Instrumental and Ultrasonic Techniques in Quality Evaluation of Fresh Fruit and Vegetables

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The candidate confirms that the work submitted is her own, except where work has formed part of jointly authored publications has been included. The contributions of the candidate and the other authors to her work have been explicitly indicated below. The candidate confirms that appropriate credits have been given where reference has been made to the work of others.

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Abstract

Non-destructive ultrasonic pitch and catch ultrasound measurement of sound velocity was used to assess ripeness in ‘Envy’ apples during storage and to detect brown heart in swede. Ultrasonic group velocity was measured (path length over the transit time) through intact apples along the axial and radial directions of the mature and more mature fruit every two weeks for eight weeks at 4°C and 20°C. The velocity measurement was also conducted on the defective and non-defective Brown Heart (BH) swedes in an axial direction. Compression, puncture, and sugar level tests were also carried on the two maturity fruit groups, together with a puncture test on the vegetables. The differences between the ultrasonic velocity measured in the axial and radial directions in apples was significantly correlated with the firmness (as assessed by the compression and puncture tests) of the fruit and this is possibly due to increased homogeneity of the fruit during senescence.

The correlations between ripeness and ultrasonic velocity in apples, and BH and ultrasonic velocity in swede were supported by the hypothesis of changes of volume fraction of air-water in the parenchyma. The parenchyma of the ripening apple was suggested to have undergone changes of cell compositions of the starch-sugar conversion, cell walls disassembly, and middle lamella disintegration during storage. These changes caused the accumulation of air-water mixtures in the cells, indicating the ripening process in apples. The PCA clearly discriminated the ripening apples based on the weeks of storage (weeks 2 to 8), the maturity levels (mature and more mature fruit), and the orientations of ultrasonic velocity measurements (the axial and radial directions). Meanwhile, the defective BH was suggested to cause the increasing ‘water-core’ of the internal volume in swede parenchyma. This finding was supported by the dissimilar TPA curves between the BH and the healthy swedes. The ultrasonic technique offers an alternative online, fast, economical, non-destructive assessment of firmness for the apples at different ripening stages, storage durations, and storage temperatures. It may assess the fruit ripeness along the postharvest chain and can evaluate the presence and levels of BH of an individual swede. Therefore, this technique signifies cost savings and high standard quality in fruit and vegetables.

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λ: wavelength, 81
xrec: mean recovery, 112
BH: Brown Heart, 118
BHI: Brown Heart Index, 120
c: Velocity, 81
CO_2: Carbon Dioxide, 3
f: frequency, 81
FFT: Fast Fourier Transform, 91
M: Mature, 99
MM: More Mature, 99
O_2: Oxygen, 3
PCA: Principle Component Analysis, 118
PRF: Pulse Repetition Frequency, 130
PUNDIT: Portable Ultrasonic Non-destructive Digital Indicating Tester, 86
Q_{10}: Temperature Coefficient, xxiv
RSD: Relative standard deviation, 111
TA: Texture Analyser, 118
u: uncertainty value from the sources of uncertainty, 110
$u(A)$: uncertainty of the method accuracy, 111
$u(I)$: uncertainty of the instruments' specifications, 112
$u(P)$: Uncertainty of the method precision, 111
U_c: Combined uncertainty, 110
U_e: Expanded Uncertainty, 110
UVM: Ultrasonic Velocity Meter, 107

Chapter 1
Introduction

1.1 Problem statement

Maintaining quality is one of the important issues of fresh fruit and vegetables along the supply chain. This issue is due to the increasing demand, availability of the produce, and customer awareness towards food quality. Moreover, the perishable produce and the selections of unrepresentative techniques for quality evaluation can lead to high possibility of postharvest losses, customer dissatisfaction, and unbalance of the supply and demand. These factors can affect the quality and the performance of food sector, global trade, and economy.

1.1.1 Supply and demand

Global supply and demand of fresh fruit and vegetables has been dramatically increasing in recent years. FAO (2015) and OEDC-FAO (2014) report that the global exports and imports of food have grown by over 200% from 2000 to 2014, with almost 50% of the increasing production in fruit and vegetable sectors. The reports also suggest that the trend of the global supply and demand for the produce will continue to grow in the near future.

This increasing trend in supply and demand for fresh fruit and vegetables has led a challenge to food industry ensuring the delivery of high standards of quality produce along the supply chain. Determination on the optimum harvest maturity for the produce, choice of grading parameters and monitoring of quality and ripening of fresh fruit and vegetables are critical, until the produce reaches customers. Kader (1999) pinpoints that this is because maturity index (a set of quality measurements for a specific produce to verify that the produce are mature and fit to be harvested) and ripeness are frequent quality indicators for high quality produce.

