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**STUDY OF MEALINESS AND OTHER PHYSICAL
PROPERTIES OF APPLE**

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STUDY OF MEALINESS AND OTHER PHYSICAL PROPERTIES OF APPLE

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ABSTRACT

Quality is an increasingly important factor in the production and marketing of fresh fruit and vegetables. After harvesting, fruit and vegetables are placed in storage for a few days or a few months, serving as a means to extend the season and provide a reserve for more uniform retail distribution. Changes in texture, particularly the development of mealiness, will greatly affect fruit eating quality, and thus its acceptability to consumers. Mealiness occurs extensively in both pome and stone fruits. A special concern in this study was the problem of mealiness measurement of "Braeburn" apples. This cultivar was New Zealand's most popular export apple variety in the June 1996 year, valued at \$102.2 million.

The objective of this study was to investigate the development of apple mealiness and to develop an apple mealiness indicator by (1) providing a detailed review of literature on fruit mealiness, (2) studying the changes in physical properties with the development of apple mealiness, (3) comparing the physical properties of mealy with non-mealy and aged apples, and (4) investigating the relationships between objective tests and subjective tests to develop good mealiness indicators.

A review of the literature showed that mealiness was not just the loss of water, but was related to the way water is bound or tied up chemically within the fruit, which can make it difficult to extract. Many studies have been done on the objective measurement of fruit mealiness, but until now there are no good indicators based on fruit physical properties which have been adopted by the fruit industry. Some parameters may be effective for measuring fruit mealiness, such as internal air space, extractable juice, but further research work needs to be done to correlate these parameters with subjective tests, to investigate the correlation with other physical parameters and to monitor changes in values with the development of fruit mealiness.

Experimental studies on the effect of storage conditions showed that high temperature hastened apple ripening and mealiness processes. Texture deterioration was a main quality concern in apples stored under high humidity conditions (>90% at 20°C and >

95% at 0°C), but shrivel was also a major quality factor in fruit stored at low humidity conditions (<90% at 20°C and <95% at 0°C).

By monitoring the physical property changes during the development of apple mealiness, the following results were obtained:

Mealy apple density was significantly lower than that of fresh apple. This decrease was mainly affected by the development of apple mealiness. However due to the wide variation in density change, it was not a reliable mealiness predictor.

In the initial stage of the experiment twist strength declined with storage time. Thereafter changes were related to humidity conditions. The value of mealy apples was significantly lower than that of aged, shrivelled but not mealy apples. Multiple regression indicated that twist strength change was not only affected by mealiness development, but also by different treatments and apple age, so it was not a suitable mealiness indicator.

Elastic modulus changes did not respond well to the development of apple mealiness. When apples shrivelled, elastic modulus declined greatly. Multiple regression results indicated that both apple mealiness development, and apple age affected the change of elastic modulus. Low R^2 values indicated that elastic modulus were weakly related to the treatments, apple age and mealiness development. It could not be used as an apple mealiness indicator.

Both compressive energy and fracture strength declined with storage time. Multiple regression results indicated that both changes of compressive energy and fracture strength were not only affected by mealiness development, but were also equally affected by different treatments and apple age, so they were not suitable mealiness indicators.

Based on literature studies, shear strength is mainly determined by the strength of the cell wall. Shear gradient declined with storage time and the development of apple mealiness. Multiple regression results indicated that changes of shear gradient were only

affected by mealiness development, and so it may be a suitable mealiness indicator. However, the regression coefficient was relatively low (0.43), and further research work would need to be done to attempt to improve its reliability.

Keywords: Fruit texture, texture analyser, fruit mealiness, density, twist strength, elastic modulus, fracture strength, compressive energy, shear gradient.

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CHAPTER ONE

GENERAL INTRODUCTION

1.1 STATEMENT OF THE PROBLEM

The market of fruit and vegetables, especially in many industrial countries, is constantly changing and evolving. With this market development, quality becomes an increasingly important factor in production and marketing. Suppliers must meet the elevated quality demand of the consumers to retain or increase their market share.

According to Kader (1992), consumers consider good quality fruits to be those that look good, are firm, and offer good flavour and nutritive value. Although consumers buy on the basis of appearance and feel, their satisfaction and repeat purchases are dependent upon edible quality. As described by Rayll and Pentzer (1974), fruit quality is a combination of characteristics, attributes, and properties that give the commodities value for food. It may be described in terms of general attributes such as appearance, flavour and texture. These attributes affect product attractiveness to the consumer and therefore are main quality attributes in the fruit industry.

Fruit texture, as an important quality component, is related to those attributes of quality associated with sensation of feeling, as experienced by fingers, hands, or the mouth. Included in texture are such sensations as firmness, crispness, mealiness, juiciness and toughness (Studman, 1994). Flavour may also be affected by texture because release of taste components in the mouth is related to tissue structure (Rayll and Pentzer, 1974). The texture and softening of harvested fruit are of considerable physiological importance and of horticultural concern. From a horticultural standpoint, texture serves as an important determinant of quality in harvested fruit and thus influences the methods by which commodities are harvested and handled in market channels (Huber, 1983). An extensive review has recently been completed by Harker *et al.* (1997).

Firmness measurements in fruit and vegetables have been used for many years as a guide to the quality of the product (Abbott *et al.*, 1976; Bourne, 1979). This method has not proven entirely satisfactory since it damages the specimen. A rapid objective method which measures texture without damaging the fruit would be desirable because it permits testing of large samples or perhaps all of the fruit and could be incorporated in the sorting apparatus of the packing or processing plant (Abbott *et al.*, 1982). These non-destructive techniques would also greatly enhance management decisions during picking, grading and storage operations (Armstrong *et al.*, 1990). According to Chen and Sun (1991), a number of these methods have been developed by different researchers over the past decades. These methods are based on the detection of various physical properties which correlated well with certain quality factors of the product. The sophistication of non-destructive methods has evolved rapidly with modern technologies. The combination of new imaging acquisition and high-speed image processing techniques has provided new tools for researchers to develop many new and improved techniques for non-destructive quality evaluation of agricultural products. Although the non-destructive tests are very important for the agricultural produce industries, destructive quality tests are still useful tools. Compared to non-destructive tests, destructive tests are more straightforward to interpret (Studman and Boyd, 1994).

New Zealand is a small country, whose economy has depended upon the export of biological products for the past 150 years (Studman, 1994). Pipfruit, including apples and pears, is one of its most successful horticultural produces. The pipfruit exports during the early 1990s were relatively stable at around 11 million cartons (200,000 tonnes). This increased to 17 million 18-kg cartons in 1995, and almost 18 million in 1996 (New Zealand Official Yearbook 1997). Fresh apples were the second largest of all fruit and nuts exported in the June 1996 year, valued at \$338.6 million (New Zealand Official Yearbook 1997). The continued success of the New Zealand apple industry has been partially attributed to its high quality.

The reputation of apple exports from New Zealand has been built upon a high dessert apple quality with crisp and juicy characteristics. However during cool storage for transportation overseas or for year-round marketing, the texture of apples can

deteriorate, resulting in soft, dry, mealy fruit. These changes in texture will greatly affect fruit eating quality, and thus its acceptability to consumers. According to Rayll and Pentzer (1974), a mealy apple is a more serious defect resulting in less desirability than an apple which is below standard in colour. As discussed in detail in Chapter two, mealiness not only affects apples, but is also a serious economic problem in stone fruits (Kailasapathy and Melton, 1992), including peaches (Von Mollendorff and De Villiers, 1988a), and nectarines (Dawson, Watkins and Melton, 1995).

The problem of apple mealiness is by no means a new problem in New Zealand and other apple producing areas in the world. Since at least 1943, researchers have investigated the causes of mealiness and poor quality in Delicious apples (e.g. Fisher 1943), but the causes are still poorly understood. In particular, despite over 50 years research into its courses, no mealiness indicator tests based on physical properties has been developed.

1.2 OBJECTIVES OF THE STUDY

The main goal of this study was to develop an apple mealiness indicator which can help the apple industry to monitor the development of fruit mealiness and to detect mealy apples before they are put on the shelf, thereby improving marketing apple quality and consumer acceptability. The specific objectives of this study were:

- (1) To provide a detailed review of literature on fruit mealiness.
- (2) To study the changes in physical properties with the development of apple mealiness.
- (3) To compare the physical properties of mealy apples with non-mealy and aged apples.
- (4) To investigate the relationships between apple physical properties and sensory tests to develop good mealiness indicators.

The approach taken was to store apples under different conditions, and to monitor the physical properties and mealiness changes during the storage period. The "Braeburn" cultivar was chosen because of its importance to the New Zealand export industry. According to New Zealand Official Yearbook 1997, Braeburn apple was New Zealand's most popular export apple variety in the June 1996 year, valued at \$102.2 million.

CHAPTER TWO

LITERATURE REVIEW

2.1 INTRODUCTION

Fruit ripening involves a complex series of changes including cell wall degradation, alteration of membrane condition and function, changes in compartmentation of solutes, and metabolic changes associated with climacteric changes (Brady, 1987). For most of the fruits, these changes result in the softening of mesocarp tissue and development of juicy texture. But sometimes the fruit fails to degrade according to the usual pattern due to chilling injury or other reasons, and although the mesocarp softens, the texture of the tissue becomes mealy (Ben-Arie and Lavee, 1971; Harker and Hallett, 1992; and Dawson *et al.*, 1992).

2.2 DESCRIPTION OF MEALINESS

Mealy fruit are dry and flavourless. Mealiness in apples means the breakdown of flesh into small pieces in the mouth, which feel dry to the palate (Harker and Hallett, 1992). Fruit mealiness is a physiological storage disorder. For summer fruit, it has been generally regarded as a low-temperature disorder sometimes referred to as chilling injury (Bramlage, 1982) which has reduced postharvest shelf life. It is characterised by abnormal ripening after cold storage and inedible, flavourless fruit. Senescent breakdown, as one kind of physiological disorder, may cause the whole apple to become dry and mealy which may occur in any apple cultivar (Meheriuk *et al.*, 1994). It is not the loss of water, but just the way the water is bound or tied up chemically within the fruit which makes it difficult to extract juice. It is the lack of ability to get juice out of the fruit which gives it the sensation of dryness on the palate, not the sense of dehydration (Rowe, 1986).

Fruit mealiness, as a kind of texture deterioration, is affected by cellular anatomy, the water relations of cells, and the composition of cell walls. Fruit cells retain their normal osmotic properties during ripening (Simon, 1977) and there is little loss of turgor pressure, although this may be affected by dehydration if fruit is stored at a low humidity. The cells of fruit are frequently large parenchymatous cells with large air spaces between them. Changes in the degree of contact between cells as well as in the structure of the cell walls themselves will affect texture (Rhodes, 1980; Hatfield and Knee, 1988).

2.3 FRUIT CELL WALLS AND MIDDLE LAMELLA

The cell wall lies outside the plasma membrane, which defines the boundaries of the cell itself. Each cell is connected to adjacent cells by a pectin-rich middle lamella. Cells present in fruit pulp are generally thought to contain only primary cell walls (John and Dey, 1986).

The primary wall consists of cellulose microfibrils embedded in a matrix of pectic and hemicellulose polysaccharides and hydroxyproline-rich glycoprotein (Northcote, 1972). It is thought that these polymers are held together in the three-dimensional cell wall by a variety of covalent and non-covalent bonds (Fry, 1986). Cellulose fibrils are held together by hydrogen bonds, and similar bonds account for the interaction of cellulose with hemicelluloses (Tucker, 1993). Apart from the vascular tissue and specialized cells, for example the stone cells in pears, the cell wall of the fleshy parts of most fruits are unlined. They also contain a low proportion of hydroxyproline rich protein and small amounts of xylose and mannose residues which are characteristic of hemicellulose polymers; on the other hand, they contain a high proportion of galacturonic acid, galactose and arabinose residues which are typical of pectic polymers (Knee and Bartley, 1981).

The individual cells are cemented together by an amorphous layer external to the primary wall and called the middle lamella, or sometimes interlamellar layer. It consists principally of the calcium salts of polymers of galacturonic acid that have been partially esterified with methyl alcohol, and is known as pectic material (Bourne, 1976). The middle lamella area between primary cell walls of adjoining cells forms a continuous

intercellular matrix. This layer is particularly rich in pectic polysaccharides and is believed to be the region of the wall most affected during fruit softening (Huber, 1983).

2.4 TEXTURE AND CELL WALLS

The texture of fruit is highly dependent on the chemical and physical properties of the cell wall (Van-Buren, 1979; Jackman and Stanley, 1995). When a fruit is eaten, its texture is determined, primarily by the manner in which the cell-wall skeleton deforms and ruptures (Waldron *et al.*, 1997). How a plant tissue deforms during mastication depends on (1) the forces of oral mastication and (2) the structural characteristics of the fruit. Fruit mechanical properties will depend on contributions from the different levels of structure, and how they interact with one another. These levels are shown in **Figure 2-1**.

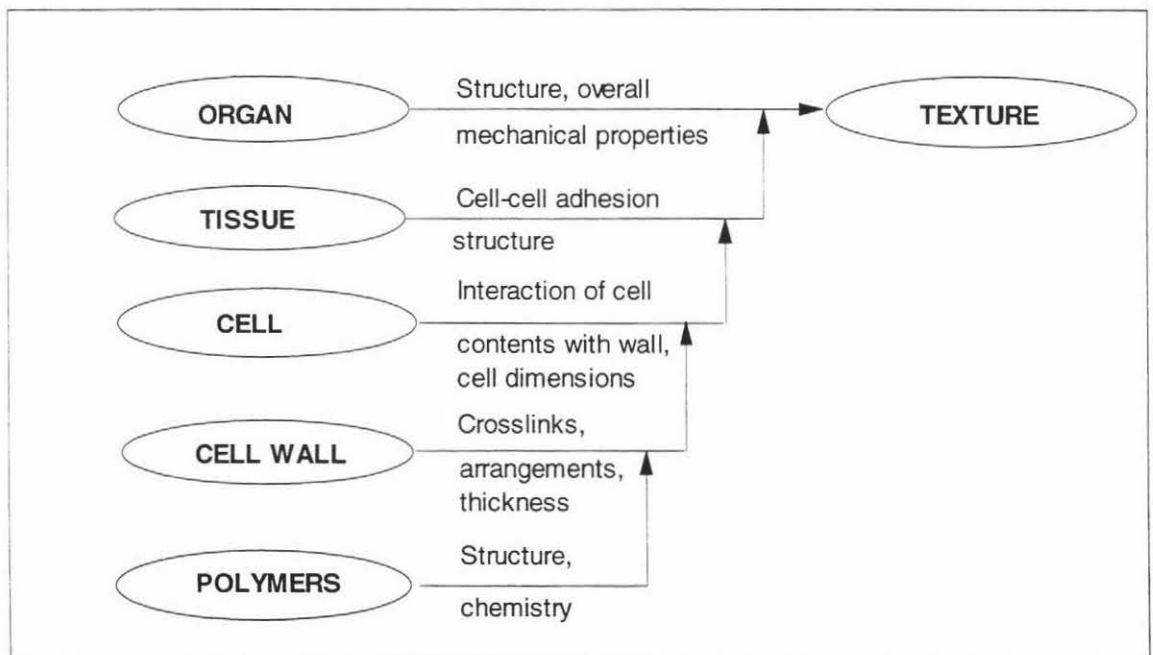


Figure 2-1 Schematic representation of the levels of structure that contribute to the mechanical properties of plant tissues (Source : Waldron *et al.*, 1997).

The mode of fruit deformation and rupture classically involves cell separation or cell breakage. If the forces holding the cells together are stronger than the cell walls, failure

occurs in the cell walls; if the forces holding the cells together are weaker than the cell walls, the cells will separate.

As described by Harker and Hallett (1992), the ease with which cells in fruit tissue burst is probably related to the strength and elasticity of the cell wall and plasma membrane as well as to the strength of adhesion between neighbouring cells. Adhesion might be expected to influence the way fruit tissue breaks when chewed by determining whether 1) the fruit tissue breaks into small fragments containing undamaged cells or 2) individual cells rupture when crushed, leaving a pulp of connecting cell wall material.

2.5 FRUIT MEALINESS AND CELL DAMAGE

Described by Waldron *et al.* (1997), in unripe fruit, cell adhesion is strong and tissue fracture involves rupture across the cell walls, breaking open the cells and releasing the cell contents, so the fruit are juicy. With the development of fruit ripening, the fruit tissue becomes considerably soft. This softening is usually consequent to the dissolution of wall polymers, many of which are involved in cell adhesion. In highly softened tissues such as over-ripe, mealy apples, the cells become completely separated. The textural properties of such tissues no longer reflect those of unified structure, but approach those of a soft solid comprising many single cells. It does not follow that a weak structure will fail only by cell separation. In principle, breakage or separation may occur at low or high strengths, depending on which mode is the low-stress route. The failure of a soft juicy fruit will still involve a considerable degree of cell rupture. However, cell separation is an important element in tissue softening.

Harker and Maindonald (1994) used electrical impedance measurement to study the changes in the cell wall, vacuole, and membranes of nectarines, and found that the quality of cell contents (juice) was not different in woolly (or mealy) and non-woolly fruit. The structural integrity and area of membranes were also expected to be the same for woolly and non-woolly nectarines since membrane capacitances were similar. The main difference between woolly and non-woolly nectarines was in cell wall resistance, which was higher in woolly than in non-woolly tissues.

Ben-Arie *et al.* (1989) indicated that with the development of fruit ripening, there are two kinds of cell separation occurring in fruits. The first or the more obvious case is the gradual separation, which develops with the onset of ripening, between the peel and the flesh of certain fruits. In stone fruit, for example, this appears to be a separation between adjacent tissues by dissolution of part of the intervening tissue or cell wall, such as occurred in abscission. Although the two tissues differ structurally, they are cemented to each other in the immature fruit, but at a certain stage of maturity they become easily separable. The final cleavage between them might conceivably be the result of cell wall degradation or cell disintegration. The second instance of total cell separation occurring in fruits has been suggested in relation to the development of mealiness in over-ripe pome fruit. Mealiness, i.e., the apparent loss of juice in the fruit flesh, may occur also as a result of chilling injury, e.g. woolliness in peaches. As there is no excessive loss of water, which could explain the loss in juiciness in either case, it has been suggested that mealiness in pears results from both increased dissolution of pectin in the middle lamella and increased rigidity in the cell wall due to restricted cellulose and hemicellulose degradation. Therefore, when the fruit is exposed to any kind of mechanical stress, the cells separate and slip easily past each other, instead of breaking and releasing their juice contents.

Examination of fracture surfaces by Harker and Sutherland (1993) indicated that tissue failure in unripened nectarines at harvest occurred when cells were ruptured. Once the fruit had ripened the fracture surface was obscured by a coating of juice. They observed the convex shapes of cell surface protruding through the surface juice, and this suggested that neighbouring cells had separated from each other without rupturing. Tissue plugs from mealy fruit broke apart when neighbouring cells separated leaving undamaged cells exposed at the fracture surface. Little juice was found on the fracture surface.

Harker and Hallett (1992) found that juiciness in apples seemed to be related to rupture and release of cell contents, and apples became mealy when reductions in cell adhesion and increased cell rigidity reduced the propensity for cells to become broken during

chewing. Previous studies (Ben-Arie *et al.* 1989; King *et al.*, 1989) have led to speculation that mealiness in stone fruit may be associated with reductions in the adhesion between neighbouring cells, as is the situation with mealiness in apples (Harker and Hallett, 1992).

Tensile measurements undertaken by Harker and Sutherland (1993) established that cell adhesion decreased when stone fruit were allowed to ripen. In contrast, they found that once the fruit has ripened there was no significant difference in the tensile strength between mealy and non-mealy fruit. They suggested that cell adhesion did not account for the difference in the texture of non-mealy and mealy nectarines. Visual examination of the fracture surfaces tended to confirm this, as cell separation in both mealy and non-mealy fruits occurred without causing cell damage. However a more obvious difference between the fracture surfaces was the presence of juice in non-mealy tissue and absence of juice in mealy tissue. They gave two possible explanations for the presence of juice on the fracture surface of normally ripened non-mealy mesocarp tissue in which the cells themselves remain intact: (A) the juice naturally occurs in the apoplast of ripened nectarine tissue, or (B) intracellular solution is somehow exuded from the cells during the application of tensile force without causing any visible indication of cell damage. No matter which mechanism is involved, it is clear that in mealy tissues the natural or induced exudation of juice into extracellular spaces must be inhibited, perhaps through blockage of pathways across the plasma membrane or cell wall. So while both mealy apples and mealy nectarines had a similar dry unpalatable texture, in apple the dryness was associated with the failure of cells to burst, whilst in nectarines, another mechanism such as gel formation which reduced the quantity of liberated juice was required. Evidence for this is discussed below.

2.6 APPLE MEALINESS AND ITS INTERNAL AIR SPACE

Wilkinson (1965) found that an increase in the air space in fruit is expected to lead to easier cell separation and a change in texture. Further research was conducted by Hatfield and Knee (1988). They reported that the loss of cell cohesion derived from the increase in air space in fruit. This increase implied a decrease in the average area of

contact between cells. Cell contact area should have been maintained in the initial weight loss fruit because the air space did not increase. According to the sensory panel the initial weight loss apples were firmer, tougher and less mealy than controls. The absence of discrimination in terms of juiciness may indicate that, while the cohesion of tissue was affected by weight loss, the ease of cell breakage (to liberate juice) was not. They suggested that the cause of these large air spaces and mealy texture was probably related to the degradation of the middle lamella and corresponding reduction in cell adhesion. The cells were then able to expand slightly and become more rounded, with the result that air spaces increased and cell contact areas decreased. This change in cell shape and cell expansion seemed to be a turgor-driven process. They concluded that, during cool storage, cells from high-maturity fruit tended to lose cell-to-cell adhesion but maintained cell wall strength, so that individual cells were difficult to rupture. In contrast, cells from low-maturity fruit tended to maintain relatively high cell-to-cell adhesion, but the strength of the cell wall declined so that the cells were easily ruptured. This suggested that during chewing, tissue from the high-maturity apples may be perceived as mealy because it breaks down into small clumps of undamaged cells. In comparison, cells from non-mealy apples, which are relatively strongly attached to each other and are relatively easy to rupture, would be crushed and release their contents during chewing.

Fisher (1943) and Harker and Hallett (1992) confirmed that high-maturity apples become mealy during cool storage, while low-maturity apples did not. Further investigation was conducted by Harker and Hallett (1992). They studied physiological changes associated with the development of apple fruit mealiness and found that the volume of air space was greater in apples harvested at high maturity than at low maturity, and although air space expanded during cool storage this relative difference was maintained. The cellular characteristics particularly associated with mealy tissue were high volume of air space and lack of juice on the surface of tissue fractured during tensile tests (Harker and Sutherland, 1993). High volumes of air space are thought to be symptomatic of mealiness in apples (Harker and Hallett, 1992), and have been observed during other studies on stone fruit (Luza *et al.*, 1992).

Vincent (1989) found that apple flesh contained air space or channels which were orientated radially from the centre and did not communicate very much laterally, though they probably connected radially. He suggested that in a mealy apple the cells and the space between them had increased so much that the radial channels were easily distinguished. This would imply that mealy apples should display an increase in mechanical anisotropy.

2.7 PHYSIOLOGY AND BIOCHEMISTRY OF FRUIT MEALINESS

Fruit mealiness is the result of physiological and biochemical changes during storage. For summer fruit it can be a chilling injury disorder. In early research on 'Elberta' peaches, Pentzer and Heinze (1954) suggested that the occurrence of chilling injury in fruits was the result of a deviation in the equilibrium between two types of reaction taking place in the cells, namely the accumulation and the breakdown of substances toxic to cell structure. At the critical temperature specific to each cultivar, the rate of both reactions is equal, but below this temperature the accumulation of toxic substances was more rapid than their breakdown, causing injury to the tissue. Based on this theory, Ben-Arie *et al.* (1970) described the occurrence of mealiness (woolliness) in a two stage process. When fruits were in cold storage, certain changes occurred in their tissue but the cumulative harmful effects could be reversed by exposing them to temperatures above critical for normal ripening. In the second stage, which was the result of the accumulated effects of the first stage, there appeared to be an altered metabolism of pectic substances and the occurrence of mealy (woolly) breakdown. At this stage the process is irreversible and a transfer of the fruit to warmer temperatures only hastened the intensity of mealiness.

