$ for all parameters except (D_F-D_I), n.s.d.; analyses by t-tests and Mann-Whitney rank sum tests). Dehydrated fruit showed much greater deformation under a given load, resulting in higher initial, final and residual deformation values (Fig. 4.3.1 and 4.3.2). In general, dehydrated fruit tended to deform around twice as far as hydrated fruit. The creep process (gradual deformation over time (D_F-D_I)) was significantly affected by hydration under 400 g compression, with dehydrated fruit showing a greater degree of creep (Mann Whitney rank sum test, $p=0.023$). However, under 800 g compression, hydrated and water stressed fruit did not differ in creep. The degree (E_D) and modulus of elasticity (E_M) were higher in the hydrated fruit, which showed a greater recovery of shape proportional to the maximum deformation.

The magnitude of the load had an obvious effect on the mechanics of deformation. The 800 g load resulted in significantly greater initial, final (Fig 4.3.3) and residual deformation than 400 g (in hydrated fruit, D_I and D_F $p<0.001$, D_R $p=0.038$; in dehydrated fruit D_I, D_F and D_R $p<0.001$; analyses by t-tests). An increased load resulted in greater creep in hydrated fruit (t-test $p=0.004$), but not in water stressed fruit. The degree of elasticity was significantly affected by load in hydrated fruit, with the 800 g load giving a slightly higher value (t-test $p<0.001$). This difference is probably due to the slightly greater delay in measuring residual deformation using the 800 g load, as more weights needed to be removed.

A heavier load resulted in a significantly higher modulus of elasticity in dehydrated fruit (t-test $p<0.001$). The calculation of the modulus of elasticity relates the initial deformation to the force applied. However, this is not a linear relationship, as greater resistance from the tissue is encountered with increasing deformation, and hence increasing load. The application of an 800 g load did not result in twice the deformation of the 400 g load. In hydrated fruit, the 800 g load resulted in initial deformation 1.66 times greater than 400 g, while in dehydrated fruit the heavier load gave 1.78 times more deformation. These differences resulted in the slightly higher moduli of elasticity observed under heavier loads.

The creep phenomenon resulted in some significant differences due to the duration of load. Creep occurs where the load on the fruit is constant, but the tissue continues to deform over time (Sitkei 1986). The detection of significant differences in final deformation between 10 and 20 minute treatments was hampered by the low level of replication. However, the 20 minute treatments provided paired data of deformation at 10 and 20 minutes, showing a significant increase in deformation with duration (signed rank test, $p<0.001$). Doubling the load duration increased deformation around 9% on average. This is a minimal effect compared to load magnitude and hydration. The percentage increase in deformation from 10 to 20 minutes appeared to be higher in hydrated fruit, and slightly lower under the heavier weight (Fig. 4.3.4). The residual deformation was consistently higher after a longer load duration, but this was significant only in D-800 and H-400 treatments (t-tests, $p=0.039$ and 0.031, respectively). The degree of elasticity was consistently slightly lower after longer load duration, significant only in the H-800 treatment (t-test, $p=0.039$). The residual deformation measured in this experiment includes both creep and plastic deformation. Elastic deformation is recovered immediately after the removal of the load, and creep deformation is slowly recovered after the load is removed, while plastic deformation is permanent. The slightly lower degree of elasticity after longer load duration may simply result from a greater proportion of creep deformation.

Creep has been observed to typically occur in three stages; an initial rapid rate of deformation, slowing to a linear phase, and a third increasing rate, which ends with rupture (Sitkei 1986). Lychee fruit subjected to 400 and 800 g loads tended to show only the first two phases. A slight increase in the final reading was observed in some fruit subjected to 800 g load (Fig. 4.3.5). This may have been the start of the third phase, but was more likely to have simply been a single aberrant reading. Greater extent and/or duration of load are required to cause phase three of the creep process, leading to rupture.

4.3.3 Conclusions

Fruit hydration strongly affected the mechanical properties of the tissue under compression. Moderate water stress resulted in substantial increases in deformation and loss of elasticity. Deformation also increased with magnitude of load. Doubling the load resulted in an average increase in deformation of around 70%. In contrast, doubling the load duration resulted in an average 9% increase. Phase three of the creep process, involving a rapid rate of deformation ending in rupture, was not apparent in this experiment. Load duration would be expected to be of greater importance in compression incidents involving phase three. Fruit hydration, load magnitude and compression duration are important factors that need to be considered in designing postharvest operations to avoid compression damage.

4.4 Effect of Fruit Hydration on Lychee Vibration and Abrasion Damage

Hydration strongly affects the mechanical properties of fruit tissue, and is hence a key factor in the response of fruit to injury. In pears, reduced hydration of the fruit resulted in greater susceptibility to friction discolouration (Amarante *et al.* 2001). A trial was carried out to study the effect of fruit hydration on the response of lychee to abrasion and vibration injury. A strong hydration effect on lychee mechanical damage may influence the design of postharvest operations, with certain stages requiring greater care.

4.4.1 Methods and Materials

‘KMP’ fruit grown in Yandina were harvested directly into water, and soaked for at least one hour to ensure turgidity. Fruit water content was manipulated by storage at five different levels of RH (10, 33, 53, 75 and 97%) for 3 days at 20^{o}C. Individual fruit weights were measured before and after the humidity treatment to determine water loss. A further 6 to 8 fruit from each storage treatment were used to measure water potential after storage, using a pressure bomb.

After the 3 day storage period, 13 to 15 fruit from each humidity treatment were subjected to mechanical damage treatments, in a factorial randomised control trial:

- Vibration was applied for 10 minutes at setting 4 using a flask shaker. A high level of vibration damping occurred, as the treatment was applied on a raised floor. As a result, the injury severity was very low in this experiment;
- Abrasion was applied by three light 15 cm rubs against a metal file, spread over the centre of the cheek;
- Control fruit were stored in the same conditions, but were not mechanically injured.

The application of treatments was spread over two days in this experiment. Fruit were stored at 20°C and 53% RH after injury. For abrasion and control fruit, colour readings were taken immediately before and after injury, and then after 3 days storage. Colour readings were taken only before and after injury for the vibration treatment. Single colour readings were taken in the cheek centre for control and abrasion treated fruit. For vibration injured fruit, seven colour readings were taken on each fruit, of both cheek centres, the sides of the fruit, the base and the shoulders.

Loss of moisture resulted in direct changes in colour during subsequent storage, unrelated to the response of the tissue to injury. The direct effects of hydration on colour were eliminated by subtracting average control values (for the same period and humidity level) from each raw measurement. The corrected colour changes were solely due to the effect of fruit hydration on response to injury. Vibration measurements were taken immediately before and after injury, and therefore did not require this adjustment.

4.4.2 Results and Discussion

The RH treatments resulted in water loss ranging from 0.9 to 7.3%, translating into water potential values of –1.1 to –2.2 MPa (Fig. 4.4.1). The 33% RH treatment tended to result in less moisture loss than 54% RH, possibly due to slower equilibration.

Fruit hydration significantly affected the response to abrasion, with water stressed fruit showing greater yellowing, darkening and loss of chroma (analyses by t-tests $p<0.05$) (Fig. 4.4.2). Darkening and loss of chroma due to abrasion were strongly affected by hydration. Fruit with up to 2% loss showed minimal darkening and loss of colour saturation. Moisture loss of 4 to 5% resulted in a substantial increase in both darkening and loss of chroma after abrasion. Further moisture loss, totaling 7%, resulted in an even stronger colour change in response to abrasion. Changes in hue were not as strong, but showed the same trend of increasing damage with loss of hydration. Trends were confirmed by strong correlation against fruit moisture loss, with all colour change indices significantly affected (for L-value, hue and chroma, Spearman correlation co-efficients = -0.477, 0.280, -0.694 and $p<0.0005$, $p=0.0210$, $p<0.0005$, respectively).

Vibration colour change showed less definite trends, due to very low and variable levels of damage (Fig. 4.4.3). No significant differences were detected between the RH treatments. However, significant correlation was detected between fruit moisture loss and the changes in L-value (co-efficient= -0.315, $p=0.00604$) and chroma (co-efficient= -0.380, $p<0.0005$). The trends reflected those shown in abrasion injured fruit, with water stress tending to increase colour change in response to injury. The results are consistent with previous research on pears, where water stress was shown to increase friction discolouration (Amarante *et al.* 2001).

Greater cellular damage or disruption in water stressed fruit is the probable cause of the hydration effect on colour. Loss of turgor reduces cell elasticity, increasing the plastic deformation under a given strain. This would be expected to result in increased disruption of cell contents due to mechanical injury. Loss of turgor also increases the contact surface area, which may increase the area of damage in an abrasion injury.

4.4.3 Conclusions

Loss of turgor resulted in greater colour change in response to abrasion and vibration. Significantly greater relative changes in yellowing, darkening and loss of chroma were shown in water stressed fruit. Loss of around 4% moisture resulted in greater susceptibility to damage, while around 2% moisture loss had no significant effect. More careful handling would be recommended when fruit are less turgid, in particular towards the end of the postharvest chain. The greater resistance of well hydrated fruit to abrasion and vibration damage gives further incentive to the minimisation of postharvest moisture loss.

4.5 Effect of Surface Wetness on Abrasion and Vibration Injuries

In previous experiments on vibration, it was observed that damage tended to be more severe when condensation was present on the fruit surface. A trial was carried out to determine whether surface wetness had a significant effect on abrasion and vibration damage. A difference in injury levels due to surface wetness would have implications in postharvest handling, particularly during mechanical destalking and transportation.

4.5.1 Methods and Materials

‘KMP’ fruit were harvested in Byron Bay, and transported to Brisbane by car in damp paper towel over-wrapped with plastic. The fruit were placed into 5^{o}C storage for around one and a half weeks prior to treatment. The fruit were warmed to room temperature prior to treatment.

In a randomised control trial, wet and dry fruit were treated by abrasion or vibration injury. Wet fruit were dipped in water prior to treatment. The abrasion treatment was applied as three 15 cm rubs against a metal file, spread over the fruit cheek. The treatment was applied to a total of 15 dry and 15 wet fruit. When wet fruit were injured, the metal file was also wet with distilled water. The vibration treatment consisted of 10 minutes vibration using a flask shaker on setting 4. Separate boxes were used for wet and dry fruit, with 3 replicates of 10 fruit each. An additional 15 fruit were not subjected to any mechanical injury to act as a control. After treatment, fruit were stored at RT and 53% RH.

An additional 20 fruit from the same harvest were later subjected to additional abrasion treatments. The same technique was used, but up to 4 sites were treated on each fruit where possible. This resulted in 25 additional measurements of the effect of wet and dry abrasion.

Colour readings were taken in dry condition for all fruit prior to treatment, and after 24 hours storage. For vibration injured fruit, the perpendicular diameters of areas of scuff injury and darkened tips were recorded. Scuffs were defined as distinct severe areas of vibration injury, affecting more than 80% of the measured surface area. A score for severity was also recorded for each injury (1 = mild, 2 = moderate, 3 = severe and 4 = very severe).

4.5.2 Results and Discussion

Abrasion injury applied to wet fruit was shown to result in a greater change in hue, or more yellowing of colour than in dry fruit (t-test p=0.045) (Fig. 4.5.1). However, surface wetness did not significantly affect darkening of colour, or loss of chroma due to abrasion. It was observed that concise yellowish scuff marks were more likely to form when surface wetness was present, explaining the strong trend in yellowing.

For vibration treated fruit the analysis of colour changes was carried out on average values for each replicate box, resulting in only 3 values per treatment. This resulted in a large degree of error, and no significant differences were detected. The trends shown suggested a possible increase in hue and darker colour in fruit injured when wet (Fig. 4.5.2). Further experimentation, with a greater number of replicates, would be required to detect a significant effect.

The standard error between replicate boxes was consistently much higher in fruit injured when wet (Fig. 4.5.2). However, the variation observed between individual fruit within each run was lower when fruit were injured wet. For average hue values of each fruit, those injured when wet showed a standard error of 0.107 between fruit, while dry fruit had a standard error of 0.567. In contrast, the average variation within each fruit (between the 7 sites measured on each fruit) tended to be higher in wet injured fruit, with a hue standard error of 1.12, compared to dry fruit at 0.69. High variation between sites on an individual fruit suggests that greater localisation of injuries within each fruit may occur when fruit are injured wet. A group of fruit exposed to vibration treatment when wet would each have a similar degree of injury, but damage may be more likely to be localised to one severe injury on each fruit. In contrast, dry injured fruit may be more likely to show light marking scattered over the whole fruit (Fig. 4.5.3).

Figure 4.5.3 Typical vibration injuries on lychee fruit injured while dry (A) and wet (B, C) and an undamaged control (D).

Observation of the injury types occurring on vibration treated fruit showed that yellowish scuff injuries were more prevalent when fruit were wet (t-test p=0.013) (Fig. 4.5.4). Scattered darkening of protuberance tips was observed on many fruit as a symptom of vibration damage. Compared to the tip darkening caused by impact, this injury was lighter in colour, diffuse and less severe. The extent of vibration tip darkening occurring on the fruit was not affected by surface wetness.

The mechanism of surface wetness influencing damage is not clear. Research examining this effect in other types of fruit or vegetable could not be found. The response may thus be due to a specific feature of the lychee pericarp. It was thought that surface water may act as a lubricant during abrasion, thus decreasing the amount of energy required for the abrasive surface to slide across the fruit. Due to energy partitioning, this could result in more energy being dispelled into the tissue during an abrasion event, leading to increased damage. However, it is not clear why this effect would occur in lychee, but not have been observed in other fruit and vegetable varieties.

The effect of surface wetness on vibration injury has major implications in postharvest management. On some farms, lychees are deliberately kept wet during postharvest handling, in an effort to reduce desiccation browning. Ironically, this may actually increase browning, due to greater severity of vibration damage. In other cases, wetness may occur due to dew, rainfall, postharvest sprays or dips, or condensation. To minimise damage, it is important that procedures causing vibration are recognised, and that fruit are dry when vibration events occur. Mechanical destalking and transportation (both on-farm, and after packaging) would be the most common procedures causing vibration. Wet handling could still be a viable option, if fruit are not subjected to damaging levels of vibration or abrasion while wet.

Drying wet fruit prior to operations involving vibration would reduce damage, but could be difficult and time consuming. Uniform drying of the fruit surface in a short period would require some effort, and would increase moisture loss from the fruit. Prevention of wetness may be more easily achieved, as opposed to a drying process. Wetness prevention may involve harvesting fruit slightly later in the day, after dew has evaporated. The advantages of reduced vibration damage would need to be balanced against the lower moisture content of the fruit when picked later in the day. Avoiding harvest during or after rainfall would also prevent surface wetness. This is already practiced on some farms, as it has been observed by growers that harvesting in the wet can increase postharvest browning (Amos 2000, personal communication). The observed browning is likely to have been due to vibration damage. Prevention of wetness prior to vibration is the simplest method of minimising damage. Minimising wet vibration damage may be as easy as changing the order of postharvest procedures. For example, on one farm, fruit were placed into the destalker after pre-cooling, still damp with condensation. Pre-cooling is an important step, but may need to be relocated to a later point in the handling chain. Packing fruit dry and preventing subsequent condensation are important in avoiding rot development. The value of keeping fruit dry is further reinforced by the effect of surface wetness on vibration and abrasion damage.

4.5.3 Conclusions

The results clearly show that both vibration and abrasion injury can be exacerbated by surface wetness on the fruit. Surface wetness appeared to promote greater yellowing of the fruit skin in response to injury. Fruit damaged while wet showed an increased incidence of concise scuff marks. These results suggest that agitation of fruit should particularly be avoided when fruit are wet. This type of injury may be likely when wet fruit are placed into a mechanical destalker. It may also occur when fruit are harvested wet, and then transported by truck to the packing shed packed loosely in crates. These results may explain the observation made by some growers that the harvesting of wet lychee fruit often results in poor appearance and skin browning. The results also place additional importance on the control of condensation. Surface wetness on the fruit as a result of condensation is likely to increase the susceptibility of lychees to vibration and abrasion injuries during transport.

4.6 Effect of Fruit Temperature on Injury Development

Temperature generally has a large impact on the mechanical properties of a product, and thus on its sensitivity to damage. Temperature has been shown to have a significant effect on response to mechanical damage in many crops, including carrots (Kokkoras 1995), potatoes (Sitkei 1986), and cherries (Crisosto *et al.* 1993). Temperature effects can vary between crops, but generally cooler fruit are more easily damaged, due to brittle failure in the tissue (Hyde 1999). Temperature effects may also vary for different types of damage. In sweet cherries, cooler fruit were less susceptible to compression injury (Patten and Patterson 1985), and more prone to impact damage, while vibration injury was unaffected by temperature (Crisosto *et al.* 1993). This experiment aimed to determine the effect of fruit temperature on the development of mechanical injury in lychee.

4.6.1 Methods and Materials

The experiment was carried out on mature fruit of cultivar ‘KMP’, harvested in Yandina. Fruit were harvested into water and remained in water for at least one hour to ensure a high level of hydration prior to treatment.

Fruit were stored overnight at either 5 or 25^{o}C storage, with moist paper towels packed around the fruit to maintain a high RH. In a randomised control trial, mechanical damage treatments were applied to 30 fruit of each temperature;

- Impact by pendulum from level 2, using a round head with no added weights. This was estimated to apply an equivalent energy per unit area as a 67 cm fall onto a flat hard surface;

- Abrasion by 3 light 15 cm rubs against a metal file to each fruit, spread over the centre of one cheek;
- Compression using a penetrometer with a contact surface of approximately 6 mm diameter, holding a displacement of 6 mm depth for 10 seconds;
- Control fruit were not mechanically damaged.

From each treatment, 15 fruit were then placed into further storage at 5 or 25^{o}C with RH maintained at approximately 75%. After 24 hours post-injury storage, all fruit were transferred to 25^{o}C. The factorial experimental design thus incorporated 4 temperature regimes (Table 4.6.1), combining injury temperature (2 levels) and storage temperature (2 levels), tested for each of the 4 damage treatments, using a sample size of 15 fruit.

Table 4.6.1 Temperature regime treatments used in studying the effect of temperature on lychee mechanical damage.

Treatment	*Injured at:*	*Stored at:*	*Transferred to:*
1 (5/5)	5^{o}C	5^{o}C	25^{o}C
2 (5/25)	5^{o}C	25^{o}C	25^{o}C
3 (25/5)	25^{o}C	5^{o}C	25^{o}C
4 (25/25)	25^{o}C	25^{o}C	25^{o}C

Injury development was measured through single colour readings on the cheek centre. Readings were taken immediately before and after injury, and after 4, 24 and 48 hours.

For impact and compression treated fruit additional data were recorded three days after injury, including:

- the number and darkness of injured tips;

- the length and width of cracks, and any signs of weeping;
- injury dimensions, incidence of rots and discolouration.

4.6.2 Analysis

The analysis of the data was complicated by the direct effect of temperature on lychee skin colour. This effect was significant, as shown by comparison of the colour reading values prior to treatment (Fig. 4.6.1). Warmer fruit showed significantly lighter, more yellow and less intense colour than cool fruit (t-test $p<0.001$ for L-value, hue and chroma). The direct effect of temperature on colour was further complicated by the different temperature regimes, with each colour index showing a different response to temperature changes. In order to keep the analysis as simple as possible, colour reading values were corrected for the direct temperature effect by use of average control values. For each individual colour reading, the average control values from the same regime and time period were subtracted prior to analysis.

4.6.3 Results and Discussion

4.6.3.1 Direct Response of Pericarp Colour to Temperature Changes

The colour changes observed in control fruit were an interesting sideline in the experiment. The cooling of fruit had an immediate effect on colour, with 5^{o}C fruit appearing darker, more intense and more red-purple in hue (t-tests $p<0.001$) (Fig. 4.6.1). This would be assumed to improve visual appeal, as bright red colour is considered a desirable attribute in the fruit. The increased visual appeal of chilled fruit gives additional impetus to refrigeration of lychee fruit at the retail level.

The reason for the change in colour with varying temperature may simply be surface wetness caused by condensation. Cooler fruit would be expected to show condensation when removed to room temperature for measurement. While fruit were dabbed dry with tissue prior to colour measurement, the lychee surface remained slightly damp, and this may have been the major cause of the colour change observed. The influence of temperature on pigments may also have played a role. The colour change is unlikely to be related to moisture loss, as fruit were stored in very high humidity conditions during temperature equilibration.

Differences in moisture loss between the two storage temperatures may have occurred during later storage. Saturated sodium chloride was used to keep the RH close to 75% in both storage temperatures. At equilibrium, sodium chloride gives 75.65% RH at 5^{o}C, and 75.29% at 25^{o}C (Wexler 2000). However, in addition to RH, both fruit and air temperature affect the vapour pressure deficit, and hence the rate of moisture loss. Under conditions of fairly high humidity and no exposure to air currents, the maximum difference in moisture loss over a 24 hour period due to this effect would be around 1% of fruit weight, which is unlikely to result in a significant colour change unless fruit have already suffered substantial water loss.

Cool temperatures tended to cause darkening, (Fig. 4.6.2), with the 5/5 treatment significantly darker than the 25/25 (at 24 hrs, t-test p=0.026). This darkening remained significant even after all fruit had been shifted back to 25^{o}C for 24 hours (at 48 hrs, Mann-Whitney rank sum test p=0.042). In contrast, fruit kept at 25^{o}C throughout the storage period showed very little change in colour darkness.

Hue tended to be more red-purple at cool temperatures, with the 25/25 treatment significantly more yellow than the 5/5, from 0 to 24 hours (t-test p-values<0.023). Cool storage had no lasting effect on colour, with no significant differences in hue observed at 48 hours.

Chroma, or colour intensity, was significantly affected by the various temperature regimes (Figure 4.6.2), with all treatments significantly different from each other at 24 hours (t-tests and Mann-Whitney rank sum tests, p-values<0.03). Fruit stored at 5°C showed much greater intensity of colour, which was quickly lost once the fruit were warmed. The 25/5 treatment was significantly less intense in colour than the 5/5 treatment at 24 hours, despite having spent 24 hours at 5°C. It is possible that chilling of fruit soon after harvest may be of greater benefit in improving colour intensity than cooling after 24 hours warm. The various temperature regimes had no permanent effect on colour saturation, with very similar chroma values observed when fruit were returned to 25°C.

4.6.3.2 Effect of Temperature on Abrasion Injury

There were few significant effects observed in the analysis of abrasion response to temperature. The immediate lightening of colour observed after abrasion injury (Fig. 4.6.3) was significantly greater in cool fruit (analysis by t-test, $p=0.012$). However, this was a transient effect, and was not detrimental to fruit appearance. Cool fruit tended to show greater darkening in response to abrasion from 4 to 24 hours (t-tests between 5/5 and 25/25 treatments, $p<0.02$), this was not maintained when fruit were shifted to 25^{o}C. It is possible that this effect is an exaggeration of the direct effect of temperature on fruit colour. When compared to undamaged pericarp, the abrasion injury site may show a greater degree of colour fluctuation in response to temperature or dampness.

Temperature did not strongly affect the change in hue after abrasion, with no significant injury or storage temperature effects. Changes in chroma also showed few significant differences due to temperature. The 25/5 treatment showed significantly less loss of chroma than 5/25 and 25/25 treatments at 48 hours (t-tests $p<0.05$). This would suggest that warm injury temperature, followed by cool storage tended to minimize loss of chroma in response to abrasion.

4.6.3.3 Effect of Temperature on Compression Injury

The compression injury applied in this experiment involved the application of a constant displacement of 6 mm for 10 seconds. It was assumed that fruit temperature would not affect displacement under a given load, but this effect has not been tested. If temperature does affect displacement, the treatment effect may be invalid, being equivalent to different load rates.

Storage temperature appeared to affect the colour change resulting from compression injury (Fig. 4.6.4). Darkening of the tissue significantly increased when fruit were stored in warm conditions (at 48 hrs, Mann-Whitney rank sum test p=0.018), although this effect was evident only in fruit injured at 5°C. A significant response to storage temperature was also shown in the loss of chroma (Fig. 4.6.4). All treatments showed a similar loss of chroma at 24 hours, but after being returned to 25°C, a storage effect emerged. Fruit stored at warm temperatures showed a substantial loss of chroma from 24 to 48 hours, while fruit stored at 5°C showed very little change (t-test of storage effect at 48 hours, p = 0.031, data transformed by adding 13, then squaring). This effect was almost identical in fruit injured at both 5°C and 25°C. The increased loss of chroma from 24 to 48 hours, caused by storage at 25°C for 24 hours following injury, may be a lag response resulting from increased speed of biochemical reactions at warm temperatures.