1.1.2 Quality issue of fresh fruit and vegetables

1.1.2.1 Postharvest losses

Studies have consistently shown that significant amounts of postharvest losses have been identified along the food supply chain, despite the increasing global trend in the supply and demand for fresh fruit and vegetables. This is mainly due to their physiological properties, biological variations, handlers and long distance transportations (Moretti et al., 2010; Florkowski et al., 2009; Shewfelt and Prussia, 2009; Kader and Rolle, 2004; Aked, 2002; Kader, 2002a; Bourne, 1977). Fresh fruit and vegetables undergo a complex handling throughout the food supply chain. These products are delivered from point to point by their supply chain handlers: producers (growers and processors), exporters, international brokers (wholesale distributors), retailers, and consumers. The food handlers involve in managing and controlling these products throughout the chain. They evaluate specific qualities of the produce at the designated transaction point and select or grade the produce by looking at different physical and/or biological properties (Gustavsson et al., 2011; Florkowski et al., 2009; Aitken et al., 2005; Hui et al., 2003). Inadequate understanding of the food handlers towards physiological and biological variations and improper handling of the produce are some main factors contributing to postharvest losses (Vorst et al., 2011; Tijskens et al., 2003).

Examples of the important strategies to reduce risks associated with postharvest losses of the fresh produce are the determination of right maturity stage for harvest time and monitoring ripening quality of the produce during shelf life. This determination and monitoring steps can be achieved by selecting the right instruments (techniques) that measure quality-related attributes of interests of fresh fruit and vegetables (Chen and Opara, 2013; Cho, 2011; Butz et al., 2005). The study conducted by Parfitt et al. (2010) shows that the postharvest losses start at the beginning of harvest and affect the quality of the product along the consecutive transaction points through the food supply chain. The magnitude of the problem accumulates until the product reaches consumers. Another study reveals that the largest percentages of global postharvest losses of fresh fruit and vegetables came from Asia and Latin America with percentage losses more than 30%. Indeed, approximately between 5% and 25% of losses of perishable fruit and vegetables occur in the developed countries. In developing countries, the percentage of losses is even higher valued from

20 to 50 % (Kader, 2002a). In another report, the losses of global postharvest fruit and vegetables were estimated 30 to 40% in the developing countries (Panhwar, 2006).

The fresh fruit and vegetables themselves are complex biologically and physically as well as their postharvest handling. Therefore, those influencing factors postharvest losses should be understood, investigated and managed when handling perishable products.

1.1.2.2 Perishable produce

One of the factors influencing the physiologically and physically complex fruit and vegetables is due to the climacteric rise. The climacteric rise is detected based on the dramatic increasing of respiratory carbon dioxide CO_2 exhibited by the ethylene production (the ripening hormone) prior to the fruit ripening phase. The CO_2 rise followed by its sharp decrease accelerates the senescence phase, indicating the deterioration of the fruit. The CO_2 correlates with the oxygen O_2 change rate during respiration at the stage of maturity and ripening. The increasing of the ethylene production and respiration activity increase during ripening stages indicates the climacteric behaviour of fresh produce. The storage life dramatically decreases during ripening stage and it continuously declines during senescence. This CO_2/ O_2 exchange rate is influenced by the three environmental conditions. The first condition is the effect of light, related to the photosynthesis activity. The second condition is the effect of temperature, based on the principle of Temperature Coefficient Q_{10}. The rate of respiration is doubled for every increment of 10°C between 0° and 30°C. The third is the effect of O_2 availability. It depends on the ratio of water displacement and O_2 supply/ diffusion through the intercellular air spaces of the products' cell tissues. Other plant biological related issues such as water and solutes, biochemistry and embolism, and growth and development of the cellular cells would speed up the ripening and senescence after the harvest stage (Brummell, 2010a; Kader, 2010; Moretti et al., 2010; Jackson, 2003; Kader, 2002a; Taiz and Zeiger, 2002; Downing, 1989). Texture is perceived as one of the important indicators of high quality of fresh fruit and vegetables. The texture of the produce can be measured by their flesh firmness to indicate the stage of postharvest ripeness and the quality grade. This textural property is influenced by the mechanical characteristics, such as elasticity and compressibility. These mechanical characteristics associate with the ripening progress of the produce, demonstrating softer flesh. The softening (senescence) is mediated by the biological changes of the cells (Bollen and Prussia, 2009; Abbott, 2004; Bourne, 2002; Abbott, 1999; Kramer and Szczesniak, 1973a). The softening corresponds to degradation of starch to sugar constituents, cell wall integrity and middle lamella bindings (Ng et al., 2013b; Ruiz-May and Rose, 2013; Terasaki et al., 2013; Vicente et al., 2007; Waldron et al., 2003; Redgwell and Fischer, 2002; Taiz and Zeiger, 2002; Knee, 1993; Tucker, 1993; Van Buren, 1979). The ability to dictate the ripening is critical because it will affect the product quality along the supply chain and can lead to postharvest and economic losses as well as food-waste.

Moreover, the degree of variability in quality within and among individuals of fruit and vegetables can differ considerably. For example, the variation of fruit may also come from the same fruit itself. However, understanding merely on the biological and physical changes in a product individually is insufficient. Most quality monitoring systems are based on the statistical analysis of the mean of the limited random sampling and destructive measurements, representing the whole population of the products. The data provides information to determine the quality status for the harvest time, storage conditions, and shelf life (Tijskens et al., 2003). This statistical method is not entirely sound to interpret the variation in biological data of the ripening changes in fresh fruit and vegetables due to the lack on repeatable quality measurements on the same sample. Consequently, lack of this interpretation of the biological and physical properties of the products can affect the product quality and can influence the consumers' acceptability levels.

Another important aspect of minimizing the deterioration of the fruit and vegetables is choices of measurement techniques for the quality evaluations. Various instruments using specific techniques have been implemented to measure specific quality parameters and aim to determine and monitor the postharvest shelf life quality of the fresh fruit and vegetables.