More recent studies showed that during normal ripening of stone fruit, there is an increase in the concentration of soluble pectic polysaccharides (Bartley and Knee, 1982; Huber, 1983) caused by the degradation of the cell wall polysaccharides that cement the cells together. The polygalacturonides (major cell wall polysaccharides) are repeating polymers with a backbone of alpha-D-(1-4) linked galactopyranosiduronic acid residues, interrupted by L-rhamnose units (Albersheim, 1976; Aspinall, 1980). The increase in the

concentration of soluble uronic acid residues is often correlated with an increase in the polyuronide hydrolysing enzymes, especially endopolygalacturonase and poly (1-4- α -D-galacturonide) glycanohydrolase, (EC.3.2.1.15). The activity of polygalacturonases (PG) was not detected in unripe peaches (Pressey, Hinton and Avants, 1971). The extent of pectin solubilization by PG also depends on whether it is an endo-PG or an exo-PG, the endo-PG solubilizing more pectic material (Pressey and Avants, 1978).

The formation of woolliness or dry texture has been associated with impaired solubilization of pectic substances (Ben-Arie and Lavee, 1971; Ben-Arie and Sonogo, 1980) due to improper functioning of PG. The initial low PG-activity in peaches (which eventually become woolly), followed by a sudden and marked increase during the ripening process, resulted in an accumulation of pectic substances with a high relative molecular masses in the inter-cellular spaces, and this was probably the primary cause of woolliness in peaches (Von Mollendorff and De Villiers, 1988b). Ben-Arie and Lavee (1971) indicated that pectinesterase (PE) demethylated pectins during low temperature storage which resulted in an insoluble low methoxyl pectin of high molecular weight that held water in a gel. Thus, juiciness was reduced and the symptom of woolliness was expressed. This would account for why the juice is bound in mealy nectarines, as discussed above (Harker and Sutherland, 1993). Buescher and Furmanski (1978) also found that woolliness was associated with reduced levels of soluble pectins and enhanced levels of insoluble pectins in ripened fruits. Reduced PE and PG activities appeared to account for reduced depolymerization and reduced solubilization of pectic substances. Ben-Arie and Lavee (1971) have indicated that insoluble low methoxyl pectins were high in woolly fruit due to continuous demethylation by PE. This suggested that PG was responsible for releasing cell wall bound enzymes. Dawson *et al.* (1992) Analysed pectic and hemicellulosic polysaccharides changes of nectarines, they found that fruit mealiness predominantly affected the pectic component of the cell wall. Solubilization of uronic acid-rich polymers in mealy fruit was altered relative to that in normally ripened fruit. Solubilized polymers were of high molecular weight and were not depolymerized to lower molecular weight species. During cool storage, pectins in the cell wall were de-esterified, suggesting continued activity of PE at lower temperature.

Branched pectins accumulated in the cell wall material and galactan side chains, which were removed from the pectic backbone during the initial stages of normal ripening, remained attached to the backbone in mealy fruit. Ben-Arie and Sonego (1980) also suggested that the development of woolly breakdown in cold-stored peaches resulted from an imbalance in pectolytic activity, whereby low temperatures induced pectinesterase to cause the accumulation of de-esterified pectate (soluble in EDTA) and inhibited polygalacturonase from degrading this substrate.

2.7.1 Evidence for Gel Formation in Mealy Stone Fruit.

Von Mollendorff and De Villiers (1988a) found during the process of peach mealiness, free moisture (juice) was converted into bound moisture without any change in the total moisture content. They also found that the respiration rate of woolly fruit was lower than that of normal fruit. This might be due to gel formation (Ben-Arie and Sonego, 1980), which prevented gas exchange between cells, or the metabolic activity of the tissue of woolly peaches might be lower than that of sound fruit. The viscosity of soluble pectins attained a maximum value during the development of woolliness. It was directly proportional to the concentration, relative molecular mass, degree of esterification of the pectins, presence of electrolytes and the PH of the medium (Von Mollendorff and De Villiers, 1988b). Ben-Arie and Lavee (1971) had also reported that PE demethylated pectins during low temperature storage which resulted in an insoluble low methoxyl pectin of higher molecular weight that held water in a gel. In the case of chill-injured peaches it has been reported that there was very low activity of PG but there was continued activity of PE (Ben-Arie and Lavee, 1971). The prolonged activity caused the accumulation of insoluble high molecular weight low methoxyl pectins that held water in gel. Hence juiciness was reduced and overall woolly texture was expressed in the fruit tissue. Thus in cold storage peaches, the development of woolly breakdown was accompanied by the continuous activity of PE but a simultaneous inhibition of PG activity. With intermittent warming of the fruit, which delayed the development of woolliness, the activity of PG increased to levels comparable to that of a normally ripened fruit (Ben-Arie and Sonego, 1980). However cold storing the fruits at 0°C might reduce the activity of PE substantially and hence prevented the formation of a gel

due to the binding of water (usually released during fruit ripening) by the low methoxyl pectin of high molecular weight. Ben-Arie and Lavee (1971) showed that the total amount of pectic substances decreased in healthy peach fruit due to degradation of the soluble pectin in addition to a continued decrease in protopectin. In mealy fruit, levels began to rise as a result of an increased quantity of the water-insoluble fractions (protopectin and pectates) in spite of a continued decrease in water-soluble pectin. These findings also indicated that the loss of juiciness in fruit might be due to the formation of a gel-like structure in the fruit tissue (Kailasapathy and Melton, 1992).

In studies on cell wall changes in ripening peaches, Ben-Arie *et al.* (1989) indicated that instead of cell separation, as has been shown in mealy apples (Ben-Arie *et al.*, 1979), the intercellular spaces in the fruit pulp became filled with a fairly dense matrix and the middle lamella appeared to widen and become less dense. They indicated that some of the insoluble pectin was solubilized but the main change occurred in the calcium pectate fraction, which almost doubled in quantity during 3 days at 20°C after 1 month at 0°C. They also found that the distribution of calcium showed a concomitant increase in the calcium pectate fraction. This increase appeared to be due to a reversed migration of calcium from both the soluble and insoluble pectic fractions to the middle lamella. The composition of the neutral sugars in the calcium pectate fraction supported the hypothesis that insoluble pectin from the primary cell wall had probably been de-esterified and then precipitated with available calcium as calcium pectate, causing an expansion of the middle lamella region and a filling of intercellular spaces with a calcium pectate gel. The appearance of rhamnose and the almost three-fold increase in the percentage of galactose in the calcium pectate fraction were supporting indications of this process. Dawson *et al.* (1993) studied the calcium uptake and efflux, ion leakage, internal air space and cation exchange capacity in relation to mealiness in nectarine tissue. They measured calcium uptake from 0.5M mannitol, 1.0 mM CaSO₄ with 0.5 mM mercaptobenzothiazole (MBT) and containing ⁴⁵Ca (specific activity 0.4 µCi µmol⁻¹ Ca) at 25°C by nectarine tissue. They found that calcium uptake by discs from fruit stored at 0°C for 6 weeks without ripening was similar to uptake from fruit at harvest. However, tissue from fruit that had been ripened, either immediately after harvest or after 6 weeks of cool storage, showed a marked decrease in the rate and extend of

calcium uptake. This was especially evident in ripe fruit where calcium uptake into discs was 20% of that in discs from fruit at harvest after 6h. They concluded that in mealy fruit, calcium uptake was greater than for fruit that ripened normally and more like that of fruit at harvest. The cell walls of mealy fruit had a similar degree of esterification to those of ripe fruit. They suggested that if pectic solubilisation was impaired in mealy fruit, there would be an overall increase in sites available for binding calcium, hence the greater calcium uptake in mealy versus ripe fruit. The internal air space of the tissue decreased during normal ripening but increased in mealy fruit.

Research on cell wall changes in ripening nectarines (Lurie *et al.* 1993) found that the loss of arabinose from the Na_2CO_3 fractions during air storage might play a role in the lack of solubilization of this fraction during ripening. On the other hand, the CDTA and water soluble fractions were smaller in molecular weight than from normally ripened fruit. One or both of these results might contribute to the development of mealiness.

2.8 PHYSIOLOGICAL CHANGES

Fruit mealiness, which is caused by physiological disorder, is related to the fruit ripening and senescence processes. Von Mollendorff and De Villiers (1988a) studied the physiological changes associated with the development of woolliness in 'Peregrine' peaches. They measured the respiration rate and ethylene production and found that the respiration rates of all peaches decreased gradually during storage. During the ripening stage significant differences were evident between the respiration rates of mealy and non-mealy peaches. At the end of the ripening period the respiration rate of fruit which developed woolliness was at least $20\mu\text{l O}_2 \text{ g}^{-1}\text{h}^{-1}$ lower than that of normal fruit. Peaches produced very little ethylene but the production rate of non-mealy peaches was significantly higher than that of mealy peaches. The normal ripening fruit appeared to go through a respiration 'climacteric' which coincided with the increase in ethylene production. However, the fruit which developed the woolliness disorder did not show a typical climacteric rise immediately following transfer to high temperature, although it did show a marked increase in ethylene production.

2.9 FACTORS AFFECTING MEALINESS DEVELOPMENT

Fruit mealiness, as a storage disorder, is mainly affected by postharvest factors, such as storage temperature, humidity, and controlled atmosphere (CA) condition. It is also affected by other factors such as maturity of the fruit, and field temperature during harvesting etc. A summary of these factors is presented in Table 2-1.

2.10 MEALINESS MEASUREMENTS

In order to maintain high quality of fruit, mealiness should be assessed before putting fruit on the market-shelf. Visual assessments is not suitable to detect this disorder, because for most of the fruit the appearance of the mealy one is the same with the normal one. Various methods has been investigated to measure the development of fruit mealiness.

2.10.1 Texture Assessment

There are different methods used to assess fruit mealiness. Early researchers assessed fruit mealiness by observations for apples (Fisher, 1943) and peaches (Fisher *et al.*, 1943; Weaver and Jackson, 1966; Ben-Arie *et al.*, 1970). With the development of fruit mealiness research, the amount of extractable juice was used to express fruit mealiness for peaches (Ben-Arie and Lavee, 1971) and nectarines (Dawson *et al.*, 1993; Dawson *et al.*, 1995). Ben-Arie and Sonego (1980) determined the peach fruit mealiness (woolly breakdown) visually in the halved fruit and graded as slight (<25% of surface), moderate (25-50%) or severe (>50%). The woolly breakdown index was calculated as follows:

$$\text{W.B. index} = ((\% \text{ fruit with slight breakdown} \times 1) + (\% \text{ of fruit with moderate breakdown} \times 2) + (\% \text{ of fruit with severe breakdown} \times 4)).$$

Nectarine was cut halves and squeezed, if the juice appeared thick and did not flow freely, the fruit was considered mealy (Von Mollendorff *et al.*, 1992; Lurie *et al.*, 1993, 1994). Due to the relation of mealiness with chilling injury for stone fruit, storage time below certain temperature (2°C) was used to indicate the occurrence of fruit mealiness for nectarines (Anderson and Penney, 1975; and Dawson *et al.*, 1992; 1993; Harker and Dunlop,

Table 2-1. Summary of factors affecting fruit mealiness

Fruit type	Factors which induced mealiness	Factors which reduced mealiness	Reference
Delicious apple	High maturity induced mealiness.	Low storage temperature delayed or reduced the development of apple mealiness.	Fisher, 1943
Peaches or nectarines	Stored at 7.8°C mild chilling symptoms appeared.	stored at 10°C no chilling injury symptoms appeared.	Mitchell <i>et al.</i> , 1974.
Peaches or nectarines	Stored at 2.2-5°C resulted in a rapid development of mealiness, often within one to two weeks.	Stored at 0°C delayed chilling injury symptom or made it less severe.	O'Reilly, 1947. Mitchell <i>et al.</i> , 1974. Anderson, 1979.
Peaches		Delayed storage for 2-5 days at 24°C before cold storage prevented chilling injury.	O'Reilly, 1947.
Peaches		Delayed storage for 2-3 days at 26°C extended storage life by 10-15 days.	Lill <i>et al.</i> , 1989.
Peaches	As weight loss increased chilling symptom (mealiness) increased.	holding at 20°C for 2 days prior to seven weeks at -0.5°C significantly reduced chilling injury symptom.	Scott <i>et al.</i> , 1969.
Peaches		Delayed storage for 2 days at 23°C decreased the proportion of 'Peregrine' peaches becoming mealy markedly.	Von Mollendorff and De Villiers, 1988a.
Nectarines		Stored nectarines at 0°C with intermittent warming at 20°C at 2-week intervals alleviated the development of mealiness.	Dawson <i>et al.</i> , 1995.
Apples		Initial weight loss apples were firmer, tougher, and less mealy	Hatfield and Knee, 1988.
Peaches and nectarines	High humidity (95-99%) had a higher incidence of mealiness.	Carbon dioxide concentration of at least 5% in CA storage was necessary to delay the appearance of chilling symptoms.	Lill <i>et al.</i> , 1989.
Nectarines		5 % carbon dioxide could store fruit for 6 weeks without appearance of chilling symptoms.	Olsen and Schomer, 1975.
'J H Hale' Peach		20% carbon dioxide without oxygen reduction completely controlled chilling symptoms for 6 weeks.	Wade, 1981.
'Fay Elberta' Peach	5% carbon dioxide increased chilling injury.		Kader <i>et al.</i> , 1982.
Stone fruit	low maturity were more susceptible to mealiness disorder. Higher field temperature during harvesting increased the incidence of fruit mealiness.		Kailasapathy and Melton, 1992.

1994), peaches (Ben-Arie, *et al.* 1989; and Luza *et al.*, 1992). Internal air space was used as the mealiness indicator for Granny Smith apples (Tu *et al.*, 1996).

Taste assessment, as a kind of sensory test, was used by many researchers to assess fruit mealiness. Buescher and Furmanski (1978) established a sensory panel with six members to evaluate the texture of peaches. Samples were rated on a scale from 1 (poor) to 10 (excellent). Lill and van der Mespel (1988) used 10 untrained judges as taste panel. They asked the panel to rate test sample of nectarines on a 5-point hedonic scale for mealiness. Hatfield and Knee (1988) used a laboratory panel of 15 people for paired comparison of apple mealiness tasting of control apples and apples after initial weight loss. Harker and Hallett (1992) assessed apple mealiness by an informal tasting of one assessor. The same method was used to assess mealiness of nectarines (Harker and Sutherland, 1993).

Mealiness assessment is the base of objective tests. Many objective tests have been investigated.

2.10.2 Extractable Juice

Fruit juice release is directly related to the sensation of mealiness. Lill and van der Mespel (1988) found that there was a highly significant relationship between apparent juice content and taste-panel response. Ben-Arie and Lavee (1971) measured the loss of juiciness in Elberta peaches suffering from mealy (woolly) breakdown, and found that the loss of juiciness accompanying woolly breakdown of stored peaches was unconnected with the loss of water vapour from fruit, but was correlated with the amount of expressible juice. Buescher and Furmanski (1978) reported that juiciness was markedly reduced in peaches when it became mealy. The same result was found for nectarines by King *et al.*(1989), and Dawson and co-workers (1992, 1993). Von Mollendorff and De Villiers (1988a) found that regardless of storage temperature the extractable juice in peaches, which were first subjected to a 2-day delay at 23°C before storage followed by ripening for 12 days at 10°C, decreased gradually from 60% to about 42%. In contrast, the percentage extractable juice in the peaches stored

immediately remained fairly constant near 62% over the entire 3-week storage period. In both cases the extractable juice content decreased rapidly in the subsequent 12-day ripening period. The extractable juice content of peaches which became woolly was significantly higher than that of sound fruit during most of the storage period. However, woolly peaches contained significantly less extractable juice after ripening than normal ripening peaches. Lurie and co-workers (1993) confirmed that nectarines during cool air storage (8 weeks) had slightly higher extractable juice content than that of fruit after harvest. After ripening, the cool storage nectarines had much less extractable juice content than that of harvest fruit. Dawson and co-workers (1995) found that the extractable juice content of fruit was low during storage at 0°C, whether continuous or with warming. Ripening immediately after harvest resulted in a considerable increase in the extractable juice content, whereas mealy fruit showed no increase from levels during storage.

There are many kinds of extractable juiciness measurement. Ben-Arie and Lavee (1971) placed peaches under constant pressure, with the aid of a "succulometer" to measure the expressed juice. Buescher and Furmanski (1978) estimated expressible juice by determining weight loss from tissue disks (18 mm diameter, 3 mm thickness) after centrifuging for 10 min at 1700×g. Tissue disks were supported over a wad of absorbent paper by polyethylene net during centrifugation. They found that results from this method correlated very well ($r = 0.88$) with sensory scores for juiciness.

Lill and van der Mespel (1988) used the following method to measure the extractable juice of nectarine fruit. Segments of tissue (approximately 1.5 g) were taken from nectarines after the skin had been removed. Each segment was placed in a 5 ml disposal syringe with no needle and forced through the luer hub (1.9 mm internal diameter, 10 mm long) to achieve gentle homogenisation. The homogenate was collected in an Eppendorf centrifuge tube, weighed and centrifuged (12000×g). The weight of supernatant juice was expressed as a percentage of the weight of the sample. This value was considered to be the apparent juice content. Through studying the affect of centrifugation time and syringe aperture diameter, they suggested that centrifugation time should not be less than 5 min, and syringe apertures should range from 1.95 to 4.3

mm. This method was used by Lurie *et al.* (1993, 1994). A modification of this method was used by Dawson and co-workers (1993, 1995) to measure expressible juice of nectarines. They inserted a 1 cm diameter core from the mesocarp into a 2.5 ml syringe barrel with glass wool plugs and centrifuged for 5 min at 5000g, after which the extractable juice was weighed.

Von Mollendorff and De Villiers (1988a) measured the amount of extractable juice (as an indication of free moisture) of peaches using an electric liquidizer. Juice was separated from solid phase of ten peaches by a fast-rotating sieve. The weight of the juice thus obtained was expressed as a percentage of the weight of the fresh fruit. The total moisture content was determined by drying a known weight of fresh tissue at 80°C for 14 days.

Von Mollendorff and co-workers (1992) used 20 fruits to determine extractable juice by homogenising 200 g of tissue for 60 sec in a washing blender. The suspension was allowed to stand for 15 min before centrifugation for 10 min at $1000 \times g$. The clear decanted liquid was weighted and expressed as a percentage of the fresh fruit weight. The lowest values for extractable juice coincided with the highest incidence of mealy fruit. No further analysis was conducted.

2.10.3 Electrolyte Leakage and Internal Conductivity

2.10.3.1 Electrolyte leakage

Electrolyte leakage is the measurement of electric conductivity which represents the leakage of ions (Furmanski and Buescher, 1979). It is increased mainly by chilling injury, which is presumably due to increased membrane permeability of the tissue (Creencia and Bramlage, 1971). The typical objective of electrolyte leakage studies is to assess injury, presumably at the membrane level, resulting from environmental stress.

Furmanski and Buescher (1979) used the mesocarp tissue to measure peach fruit electrolyte leakage. The mesocarp tissue was prepared by slicing cylinders extracted

with a cork borer into discs (2 mm thickness and 12 mm diameter), washing 3 times with distilled water, and incubating in 200 ml of 0.4 M mannitol for 3 hr at 30°C in a shaking water bath. Electrolyte leakage was determined with a conductivity bridge (cell constant of 1.0) after incubating the discs and again after boiling for 15 min and cooling to 30°C (total electrolytes). Electrolyte leakage after 3 hr was calculated as percentage of total electrolytes. They found electrolyte leakage remained low and did not appear to be altered while peaches were held at 1°C. Fruit ripening dramatically enhanced the electrolyte leakage, and the appearance of woolliness in ripened fruit was associated with reduced electrolyte leakage rather than its initial enhancement.

2.10.3.2 Internal conductivity

The impedance of a biological system is related to the resistance and capacitive reactance of the tissue. Recent studies have demonstrated that plant tissue conforms to an electrical double-shell model which includes resistive and capacitive components associated with the vacuole, cytoplasm, plasma membrane and extracellular space (Zhang and Willison, 1991). Both intracellular and extracellular current pathways can be assessed using impedance measurements (Stout, 1988; Harker and Dunlop, 1994). The extracellular pathway is predominant when low frequency (50-100 Hz) alternating current is passed through the tissue, while the intracellular pathway predominates at high frequencies (100 kHz). Generally, the impedance of the high frequency (intracellular) pathway is lower than that of the low frequency (extracellular) pathway. Presumably, the decrease in impedance of the intracellular pathway (mainly associated with the ionic content of the protoplasm) is little altered by membrane damage. However, low frequency current which is unable to cross the plasma membrane of undamaged tissue, gains access to the entire cell once the plasma membrane becomes damaged. Thus the resistance of the low frequency pathway decreases as a greater proportion of the tissue becomes accessible to electrical current.

Weaver and Jackson (1966) measured peach impedance with a portable transistorised impedance tester, Model ZP2 manufactured by Sennheiser Electronics, Germany. The electrodes consisted of fine nickel-plated needles which were embedded in a rubber

insulator with a separation of 5 mm and a length of 7 mm. Measurement at 250 and 4000 Hz were taken by inserting the electrodes into the cheek of the fruit and the impedance value was immediately recorded. This method was used by Furmanski and Buescher (1979) and Von Mollendorff and co-workers (1992) to measure the conductivity ($1/\text{impedance}$) of fruit flesh (whole fruit).

Harker and Dunlop (1994) conducted research on nectarines. They measured fruit impedance using the method illustrated in **Figure 2-2**.

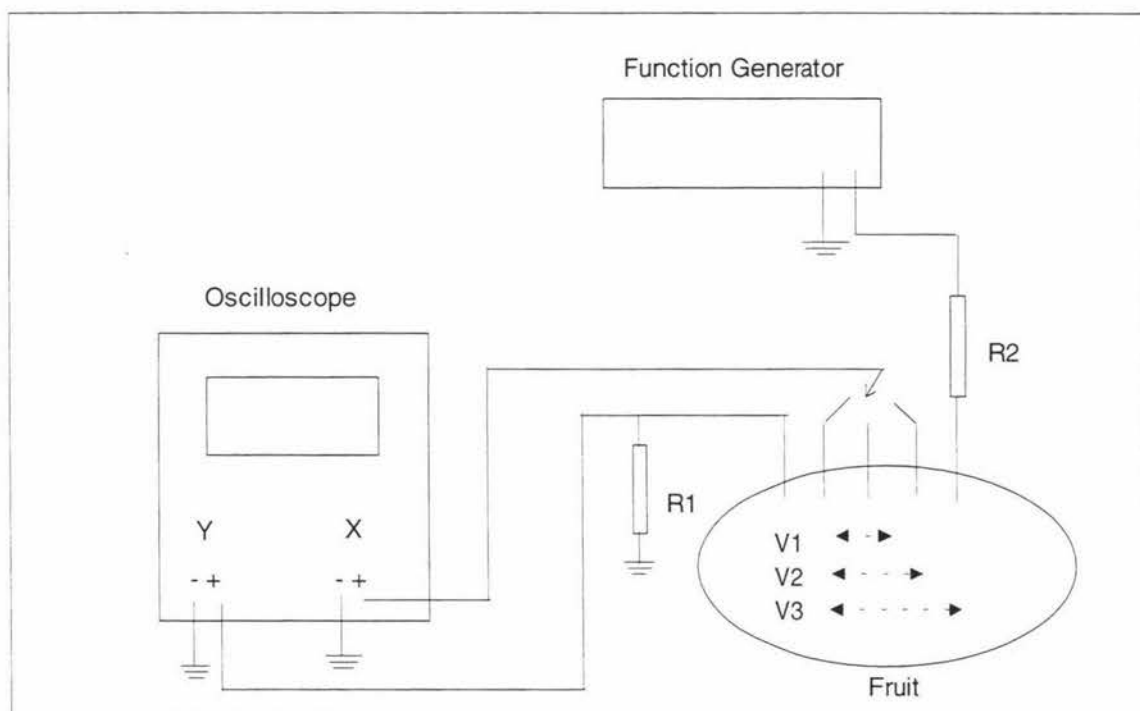


Figure 2-2 Circuit diagram showing the arrangement of equipment used for impedance measurements.

A function generator (Model F32, Interstate Electronic Corporation), an Oscilloscope (Model 50103N, Tektronix), and array of 4 parallel stainless steel (0.45 mm diameter) or silver (0.6 mm diameter) electrodes were connected as shown in **Figure 2-2**. The electrodes were spaced at 1 cm intervals to allow impedance measurements across 1 cm, 2 cm and 3 cm of tissue. A tangential slice was removed from the cheek of the nectarine and the electrode array inserted to a depth of 6 mm into the flesh so that the electrodes

were spaced along the fruit equator. A sine wave (50 Hz to 100 kHz frequency) was fed into Y input on the oscilloscope whilst V_1 , V_2 or V_3 (depending on the position of the switch) was fed into the X input of the oscilloscope resulting in an elliptical figure which was traced out on the oscilloscope. The resistance (r_j) and reactance (x_j) of the fruit was determined from the following formulae:

$$r_j = Z_j \cos\theta_j - R_0 \quad (1) \quad \text{and} \quad x_j = Z_j \sin\theta_j \quad (2)$$

$$\text{where } Z_j = V_j R_0 / V_0 \quad (3) \quad \text{and} \quad \sin\theta_j = V_0' / V_0 \quad (4)$$

where V_j , V_0 and V_0' are dimensions used to characterize the ellipse traced out on the oscilloscope, and R_0 is 2022 ohms.