In fruit stored at 25°C after injury, the number of tips darkened by compression was slightly higher in cold-injured fruit (Mann-Whitney rank sum test, p=0.018) (Fig. 4.6.5). This effect was not shown in fruit stored at 5°C. Fruit injured while warm showed significantly darker tip injury than cold-injured fruit when subsequently stored at 5°C (Mann-Whitney rank sum test, p<0.001). This effect also resulted in a significant difference in the index of tip darkness between warm and cold-injured fruit stored at 5°C t-test, p=0.019). However, in fruit stored at 25°C, injury temperature had no effect on tip darkness. The mechanism of this effect is not clear, as cold storage temperature would be expected to slow the development of damage, thus obscuring the effect of injury temperature. Tip darkening due to compression was less severe when fruit were injured and stored at the same temperature, with the 5 / 5^{o}C and 25 / 25^{o}C treatments showing the lowest damage indices.

Injury area due to compression was unaffected by injury temperature (Fig. 4.6.5). Storage temperature significantly affected injury area in fruit injured at 5^{o}C (Mann-Whitney rank sum test, p=0.034), with fruit stored at 25^{o}C showing a greater area of injury. This was probably due to the faster rate of biochemical reactions at warmer storage temperature.

The compression injury applied was generally not severe enough to cause cracking of the pericarp. Cracks were shown in a total of only 5 fruit. Significant differences between treatments were not detected due to the small number of fruit involved. The effect of temperature on compression cracking thus could not be established.

4.6.3.4 Effect of Temperature on Impact Injury

In general, colour changes resulting from impact injury were not strongly affected by fruit temperature (Fig. 4.6.6). Injury temperature did not significantly affect changes in colour in response to impact. Warm storage temperature resulted in greater yellowing in fruit injured at 5^{o}C after 24 hours (t-test, p=0.004). However, the effect was not shown in fruit injured at 25^{o}C. The greater yellowing observed in the 5 / 25^{o}C treatment may have resulted from faster biochemical processes at the warmer storage temperature. Impact injury can result in either increased or decreased hue, making it difficult to classify increased hue as a symptom of injury development. After fruit were returned to 25^{o}C, the various treatments showed no significant differences in hue.

In fruit injured at 25°C, storage temperature had a significant effect on loss on chroma. Storage at 25°C resulted in increased loss of chroma at 4, 24 and 48 hours after injury (t-tests, p-values = 0.047, 0.024 and 0.030, respectively). This trend is not repeated in fruit injured at 5°C and storage temperature showed no significant effect on tip damage. It is not clear why storage temperature differently affected fruit injured at different temperatures.

A strong response to injury temperature was observed in measurements of tip darkening (Fig. 4.6.7). Impact damage occurring at 25°C resulted in significantly darker discolouration (Mann-Whitney rank sum test $p<0.001$), and a greater number of tips injured (Mann-Whitney rank sum test $p=0.013$), compared to injury at 5°C. As a result, the tip darkening index (calculated as number of tips affected multiplied by darkness score) showed a very strong response to injury temperature, with fruit injured at 25°C showing significantly increased damage compared to fruit injured when chilled (Mann-Whitney rank sum test $p<0.001$). Injury at warm temperatures could result in greater damage due to changes in the mechanical properties of the tissue. Warm fruit may show greater tissue deformation than cool fruit (Somner *et al.* 1960), resulting in greater disruption to cell contents, and hence a stronger darkening response.

Storage and injury temperature did not significantly affect pericarp cracking (Fig. A4, in the Appendix). With only around one third of fruit showing any cracking in this experiment, data was dominated by zero values, making it difficult to detect any temperature effects. For this reason, an additional experiment was carried out to study the effect of injury temperature on impact cracking in greater depth.

4.6.4 Conclusions

Temperature had a significant transient effect on fruit colour, with chilled fruit appearing darker, brighter and more red-purple in hue. The visual appeal of fruit could be enhanced by refrigeration at retail level, to improve hue and colour saturation.

Injury and storage temperature had little effect on the development of abrasion injury. Cooler temperatures resulted in transient darkening of the injury site. It is likely that this effect resulted from greater fluctuation of colour in injured tissue, in response to either low temperature or surface wetness.

The effects of temperature on compression damage were complex. Storage at warm temperatures for 24 hours after injury resulted in substantial loss of chroma in the following 24 hours. Exposure to warm temperatures tended to increase darkening and injury area. Constant cool temperatures seemed to be optimal for minimising the symptoms of compression damage. Sweet cherries similarly show reduced compression injury at cool temperatures (Patten and Patterson 1985).

Temperature had little effect on colour changes in response to impact, however tip injury was observed to be more severe in warm-injured fruit. Injured protuberance tips were greater in number and darker in colour in fruit injured while warm. A similar response is shown in pears, with cool temperatures reducing impact damage (Baritelle and Hyde 2001). Changes in temperature affect the mechanical properties of the tissue. In lychee fruit, it appears that warm temperatures result in greater tissue damage in response to impact, possibly due to greater deformation.

There is some potential for reducing mechanical damage by temperature management. The benefits of temperature management would depend on the risk and type of damage and the resultant loss of value. For example, cool temperatures could be used to reduce protuberance damage due to compression and impact. However, given the limited commercial importance of these injuries, cooling fruit specifically to avoid tip darkening is unneccesary.

4.7 Effect of Fruit Temperature on Cracking Due to Impact

Cracking of the lychee pericarp is a major defect, capable of rendering fruit unmarketable when severe. Previous research has shown that warmer lychee fruit are more susceptible to protuberance tip darkening (Section 4.6.3.5). This experiment was carried out to determine whether temperature significantly affects cracking. Knowledge of the effect of fruit temperature on cracking susceptibility could be used to improve postharvest handling procedures. Fruit temperature is easily manipulated, and could be adjusted to minimise the chance of impact cracking during high-risk procedures.

4.7.1 Methods and Materials

Fruit of cultivar ‘KMP’ were harvested at Byron Bay. Fruit were tightly packed in damp paper towel, over-wrapped with plastic, and were transferred to Brisbane by car. After storage at 5^{o}C for one week, the fruit were transported to the Sunshine Coast. Randomly sorted fruit were packed in damp paper towel and stored overnight, at either 5 or 25^{o}C. In a randomised trial, a total of 55 fruit from both 5^{o}C and 25^{o}C storage were subjected to a single impact to the cheek centre, using a round head with no added weights from level 4. The impact energy per unit area was estimated to be equivalent to a 114 cm fall onto a flat surface. After impact, fruit were stored at 53% RH at RT.

Crack width and length were recorded immediately after injury, and after 24 hours storage. A score was given for the extent of penetration of the crack through the pericarp (1 = closed crack, not penetrating through to the aril, 2 = weeping or open crack, penetrating the full thickness of the pericarp, but with aril not visible, 3 = severe open crack, with aril visible). Tip damage was assessed 2 days after injury. The number of tips at each level of severity score (1 = light, 2 = light-moderate, 3 = moderate, 4 =

moderate-severe, 5 = severe) was recorded. The severity of tip injury was based on both the area affected, and the darkness of colour. A tip damage index was calculated as the sum of the number of tips affected multiplied by severity score.

4.7.2 Results and Discussion

Fruit showed greater susceptibility to tip damage when injured while warm (Fig. 4.7.1), confirming the trend previously observed (Section 4.6.3.5). Fruit injured at 25^{o}C showed a greater number of tips in the score range 3-5 (moderate-severe) (Mann Whitney rank sum test $p<0.001$). The index of tip darkening was also significantly higher (t-test $p<0.001$).

Analysis of total crack area detected no significant temperature effect (Fig. 4.7.2). Over the 24 hour period following injury, cold-injured fruit showed a substantially greater increase in the number of closed cracks (Mann Whitney rank sum test, $p<0.001$). On average, each cold fruit showed 1.75 new cracks, while warm fruit showed only 0.67 new cracks per fruit. This suggests that closed cracks were more likely to be overlooked while fruit were chilled. The reduced visibility of cracks may be due to condensation, with surface wetness masking cracks. This observation has implications in packing line procedures. Sorting for cracks may be more effective if carried out on dry fruit. Closed cracks tended to become larger and more visible with storage. It appeared that drying of the pericarp in storage pulled the edges of the crack apart. In contrast, open wounds tended to decrease in area. This was possibly due to the loss of turgidity in the aril. In fresh wounds, the pressure of the aril often forced the edges of the crack apart.

The percentages of fruit affected suggest increased open cracking in chilled fruit (Fig. 4.7.3). At the 24 hour assessment, open cracks occurred in around 43% of chilled fruit, compared to 30% of warm fruit. The incidence of cracking resistance was higher in warm fruit, with 5% showing no cracks 24 hours after injury. In comparison, all chilled fruit showed some signs of cracking after 24 hours. There was little difference in the incidence of closed cracks between the two injury temperatures.

4.7.2.1 Correlations

There was a strong negative correlation between the area of open and closed cracks (at 24 hours, Spearman correlation co-efficient = -0.674, p<0.0005). If an open crack resulted from the impact, there was a reduced chance of closed cracks also occurring. Some negative correlation was also shown between cracks and tip damage. Immediately after injury, the total crack area was negatively correlated to the number of tips of severity score 4 to 5 (Spearman correlation co-efficient = -2.37, p=0.0121). Similarly, 24 hours after injury, the area of open cracks was negatively correlated to the number of tips of severity score 4 to 5 (Spearman correlation co-efficient = -0.222, p=0.0190). Crack area was not significantly correlated to the index of tip damage, but p-values of less than 0.1 were observed at 24 hours, suggesting a possible weak correlation. The negative correlation between different types of damage probably occurs due to energy partitioning. Each fruit received the same load, or amount of energy. If energy is spent creating a crack, less is available to deform the tissue. The formation of a crack, particularly an open crack, also releases the tension on the skin, effectively cushioning the impact.

4.7.3 Conclusions

The results confirm that lychee fruit at warm temperatures sustain greater tip injury due to impact. A similar response is shown in pear, with reduced impact damage occurring at cool temperatures (Baritelle and Hyde 2001). A significant effect of fruit temperature on the area of pericarp cracking could not be detected. However, comparison of the percentage of fruit affected by cracking revealed that cold fruit are slightly more susceptible to open cracking. This response is similar to that shown in potato, with the risk of brittle failure increasing at cool temperatures (Hyde 1999). A negative correlation was shown to exist between cracking and tip damage. Avoidance of cracking is a higher priority than reducing tip damage, due to the severe influence of cracking on fruit quality. Therefore, in procedures involving some risk of impact damage, it is recommended that fruit be handled in a warm state to minimise open cracking.

4.8 Effect of Fruit Temperature on Vibration Injury

Temperature can affect the mechanical properties of fresh produce, and hence their response to mechanical damage. Research was carried out to determine whether the extent of lychee vibration damage was influenced by injury temperature.

4.8.1 Methods and Materials

'Wai Chee' fruit were harvested from Byron Bay, and were soaked in water for an hour immediately after harvest. They were then dried, and packed in damp paper towel for transport to the lab. The fruit were numbered and weighed, and colour was measured at seven sites on each fruit (base, cheeks, sides and shoulders). Fruit were randomly allocated to either 5^{o}C or 25^{o}C storage overnight.

The next day the fruit were treated by vibration, using a flask shaker at setting 4 for five minutes, with 16 fruit (approximately 315 g) in each box. In a randomised control trial, fruit were treated in groups of 4, with treatments applied in random order. The vibration treatment was carried out at the same temperature as storage. The additional 12 fruit in the box during each run were at the same temperature as the sample. The 5^{o}C treatment was carried out within a cool room, with a marble slab used to reduce dampening of the vibration. A total of 40 fruit (10 replicate runs of 4 fruit) were treated at 5^{o}C and 25^{o}C. An additional 16 fruit at each temperature were treated as controls. The controls were subjected to the same changes in temperature as the treated fruit, but were not injured by vibration.

After treatment, the fruit were packed in damp paper towel, and stored at 25^{o}C. Colour readings were taken when the fruit were at room temperature and air dry. On each fruit, vibration damage was scored for severity from 0-6 (0=none, 1=very mild, 2=mild, 3=mild-moderate, 4=moderate, 5=moderate-severe, 6=severe), and the surface area of injury was estimated. Scuffs, defined as concise areas of moderate to severe damage, were also scored for severity, and two perpendicular diameters were measured. Indices were calculated by the sum of score multiplied by area.

4.8.2 Results and Discussion

There was little difference in the colour change response to vibration between fruit at 5^{o}C and 25^{o}C (Fig. 4.8.1). Changes in hue and L-value were not significantly affected by temperature. The loss of chroma was significantly greater in fruit injured at 25^{o}C than at 5^{o}C (t-test, $p<0.001$). However, this difference was primarily due to the direct effect of temperature on colour. When average control values for each temperature were subtracted from individual sample values, this difference was not significant.

Damage indices were also fairly similar in fruit injured at 5^{o}C and 25^{o}C (Fig. 4.8.2). Fruit injured at 25^{o}C showed significantly greater area of vibration damage than those injured at 5^{o}C (t-test, $p=0.028$). The severity of vibration damage, total damage index and scuff index were not significantly influenced by temperature.

Fruit weight significantly influenced damage to fruit in this treatment, with smaller fruit showing greater damage. Weight was correlated with changes in both chroma (Spearman correlation coefficient= 0.356, $p<0.0005$) and hue (coefficient= -0.320, $p<0.0005$). This effect may be specific to the size of the box used in treatment, as smaller fruit would be more capable of moving about within the box, thus potentially sustaining more damage.

4.8.3 Conclusions

Fruit temperature does not appear to have a substantial effect on vibration damage in lychee. Warmer fruit tended to show slightly greater damage, but the effect was not strong, with significant differences observed only in the surface area of damage. Similar results were observed in sweet cherries, with temperature having little effect on vibration injury (Crisosto *et al.* 1993).

4.9 Effect of Individual Fruit Morphology on Cracking Susceptibility

In previous experiments, it was observed that cracking susceptibility during impact varied widely between seemingly similar fruit. A trial was carried out to study the effect of individual fruit morphology on cracking susceptibility. A greater understanding of the influence of fruit morphology on cracking susceptibility may explain observed cultivar differences, and assist in predicting crack resistance in cultivars not yet tested.

4.9.1 Methods and Materials

‘Brewster’ lychees were purchased from the Brisbane markets, and were stored at 4^{o}C for 2 days, then at RT for 2 days prior to treatment. Fifty fruit were numbered and weighed, and the dimensions of each fruit were measured using digital calipers. The length from shoulder to base, and two perpendicular widths, across sides and cheeks were noted. Each fruit was scored for spikiness of the skin (1 = almost flat surface; 5 = very spiky), overall fruit shape, or fill (1 = unfilled, slightly hollow appearance; 5 = full, rounded) and angles of curvature (looking at the impact site both from the side, and from above). Angle of curvature scores were estimated by holding the fruit against a range of drawn curves. The equivalent radii of the scores are shown in Table 4.9.1.

Table 4.9.1 Angle of curvature scores for lychee and range of equivalent radii.

Score	Equivalent radii (cm)
1	1.80 – 1.98
2	1.98 – 2.26
3	2.26 – 2.70
4	2.70 – 5.10
5	>5.10

Each fruit was subjected to a level 2 pendulum impact to the cheek, using a flat head and 2 added weights, estimated to be equivalent to a 45 cm fall. Fruit that cracked were deemed susceptible, and were not exposed to further impact. Uncracked fruit were subjected to a further 2.5 level impact (equivalent to a 60 cm fall). Fruit that cracked at this point were deemed to be of average susceptibility. Uncracked fruit were deemed resistant, and were exposed to a further level 3 impact (81 cm). Crack dimensions were recorded. Fruit were then cut open, and the seeds were cleaned, air-dried and weighed.

An additional experiment was carried out on ‘Wai Chee’ and ‘KMP’ fruit to determine whether seed size affected cracking in other cultivars. A group of fruit (23 ‘KMP’ and 25 ‘Wai Chee’) were each subjected to a single impact (level 4, using a flat head with 2 added weights, equivalent to 122 cm fall). Fruit were scored as either having no cracks, closed or open cracks (none = 0, closed = 1 and open = 2). Seeds were removed, cleaned and air-dried prior to weighing.

4.9.2 Results and Discussion

4.9.2.1 The Effect of Seed Size on Pericarp Cracking

Factors affecting cracking susceptibility included seed weight, fruit diameter and the spikiness of the skin. Seed weight had a surprisingly strong effect on the height at which cracking occurred (ANOVA by ranks $p=0.001$). Large seed size was strongly associated with cracking resistance (Spearman correlation co-efficient=0.477, $p<0.0005$) (Fig. 4.9.1). Seed size may influence the mechanics of impact, providing structural stability at the core of the fruit. A fruit with a very small seed would probably undergo greater deformation of the pericarp, due to the highly elastic properties of the aril. In contrast, a large seed would not deform substantially, thus reducing displacement of the surface tissues during impact. No other measured factor showed as strong a correlation with cracking susceptibility as seed size.

The ‘Brewster’ cultivar shows a large degree of variation in seed size, with around half of all fruit in this experiment showing an aborted seed, commonly referred to as “chicken tongue” (Fig. 4.9.2). Despite lower variation, seed size also significantly affected cracking resistance in ‘KMP’ and ‘Wai Chee’ (Fig. 4.9.3). In ‘KMP’ fruit a strong correlation existed between seed size and cracking susceptibility (Spearman correlation co-efficient= -0.613, p=0.00193). In ‘Wai Chee’, the correlation was not significant, but fruit showing open cracks had a significantly smaller seed size than fruit with closed or no cracks (t-test p=0.040). The observation of the same trend in three different cultivars confirms that seed size has a consistent influence on lychee pericarp cracking.

Figure 4.9.2 Large seed (A) and “chicken tongue” seed (B) in lychee.

Small seed size is commercially attractive, due to the increased proportion of pulp. Cultivars with a high percentage of chicken tongue seeds are generally well regarded. ‘Salathiel’, ‘KMP’ and ‘Wai Chee’ generally have fairly high levels of chicken tongue seed (Menzel and Simpson 1986). These cultivars may require extra care to avoid pericarp cracking. In contrast, cultivars with large seeds, such as ‘Bengal’, may be able to tolerate greater falls. The proportion of fruit with chicken tongue seed can vary substantially between orchards and over seasons (Menzel and Simpson 1986). It is probably necessary to manage impact thresholds assuming small seed size, as most growers handle a range of cultivars using the same equipment, and most cultivars show a significant level of chicken tongue seed.

The seed size effect provides some rationale to the variation between similar fruit, but is obviously not the only factor affecting cracking susceptibility. Around 25% of the ‘Brewster’ fruit used in this experiment showed open cracks after an impact equivalent to only 45 cm. In contrast, the ‘KMP’ and ‘Wai Chee’ fruit showed around 35% open cracking at 122 cm fall height. The huge difference in cracking susceptibility between cultivars was not explained by any of the characteristics measured in this experiment. Other factors, such as the shape, adhesion and arrangement of cells in the pericarp may be substantially more important than the gross features of the fruit.

4.9.2.2 Other Factors Affecting Skin Cracking

Fruit dimensions were significantly correlated with crack susceptibility (Fig. 4.9.1), with fruit length showing a particularly strong relationship (Spearman correlation co-efficient=0.394, p=0.00487). Crack resistant fruit had a significantly larger average diameter than other fruit (ANOVA p=0.003). Fruit width measured from side to side (co-efficient=0.365, p=0.00934) had a greater influence than the width across the cheeks (co-efficient=0.315, p=0.0260). In all cases larger fruit showed a reduced susceptibility to cracking. Fruit dimensions were very strongly correlated with seed size (for the average diameter, co-efficient=0.770, p<0.0005), so the correlation between cracking susceptibility and diameter may occur simply due to this relationship.

The spikiness of the skin surface was the only other measured characteristic showing a significant correlation with the height at which cracking occurred (co-efficient= -0.362, p=0.00997). Spiky textured fruit were more susceptible to cracking.

However, the spikiness score was also correlated with seed size (co-efficient= -0.463, p<0.0005). Spiky fruit tended to have smaller seeds, so this may have been the mechanism of greater cracking susceptibility.

Other characteristics measured were not significantly correlated with cracking susceptibility. Despite being correlated with seed size and diameter, fruit weight was not significantly correlated with cracking susceptibility. Fruit density also had no significant influence. Skin thickness, measured at the protuberance tips, did not affect cracking. Thickness at the protuberance bases would have been more relevant, as failure during cracking occurs between protuberances, but measurements were not taken. Angles of curvature were not a significant factor in cracking susceptibility, but this was probably due to the limited size of the impacting head. Sharper angles tend to concentrate the impact load into a smaller contact surface area, hence increasing the potential damage. At the impact levels used in this experiment, the full face of the impact head would have been in contact with the fruit surface. The contact surface area would therefore be expected to be constant, at around 3.1 cm^2 for all impacts. In an impact against a larger surface, as would occur in practice, the angle of curvature would be expected to have a significant effect on contact surface area, and hence impact damage.

When cracks did occur, numerous factors were related to the crack width. In contrast, crack length appeared to be unaffected by any of the parameters measured. Cracks tended to be narrower in fruit with larger seeds (co-efficient= -0.555, p<0.0005) and greater average diameters (co-efficient= -0.369, p=0.00867). Fruit that were wider across the sides than the cheeks (higher width 1:width 2 ratio) showed reduced crack width (co-efficient= -0.413, p=0.00298). This may be due to increased transverse growth in fruit with fuller cheeks. Transverse growth in lychee occurs due to expansion of the aril, and results in greater pressure on the pericarp (Kumcha 1998). The internal pressure from the aril would be expected to push the crack edges apart, resulting in a wider wound. The angle of curvature at the impact site was correlated with crack width (co-efficient= -0.347, p=0.0139), with sharper curves tending to result in wider cracks. This may also be related to the pressure of the aril, as the average angle of curvature was strongly positively correlated with the width ratio.

4.9.3 Conclusions

Seed size was a major factor influencing cracking susceptibility in lychee, with larger seeds giving improved resilience. The relationship was consistent in the three cultivars tested. In contrast, larger seed size in peaches increased impact damage (Menesatti *et al.* 1999). The differing mechanisms of impact damage are the likely cause of these contrasting results, with peaches showing largely internal injury, while lychee injury is to the pericarp. It was theorised that in lychee the mechanical properties of the seed could reduce deformation of the surface tissues by acting as a solid core.

Fruit diameter and skin spikiness were correlated with cracking susceptibility in lychee. However, these characteristics were also strongly related to seed size, and this may have caused a significant effect. The results provide some explanation of the inherent variation in cracking susceptibility. However, the variation in resilience between the cultivars tested could not be explained by any of the parameters measured.

4.10 Variation Between Cultivars in Susceptibility to Mechanical Damage

A large degree of variation in response to mechanical damage between cultivars has been observed in previous experimental results. For example, fruit of cultivar ‘Brewster’ appeared to be highly susceptible to impact cracking, with many fruit cracking due to a 45 or 60 cm fall (Section 4.9.2.1). In the same experiment, ‘KMP’ and ‘Wai Chee’ fruit required an impact equivalent to a 122 cm fall to show a similar level of cracking. Research was carried out to compare susceptibility to mechanical damage in a range of commonly grown varieties.

4.10.1 Methods and Materials

Fruit from four different cultivars grown at Sippy Downs, SE Queensland, were harvested over a three week period. In order of harvest date, the cultivars were ‘Bengal’, ‘KMP’, ‘Wai Chee’ and ‘Salathiel’. Conditions at harvest were fairly uniform, all occurring under scattered cloud, with temperature ranging from 26-30^{o}C and RH from 45-57%. The fruit were harvested into water and soaked for at least 30 minutes, and were dried and packed in damp paper towel for transport.