1.1.3 Problems in choosing suitable techniques for objective quality evaluation

Many destructive and non-destructive measurement techniques are implemented to evaluate the quality properties of foods. However, identification on the right quality attributes, measurements on the attributes of interest, and interpretation on the quality attribute correlated to consumers' interest are challenging. Some techniques are subjective and not repeatable (Zou and Zhao, 2015; Nicolaï et al., 2014; Zdunek et al., 2014; Chen and Opara, 2013; Schmilovitch and Mizrach, 2013; Kong and Singh, 2012; Mizrach, 2011; Zou et al., 2010; Camps and Christen, 2009; Abbott, 1999; Chen and Sun, 1991). A study by Tijskens et al. (2003) indicates three suggestions for addressing these challenges. Firstly, the measurements must be objective to evaluate the specific quality attribute of the produce. The data of the correct technique is able to represent the biological variation within a batch and individual unit of fresh fruit and vegetables. Secondly, the food handlers must be competent to interpret the values of data analysis meaningfully. Thirdly, these food handlers must follow a standard guideline of the suitable food attributes associated with the customers' perceived quality during the purchasing. In other studies, a correlation between food attributes and food properties has been examined for biological variations of cucumbers and tomatoes by combining multiple measurement techniques to explain one food quality attribute (Schouten et al., 2004; Schouten et al., 2002).

The majority of the quality of fruit and vegetables are measured destructively. This means that reliability and reproducibility of the quality assessment on the same products cannot be performed. Hence, the techniques rely upon limited sampling and contribute large variability that may not represent the quality of the whole batches (Alfatni et al., 2013; Chen and Opara, 2013; Abbott, 1999; Wallace, 1946). Therefore, determination on the status of the produce quality by using non-destructive techniques can be alternative due to their practical and cost-effective advantages.

As a result, researchers and stakeholders have been striving to bridge the questions between quality of fresh fruit and vegetables and objective assessments of the quality. The following issues have been raised;

(1) suitable number of quality properties (variables/ parameters) to be measured;
(2) suitable measurement techniques to be selected to evaluate the quality properties;
(3) sufficient number of quality attributes to be interpreted;
(4) correlations between measured quality properties and selected quality attributes of the fruit and vegetables; and
(5) mechanisms (data analysis based on statistical tools) to translate back this correlation to the quality accepted by consumers.

Therefore, the key problems with these evidences are to evaluate and dictate the quality of these fruit and vegetables until they reach their consumers. The evaluation of the quality can be handled by measuring the main quality properties of fresh produce, which prominently influence the ripening (senescence) along the food supply chain. To evaluate those critical quality properties, right techniques must be selected properly that are objective and repeatable for measurements of the quality attributes of interests (Kader and Rolle, 2004; Luning et al., 2002; Bourne, 1977).

1.2 Aim of the research

Based on the problem statement phase, this research aimed to evaluate the quality of fruit and vegetables by using the instrumental and ultrasonic techniques by

(1) developing a methodology for the experimental ultrasonic measurement techniques,
(2) investigating the feasibility of the ultrasonic measurement techniques in quality evaluation of ripeness and internal qualities of the selected produce, and
(3) understanding a correlation between the textural properties by using the ultrasonic measurement techniques together with other designated measurements and ripeness and internal qualities of the produce.

The conceptual framework and aim of this research together with the research questions and hypothesis were reformulated at the end of this chapter after further discussions on the respective literature.

1.3 Outlines of the thesis

This thesis is outlined as follows:

Chapter 1 provides the statement of the problem and a set of general research objectives, research questions, and hypothesis. It also gives a background of the problem of this study by reviewing respective literature related to the evaluation of the quality of fresh fruit and vegetables by instrumental and ultrasound techniques. It aims to define the gaps in the subject area. Literature pertains to the areas of research on food quality attributes, sensory attributes, and textural properties of the products. In this chapter, instrumental techniques, measuring the quality of the textural properties of the fruit and vegetables, are also presented, focussing on ultrasound techniques.

Chapter 2 covers materials and methods for an experimental study of apples and swedes using ultrasound means and for data analysis conducted in this study. The first part of the study demonstrates a setup of ultrasonic instruments and its protocol, providing ultrasonic velocity data (based on the distance of ultrasonic propagated wave through the tested medium over the time of flight). The second part of the study discusses the assessment of ripeness of apples during storage using the ultrasonic measurement developed technique. The assessment is based on the orientations of the measurements (anisotropy of apple cell structures: axial and radial directions), two maturity levels (mature and more mature samples), and 8-weeks storage durations (weeks 0, 2, 4, 6, and 8). The fruit firmness is also measured, using compression and puncture tests, and sugar content, using a hand-held refractometer to establish the correlations among these multiply testing. The third part of the study is the detection of Brown Heart (BH) in swede, using the ultrasonic measurements (velocity) and the laboratory-developed BH Index (ten levels of severe BH based on a visual inspection). Texture Profile Analysis (TPA) is also conducted. The procedures of sampling, analysis, and data processing are elaborated.

Chapter 3 presents and discusses on the results of the three experimental studies. The data analysis is based on descriptive statistical analysis, linear regressions, *t*-Test, Pearson’s Correlation and Principle Component Analysis, PCA.