The electrode and tissue resistance and reactance were determined by plotting r_j and x_j against interelectrode distance. The y-axis intercept gives resistance and reactance of the electrode, whilst the slope provides the resistance and reactance of the tissue. They investigated the effects of current frequency, needle material, whole fruit and fruit tissue, and found that tissue resistance decreased in a sigmoidal fashion as frequency of the current was increased. The greatest resistance was achieved at low frequencies and there was only a small decrease in resistance as the frequency was lowered from 100 to 50 Hz. The resistance and reactance of stainless steel electrodes was far higher than those of silver electrodes. Changes in solution PH could influence the potential of stainless steel electrodes. Tissue resistance was higher when blocks of excised tissue were used instead of whole fruit, and values for whole fruit resistance measurements may be affected by fruit size.

Electrical impedance measurements have been used as maturity indices during fruit development (Weaver and Jackson, 1966). Furmanski and Buescher (1979) used this method to study the physiological changes during storage and ripening of fruit. They found that impedance of the fruit decreased during ripening, but once ripening was completed, the impedance was higher in mealy than non-mealy fruit. Von Mollendorff and co-workers (1992) showed that although a rapid increase in internal conductivity occurred during nectarine ripening, the onset of the increase was advanced in mealy

fruit. Harker and Dunlop (1994) conducted research on nectarines. They found that, at low frequencies (extracellular pathway), the resistance of unripened nectarines increased as the fruit were stored from 0 to 3 weeks, then decreased as the fruit were stored for 8 weeks (after which time nectarines were mealy). The resistance of fruit stored for 0 to 3 weeks decreased to similar levels at low frequency when they were allowed to ripen for 5 days. However, the resistance of fruit stored for 8 weeks increased by about 200 ohms when allowed to ripen, although the final resistance was still lower than that of fruit ripened following shorter periods of coolstorage. Harker and Maindonald (1994) used electrical impedance measurements to study the changes in the cell wall, vacuole, and membranes, and found that electrical impedance measurements were related to changes in fruit texture assessed by flesh firmness and apparent juice content. They also found that changes in tissue resistance measured using low frequencies of alternating current were closely related to flesh firmness. The main difference between these woolly and non-woolly nectarines was the resistance of the cell wall, which was higher in woolly tissue than in non-woolly tissue.

2.10.4 Mechanical Measurements

Different ripening processes may modify cell wall polymer differently, which will lead to changes in intermolecular interactions and hence affect cell wall characteristics such as strength. King *et al.* (1989) found that mealiness (pastiness) of 'Fantasia' nectarine was associated with separation of the middle lamella without extensive degradation of the cell wall. Holt and Schoorl (1984) studied the mechanical properties and texture of stored apples, and found that tensile testing of apple tissue was a reliable way of quantifying texture deterioration during storage. They found that ultimate tensile showed strong linear negative correlations with time. Luza and co-workers (1992) proved this finding and indicated some of the major structural changes related to softening, and later to chilling injury (mealy fruit), were those related to loosening of cell walls, loss of wall cohesion, development of an intercellular matrix with new carbohydrates and pectins, and apparent cell wall synthesis. Harker and Hallett (1992) suggested that adhesion between neighbouring cells can be measured by the application of tensile tests of plugs of fruit tissue. They conducted research on apple mealiness, and

found that changes in tensile strength of apple tissue were related to the way in which cells separated from each other, and this conclusion is supported by the observation of fracture surfaces. They also found that tensile strength of mealy apples was much lower than that of non-mealy apples. Harker and Sutherland (1993) continued their studies on nectarines, and found that during ripening, adhesion between cells decreased, turgor pressures decreased, and propensity of cells to rupture increased. These changes occurred in both non-mealy and mealy nectarines and the magnitude of the changes in tissue strength were the same for both mealy and non-mealy nectarines. The principle differences between non-mealy and mealy nectarines was the presence or absence of juice on the fracture surface following the application of tensile tests. Tu *et al.* (1996) measured the Grammy smith apple tensile strength, and indicated that tensile tests might not conclusively indicate the degree of mealiness but might possibly indicate if apple was mealy or not as the apple flesh strength fails below a certain level.

There are different ways to measure tensile strength. Holt and Schoorl (1984) carried out tensile test at a loading rate of 50 mm min^{-1} in a Model 1122 Instron Universal Tester on specimens cut from slices of apple tissue and gripped in standard jaws. The shape of the specimens is shown in **Figure 2-3**. The specimen was loaded to failure. A force-deformation curve was recorded for each specimen, and the cross-section of the specimen were measured. The maximum force and energy input for each specimen were measured from the force-deformation records.

Stow (1989) used the following method to measure force required to cause failure in tissue subjected to tension (rupture force). A 30 mm thick equatorial slice was taken from each fruit and 14 mm diameter plugs were removed with a cork borer. Plugs were cut to produce an 'H' shape with the ends each 14 mm in diameter and 15 mm long joined by a $4 \text{ mm} \times 12 \text{ mm}$ stripe (as shown in **Figure 2-4**). Plugs were inserted into a 4 mm wide groove cut into two 5 mm thick plates attached to the motor platform and bar of a recording penetrometer. When the platform was driven down at 0.6 mm min^{-1} the bar was pulled down via the 4 mm strip in the middle of the apple plug, until the strip failed. The force at this point was recorded and is reported as the 'rupture force'.

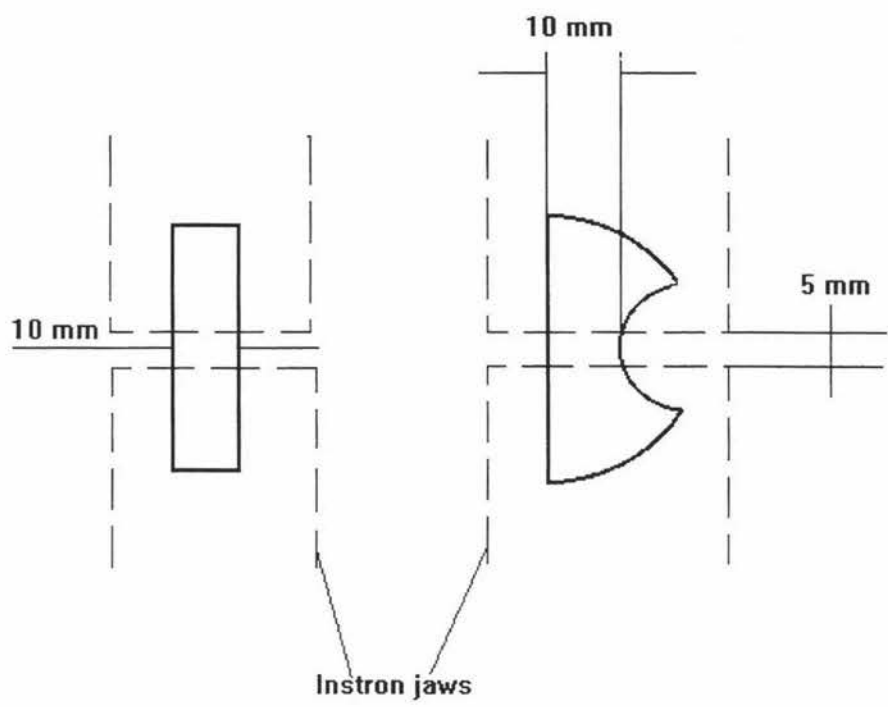


Figure 2-3 Apple specimen cut from slice through whole apple

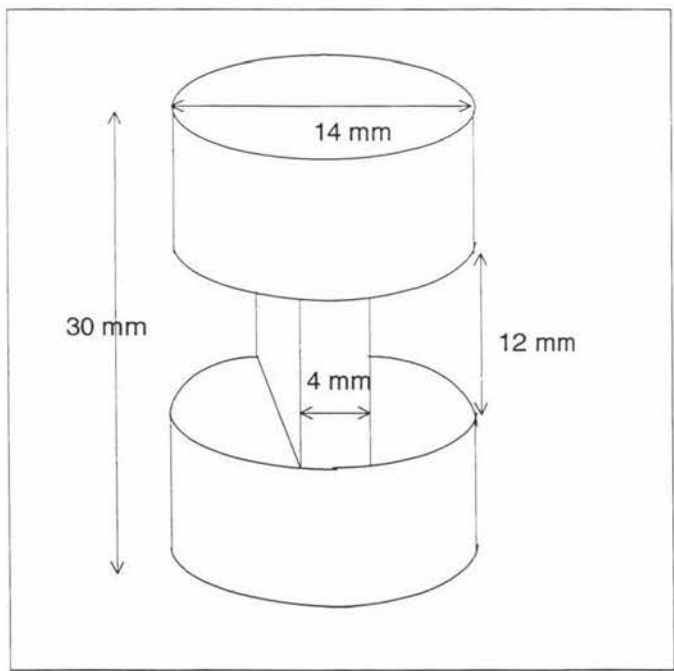


Figure 2-4 Dimensions of apple tissue plug used for tensile test

Harker and Hallett (1992) used the same method to measure apple cell cohesion. They inserted a 10 mm diameter cork borer radially and two plugs of cortical tissue were removed around the equator of each apple. Each plug was then cut into an H-shape section. The waist of the section was 5 mm in diameter. The tensile test was applied by inserting the section between two sets of claws of an Instron model 4301 materials testing machine (Instron, Canton, Mass.). The sets of claws moved apart at the rate of 2 mm min^{-1} , stretching the tissue section until it snapped across the waist. The force required for tissue failure was recorded, and this value was divided by the waist cross-sectional area to give results as Newtons per square centimetre.

Harker and Sutherland (1993) measured tensile strength of nectarines. They used an 11 mm diameter borer. The plugs (approximately 30 mm long) were prepared for use in tensile tests by cutting semicircular notches off opposite sides about 15 mm along the plug using a cutter made from two 5 mm diameter cork borers set 4 mm apart. The notches were originated along a radial plane in the original fruit (as shown in **Figure 2-5**). The ends of the plug were then fixed to metal strips using a cyanoacrylate adhesive in a gel form (Selleys Supa Glue Gel) and the plug was then inserted between two sets of claws on an Instron model 4301 materials testing machine. At the start of the tensile test the claws moved apart at a rate of 10 mm/min, strengthening the tissue until the plug snapped across the waist. The force required for tissue failure was recorded.

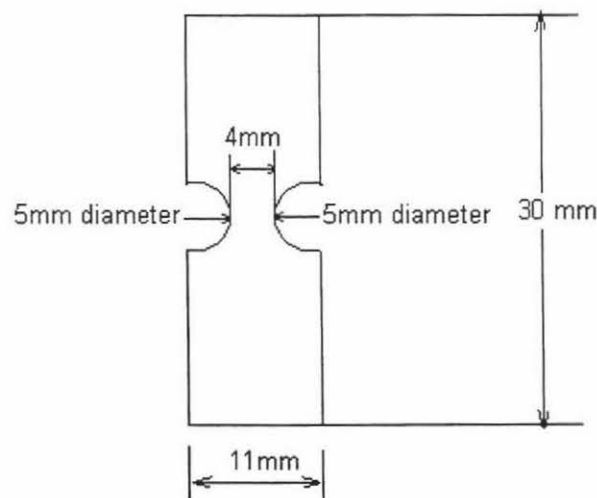


Figure 2-5 Nectarine tissue plug used for tensile test (Harker and Sutherland, 1993)

Tu and co-workers (1996) measured the tensile strength of Grammy Smith apples. They used ring-shaped samples subjected to radial loading. The test device consisted of two half ring shaped cylinders over which the ring shaped sample can slide, as shown in Figure 2-6.

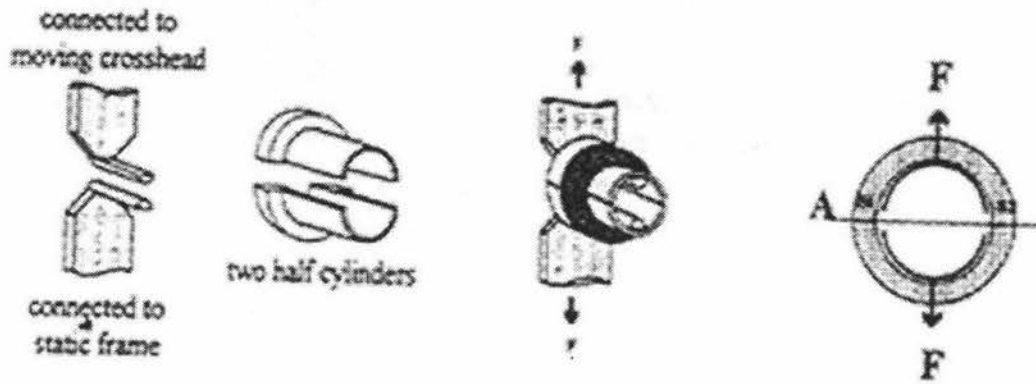


Figure 2-6 The tensile measurement set up

During the test, the moving crosshead moved at the test speed and the ring-shaped sample deformed and eventually broke. The force-deformation curve was recorded. It was assumed that the deformation ϵ was the same in both 'loop' sides since the material in both sides was the same. The tensile stress was calculated by dividing the measured force F by the sum of the cross-sections S_1 and S_2 of each 'loop': $\sigma = F/(S_1 + S_2)$. They suggested that this type of test avoided clamping problems of the apple specimen as well as damage to the sample texture.

2.10.5 Strength and Elasticity of the Cell Wall

Strength and elasticity of the cell wall and plasma membrane are important for the changes of fruit texture. Harker and Hallett (1992) found that there was little difference in the turgor pressures between low and high maturity apples at harvest, but after cool storage, the pressures required to rupture cells from low maturity (non-mealy) fruit had decreased 40%, while there was no change in the pressure required to burst cells from high-maturity (mealy) fruit. Harker and Sutherland (1993) found that when discs of fruit

tissue were incubated in solutions containing varying concentrations of mannitol, the weight of the disks changed due to infiltration of extracellular regions, osmosis, solute leakage and cell rupture, as well as other unknown causes. The only difference between ripened and unripened tissue was the ability of tissue to hydrate. When discs were incubated in isotonic solutions (tonicity of the tissue determined by dewpoint psychrometer), the weight of discs from unripened tissue increased by about 10%, whilst discs from ripened tissue increased in weight by less than 5%.

Harker and Hallett (1992) measured the water potential at the point of bursting and osmotic potential of apple. The turgor pressure required to rupture cells were determined on disks of cortical tissue (10 mm in diameter, 3 mm thick) cut from five apples. Disks were combined, then rinsed in deionized water (30 sec) and blotted dry. Disks of about 1 g were weighted to a precision of ± 0.001 g into duplicate 50-ml flasks containing 20 ml of 0.1, 0.2, 0.3, 0.4, or 0.5 molar mannitol with 0.5 mM mercapto-benzothiozole and 0.5 mM CaCl_2 . The disks were incubated in these solutions for 3 h before being filtered, blotted, and reweighed. The lowest concentration of osmoticum below which cells started to burst was determined from plots of concentration against changes in disk weight. The maximum changes in disk weight indicated uptake of water by osmosis, and cells burst when the disks were placed in lower concentrations of mannitol. The turgor pressure required to burst the cells was then estimated using the formula $P = \psi - \pi$, where P is the turgor pressure, ψ is the water potential at the point of bursting, and π is the osmotic potential of the cell sap. Osmotic potential of the apple cortical tissue was estimated from SSC (Soluble Solid Content), assuming the soluble sugars were the main contributor to the osmotic potential and ignoring the effects of other solutes.

Harker and Sutherland (1993) used the same method on nectarine tissues. Instead of calculating turgor pressure, they determined the water potential of nectarine tissue as the change in disc weight which was expressed as a % of the initial disc weight.

2.10.6 Firmness Measurements

Penetrometers, as a force measuring instrument, have been used for over 60 years. The test consists of measuring the force required to push a probe or punch into fruit to a depth that causes irreversible crushing or flow of the fruit (Bourne, 1979). This force is defined as firmness. Firmness is a physical properties that is widely used for evaluating the quality of fruits. Firmness of some fruits decreases gradually as they become mature and decreases rapidly as they ripen (Chen, P. and Sun, Z. 1991).

Many researchers have measured fruit firmness changes during woolly breakdown. Ben-Arie and Sonogo (1980) found that with the development of woolly breakdown there was no additional fruit softening, in contrast there was a decline in fruit firmness for healthy ripening fruit. Von Mollendorff and De Villiers (1988a) measured peach firmness, and reported that the firmness of peaches stored after the delay period decreased sharply during the 3-week storage period. It decreased further during subsequent ripening period. Fruit stored without delay remained firm for the whole storage period followed by a sharp decrease during ripening especially in the first three to six days, coinciding with the onset of woolliness. Dawson and co-workers (1993) reported that during ripening after cool storage, fruit softened to the same extent as did those ripened without cool storage. Dawson and co-workers (1995) continued their research and indicated that fruit stored continually at 0°C softened only slightly during storage. Ripening for 6 days at 20°C resulted in a loss of flesh firmness for all fruit, whether ripened immediately after harvest or ripened after storage, and mealy fruit softened to a similar extent as fruit that ripened normally. Lurie and co-workers (1994) got the same results. However, a contrary result was reported by Ben-Arie and co-workers (1989). They found that during cold storage there was very little noticeable change in peach firmness or ripening. The symptoms of the disorder became apparent upon transfer to higher temperatures for ripening. The fruit lost some of its ability to soften and it failed to become juicy. This was proved by Harker and Dunlop (1994). They found that firmness of mealy nectarines is higher than that of normal fruit after ripening.

Although the principle of firmness measurements was the same, the equipments especially the probes used for summer fruit firmness measurement were different. Ben-Arie and Sonego (1980) determined the firmness changes of stored peaches with a Hunter penetrometer equipped with an 11-mm tip on two pared cheeks of each fruit. Von Mollendorff and De Villiers (1988a) used an Effigi penetrometer fitted with an 11 mm point to measure peach firmness. Lill and Der Mespel (1988) measured nectarine firmness with a Chatillon force gauge with an 8 mm diameter plunger. Dawson *et al.* (1993) and Dawson and co-workers (1995) used an Effigi penetrometer (FT011) fitted with an 7 mm diameter plunger to measure the firmness of nectarines. Harker and Dunlop (1994) used an Atago handheld penetrometer with an 7.9 mm diameter prober measured nectarine firmness. Lurie and co-workers (1994) used an 11.1 mm tip penetrometer to measure nectarine firmness.

2.10.7 Internal Air Space

The parenchyma of a mature apple has a density less than unity indicating that there are spaces between the cells, estimated in cv Granny Smith to be about 27% of the total volume (Bain and Robertson 1951). A similar figure has been derived for the other varieties of apple (Reeve, 1953). The parenchyma of apples in store, especially in high humidity, tended to increase in permeability, indicating a further increase in the volume of air space and leading to a 'mealy' texture which was due to the almost complete separation of cells (Wilkinson, 1965). Vincent (1989) measured apple voids and found that apple flesh contained air spaces or channels which radiated from the centre. Harker and Hallett (1992) measured the % air space in apple, they indicated that the volume of air space was greater in apples harvested at high than at low maturity, and although air space expanded during cool storage, this relative difference is maintained. They indicated that high levels of air space are symptomatic of mealiness. The cause of these large air spaces and mealy texture is probably related to the degradation of the middle lamella and corresponding reduction in cell adhesion. Harker and Sutherland (1993) also reported that the cellular characteristics particularly associated with mealy tissue were high volumes of air space. Dawson and co-workers (1993) found that the percentage internal air space of nectarine tissue decreased during normal ripening but was

significantly increased in mealy fruit. Tu and co-workers (1996) studied the internal air space of Granny Smith apples. They found that internal air space increased during the storage, and when the internal air space increased around 20% Granny Smith and Elster Jonagold apples tended to taste mealy. Further study (Tu and De Baerdemaeker, 1996) indicated that although the internal air space for different apple varieties might be different, it seemed to be a good indicator of apple mealiness. The higher IAS ($\geq 21\%$) normally corresponded to poor, mealy apple texture.

Hatfield and Knee (1988) described a method to measure the apple internal air space (IAS) as: $IAS = 1 - SG/J$ where SG is the apple specific density, and J is the specific density of apple juice. for Cox's apple juice, the specific density was taken to be 1.059. This value was the average of a number of estimations using a pycnometer. They also indicated that a change of 10 mg g^{-1} in the sugar content of apples would cause a change of 0.004 in specific density of apple juice J; in a fruit with an IAS of 0.2, neglect of this change in sugar would introduce an error of about 0.003, or 1.5% of the estimated IAS. Respiration during storage would deplete sugar concentration by about this amount, but this would be offset by the concentrative effect of any water loss. The same method was used by Harker and Hallett (1992), Tu and co-workers (1996), and Tu and De Baerdemaeker (1996).

Harker and Sutherland (1993) used the same method to measure nectarine internal air space. The tissue density was determined by weighing the plugs whilst floating and again when submerged in a solution of 10% sucrose (w/v). Plugs were cut from nectarines using a 10 mm diameter cork borer. Sucrose solutions were required as the tissue had a specific gravity close to that of water. The apparent juice of each nectarine was determined using the method of Lill and van der Mespel (1988). Plugs of mesocarp tissue (10 mm diameter) were collected using cork borer then macerated by forcing them through a 5 ml plastic hypodermic syringe into preweighed 2.5 ml microfuge tubes. The macerated tissue was centrifuged for 5 min at 10000 rpm, and the supernatant was removed. The supernatant weight was expressed as a percentage of the initial weight of macerated tissue to give the apparent free juice content. The juice was frozen in liquid nitrogen and stored at -20°C . The specific gravity of juice (SG juice) was determined by

collecting 200 μ l of thawed juice into a preweighed micropipette tip. The tip was then removed from the micropipette and reweighed. Dawson and co-workers (1993) used the same method to measure nectarines internal air space.

Vincent (1989) determined apple voids by subjecting slices of parenchyma (cut 2 mm thick, equatorially normal to the core and with the peel removed) to varying degrees of vacuum, and allowing them to take up 5% w/v aqueous mannitol into the evacuated air spaces on return to atmospheric pressure. Vacuum was generated by a water pump attached to a mains supply of water and was measured with a simple mercury manometer. The samples were contained in a Buchner flask and were subjected to the vacuum for about a minute during which time the flask was shaken so as to remove the gas bubbles from the surface of the apple slice. The slice was left in the mannitol for about 10 min after returning to atmospheric pressure to ensure full penetration of the evacuated spaces. The slices were then photographed in transmitted light so that the translucent areas where the mannitol had penetrated between the cells appeared light and the opaque areas, which still contained air, appeared dark.

2.11 RELATIONSHIP BETWEEN FRUIT MEALINESS WITH OTHER MECHANICAL PROPERTIES

Fruit texture, as a general term, is often used to encompass both sensory reactions and mechanical responses of the food material to applied forces. Most horticultural studies of texture have related mechanical measurements to fruit maturity or to resistance to mechanical injury (Abbott *et al.*, 1984). The mechanical tests applicable to food materials include compression, shear, torsion, bending, tensile etc. (Mohsenin, 1986b). Some of these tests has been widely used in the fruit industry, such as firmness.

In addition to tensile strength for apples, fruit mealiness may be related to other mechanical properties. Tu *et al.* (1996) found that compressive rupture force for Granny Smith apples changed linearly with time. Compressive rupture and tensile force have nearly the same change pattern. Tu and De Baerdemaeker (1996) used the texture profile analysis (TPA) to study the apple mealy texture and found that fresh non-mealy

apple did not show adhesiveness during texture profile analysis but the ripened, mealy apple did show some adhesiveness.

Biological materials are commonly anisotropic, hence their mechanical properties differ according to the orientation in which it is tested (Khan and Vincent, 1993b). Khan and Vincent (1990) studied the anisotropy of apple parenchyma, and found that the apple cells immediately underneath the surface were small and radially flattened with their maximum dimension of about 50 μm . Progressing towards the core of the cells gradually increased in size until they reached a maximum of about 200-300 μm in diameter (depending on the variety of apple) at 5-10 mm from the surface. The inner cells became increasingly radially elongated and began to be organised into radial columns diverging from near the centre of the fruit towards the periphery. In polar direction the cells appeared rounded and showed no orientations since the columns of cells were being viewed from one end. Due to this structure difference, when force was applied at the equatorial direction, the apple was compressed along the cell columns, the deformation resistance was higher in this direction. When the force was applied in polar direction, the apple cell was compressed in the right angle of the cell column, the deformation resistance in this direction is lower than that in column direction.