Ten fruit from each cultivar were randomly selected, and placed into 20^{o}C storage overnight in damp paper towel. These fruit were used for the measurement of gross anatomy and Brix : acid ratio. Gross anatomy measurements were taken after overnight storage, and included the weight and dimensions of each fruit (length and two perpendicular widths, across the sides and cheeks). A disc of skin was removed from the cheek centre of each fruit using a cork borer, and the number of protuberance tips counted. The skin thickness at protuberance tip and base was measured using digital callipers. The seed was removed from each fruit, cleaned, air-dried and weighed. Arils were stored in plastic bags for up to one week at 4°C prior to measurement of Brix : acid ratio. Acid levels were measured by titrating 5 ml samples of aril juice against 0.1 M NaOH to an endpoint of pH 8.2.

All other fruit were stored at 20°C in damp paper towel for 2 days prior to treatment, and were then removed and allowed to come to room temperature. Colour was measured on both cheeks of each fruit prior to treatment. For vibration treated fruit colour readings were taken at 7 sites (cheeks, sides, shoulders and base). In a randomised control trial, treatments were applied to 20 replicate fruit of each cultivar:

- Impact by pendulum using the flat impacting head with 2 added weights from level 2.5, estimated to be equivalent to a 60 cm fall;
- Vibration for 10 minutes at setting 4, treating 5 fruit per run and with a total of approximately 370 g fruit in each run;
- Abrasion against a metal file, applied by three 15 cm rubs spread over one cheek;
- Compression of 1 kg weight applied for 2 hours, using the compression device described in Section 1.6.4.5.1.
- Control treatment, with no mechanical injury applied.

Water potential was measured in a total of ten fruit from each cultivar, with measurements taken at regular intervals during the application of treatments. After injury, fruit were stored at 20°C and 53% RH. Colour was re-measured after 24 hours, and any cracks were recorded.

The impact treatment, equivalent to a 60 cm fall, caused very low levels of cracking. For each cultivar, additional fruit were subjected to a range of impact levels as a gauge of cracking susceptibility. Between 45 to 65 fruit of each cultivar were treated by a single impact, using a flat head and 2 extra weights. Impact heights were either level 3 (81 cm), level 4 (122 cm), level 5 (168 cm) or level 6 (223 cm). Open and closed cracks were noted when they occurred.

4.10.1.1 Analysis

Due to the presence of a direct cultivar effect on colour change, average control values for each cultivar were subtracted from individual measurements. The values analysed were therefore purely resulting from the response to mechanical injury. Vibration injured fruit were analysed using the average values from each run (N=4).

4.10.2 Results and Discussion

4.10.2.1 Cultivar Characteristics

Water potential was fairly consistent across cultivars (Fig. 4.10.1). Previous water potential measurements on fully hydrated fruit (-1 MPa (Fig. 4.1.1)) suggest that the fruit in this experiment had not suffered significant moisture stress. ‘KMP’ showed significantly greater water stress than ‘Salathiel’ and ‘Bengal’ (ANOVA p=0.002, comparison by Tukey test). Based on relationships observed in the previous experiment (Fig. 4.1.1), this may equate to around 1.4% more weight loss (as a percentage of fruit weight) having occurred in the ‘KMP’ fruit. The difference is likely to have resulted from reduced water availability prior to harvest.

The typical characteristics of each cultivar are summarised in Table 4.10.1. ‘KMP’ fruit had the largest average weight, and ‘Salathiel’ were considerably smaller than other cultivars. ‘Bengal’ fruit had the largest diameter, and substantially lower density than other cultivars. ‘Bengal’ and ‘Salathiel’ fruit had a more elongated shape than the almost spherical ‘Wai Chee’ and ‘KMP’. ‘Bengal’ fruit tended to have the thickest skin and greatest height and density of protuberances. ‘Salathiel’ fruit had thin skin, and the lowest protuberance height. Brix : acid ratios were similar in all cultivars, indicating that fruit maturity was not substantially different. Seed size varied widely between cultivars. The level of chicken tongue (aborted) seeds ranged from 100% in ‘Salathiel’ to none in the ten ‘KMP’ fruit tested. ‘Bengal’ fruit showed 30% chicken tongue, and ‘Wai Chee’ only10%. Average seed size was greatest in ‘Bengal’, with seeds generally weighing 3 to 4 g when not aborted. Non-aborted seeds in ‘KMP’ and ‘Wai Chee’ were of moderate size, averaging 2.28 and 1.88 g respectively. These observed differences in the properties of the fruit may help to explain differences in response to mechanical injury.

Table 4.10.1 Basic characteristics of the lychee cultivars tested.

Cultivar:	**‘Bengal’**		**‘KMP’**		**‘Wai Chee’**		**‘Salathiel’**	
Characteristic:	Avg.	SE	Avg.	SE	Avg.	SE	Avg.	SE
Fruit weight (g)	21.86	1.17	22.67	0.51	20.58	0.62	15.94	0.52
Average diameter (mm)	36.47	0.69	34.95	0.24	33.84	0.36	30.84	0.35
Length: width ratio	1.22	0.02	1.01	0.01	0.99	0.01	1.13	0.01
Density (gcm^{-3})	0.87	0.01	1.01	0.01	1.01	0.01	1.04	0.01
Protuberance tips / cm^2	11.6	0.59	7.9	0.28	6.49	0.48	7.28	0.64
Base skin thickness (mm)	1.64	0.08	1.19	0.05	1.35	0.04	1.11	0.03
Tip height (mm)	1.08	0.06	0.80	0.07	0.56	0.06	0.42	0.07
TSS (Brix^0)	17.6	0.36	17.7	0.36	17.0	0.28	16.7	0.27
Brix : acid ratio	73.5	4.32	70.5	7.13	75.7	3.77	71.6	2.09
Seed weight	2.78	0.54	2.28	0.07	1.72	0.21	0.34	0.02

N=10.

4.10.2.2 Colour Changes in Control Fruit

Significant differences existed between cultivars in the colour change occurring in control fruit during storage (Fig. 4.10.2). All cultivars tended to show slight darkening of colour, but this was significantly less in ‘Bengal’ compared with ‘Salathiel’ and ‘KMP’ (ANOVA p=0.005, comparison by Tukey test). ‘Bengal’ and ‘KMP’ showed a trend towards more red-purple colour over the 24 hours storage. In contrast, ‘Salathiel’ and ‘Wai Chee’ showed minimal change in hue. A slight loss of chroma was shown in all cultivars except ‘Bengal’. Average control values were subtracted from each individual measurement to remove these direct cultivar effects on colour change.

4.10.2.3 Colour Changes in Response to Vibration and Abrasion

Changes in darkness and hue in response to abrasion did not significantly differ between cultivars (Fig. 4.10.3). ‘Salathiel’ showed significantly greater loss of chroma after abrasion injury than ‘KMP’ or ‘Wai Chee’ (ANOVA p=0.003, comparison by Tukey test). This trend was more extreme in response to vibration injury, with ‘Salathiel’ showing substantially greater loss of colour intensity (ANOVA p<0.001, comparison by Tukey test) (Fig. 4.10.4). Loss of chroma was more than three times greater in ‘Salathiel’ than other cultivars. ‘Salathiel’ fruit also showed much stronger yellowing of colour due to vibration, which was at least twice that of other cultivars.

The cultivar ‘Salathiel’ showed a strong susceptibility to both abrasion and vibration. This may have occurred partially due to the protuberance shape, which was the lowest in height of all cultivars tested. An abrasion injury could be exacerbated by flatter protuberances, as more skin surface area would be in contact with the abrasive surface. In contrast, a very spiky textured fruit would sustain damage only to a small area on the protuberance tips. ‘Salathiel’ fruit appeared to be particularly susceptible to vibration. This may have resulted from the small size of the fruit. The total weight of fruit in each box during vibration treatment was kept constant across cultivars. However, the small size of ‘Salathiel’ fruit would allow them to move more freely within the box, potentially leading to greater damage. This effect would be expected to be less extreme in practice, as fruit would be more tightly wedged into the box.

4.10.2.4 Colour Changes in Response to Impact and Compression

The compression treatment caused very minimal change in colour, generally resulting in slight darkening and loss of chroma (Fig. 4.10.5). The ‘Bengal’ fruit tended to show slightly greater loss of chroma, resulting in a significant difference to ‘KMP’ (ANOVA p=0.033, comparison by Tukey test). The impact treatment used was not severe enough to cause any substantial changes in colour, and no significant differences between cultivars were detected (Fig. 4.10.6). ‘Bengal’ and ‘Wai Chee’ tended to show the greatest loss of chroma in response to both impact and compression. The analysis of combined impact and compression data detected a significant difference. ‘Bengal’ fruit showed significantly greater loss of chroma than both ‘Salathiel’ and ‘KMP’ (ANOVA, p=0.002, comparison by Tukey test). This could result from biochemical differences in the tissue. A greater number of replicates, and more extreme impact and compression loads would be needed to clarify cultivar differences. However, tip darkening type injuries have little impact on fruit appearance, and are likely to be of limited commercial importance.

4.10.2.5 Cultivar Differences in Cracking Response Due to Impact and Compression

The compression injury resulted in cracking in only 2 of the 80 fruit treated, and the cracks were minor, not penetrating through to the aril. The limited number of fruit affected prevents any comparison of cultivars.

Cracking due to impact did not vary hugely between cultivars, but slight differences were observed. ‘Bengal’ fruit showed no cracking at an impact height equivalent to 60 cm, while ‘Wai Chee’, ‘KMP’ and ‘Salathiel’ showed closed cracking to 25, 15 and 5% of fruit respectively (Fig. 4.10.7). An impact equivalent to 122 cm resulted in cracking to 43% of ‘Bengal’ fruit, but caused 80 to 83% cracking in other cutlivars. Open cracking was not observed at the level 2.5 (60 cm) impact, but emerged at level 3 (81 cm) (Fig. 4.10.8). The percentage of fruit affected by open cracks at level 3 ranged from 5% in ‘Wai Chee’ to 10% in ‘KMP’. Overall, ‘Bengal’ fruit were the most resistant to severe cracking, with 64% of fruit showing open cracks after an impact equivalent to 168 cm. In all other cultivars, this level of impact resulted in open cracks to all fruit treated.

Numerous differences existed in the gross anatomy of the ‘Bengal’ fruit compared to other cultivars, including lower fruit density, more elongated shape, thicker skin, heavier seed and greater concentration and height of protuberances, giving a more spiky texture. The thickness of the skin at the protuberance base may contribute to the resilience of the ‘Bengal’ cultivar to cracking. ‘Bengal’ fruit showed an average thickness of 1.64 mm, compared to 1.35, 1.19 and 1.11 mm in ‘Wai Chee’, ‘KMP’ and ‘Salathiel’, respectively. Greater skin thickness may give increased strength to prevent skin cracking. Further experimentation, with measurements of base skin thickness and cracking on numerous individual fruit would be useful to confirm this effect. The large seed size of the ‘Bengal’ fruit may also be important, as seed size confers some cracking resistance (Section 4.9.2.1). However, ‘Salathiel’ fruit did not show greater susceptibility than ‘KMP’ and ‘Wai Chee’, despite a much smaller average seed size. Many interacting factors could potentially affect skin cracking, making it difficult to isolate individual variables.

4.10.2.6 Incidental Evidence of Cultivar Variation in Cracking Susceptibility

Data from numerous experiments (Sections 2.1, 2.8, 2.9, 2.10, 2.11, 2.13, 3.1, 3.4, 3.5, 3.6, 4.1, 4.2, 4.6, 4.7 and 4.9) was compiled to determine whether any strong trends in cracking susceptibility were apparent. The data were dominated by cultivars ‘KMP’ and ‘Tai So’, and did not reveal any strong cultivar differences in total cracking (Figure 4.10.9) or open cracking (Figure 4.10.10). Hydration was roughly indexed by categorising fruit as either soaked or unsoaked. As previously mentioned, ‘Brewster’ fruit showed a high level of susceptibility, particularly for dehydrated fruit. However, the cultivar was tested in only one experiment (Section 4.9). The large variation across experiments may have resulted from factors such as hydration, method of impact application and impact head shape. The compilation of data does not provide any additional evidence of consistent cultivar variation in susceptibility to impact cracking.

4.10.3 Conclusions

‘Salathiel’ fruit were highly susceptible to abrasion and vibration injuries, showing a greater level of damage than other cultivars. The increased susceptibility may have resulted from low protuberance height allowing greater contact with the abrasive surface. Small fruit size in ‘Salathiel’ may also contribute to greater vibration damage by allowing greater movement within the box. There was some evidence of greater loss of chroma in ‘Bengal’ in response to impact and compression injuries, which may have occurred due to biochemical differences in the tissue. ‘Bengal’ fruit showed substantial resistance to cracking compared to the other cultivars tested. The large seed size and thick skin of this cultivar may have contributed to its resilience. These results support previous research showing that strong cultivar differences can occur in response to mechanical damage, as seen in cherries (Burton and Schulte-Pason 1987). Replication across seasons and farms would be required to draw solid conclusions regarding cultivar susceptibility. The addition of other cultivars would also be valuable. However, the experiment gives some indication of the variation in resilience between some commonly grown cultivars.

4.11 Overview - Fruit Properties Affecting the Response of Lychee to Mechanical Damage

Fruit water content strongly affected the response to impact injury, with turgid lychees more susceptible to impact cracking. A similar response is shown in potato, with cracking more common in hydrated tubers (Thornton *et al.* 1973). For lychee, both the incidence and size of cracks were reduced in mildly water stressed fruit (Section 4.2). Mild water stress also reduced tip darkening injury, and in some cases increased background colour change (Section 4.1). Turgid fruit require more careful handling and lower tolerable fall thresholds (less than 40 cm fall height).

The mechanical properties of lychee tissue under compression were strongly affected by hydration (Section 4.3). Moderate water stress resulted in loss of elasticity and greater deformation of tissue, a typical response (Baritelle and Hyde 2001).

Water stressed lychee fruit showed increased yellowing, darkening and loss of chroma in response to abrasion and vibration damage (Section 4.4). Similar results have been described in pears, with reduced hydration resulting in greater susceptibility to friction discolouration (Amarante *et al.* 2001). In lychee, around 2% moisture loss caused little change, but 4% loss from the fruit exacerbated damage.

Surface wetness on the fruit also strongly affected abrasion and vibration damage (Section 4.5). Wet fruit showed increased yellowing of skin colour and greater incidence of scuff. No evidence of this type of response in other fruits could be found in previous research. These results further increase the importance of condensation control in lychee.

Fruit temperature had an immediate effect on colour, with chilled fruit appearing darker, brighter and more red-purple in hue (Section 4.6). Temperature had little effect on abrasion (Section 4.6) and vibration injury (Section 4.8). Temperature effects on compression were complex, with constant cool temperatures seeming to minimise damage. Impact tip injury was lessened at cool temperatures (Section 4.6), while open cracking appeared to increase slightly in cold fruit (Section 4.7). An increase in brittle failure is a typical response to cooler temperatures in fruit and vegetables (Hyde 1999).

Large seed size was shown to strongly reduce cracking susceptibility, possibly due to a solid core in the fruit reducing surface tissue deformation (Section 4.9). The small-seeded “chicken tongue” fruit most desired by consumers may require careful handling to avoid cracking. Large fruit diameter and reduced skin spikiness were also associated with cracking resistance, but these characteristics were strongly correlated with seed size. None of the parameters tested explained the major differences in cracking susceptibility observed between cultivars.

Cultivar differences were observed in susceptibility to vibration, abrasion and impact injury (Section 4.10). Small smooth-skinned ‘Salathiel’ were susceptible to vibration and abrasion damage. ‘Bengal’ fruit, with large seeds and thick skin, were strongly resistant to impact cracking, while ‘Brewster’ were susceptible (Section 4.9).

Various fruit properties were shown to affect the response of lychee fruit to mechanical damage. The management of factors such as fruit wetness, temperature and hydration could be adapted to minimise the risk of fruit injury.

CHAPTER 5. LYCHEE POSTHARVEST DESICCATION

Skin browning associated with desiccation is identified as one of the major causes of postharvest loss in the lychee industry (Snowden 1990). Substantial research has been published on the physiological, anatomical and biochemical processes of skin browning (Jiang 2000; Jiang and Fu 1999a; Pang *et al*. 2001; Underhill 1993; Zhang *et al.* 2001), but quantitative research on water relations has been relatively limited. Chinese journal articles have been published on lychee desorption and absorption characteristics (Chen *et al.* 1995; Yu 1998), but are not readily accessible. A further study of moisture loss by Zhao et al. (1999) provided practical data on water relations, but related to high temperature drying of the fruit. Recent research has explored practical aspects of water relations, including time of harvest and postharvest soaking effects on turgor (Olesen 2001). While avoidance of air currents is often recommended (Bagshaw *et al.* 1991), the relationship beween air speed and moisture loss has not been defined. The estimation of skin resistance properties and quantitative analysis of air current effects will contribute to the development of a water loss model for lychee. The development of a model will assist in the identification of important sites of loss in the postharvest chain.

The effects of maturity and cultivar on susceptibility to moisture loss and browning have not been extensively researched. The levels of enzymes associated with browning can significantly vary between lychee cultivars (Chen *et al.* 2001; Underhill and Critchley 1995). Chen *et al.* (2001) showed a significant difference in browning susceptibility associated with this difference in enzymes in the Chinese cultivars ‘Nuomici’ and ‘Guiwei’. Wu *et al.* (2001) explored the effect of lychee maturity on moisture loss, and detected slight differences. However, the categories used were more extreme than would occur in commercial harvests, and desiccation browning associated with the loss was not assessed. Differences in desiccation browning susceptibility due to maturity or cultivar may assist in the reduction of losses through selected plantings, breeding or adjusting the timing of harvest.

Physiological processes of moisture loss, such as the pathway of moisture loss from the fruit and the importance of epicuticular waxes in protection have not previously been explored. Similarly, the effect of moisture loss on CO_2 and ethylene generation has not been established. Study of these processes will further develop an understanding of the physiology of lychee moisture loss.

5.1 Variation Between Cultivars in Susceptibility to Moisture Loss

Anatomical and physiological differences between cultivars could potentially result in different rates of postharvest moisture loss. Characteristics such as epicuticular wax structure and composition are not visible to the naked eye, but can substantially affect water loss. A study of moisture loss in some of the more commonly grown lychee cultivars was undertaken to detect any consistent differences between cultivars.

5.1.1 Methods and Materials

5.1.1.1 Experiments 1, 2 and 3 - 1999/2000 Season

For each cultivar, 8 fruit in good condition were harvested from each of 5 trees. Fruit were harvested into damp paper towel, and were transported immediately to a mobile laboratory. TSS and titratable acidity were measured for each cultivar on two blended samples of 5 fruit (total measurement the average of 10 fruit, using 2 from each tree). The remaining 30 fruit were stored at RT, at either 70% RH (Experiment 1 (E1)) or

57% RH (Experiments 2 (E2) and 3 (E3)). In a longitudinal study, colour, weight and score (where 1 = 0 to 10% of the SA brown and 10 = 90 to 100% brown) were recorded regularly until a severe rot developed, or until all fruit in the cultivar were brown.

- E1 was carried out on ‘KMP’ and ‘Wai Chee’ harvested from Childers on 18/1/00.
- E2 used ‘KMP’, ‘Wai Chee’, ‘Salathiel’ and ‘Bengal’ from Childers, harvested on 28/1/00.
- E3 studied ‘KMP’, ‘Wai Chee’ and ‘Salathiel’ harvested from Nambour on 15/2/00.

5.1.1.2 Experiment 4 - 2001/2002 Season

Fruit of 4 different cultivars (‘Bengal’, ‘KMP’, ‘Wai Chee’ and ‘Salathiel’) grown at Sippy Downs were harvested over a 3 week period. Fruit were picked directly into water, and were soaked for at least 30 minutes. Fruit were dried, and packed into damp paper towel for transport to the laboratory. Gross anatomy and Brix: acid measurements were made on 10 randomly selected fruit from each cultivar, as previously described (Section 4.10.1). For each cultivar, 30 fruit were stored at 33% RH (using saturated MgCl) and 20^{o}C. In a longitudinal study, colour on both cheeks and fruit weight were measured daily until the majority of fruit were brown. At the commencement of the storage period, water potential was measured in 10 fruit.

The colour and browning score data are presented separately in Section 5.2.

5.1.1.3 Other Experiments

Some results are calculated using data from other unrelated experiments. Where treatments were applied that may have affected moisture loss, such as mechanical injury, only control data were used. Methods and materials for these experiments have been previously described, as referenced in the text.

5.1.1.4 Calculation of Specific Moisture Loss (SML)

The specific moisture loss (SML) is an index of fruit resistance to moisture loss that compensates for different storage conditions. It can be used as a basis for comparison between different experiments, despite variation in RH and temperature.

SML = ML / (M x dP x t) ***…Equation 5.1.1***

Where, SML = specific moisture loss ($kgkg^{-1}Pa^{-1}s^{-1}$), ML = moisture loss (kg), M = fruit mass (kg), dP = vapour pressure deficit (Pa) and t = time (seconds) (equation adapted from van Beek and Lamers (1979), as described by Scheer (1994)).

Vapour pressure deficit can be calculated from RH and temperature:

$dP = e^{o} – (e^{o} \times RH)$ ***…Equation 5.1.2a***

$e^{o} = 610.8 \times 2.7183\text{^} [17.27 \times T / (T + 237.3)]$ ***…Equation 5.1.2b***

Where, dP = vapour pressure deficit (Pa), e^{o} = saturation vapour pressure (Pa), RH = relative humidity and T = temperature (^{o}C) (equations from Tetens (1930), as described by Schaefer (1989)).

5.1.2 Results and Discussion

5.1.2.1 Specific Moisture Loss

SML was fairly consistent across experiments, with the exception of Experiment 4 (E4) (Fig. 5.1.1). The average SML observed in E4 fruit was at least 60% greater than other experiments. Comparing the overall averages of each experiment, only the first two experiments, carried out at the same location, did not significantly differ (ANOVA on day 2 data, $p<0.001$, comparison by Tukey test). The significant difference between the Nambour experiment (E3) and experiments at Childers (E1 and 2) show that SML can be affected by location as these experiments were carried out in the same season. The very high SML in E4 was possibly a site effect, as similar high SML was also shown in other experiments on fruit from this orchard (data not shown). However, a seasonal effect could also have contributed, as this was the only orchard tested in the ’01 / ’02 season. Fruit from this orchard were not tested in other seasons, so it cannot be determined whether this higher SML is due to seasonal variation or the individual orchard.

The SML is largely determined by cuticle properties, in particular the arrangement of epicuticular waxes (Chambers and Possingham 1963; Denna 1970). The characteristics of these waxes could be affected by conditions during development, including temperature, nutrient supply and moisture availability. The SML could therefore be influenced by climate, soil qualities, fertiliser programs and irrigation management. Greater weathering or damage to the protective waxes could also affect the SML. Damage to waxes could result from wind rub. This is unlikely to have caused the observed differences, as the orchard site for E4 was well protected from wind, and signs of damage were not observed on the fruit. Extreme temperatures and the use of certain sprays may also potentially damage wax structure. Microcracking is another possible factor, and is strongly influenced by preharvest conditions. Three days of moderate water stress at the late stage of fruit development was shown to increase the extent of microcracking in lychee by 65% (Kumcha 1998). Microcracks in the apple cuticle were estimated to be 12 times more permeable than the intact cuticle (Maguire *et al.* 1999b), so substantially higher moisture loss would be possible. The cause of increased SML in season 3 fruit cannot be identified, and may have resulted from numerous management or climatic factors.

Within each orchard, there were some significant differences between cultivars, but these were generally slight and were not consistent across a variety of orchards (Fig. 5.1.1). ‘Salathiel’ showed significantly higher SML than other cultivars in Childers (ANOVA $p<0.001$, comparison by Tukey test) , but had the lowest SML in Sippy Downs. The lack of consistent difference shows that resistance to moisture loss does not substantially vary due to cultivar genome for the range of varieties tested. Significant variation within each farm could result from the suitability of each cultivar to the orchard conditions. A tree well suited to the orchard conditions may produce fruit with slightly better SML.

5.1.2.1.1 The Effect of Storage RH on SML

Extreme levels of storage RH significantly affected the calculation of SML (Fig. 5.1.2, data from Section 5.5). Using data from previous experiments (Section 4.1 and Section 5.5), SML could be estimated over a range of humidity levels from 11 to 97%. In all experiments RH was controlled using saturated salts. A relative humidity of 97% consistently resulted in very high SML. This is due to the fact that a flat value of 97% RH was used in calculation of SML, while in reality the humidity would slowly increase towards the equilibrium point of 97%. The equilibration period has a strong effect due to the large discrepancy between the ambient and controlled RH. The SML was also slightly high at 85% RH, but was significantly different only to 65% RH (t-test, p=0.017). Low humidity has the reverse effect, resulting in a lower SML, as the actual humidity is higher than predicted. No consistent trend in values occurred in the range of 39-75% RH. The variation in values shown in this range could result from different speed of equilibration in the salts used. The SML values used in analysis are from RH in the range 33-75%, and should therefore give an accurate measure of the SML.