Chapter 4 summarizes the research findings and provides conclusions from the findings in this experimental study. It also presents implications and limitations of the work. Recommendations for future research are included.

The next sections consist of literature reviews of evaluation of quality of fresh fruit and vegetables by instrumental and ultrasound techniques. It covers food quality attributes, sensory attributes, and textural properties of the products, and instrumental techniques to measure the quality of the textural properties of the produce.

1.4 Food quality

1.4.1 Maintaining quality

Literature has shown that adequate food handling is one of the most challenges in keeping acceptable quality through the food supply chain after harvest. Ability to prolong quality of fresh fruit and vegetable is associated with the shelf life, the handling of the product and physical storage conditions (Gustavsson et al., 2011; Hubert et al., 2010; Kader, 2010; Parfitt et al., 2010; Luning and Marcelis, 2007; Shewfelt and Henderson, 2003; Kader, 2002a; Luning et al., 2002; Juran, 1998).

1.4.2 Postharvest Deterioration (quality degradation)

Postharvest deterioration (quality degradation) is associated with four common causes. Firstly, physiological changes of fresh fruit and vegetables is due to ripening (senescence) (been discussed in sub-sections 1.1.2.1 Postharvest losses and 1.1.2.2 Perishable produce) and 'chilling injury' due to low temperature storage (such as dark spots in bananas, degrading their appearance quality). Senescence and 'chilling injury' leads to irreversible damage to the cell tissues and exposed to microbial decay. Secondly, physical damage occurs before, during and after harvest potentially due to climatic conditions, food-handling practices, and storage and transportation conditions. Impacts bruising during harvest (such as kiwifruits, apples, and potatoes), compression brushing during grading and packaging (such as grapefruit), and vibration damage during transportations are the examples of the physical damage. These damages promote progressive increase of respiration rate and heat, moisture loss and exhibit of ethylene production that leads to microorganism invasion causing contagious decays of among cell tissues. Thirdly, insecticides, fungicides, herbicides, growth, and nutritional applications cause chemical injury. The chemical reactions within itself and its act as a catalyst to another reaction have been observed in in two occasions (disfigured onions caused by the reaction of water on the brown papery scales and bleaching in grapes caused by over-concentrations of sulphur dioxide). Fourthly, pathological decay (postharvest diseases) is caused by virus, bacteria and fungi (Snowdon, 2010). Virus infection is not common although the infection could be identified before harvest. Bacteria normally survive in a medium between pH 4.5 to 7.0, whereas fungi medium is between pH 2.5 to 6.0. As a result, bacterial infection is seldom in acidic fruits (such as citrus) but it is usually in few selected vegetables. In contrast, fungi infection is the most common cause of the postharvest diseases (Thompson, 2015). The infection process starts with spore germination. The germination is triggered by favourable temperature and atmosphere together with oxygen, metabolised organic compound solutions. Next, the spores are swelled and disseminated by rain and windborne mist as well as soil water, deposited driving rain on lower positioned fruit, sprinkler irrigation systems, human and animals (such as insects and birds). The pathogens enter fruit and vegetables through cut skin due to bruises, punctures, rubbed areas. Then, they invade the tissue of the wounded fruit. Another pathogen penetration is through direct entrance to fruit cuticle and epidermis by sickle-shape protruded pathogens. Later, they attack the fruit flesh by toxic substances and lead to the dead of fruit cells (ripening fruit accelerates the infection process). The pathogens in the infected cells will regenerate other cycles of spore productions (Kader, 2002a). Examples the common postharvest diseases are stem-end rots (avocado, citrus fruits and mango), botrytis (apple, pear and kiwifruit), and anthracnose (banana, mango, papaya, melon, apple, strawberry and avocado) (Thompson, 2015; Snowdon, 2010; Ladaniya, 2008; Kader, 2002a).

1.4.3 Potential sources influencing quality deterioration

Various potential sources of variations influence quality of the fresh fruit and vegetables because their quality deteriorates starting right after the harvest. To understand the quality variation of the produce within individual and among batches, the food handlers must identify important locations of transactions where the deterioration is likely to occur along the food supply chain (Kader, 2010; Luning and Marcelis, 2009a; Hewett, 2008; Thompson, 2003; Luning et al., 2002). For this reason, some potential sources of variation influencing quality and location along the chain are summarised in Table 1 (substantiated with the respective literature). It highlights that improper food handling could decrease the quality of fresh and vegetables at the beginning of the food transactions. The influencing quality attributes include appearance, texture, flavour, shelf life, and/or nutritional value. As early as the grower stage, choosing the right cultivar is crucial to minimise postharvest deterioration and extend the shelf life of the produce. Managing the climatic conditions and implementing right cultivation practices are the next possible sources of variations due to a correlation between the physiochemical properties and temperature, light, wind and rainfall. Poor determinations of harvesting time, ripeness and handling can also affect the quality of the produce at the later phases. Subsequently, the lack of good handling of fresh produce (treatment, packaging and coding and grading), maintaining storage conditions and having a proper transport vehicle (storage condition: temperature setting and relative humidity) can also affect the quality of the products at the subsequent transaction points.