With the development of fruit mealiness research, mechanical properties such compressive, fracture and shear properties may be found to be related to the development of fruit mealiness. According to Khan and Vincent (1993a), early season apples had less resistance to deformation and fracture test and thus needed less energy to compress. Crack opening tests have shown that in early apples fracture was a result of cell separation and in late ones of similar age it was a result of cell breakage (Khan and Vincent 1993b). This was consistent with the fact that early or mid season apples (Cox and Bramley) tend to become mealy sooner than late varieties (Norfolk Beefing) (Khan and Vincent, 1993a).

2.12 SUMMARY OF FINDINGS

Mealiness occurs widely in many cultivars of apples and stone fruits. Research in this area began in the early part of this century (Harding and Haller, 1932; 1934; Fisher, 1943). Mealiness is defined as a texture deterioration. For stone fruit it is similar to chilling injury symptoms. It is not the loss of water, but just the way the water is bound or tied up chemically within the fruit which makes it difficult to extract juice (Rowe, 1986). These texture changes are related to cell wall changes and cell pectin substance changes.

Some postharvest factors affect the development of fruit mealiness, such as temperature, humidity, oxygen and carbon dioxide concentration, and fruit maturity. Some techniques may be useful to prevent or alleviate this disorder, such as intermittent warming and delayed storage for stone fruit. Controlled atmosphere is useful for reducing the development of fruit mealiness and prolonging fruit storage life.

Several assessment methods have been used to assess fruit mealiness, e.g. extractable juice, storage time under certain storage conditions for stone fruit, taste assessment, etc. Among these assessment methods, taste assessment is the most interpretable one and so it is the basic one to other assessment methods.

Several test parameters are related to fruit mealiness development, such as extractable juice, internal conductivity, cellular adhesion and turgor pressure, and internal air space. Some of these parameters are useful indicators for specific fruit, but not useful for others. For example cell adhesion, measured by tensile test, may be useful for apple mealiness development (Harker and Hallett, 1992), but not related to the mealiness of nectarines (Harker and Sutherland, 1993). Internal electrical conductance (or impedance) is useful to measure peach mealiness (Furmanski and Buescher, 1979), but no research have been done to investigate the usefulness of this parameter to measure apple fruit mealiness. Internal air space was suggested to be a useful indicator for the mealiness of apples (Harker and Hallett, 1992) and nectarines (Harker and Sutherland 1993). It is suggested that the only problem is what kind of juice should be used to

measure the density. Although many researchers used juice obtained by the method described by Lill and van der Mespel (1988), but centrifugation equipment, centrifuged speed, and time were quite different. These difference may cause density measurement inaccuracy and thus may be unstable.

2.13 JUSTIFICATION AND CONTRIBUTION OF PRESENT STUDY

As described above, many studies have been done on the objective measurement of fruit mealiness, but as yet there are no good indicators based on fruit physical properties which have been developed and used in fruit industries. Some of the parameters may be effective to measure the fruit mealiness, such as internal air space, extractable juice, but further research work needs to be done to correlate these parameters with subjective tests, to investigate the correlation with other physical parameters and to monitor changes in values with the development of fruit mealiness.

This study is intended to contribute to a better understanding of fruit mealiness, which may help the fruit industry to better manage fruit storage according to fruit variety and maturity.

In this study, apple physical property, especially mechanical property, changes during the development of mealiness were investigated. Through comparison, the difference between mealy apple and aged but not mealy apples was studied. Finally through regression and statistical analysing, possible apple mealiness indicators have been suggested and assessed. This may help the apple industry to detect apple mealiness development before fruit are put onto the market, and so to provide high quality fruit to the consumer. This study may also be useful for other researchers in developing mealiness indicators for other kinds of fruits.

CHAPTER THREE

MATERIALS AND METHODS

3.1 EXPERIMENTAL DESIGN

This experiment consisted two parts: One was the investigation of apple physical property changes with the development of mealiness; and the other part was to compare physical properties of apples stored in different conditions, and to correlate apple physical properties with sensory tests conducted at the same day and on the same fruit, to find an apple mealiness measurement indicator.

3.1.1 Samples

Based on the availability and importance in New Zealand fruit industries, “Braeburn” apples were chosen. This cultivar is not only the most popular export apple variety in June 1996 year, but also the most commonly planted apple tree for export production in New Zealand (New Zealand Official Year Book, 1997).

The fruit for the first experiment was produced in the Waikato area. It was harvested at the end of April, 1997, and stored in cold storage (about 0°C). The fruit for the second experiment was supplied by Massey University Fruit Crop Unit, Palmerston North. The apples were harvested in late April, and stored in cold storage until required.

3.1.2 Treatment

In the first experiment, Braeburn apples, taken from cold storage, were put in perforated plastic bags to achieve high humidity conditions, and held at $20 \pm 2^{\circ}\text{C}$ in a controlled temperature laboratory. This storage condition was chosen to make apples mealy quickly.

In the second experiment, Braeburn apples from the Fruit Crops Unit, Massey University, were divided into four groups. The first two groups were stored at high temperature (20°C) but in different humidity conditions. One group was put in the same storage condition with the first experiment to make the apples mealy. The other group was stored at $20 \pm 2^{\circ}\text{C}$ but under low humidity ($50 \pm 15\%$ R.H.) to make the apples shrivelled. The other two groups of apples were put into low temperature storage conditions (0°C). One group was put into an apple carton directly, and the other group was put in the carton with a perforated plastic bag cover to achieve high humidity condition.

3.1.3 Sample Size

There is no correct sample size that can be determined without additional information. The size of the sample required for a given experiment is influenced by the values selected for alpha (α) and beta (β) risks (see below), by the selection of an important increment of test response (δ), and by the value, or values, of population variances (σ^2).

α and β represent two kinds of errors in any experiment:

Alpha (α) error --- the experiment accepts the alternative hypothesis (H_a) as being true when the null hypothesis (H_0) is actually true.

Beta (β) error --- the experiment accepts the null hypothesis as being true when the alternative hypothesis is actually true.

For this study, the experiment is a simple comparative experiment. The mean value and the population variances are unknown, but the population variance of every test sample should be the same because all the sample was from the same source. So the object of the experiment can be assumed as $H_0: \mu_1 = \mu_2$ and $H_a: \mu_1 \neq \mu_2$; $\sigma_1 = \sigma_2$ but unknown. Assuming $\alpha = \beta = 0.10$. but δ is specified in terms of σ , that is $\delta = f(\sigma)$. suppose that $\delta = \sigma$.

According to Diamond (1981) U_α can be obtained from his table 2 (double sided table) (Appendix 1), which is 1.645, and U_β can be obtained from Table 1 (single sided table) (Appendix 1), which is 1.282.

$$\text{so } N = 2 * (U_\alpha + U_\beta)^2 \sigma^2 / \delta^2 = 17.135.$$

Compute ϕ using $\phi = N + N - 2 = 32.27 \approx 32$.

Using ϕ to compute $N_t = 2 * (t_\alpha + t_\beta)^2 * \sigma^2 / \delta^2$

t_α is obtained from table 4 (double sided t distribution table) (Appendix 1), which is 1.698, and t_β can be obtained from table 3 (single sided t distribution table) (Appendix 1), which is 1.3093.

So $N_t = 18.08 \approx 18$, which means the sample should be more than 18 apples.

Based on this calculation, the sample size for this experiment was 20 apples.

3.2 MEASUREMENT OF PHYSICAL AND MECHANICAL PROPERTIES OF FRUIT

Each sample of fruit was selected at random. The selected fruit were numbered using indelible markers.

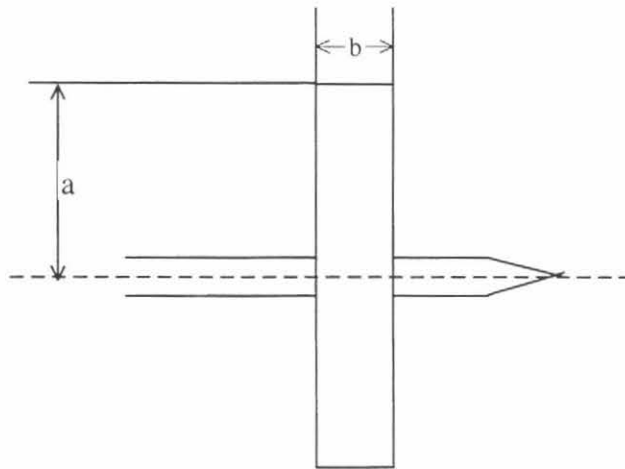
3.2.1 Fruit Density

Fruit density was measured by Archimedes principle. A METTLER PM6100 desk-top balance was used for weight measurement. The specific density of apples was estimated by weighing a beaker containing a certain amount of water (W_1) and reweighing the beaker with an apple (W_2). A third weight (W_3) was recorded with the apple held below the water surface by three fine needles which did not themselves influence the balance reading. The specific density was calculated from

$$\text{SG(specific density)} = \frac{\text{Weight of apple}}{\text{Weight of water displaced}} = \frac{W_2 - W_1}{W_3 - W_1} \quad (1)$$

3.2.2 Fruit Firmness

The Massey Twist Tester was used to measure the crush strength of fruit flesh. This device measures the moment required to crush fruit cells using a blade and this moment is converted to a crushing stress figure for the tissue by calculation (Studman and Yuwana, 1992). The blade size used for this experiment is shown in **Figure 3-1**. The twist tester is connected to a computer, which recorded the results.



a = blade radius, and b = blade width; In this experiment $a = 6$ mm and $b = 2.5$ mm.

Figure 3-1 Enlargement of the blade.

3.2.3 Wholefruit Elastic Modulus

Elastic modulus was measured by a TA-XT2 Texture Analyser. The whole intact apple was put on the heavy duty platform. Uniaxial compression (the sample was compressed in one direction while unrestrained in the other directions) was conducted. A flat ended 5 mm stainless steel cylinder punch was used to compress the whole fruit to 2% strain at a constant deformation rate of 1 mm per second. The load was held for 1 second and then unloaded at 1 mm per second. The force required, the fruit height were recorded automatically. By running a macro all of these data were put into a result spreadsheet. In the spreadsheet the elastic modulus was calculated automatically by the formula (Mohsenin, 1986a):

$$E \text{ (elastic modulus)} = \frac{F / A}{\Delta L / L} \quad (2)$$

where

F = force

A = cross-sectional area of the punch

ΔL = deformation corresponding to force F

L = fruit height.

Each apple was tested twice by this method. One was in the polar (stem-calyx) direction (P direction), and the other was in cheek to cheek (C direction) as shown in **Figure 3-2**. In the P direction the punch contacted the fruit at its shoulder, while the base rested on the flat platform. For the C direction test, the load was applied at the cheeks and hence through the centre of the fruit.

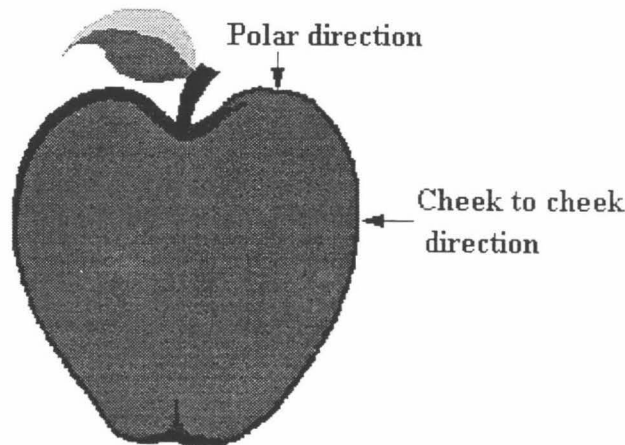


Figure 3-2 Apple direction definition

3.2.4 Fracture Strength

Fracture strength was measured as the force required to cause the break of the tissue subjected to three point bend test. A 12 mm diameter and 40 mm long plug was removed from each fruit in the equatorial direction with a cork borer, as shown in **Figure 3-3**. Plugs were cut with a razor blade to produce a 3 mm notch in the diameter in order to control the fracture position. The notch was cut in the edge closest to the

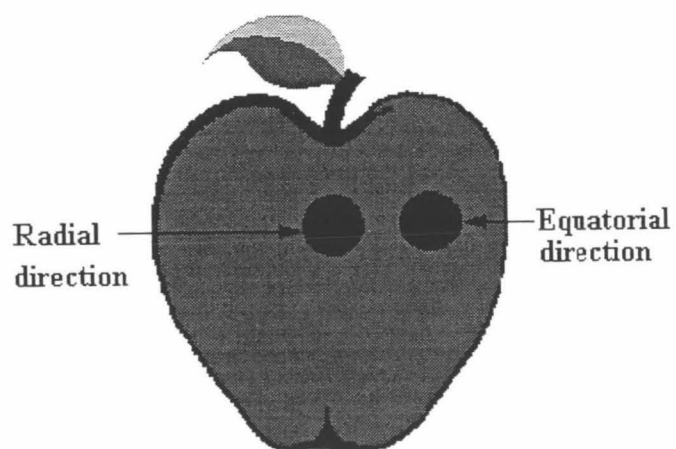


Figure 3-3 Apple direction definition

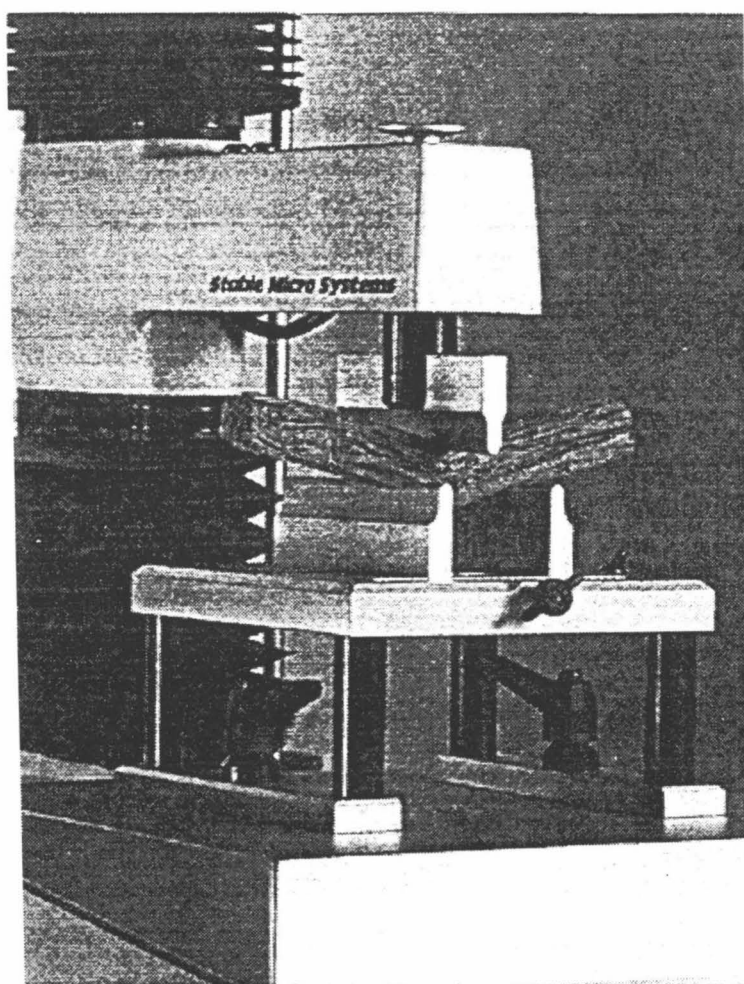


Figure 3-4 Three point bend rig.

core of the apple, and loaded so that the tensile stress occurred at this point in the three point bent test (ie. at the bottom in **Figure 3-4**). The TA-XT2 Texture analyser was used for the test. The sample was put on the two supports, which were mounted on the heavy duty platform. The third support, which was mounted on the probe location of the texture analyser, pressed the sample at 1 mm per second deformation rate until failure. The equipment diagram is shown in **Figure 3-4**. The maximum force was defined as the fracture strength. The results were recorded by the connected computer and transferred to Microsoft Excel for analysis.

3.2.5 Shear Gradient

Shear gradient was measured as the gradient of the force change with distance as a flat probe compressed a plug of tissue. A plug 12 mm in diameter and about 20 mm long was obtained from the apple radial direction using a cork borer (**Figure 3-3**). Measurement was carried on by TA-XT2 Texture Analyser. Plugs were put on the heavy duty platform, and the Jacobs Chuck probe, which was mounted on the TA-XT2 Texture Analyser probe location, compressed the specimen through the whole apple plug diameter at a constant speed of 1 mm per second. The testing diagram is shown in **Figure 3-5**.

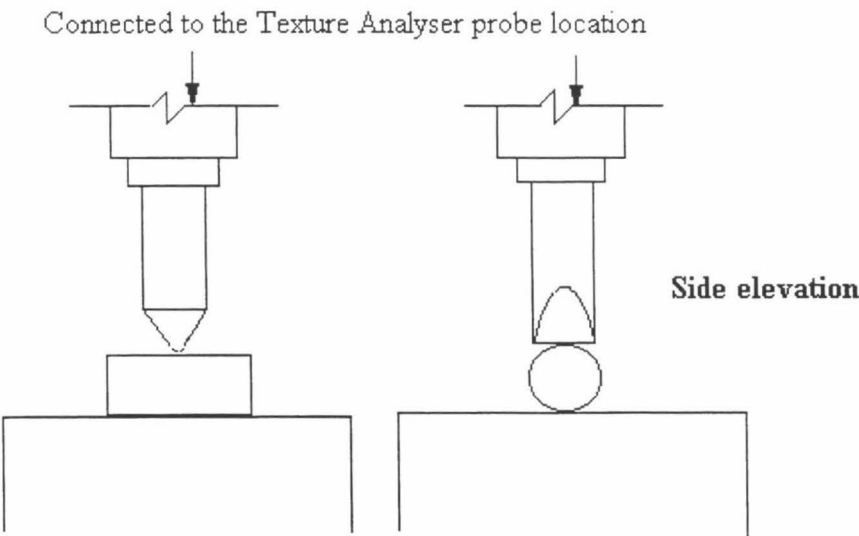


Figure 3-5 Jacobs Chuck for shear gradient test.

The force change with the compression of the apple plug was recorded automatically by the computer. The gradient of the force change from zero to the maximum value with the distance the probe moved was defined as the shear gradient. The results were recorded and calculated by running a macro, and is transferred to Microsoft Excel for analysis.

3.2.6 Compressive Energy

Compressive energy is defined as the area under the curve of force with distance. This was measured with TA-XT2 Texture Analyser. A 12 mm diameter plug was obtained from the apple radial direction with a cork borer, and was cut to 13 ± 1 mm height. The sample cylinder was put on the heavy duty platform vertically. The 35 mm diameter cylinder probe was used to press the sample to a maximum of 12 mm deformation at a constant speed of 1 mm per second. The area under the curve for the probe moving from 0 to 10 mm was taken as a measure of the compressive energy. The test results were recorded and processed automatically by running the TA-XT2 software, and data was transferred to Microsoft Excel for further analysis.

3.3 SENSORY TEST OF APPLE MEALINESS

Economical and practical constraints meant that it was not possible to establish a full taste panel to determine mealiness. Instead texture assessment test was conducted by the researcher and one volunteer to measure the fruit mealiness. The test scale was from 0 to 10, where 0 meant the fruit was fresh and not mealy, and 10 meant the fruit was very mealy. The test panel of two persons assessed the fruit mealiness one by one and determined the mealiness scale separately for every fruit. The final texture assessment value was taken as the average value of these two assessments.

3.4 EXPERIMENTAL PROCEDURE

Fruit stored at high temperature (20°C) were assessed every 7 days for the first two weeks. After two weeks of storage, fruit properties changed quickly with storage time,

and so fruit were assessed every 4 days. Due to the slower changes of physical properties in low temperature condition, the fruit stored at low temperature (0°C) were assessed every 16 days.

Before each assessment, test samples (20 apples from each treatment) were taken out of the storage condition and put in the test laboratory for 8 to 10 hours to reach 20°C, in order to avoid any effects of fruit temperature change during the testing period.

At every assessment, fruit density was measured first. The results were recorded manually. After measuring density, whole fruit elastic modulus was measured. The force and probe height of the TA-XT2 Texture Analyser were calibrated before measuring. Elastic moduli in two directions (one in a polar direction and the other in a cheek to cheek direction) on each apple were measured, as described above.

The first destructive test was fruit firmness. Next the Massey Twist Tester was used to measure the maximum twist strength, and then three kinds destructive measurements were carried out using the TA-XT2 Texture Analyser with different probes as described above (fracture strength, compressive energy and shear gradient).

After finishing all these objective tests for all fruit to be tested, the fruit were used for sensory mealiness assessment.

3.5 ANALYSIS OF EXPERIMENTAL DATA

Experimental data were analysed for variance, means and standard errors using the Statistical Analysis Systems (SAS) programmes (Cody and Smith, 1991). In order to investigate physical property changes with the development of mealiness, test results from all the treatments were put together, and were sorted by texture assessment values using SAS software. Means were compared using Duncan's Multiple Range Test. The mean value, the standard deviation, standard error and observation numbers were given. To get the mealiness indicator, multiple regression analysis was used to regress every physical property with sensory test, storage time and treatment.

CHAPTER FOUR

RESULTS

4.1 INTRODUCTION

This chapter presents the results of apple physical property changes under different storage conditions, including texture assessment tests and effects of storage conditions on the development of apple mealiness. Firstly the results showing changes in physical properties with time will be presented in this Chapter. In Chapter 5 the data will be analysed in relation to the mealiness assessment.

4.2 EFFECTS OF STORAGE TIME ON TEXTURE AND PHYSICAL PROPERTIES

Storage condition not only affected apple mechanical properties, but it also affected the change of stored apple appearance. Most low humidity stored apples were shrivelled after a period of storage, especially those at high temperature. However high humidity stored apples were never found to be shrivelled. The appearance of these apples did not show much difference from the high maturity fresh apples, which had a yellow background colour.

4.2.1 Apple Density Changes During Storage

Apple density changes during storage are shown in **Figure 4-1** and **4-2**. Apple density declined during storage at high temperature and both high and low humidities (HH and HL) by around 2% over 40 days. Storage at low temperature and low humidity (LL) had only a small effect on apple density. There were no density changes in LL (low temperature, low humidity) fruit in the first 60 days of storage, followed by a drop of around 0.7% after 130 days. LH (low temperature, high humidity) fruit density fell steadily from 40 days onwards, with a 2.6% drop after 130 day's storage.

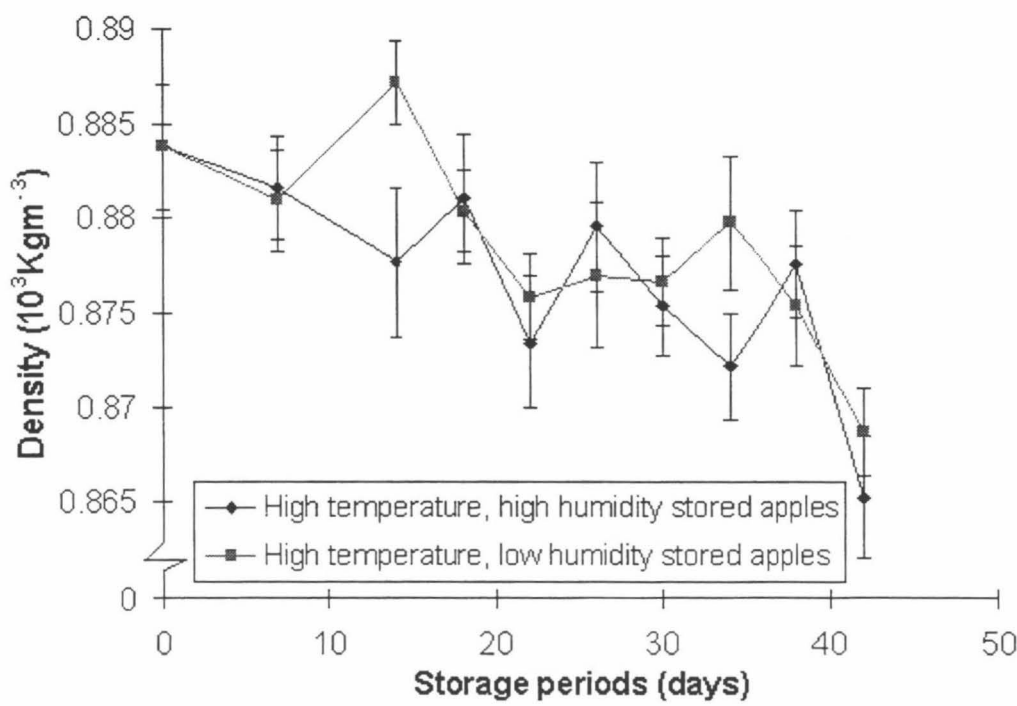


Figure 4-1 Density (whole fruit) changes during storage for high temperature stored apples. Vertical bars represent the standard errors of the means.