5.1.2.1.2 Effect of Fruit Size on SML

Overall, a trend toward higher SML in smaller fruit was shown (for all SML data, Spearman correlation co-efficient = -0.131, p<0.0005). Greater SA: volume ratio in smaller fruit can result in more rapid moisture loss on a percentage of weight basis, as observed in capsicums (Lownds *et al.* 1993). Analysis of individual lychee cultivars within experiments rarely showed a significant correlation, with little variation in size occurring. The tendency for higher SML in smaller fruit may have contributed to the faster loss of moisture in ‘Salathiel’ at Childers. However, similar small fruit size is shown in other cultivars without any accompanying increase in SML.

5.1.2.1.3 The Effect of Harvest Date on SML

On a few occasions, the SML could be estimated over a range of harvest dates for a single cultivar at one location. This overlap of data allowed the effect of harvest date on postharvest moisture loss to be examined. Fruit of cultivar ‘KMP’ in Childers were harvested on 4 different dates in the ‘99/’00 season, 18th, 28th and 31st January and 18th February. At two of these dates (18th and 31st January), SML was calculated in two distinct experiments, and is hence presented as separate data points. A consistent trend over time was not observed (Fig. 5.1.3), although SML did appear to decline over the month of January. Significant differences in SML were shown between early harvest dates (18th January) and mid-season dates (28th and 31st January) (t-tests $p<0.025$). The data would suggest a harvest date of late January to early February for optimal resistance in this instance. Typical harvest dates for ‘KMP’ in Childers would be early-mid January (Greer 1990). However, the ‘99/’00 season was reported to be almost one month late, with cool weather delaying fruit ripening (Dixon, 2000). The commercial harvest would therefore have occurred in early-mid February, close to the period of minimal SML.

Data collected for ‘KMP’ in Sippy Downs shows a similar, but much more extreme trend, with a rapid decline in SML from January to mid-February (Fig. 5.1.4), followed by a sharp increase in early March (ANOVA $p<0.001$, comparison by Tukey test). The commercial harvest of ‘KMP’ typically occurs in mid-late January in this area (Greer 1990). Only three dates were collected for ‘Wai Chee’ at Sippy Downs, but a similar sharp rise in SML with over-maturity is shown (ANOVA p<0.001, comparison by Tukey test) (Fig. 5.1.5). It would appear that significant benefit could be gained by timing harvest for the lowest postharvest moisture loss, with rates capable of being halved.

Variables such as colour and flavour development and the prices obtained at market generally determine the selection of a harvest date. These factors can vary substantially from week to week, are easy to estimate, and are commercially important. In contrast, SML would be difficult to gauge, and would be unlikely to translate into any tangible economic benefit to the grower. The variation in SML with harvest date is therefore unlikely to be of practical importance. However, considering the importance of desiccation browning in postharvest losses, the substantial changes in SML shown warrant further research.

5.1.2.1.4 Changes in SML During Postharvest Storage

Changes in SML over the course of postharvest storage were observed in some experiments, but these were not consistent. For example, in E1 the SML showed a continuous upward trend (data not shown). As the experiment progressed, fruit lost moisture more rapidly. In contrast, the opposite trend occurred in E4, where the SML declined over storage (data not shown). These differences probably occurred due to storage RH, which was 70% in E1 and 33% in E4. In fruit stored at low RH, any readily removed water (in particular, from the pericarp) would be quickly lost from the fruit. Water stress would have progressively reduced the rate of moisture loss, due to physiological changes, such as stomatal closure, and the fact that more energy is required to pull water from desiccated tissue. The endocarp barrier to moisture movement between pericarp and aril strengthens the tendency for rates of moisture loss to decline with increasing dehydration. The higher humidity level in E1 would have resulted in much slower moisture loss, where it may have taken several days to pull the readily available water from the pericarp. The SML may have gradually increased due to postharvest microcracking. Trends in SML over time were similar in various cultivars within each experiment, but varied with storage RH.

5.1.3 Conclusions

Cultivar differences in postharvest moisture loss were shown to exist, but these were minimal, and were specific to location. Variation in specific moisture loss (SML) due to unknown location or seasonal variables overwhelmed the minor cultivar differences. A much higher rate of loss within one experiment may have resulted from numerous climatic or management factors. Harvest date was shown to have a significant effect on SML. Rates of loss tended to decline in mid-season, but later increased with over-maturity. Additional research is required to confirm and explore this relationship. Research into the pre-harvest factors affecting postharvest moisture loss in lychee would be highly beneficial. Improved pre-harvest management may have the potential to decrease rates of moisture loss, giving significant increases in shelf life.

5.2 Variation Between Cultivars in Susceptibility to Desiccation Browning

A study of moisture loss and accompanying browning was undertaken in a variety of lychee cultivars to determine whether substantial differences occur between varieties. The desiccation browning response could vary between cultivars due to structural or biochemical differences in the pericarp. Substantial cultivar variation could necessitate more careful management of highly susceptible varieties. Tolerant cultivars may be preferentially planted, or used for future breeding programs or genetic manipulation.

5.2.1 Methods and Materials

Methods are as described in Section 5.1.1. In a longitudinal study, regular measurements of weight, colour and browning score (1 = 0-10% to 10 = 90-100% brown) were carried out on a range of cultivars in 4 experiments. Browning scores were not recorded in experiment 4 (E4), but estimates were derived from changes in colour (using previous browning score and colour relationships from the same cultivars).

5.2.2 Results and Discussion

5.2.2.1 Relationship Between Moisture Loss and Browning Scores

The relationship between moisture loss and browning followed a similar pattern in most cultivars (Fig. 5.2.1). Two rough groupings were apparent, with around half of the samples showing early browning (‘KMP’ from E1, E2 and E4 and ‘Salathiel’ and ‘Wai Chee’ from E4). Other samples showed browning at slightly higher levels of moisture loss (‘Wai Chee’ from E1, E2 and E3, ‘Bengal’ from E2 and E4, and ‘Salathiel’ and ‘KMP’ from E3). There was no consistent division between cultivars or seasons in these groupings, but all fruit harvested at Nambour were in the late browning group.

For the more tolerant samples, the SA of browning generally remained low until around 7% moisture loss, after which a gradual steady increase in browning occurred. All samples in this group reached 10-20% SA browning within the very small range of 9.5 to 11% moisture loss. In the samples that browned at lower moisture loss, 10-20% browning occurred from around 5 to 8% weight loss. The tolerant samples continued to show a very similar relationship until 30-40% browning, which generally occurred at 12-13% moisture loss. At scores greater than 4, these samples showed a greater degree of variation, with the browning of ‘Bengal’ fruit from E2 particularly slower than other samples. The early browning samples showed 30-40% browning at 8-10% moisture loss. Overall, an average of 10% moisture loss was required to cause any substantial browning (>20% SA), with a further 4.5% loss resulting in severe browning (>70%).

The fruit that browned early may have suffered from slight moisture stress at harvest, resulting in moisture levels reaching a critical threshold earlier. Within both E1 and E2 all cultivars were harvested on the same morning, so climatic conditions prior to harvest were not variable. However, different cultivars were located in different sections of the orchard, with soil properties or irrigation management possibly varying. E3 fruit were consistently tolerant to moisture stress. These fruit were harvested in cool, wet conditions (19^{o}C with drizzle), so fruit moisture content at harvest was likely to have been high. While this partially explains the tolerance shown by E3 fruit, other unidentified factors may also have contributed. In these fruit browning commences at a higher levels of moisture loss (around 11.8%, compared to 9.2% in other sites (Fig. 5.2.2)), which may be explained by higher moisture content. However, the progression of browning also occurred at a slower pace, taking an average 6.1% loss to change from score 3 to score 8, while other sites averaged 3.9% (t-test, p=0.030). This cannot be explained by moisture content, but is more likely to result from biochemical and structural differences. The results therefore provide some evidence that location or management can influence the browning response. The mechanisms of this effect are not clear.

Some variation due to cultivar does appear to exist, with consistent ranking of cultivars within each experiment (Fig. 5.2.3). In all cases, ‘KMP’ and ‘Salathiel’ were the earliest to show browning, followed by ‘Wai Chee’, then ‘Bengal’. This relationship suggests that ‘KMP’ and ‘Salathiel’ are slightly more susceptible to browning, while ‘Bengal’ is more resistant. The differences between cultivars do not appear to relate to fruit water potential. In E4, water potential was lowest in ‘KMP’ and highest in ‘Salathiel’ (Fig. 5.2.4), but these cultivars showed a very similar browning response (Fig. 5.2.3). This would suggest that the differences between cultivars result from structural or biochemical properties of the pericarp. It is possible that skin thickness contributed to browning susceptibility. The base skin thickness in the four cultivars followed the same trend as browning susceptibility (Fig. 5.2.4), with thick-skinned fruit more resistant. This could be explained by a theoretical browning “trigger value” of water content in the skin. Thin-skinned fruit have less skin volume, and therefore would need to lose less moisture (on a whole fruit basis) before a threshold value was reached. This effect would be exaggerated by the fact that the majority of early moisture loss in lychee occurs from the pericarp (Zhao *et al.* 1999).

5.2.2.2 Relationship Between Moisture Loss and Colour Changes.

Differences in freshly harvested skin colour between cultivars were significant (Fig. 5.2.5). Analysis of combined site data for each cultivar showed that ‘KMP’ had the lightest and most yellow colour, with values substantially different to other cultivars. ‘Bengal’ was slightly lighter in colour than ‘Wai Chee’ and ‘Salathiel’. ‘Salathiel’ showed the most red-purple hue and most saturated colour. ‘Wai Chee’ fruit had the dullest colour, and were fairly dark and red-purple in hue. Colour significantly varied within each cultivar between the different experiments (Figures 5.2.6, 5.2.7 and 5.2.8), but differences were not consistent.

As would be expected, colour changes measured by the colour meter showed very similar trends to browning scores, but allowed the separate observation of darkening, loss of chroma and hue changes. Colour darkened gradually even at very low levels of moisture stress, with the rate of darkening tending to accelerate after around 7% moisture loss (Fig. 5.2.9). ‘KMP’ and ‘Salathiel’ showed similar rates of darkening, normally more rapid than ‘Wai Chee’ or ‘Bengal’. ‘Wai Chee’ fruit showed the slowest darkening of colour, and the least total darkening in most cases. The irregular shape of the ‘Bengal’ curve, as observed in browning scores (Fig. 5.2.3) is also seen in L-values. Browning in ‘Bengal’ fruit seems to occur slightly erratically compared to the regular curve shapes of other cultivars. ‘KMP’ and ‘Wai Chee’ showed lightening of colour at extreme desiccation (10-12% loss for ‘KMP’, and 12-15% for ‘Wai Chee’) at Childers and Sippy Downs, but this did not occur at Nambour. The cause of this lightening of colour is not clear.

Gradual loss of chroma was observed at very low levels of moisture loss, with the rate of loss increasing as dehydration continued, but slowing after extreme desiccation

(Fig. 5.2.10). ‘Salathiel’ fruit showed the most intense colour prior to dehydration (Fig. 5.2.5), but also tended to show the fastest rate of chroma loss, and the greatest total loss of chroma (Fig. 5.2.10). Despite having fairly intense colour at harvest (Fig. 5.2.5), ‘Bengal’ fruit tended to show slow, or slightly delayed loss of colour saturation (Fig. 5.2.10). This provides some further evidence of the greater resistance of ‘Bengal’ fruit to desiccation browning. ‘KMP’ fruit from E2 and E4 showed much less total loss of chroma than other cultivars due to lower initial chroma values (Fig. 5.2.7). ‘KMP’ and ‘Wai Chee’ fruit tended to show similar rates of chroma loss.

‘Bengal’ fruit showed distinctly different changes in hue compared with other cultivars (Fig. 5.2.11). A substantial colour change towards red-purple occurred until moisture loss reached around 9%. A small change in hue towards red-purple also occurred in ‘Salathiel’ fruit from E2. The mechanism of this change is not clear, but is likely to relate to biochemical changes in the skin. After the initial dip in hue, ‘Bengal’ fruit showed some yellowing of colour, but this reached a maximum at an increase in hue angle of around 5-6. In contrast, many other samples reached hue angle changes of over ten. ‘KMP’ tended to undergo the most rapid yellowing. ‘Wai Chee’ and ‘Salathiel’ showed similar rates of yellowing, slightly slower than ‘KMP’. The degree of yellowing occurring after extreme dehydration varied substantially between cultivars and experiments with no clear reason. In particular, ‘KMP’ and ‘Wai Chee’ from E2 and E4 showed large changes in hue, while ‘KMP’ in E1 and E3 and ‘Salathiel’ in E4 showed very low values. These differences did not relate to initial hue values.

5.2.3 Conclusions

A roughly sigmoid relationship was generally shown between moisture loss and browning. Some extent of variation between samples may have resulted from different moisture content at harvest, but this did not appear to be an overriding effect. The comparison of cultivars within each experiment revealed a consistent order of browning susceptibility. ‘KMP’ and ‘Salathiel’ were most prone to browning, ‘Wai Chee’ showed moderate susceptibility, and ‘Bengal’ fruit were most resistant. It was theorised that this may have resulted from differences in skin thickness, which followed the same trend as browning susceptibility. Small losses of moisture would have a greater impact on the pericarp water content in thin-skinned fruit, due to its reduced comparative volume. There was some evidence of a location effect, with the progression of browning significantly slower in Nambour fruit. However, there were generally no consistent site effects. Large variations between samples appeared to result from factors other than cultivar, season and orchard. Additional research would be beneficial, in order to confirm the cultivar effect suggested by this data, to test the skin thickness hypothesis, and to compare additional varieties. The research shows a large degree of variation due to unknown factors, warranting further exploration. The management of desiccation browning may be improved by greater understanding of the underlying factors influencing the process.

5.3 The Effect of Maturity on Moisture Loss and Desiccation Browning

Lychee maturity has been shown to affect postharvest moisture loss, but differences were minimal across a wide range of maturity levels (Wu *et al.* 2001). Rates of loss were slightly lower in half-red fruit, compared to fully mature and very immature fruit. For example, at 72 hours, the half-red fruit had lost 8.1%, while fully mature fruit lost 8.8%. The rate of browning in these fruit was not noted, although it would be expected that slightly faster browning would accompany more rapid moisture loss. A study of the effect of maturity on postharvest browning was reported (Sittigul *et al.* 1994), but related to storage at 5^{o}C and 90-95% RH, conditions in which desiccation browning would be unlikely to occur. The current experiment was carried out to develop a greater understanding of the effect of fruit maturity on lychee moisture loss and desiccation browning.

5.3.1 Methods and Materials

A bulk sample of 50 ‘KMP’ fruit of varying maturity was harvested from Nambour. Fruit were separated into 5 maturity grades based on the amount of green colour in the skin (Table 5.3.1) resulting in slightly uneven replication (9-11). TSS and titratable acidity were measured using 10 fruit from each grade, in 2 bulk samples.

Table 5.3.1 Brix : acid ratios and percentage green SA in the lychee maturity grades.

Maturity grade	Green SA (%)	Brix : acid
M1	>10	39:1
M2	5-10	43:1
M3	2-5	48:1
M4	1-2	54:1
M5	<1	63:1

For each fruit, the percentage of green colour on each side of the fruit was estimated at the start of the experiment. Fruit were stored at RT and 70% RH. In a longitudinal study, fruit weight, colour at each cheek centre and browning scores on each side (1 = 0-10% brown to 10 = 90-100%) were recorded regularly until fruit were completely brown.

5.3.2 Results and Discussion

Maturity levels varying from slightly immature to fully red showed no significant difference in rates of moisture loss (Fig. 5.3.1). There was no consistent trend across the range tested, and no significant correlation between moisture loss and percentage SA green. The conflict with previous research (Wu *et al.* 2001) may have occurred due to cultivar differences or the narrow range of maturity levels tested in this experiment.

Analysis of browning scores showed that greener fruit browned faster than red fruit (Fig. 5.3.2). Fruit more than 10% green showed significantly more rapid browning than other grades. Maturity grades M2 and M3 (2-10% green) browned significantly more rapidly than M4 and M5 (0-2% green) (Analyses by Mann Whitney rank sum tests and t-tests at $p<0.05$). However, the analysis of colour readings showed that this was merely an error of perception. Greener fruit consistently showed a more yellow hue than red fruit, and the early fading of green colour was perceived as browning.

Obviously, the colour of the maturity groups varied substantially prior to storage, as colour was used to grade the fruit (Fig. 5.3.3). Greater green SA was accompanied by significantly lighter, less saturated colour, further towards yellow in hue (t-tests and Mann-Whitney rank sum tests $p<0.05$). Mature fruit were darker, brighter and moved towards a red-purple hue. In addition to improved flavour, the marketing of fully mature fruit has the advantage of better visual appeal, with fruit appearing brighter and more intensely red in colour.

Changes in colour due to moisture loss also varied with maturity. More immature fruit tended to show greater total darkening, particularly at extreme levels of moisture loss (Fig. 5.3.4). The lighter colour of these fruit prior to desiccation probably results in greater expression of tissue browning. Loss of chroma tended to occur at a similar rate regardless of maturity (Fig. 5.3.5). Immature fruit (>10% green) showed minimal yellowing of colour (Fig. 5.3.6). Fruit of maturity grade M2 showed a late increase in hue, less substantial than that shown in more mature fruit. Fruit of maturity grades M3, M4 and M5 showed similar strong yellowing in response to moisture loss, commencing at around 7% weight loss. Greener fruit continue to show a more yellow hue than red fruit throughout the experiment, but the difference lessened later in the experiment, due to the stronger yellowing of more mature fruit. In all colour indices, the changes are fairly similar across all maturity levels until 120 hours storage (around 7% moisture loss).

5.3.3 Conclusions

Fruit ranging from slightly immature to fully mature showed no substantial variation in the rate of postharvest moisture loss. Browning scores increased more rapidly in greener fruit, probably due to fading of green being perceived as browning. Colour readings revealed similar patterns of change until around 7% moisture loss. After this point, greener fruit tended to show greater darkening, but less yellowing of colour than more mature fruit. These changes appeared to relate to initial differences in colour, with greener fruit showing lighter and more yellow colour at the start of the experiment.

The small differences between the maturity levels tested are unlikely to be of any commercial significance, occurring principally at high levels of moisture loss.

5.4 Effect of Air Currents on Lychee Moisture Loss

The effect of air movement on moisture loss varies widely between commodities. For some fruit, such as apples, air currents have very little effect on water loss, while in others slight air movement has a major impact (Van Beek and Lamers 1979). For example, water loss from carrots increased by a factor of nine when slight air currents (0.05-0.15 m/sec) were applied (Van Beek and Lamers 1979). This experiment aimed to clarify the effects of various air current speeds on lychee moisture loss.

5.4.1 Methods and Materials

The experiment was repeated three times using both ‘KMP’ (Runs 1 and 3) and ‘Wai Chee’ (Run 2) fruit harvested from Sippy Downs. Fruit were harvested into water and soaked for at least 45 minutes. They were then tightly packed into damp paper towel and transported to the laboratory. Fruit were weighed prior to treatment, and were then exposed to various levels of wind speed during storage. Fruit weights were re-measured after various periods of storage. In a randomised control trial, 4 treatments were each applied to 20 fruit:

- Control, exposed to no air currents;
- Fan setting 1, with a circle of cardboard (16.5 cm diameter) taped over the fan face, giving an average wind speed of 0.59 msec^{-1};
- Fan setting 1, average wind speed of 1.17 msec^{-1};
- Fan setting 2, average wind speed of 1.55 msec^{-1}.

On the Beaufort scale, the lower wind speed treatments were scale 1 (light air), and the most severe treatment was at the lowest end of scale 2 (light breeze).

Each treatment group was stored in an identical plastic crate, with dishes of saturated NaCl used to regulate humidity to around 75%. Fruit were stored at room temperature, which ranged from 22-32^{o}C during the course of the experiments. Fruit were arranged in a staggered grid, with approximately a 1 cm buffer of space around each fruit (Fig. 5.4.1). Small upturned lids (22 mm in diameter and 12 mm high) were used to raise the fruit, and keep them in place. “Cool Breeze” 15 cm diameter fans (CBD15) with 2 speed settings were used in the experiment. The fans were placed approximately 2 cm from the first row of fruit and were angled downwards, towards the fruit, during treatment. Air velocity was estimated using an anemometer (C.F. Cassela & Co. Ltd.) within the sealed crate, held at the same height as the fruit.

Figure 5.4.1 Arrangement of lychee fruit within the crate, with dishes of saturated salts and fan.

5.4.2 Results and Discussion

In the first run, using ‘KMP’ fruit, fruit weight was re-measured after 27.5 hours storage. At this point, weight loss in fruit exposed to air currents was extreme, averaging 13.4% in the low air speeds and 15.1% under a wind speed of 1.55 msec^{-1} (Fig. 5.4.2). In contrast, control fruit had lost only 2.4% of weight. Differences between the wind speeds were obscured, as rates of water loss slow down substantially at these high levels of desiccation. Analysis by ANOVA showed that the four treatment groups were significantly different from each other, except for the two low speed settings (p-value of analysis <0.001, comparison by Tukey test).

The second run, using fruit of cultivar ‘Wai Chee’, gave similar results, with slightly greater differences between treatments due to earlier measurement (Fig. 5.4.2).

All treatments were significantly different to each other (Mann Whitney rank sum tests, $p<0.001$). The relationship between air speed and water loss was close to linear ($R^2 = 0.95$). The varying rates of moisture loss were strongly reflected in colour (Fig. 5.4.3).

Figure 5.4.3 Effect of air currents on lychee appearance after 18 hours at RT (20-30^{o}C). A – control (no current), B – 0.59 msec^{-1}, C – 1.17 msec^{-1} and D – 1.55 msec^{-1} (Run 2).

A stronger linear relationship ($R^2 = 0.99$) was found by measuring weight loss after 6 hours in the third run (Fig. 5.4.4). The relationship was defined by the equation:

$W = 1.074 + 3.731 \times S$ ***…Equation 5.4.1***

Where, W = weight loss (%) and S = wind speed (msec^{-1}).

Analysis showed that all treatments were significantly different (ANOVA using ln transformed data, $p<0.001$, comparison by Tukey test). The highest wind speed tested (1.55 msec^{-1}) increased moisture loss by a factor of seven. Exposure to this wind speed would therefore result in desiccation browning emerging seven times faster. A wind speed of around 0.3 msec^{-1} would be predicted to double the rate of weight loss.

For most commodities, the effect of increasing wind speed declines above a certain point. For example, in well-suberized potatoes, wind speeds greater than 0.07 msec^{-1} do not result in any further increase in the rate of moisture loss (Rastovski 1987 (In: Scheer 1994)). For lychee, wind speed continued to affect the rate of water loss until at least 1.55 msec^{-1}. Further experimentation would be required to determine the point at which this relationship levels off.

The strong effect of air currents on moisture loss reveals the importance of boundary layers in lychee resistance to water loss. The spiky shape of the lychee skin would encourage the formation of a humidity gradient near the skin surface, with air movement restricted in the valleys formed by protuberance bases. Even at wind speeds of greater than 1 msec^{-1}, a boundary layer appears to be capable of forming.

Measurements of moisture loss over time clearly show a decline in rate of loss as desiccation increases (Fig. 5.4.5). This is a common reaction, occurring due to tissue responses to moisture stress and a lag effect in moisture being drawn to the surface of the tissue (Van den Berg 1987). In lychee, the lag effect would be exaggerated by the composite structure of the fruit, as the movement of moisture from aril to pericarp is restricted (Zhao *et al.* 1999).

The relationship between previous weight loss and subsequent rate of loss was fairly consistent in the treatments exposed to air currents. The rate of loss declined by around 20% after moderate (5%) weight loss. When fruit were severely desiccated (10% weight loss), the rate of weight loss was 40-50% lower than in turgid fruit. However, the control fruit, not exposed to any air currents, showed a substantial decline in the rate of loss despite still being well-hydrated. In control fruit, the benefits of a small reduction in water loss (for example, due to stomatal closure) would be proportionally greater, as the total rates of loss are lower. This may explain the faster decline in rates of moisture loss during storage.