Table 1: Relationships between locations and possible sources of variation influencing quality attributes

Where does the variation occur?	What are possible sources of variation influencing quality attributes?	What are the influenced quality attributes	Related literatures
Growers	*Deciding on the right cultivar* • associated with quality expectation of consumers (outcome) : because not all cultivars are suitable for the export conditions (for long distance transport and storage before their consumption)	Appearance (A) Texture (T) Flavour (taste and smell) (F) Shelf-life (S) Nutritional value (N)	(Lugaric et al., 2016; Ng et al., 2013b; Toivonen, 2011; Hewett, 2008; Aitken et al., 2005; Hewett et al., 2005; Schouten et al., 2004; Manolopoulou and Papadopoulou, 1998)
	Handling climatic conditions (temperature, light, wind and rainfall) • unlikely that those two consecutive seasons would have identical soil and climatic conditions meaning wide variations in physiochemical properties of the fruit		(Kader, 2010; Kader, 2008; Léchaudel and Joas, 2007; Mowat and Kay, 2006; Richardson and Currie, 2006; Kader, 2005; Lee and Kader, 2000)
	Practicing right cultivation practices (tree pruning, irrigation management and harvesting time and handling fruit during harvest) • Lack of right determination of harvest time and inadequate handling of fruit during harvest can lead to poor fruit storage quality. Thus, it influences the shelf life in the latter phases.		(Fawole and Opara, 2013; Kader, 2010; Burdon et al., 2009; Florkowski et al., 2009; Shewfelt and Prussia, 2009; Antunes et al., 2007; Aviara et al., 2007; Léchaudel and Joas, 2007; Mowat and Maguire, 2006; Aitken et al., 2005; Kader and Rolle, 2004; Strik, 2004; Kumar et al., 2003; Thompson, 2003; Aked, 2002)
Collectors/ Handlers/ Transporters/ distributors	*Maintaining storage conditions* • Temperature: Low temperature slows down the respiration rate; below ±1°C causes chilling injury and other cold related disorders; large fluctuations in temperature cause water condensation on the fruit, which can lead to high water loss from the fruit. • Relative humidity (RH): fresh fruit stored at different setting of temperatures influencing variable fruit properties; higher RH level from prescribed range inducing the growth of mold or microorganisms and surface cracking on the fruit •		(Vorst et al., 2011; Kader, 2010; Parfitt et al., 2010; Vorst et al., 2007; Boyd and Barnett, 2006; Aitken et al., 2005; Hewett et al., 2005; Kader and Rolle, 2004; Maguire et al., 2004)
	Managing good handlings of the fresh produces • treatment, packaging and coding and grading		(Alfatni et al., 2013; Trienekens and Zuurbier, 2008; Aitken et al., 2005; Hewett et al., 2005)
	Having a proper transport vehicle • Suitable temperature and relative humidity managements • Suitable frequency of inspection of the products		(Bhat, 2012; Pimentel and Pimentel, 2008; Kader, 2005)

1.5 Food quality attributes and customers’ perception on quality

1.5.1 Perception of food quality attributes

Good quality of fresh fruit and vegetables as perceived by consumers is based on food quality attributes. These attributes are divided into two major categories: intrinsic and extrinsic quality attributes. Firstly, intrinsic quality attributes are associated with the physical properties of the product. They are sensory properties (appearance, texture, flavour, odour and sound), product safety and health aspects (nutrition and safety) and shelf life as well as product reliability and convenience. Secondly, the extrinsic quality attributes are related to the external characteristics of the product. It comprises of production system characteristics, environmental aspects and marketing (Luning et al., 2002). The sensory properties of food in the intrinsic quality attributes are an important factor consumer acceptability of the product. They indicate the interdependence of quality and customer perception towards food (Kuipers et al., 2013; Bourne, 2002).

The similar trend of the domination of sensory properties is also found in quality components of fresh fruit and vegetables (Table 2). The first factor is associated with the appearance mainly focussing on the visual aspects of the product: size (e.g. dimensions, weight, and volume), shape and form (e.g. diameter, ratio), colour (e.g. intensity, uniformity), gloss and defects (e.g. external internal, physical, chemical). The second factor is related to the textural properties: firmness, hardness, softness, juiciness, mealiness, and toughness of the produce. Meanwhile, flavour is dominated by taste and smell of the food such as sweetness and acidity. Nutritional value of the produce is represented commonly by the contents of carbohydrates, proteins, and vitamins. Lastly, safety frequently covers the aspects of toxicity and contamination of food (Kader, 2002a). Therefore, studies on food sensory properties demonstrate that these properties are important due to their correlation with customers’ perception towards quality of food.

Table 2: Quality components of fresh fruit and vegetables (Kader, 2002a)

Main factors	Components	
Appearance (visual)	Size	Dimensions, weight, volume
	Shape and form	Diameter/depth, ratio etc.
	Colour	Intensity, uniformity.
	Gloss	Nature of surface wax.
	Defects	External, internal, morphological, physical, chemical, etc.
Texture (feel)	Firmness, hardness, softness, juiciness, mealiness, toughness.	
Flavour (Taste and smell)	Sweetness, acidity, astringency, etc.	
Nutritional value	Carbohydrates proteins , vitamins, etc.	
Safety	Naturally occurring toxicants, contaminants, mycotoxins	

1.5.2 Sensory properties

A number of studies have emphasized that customers perceive a quality through the sensory properties: appearance, flavour and texture in food (Moskowitz et al., 2012; Barrett et al., 2010; Varela et al., 2006; Kilcast, 2004; Schroder, 2003; Bourne, 2002; Reid, 2002; Meilgaard et al., 1999; Moskowitz, 1995; Moskowitz and Krieger, 1995; Szczesniak, 1986). The emphases on sensory characteristics in those studies are summarized in Table 3.