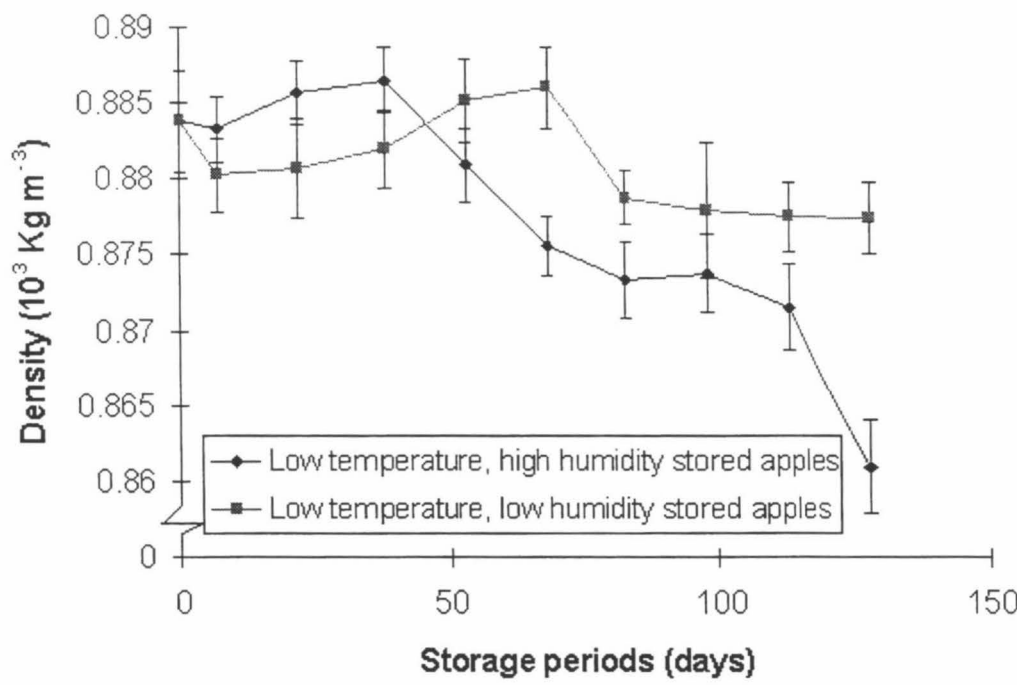


Figure 4-2 Density (whole fruit) changes during storage for low temperature stored apples. Vertical bars represent the standard errors of the means.

4.2.2 Apple Twist Strength Changes During Storage

Apple twist strength changes during storage are shown in **Figure 4-3** and 4-4. Twist strength decreased with storage time. Both temperature and humidity had effects on the this decline. High temperature decreased apple twist strength quickly. In 7 days twist strength decreased more than 200 kPa (around 20%) in high temperature conditions, but it decreased less than 185 kPa (around 15%) in 53 days in low temperature storage conditions. In the initial stage humidity had little effect on the change of apple twist strength, but later, high storage humidity (both HH and LH) reduced the apple twist strength further than for the low humidity (HL and LL) stored fruit. After 42 days high temperature storage, twist strength of HH fruit declined from 1220 kPa to 629 kPa, a decrease of 48%, but the value of HL fruit declined from the same initial value to 780 kPa, a decrease of 36.13% (**Figure 4-3**). Twist strength of LH fruit declined 28% in 128 days of storage, but the value of LL fruit retained the same value after an initial decline in the first 20 days storage (**Figure 4-4**).

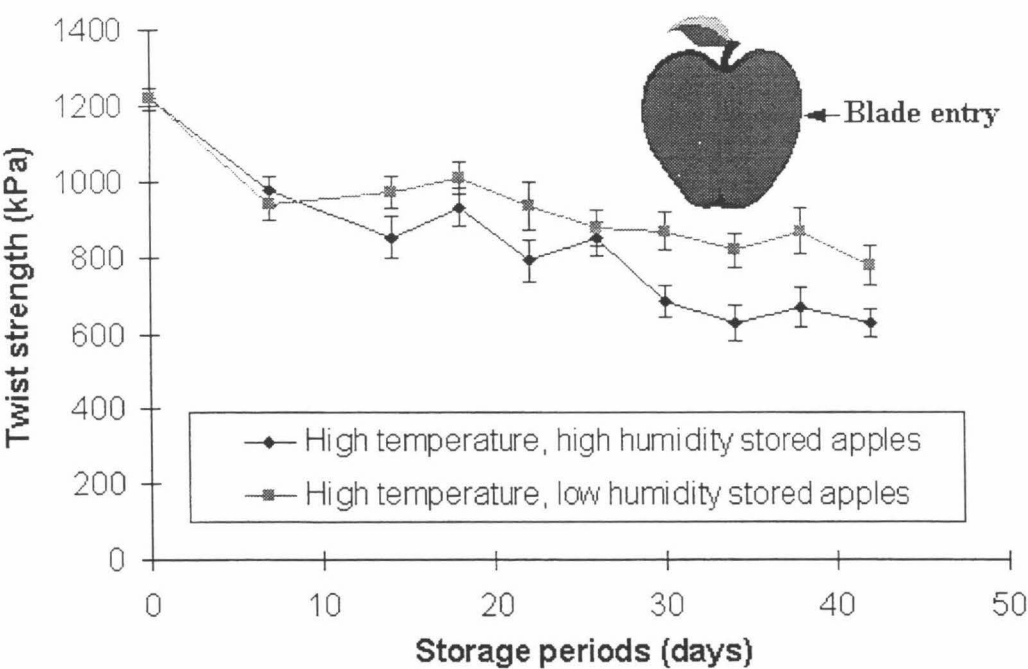


Figure 4-3 Twist strength (whole fruit) changes during storage for high temperature stored apples. Vertical bars represent the standard errors of the means.

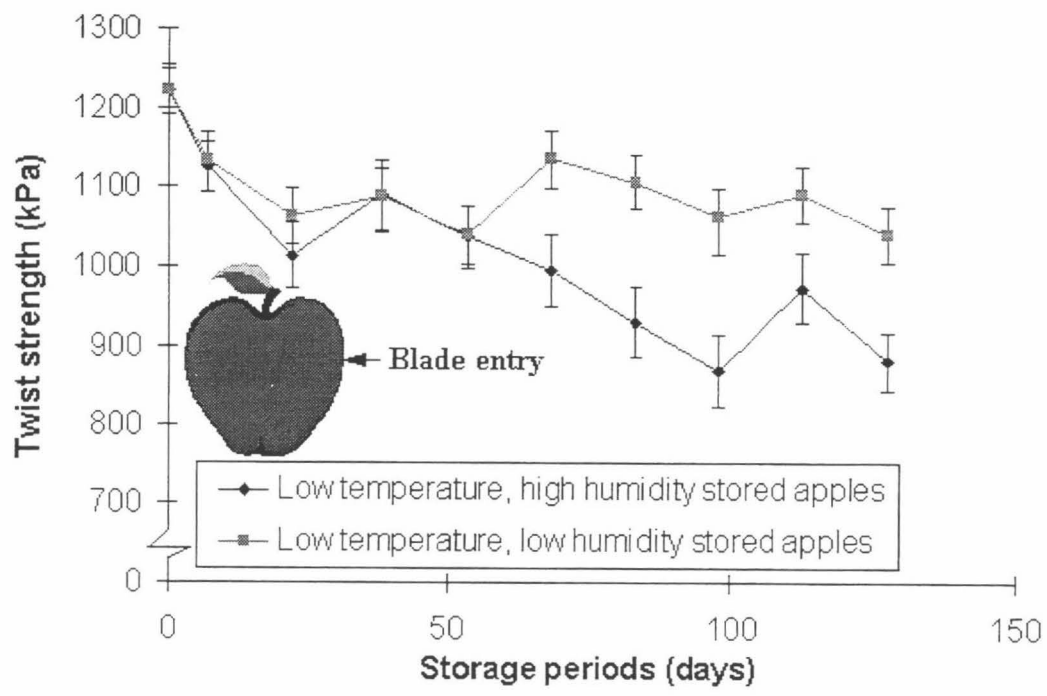


Figure 4-4 Twist strength (whole fruit) changes during storage for low temperature stored apples. Vertical bars represent the standard errors of the means.

4.2.3 Elastic Modulus Changes During Storage

Elastic modulus was measured by TA-XT2 Texture Analyser. Whole apple was put on the heavy duty platform. The 5 mm cylinder probe was used to compress the apple to 2% strain at a constant speed of 1 mm per second, then holding on that position for 1 second and unloading at 1 mm per second. **Figure 4-5** shows a typical loading curve for both fresh apple and apple which is both aged and had a mealy texture (aged and mealy apples). Compressive force increased nearly linearly with deformation. During the holding period, the apple compressive force slightly declined. Compared with fresh apples, aged and mealy apples had lower compressive force under the same strain (2%), and a lower loading gradient.

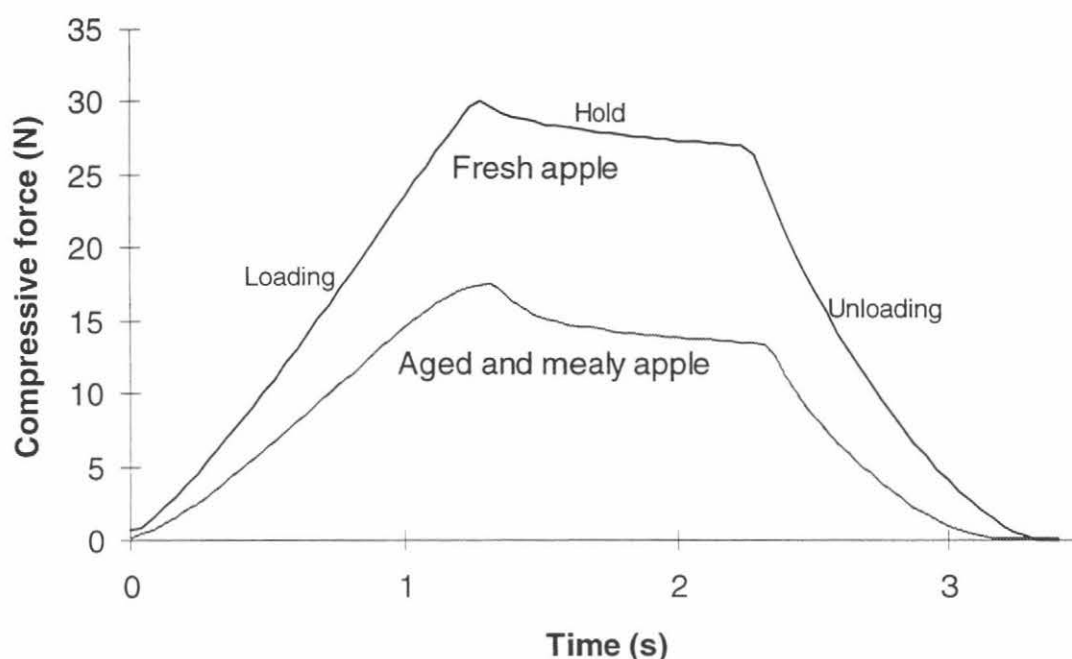


Figure 4-5 Typical force-time graphs for compression test on whole fresh and mealy apples.

For some of the tests the probe punched the fruit and caused damage, especially for fresh and high humidity stored apples. About 5 - 10% of fresh and high humidity stored apples were punched during the tests. For the test which fruit was punched, the maximum force was taken to calculate the elastic modulus.

Elastic modulus changes both in cheek to cheek and polar directions are shown in **Figure 4-6** and **4-7**. Both temperature and humidity had an effect on the rate of decrease of apple elastic modulus in cheek to cheek and polar directions. High temperature decreased elastic modulus quickly and to a lower value, as shown in **Figure 4-6** and **4-7**. Humidity had the greatest effect on the decline of the apple elastic modulus both in cheek to cheek and polar directions. Elastic modulus of low humidity stored apples declined much faster than that of high humidity stored apples. After 42 days storage, elastic modulus of HL stored apple declined 93% in P direction, and 77% in C direction. But values of HH stored apple declined about 34% in P direction, and 32% in C direction. HL stored apples began to shrivel after 14 days storage. Elastic modulus of LL stored apples declined about 26% both in P and C directions after 128 days storage.

But in the same storage period the values of LH stored apples declined only about half (13%) of that LL stored apples both in P and C directions. Shrivelled apples had a lower elastic modulus in both P and C directions. For high humidity stored apples, elastic modulus between cheek to cheek and polar directions were not significantly different ($P > 0.1$). For low humidity stored apples, statistical analysis showed that they were not significantly different for $P \leq 0.05$, but were significantly different under $P \leq 0.10$. Elastic modulus in cheek to cheek direction was a little higher than that in the polar direction.

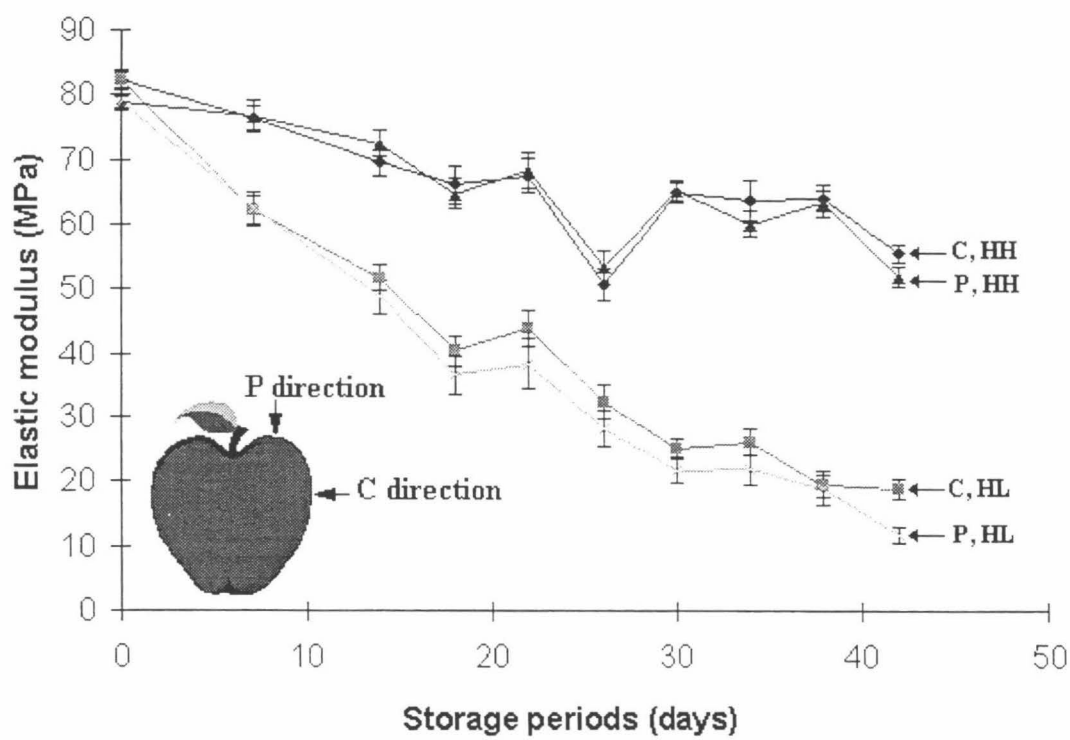


Figure 4-6 Elastic modulus (whole fruit) changes during storage of high temperature stored apples in both cheek to cheek and polar directions. Vertical bars represent the standard errors of the means.

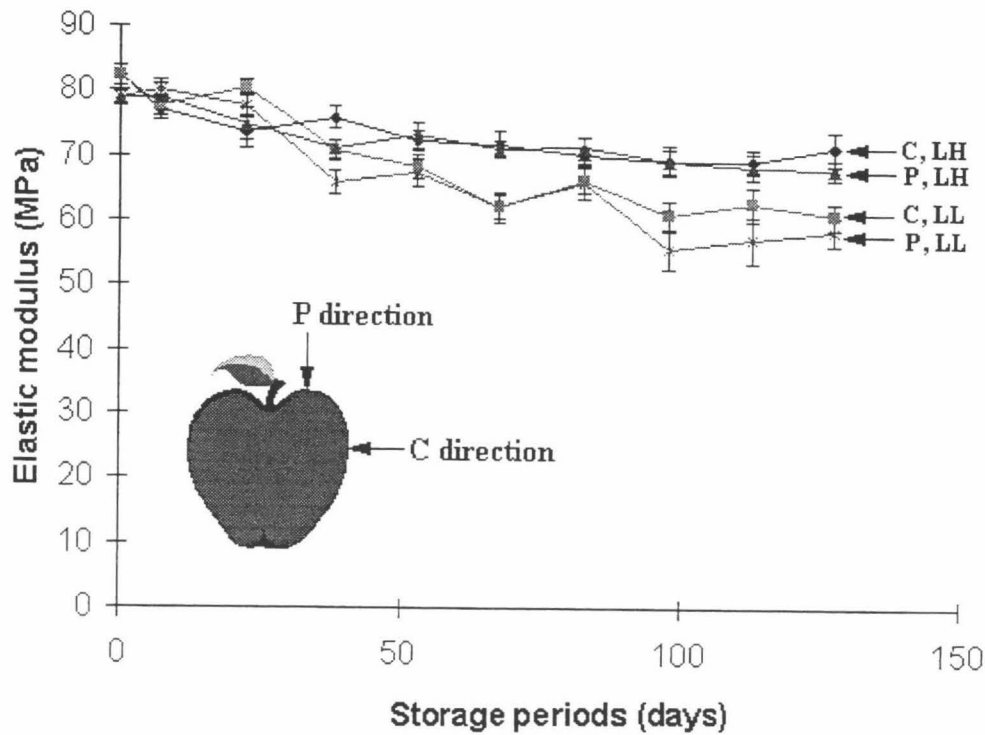


Figure 4-7 Elastic modulus (whole fruit) changes during storage of low temperature stored apples in both cheek to cheek and polar directions. Vertical bars represent the standard errors of the means.

4.2.4 Compressive Energy Changes with Storage Time

As discussed in the methods section, compressive energy was measured using 13×12 mm diameter samples, cut in the radial (R) direction. Samples were put between two flat plates and compressed at a constant speed of 1 mm per second. Typical loading curves for fresh and for aged, mealy apples are shown in **Figure 4-8**. As the figure shows, when the apples were fresh, the compressive force increased linearly with deformation. Failure occurred by a sudden and unstable collapse of just one or two layers of cells, usually in the middle of the specimen, at right angles to the applied force. This was accompanied by a sudden fall in the force reading. The cells around the collapsed zone remained undamaged. When the test continued, after a while a second layer of cells would collapse. Fracture was usually distributed throughout the entire width of the specimen. When specimens from mealy apples were compressed, force increased linearly but at a lower gradient than that of fresh apples. Failure strain was roughly the same but failure

stress was considerably lower, that is, the failure force was significantly lower than that for fresh apples. The force fell slowly as compression continued.

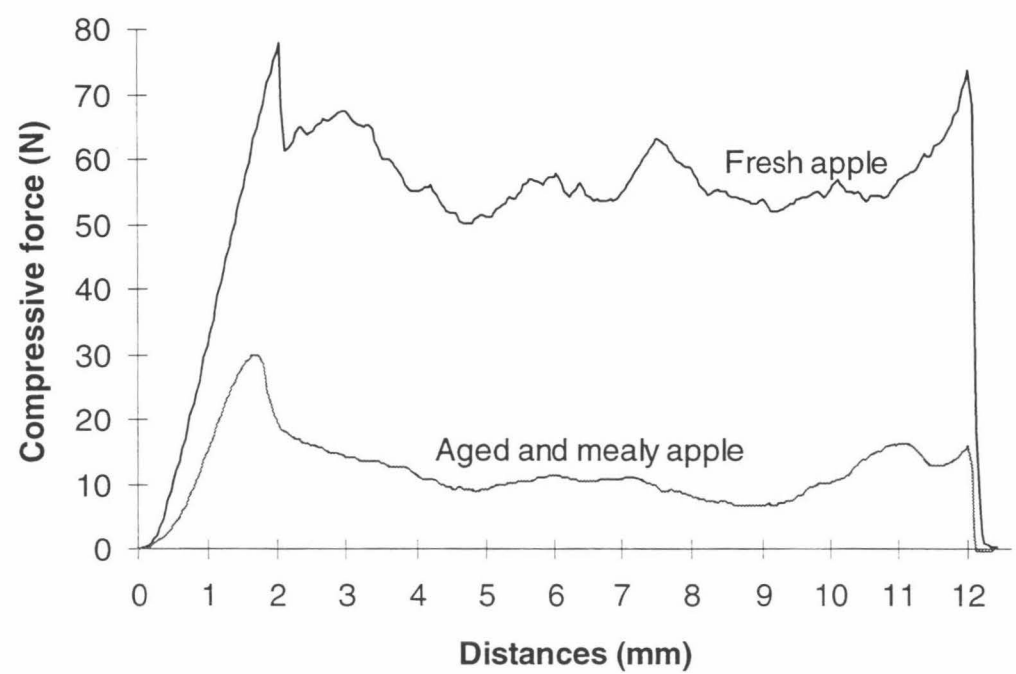


Figure 4-8 Typical force-distance graphs for compressive test on cylindrical samples of 13×12 mm diameter of fresh and of aged and mealy apple.

Figure 4-9 and **4-10** show the changes of compressive energy with storage time. Compressive energy declined with storage time for all four storage conditions. High temperature made the compressive energy value declined quickly. In 42 days it declined to less than half of its initial value. In low temperature conditions, compressive energy values reduced only about 1/3 in 128 days of storage time. Humidity had little effects on the changes of compressive energy as shown in **Figure 4-9** and **4-10**.

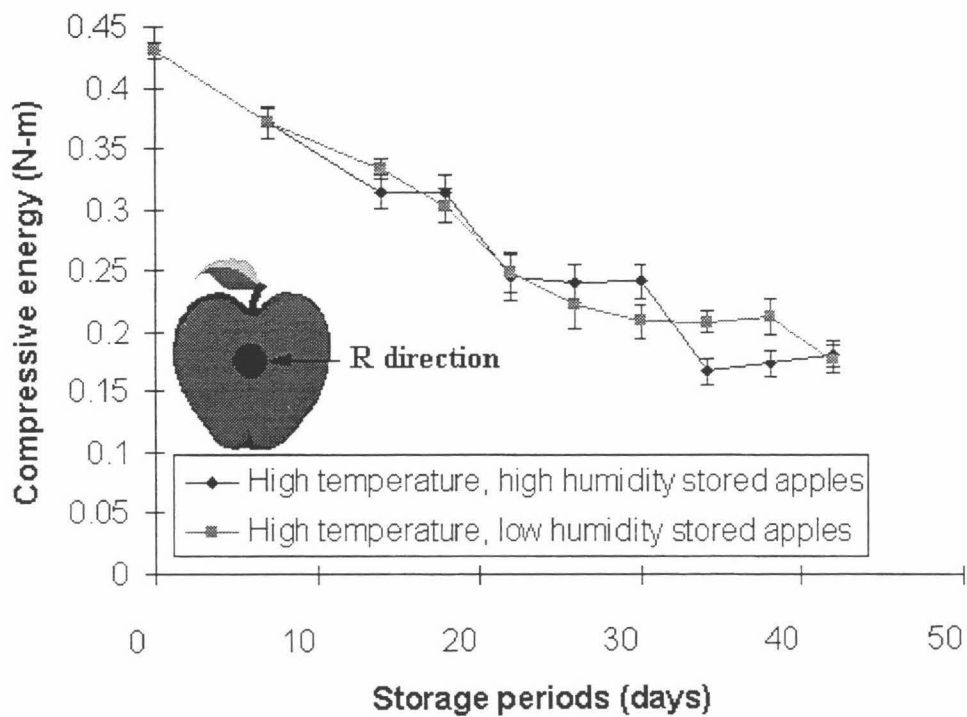


Figure 4-9 Compressive energy changes with storage time for high temperature stored apples. Vertical bars represent the standard errors of the means. Test samples were cylindrical of 12 mm in diameter and 13 ± 1 mm high cut from radial (R) direction. Energy to compress specimen by 10 mm was calculated.