5.4.3 Conclusions

Air currents had a very strong effect on rates of water loss from the fruit, capable of increasing loss by at least seven times. The relationship between wind speed and moisture loss was shown to be linear in the range tested. Further experimentation would be required to determine the point at which this relationship levels off. Rates of moisture loss were shown to decline with increasing desiccation. This response occurred at low levels of moisture loss in fruit not exposed to air currents, giving further benefit to the avoidance of air movement.

The massive increase in water loss resulting from air currents has major implications in postharvest management. Minimising exposure of the fruit to air currents would be a key step towards preventing desiccation browning. The benefits of forced-air cooling may be compromised by accelerated moisture loss. At the retail level, current postharvest practice typically involves displaying fruit open to the air. Fruit are exposed to both air movement and low humidity, greatly accelerating the rate of moisture loss. The results of this experiment add further impetus to the use of punnets, or protective display cases.

5.5 Effect of Storage RH on the Relationship Between Moisture Loss and Desiccation Browning

In rambutan, storage RH affected the relationship between weight loss and skin browning. At 60 to 80% RH, browning of the skin occurred at similar levels of weight loss, regardless of humidity. Higher RH of 90 and 95% resulted in browning later in postharvest storage, but at lower levels of weight loss (Landrigan *et al.* 1996b). It was not specified whether the browning was related to disease, senescence or desiccation. In contrast, a previous study on lychee showed no substantial RH effect on the relationship between lychee browning and pericarp moisture loss (rearranged data from Jiang and Fu 1999). However, the study focussed only on the first 3 days of storage. The current research was carried out to further define the relationship between moisture loss and colour change in lychee, and to determine whether storage RH affects this relationship.

5.5.1 Methods and Materials

Uniform lychees of cultivar ‘KMP’ were harvested and randomly placed into 6 groups of 10 fruit. Each group was placed into a punnet within a sealed container at RT (20-30^{o}C), with saturated salt solutions used to manipulate the humidity of each container to various levels (33, 53, 65, 75, 84 and 97% RH). In a longitudinal study, fruit were weighed daily, colour readings were taken at the cheek centre and a browning score was noted (1 to 10, where 1 = 0-10% SA brown and 10 = 90-100%). Fruit were assessed daily until a severe rot formed, or until all fruit within the sample were brown.

5.5.2 Results and Discussion

As would be expected, fruit browned rapidly at low RH (Fig. 5.5.1). Fruit stored at 33% RH were fully brown within 93 hours. Higher storage RH resulted in slower development of browning.

The relationship between moisture loss and browning score was fairly consistent across a wide range of RH levels, with no substantial differences from 33 to 84% (Fig. 5.5.2). The 97% RH treatment resulted in browning at substantially lower levels of moisture loss. This was primarily due to rot development in the later stages of storage. However browning scores were higher in 97% RH fruit even very early in storage. At 70.5 hours the 97% RH fruit show more browning than low RH treatments, but rots were not observed until 194 hours. The 84% and 75% RH fruit also show a slight rise in score very early in storage, but after 5% moisture loss these fruit followed the same pattern as low RH fruit. Analysis of estimated scores at 3% moisture loss showed that the highest RH treatments (84 and 97%) were significantly more brown than lower RH treatments (39 to 65%) (Analysis by t-tests and Mann-Whitney rank sum tests, $p<0.05$) (Fig. 5.5.3). The reason for greater browning in high RH fruit early in storage is not clear, but may relate to changes in fruit colour due to dampness. For example, vibration damage tends to appear more brown in colour when fruit are damp.

The relationship between moisture loss and browning score followed a roughly sigmoid shape, with a more rapid rate of browning occurring after around 6% moisture loss. At around 12.5% weight loss, most fruit were brown, resulting in a plateau in the curve. Around 8% weight loss was required to cause browning to greater than a third of the fruit. In comparison to other fruit and vegetables, lychee would be considered fairly tolerant of water loss. Typical levels at which produce becomes unsaleable are around 5-8% (Ben-Yehoshua 1987).

In contrast to the fairly consistent changes in browning score, changes in colour readings with moisture loss showed substantial differences between RH levels. Fruit at higher RH tended to show extreme darkening of colour with moisture loss (Fig. 5.5.4). Fruit at 65, 75 and 84% RH showed a similar pattern of gradual decline, followed by more rapid darkening after 8-9% moisture loss. This was most rapid in 84% RH fruit, and was not due to rot formation. In contrast, the 33% RH treatment resulted in minimal darkening, followed by lightening of colour after 10-13% moisture loss. Fruit at 53% RH also showed a slight lightening of colour after around 13% loss. Trends in chroma were similar, although less extreme, with higher storage RH resulting in greater loss of colour saturation with moisture loss (Fig. 5.5.5). Changes in hue were greatest in the 33% RH treatment, but did not follow a consistent trend, with 65% RH fruit showing the least yellowing (Fig. 5.5.6). The mechanisms behind these different relationships between colour change and moisture loss are not clear. It is possible that processes such as wound healing or microcracking may be affected by RH. The differences in colour change occur primarily after 10% moisture loss, when browning is already substantial, and are therefore of little practical significance.

5.5.3 Conclusions

Browning scores showed a consistent, roughly sigmoid relationship to moisture loss at RH ranging from 33-84%. At 97% RH, browning due to rot development late in storage resulted in a substantially different relationship. A similar darkening response at high RH was shown in rambutan (Landrigan *et al.* 1996a). In lychee, high RH treatments tended to show greater browning at low levels of moisture loss, unrelated to rot development. This may have been due to the effect of dampness on the perception of fruit colour. Changes in L-value, chroma and hue with moisture loss were generally fairly similar in the 33-84% RH range until 10% moisture loss. At extreme levels of moisture loss, the relationships between colour change and weight loss varied substantially with RH. High RH fruit tended to show greater darkening and loss of chroma, but less yellowing with increasing moisture loss over 10%. The reason for this response is not clear, but is of little practical significance given that the fruit are generally unmarketable at 10% loss. Overall, storage RH ranging from 33 to 84% appears to have little effect on the relationship between moisture loss and desiccation browning.

5.6 Pathways of Moisture Loss

Localised cuticle degradation on the protuberance tip, possibly caused by mechanical injury, has been observed by microscopic study of lychee pericarp (Underhill and Simons 1993). The protuberance apex is also the first area in which desiccation browning is observed (Underhill and Critchley 1995). It is possible that disruption of the cuticle and epicuticular waxes on the protuberance tip increases permeability to water vapour, enhancing moisture loss from this region. Stem ends are often an area of high water loss in fresh fruit and vegetables. The stem end of citrus is often sealed with wax to reduce moisture loss. This trial compared the importance of various areas of the lychee surface as pathways to moisture loss.

5.6.1 Methods and Materials

Petroleum jelly was applied to various areas of the fruit to block water loss, and subsequent changes in colour or weight were monitored. Treatments included application of petroleum jelly to the protuberance tips, to the stem scar and to the entire fruit. Petroleum jelly was applied using a paintbrush. The area of the fruit surface covered by the tip treatment was estimated at 5%.

5.6.1.1 Weight Loss in Marketed Fruit

Fruit of cultivar ‘KMP’ were purchased from Brisbane Markets, Rocklea. In a randomised control trial, 4 treatments (control, all protuberance tips, whole fruit and stem scar) were applied to 10 fruit each. Browning score (1 to 10, where 1 = 0-10% and 10 = 90-100% brown) and weight were recorded for each fruit twice daily for 4 days at room temperature and RH (approximately 20-24^{o}C and 30-40% RH). A cardboard box was used overnight to minimise the effect of air currents on the fruit.

5.6.1.2 Weight Loss in Fresh Fruit

Panicles of cultivar ‘KMP’ were harvested using secateurs, and were kept moist with damp paper towels until treatment, less than 2 hours later. The stems were removed by hand. In a randomised control trial, 3 treatments (control, stem scar and all protuberance tips) were applied randomly to 10 fruit each. Fruit were weighed, and stored at RT (20-30^{o}C) and 70% RH. Each fruit was stored in a pre-weighed plastic dish, to avoid the removal of petroleum jelly with handling. The weight of each fruit was measured every second day for 11 days. On the final day the number of rots on each fruit was recorded.

5.6.1.3 Colour Effects

‘Tai So’ lychees were obtained from the Brisbane Markets. The most uniformly ripe and healthy were selected for the experiment. However, fruit in this experiment did differ in maturity, and some were immature. In addition, browning had already commenced at the start of the experiment, and moisture loss may have already been very significant. In a randomised control trial, 4 treatments (control, all protuberance tips, whole fruit and stem scar) were each applied to 20 fruit. Treatments were applied to approximately 1/3 of the fruit surface. The fruit were stored at room temperature and humidity, covered with a cardboard box. Colour readings were recorded over the course of storage.

5.6.1.4 Microscope Observations

Using ‘KMP’ fruit purchased from Brisbane Market, the effect of petroleum jelly tip treatment on colour change was observed under a dissecting microscope at 30X magnification. Within an area of around 50 protuberances on the cheek of a single lychee, half of the tips were randomly coated with petroleum jelly to block moisture loss.

After 24 hours storage at RT and low RH, each protuberance was given a browning score (1-10).

5.6.2 Results and Discussion

In fruit purchased at market, blocking of the protuberance tips substantially reduced loss of moisture from the fruit (Fig. 5.6.1). Covering of protuberance tips with petroleum jelly resulted in total moisture loss consistently 20 to 30% lower than control fruit. Significant differences were detected throughout the storage period (t-tests $p<0.005$). Per unit of surface area, the rate of water vapour movement was estimated to be 4 to 5 times higher from the tips than other areas of the fruit surface. Browning of the skin was similarly substantially reduced by application of petroleum jelly to the tips (Fig. 5.6.2). Localised cuticle damage to the protuberance tips, as previously observed by Underhill and Simons (1993), is the likely cause of increased moisture loss.

In carefully harvested fruit, blocking moisture loss through the tips did not significantly decrease total fruit moisture loss (Fig. 5.6.3). The tip treated fruit lost 5 to 6% less weight than the controls. Rates of loss per unit surface area were estimated to be similar in tips to other areas of the fruit surface. Standard postharvest handling practice probably damages the protuberance tips, accelerating the rate of moisture loss from this area. Tip damage during handling may have resulted in greater proportional moisture loss from this area in the marketed fruit, compared to carefully harvested fruit.

Petroleum jelly application caused immediate changes in colour that were unrelated to moisture loss. An immediate change in hue towards red-purple, loss of chroma and slight lightening of colour were observed (Fig. 5.6.4). The treatments subsequently affected colour in a manner that cannot by explained by moisture loss.

Rapid strong darkening was recorded in tip treated fruit, and petroleum jelly appeared to induce greater loss of chroma during storage. These changes may have been related to pH effects or changes in gaseous exchange in the treated tissue. A reduced rate of yellowing in treated fruit may have resulted from prevention of moisture loss. The tip treatment slowed postharvest yellowing. Covering the whole fruit surface in petroleum jelly resulted in very slow yellowing during storage. The complex effects of petroleum jelly on colour limit the value of this data as an index of moisture loss.

Colour changes observed under the dissecting microscope tended to confirm the importance of protuberance tips as a pathway for moisture loss. Treated protuberances showed significantly less browning (t-test $p<0.001$), with an average score of 3.32, while control protuberances averaged 4.55. Generally, the protuberances with tips covered retained slightly darker red colour in the channels leading up to the tip, an area not treated with jelly. From a distance, this gave them a noticeably redder appearance. This effect allowed treated protuberances to be distinguished from untreated by the naked eye, despite the fact that the jelly was not visible.

Throughout the course of experimental work, it was observed that browning effects were highly localised within the protuberance. Browning in response to air currents or low humidity was localised to exposed protuberances, and did not spread to neighbouring protuberances that were subject to more favourable conditions. The reverse was also observed to hold true; where moisture loss was blocked, the reduction in browning was localised to the treated protuberance. The independent reaction of each protuberance suggests that movement of moisture is restricted between protuberances. By observation under the dissecting microscope, it was noticed that the tips of protuberances with bases covered in petroleum jelly were consistently much redder in colour than controls. Similarly, the bases of tip treated protuberances retained more red colour than controls. These changes suggest that the effect of moisture loss on browning is not localised to a scale smaller than the single protuberance.

In addition to being a pathway for moisture loss, the protuberance tips may play a particularly important role in pathogen resistance. The incidence of rots in freshly harvested fruit was substantially lower in fruit with petroleum jelly covering the protuberance tips than in control fruit (Mann Whitney rank sum test p=0.004). Tip treatment resulted in only one rot in 10 fruit after 9 days storage. In contrast, control fruit showed a total of 16 rots in the 10 fruit, and stem treatment gave a similar total of 18 rots. This substantial reduction in rots suggests that the protuberance tips are a particularly important point of entry for pathogens.

Sealing the stem end did not result in any significant differences in moisture loss (Figures 5.6.1 and 5.6.3), browning score (Fig. 5.6.2) or colour change (Fig. 5.6.4) compared to the control. The stem scar was not found to be a significant point of moisture loss in any analysis. It seems likely that a small amount of moisture is lost through the stem scar, particularly immediately after harvest. The amount of moisture lost through the stem does not appear to be significant, compared to the scale of moisture loss through the pericarp. The natural abscission zone in the lychee stem may minimise moisture loss through this area in harvested fruit.

5.6.3 Conclusions

Initial rates of moisture loss appear to be fairly even over the fruit surface. However, injury inflicted through harvest and the marketing chain may cause an increase in the rate of moisture loss through the protuberance tips. The proportion of loss from the tips ranged from 5 to 6% in carefully harvested fruit, and from 20 to 30% in purchased fruit. Blocking of the tips in carefully harvested fruit significantly reduced rot development, suggesting that even when relatively undamaged, the tips may be an important access point for pathogens. The stem of the harvested lychee did not appear to be a major point of moisture loss.

5.7 The Importance of Epicuticular Waxes in Protection Against Moisture Loss

Epicuticular waxes, embedded within the cuticle and deposited on its surface, are particularly important in the protection of fruit from moisture loss (Chambers and Possingham 1963). Complex wax shapes force water vapour to move in a longer path through the fruit skin, thus providing a barrier to moisture loss (Pantastico 1975). Wax structure can be damaged by mechanical injury, potentially decreasing the ability of the fruit to resist moisture loss. In apples, the removal of waxes resulted in a 30-90 fold increase in water loss (Horrocks 1964). Research was carried out to gain some understanding of the importance of epicuticular waxes in lychee moisture loss.

5.7.1 Methods and Materials

‘Tai So’ fruit were purchased from Brisbane Markets, and were stored wrapped in damp paper towel at 4^{o}C for 2 days, prior to treatment. In a randomised control trial, 4 treatments were each applied to 40 fruit after weighing. Treatments included soaking in acetone for 30 seconds, 1 minute or 5 minutes, and a control, with fruit soaked in water for 30 seconds. Fruit were placed into 20 mL of acetone or water, and were gently agitated during soaking. The fruit were removed, allowed to briefly air dry, and then re-weighed. The washings were placed into pre-weighed glass bottles, and were evaporated to dryness at RT for acetone and 60^{o}C for water controls. The bottles were re-weighed when dry to measure the weight of wax removed. Fruit were stored at RT and 33% RH, and were re-weighed approximately 24 and 48 hours after treatment.

5.7.2 Results and Discussion

Increased soaking time resulted in a greater weight of wax being removed (Fig. 5.7.1), with a strong positive correlation detected (Spearman correlation co-efficient=0.808, p<0.0005). All treatments except the 30 second and 1 minute treatment significantly differed in the amount of wax removed (ANOVA by Ranks, p<0.001, comparison by Dunn's method). The loss of fruit weight from before to after treatment was also positively correlated with both wax weight (co-efficient=0.434, p<0.0005) and soaking time (co-efficient=0.262, p<0.0005).

Loss of moisture during the first 24 hours of storage was increased by the removal of waxes (Fig. 5.7.2a). An increase in soaking time from 30 seconds to one minute appears to cause a sharp increase in moisture loss, despite not causing any substantial increase in waxes removed. This could occur due to a critical breakdown of structure in this period. The 30 second treatment resulted in an average 15% increase in moisture loss, but this was not significantly different to the control (Fig. 5.7.2b). One and five minute treatments resulted in 37% and 45% increases in moisture loss compared to the control, which were significantly higher (ANOVA on ln transformed data, p<0.001, comparison by Tukey test). The increases in moisture loss resulting from removal of epicuticular waxes are very minimal compared to apples, which showed a 30-90 fold increase (Horrocks 1964). Epicuticular waxes may be substantially less important in the regulation of moisture loss in lychee, compared to other fruits. This may be due to other protective features, such as the composite structure of the fruit.

Later in storage, the fruit appeared to recover from the damage inflicted (Fig. 5.7.3a). Rates of moisture loss from 24 to 48 hours were similar in all treatments, with control fruit showing the highest average values (Fig. 5.7.3b). The control fruit showed significantly higher moisture loss in this period than fruit soaked in acetone for 1 minute (t-test on ln transformed data, p=0.013). This could result from the deposition of new waxes in response to injury. In citrus, signs of recovery from mechanical damage are observed in as little as 15 hours (Brown and Barmore 1981). It is therefore possible that the formation of an additional protective barrier in response to injury has resulted in decreased 24 to 48 hour rates of moisture loss in damaged fruit. Alternatively, this effect could result from decreased importance of waxes in regulating moisture loss. Limited availability of moisture in the pericarp would lessen the importance of epicuticular waxes in controlling moisture loss. The fruit used in this experiment were purchased after commercial handling, and were likely to have been mildly water stressed prior to treatment. A further loss of around 2% of fruit weight would have reduced the pericarp water content substantially, thus decreasing the importance of epicuticular waxes in protection. When severe moisture stress has occurred, other factors may become more important in controlling moisture loss. In particular, the ability of moisture to move from aril to pericarp would be likely to increase in importance when the pericarp is dehydrated.

The weight loss data in treated fruit were positively skewed, while control fruit data were normal. The extreme responses causing the positive skew can be observed in Figure 5.7.4. These fruit may have suffered less previous water stress than others, thus having more moisture to lose. The importance of waxes in regulating moisture loss may have been underestimated by this experiment, due to the poor state of hydration at commencement. It is likely that epicuticular waxes would show the greatest control over moisture loss when fruit are in a turgid state.

5.7.3 Conclusions

Epicuticular waxes contribute significantly to the protection of lychee fruit from moisture loss, but are not nearly as important as in apples. When severe, the removal of waxes can result in increases in moisture loss of around 45%. The contribution of waxes to protection would be expected to be greatest in turgid fruit, so this experiment, carried out on partially dehydrated fruit, may have substantially underestimated their role.

5.8 Effect of Moisture Stress on Generation of CO_2 and Ethylene

Moisture stress generally results in increased rates of ripening and senescent degradation in fruit and vegetables (Van den Berg 1987). These processes are typically accompanied by increased rates of CO_2 and ethylene synthesis, and result in reduced shelf life. A preliminary experiment was carried out to determine whether moderate to severe moisture stress strongly affects ethylene and CO_2 evolution in lychee.

5.8.1 Methods and Materials

Methods were as described in Section 2.9.1, with the moisture stress treatment included only in the first run of the experiment. In a randomised control trial, fruit were treated by storage at low humidity of approximately 11%, for 24 or 48 hours, at 20^{o}C. This was estimated to result in 4-5% and 9-10% moisture loss, respectively. After treatment, 10 fruit were immediately placed into small containers, from which gas was sampled 24 hours later. Control fruit were kept constantly at 75% RH and 20^{o}C. Due to problems with equipment, the number of replicate measurements ranged from 4-5 for the low RH treatment, and 4-6 for the controls.

5.8.2 Results and Discussion

The low RH treatment significantly increased ethylene synthesis in the 24 hours following moisture stress (t-test p=0.025). The effect was highly significant after 48 hours low RH storage (t-test p=0.001) (Fig. 5.8.1). Severe moisture stress, of around 9-10% weight loss, caused an ethylene evolution rate 50 times greater than in the control. A significant effect could not be detected after 24 hours low RH storage, despite the rate of ethylene synthesis being more than 7 times higher than the control, due to the small number of replicates (t-test, p=0.068). It is likely that moisture loss of 4-5% substantially increases ethylene production, but further experimentation is required to confirm this finding. Moisture loss has similarly been shown to increase ethylene release in many fruits and vegetables (Aharoni *et al.* 1975; Littmann 1972; Kader 1985).

Moisture stress did not strongly affect CO_2 evolution, with the low RH treatments not significantly different to the control (Fig. 5.8.1).

5.8.3 Conclusions

This preliminary study shows that further experimentation in this area is warranted. Moisture stress was shown to significantly increase ethylene production, with severe desiccation resulting in rates 50 times higher than hydrated fruit. A significant effect on production of CO_2 could not be detected. Further experimentation, using a greater number of replicates and a variety of levels of water stress would be beneficial, as would research into the effects of ethylene on the lychee fruit.

5.9 The Development of a Water Loss Model for Lychee

A basic model using Microsoft Excel was developed to predict the loss of moisture from lychee during postharvest operations. The model was based on properties of lychee previously measured, including specific moisture loss and response to air currents.

5.9.1 Methods and Materials

The model was based on specific moisture loss (SML), an index of the ability of the fruit to resist moisture loss. This parameter varies substantially in lychee, but generally averages around 2×10^{-10} $kgkg^{-1}Pa^{-1}s^{-1}$ (Section 5.1.2.1). The vapour pressure deficit (VPD) was calculated by temperature and RH parameters. The duration of exposure, SML and VPD can be used to estimate moisture loss. The effects of initial fruit water content and air currents on moisture loss were also considered.

Air saturation vapour pressure was calculated by;

$e_o = 610.8 \times 2.7183^{[17.27 \times T / (T + 237.3)]}$ ***…Equation 5.9.1***

Where, e_o = saturated air vapour pressure (Pa) and T = air temperature (^oC).

The air vapour pressure was then calculated by;

$VP_A = e^o - (e^o \times RH / 100)$ ***…Equation 5.9.2***

Where, VP_A =air vapour pressure (Pa) and RH = relative humidity (%).

Similarly, fruit vapour pressure was calculated by;

$VP_F = 610.8 \times 2.7183^{[17.27 \times (0.2 \times FT + 0.8 \times T) / (0.2 \times FT + 0.8 \times T) + 237.3]}$ ***…Equation 5.9.3***

Where, VP_F = fruit vapour pressure (Pa) and FT = core fruit temperature (^oC) (equations adapted from Schaefer (1989)).

The vapour pressure deficit (VPD) was therefore simply;

$VPD = VP_F - VP_A$ ***…Equation 5.9.4***

Moisture loss was then calculated by;

$ML = VPD \times t \times SML \times AC \times PML \times 100$ ***…Equation 5.9.5***

Where ML = moisture loss (% fruit weight), t = time (seconds), SML = specific moisture loss ($kgkg^{-1}Pa^{-1}s^{-1}$), AC = air current factor and PML = previous moisture loss factor (equation adapted from van Beek and Lamers (1979), as described by Scheer (1994)).

The effect of air currents on moisture loss were estimated from a previous experiment (Section 5.4.2), where a linear equation was found to relate relative moisture loss to air speed;

$RL = 4.2257 \times S + 0.2165$ ***…Equation 5.9.6***

Where, RL = relative level of moisture loss and S = air speed (msec^{-1})

Based on results from the same experiment, a correction for previous moisture loss was also added. Loss of moisture declined as water stress increased, according to the equation;

PML = -5.0244 x ML + 100 ***…Equation 5.9.7***

Where, PML = previous moisture loss factor (subsequent moisture loss compared to the initial rate (%)) and ML = total moisture loss, as a percentage of fruit weight.

The model was based on several assumptions that limit its value. Fruit were assumed to be in a single layer, without any protective packaging. More complex modeling is required for bulk or packaged produce. Heat transfer was not included in the model, but it was assumed that the fruit surface would more closely mimic air temperature than internal pulp temperature. This was weighted by 80% to air and 20% to core temperature in equation 5.9.3. The proposed model has not been validated by experimentation.