Studies have widely focused on appearance and flavour to evaluate the quality of food. However, studies on the correlation between textural properties and food quality have not been consistent among different classification of fresh fruit and vegetables. The textural properties are unique because the properties are mixed between appearance and flavour. The terminology of the word, texture, varies and it is influenced by geographical and cultural factors. Consequently, measurements and standardisations on food textural properties are challenging. In the subsequent section, textural properties are discussed.

Table 3: Sensory properties of food – appearance, texture, and flavour

Sensory characteristics	Sources of senses	Influencing components	Impact	Examples of Produce	References
Appearance	Sight (visual)	Shape Pattern Size Colour	Product Packaging	*Ratio size:* Tea leaf *Colour:* Bananas Capsicum, Tomato *Glossy and free from physical damage:* Apples, Kiwifruit	(Toivonen, 2011; Barrett et, al., 2010; Schroder, 2003; Tijskens et al., 2003; Johnston et al., 2001)
Texture * Sound is related to textural properties	Touch Movement Sight * Sound	Attributes: *1. Mechanical* Hardness Fracturability Chewiness Gumminess *2. Geometrical (Visual Texture)* Size, Shape Pattern *3. Surface* Level of moisture/ fat *4. Auditory ** Crispiness Crunchiness	Product	*Crunchiness:* Apples	(Barrett et al., 2010; Tiplica and Vandewalle, 2010; Zdunek et al., 2011; Zdunek et al., 2010; Varela et al., 2007; Shmulevich et al., 2003)
Flavour	Taste Odour	Sour Bitter Salty, Umami	Product	*Sweetness:* Kiwifruit	(Barrett et al., 2010; Harker et al., 2009; Kader, 2008; Kader, 2002b)

1.6 Importance and classifications of textural properties

Texture is a major factor in customers' perception of good quality of product. It can be measured by selected instruments due to its association with mechanical aspects of food. Studies stress that understanding of textural properties of fresh fruit and vegetables is important, such as firmness and internal defects (Chen and Opara, 2013; Kilcast, 2013; Engler and Randle, 2010; Aguilera, 2005; Kilcast, 2004; Bourne, 2002; Luning et al., 2002; Kilcast and Fillion, 2001; Aguilera and Stanley, 1999; Kilcast, 1999; Rosenthal, 1999; Kramer and Szczesniak, 1973a; Muller, 1969). In addition, sound such as crunchiness and crispness of food is reported as another indicator in evaluation in textural properties after firmness. Other studies show that sound has a relationship with firmness of fruit and vegetables (São José et al., 2014; Tunick, 2011; Varela et al., 2006; Chen et al., 2005; Duizer, 2004; Aked, 2002; Kilcast and Fillion, 2001). From these studies, sound characteristics are suggested that can be used to assess ripeness or internal defect of the produce. As a result, techniques by using sound are worth to be explored and be further understood.

However, standardising and interpreting the correlation among textural properties, the interest quality attributes and consumers' preferences of food product have few successful attempts. This is because the researchers have found that a general agreement on the terminology referring to textural mechanical characteristics is challenging to be standardised due to the different geographical, cultural, and linguistic factors. Consequently, consensus statements on the terminology used in food sensory textural characteristics is limited to interpret the quality attributes of the fresh produce. The following discusses about classifications of textural properties, various ranges of terminologies in description of texture in food, associations between texture and plants, ripening stages, mechanical characteristics, and sound.

Many approaches explain food texture. According to Bourne (2002), texture is referred as textural properties because the words themselves convey various interdependent characteristics in foods. The textural properties are defined by physical properties built by the structure of foods and mechanical properties. The properties may be sensed by hand and mouth and they are not affected by chemical aspects of taste and odours. These properties can be measured as a function of mass, time and distance.

Textural properties are mostly associated by more than one characteristic. The article, ‘Classification of Textural Characteristics’, defines textural properties as the following:

“Texture is the sensory and functional manifestation of the structural, mechanical and surface properties of foods detected through the senses of vision, hearing, and touch and kinaesthetic.” (Szczesniak, 2002).

In addition, The International Organization for Standardization (ISO) (1994) in Sensory analysis — Methodology — Texture profile defines texture as the followings:

“All the mechanical, geometrical and surface attributes of a product perceptible by means of mechanical, tactile, and where appropriate, visual and auditory receptors.”

An overview of textural properties is summarized in Table 4 (Bourne, 2002; Szczesniak, 1962) with additional noise or sound characteristics (Meilgaard et al., 1999). The properties consist of mechanical, geometrical, noise and other characteristics. Each of these characteristics is classified in primary and secondary sub-categories, and their popular terms are given.