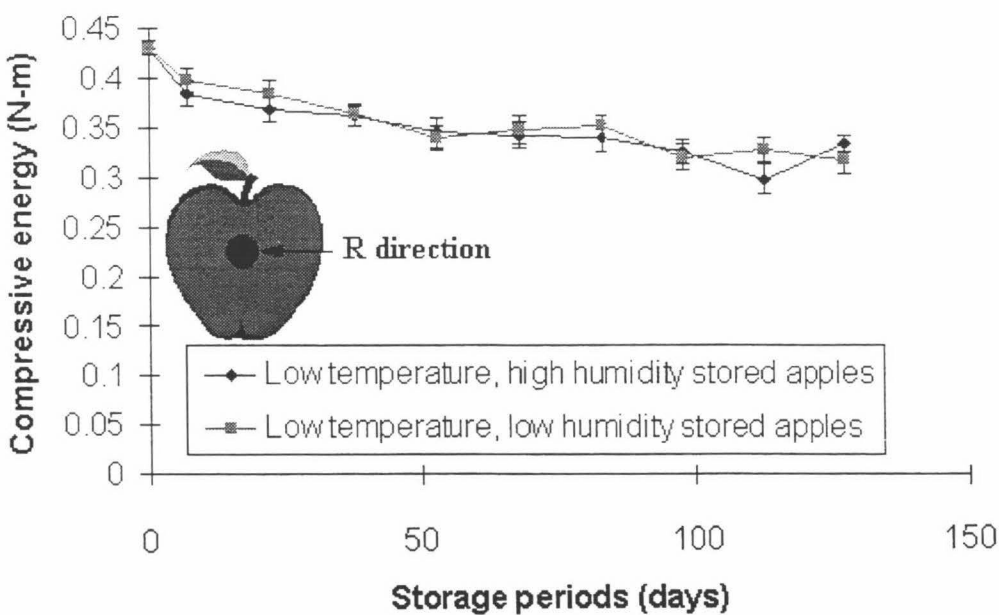


Figure 4-10 Compressive energy changes with storage time for low temperature stored apples. Vertical bars represent the standard errors of the means. Test samples were cylindrical of 12 mm in diameter and 13 ± 1 mm high cut from radial (R) direction. Energy to compress specimen by 10 mm was calculated.

4.2.5 Fracture Strength Changes During Storage

As described earlier (3.2.4), fracture strength was measured using 12 mm diameter samples cut from the apple equatorial (E) direction, with a 3 mm sharp notch, and then was put on the two supports. The third support pressed the plug sample at 1 mm per second deformation rate until failure at the notch. Typical loading curves of fracture tests for fresh and for aged and mealy apples are shown in **Figure 4-11**. The figure shows that bending force increased nearly linearly with deformation. Fracture occurred by a sudden collapse. Loading rates for aged, mealy apples were lower than that for fresh apples, but the deformation at failure was roughly the same. Failure modes were quite different between fresh apples and aged, mealy apples. Fresh apples had a smooth broken plane at the notched surface. Aged apples usually broke at the notched position with an irregular surface as shown schematically in **Figure 4-12**.

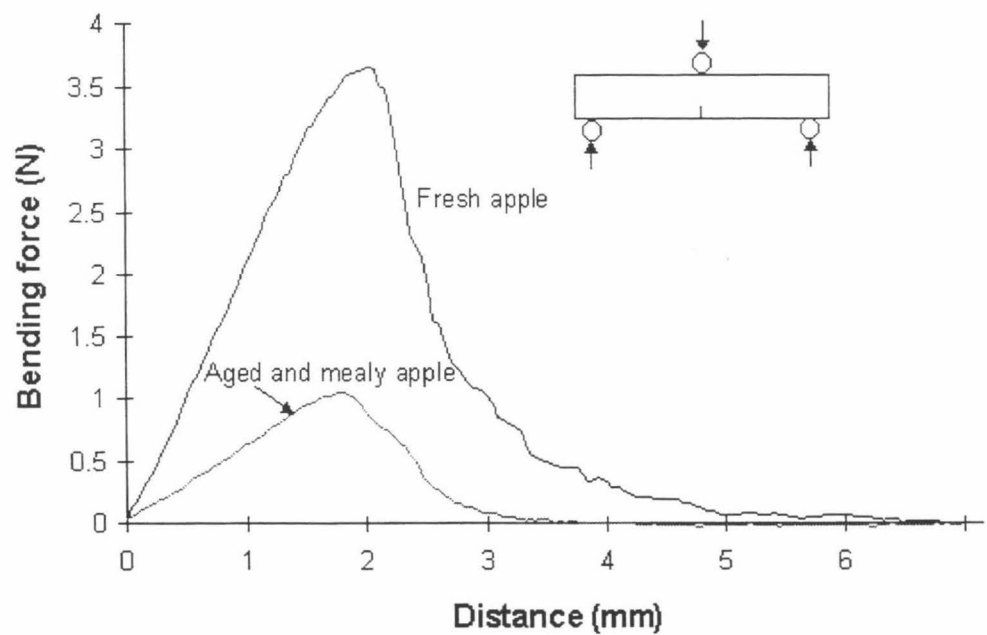


Figure 4-11 Typical bending force-distance graphs for three point bending test of fresh and of aged and mealy apples.

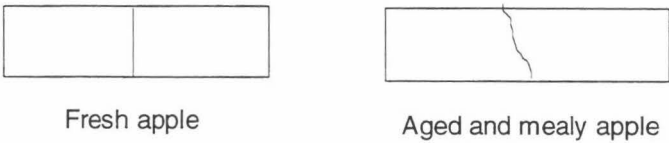


Figure 4-12 Typical specimen fracture failure mode for fresh and mealy apple.

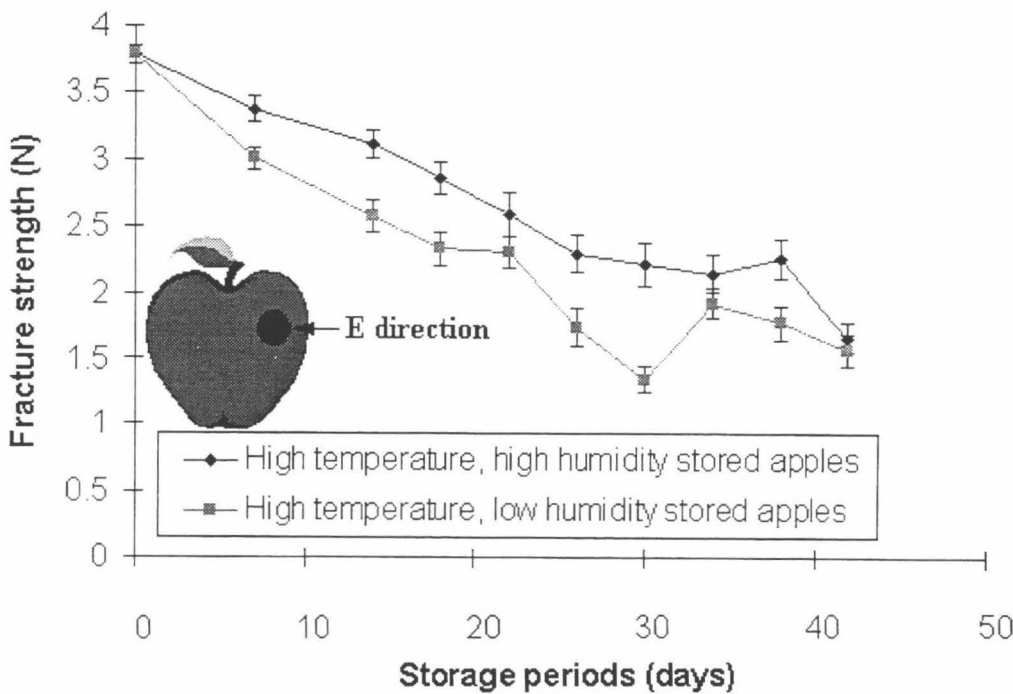


Figure 4-13 Fracture strength changes for high temperature stored apples. Vertical bars represent the standard errors of the means. Test samples were 12 mm diameter cylindrical cut from equatorial (E) direction.

Changes of fracture strength during storage are shown in **Figure 4-13** and 4-14. Temperature had a great effect on the decrease of apple fracture strength. As shown in **Figure 4-13** and 4-14, fracture strength declined more than 55% during 42 days of storage at high temperature, but fell by less than 30% during 128 days at low temperature. Humidity had different effects on high and low temperature stored apples. In HL apples fracture strength declined quickly as shown in **Figure 4-13**, but at low temperature humidity had little effect on the changes of fracture strength as shown in **Figure 4-14**.

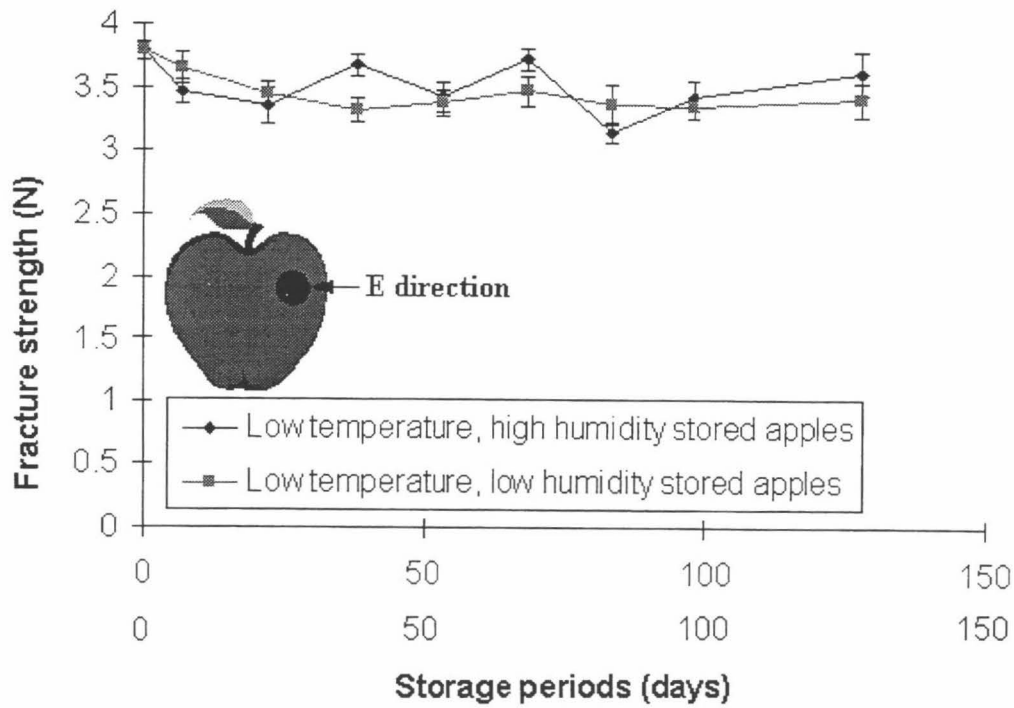


Figure 4-14 Fracture strength changes for low temperature stored apples. Vertical bars represent the standard errors of the means. Test samples were 12 mm diameter cylindrical cut from equatorial (E) direction.

4.2.6 Shear Gradient Changes During Storage

As described in Chapter 3 (3.2.5), shear gradient was measured using a 12 mm diameter specimen cut from apple in the radial (R) direction. A probe was pressed through the plug diameter at a constant speed of 1 mm per second till 10 mm. Typical loading curves for shear gradient test of fresh and of aged, mealy apples are shown in **Figure 4-15**. As shown in **Figure 4-15**, the failure strength of aged, mealy apples were lower than that of fresh apples. The failure distance moved by the probe before failure was greater in fresh apples than in aged, mealy apples. The failure surface was a flat plane for fresh apples, but not a plane for mealy, aged apples (as shown in **Figure 4-12**).

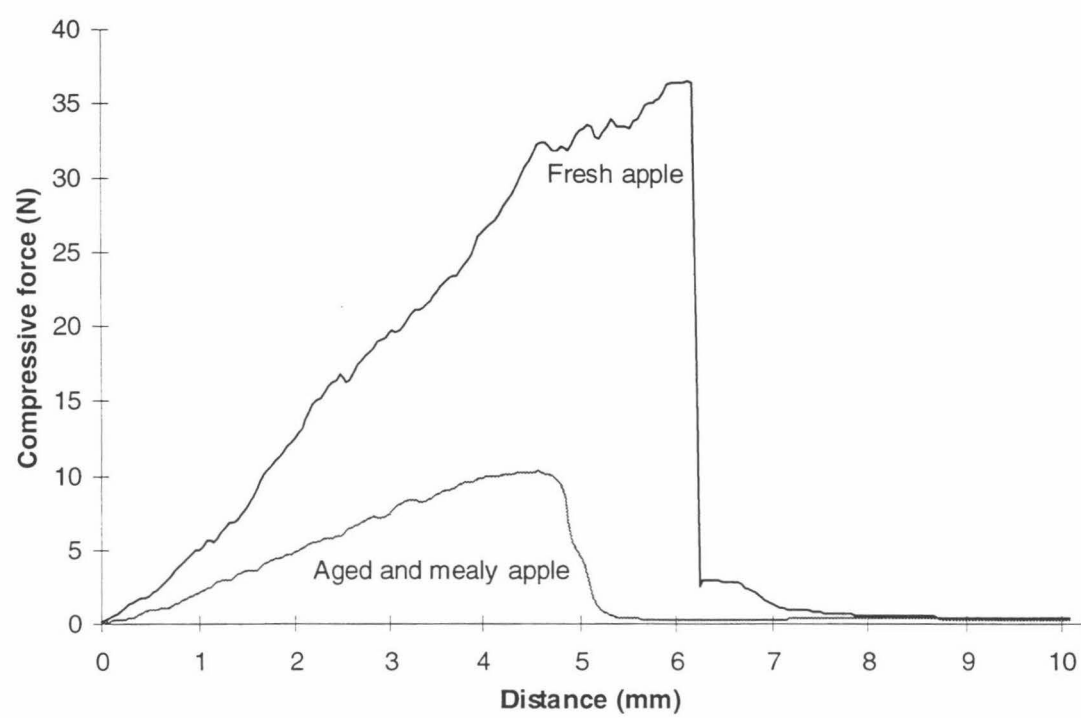


Figure 4-15 Typical compressive force-distance graphs for shear gradient test of fresh and of aged and mealy apples.

Shear gradient changes during storage are shown in **Figure 4-16** and 4-17. Shear gradient decreased during storage. Compared with low temperature storage, high temperatures made apple shear gradient decline faster. Shear gradient declined about 43% in 42 days storage for both HH and HL fruits. It only declined about 31% for LH fruit and 21% for LL fruit in 138 days of storage. For high temperature stored apples, humidity had no significant effects on the decrease of shear gradient as shown in **Figure 4-16**. However under low temperature condition, humidity had some effects on the change of apples shear gradient as shown in **Figure 4-17**. Shear gradients of LL fruits were significantly higher ($P \leq 0.05$) than that of LH apples.

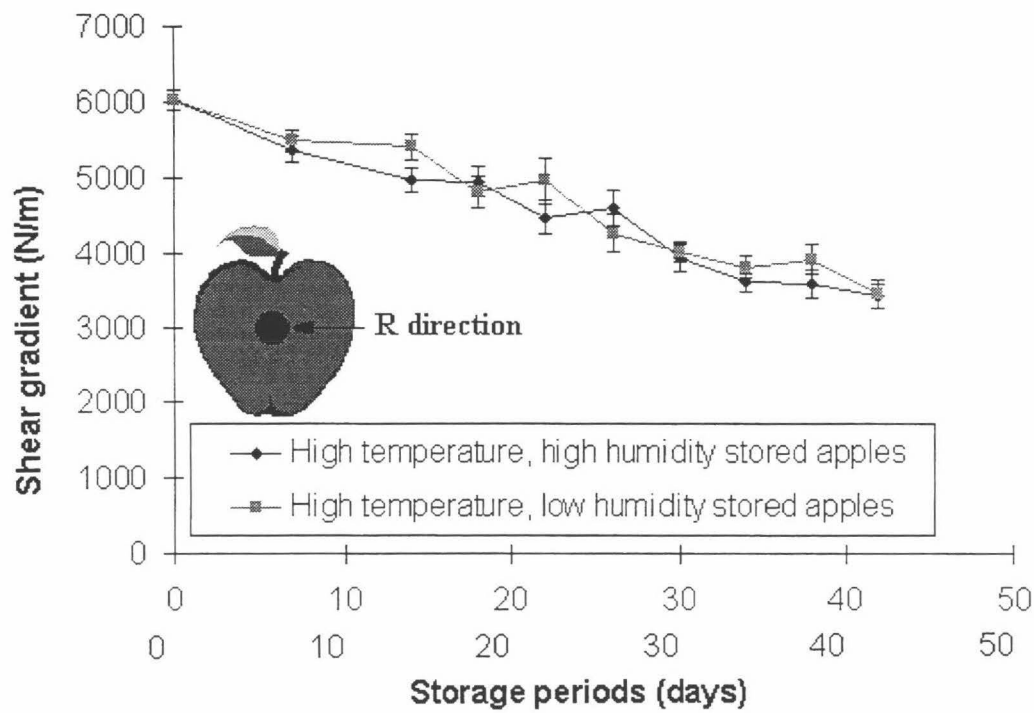


Figure 4-16 Shear gradient changes during storage for high temperature stored apples. Vertical bars represent the standard errors of the means. Test samples were 12 mm diameter cylindrical cut from radial (R) direction.

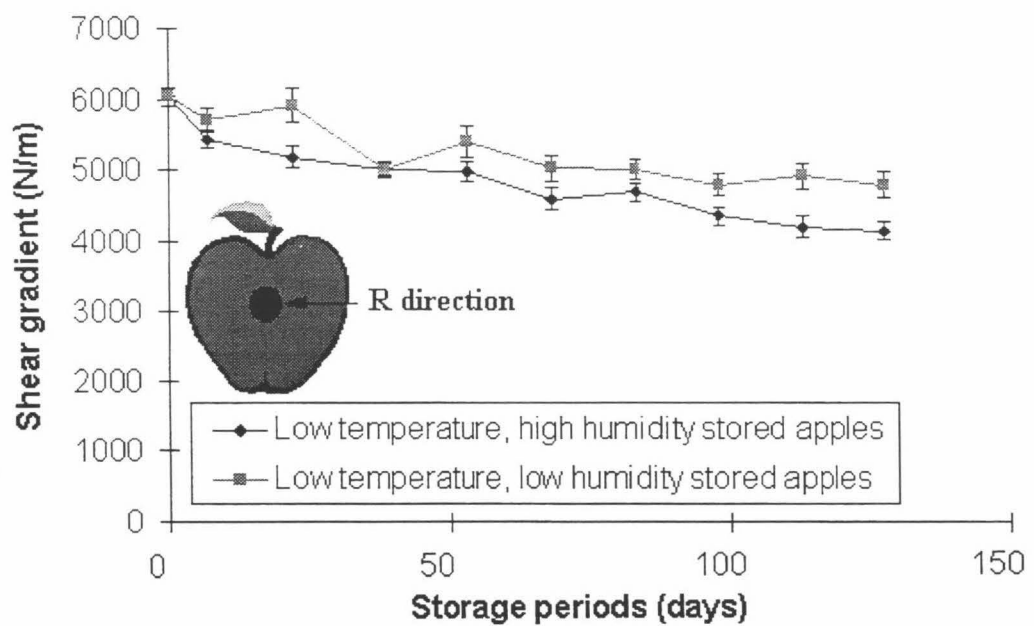


Figure 4-17 Shear gradient changes during storage for low temperature stored apples. Vertical bars represent the standard errors of the means. Test samples were 12 mm diameter cylindrical cut from radial (R) direction.

4.3 TEXTURE ASSESSMENT

As described in the last chapter (Chapter 3), a taste panel consisting of two persons assessed the texture value for every tested fruit. The assessment value was graded from 0 to 10, with 0 representing fresh, non-mealy apples, 10 representing very mealy apples. The assessment value differences between these two persons were shown in **Figure 4-18**. In general, the texture assessment values between these two test persons were not significantly different ($P>0.1$). As shown in **Figure 4-18**, taste differences for low temperature stored apples were less than for high temperature stored apples. The largest difference between two taste persons was for HL stored apples. The standard errors of this taste difference for different measurement are shown in **Figure 4-19**. The standard error values were from 0.04 to 0.26. The maximum standard error value (0.26) was for HL stored apples, and the minimum value was for LH stored apple.

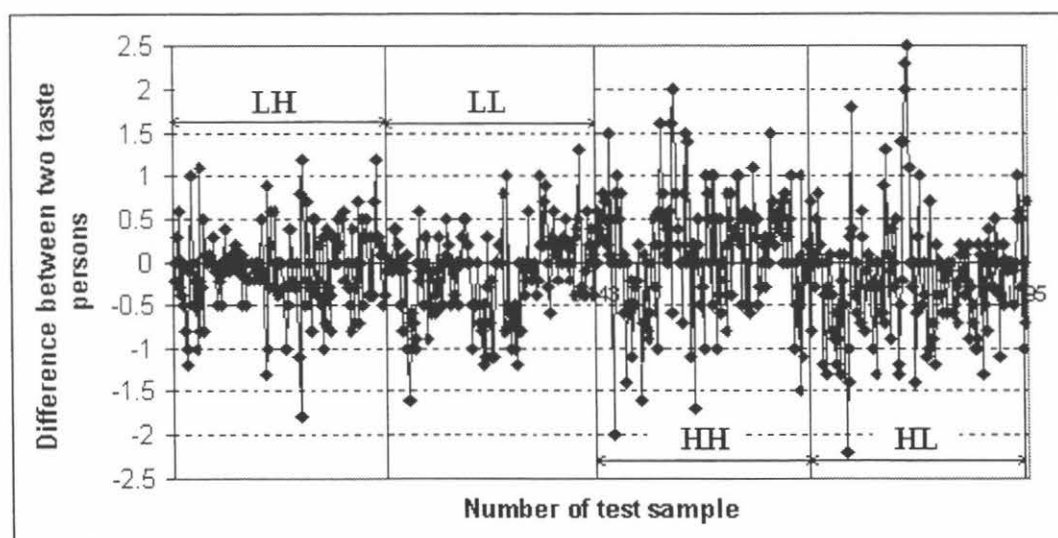


Figure 4-18 Texture assessment value difference between two persons. LH, LL, HH, HL represent four different treatments.

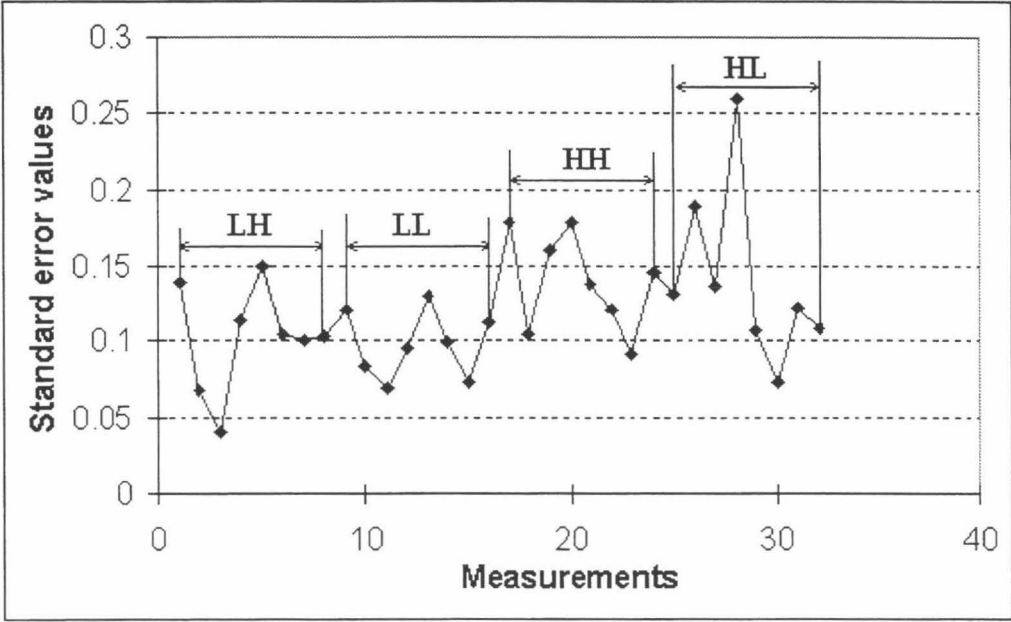


Figure 4-19 Standard error values for different measurement. LH, LL, HH, and HL represent four different treatments.

4.4 EFFECT OF STORAGE CONDITIONS ON THE DEVELOPMENT OF APPLE MEALINESS

As described in the last chapter (Chapter 3), four treatments were used to investigate the apple mealiness development. **Figure 4-20** shows apple mealiness development in different storage conditions. High storage temperature ($20 \pm 2^{\circ}\text{C}$) made the apple mealy quickly. High humidity made apple mealy to a higher extent both for high and low temperature stored fruits, as shown in **Figure 4-20**. All low humidity stored apple showed a shrivelled appearance in a different extent after a period of storage, especially under high temperature storage conditions. However high humidity stored apples were never found to be shrivelled. The appearance of these apples did not show much difference from the high maturity fresh apples, which has a yellow background colour.