5.9.2 Results and Discussion

The model suggests that recommended handling practices should not cause desiccation browning. Fruit following recommended handling practices were assumed to spend 2 hours in the field at harvest (30^{o}C and 65% RH with a light breeze, 1.5 msec^{-1}). The fruit were then hydro-cooled and packaged using a liner. The RH surrounding packaged fruit was assumed to be 75%. Fruit were stored in the farm coolroom at 5^{o}C for 6 hours and then transported at 7^{o}C for 12 hours. At wholesale level, fruit were assumed to be stored in coolrooms at 5^{o}C for 3 hours and then displayed at 25^{o}C for 2 hours. Retail transport (7^{o}C) lasting 2 hours was followed by retail display at 20^{o}C and

40% RH, with air currents (1 msec^{-1}) for 6 hours. Total moisture loss after this regime was estimated by the model at 4.4% of fruit weight, a level unlikely to result in browning.

Harvest conditions measured during the lychee season on a variety of farms were typically 25-30^{o}C and 60-75% RH. When exposure to these conditions is limited to 2 hours without any wind, estimated moisture loss ranges from 0.1 to 0.25%. However, a light breeze (1.5 msec^{-1}) increases the range from to 0.7 to 1.6% moisture loss. Obviously, any increases in time directly influence moisture loss. An additional 2 hour delay at the packing shed under these conditions increases estimated moisture loss to 1.4 to 3.2%. Greater wind speed may also substantially increase loss. The effects of wind speeds over 1.55 msec^{-1} have not been determined. Protecting fruit from air currents in the field would be likely to be highly beneficial.

Pre-cooling in a coolroom typically causes substantial moisture loss. For example, 5 hours at 5^{o}C and 50% RH with 1 msec^{-1} air speed results in an estimated 1.2% loss of weight. In contrast, hydro-cooling causes negligible weight loss, and may even increase water content by absorption (Olesen 2001). Forced air cooling substantially reduces the duration of pre-cooling, therefore reducing moisture loss. However, the balance between reduced duration, and the increase in air speed under forced air may limit the benefits for lychee. A decrease in pre-cooling time to 1 hour, under greater air speed (2 msec^{-1}) results in an estimated 0.5% moisture loss (assuming a further linear increase in response to air currents). Increased moisture loss through greater air speed may reduce the benefits of forced air cooling.

After packaging, the liner maintains a high level of humidity around the fruit and protects against air currents, reducing the subsequent rate of moisture loss. Low temperatures combined with plastic packaging result in minimal moisture loss. For example, 24 hours at 5^{o}C and 75% RH results in an estimated weight loss of only 0.4%. Delays while fruit are cooled, stored at low temperature and packaged appear to be of minimal importance.

Transport is a potential location of moisture loss due to temperature fluctuation. A steady 7 or 12^{o}C transport period of 12 hours is predicted to cause around 0.2% loss of weight. However, increases in temperature reduce RH, due to the limited capacity of cool air to hold moisture. For example, a sealed space at 5^{o}C and 90% RH changes to around 65% RH at 10^{o}C and 35% RH at 20^{o}C. Additional moisture is drawn from the fruit by this process. Re-cooling then results in condensation of the lost moisture. Cycling of temperature can cause substantial moisture loss by this process. Similar large temperature fluctuations can also occur during wholesale and retail marketing. These effects are beyond the scope of this model, but may contribute significantly to desiccation in practice.

The retail environment has the potential to cause substantial moisture loss, as fruit are removed from protective packaging and exposed to low RH and air movement. Storage conditions vary widely between stores, but air conditioning typically results in 20 to 25^{o}C, 30 to 50% RH and light air movement (estimated at 0.1 to 0.5 msec^{-1}). Over 6 hours, this was estimated to result in anywhere from 0.3 to 2% loss. Refrigerated storage substantially reduces the risk of desiccation. Under 40% RH and 0.5 msec^{-1} air speed, 5^{o}C storage results in an estimated loss of 0.5% over 6 hours, while 25^{o}C results in 1.7%. Storage duration is obviously also important. A staff member at a large supermarket suggested a rate of turnover of around 6 lychee boxes per day, with 2 boxes constantly on display. Each box would therefore be open for roughly 4 hours. The boxes were refrigerated, so rates of moisture loss in these conditions would be minimal. However, the fruit were left on display overnight, and were not covered, which would result in more substantial loss.

5.9.3 Conclusions

The model emphasised the importance of storing fruit at constant low temperatures and protecting from air currents. Major risk areas for moisture loss included harvest, pre-cooling, transport and retail marketing. Optimal practices to minimise moisture loss include protecting fruit from air currents after harvest, rapidly hydro-cooling, packaging in plastic and maintaining cold temperatures throughout the postharvest chain. Further research to validate this model would be beneficial.

5.10 Overview - Lychee Postharvest Desiccation

Cultivar differences in moisture loss were not significant, and were overshadowed by huge variation due to unknown pre-harvest factors (Section 5.1), Further research into the influence of pre-harvest variables on postharvest moisture loss may be of significant economic value in reducing desiccation browning. A harvest date effect was detected, with both under-mature and over-mature fruit showing higher rates of postharvest moisture loss. The relationship between moisture loss and browning was significantly affected by cultivar, but also showed a large degree of variation due to unknown variables (Section 5.2). Cultivars with thicker skin tended to lose more moisture before showing browning. The smaller relative volume of the pericarp in thin-skinned fruits may result in a lower threshold of moisture loss.

Fruit spanning the typical commercial maturity range showed no significant variation in moisture loss (Section 5.3). Browning was generally unaffected by maturity, although some differences in colour change were observed at high levels of moisture loss.

Air currents strongly affected moisture loss, with a light breeze capable of increasing loss by a factor of seven (Section 5.4). These results confirm previous observational reports of air currents accelerating lychee desiccation (Bagshaw *et al.* 1991). Over the range of air speeds tested, speed was linearly related to rates of moisture loss. The strong effect of air currents on moisture loss gives further impetus to the use of protective packaging such as punnets. The relationship between moisture loss and browning was unaffected by RH (Section 5.5).

Rates of moisture loss occurred fairly evenly over the surface of the fruit, with the stem not contributing significantly (Section 5.6). Commercially handled fruit showed a greater proportional loss of moisture from the protuberance tips, suggesting that tip damage may enhance loss. Previous microscopy research has shown localised cuticle degradation on the protuberance tips, thought to result from mechanical injury (Underhill and Simons 1993). Blocking lychee protuberance tips with petroleum jelly substantially reduced rot development, suggesting that this may be a crucial location in pathogen entry. Epicuticular waxes were shown to contribute significantly to protection against moisture loss (Section 5.7), but were not nearly as important as in apple (Horrocks 1964).

Moisture loss was shown to significantly increase ethylene evolution, with rates 50 times higher observed in severely water stressed fruit (Section 5.8). Increased ethylene release is a typical response to moisture stress (Kader 1985). Carbon dioxide generation in lychee was slightly raised in water stressed fruit, but a significant effect was not detected.

The development of a water loss model for lychee emphasised the importance of constant low temperatures and protection from air currents in minimising moisture loss (Section 5.9). Major risk areas in postharvest handling, such as harvest, pre-cooling, transportation and retail display require particular attention to avoid desiccation browning.

CHAPTER 6 - GENERAL DISCUSSION

Mechanical damage was revealed as a significant issue in the maintenance of lychee quality after harvest. Impact and vibration damage were shown to occur in practice, and were capable of causing a significant decline in fruit quality. Some simple changes to postharvest management may minimise the detrimental effects of these mechanical injuries.

6.1 Impact Damage

Impact was capable of causing open cracking of the lychee pericarp, resulting in significant degradation of visual appeal and reduced shelf life. Open cracking caused a significant increase in ion leakage (Section 2.8), ethylene output (Section 2.9) and pathogen establishment (Section 2.13). These types of responses are commonly observed in injured fruit and vegetables (Ruiz Altisent 1991). A very low incidence of open cracking was observed at wholesale level (Section 2.14), showing that this type of injury can occur in practice, but would generally be removed during sorting. The threshold fall height for open cracking varied with factors such as cultivar and fruit hydration, but tended to be in the 40-70 cm range (Sections 3.1, 4.1 and 4.10).

Low impacts caused protuberance tip darkening and closed cracking. These symptoms are unlikely to be of commercial significance, given the minimal effect on fruit appearance. Low levels of closed crack and tip darkening damage were observed at wholesale level (Section 2.14), suggesting that impact injury does occur, but is not a major concern.

Ethylene synthesis (Section 2.9), aril quality (Section 2.1) and shelf life (Section 2.13) appeared to be unaffected by impacts not causing open cracking. However, impact damage not causing open cracking resulted in some significant physiological changes in the fruit. Moisture loss from the fruit was significantly increased by impact, particularly in the 24 hours following injury (Section 2.10). Accelerated moisture loss has been noted as a typical response to mechanical injury in fruit and vegetables (Wills *et al.* 1998). Carbon dioxide output was increased by impact, and was not correlated with cracking (Section 2.9). This response is likely to have resulted from the decarboxylation of malic acid spilled from damaged cells (Hung 1993). Significant changes in skin colour, including darkening and yellowing, were observed after impact (Section 2.1). The extent of colour change increased gradually with fall height (Section 3.1). Impact injury caused a significant decline in phenolics, possibly due to browning reactions in the tissue (Section 2.7). A similar decline in phenolics is observed during desiccation browning in lychee (Jiang and Fu 1999; Zhang *et al.* 2001). Cuticle weight declined after impact, possibly due to weakened tissue being more easily lost through mild abrasion during storage (Section 2.6). A possible wound healing response was observed, with the average weight of cuticles in damaged fruit increasing in the days following injury (Section 2.6). Impact injured fruit without open cracks tended to show increased resistance to stem end rot and mycelial growth, suggesting a possible defence mechanism triggered by impact (Section 2.13). Mechanical injury was similarly observed to trigger defences against pathogens in oranges (Stange *et al.* 1993a).

Fruit characteristics such as hydration, cultivar and morphology were found to affect susceptibility to impact damage. Mild water stress decreased tip darkening (Section 4.1) and the incidence and size of cracks resulting from impact (Section 4.2). A similar response is shown in potato, where shatter cracking is more common in hydrated tubers (Bland *et al.* 1987). Chilled fruit showed reduced tip darkening, but may have been slightly more susceptible to open cracking (Section 4.6). Cold fruit are generally more susceptible to brittle failure (Hyde 1999). In lychee, large seed size (Section 4.9) and thick skin (Section 4.10) appeared to confer resistance to cracking. The thick-skinned cultivar ‘Bengal’ showed greater tolerance to impact than ‘Salathiel’, ‘KMP’ and ‘Wai Chee’ (Section 4.10).

Load factors such as application method, site, cushioning and repetition affected the degree of damage sustained due to impact. Pendulum impacts resulted in a greater area of cracking than normal falls, possibly due to the slight compressive force of the clamp (Section 3.4). Impacts to the cheek resulted in greater cracking than other sites on the fruit surface. A second impact caused significant colour change and tip darkening, with changes halved compared to the initial impact (Section 3.5), in a pattern similar to that described by Sitkei (1986). Subsequent impacts did not cause further significant damage. Cracking did not significantly increase with repeated impacts. The use of padding can often significantly reduce damage levels in fruit (Brown *et al.* 1990), and this was confirmed in lychee, with cushioning reducing impact damage, including open cracking (Section 3.6).

6.2 Compression Damage

Contrary to previous reports (Batten and Loebel 1984; Lindsay and Cull 1986), compression injury did not appear to be a major concern in lychee. Compression injury generally resulted in slight tip darkening and changes in skin colour, but the aril appeared unaffected. Rapid darkening and loss of chroma were shown within 4 hours of injury (Section 2.2). Puncture, shape distortion and cracking could occur due to compression, but required extreme loads (Section 2.2), and were not observed in practice (Section 2.14). Generally, fruit could tolerate 1 kg load for 48 hours with minimal loss of quality (Section 3.3). This translated into a stacking depth of at least 1 metre, in contrast to the 30 cm depth recommended by Batten and Loebel (1984). Physiological effects of compression injury were minimal, with no significant changes in phenolics (Section 2.7), ion leakage (Section 2.8) or ethylene synthesis (Section 2.9). Increased CO_2 release was observed after compression, but a sustained rise in respiration did not occur (Section 2.9).

Fruit hydration strongly affected the mechanical properties of lychee under compression (Section 4.3). Moderate water stress resulted in loss of elasticity, and increased deformation under a given load, a typical response in fruit and vegetables (Sitkei 1986). Temperature influenced symptom development to some extent, with cool temperatures minimising damage (Section 4.6). Slight differences in response to compression were observed between cultivars, with ‘Bengal’ showing greater loss of chroma than ‘KMP’ (Section 4.10). ‘Bengal’ also showed greater loss of chroma after impact, suggesting possible biochemical differences in the tissue.

6.3 Vibration and Abrasion Damage

Although abrasion alone had little impact on visual appeal, the repeated abrasion damage resulting from vibration severely degraded fruit appearance (Section 2.3). Vibration damage was characterised by strong yellowing of colour, in addition to darkening and loss of chroma. Scuff marks and a powdery appearance were also typical. The injury was cosmetic, with no noticeable change in aril quality.

Yellow residue on the fruit surface after vibration was thought to result from the crystallisation of leaked cell contents. The chemical properties of the residue were generally consistent with pericarp juice (Section 2.4). SEM study revealed accretions of tissue scattered over the fruit surface, giving the typical powdery appearance of vibration damage (Section 2.5). The powdery appearance of the surface, enhanced by dissecting microscope or hand lens, was identified as a valuable tool in differentiating desiccation and vibration browning.

Vibration damage caused some significant physiological changes. Carbon dioxide and ethylene output were significantly increased by vibration (Section 2.9). Significant acceleration of postharvest weight loss was also observed, particularly immediately following injury (Section 2.12). These changes in physiology are typical in mechanically injured fruit (Wills *et al.* 1998). Increases in both ethylene and weight loss were related to the SA of damage, but not the severity of injury. Vibration did not significantly affect levels of phenolics (Section 2.7) or ion leakage (Section 2.8). No significant effect on pathogen invasion was detected in mildly damaged fruit (Section 2.13), but incidental data from severely damaged fruit suggested that strong vibration damage encourages rots (Section 2.3).

Vibration damage was confirmed to be a major quality issue in practice. Vibration damage was observed in the wholesale survey, with most fruit affected to some extent (Section 2.14). The damage appeared to occur both before and after packing. Mechanical destalking was confirmed to have the potential to inflict severe vibration damage (Section 3.8). The lack of previous reference to vibration injury is likely to have occurred due to confusion with desiccation browning.

Fruit hydration, surface wetness and cultivar were shown to significantly affect the fruit response to abrasion and vibration damage. Water stressed fruit showed increased colour change due to injury, including darkening, loss of chroma and yellowing (Section 4.4). A similar response was observed in pears, with reduced hydration resulting in greater susceptibility to friction discolouration (Amarante *et al.* 2001). In lychee, the response was significant after as little as 4% weight loss. Surface wetness strongly increased yellowing due to abrasion and vibration, and increased the incidence of scuff injuries (Section 4.5). Reports of surface wetness affecting mechanical injury in other fruits have not been located. Fruit temperature had little effect on vibration damage (Section 4.8). Cultivar variation was shown to occur, with smooth-skinned ‘Salathiel’ fruit highly susceptible to vibration damage (Section 4.10).

Within the range tested, increased vibration duration and frequency both increased the scale of damage (Section 3.2). At low frequency, extending the duration had little effect, while at high frequency a longer duration strongly exacerbated damage. It was anticipated that box liners could increase vibration damage where surface wetness occurred, but liners did not affect the scale of damage in dry fruit (Section 3.7).

6.4 Pepperspot

Pepperspot has been referred to as a purely cosmetic disorder, with no effect on shelf life (Drew 1999). However, increased pepperspot resulted in more rapid establishment of some postharvest fungi, characterised by low, dense mycelial growth white to pale green in colour (Section 2.13). The more rapid rot may occur through pepperspot weakening the pericarp defences. Alternatively, the characteristics that render the fruit susceptible to pepperspot may also confer poorer resistance to rots.

Moderate pepperspot reduced shelf life at RT by around one day due to the more rapid establishment of pathogens. However, pepperspot did not affect ethylene or CO_2 production (Section 2.9), or postharvest moisture loss (Section 2.11).

6.5 Desiccation Browning

Cultivar differences in moisture loss were inconsistent, and were eclipsed by variation due to unknown seasonal or location factors (Section 5.1). The relationship between moisture loss and browning varied consistently between cultivars, with thicker skin appearing to convey greater resistance to browning (Section 5.2). Harvest date was shown to influence postharvest weight loss, with higher rates occurring in immature and over-mature fruit (Section 5.1). Wu *et al.* (2001) previously showed slight differences in lychee moisture loss with extreme changes in maturity. However, fruit over a commercially acceptable range of maturity levels showed no significant variation in weight loss or browning (Section 5.3).

The success of protective barriers in reducing lychee desiccation browning (Bagshaw *et al.* 1991; Bagshaw 1995; Huang and Scott 1985; Scott *et al*. 1982; Wong *et al.* 1991) suggests that RH and air currents may be important in the process. Air currents were revealed as a key factor in moisture loss, with a light breeze increasing the rate of loss by a factor of seven (Section 5.4). Over the range tested, a linear relationship between air speed and moisture loss was shown.

Rates of moisture loss appeared to be fairly even over the lychee fruit surface, including the stem (Section 5.6). However, postharvest handling appeared to increase the proportion of loss from the protuberance tips, possibly through mild mechanical injury. This evidence correlates with the localised cuticle damage previously observed by Underhill and Simons (1993) on the protuberance tips.

6.6 Recommendations

Many simple management strategies were identified to maximise fruit quality. Mechanical damage can be reduced by minimising the mechanical loads acting on the fruit, particularly when fruit are in a susceptible condition. The management of air currents was shown to be crucial in avoiding desiccation browning.

6.6.1 Impact

The key objective in managing impact damage is avoidance of open cracking, as this injury renders the fruit unsaleable. Impacts greater than 40 cm have the potential to cause open cracking in lychee, and could be avoided by improved design of packing lines and education of fruit pickers. Cushioning has the potential to reduce cracking, and could be added to existing packing lines to reduce injury. Lychee fruit require more careful handling when turgid, as the fruit are more susceptible to impact cracking. Soaking treatments can increase fruit turgor (Olesen 2001), and are therefore optimally located after impact events in the packing line. Impact damage without open cracking can cause detrimental physiological changes in the fruit. The minimisation of impact events is therefore recommended for maximum quality.

6.6.2 Vibration

Vibration injury causes a major decline in visual appeal, and is a key issue in lychee postharvest quality. To prevent damage, it is optimal to avoid vibration where possible, or to otherwise minimise the duration or frequency. Mechanical destalking can cause substantial vibration damage. It is recommended that growers perform a simple check of their equipment by comparing fruit sampled before and after destalking. Hand destalking is optimal to minimise vibration damage. Wet fruit are particularly susceptible to vibration damage. Harvesting of fruit in dry condition is recommended, as exposure to vibration is likely to occur during transport to the packing shed. Condensation control during transport is also important in reducing the risk of vibration injury. Smooth skinned fruit are more susceptible to vibration injury, and require greater care. Water stressed fruit also show greater susceptibility to vibration damage. Deeper picking trays and cartons may reduce vibration damage, as there are fewer fruit in the vulnerable top layer. The resistance of lychee fruit to compression injury allows cartons of greater depth to be used. The high level of vibration damage observed in practice, and the lack of published information suggests that the injury has previously been misdiagnosed as desiccation browning. It is recommended that hand lenses or dissecting microscopes are used as diagnostic tools to differentiate characteristically powdery vibration injury from moisture loss.

6.6.3 Desiccation

The avoidance of air currents is crucial to the management of desiccation browning in lychee. This could be achieved through the use of punnets, which protect fruit from air currents throughout the postharvest chain. Problems such as cost, wholesaler preference, rots and condensation control have limited the adoption of punnets. Alternatively, exposure to air currents could be minimised by the use of lined cartons, combined with a protective case at the retail level (Bagshaw 1995). Harvest at optimal commercial maturity is recommended to minimise moisture loss, as over-mature and immature fruit show increased postharvest weight loss (Section 5.1). Further research is warranted to compare hydrocooling and forced-air cooling, given the strong effect of air currents on lychee browning.

6.6.4 Other Issues

Basic quality issues, such as insect feeding damage, rot control and fruit maturity were confirmed as continuing problems in the lychee industry. Further extension work in these areas, to develop grower awareness would be valuable in improving the overall quality of lychee fruit.

6.7 Future Research Directions

Desiccation browning is a major cause of postharvest loss, and could most simply be avoided through the protection of fruit from air currents. The development of strategies to refine lychee punnetization would be an important step towards the minimisation of desiccation browning. Further studies on the effects of air currents on lychee moisture loss would be of interest, particularly under actual industry conditions. Unidentified pre-harvest factors appeared to have a major effect on postharvest moisture loss, warranting further research. The harvest date effect on moisture loss requires further research to confirm and explain the trends observed.

A wide variety of cosmetic blemishes were observed on fruit, with many severe enough to cause downgrading. Further study to identify the causes of cosmetic blemishes would be valuable in maximising the visual appeal of the fruit.

Studies to expand upon the research presented in this thesis may include research into aril quality, wound healing and the effects of skin properties on cracking. Visually, the aril appeared to be affected by mechanical injury only when an open crack occurred. The testing of aril quality after mechanical injury, by taste and chemical tests, would be valuable in completely defining the effects of injury on the fruit. Some evidence of wound healing in lychee was observed, warranting further study, including microscopy, to confirm and explore the process. Further study of the effects of skin properties on crack susceptibility would be of interest, particularly pericarp thickness at protuberance base and structural properties such as cell size and arrangement.

Practical issues in lychee mechanical damage require some further exploration. A survey of fruit rejected during sorting would give a greater understanding of the causes of postharvest loss, including mechanical damage. For example, open cracking due to rough handling at harvest may cause significant packing line losses, but this has not yet been assessed. Further comparison of cultivars in the response to mechanical damage is recommended, to develop awareness of varieties requiring particular care. The comparison of commercial cushioning products, including thin steel plates, may allow existing packing lines to be adapted to avoid impact damage. Tight filling of boxes and the use of deeper cartons could substantially reduce vibration damage, but practical trials would be required to confirm the benefits. Further study of mechanical damage in lychee may provide additional strategies to improve quality and reduce losses.

Reference List

Abbott J.A. and Lu R. (1996) Anisotropic mechanical properties of apples. Transactions of the American Society of Agricultural Engineers (ASAE), **39(4)**, 1451-1459.

Aharoni N., Ben-Yehoshua S. and Richmond A.E. (1975) Effects of water stress upon ethylene and endogenous content of abscisic acid and gibberellins in detached lettuce leaves (*Lactuca sativa* L.) Israel Journal of Botany, **24**, 55.

Akamine E.K. (1960) Preventing the darkening of fresh lychees prepared for export. Bulletin, Hawaii Agriculture Experimental Station, **127**, 3-17.

Akamine E.K. and Goo T. (1973) Respiration and ethylene production during ontogeny of fruit. Journal of the American Society of Horticultural Science, **98**, 381-383.

Alayunt F.N, Cakmak B., Can H.Z. and Aksoy U. (1998) Vibration damage trial on some fig cultivars. Acta Horticulturae, **480**, 305-310.

Amarante C., Banks N.H. and Ganesh S. (2001) Effects of coating concentration, ripening stage, water status and fruit temperature on pear susceptibility to friction discolouration. Postharvest Biology and Technology, **21(3)**, 283-290.

Bagshaw J. (1995) Development of an "in-store" dispenser system. In ‘Lychee Postharvest Handling and Marketing, Final Report 94/95’. RIRDC, 104-108.

Bagshaw J., Underhill S. and Prasad A. (1991) Lychee skin browning: its cause and control. Australian Lychee Yearbook, **1**, 9-13.

Bagshaw J.S., Underhill S.J.R. and Fitzell R.D. (1995) Lychee disorders. In ‘Postharvest Diseases of Horticultural Produce, Volume 2, Tropical Fruits’ (Eds Coates L., Cooke T., Persley D, Beattie B., Wade N and Ridgway R.) Department of Primary Industries, Qld, 43-45.

Bajema R.W., Baritelle A.L., Hyde G.M. and Pitts M.J. (2000) Factors influencing dynamic mechanical properties of red 'delicious' apple tissue. Transactions of the ASAE, **43(6)**, 1725-1731.