Most consumers select fruit and vegetables by assessing the firmness of the products to determine the ripeness or categorize the quality grade. Firmness falls under the category of mechanical characteristics of produce. It is correlated to elasticity of the produce where the decreasing of compressibility related to maturity and ripeness levels due to softening (senescence) of tissue cell structures (Bollen and Prussia, 2009; Florkowski et al., 2009; Abbott, 2004; Bourne, 2002; Abbott, 1999; Szczesniak and Kahn, 1971). Firmness is correlated with forces on an object (ripening status of fruit and vegetables) and deformation as a function of time (storage time, shelf-life). Force is quantified by force per unit area (stress), and deformation is quantified by the change in length of an object (strain) (Table 5). Three main elasticity responses depending on the food medium are compressibility (elastic modulus, E), shear (shear modulus, G) and Poisson directional effects (Poisson ratio, μ). The elastic modulus is frequently used in describing the textural properties in food (Dobraszczyk and Vincent, 1999; Van Vliet, 1999). The references suggest that the quality evaluation of textural properties of fresh fruit and vegetables can be explained through the mechanical perspectives.

Firmness is identified as the most important mechanical properties of the produce and associated with ultrasound propagation parameters such as velocity (Chen and Opara, 2013; Mizrach, 2008b). Therefore, this characteristic is a prospective indicator to assess the quality status of fresh fruit and vegetables. Subsequently, the mechanical parameters of texture are divided into two properties: primary and secondary properties (Table 6). The primary properties of food are hardness (firmness), viscosity, springiness, and cohesiveness. The secondary properties include fracturability, chewiness, gumminess, crispiness, and crunchiness. Each property is defined by its physical and sensory aspects (Bourne, 2002; Szczesniak, 2002; Vickers and Bourne, 1976a; Vickers and Bourne, 1976b; Kramer and Szczesniak, 1973a).

Despite the presentation on the mechanical textural properties, terminologies for these properties vary among consumers and they are dependent on geographical and cultural factors. As a result, a challenge arises to standardise the terminologies so that the textural properties of food can be objectively measured (been mentioned previously) (Bourne, 2002; Szczesniak, 1988; Kramer and Szczesniak, 1973a). The terminologies of the textural properties used in different languages are presented in Appendix 1.

As been discussed earlier, textural properties are not only associated with physical (mechanical) aspect but also with biological aspect of fruit and vegetables. The next section discuss on a relationship between of the textural (physical) and biological properties of the produce.

Table 4: Classification of Textural Characteristics and their Popular Terms (Bourne, 2002; Szczesniak, 2002; Vickers and Bourne, 1976a; Szczesniak and Kahn, 1971; Szczesniak, 1962)

Mechanical characteristics					
	Primary	Secondary	Popular terms		
Rheology Young modulus Shear modulus Poison ratio	Hardness, Firmness	Fracturability	Soft → firm → hard		
	Cohesiveness	Brittleness *	Crumbly → crunchy → brittle		
		Chewiness	Tender → chewy → tough		
		Gumminess	Short → mealy → pasty → gummy		
	Viscosity		Thin → viscous		
	Elasticity *		Plastic → elastic		
	Adhesiveness		Sticky → tacky → gooey		
Geometrical characteristics	Class		Examples		
	Particles size and shape		Gritty, grainy, coarse etc.		
	Particle shape and orientation		Fibrous, cellular, crystalline, etc.		
Other (Surface) characteristics	Primary	Secondary	Popular terms		
	Moisture content	-	Dry → moist → wet→ watery		
	Fat content	Oiliness	Oily		
		Greasiness	Greasy		
Common *Noise Characteristics			Sound or non-oral		
	Primary	Secondary	Foods	Skincare	Fabric
* Noise properties are perceived sounds (pitch, loudness, persistence) and auditory measurement	Pitch	-	Crispy	Squeak	Crisp
	Loudness	-	Crunchy		Crackle
	Persistence	-	Squeak		Squeak

.

Table 5: Definition, expression and equation of mechanical properties

Rheology properties	Definition	Expression	Equation
Young Modulus, *E*	Elasticity of materials as a ratio of stress (σ) and strain (ξ).	Stress is quantified by **F**orce over **A**rea (units of pressure) and strain is quantified as range of displacement**, *ΔL***, over **L**ength. It occurs in uniaxial compression (direct compression on a plane).	$E = \frac{Stress}{Strain} = \frac{\sigma}{\xi} = \frac{F/A}{\Delta L/L}$ Elasticity Modulus applies on the first portion of the stress - strain curve where the elasticity is linear. It is also called a tangent modulus in solid material (e.g. Fruit and vegetables) and can be quantified by its slope.
Shear Modulus, *G*	Shearing stress proportional to shear strain	Shearing stress is quantified by Force over Area (units of pressure). Shearing strain is quantified as greatest displacement of the vertical face of the material, γ, over Length Shearing occurs in the lateral over the sideways of the solid material while the base of the material is remained motionless. This brings a change in angle, vertical face that is $\tan\gamma = \frac{\Delta L}{h}$. When the amount of shearing stress and shearing strain is equal, it becomes $\tan\gamma = \gamma$	$G = \frac{shearing\ stress}{shearing\ strain} = \frac{F/A}{\gamma/L}$
Poisson Ratio, μ		Ratio of a change in width per unit width (transverse/ lateral strain) ($d\varepsilon_{trans}$) over change in length per unit length (axial strain) ($d\varepsilon_{axial}$)	$\mu = \frac{d\varepsilon_{trans}}{d\varepsilon_{axial}}$