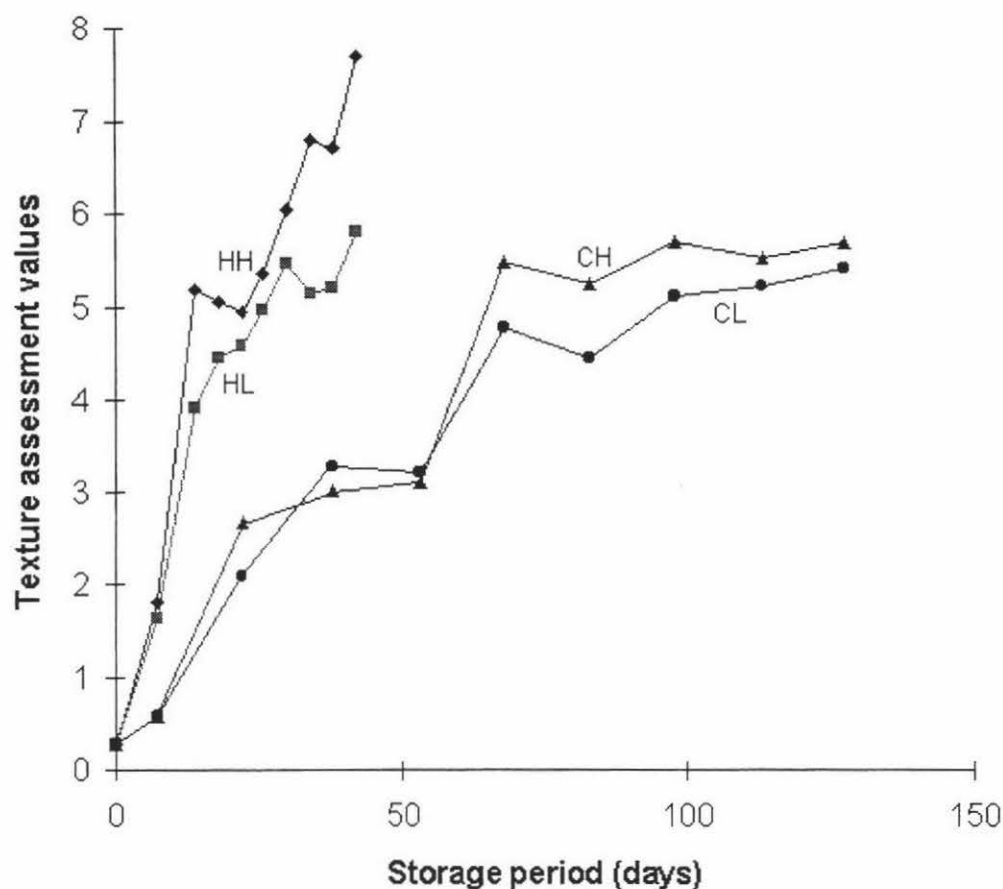


Figure 4-20 Effects of storage conditions on the development of apple mealiness.

HH = High temperature ($20 \pm 2^\circ\text{C}$), high humidity ($>90\%$ R.H.).

HL = High temperature ($20 \pm 2^\circ\text{C}$), low humidity ($50\% \pm 15\%$ R.H.).

LH = Low temperature (about 0°C), high humidity (about 99% R.H.).

LL = Low temperature (about 0°C), low humidity (about 92% R.H.).

Each point represents mean value of 20 measurements.

CHAPTER FIVE

ANALYSIS OF RESULTS

5.1 INTRODUCTION

In order to investigate physical property changes with the development of apple mealiness, the results from all four storage treatments were combined and sorted by texture assessment values (ie. mealiness scale) using SAS software. Texture assessment values were treated as integers, which means the texture assessment value of i represents a range of taste values of $(i-0.5 < i \leq i+0.5)$. The results are shown in Table 5-1. Nearly all of the fruits were in the range from 1 to 7, and most were between 3 and 6. Except for twist strength, all of the test parameters were not significantly different at 5% level for texture assessment values from 0 to 3. When the texture assessment value (i.e. mealiness scales) changed from 3 to 6, compressive energy, fracture strength and shear gradient decreased significantly ($P \leq 0.05$), while density and twist strength did not show significant differences. Modulus in both equatorial and polar directions declined and then increased again. These changes will be discussed in the next chapter.

5.2 APPLE DENSITY

Figure 5-1 shows density changes with the development of apple mealiness. There were no significant differences of apple density during the initial period of fruit mealiness development. When the fruit mealiness was significant, which means that the assessment values exceeded 4.0, apple densities declined progressively. When the fruit was quite mealy (texture assessment value exceeding 7.0) density values were more scattered, which is shown on the graph by the increasing size of the error bar.

Table 5-1. Sensory and physical properties distributions and mean values

Texture Assessment Values	Number of Fruits	Mean values						
		Compressive Energy (N-m)	Fracture Strength (N)	Modulus (E) (MPa)	Modulus (P) (MPa)	Shear Gradient (N/m)	Twist Strength (kPa)	Density (g/cm ³)
0	18	0.42810a	3.7785a	82.360a	79.837a	6060.6a	1213.15a	0.884850a
1	43	0.39420ab	3.5821a	77.780a	78.776a	5602.0ab	1136.34ab	0.881909a
2	64	0.38394ab	3.3094ab	73.202ab	72.173ab	5569.1ab	1005.74bc	0.882502a
3	93	0.35302bc	3.3787ab	70.016abc	68.725abc	5148.3b	1054.20bc	0.883342a
4	93	0.31788cd	2.8461bc	54.628d	52.192d	5059.1bc	1044.00bc	0.882981a
5	223	0.29105de	2.6944c	51.814d	49.605d	4542.8cd	978.26c	0.877970ab
6	137	0.25562e	2.6191c	59.423cd	58.230cd	4189.1de	821.07d	0.872725bc
7	38	0.19839f	2.3935c	56.916d	54.931cd	3640.1ef	594.93e	0.868053cd
8	20	0.19010f	1.8482d	62.293bcd	59.447bcd	3433.3f	580.19ef	0.867580cd
9	8	0.15452fg	1.4134de	56.523d	49.829d	3261.5fg	527.30ef	0.860000d
10	3	0.12758g	1.0072e	56.655d	57.679cd	2733.2g	450.86f	0.859933d

Means on each values followed by different letters are significantly different at $P \leq 0.05$.

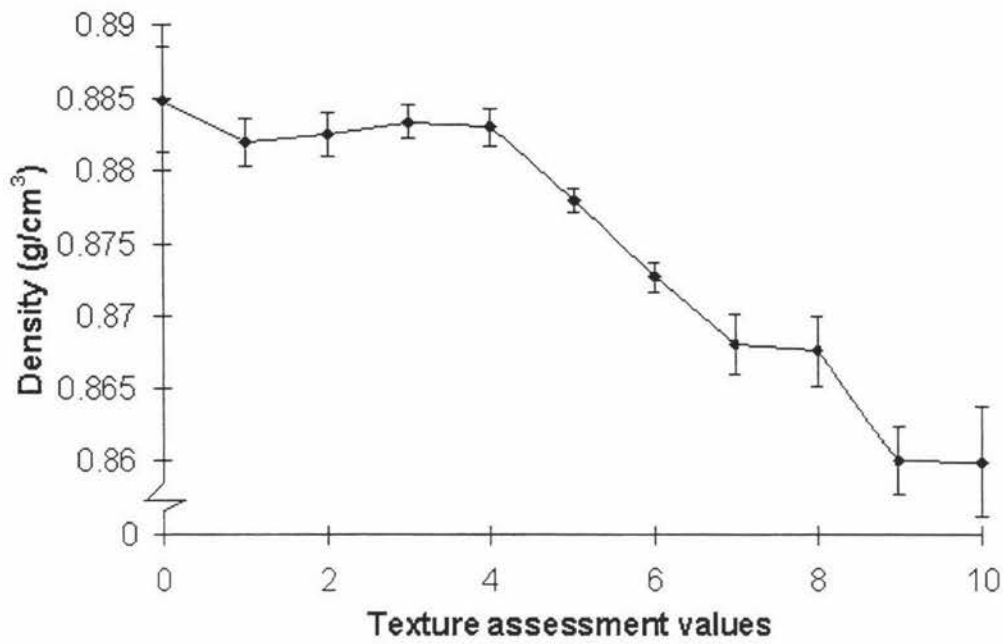


Figure 5-1 Apple density (whole apple) changes with the development of mealiness. Vertical bars represent the standard errors of the means.

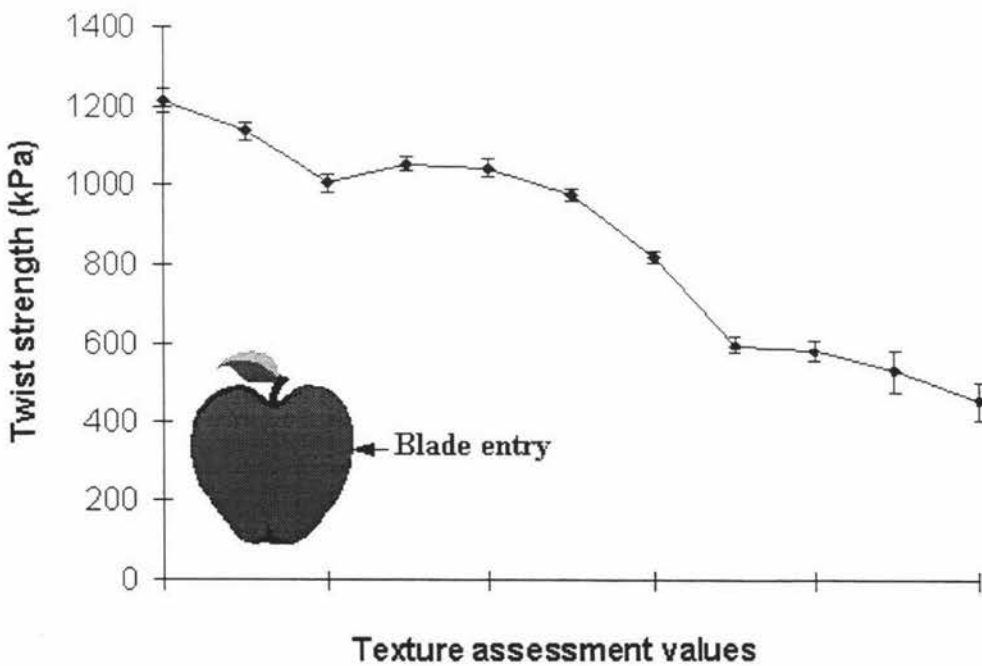


Figure 5-2 Twist strength (whole fruit) changes with the development of apple mealiness. Vertical bars represent the standard errors of the means.

5.3 TWIST STRENGTH

Figure 5-2 shows the twist strength changes with the development of apple mealiness. Twist strength declined with the development of apple mealiness. When apples became quite mealy (texture assessment values exceeding 7.0), the twist strength declined more than half (>51%) of the initial strength.

5.4 ELASTIC MODULUS

Figure 5-3 shows the changes of elastic modulus with the development of apple mealiness. Elastic modulus did not change markedly with the development of apple mealiness, and elastic modulus in both polar and cheek to cheek directions were similar. However during the experimental periods, elastic modulus of shrivelled apples were much lower than that of non-shrivelled apples.

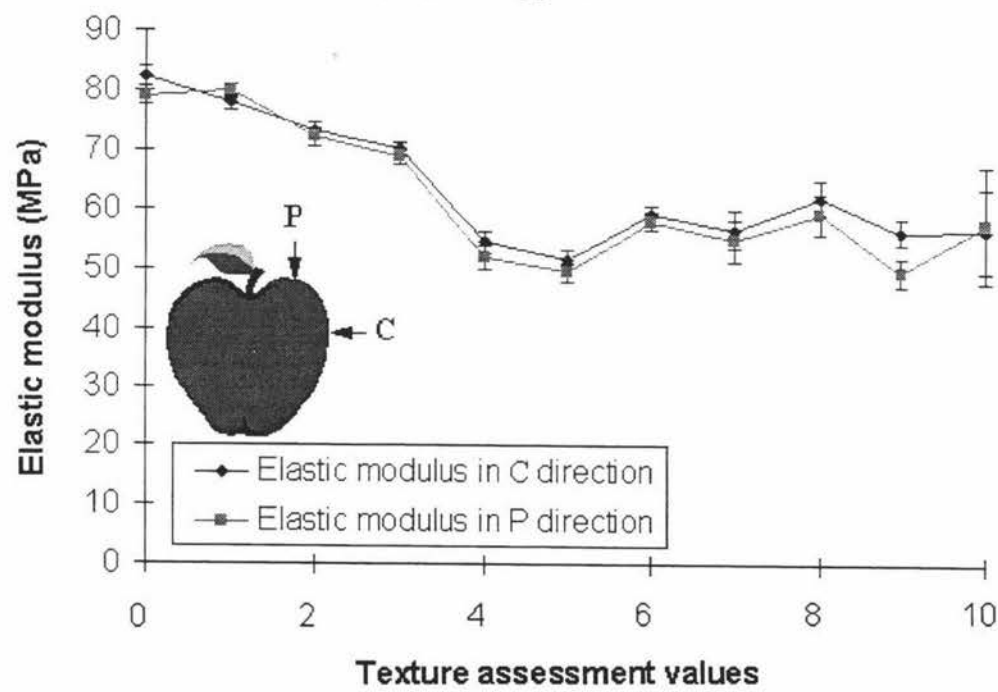


Figure 5-3 Changes of elastic modulus (whole fruit) with the development of apple mealiness. Vertical bars represent the standard errors of the means.

5.5 COMPRESSIVE ENERGY

Figure 5-4 shows the changes of compressive energy with the development of apple mealiness. As shown in **Figure 5-4**, with the development of apple mealiness, the required energy to compress the 13 mm high sample to 3mm declined. During the experimental period, the patterns of the sample breakdown were different between fresh and mealy apples.

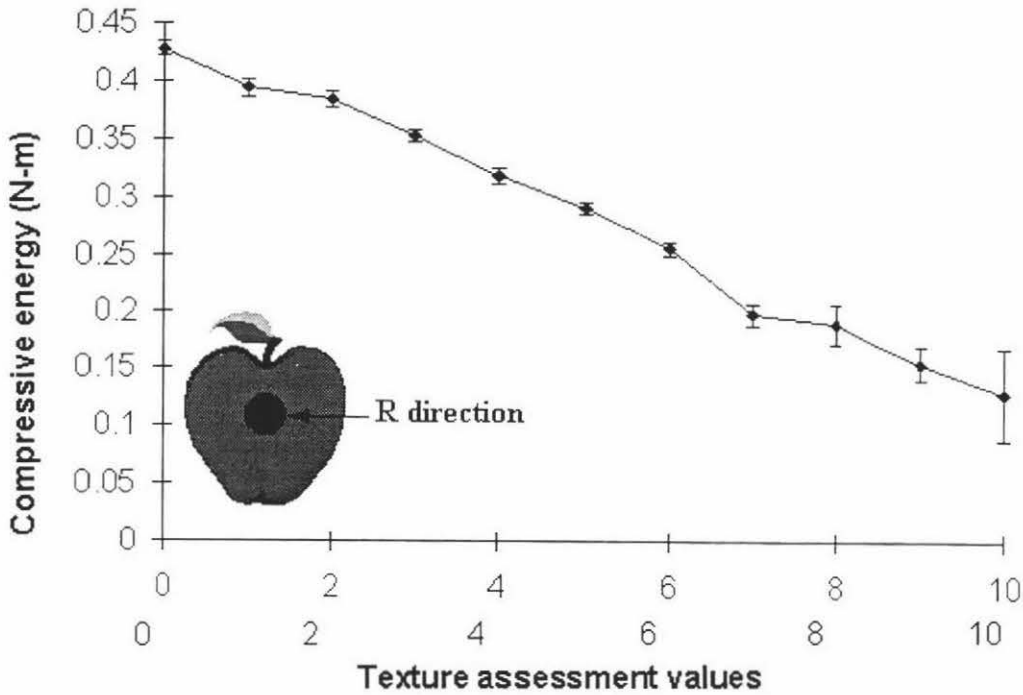


Figure 5-4 Changes of compressive energy with the development of apple mealiness. Vertical bars represent the standard errors of the means. Test samples were cylindrical of 12 mm in diameter and 13 ± 1 mm high cut from radial (R) direction. Energy to compress specimen by 10 mm was calculated.

5.6 FRACTURE STRENGTH

Figure 5-5 shows fracture strength changes with the development of apple mealiness. With the development of apple mealiness, fracture strength declined at first slowly and then progressively (mealiness values from 7 to 10).

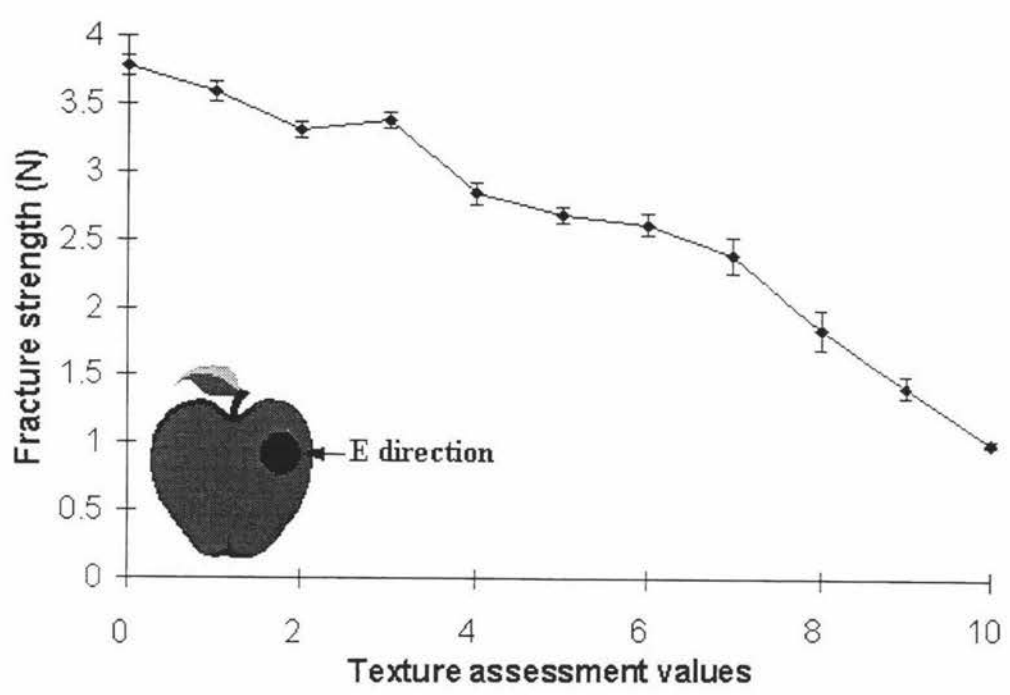


Figure 5-5 Fracture strength changes with the development of apple mealiness. Vertical bars indicate standard errors of the means. Test samples were 12 mm diameter cylindrical cut from equatorial (E) direction.

5.7 SHEAR GRADIENT

Figure 5-6 shows shear gradient changes with the development of apple mealiness. Shear gradient decreased nearly linearly with the development of apple mealiness. Shear failure distance decreased with the development of apple mealiness, as shown in **Figure 5-7**.

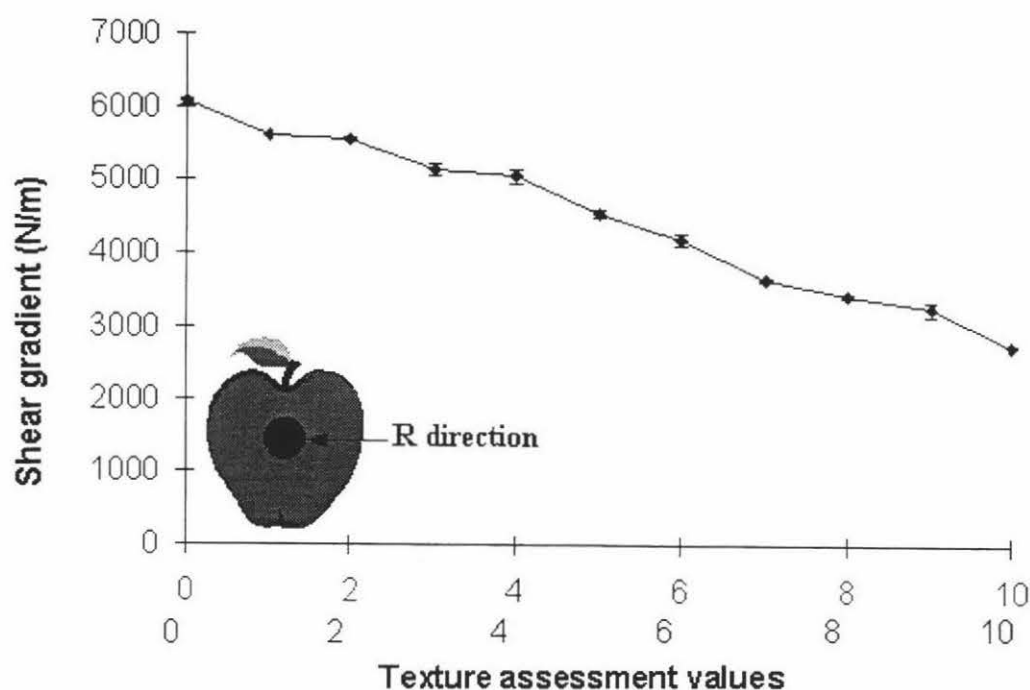


Figure 5-6 Shear gradient changes with the development of apple mealiness. Vertical bars indicate standard errors of the means. Test samples were 13×12 mm in diameter cylindrical cut from radial (R) direction.

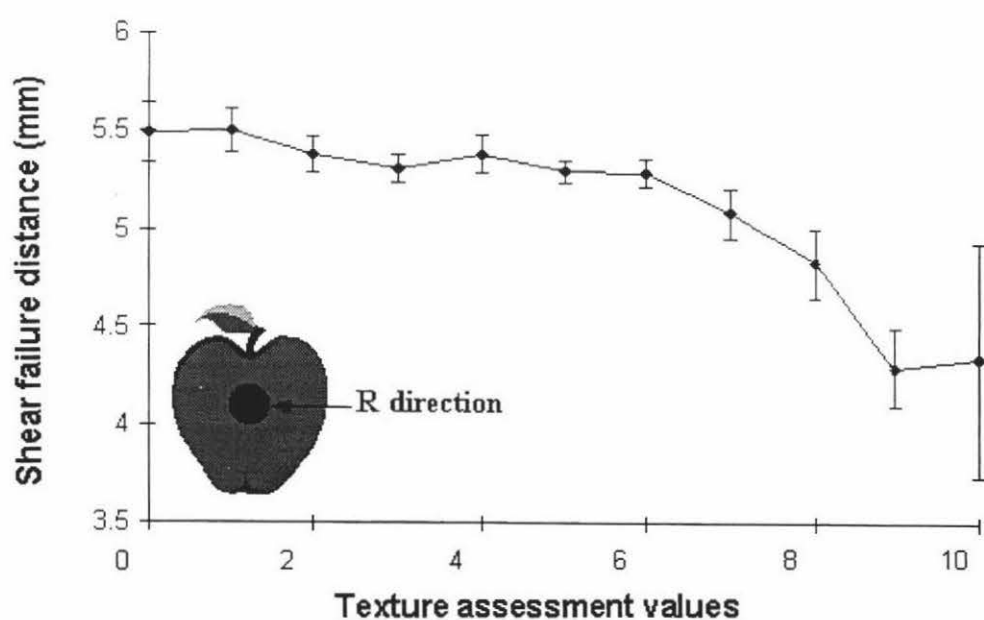


Figure 5-7 Shear failure distance change with the development of apple mealiness. Vertical bars indicate standard errors of the means. Test samples were 13×12 mm in diameter cylindrical cut from radial (R) direction.

5.8 REGRESSION RESULTS BETWEEN PHYSICAL PROPERTIES AND SENSORY TESTS

As discussed above, changes of apple physical property were affected by apple age, mealiness development and treatment. In order to separate the effects of different factors, physical properties were analysed by multiple regression of each individual fruit value from all of the treatments (total 740 tests). Linear regression was used to regress the results between sensory tests and physical properties. The regression results are shown in Table 5-2.

Table 5-2 Multiple regression results between taste assessment values and apple physical properties

Parameter	Analysis of Variance		Estimates of regression coefficient and its p-value							
	p-value	Adjusted R ²	Intercept	p-value	Treatment	p-value	Mealiness	p-value	Age	p-value
Density (g/cm ³)	0.0001	.01190	0.885519	0.0001	0.000608	0.2596	-0.001842	0.0001	-0.000029849	0.1537
Twist Strength (kPa)	0.0001	0.4352	1159.81775	0.0001	24.349575	0.0059	-79.624058	0.0001	1.695309	0.0001
Modulus (E) (MPa)	0.0001	0.2318	80.847428	0.0001	-1.134291	0.1692	-5.617589	0.0001	0.168991	0.0001
Modulus (P) (MPa)	0.0001	0.2146	80.740489	0.0001	-1.569446	0.0738	-5.813246	0.0001	0.173521	0.0001
Comp. Energ. (N-m)	0.0001	0.5731	0.420689	0.0001	0.004287	0.1460	-0.037340	0.0001	0.000802	0.0001
Fract. Strength (N)	0.0001	0.4528	3.677887	0.0001	0.030293	0.3307	-0.313049	0.0001	0.010705	0.0001
Shear Gradient (N/m)	0.0001	0.4318	5862.61298	0.0001	82.280941	0.0301	-316.81735	0.0001	0.729606	0.5764

CHAPTER SIX

DISCUSSION

6.1 INTRODUCTION

As shown in chapter 4 and chapter 5, some physical properties changed with the development of apple mealiness, and some did not. This relationship will be discussed in this chapter.