Banks N.H. and Joseph M. (1991) Factors affecting resistance of banana fruit to compression and impact bruising. Journal of the Science of Food and Agriculture, **56**, 315-323.

Bardaie M.Z. and Hitam B. (1981) Fruit damage during transportation. Conference Proceedings, Agricultural Engineering in National Development, Malaysia, September 1979, 175-178.

Baritelle A.L. and Hyde G.M. (2001) Commodity conditioning to reduce impact bruising. Postharvest Biology and Technology, **21(3)**, 331-339.

Baryeh E.A. (2000) Strength properties of avocado pear. Journal of Agricultural Engineering Research, **76(4)**, 389-397.

Batten D.J. (1986) Harvesting lychees. In ‘The Potential of Lychee in Australia. Proceedings of the First National Lychee Seminar, 14-15 February 1986’ (Eds Menzel C.M. and Greer G.N.) Sunshine Coast Tropical Fruits Association, Nambour Qld, 73-75.

Batten D.J. and Loebel M.R. (1984) Lychee harvesting and post-harvest handling. Agfact H6.4.1, Department of Agriculture, NSW.

Ben-Yehoshua S. (1987) Transpiration, water stress and gas exchange. In ‘Postharvest Physiology of Vegetables’ (Ed Weichmann J.) Dekker, New York, 113-170.

Ben-Yehoshua S. (1969) Gas exchange, transpiration and the commercial deterioration of stored orange fruit. Journal of the American Society for Horticultural Science, **94**, 524.

Bhullar J.S., Dhillon B.S. and Randhawa J.S. (1983) Extending the postharvest life of litchi cultivar Seedless Late. Journal of Research - Punjab Agricultural University, **20**, 467-470.

Bland W.L., Tanner C.B. and Maher E.A. (1987) Vapour conductance of wounded potato tuber tissue. American Potato Journal, **64**, 197-204.

Brouillard R. (1982) Chemical structure of anthocyanins. In ‘Anthocyanins as Food Colours’ (Ed Markakis P.) Academic Press, New York, 1-40.

Brown B.I. (1986) Postharvest handling and storage of lychees. In ‘The Potential of Lychee in Australia. Proceedings of the First National Lychee Seminar, 14-15 February 1986’ (Eds Menzel C.M. and Greer G.N.) Sunshine Coast Tropical Fruits Association, Nambour Qld, 77-78.

Brown G.E. (1973) Development of green mold in degreened oranges. Phytopathology, **63**, 1104-1107.

Brown G.E. and Barmore C.R. (1981) Ultrastructure of the response of citrus epicarp to mechanical injury. Botanical Gazette, **142(4)**, 477-481.

Brown G.K., Schulte-Pason N.L., Timm E.J., Burton C.L. and Marshall D.E. (1990) Apple packing line impact damage reduction. Applied Engineering in Agriculture, **6(6)**, 759-764.

Burdon J. and Clark C. (2001) Effect of postharvest water loss on 'Hayward' kiwifruit water status. Postharvest Biology and Technology, **22(3)**, 215-225.

Burton C.L. and Schulte-Pason N.L. (1985) Assaying fruit impact damage using an infrared CO_2 gas analyzer. ASAE Technical Paper, **85-1564**.

Burton C.L. and Schulte-Pason N.L. (1987) Carbon dioxide as an indicator of fruit impact damage. HortScience, **22(2)**, 281-282.

Burton C.L., Schulte-Pason N.L., Brown G.K. and Timm E.J. (1987) The effect of impact bruising on apples and subsequent decay development. ASAE Technical Paper, **87-6516**.

Campbell (1959) Storage behaviour of fresh Brewster and Bengal lychees. Proceedings of the Florida State Horticultural Society, **72**, 356-360.

Chalmers D.J. and Faragher J.D. (1977) Regulation of anthocyanin synthesis in apple skin. II. Involvement of ethylene. Australian Journal of Plant Physiology, **4(1)**, 111-121.

Chambers T.C. and Possingham J.V. (1963) Studies of the fine structure of the wax layer of Sultana grapes. Australian Journal of Biological Science, **16**, 818-825.

Chen P. and Squire E.F. (1970) An evaluation of the coefficient of friction and abrasion damage of oranges on various surfaces. ASAE Paper, **70-364**.

Chen W., Wu Z., Ji Z. and Su M. (2001) Postharvest research and handling of litchi in China - A review. Acta Horticulturae (Proceedings of the First International Symposium on Litchi and Longan, Guangzhou, China, 16-19 June 2000), **558**, 321-329.

Chen W.J. and Hong Q.Z. (1992) A study on the senescence and browning in the pericarp of litchi (*Litchi chinensis* Sonn) during storage. Acta Horticulturae Sinica, **19(3)**, 227-232.

Chen Y.B., Li C.Y. and Shao Y.J. (1995) Experiment and study on the equilibrium moisture content of litchi. Transactions of the Chinese Society of Agriculture, **11(1)**, 171-174. (Abstract only).

Cobin M. (1954) Lychee in Florida. Bulletin of the Florida Agricultural Experiment Station, **546**, 1-35.

Cooke A.W. and Coates L.M. (2002) Pepper spot: A preharvest disease of lychee caused by*Colletotrichum gloeosporioides*. Australian Plant Pathology, **31**, 303-304.

Cotner S.D., Burns E.E. and Leeper P.W. (1969) Pericarp anatomy of crack-resistant and susceptible tomato fruits. Journal of the American Society for Horticultural Science, **94**, 136-137.

Crisosto C.H., Garner D., Doyle J. and Day K.R. (1993a) Relationship between fruit respiration, bruising susceptibility and temperature in sweet cherries. HortScience, **28**, 132-135.

Crisosto C.H., Johnson R.S., Luza J. and Day K. (1993b) Incidence of physical damage on peach and nectarine skin discolouration development: anatomical studies. Journal of the American Society for Horticultural Science, **118(6)**, 796-800.

Datta S.C., Sarkar K.P. and Lodh S.B. (1963) Storage behaviour of litchi *(Litchi chinensis* Sonn.). Science and Culture, **29**, 405-406.

Dean B.B., Jackowiack J. and Munck S. (1992) Tyrosine synthesis in potato tuber tissue from black-spot susceptible and resistant genotypes. Potato Research, **35**, 49-53.

Dekazos E.D. and Worley J.F. (1967) Induction of callose formation by bruising and aging of red tart cherries. Journal of Food Science, **32**, 287-289.

DeMartino G., Massantini R., Botondi R. and Mencarelli F. (2002) Temperature affects impact injury on apricot fruit. Postharvest Biology and Technology, **25**, 145-149.

Denna D.W. (1970) Transpiration of the waxy bloom in *Brassica oleracea* L. Australian Journal of Biological Science, **23**, 27-31.

Dixon T. (2000) Central Queensland delegates report. Living Lychee, **22**, 8.

Drew H. (1999) Pepper spot - a new disease affecting lychee in Australia. Proceedings of the Fifth National Lychee Seminar, Sunshine Coast, Queensland, Australia 13-15 September 1999, 21-23.

Duvenhage J.A. (1993) Control of post-harvest decay and browning of litchi fruit by sodium metabisulphite and low pH dips. South African Lychee Growers Association Yearbook, **5**, 31-32.

Duvenhage J.A., Mostert M.M. and Marais, J.J. (1995) Postharvest sulphuring and low pH treatment for retention of red skin colour of litchi fruit. South African Lychee Growers Association Yearbook, **7**, 44-46.

English H., Ryall A.L. and Smith E. (1946) Blue mold decay of Delicious apples in relation to handling practices. US Department of Agriculture Circular, **151**, 20pp.

Esaka M., Toyota A. and Kitabayashi M. (1993) Marked induction of basic chitinase in pumpkin in response to wounding. Phytochemistry, **34(3)**, 631-635.

FAO/WHO (1994) Revised draft codex standard for litchi (at step 8) Joint FAO/WHO Standards Programme Codex Alimentarius Commission Draft Report of the 5th Session of the Codex Committee on Tropical Fresh Fruits and Vegetables, Mexico City, 5-9 September 1994.

Fiore G.L., Massantini F. and Mencarelli F. (1992) The extent of mechanical damage and its physiological postharvest effects on eating tomatoes. Colture Protette (Italian), **21(1)**, 85-89.

Fischer D., Craig W.L., Watada A.E., Douglas W. and Ashby B.H. (1992) Simulated in-transit vibration damage to packaged fresh market grapes and strawberries. Applied Engineering in Agriculture, **8(3)**, 363-366.

Fitzell R.D. and Coates L.M. (1995) Lychee disease. In ‘Postharvest Diseases of Horticultural Produce, Volume 2, Tropical Fruits’ (Eds Coates L., Cooke T., Persley D, Beattie B., Wade N and Ridgway R.) Department of Primary Industries, Qld, 41-42.

Flurkey W.H. and Jen J.J. (1978) Peroxidase and polyphenol oxidase activities in developing peaches. Journal of Food Science, **43**, 1826-1831.

Fuleki and Francis (1968) Quantitative methods for anthocyanins. 2. Determination of total anthocyanin and degradation index for cranberry juice. Journal of Food Science, **33**, 78-83.

Gan-Mor S. and Mizrach A. (1990) Absorbing impact energy by elastic plate to reduce fruit damage. ASAE Paper, **90-6014**.

Golomb A., Ben-Yehoshua S. and Sarig Y. (1984) High-density polyethylene wrap improves wound healing and lengthens shelf-life of mechanically harvested grapefruit. Journal of the American Society of Horticultural Science, **109**, 155-159.

Gould K.S., Hammett K.R.W. and Steinhagen S. (1990) Mechanism of bruise resistance in pepino (*Solanum muricatum*) fruit. Annals of Botany, **66(2)**, 155-161.

Greer G.N. (1990) Growing Lychee in South Queensland. Department of Primary Industries, Qld.

Greer G.N. and Smith K.L.C. (1991) Lychee Marketing in Australia. Department of Primary Industries, Qld.

Groff G.W. (1921) The Lychee and Longan. Orange Judd Company, New York.

Guillou R. (1964) Orderly development of produce containers. Proceeding, Fruit and Vegetable Perishables Handling Conference, 23-25 March 1964, University of California, Davis, CA, 20-25.

Hawthorne B.T. and Sutherland P.W. (1991) Wound repair processes in fruit of the *Cucurbita maxima* hybrid 'Delica' and the role of scar tissue in the development of fungal rots on stored fruit. New Zealand Journal of Crop and Horticultural Science, **19**, 53-60.

Higgins J.E. (1917) The lychee in Hawaii. Hawaii Agricultura Experiment Station Bulletin, **44**.

Hilton D.J. (1994) Impact and vibration damage to fruit during handling and transportation. ACIAR Proceedings (Postharvest handling of tropical fruits), **50**, 116-126.

Hinch J.M. and Clarke A.E. (1982) Callose formation in *Zea mays* as a response to infection with *Phytophthora cinnamomi*. Physiol Plant Pathol, **21**, 113-124.

Hinsch R.T., Slaughter D.C., Craig W.L. and Thompson J.F. (1993) Vibration of fresh fruits and vegetables during refrigerated truck transport. Transactions of the ASAE, **36(4)**, 1039-1042.

Holcroft D.M. and Mitcham E.J. (1996) Postharvest physiology and handling of litchi (*Litchi chinensis* Sonn.) Postharvest Biology and Technology, **9(3)**, 265-281.

Holt J.E. and Schoorl D. (1977) Bruising and energy dissapation in apples. Journal of Texture Studies, **7**, 421-432.

Horrocks R.L. (1964) Wax and the water vapour permeability of apple cuticle. Nature, **203**, 547.

Horsfield B.C., Fridley R.B. and Claypool L.L. (1972) Application of theory of elasticity to the design of fruit harvesting and handling equipment for minimum bruising. Transactions of the ASAE, **13(6)**, 746-750.

Huang H.T. (1955) Decolorization of anthocyanins by fungal enzymes. Journal of agriculture and food chemistry, **3**, 141-146.

Huang P.Y. and Scott K.J. (1985) Control of rotting and browning of litchi fruit after harvest at ambient temperature in China. Tropical Agriculture, **62**, 2-4.

Huang S., Hart H., Lee H. and Wicker L. (1990) Enzymatic and colour changes during postharvest storage of litchi fruits. Journal of Food Science, **55(6)**, 1762-1763.

Hudson D.E. (1975) The relationship of cell size, intercellular space and specific gravity to bruise depth in potato. American Potato Journal, **52**, 9-14.

Hung Y.C. (1993) Latent damage: a systems perspective. In ‘Postharvest Handling: A Systems Approach’ (Eds Shewfelt R.L. and Prussia S.E.) Academic Press Inc., San Diego, 211-224.

Hyde G.M. (1999) Impact properties of agricultural commodities. <www.wsu.edu:8080/~gmhyde/ImpactProperties.html>

Hyodo H., Fujinami H., Okada E. and Mochizuki T. (1989) Wound-induced ethylene production and 1-aminocyclopropane-1-carboxylic acid synthase in mesocarp tissue of *Cucurbita maxima*. Advances in Agricultural Biotechnology, **26**, 229-236.

Hyodo H., Tanaka K. and Suzuki T. (1991) Wound-induced ethylene synthesis and its involvement in enzyme induction in mesocarp tissue of *Cucurbita maxima*. Postharvest Biology and Technology, **1(2)**, 127-136.

Imaseki H., Stahmann M.A. and Uritani I. (1968) Production of ethylene by injured sweet potato root tissue. Plant Cell Physiology, **9**, 757.

Ismail M.A. and Brown G.E. (1975) Phenolic content during healing of Valencia orange peel under high humidity. Journal of the American Society for Horticultural Science, **100(3)**, 249-251.

Jaensch L.E. (1996) Biophysical and biochemical aspects of bruising in potato tubers (*Solanum tuberosum* L.). Thesis, The Flinders University of South Australia.

Jiang J.M., Su M. and Lee P. (1986) The production and physiological effects of ethylene during ontogeny and after harvest of litchi fruit. Acta Phytophysiologica Sinica, **12**, 95-103.

Jiang Y.M. (2000) Role of anthocyanins, polyphenol oxidase and phenols in lychee pericarp browning. Journal of the Science of Food and Agriculture, **80(3)**, 305-310.
Jiang Y.M. and Chen F. (1995) A study on polyamine change and browning of fruit during cold storage of litchi (*Litchi chinensis* Sonn.) Postharvest Biology and Technology, **5(3)**, 245-250.

Jiang Y.M. and Fu J.R. (1998) Inhibition of polyphenol oxidase and the browning control of litchi fruit by glutathione and citric acid. Food Chemistry, **62**, 49-52.

Jiang Y.M. and Fu J.R. (1999) Biochemical and physiological changes involved in browning of litchi fruit caused by water loss. Journal of Horticultural Science and Biotechnology, **7(1)**, 43-46.

Jiang Y.M., Liu S.X., Chen F., Li Y.B. and Zhang D.L. (1997a) The control of postharvest browning of litchi fruit by sodium bisulphite and hydrochloric acid. Tropical Science, **37(3)**, 189-192.

Jiang Y.M., Zauberman G. and Fuchs Y. (1997b) Partial purification and some properties of polyphenol oxidase extracted from litchi fruit pericarp. Postharvest Biology and Technology, **10(3)**, 221-228.

Johnson G.I. (1989) Lychee disease control. Proceedings of the 2nd National Lychee Seminar, Cairns, 21-23 September, 1989, Far North Queensland Lychee Growers Association, Earlville, Qld, 90-93.

Joubert A.J. and van Lelyveld J. (1975) An investigation of preharvest browning of litchi peel. Phytophylactica, **7**, 9-14.

Joyce D. and Patterson B. (1994) Postharvest water relations in horticultural crops: principles and problems. ACIAR Proceedings, **50**, 228-238.

Kadam S.S. and Deshpande S.S. (1995) Lychee. In ‘Handbook of Fruit Science and Technology’ (Eds Salunke D.K. and Kadam S.S.) Dekker, New York, 435-443.

Kader A.A. (1985) Ethylene-induced senescence and physiological disorders in harvested horticultural crops. HortScience, **20**, 54-57.

Kader A.A. (1994) Modified and controlled atmosphere storage of tropical fruits. In ‘Postharvest Handling of Tropical Fruit’ (Eds Champ B.R., Highley E. and Johnson G.I.) ACIAR Proceedings, **50**, 239-249.

Kahn V. (1985) Tropolone- a compound that can aid in differentiating between tyrosinase and peroxidase. Phytochemistry, **24**, 915-920.

Kaiser C. (1995) Litchi (*Litchi chinensis* Sonn.) pericarp colour retention – Alternatives to sulphur. South African Lychee Growers Association Yearbook, **7**, 47-52.

Kaiser C. (1998) An overview of postharvest storage of mauritius litchi fruit. Journal of the Southern African Society for Horticultural Science, **9**, 57.

Kaiser C., Levin J. and Wolstenholme B.N. (1995) Vapour, heat and low pH dips improve litchi (*Litchi chinensis* Sonn.) pericarp colour retention. Journal of the Southern African Society for Horticultural Science, **5(1)**, 7-10.

Kawano S. and Iwamoto M. (1979) Analysis of vibrating characteristics for highly stacked corrugated fiberboard boxes. ASAE Paper, **79-6016**.

Kellicut K.Q. (1960) Compressive strength of boxes - Part III. USDA Forest Service, Note No. 13.

Ketsa S. and Atantee S. (1998) Phenolics, lignin, peroxidase activity and increased firmness of damaged pericarp of mangosteen fruit after impact. Postharvest Biology and Technology, **14(1)**, 117-124.

Kilili A.W., Behboudian M.H. and Mills T.M. (1996) Postharvest performance of 'Braeburn' apples in relation to withholding irrigation at different stages of the growing season. Journal of Horticultural Science, **71**, 693-701.

Klein J.D. (1987) Relationship of harvest date, storage conditions and fruit characteristics to bruise suseptibility of apple. Journal of the American Society for Horticultural Science, **112**, 113-118.
Kremer-Kohne S. and Lonsdale J.H. (1991) Maintaining market quality of fresh lychees during storage. Part 1: Control of browning. Australian Lychee Yearbook, **1**, 77-81.

Kubo Y., Xue Y.B., Nakatsuka A., Mathooko F.M., Inaba A. and Nakamura R. (2000) Expression of a water stress-induced polygalacturonase gene in harvested cucumber fruit. Journal of the Japanese Society for Horticultural Science, **69(3)**, 273-279.

Kumcha U. (1998) The effects of cultural practices and environmental factors on fruit development in lychee (*Litchi chinensis* Sonn.). Thesis, University of Queensland.

Kumcha U., Hetherington S.E. and McConchie C. (1996) Fruit growth and development in lychee (*Litchi chinensis* Sonn.). Australian Lychee Growers Association 4th National Seminar, **4**, 96-100.

Lallu N., Rose K., Wiklund C. and Burdon J. (1999) Vibration induced physical damage in packed Hayward kiwifruit. Acta Horticulturae, **498**, 307-312.

Landrigan M., Morris S.C. and Gibb K.S. (1996a) Relative humidity influences postharvest browning in rambutan (*Naphelium lappaceum* L.) HortScience, **31(3)**, 417-418.

Landrigan M., Morris S.C. and McGlasson B.W. (1996b) Postharvest browning of rambutan is a consequence of water loss. Journal of the American Society of Horticultural Science, **121(4)**, 730-734.

Landrigan M., Sarafis V., Morris S.C. and McGlasson B.W. (1994) Structural aspects of rambutan (*Naphelium lappaceum*) fruits and their relation to postharvest browning. Journal of Horticultural Science, **69(3)**, 571-579.

Lee H.S. and Wicker L. (1991a) Anthocyanin pigments in the skin of lychee fruit. Journal of Food Science, **56(2)**, 466-468; 483.

Lee H.S. and Wicker L. (1991b) Quantitative changes in anthocyanin pigments of lychee fruit during refrigerated storage. Food Chemistry, **40(3)**, 263-270.

Leon J.M. and Bukovac M.J. (1978) Cuticle development and surface morphology of olive leaves with reference to penetration of foliar applied chemicals. Journal of the American Society for Horticultural Science, **103**, 465-472.

Liang H.H., Ji Z.L. and Huang X.Y. (1998) Study on the techniques of package and storage for litchi fruit stored in the room temperature. Journal of Fruit Science, **15(2)**, 158-163 (Abstract).

Lichtensteiger M.J., Holmes R.G., Hamdy M.Y. and Blaisdell J.L. (1988) Impact parameters of spherical viscoelastic objects and tomatoes. Transactions of the ASAE, **31(2)**, 595-602.

Lichter A., Dvir O., Rot I., Akerman M., Regev R., Wiesblum A., Fallik E., Zauberman G. and Fuchs Y. (2000) Hot water brushing: an alternative method to SO_2 fumigation for colour retention of litchi fruits. Postharvest Biology and Technology, **18**, 235-244.

Lidster P.D. and Tung M.A. (1980) Effects of fruit temperature at time of impact damage and subsequent storage temperature and duration on the development of surface disorders in sweet cherries. Canadian Journal of Plant Science, **60**, 555-559.

Lidster P.D., Muller K. and Tung M.A. (1980) Effects of maturity on fruit composition and suceptibility to surface damage in sweet cherries. Canadian Journal of Plant Science, **60**, 865-871.

Lin Z.F., Li S.S., Zhang D.L., Liu S.Z., Li Y.B., Lin G.Z. and Chen M.D. (1988) The changes of oxidation and peroxidation in postharvest litchi fruit. Acta Botanica Sinica, **30**, 382-387.

Lindsay P. and Cull B. (1986) Lychee (*Litchi chinensis*). Fruit Growing in Warm Climates, Reed Books Pty Ltd, Frenches Forest, NSW, 40-51.

Littmann M.D. (1972) Effect of water loss on the ripening of climacteric fruits. Queensland Journal of Agricultural and Animal Sciences, **29**, 103-113.

Lonsdale J.H. (1993) Maintaining market quality of litchis overseas without SO_2. South African Lychee Growers Association Yearbook, **5**, 25-28.

Lougheed E.C. and Argue L.W. (1987) Air movement effects in storage. In ‘Postharvest Physiology of Vegetables’ (Ed Weichmann J.) Dekker, New York, 285-302.

Lougheed E.C. and Franklin E.W. (1974) Ethylene production increased by bruising of apples. HortScience, **9**, 192-193.

Lownds N.K., Banaras M. and Bosland P.W. (1993) Relationships between postharvest water loss and physical properties of pepper fruit (*Capsicum annuum* L.) HortScience, **28(12)**, 1182-1184.

Lyon G.D., Reglinski T. and Newton A.C. (1995) Novel disease control compounds: the potential to 'immunize' plants against infection. Plant Pathology, **44(3)**, 407-427.

Macfie G.B. Jr. (1955) Wrapping and packaging of fresh lychee. Florida Lychee Growers Association Yearbook, **1**, 25-28.

Macheix J.J., Fleuriet A. and Billot J. (1990) Fruit Phenolics, CRC Press, Boca Raton, Florida.

MacLeod R.F., Kader A.A. and Morris L.L. (1976) Stimulation of ethylene and CO_2 production of mature-green tomatoes by impact bruising. HortScience, **11(6)**, 604-606.

Maguire K.M., Banks N.H. and Lang A. (1999a) Sources of variation in water vapour permeance of apple fruit. Postharvest Biology and Technology, **17(1)**, 11-17.

Maguire K.M, Lang A., Banks N.H., Hall A., Hopcroft D. and Bennett R. (1999b) Relationship between water vapour permeance of apples and micro-cracking of the cuticle. Postharvest Biology and Technology, **17**, 89-96.

Markakis P. (1982) Stability of anthocyanins in foods. In ‘Anthocyanins as Food Colours’ (Ed Markakis P.) Academic Press, New York, 163-180.

Mathew A.G. and Parpia H.A.B. (1971) Food browning as a polyphenol reaction. Advances in Food Research, **19**, 75-145.

Mayer A.M. and Harel E. (1979) Polyphenol oxidase in plants. Phytochemistry, **18**, 193-215.

McDonald R.E., Nordby H.E. and McCollum T.G. (1993) Epicuticular wax morphology and composition are related to grapefruit chilling injury. HortScience, **28(4)**, 311-312.

McGuire R.G. (1992) Reporting of objective color measurements. HortScience, **27(12)**, 1254-1255.

McLaughlin N.B. and Pitt R.E. (1984) Failure characteristics of apple tissue under cyclic loading. Transactions of the ASAE, **27(1)**, 311-320.

Mellenthin W.M. and Wang C.Y. (1974) Friction discoloration of 'd'Anjou' pears in relation to fruit size, maturity, storage and polyphenoloxidase activities. HortScience, **9**, 592-593.

Mencarelli F. and Botondi R. (1992) The effects of superficial abrasion simulating the practice of brushing on ethylene production in pear fruits. Rivista di Frutticoltura e di Ortofloricoltura (Italian/English), **54(7-8)**, 69-71.

Menesatti P., Beni C., Paglia G., Marcelli S. and Dandrea S. (1999) Predictive statistical model for the analysis of drop impact damage on peach. Journal of Agricultural Engineering Research, **73(3)**, 275-282.

Menzel C., Bagshaw J., Campbell T., Greer N., Noller J., Olesen T., Waite G., Kernot I., Chapman L. and Rigden P. (2002) Lychee Information Kit, Agrilink.

Menzel C.M. and Simpson D.R. (1986) Lychee cultivars. Queensland Agricultural Journal, May-June, 125-136.

Menzel C.M., Olesen T., and McConchie C.A. (1999) Making a profit from lychees in Australia. Proceedings of the Fifth National Lychee Seminar, Sunshine Coast, Queensland, Australia 13-15 September 1999, 5-15.

Miller A.R., Dalmasso J.P. and Kretchman D.W. (1987) Mechanical stress, storage time, and temperature influence cell wall-degrading enzymes, firmness, and ethylene production by cucumbers. Journal of the American Society for Horticultural Science, **112**, 666-671.

Mohamed S. and Othman E. (1988) Effect of packaging and modified atmosphere on the shelf life of rambutans (*Nephilium lappaceum* L.) - II. Pertanika, **11**, 217-228.

Mohsenin N.N. (1986) Physical Properties of Plant and Animal Materials. Gordon and Breach Science Publishers New York.

Moretti C.L., Sargent S.A., Huber D.J., Calbo A.G. and Puschmann R. (1998) Chemical composition and physical properties of pericarp, locule, and placental tissues of tomatoes with internal bruising. Journal of the American Society for Horticultural Science, **123(4)**, 656-660.

Morris S.C., Forbes-Smith M.R. and Scriven F.M. (1989) Determination of optimum conditions for suberization, wound periderm formation, cellular desiccation and pathogen resistance in wounded *Solanum tuberosum* tubers. Physiological and Molecular Plant Pathology, **35**, 177-190.

Mukerjee P.K. (1957) Preservation of natural colour in litchis under cold storage. Science and Culture, **23**, 101-103.

Mulas M., Lafuente M.T. and Zacarias L. (1996) Lignin and gum deposition in wounded 'Oroval' clementines as affected by chilling and peel water content. Postharvest Biology and Technology, **7**, 243-251.

Nagar P.K. (1994) Physiological and biochemical studies during fruit ripening in litchi (*Litchi chinensis* Sonn.) Postharvest Biology and Technology, **4(3)**, 225-234.

Nelson C.W. and Mohsenin N.N. (1968) Maximum allowable static and dynamic loads and effect of temperature for mechanical injury in apples. Journal of Agricultural Engineering Research, **13**, 305-317.

Nip W.K. (1988) Handling and preservation of lychee (*Litchi chinensis* Sonn.) with emphasis on colour retention. Tropical Science, **28(1)**, 5-11.

O'Brien M., Pearl R.C., Vilas E.P. and Dreisbach L. (1969) The magnitude and effect of in-transit vibration damage of fruit and vegetables on processing quality and yield. Transactions of the ASAE, **24**, 452-455.

O'Hare T.J. and Prasad A. (1990) CA storage of rambutan. ACIAR Project 8844, Postharvest Handling of Tropical Fruit, Progress Report to 1 May 1990 for Core Program (Australia), 5-6.

O'Hare T.J., Prasad A. and Cooke A.W. (1994) Low temperature and controlled atmosphere storage of rambutan. Postharvest Biology and Technology, **4**, 147-157.

Olesen T. (2001) Improved postharvest handling of lychee - Preliminary results from the 2000/01 season. Living Lychee, **27**, 24-29.

Olesen T. and Wiltshire N. (2000) Postharvest results from the 1999/2000 season. Living Lychee, **23**, 16-22.

Pang W., Studman C.J. and Ward G.T. (1992) Bruising damage in apple-to-apple impact. Journal of Agricultural Engineering Research, **52(4)**, 229-240.

Pang X.Q., Zhang Z.Q, Duan X.W. and Ji Z.L. (2001) Influence of pH and active oxygen on the stability of anthocyanins from litchi pericarp. Proceedings of the First International Symposium on Litchi and Longan, Guangzhou, China, 16-19 June 2000, Acta Horticulturae, **558**, 339-342.

Pantastico Er.B. (1975) Structure of fruits and vegetables. In ‘Postharvest Physiology, Handling and Utilization of Tropical and Subtropical Fruits and Vegetables’ (Ed Pantastico Er.B.) The Avi Publishing Company, Westport, Connecticut, 1-24.

Patten K.D. and Patterson M.E. (1985) Fruit temperature effects on mechanical damage of sweet cherries. Journal of the American Society for Horticultural Science, **110(2)**, 215-219.

Patterson B.D., Jobling J.J. and Moradi S. (1993) Water relations after harvest - new technology helps translate theory into practice. Australasian Postharvest Conference, 99-102.

Paull R.E., Reyes M.E.Q. and Reyes M.U. (1995) Litchi and rambutan insect disinfestation: treatments to minimise induced pericarp browning. Postharvest Biology and Technology, **6**, 139-148.

Pereira A.V. and Calbo A.G. (2000) Elastic stresses and plastic deformations in 'Santa Clara' tomato fruits caused by package dependent compression. Pesquisa Agropecuaria Brasileira, **35(12)**, 2429-2436.

Piffaut B. and Metche M. (1991) Properties of peroxidase and polyphenol oxidase in natural complexes from walnuts (*Juglans regia*) and in active DL-DOPA copolymers. Journal of the Science of Food and Agriculture, **57**, 493-506.

Pitt R.E. (1982) Models for the rheology and statistical strength of uniformly stressed vegetative tissue. Transactions of the ASAE, **25**, 1776-17784.

Pitt R.E. and Chen H.L. (1983) Time-dependent aspects of the strength and rheology of vegetative tissue. Transactions of the ASAE, **26**, 1275-1280.

Pollack R.L. and Hills C.H. (1956) Respiratory activity of normal and bruised red tart cherries (*Purnus cerasus*). Federation Proceedings, **15**, 328.

Porat R., Lers A., Dori S., Cohen L., Weiss B., Daus A., Wilson C.L. and Droby S. (1999) Induction of chitinase and beta-1,3-endoglucanase proteins by UV irradiation and wounding in grapefruit peel tissue. Phytoparasitica, **27(3)**, 233-238.

Prusky D. (1998) Mechanisms of resistance of fruit and vegetables to postharvest diseases. Disease Resistance in Fruit, Proceedings of an International Workshop Held at Chaing Mai, Thailand, 18-21 May 1997, ACIAR Proceedings **80**, 19-33.

Prusky D. and Keen N.T. (1995) Inducible preformed compounds and their involvement in the resistance of plant pathogens. In ‘Novel Approaches to Integrated Pest Management’ (Ed Reuveni R.) Lewis Publishers, Boca Raton, Florida, 139-152.

Puchalski C. and Brusewitz G.H. (1996) Watermelon surface abrasion - a sensory method. International Agrophysics, **10(2)**, 117-122.

Quintana M.E.G. and Paull R.E. (1993) Mechanical injury during postharvest handling of 'Solo' papaya fruit. Journal of the American Society for Horticultural Science, **118(5)**, 618-622.

Ranganna S. (1977) Manual of Analysis of Fruit and Vegetable Products. Tata Hill McGraw Hill Publishing Company, New Delhi.

Rastovski A. *et al.* (1987) Storage of Potatoes. Pudoc Wageningen NL. (As referenced in Scheer (1994)).

Ray P.K. (1998) Postharvest handling of litchi fruits in relation to colour retention - a critical appraisal. Journal of Food Science and Technology Mysore, **35(2)**, 103-116.

Rodriguez L., Ruiz M. and De Felipe M.R. (1990) Differences in the structural response of 'Granny Smith' apples under mechanical impact and compression. Journal of Texture Studies, **21(2)**, 155-164.

Ruiz Altisent M. (1991) Damage mechanisms in the handling of fruits. In ‘Progress in Agricultural Physics and Engineering’ (Ed Matthews J.) CAB International, 231-257.

Ruiz Altisent M., Garcia C. and Rodriguez L. (1989) Impact bruises in Pomaceae fruits: Evaluation methods and structural features. Proceedings of the Fifth International Congress on Physical Properties of Agricultural Materials, Rostock, Germany, 698-703.

Ryan C.A. (1984) Defense responses of plants. In ‘Genes Involved in Microbe-Plant Interactions’ (Eds Verma D.P.S. and Hohn Th.), 375-386.

Sala J.M. (2000) Content, chemical composition and morphology of epicuticular wax of Fortune mandarin fruits in relation to peel pitting. Journal of the Science of Food and Agriculture, **80**, 1887-1894.

Saltveit M.E. Jr. (1984) Effects of temperature on firmness and bruising of ‘Starkrimson Delicious’ and ‘Golden Delicious’ apples. HortScience, **19(4)**, 550-551.

Santana Llado J.D. and Marrero Dominguez A. (1999) The effects of peel abrasion on the postharvest physiology and commercial life of banana fruits. Acta Horticulturae, **490**, 547-553.

Schaefer N.L. (1989) In situ measurement of plant water potential. In ‘Modern Methods of Plant Analysis – Volume 9 - Gases in Plant and Microbial Cells (Eds Linskens H.F. and Jackson J.F.) Springer-Verlag, Berlin, 134-161.

Scheer A. (1994) Reducing water loss of horticultural and arable products during long term storage. Acta Horticulturae, **368**, 511-522.

Schippers P.A. (1971) The influence of curing conditions on weight loss of potatoes during storage. American Potato Journal, **48**, 278-286.

Schoorl D. (1972a) Handling in the package. Queensland Fruit and Vegetable News, 25th May, 331-332.

Schoorl D. (1972b) Package performance evaluation. Sixth Australian Fruit and Vegetable Storage Research Conference, 64-73.

Schoorl D. (1974) Packaging of, and mechanical damage to fruit. Thesis, University of Queensland.

Schutte G.C., Botha T. and Kotze J.M. (1990) Post-harvest control of decay and browning of litchi fruit by fungicide dips and paper sheets impregnated with sodium metabisulphite. South African Lychee Growers Association Yearbook, **3**, 10-14.

Scott K.J., Brown B.I., Chaplin G.R., Wilcox M.E. and Bain J.M. (1982) The control of rotting and browning of lychee fruit by hot benomyl and plastic film. Scientia Horticulturae, **16**, 253-262.

Shewfelt R.L. (1993) Stress physiology: A cellular approach to quality. In 'Postharvest Handling: A Systems Approach' (Eds Shewfelt R.L. and Prussia S.E.) Academic Press Inc., San Diego, 257-276.

Shi J., Wang C., An X., Li J. and Zhao M. (2001) Postharvest physiology, storage and transportation of litchi fruits - a review. Proceedings of the First International Symposium on Litchi and Longan, Guangzhou, China, 16-19 June 2000, Acta Horticulturae **558**, 387-391.

Shibairo S.I., Upadhyaya M.K. and Toivonen P.M.A. (1998) Influence of preharvest water stress on postharvest moisture loss of carrots (*Daucus carota* L.) Journal of Horticultural Science and Biotechnology, **73(3)**, 347-352.

Siegelman H.W. and Hendricks S.B. (1958) Photocontrol of alcohol, aldehyde and anthocyanin production in apple skin. Plant Physiology, **33**, 409-413.

Singh P. and Singh I.S. (1995) Physico-chemical changes during fruit development in litchi (*Litchi chinensis* Sonn.) Mysore Journal of Agricultural Sciences, **29(3)**, 252-255.

Sitkei G. (1986) Mechanics of Agricultural Materials - Developments in Agricultural Engineering, Volume 8, Elsevier, Amsterdam, 487pp.

Sittigul C., Sardsud U., Sardsud V. and Chaiwangsri T. (1994) Effects of fruit maturity at harvest on disease development in lychee during storage. ACIAR Proceedings **58**, 9-14.

Slaughter D.C., Thompson J.F. and Hinsch R.T. (1998) Packaging Bartlett pears in polyethylene bags to reduce vibrational injury in transit. Transactions of the ASAE, **41(1)**, 107-114.

Smoot J.J., Houck L.G. and Johnson H.B. (1971) Market diseases of citrus and other subtropical fruits. USDA Handbook No. **398**.

Snowdon A.L. (1990) A Colour Atlas of Postharvest Disease and Disorders of Fruit and Vegetables - Volume 1, General Introduction and Fruits. Wolfe Scientific, Barcelona, Spain, 126-127.

Sommer N.F., Mitchell F.G., Guillou R. and Luvisi D.A. (1960) Fresh fruit temperatures and transit injury. Proceedings of the American Society for Horticultural Science, **76**, 156-162.

Song G.Q., Xiao C.Q. and Bing S.Y. (1997) An original study of the change of absorptive spectrum of browning litchi pericarp and the chemical control. China Fruits, **3**, 30-31.

Sonkar R.K., Ladaniya M.S. and Singh S. (1999) Effect of harvesting methods and post-harvest treatments on storage behaviour of Nagpur mandarin (*Citrus reticulata*) fruit. Indian Journal of Agricultural Sciences, **69(6)**, 434-437.

Spayd S.E., Proebsting E.L. and Hayrynen L.D. (1986) Influence of crop load and maturity on quality and susceptibility to bruising of 'Bing' sweet cherries. Journal of the American Society for Horticultural Science, **111(5)**, 678-682.

Spotts R.A., Sanderson P.G., Lennox C.L., Sugar D. and Cervantes L.A. (1998) Wounding, wound healing and staining of mature pear fruit. Postharvest Biology and Technology, **13(1)**, 27-36.

Stange R.R. Jr., Midland S.L., Eckert J.W. and Sims J.J. (1993a) An antifungal compound produced by grapefruit and Valencia orange after wounding of the peel. Journal of Natural Products, **56(9)**, 1627-1629.

Stange R.R. Jr., Midland S.A., Sims J.J. and Eckert J.W. (1993b) Evidence that wound gum, not lignin, in deposited in infection-resistant injuries of citrus peel. Acta Horticulturae, **343**, 347-352.

Steenburg B., Banks W.H. and Anderson O. (1963) Sorption isotherm of paper. EUCEPA Symposium, 65.

Stott R.A. (1959) Compression and stacking strength of corregated fibreboard containers. Journal of the Australian Pulp Paper Industry, **13**, 84-88.

Studman C.J. (2001) Computers and electronics in postharvest technology - a review. Computers and Electronics in Agriculture, **30**, 109-124.

Tetelman A.S. and McEvily A.J. Jr. (1967) Fracture of Structural Materials. Wiley, New York.

Tetens O. (1930) Uber einzige meteorologische. Begriffe. Z. Geophys., **6**, 297-309. (As referenced in Schaefer (1989)).

Thew R.K. (1986) The profitability of lychee growing in southern Queensland. In ‘The Potential of Lychee in Australia. Proceedings of the First National Lychee Seminar, 14-15 February 1986’ (Eds Menzel C.M. and Greer G.N.) Sunshine Coast Tropical Fruits Association, Nambour Qld, 84-93.

Thornton R.E., Smittle D.A. and Peterson C.L. (1973) Reducing potato damage during harvest. EB 0646, Coop. Extension Service, College of Agriculture, Washington State University, Pullman, WA 99164.

Timm E.J., Schulte Pason N.L., Brown G.K. and Burton C.L. (1989) Apple impact surface effects on bruise size. ASAE Paper, **89-6048**.

Tongdee S.C. (1998) Post-harvest technology of fresh lychee: Commercial perspective from Thailand. South African Lychee Growers Association Yearbook, **9**, 37-43.

Tongdee S.C., Sarpetch C., Roe D.J., Suwanagul A. and Neamprem S. (1998) Effect of heat-acid treatment on quality of lychee fruit. South African Lychee Growers Association Yearbook, **9**, 44-46.

Tongdee S.C., Scott K.J. and McGlasson W.B. (1982) Packaging and cool storage of litchi fruit. CSIRO Food Research Quarterly, **42**, 25-28.

Underhill S.J.R. and Critchley C. (1992) The physiology and anatomy of lychee (*Litchi chinensis* Sonn.) pericarp during fruit development. Journal of Horticultural Science, **67(4)**, 437-444.

Underhill S.J.R. and Critchley C. (1993) Physiological, biochemical and anatomical changes in lychee (*Litchi chinensis* Sonn.) pericarp during storage. Journal of Horticultural Science, **68(3)**, 327-335.

Underhill S.J.R. and Critchley C. (1994) Anthocyanin decolourisation and its role in lychee pericarp browning. Australian Journal of Experimental Agriculture, **34(1)**, 115-122.

Underhill S.J.R. and Critchley C. (1995) Cellular localisation of polyphenol oxidase and peroxidase activity in *Litchi chinensis* Sonn. pericarp. Australian Journal of Plant Physiology, **22(4)**, 627-632.

Underhill S.J.R. and Simons D.H. (1993) Lychee (*Litchi chinensis* Sonn.) pericarp desiccation and the importance of postharvest microcracking. Scientia Horticulturae, **54**, 287-294.

Underhill S.J.R., Coates L.M. and Saks Y. (1997) Litchi. In ‘Postharvest Physiology and Storage of Tropical and Subtropical Fruit’ (Ed Mitra S.K.) CAB International, New York, 191-208.

Underhill S.J.R., Critchley C. and Simons D.H. (1992) Postharvest pericarp browning of lychee (*Litchi chinensis* Sonn.) Acta Horticulturae, **321**, 718-725.

Underhill S.J.R., Johnson G.I. and Highley E. (1994) An overview of lychee postharvest technology. ACIAR Proceedings **58**, 36-40.

Underhill S.J.R., McLauchlan R.L. and Dahler J.M. (1998) Flavedo and albedo changes in 'Eureka' lemons caused by static compression and impact loading. Journal of Texture Studies, **29(4)**, 437-452.

Van Beek G. and Lamers J. (1979) De specifieke vochtagifte van tuinbouwprodukten. Rapport 2072. Sprenger Instituut Wageningen NL (ATO DLO Institute) (Referenced in Scheer (1994)).

Van den Berg L. (1987) Water vapour pressure. In ‘Postharvest Physiology of Vegetables’ (Ed Weichmann J.) Dekker, New York, 203-230.

Van Lancker J. (1979) Bruising of unpeeled apples and potatoes in relation with temperature and elasticity. Lebensmittel Wissenschaft und Technologie, **12**, 157-161.

Vaughn K.C., Lax A.R. and Duke S.O. (1988) Polyphenol oxidase: the chloroplast oxidase with no established function. Physiologia Plantarum, **72**, 659-665.

Vergano P.J., Testin R.F. and Newall W.C. Jr. (1991) Peach bruising: susceptibility to impact, vibration and compression abuse. Transactions of the ASAE, **34(5)**, 2110-2116.

Vincken J-P, Schols H.A., Oomen R.J.F.J., McCann M.C., Ulvskov P., Voragen A.G.J. and Visser R.G.F. (2003) If Homogalacturonan Were a Side Chain of

Rhamnogalacturonan I. Implications for Cell Wall Architecture. Plant Physiology, **132**, 1781-1789.

Vilasachandran T., Sargent S.A., Maul F. and Kader A.A. (1997) Controlled atmosphere storage shows potential for maintaining postharvest quality of fresh lychee fruits. Seventh International Controlled Atmosphere Conference, Proceedings, Davis, California, 13-18 July 1997, **3(17)**, 83-89.

Wade N.L. and Bain J.M. (1980) Physiological and anatomical studies of surface pitting of sweet cherry fruit in relation to bruising, chemical treatments and storage conditions. Journal of Horticultural Science, **55(4)**, 375-384.

Wang S.F., Cheng Z.M., Li Y., Wang Y.Y. and Zhen L.Y. (1996) Effect of postharvest treatments on physiology and quality of litchi and their economics. Acta Horticulturae, **429**, 503-507.

Wenian C., Duprat F. and Roudot A.C. (1991) Evaluation of the importance of the cellular tissue geometry on the strains observed on apples after a compression or an impact. Sciences des Aliments, **11(1)**, 99-110.

Wexler A. (2000) Constant humidity solutions. In ‘CRC Handbook of Chemistry and Physics, 81st Ed.’ (Ed Lide D.R.) CRC Press LLC Boca Raton, Florida USA, 25.

Wills R., McGlasson B., Graham D. and Joyce D. (1998) Postharvest - An Introduction to the Physiology and Handling of Fruit, Vegetables and Ornamentals, 4th Edition. University of NSW Press, Ltd., Sydney.

Wills R.B.H., Ku V.V.V., Shohet D. and Kim G.H. (1999) Importance of low ethylene levels to delay senescence of non-climacteric fruit and vegetables. Australian Journal of Experimental Agriculture, **39**, 221-224.

Wilson L.G., Boyette M.D., and Estes E.A. (1995) Postharvest handling and cooling of fresh fruits, vegetables, and flowers for small farms, Part 3: Handling. North Carolina State University Horticulture Information Leaflets. <www.ces.ncsu.edu/depts/hort/hil/hil-802.html>

Wong L.S., Jacobi K.K. and Giles J.E. (1991) The influence of hot benomyl on the appearance of cool stored lychee (*Litchi chinensis* Sonn.) Scientia Horticulturae, **46**, 245-251.

Woolf A.B., Wexler A., Prusky D., Kobiler E. and Lurie S. (2000) Direct sunlight influences postharvest temperature responses and ripening of five avocado cultivars. Journal of the American Society for Horticultural Science, **125(3)**, 370-376.

Wu Z., Su M., Ji Z., Chen W. and Han D. (2001) A study of the postharvest behaviour of 'Feizixiao' litchi during storage. Proceedings of the First International Symposium on Litchi and Longan, Guangzhou, China, 16-19 June 2000, Acta Horticulturae **558**, 381-386.

Wu Z.X., Su M.X. and Chen W.X. (1997) Research advance on mechanism of litchi browning. In ‘China Agricultural Products Storing and Processing Technical Annals’ China Agricultural University Publishing House, Beijing, 294-302.

York G.M. (1995) An evaluation of two experimental polysaccharide Nature Seal ® coatings in delaying the postharvest browning of the lychee pericarp. Proceedings of the Florida State Horticultural Society, **107**, 350-351.

Yu C.C. (1998) The study of desorption equilibrium moisture content of litchi. Journal of Agriculture and Forestry, **47(2)**, 113-141. (Abstract only).

Yuan Y.C. (1997) The principles and techniques for keeping fresh litchi fruit. South China Fruits, **26(3)**, 30-31.

Zauberman G., Fuchs Y., Rot I. and Weksler A (1988) Chilling injury, peroxidase and cellulase activities in the peel of mango fruit at low temperature. HortScience, **23**, 732-733.

Zauberman G., Ronen R., Akerman M., Weksler A., Rot I. and Fuchs Y. (1991) Postharvest retention of the red colour of litchi fruit pericarp. Scientia Horticulturae, **47**, 89-97.

Zhang D.L. and Quantick P.C. (1997) Effects of chitosan coating on enzymatic browning and decay during postharvest storage of litchi (*Litchi chinensis* Sonn.) fruit. Postharvest Biology and Technology, **12(2)**, 195-202.

Zhang D.L., Chen F., Liu, Li, Jiang, Guo and Qu. (1997) Effects of Pro-long coating on changes in colour and enzymes activity of postharvest litchi fruit. Journal of Tropical and Subtropical Botany, **5(2)**, 54-60.

Zhang Z., Pang X., Ji Z. and Jiang Y. (2001) Role of anthocyanin degradation in litchi pericarp browning. Food Chemistry, **75(2)**, 217-221.

Zhao H.H., Li C.Y. and Guan Z.J. (1999) Experimental research on drying charcteristics of litchi. Drying Technology, **17(9)**, 1915-1925.

Appendix