6.2 TEXTURE ASSESSMENT

As shown in **Figure 4-18**, the maximum texture assessment value difference on any apple was 2.5. **Figure 4-19** shows that the largest difference was for HL stored apples. This may due to the fact that apple softening had an effect on the assessment of mealiness. There were no significant differences ($P \leq 0.10$) between these two test persons overall. For a trained panel, there may be even less difference between persons for individual fruit.

6.3 THE EFFECT OF STORAGE CONDITIONS

Storage conditions have significant effects on the postharvest storage life and quality of fruit and vegetables. Results obtained in this study showed that although apple mealiness developed with storage time, it was also affected by storage conditions. Apple mealiness developed faster at high temperature than at low temperature storage. Apples were mealy in 30 days when held at about 20°C, compared with 68 days when held at 0°C. This is in accordance with Fisher's (1943) findings that Delicious apples were overripe and mealy in 5 to 6 weeks from picking when held at 15.5°C (60°F), and in 24 to 27 weeks when held at 0°C (32°F).

In addition to temperature effects, humidity also had great effects on apple mealiness development. High humidity made apples mealy quickly and to a high level (**Figure 4-20**). The same results were obtained by Scott *et al.* (1969) for peaches. Compared with high humidity storage condition, apples stored in low humidity were shrivelled after a period of storage, but mealy texture developed slowly and to a limited level. In commercial practice the quality control factor for low humidity stored apple is not the mealy texture but the shrivelled appearance.

As described by Harker and Hallett (1992), apple mealiness was associated with low adhesion between neighbouring cells, and a relatively high resistance to cell rupture. During postharvest storage, apple lost water and turgor pressure. Hatfield and Knee (1988) indicated that weight loss affected the cohesion of tissue, but did not affect the ease of cell breakage (to liberate juice). The slow and limited development of apple mealiness under low humidity storage conditions, as shown in **Figure 4-20**, supports the above statement.

6.4 PHYSICAL PROPERTY CHANGES

6.4.1 Apple Density

As described by Wilkinson (1965), the postharvest changes that occur in apples are both chemical and physical. Physical property changes such as those in fruit size, density and the permeability to gases are not necessarily related to senescence; they are connected with the structure of the fruit rather than with life processes. Nevertheless structural changes may indirectly affect the rates of chemical changes. Apple density was closely related to the fruit internal air space, which was recognised as a mealiness indicator by many researchers (Chapter 2). According to Hatfield and Knee (1988), internal air space can be calculated by the following formula:

$$\text{Internal Air Space} = 1 - \frac{\text{Apple Specific Density}}{\text{Specific Density of Apple Juice}}$$

The specific density of apple juice does not change much during the postharvest storage period (Hatfield and Knee, 1988). So the internal air space must increase with the

decrease of apple specific density. Wilkinson (1965) found that the weight of apples remained almost constant, whereas the volume of the fruit increased progressively as long as the apples remained intact during the storage period. The parenchyma of apples in store, especially at high humidity, tended to increase in permeability, indicating a further increase in the volume of air spaces, and a decrease in density, leading to a 'mealy' texture which was due to the almost complete separation of cells (Wilkinson, 1965).

As described in Chapter 2, mature apple parenchyma contain large intercellular spaces, up to 4000 μm in length and 100-200 μm in diameter, mostly filled with air, and clearly visible under a microscope (Reeve, 1953). The volume of the intercellular spaces has been estimated at about 20-25% (Sterling, 1963) or 27% (Bain and Robertson, 1951) for Granny Smith. As the fruit age, the volume fraction of air spaces increased (Teley, 1931) as cells grow and are pushed apart. In a very mealy apple this process can lead to complete separation of cells (Wilkinson, 1965). Some other studies (Hatfield and Knee, 1988; Vincent, 1989) have also demonstrated a relationship between large air space volumes and poor apple texture. Further studies (Harker and Hallett 1992; Tu *et al.* 1996; and Tu and Baerdemaeker 1996) reported that apple mealy texture was associated with high air space.

The results obtained in this study showed that the change of apple density depended on the storage humidity. As shown in **Figure 4-1** and **4-2**, density did not change greatly for low humidity stored apple, but declined more for high humidity stored apples. This change was related to the apple mealiness development in different storage conditions (**Figure 4-20**). This result may suggest that mealy apple density is lower, but density of aged but not quite mealy apples remained high values as shown in **Figure 4-1** and **4-2**. Further analysis of this results indicated that apple density did not change during the initial development of mealiness (**Figure 5-1**). This might suggest that at the first stage of apple mealiness development fruit internal air space remained almost constant. The main changes in this stage might be the pectin substances in the middle lamella of fruit cells. These changes made apples soften and cell connections weaken. With further development of apple mealiness, fruit density declined progressively (**Figure 5-1**). These

changes might indicate that apple internal air space increased quickly. This increase made apples mealy.

Multiple regression (**Table 5-2**) showed that density was related to the dependent variables: treatment, mealiness grade, and apple age (since the F was significant at $p = 0.0001$). Furthermore, only about 12% of the variation in density changes could be explained by these three parameters ($R^2 = 0.1190$). Further regression analysis showed that the regression coefficients for treatment and age were not statistically significantly different from zero ($p = 0.2596$ for treatment, and $p = 0.1537$ for age). It was concluded that mealiness development caused the decrease of apple density. However due to the variation in the density change, it was not a reliable mealiness predictor.

6.4.2 Twist Strength

Firmness has been used for assessing fruit maturity, storage behaviours and quality for many years. The Massey Twist Tester has been found to detect changes in physical properties, which other testers have failed to measure (Studman and Yuwana, 1992). In this study apple firmness was measured by the Massey Twist Tester, and the maximum crush strength of apple flesh was used to represent fruit firmness.

Apple firmness is closely related to the fruit ripening and senescence processes (Yuwana, 1991). Mealiness, as a storage disorder, is also related to the fruit physiological development. Mealy apples were often soft, so the firmness of mealy apples was much lower than that of fresh apples. Harker and Hallett (1992) measured apple firmness with a Effigi pressure tester, and found little difference between mealy and non-mealy apples. Results obtained in this study (**Figure 4-3** and **4-4**) showed that apple twist strength declined initially. After a period of storage (7 days for high temperature and 20 days for low temperature), twist strength had different change patterns between high humidity and low humidity storage conditions. Twist strength of high humidity stored apples declined with storage time, but twist strength of low humidity stored apples remained nearly constant. Further analysis of the results showed that apple firmness declined with the development of mealiness (**Figure 5-2**).

The multiple regression results (**Table 5-2**) showed that decrease of twist strength was related to the dependent variables: treatment, mealiness grade, and apple age (since the F was significant at $p = 0.0001$). About 44% of the variation in twist strength changes could be explained by these three parameters ($R^2 = 0.4352$). Further regression analysis showed that the regression coefficients for treatment, mealiness grade and age were statistically significantly different from zero ($p = 0.0059$ for treatment, 0.0001 for mealiness and 0.0001 for age). This indicated that twist strength change was not only affected by mealiness development, but was also affected by different treatments and apple age, so it is not a suitable mealiness indicator.

6.4.3 Elastic Modulus

As described by Plege (1987), most foods are known to be viscoelastic materials. “Viscoelastic” means that their mechanical behaviour is neither purely elastic nor purely viscous, but something in between that shares the properties of both. Apples are viscoelastic materials, which means that at a certain deformation the applied force will fall, as shown in **Figure 4-5**. This observed mechanical relaxation phenomenon is a result of molecular and structural reorientation. Apples and many other fruits and many plant materials are composed of fluid-filled cells. The stress level in a slowly deformed specimen of such materials is largely a result of hydrostatic pressure build-up. The pressure dissipation rate depends on the total resistance to the fluid outflow toward and through the specimen walls. This resistance is primarily determined by the density, porosity, and microstructure of the compressed solid matrix and the total length of the fluid path. On the other hand, elastic properties are more closely related to cell wall mechanical behaviours, particularly if measured at high loading rates.

Results obtained in this study showed that apple elastic modulus declined with storage time. The decrease of elastic modulus was affected both by storage temperature and storage humidity. As shown in **Figure 4-6** and **4-7**, elastic modulus in both cheek to cheek and polar directions were nearly the same and changed in the same way.

Further analysis of these results was shown in **Figure 5-3**. Elastic moduli in both polar and cheek to cheek directions were not well correlated with the changes of apple mealiness. Statistical analysis results showed that elastic moduli in cheek to cheek and in polar directions were not significantly different at the 0.05 significance level ($P \leq 0.05$). However they are significantly different at 0.1 significance level ($P \leq 0.10$). Elastic modulus in the cheek to cheek direction tends to be a little higher than that in polar direction, but the result was not conclusive.

Biological materials are commonly anisotropic, hence their mechanical properties differ according to the orientation in which it is tested (Khan and Vincent, 1993b). Khan and Vincent (1990) studied the anisotropy of apple parenchyma, and found that the apple cells were radially flattened. The inner cells became increasingly radially elongated and began to be organised into radial columns diverging from near the centre of the fruit towards the periphery. In the polar direction the cells appeared rounded and showed no orientations since the columns of cells were being viewed from one end. Due to this structure difference, when a force was applied in the cheek to cheek direction, the apple was compressed along the cell columns, the deformation resistance was higher in this direction. When the force was applied in polar direction, the apple cell was compressed in the right angle of the cell column, the deformation resistance in this direction is lower than that in column direction. The result in this study showed that compressive strength (elastic modulus) in cheek to cheek direction was higher than that in polar direction. This result supported Khan and Vincent (1990)'s conclusion.

The multiple regression results (**Table 5-2**) showed that the changes of elastic modulus both in E and P directions were related to the dependent variables: treatment, mealiness grade, age (since the F was significant at $p = 0.0001$). About 23% in E direction and 21% in P direction of the variation in elastic modulus changes could be explained by these three parameters ($R^2 = 0.2318$ for E direction and 0.2146 for P direction). Further regression analysis showed that the regression coefficient for treatment was not statistically significantly different from zero ($p = 0.1692$ for E direction and 0.0738 for P direction), hence mealiness development and apple storage age caused the change of apple elastic modulus. These results indicated that not only apple mealiness

development, but also apple age affected the change of elastic modulus. The low R^2 values indicate that elastic modulus was only weakly related to the treatments, apple age and mealiness development, and it is not a suitable apple mealiness indicator.

6.4.4 Compressive Energy

The compressive mechanical properties of fruit and vegetable parenchyma are related to the morphology of the material such as size, shape and orientation of cells and intercellular spaces. Apple has relatively large thin-walled, fluid-filled cells and in section it can be regarded as elongated polygons with their elongated walls lying radially (Khan and Vincent, 1990). It shows the type of failure common to large celled anisotropic foams (Gibson and Ashby, 1988). Described by Khan and Vincent (1993a), when a specimen of fresh apple was compressed in equatorial orientation it deformed along columns uniformly over its entire depth storing strain energy. The large amount of wall material orientated in the direction of the force offered greater resistance to deformation and hence stiffness in this direction. Microscopic examination of the fracture specimens showed that failure was due to fracture of the elongated cell-walls lying in the direction of the force (Khan and Vincent, 1993b).

The loading curve for compressive test was shown in **Figure 4-8**. It showed that force increased proportionately to the deformation until the point of failure. Tissue failed under normal stress. At failure the stored energy was suddenly released and fed into a single layer of cells, usually from the top of the specimen. The sample collapsed in a plane stretching across the entire area of the specimen at right angle to the force. This resulted in a sudden fall in the record force. If the test was continued a second layer collapsed independently of the first one. When a load was placed on the specimen, force will act along the cell columns and layers of cells collapsed in succession. This is in agreement with the finding of Khan and Vincent (1993a).

Results obtained in this study showed that storage temperature had a great effect on the decline of apple compressive energy, but humidity, although an important factor to mealiness development, had very little effect on it (**Figure 4-9** and **4-10**). This results

may suggest that compressive energy decreased with the progressing of apple ripening and senescence processes. It also decreased with the development of apple mealiness (**Figure 5-4**).

The multiple regression results (**Table 5-2**) showed that the decrease of compressive energy was related to the dependent variables: treatment, mealiness grade, and apple age (since the F was significant at $p = 0.0001$). About 57% of the variation in compressive energy changes could be explained by these three parameters ($R^2 = 0.5731$). Further regression analysis showed that the regression coefficient for treatment was not statistically significantly different from zero ($p = 0.1406$). Thus, both apple mealiness development and age caused the decrease of compressive energy. This indicated that change of compressive energy was not only affected by mealiness development, but it was also equally affected by apple age, so it is not a suitable mealiness indicator.

6.4.5 Fracture Strength

Fracture strength is the maximum force needed to break the specimen. This mechanical property changed with storage time.

The results obtained in this study showed that fracture strength declined with storage time for high temperature stored apples, but decreased much less for low temperature stored apples. This may suggest that the main factor affecting apple fracture strength is the storage temperature. Humidity had some effects on the change of fracture strength for high temperature stored apple, but had very little effect for low temperature stored apples (**Figure 4-13** and **4-14**). Further analysis showed that with the development of apple mealiness, the fracture strength declined linearly as shown in **Figure 5-5**. The force changes during three point tests for fresh apple and mealy apple are shown in **Figure 4-11**. Fracture strength of fresh apples was much higher (nearly three times) than that of mealy apple. The crack path followed a straight line to form a flat plane in fresh fruit, but in mealy, aged fruit the fracture surface was not straight or perpendicular (**Figure 4-12**). Different fracture characteristics were due to the physical and chemical

changes occurring in the apple cells and cell walls. In fresh apples, cell adhesion was strong and tissue fracture involves rupture through the cell walls, so the fracture surface was in one plane. Failure through the cells released the cell contents, resulting in juicy fruit. Fresh apples also had a high level of turgor pressure, which would result in such tissue being perceived as crisp. In storage fruit lost its firm texture and softened. According to Waldron *et al.* (1997), this softening was usually the consequence of the dissolution of wall polymers, many of which were involved in cell adhesion. In highly soft tissue such as over-ripe, mealy apples, the cells became completely separated. So when mealy apple specimens were compressed, the force holding the cells together were weaker than the cell walls. Failure happened mainly by cell separation, and there were very few cell breakages (depending on the mealiness development stage), less juice was released, and the fruit tasted dry and mealy. The failure surface showed characteristics of shear breakdown.

It is agreed that for aged or shrivelled but not mealy apples, both the strengths of cell cohesion and the strength of cell walls were reduced with storage time. The failure of these specimens may be due to both cell separation and cell breakage. Although the fracture strength was not significantly higher ($P \leq 0.05$), the tissue did not taste as dry as mealy apples. However, the fracture surface appearance was the same as mealy apples, indicating that some broken cell failure was involved.

The multiple regression results (**Table 5-2**) showed that decrease of fracture strength was related to the dependent variables: treatment, mealiness grade, age (since the F was significant at $p = 0.0001$). About 45% of the variation in fracture strength changes could be explained by these three parameters ($R^2 = 0.4528$). Further regression analysis showed that the regression coefficient for treatment was not statistically significantly different from zero ($p = 0.3307$). Thus both apple mealiness development and age caused the decrease of fracture strength. This indicated that change of fracture strength was not only affected by mealiness development, it was also equally affected by apple age, so it is not a suitable mealiness indicator.

6.4.6 Shear Gradient

Shear gradient was defined as the gradient of force change with distance the probe moved into the specimen. As shown in **Figure 4-16** and **4-17**, shear gradient decreased with storage time. The change of this parameter was mainly affected by temperature. Storage humidity had some effect for low temperature stored apples (**Figure 4-17**), but had no effect for high temperature stored apples (**Figure 4-16**).

The typical loading curve for fresh and mealy apples was shown in **Figure 4-15**. For fresh apples, the shear failure was clearly cleavage, with the tissue acting very much as a brittle material. The associated force-deformation curve exhibited marked episodic failure. These discontinuities probably coincided with the collapse of single bands of cells. As apples became aged and mealy the appearance of the shear failure surface changed from plane to not plane as shown in **Figure 4-12**. This may be evidence of slip (shear) failures appeared in increasing proportion. The force-deformation curve showed fewer discontinuities and curve generally smooths.

This force change and failure mode of shear testing were another demonstration of cell wall and cell middle lamella changes with the development of apple mealiness. When apple was fresh, cell wall and cell connection strength were both strong. The shear force needed to break the specimen was high, and the distance the probe moved into the specimen to break it was longer. Breakage failure involved mostly cell wall rupture. With the progress of apple mealiness, the force holding the cells together decreased markedly. The compressive force needed to break the specimen was much lower. Due to the great decrease of cell connection, failure happened mainly by cell separation. Very few cell contents were released, the fruit was dry and mealy. The distance the probe moved into the specimen to break it decreased steadily as shown in **Figure (5-7)**.

For aged or shrivelled but not mealy apples, the compressive force needed to break the specimen was also lower due to the reduction of cell connection and cell wall strength (**Figure 4-16** and **4-17**). The failure still contained some parts of cell breakage and also

parts of cell separation (Khan and Vincent, 1993a). The tissue did not taste as dry as that classified mealy, although it did not taste as juicy as that of fresh apples.

The multiple regression results (**Table 5-2**) showed that decrease of shear gradient was related to the dependent variables: treatment, mealiness grade, and apple age (since the F was significant at $p = 0.0001$). About 43% of the variation in compressive energy changes could be explained by these three parameters ($R^2 = 0.4318$). Further regression analysis showed that the regression coefficient for apple age effect was not statistically significantly different from zero ($p = 0.5764$). Thus that both apple mealiness development and different treatments caused the decrease of fracture strength. Because different treatments would cause different mealiness development, this result may indicate that change of fracture strength was only affected by mealiness development. It may be a suitable mealiness indicator. However, the regression coefficient was fairly low (0.4318), and further research work needs to be done to prove this result.

6.5 RESEARCH LIMITATION

Due to resources and financial limitation, standard taste panels were not established. Texture assessment was conducted by only two persons. This may affect the reliability and accuracy of the subjective test results. However, the two independent assessors' results were usually very similar, indicating that the mealiness parameter had some validity.

CHAPTER SEVEN

CONCLUSIONS

The primary aim of this study was to find a mechanical test indicator for fruit mealiness. Research carried out was focused on the investigation of the physical property changes with the development of apple mealiness, especially mechanical properties. Several physical properties such as density, twist strength, elastic modulus, compressive energy, fracture strength and shear gradient were studied. It has been shown that some mechanical property changes were related to the development of apple mealiness development, but some did not. Storage conditions affected the mealiness development. Research conclusions from this study are:

High temperature hastened apple ripening and the development of mealiness. Texture deterioration is the main concern for high humidity stored apples. However shrivel is the dominant quality factor for low humidity stored apples.

Density was closely related to the apple internal air space. Mealy apple density was significantly lower than that of fresh apple. Its decrease was mainly affected by the development of apple mealiness. However due to the variability of the density changes, it was not a reliable mealiness predictor.

Twist strength is a kind of mechanical property. In the initial stage of the experiment it declined with storage time. Thereafter changes were related to humidity conditions. The value of mealy apples was significantly different from that of aged, shrivelled but not mealy apples. Multiple regression indicated that twist strength change was not only affected by mealiness development, but was also affected by different treatments and apple age, so it is not a suitable mealiness indicator.

Elastic modulus changes did not respond well to the development of apple mealiness. When apples shrivelled, the elastic modulus declined greatly. Multiple regression results

indicated that not only apple mealiness development, but also apple age affected the change of elastic modulus. Low R^2 value indicated that elastic modulus were weakly related to the treatments, apple age and mealiness development. It is not a suitable apple mealiness indicator.

Both compressive energy and fracture strength declined with storage time. These parameters were determined by both the cell wall strength and the strength of cell separation. The main reason for mealiness disorder was that the cell force holding cells together was greatly reduced, but the cell wall strength was maintained. These measures declined with the development of mealiness, but they also declined with apple ripening and senescence processes. Multiple regression results indicated that both changes of compressive energy and fracture strength were not only affected by mealiness development, they were equally affected by apple age, so they are not suitable mealiness indicators.

Shear gradient was mainly determined by the strength of the cell wall. It declined with storage time and the development of apple mealiness. Multiple regression results showed that both apple mealiness development and different treatments caused the decrease of fracture strength. Because different treatments would cause different mealiness development, this result may indicate that change of shear gradient was only affected by apple mealiness development. It may be a suitable mealiness indicator. However, the regression coefficient was low (0.4318), and further research work needs to be done to test and improve this result.

FURTHER RESEARCH SUGGESTIONS

This study has provided an increased understanding of apple mealiness development with respect to the role of mechanical properties. It also provided a method to understand the correlation between subjective tests and objective tests, and the difference between mealy and not mealy but aged and shrivelled apples. Further work needs to be done to extend this research, to provide accurate texture assessment values by using standard taste panels, and to quantify the mealiness indicator.

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APPENDIX 1. Probability points distribution table

(Source: Diamond, 1981)

Table 1 Probability points of the normal distribution:
single-sided; σ^2 known.

ρ (α or β)	U
0.001	3.090
0.005	2.576
0.010	2.326
0.015	2.170
0.020	2.054
0.025	1.960
0.050	1.645
0.100	1.282
0.150	1.036
0.200	0.842
0.300	0.524
0.400	0.253
0.500	0.000
0.600	-0.253

Table 2 Probability points of the normal distribution:
double-sided; σ^2 known.

ρ (α only)	U
0.001	3.291
0.005	2.807
0.010	2.576
0.015	2.432
0.020	2.326
0.025	2.241
0.050	1.960
0.100	1.645
0.150	1.440
0.200	1.282
0.300	1.036
0.400	0.842
0.500	0.675
0.600	0.524

Table 3 Probability points of t -distribution:
single-sided; σ^2 unknown.

ϕ	ρ						
	0.005	0.01	0.025	0.05	0.10	0.20	0.30
1	63.66	31.82	12.71	6.31	3.08	1.38	0.73
2	9.93	6.97	4.30	2.92	1.89	1.06	0.62
3	5.84	4.54	3.18	2.35	1.64	0.98	0.58
4	4.60	3.75	2.78	2.13	1.53	0.94	0.57
5	4.03	3.37	2.57	2.02	1.48	0.92	0.56
6	3.71	3.14	2.45	1.94	1.44	0.91	0.56
7	3.50	3.00	2.37	1.90	1.42	0.90	0.55
8	3.36	2.90	2.31	1.86	1.40	0.90	0.55
9	3.25	2.82	2.26	1.83	1.38	0.89	0.54
10	3.17	2.76	2.23	1.81	1.37	0.89	0.54
15	2.95	2.60	2.13	1.75	1.34	0.87	0.54
20	2.85	2.53	2.09	1.73	1.33	0.86	0.53
25	2.79	2.49	2.06	1.71	1.32	0.86	0.53
30	2.75	2.46	2.04	1.70	1.31	0.85	0.53
60	2.66	2.39	2.00	1.67	1.30	0.85	0.53
120	2.62	2.36	1.98	1.66	1.29	0.85	0.53
∞	2.58	2.33	1.96	1.65	1.28	0.84	0.52

Table 4 Probability points of t -distribution:
double-sided; σ^2 unknown.

ϕ	ρ						
	0.005	0.01	0.02	0.05	0.10	0.20	0.30
1	127.00	63.70	31.82	12.71	6.31	3.08	1.96
2	14.10	9.93	6.97	4.30	2.92	1.89	1.39
3	7.45	5.84	4.54	3.18	2.35	1.64	1.25
4	5.60	4.60	3.75	2.78	2.13	1.53	1.19
5	4.77	4.03	3.37	2.57	2.02	1.48	1.16
10	3.58	3.17	2.76	2.23	1.81	1.37	1.09
15	3.29	2.95	2.60	2.13	1.75	1.34	1.07
20	3.15	2.85	2.53	2.09	1.73	1.33	1.06
25	3.08	2.79	2.49	2.06	1.71	1.32	1.06
30	3.03	2.75	2.46	2.04	1.70	1.31	1.05
60	2.91	2.66	2.39	2.00	1.67	1.30	1.05
120	2.86	2.62	2.36	1.98	1.66	1.29	1.05
∞	2.81	2.58	2.33	1.96	1.65	1.28	1.04