Table 6: Definitions of sensory textural parameters: mechanical characteristics (Bourne, 2002; Szczesniak, 2002; Vickers and Bourne, 1976a; Vickers and Bourne, 1976b; Kramer and Szczesniak, 1973b)

Sensory textural mechanical characteristics	Definition by	
	Physical aspect	Sensory aspect
Primary properties		
Firmness (Hardness)	Force necessary to attain a given deformation	It is texture characteristics explaining the resistance of food to deform or break. Force is applied between the molar teeth to compress foods.
Cohesiveness	The extent to which a material can be deformed before it ruptures.	Numbers of chewiness are applied in the food during mastication.
Viscosity	Rate of flow per unit force.	The force required when drawing a liquid from a spoon over the tongue.
Springiness	The rate at which a deformed material goes back to its un-deformed condition after the deforming force is removed	The degree to which a product returns to its original shape and it has been compressed between the teeth
Adhesiveness	The work necessary to overcome the attractive forces between the surface of the food and the surface of the other materials with which the food comes in contact	Force is applied to separate food from the mouth palate during mastication.
Secondary properties		
Fracturability	Force with which a material fractures: a product of high degree of hardness and low degree of cohesiveness	Force is needed to break foods during mastication.
Chewiness	Energy required to masticate a solid food to a state ready for swallowing: a product of hardness, cohesiveness and springiness	The time is taken for solid foods to be ready to be swallowed.
Gumminess	Energy required disintegrating a semi-solid food to a state ready for swallowing: a product of low degree of hardness and a high degree of cohesiveness.	The time is taken from semi-solid foods to be ready to be swallowed.
Crispiness	Sharp, burst and short sound upon deformation (typically producing a high pitched sound)	Initial sensation of sound produced during biting/ crushing
Crunchiness	Firm and brittle sound (typically producing a lower pitched sound, less loud and longer lasting than for crisp sound)	Cumulative sensation of sound during chewing

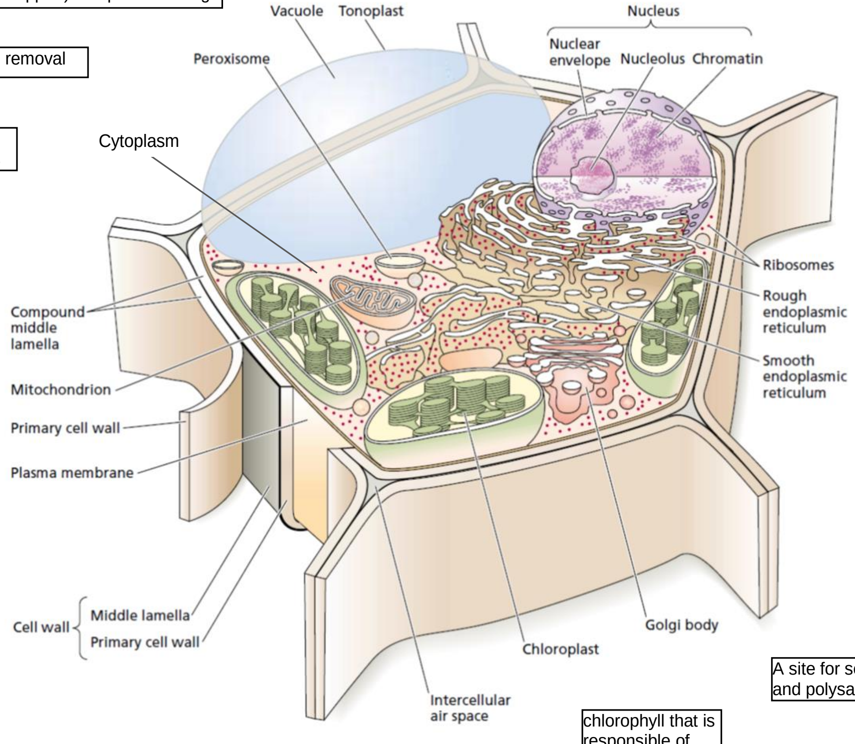
1.7 A relationship between textural (physical) and biological properties

Textural properties of the product determine quality of fresh fruit and vegetables. These properties are influenced by the cellular tissue structures of the produce (Hopkins and Huner, 2009; Taiz and Zeiger, 2002; Harker et al., 1997; Jackson and Harker, 1997; Northcote, 1972). The following sections covers plant cells, their plant tissue systems, and primary cell walls.

1.7.1 Plant cells

A plant cell consists of a nucleus, a cytoplasm, and subcellular organelles (Figure 1) and the followings give brief descriptions of the cell structure. Plasma membrane is a semi permeable layer surrounding a cell and allowing transportations of solutes. Cell walls also surround the cell and serve to protect and provide shape as well as turgidity of cells. The middle lamella segregates adjacent cells and plays a role in supporting and strengthening plant cells. The vacuole is responsible for plant defence, cell growth, turgidity (structural support), and protein storage. It contains largely water and solutes. Intercellular air space is a site for gas exchange and diffusion in photosynthesis and respiration.

Meanwhile, cytoplasm is filler to the cell and a medium of cell activities. Endoplasmic Reticulum (ER) comprises of rough and smooth ribosomes. It is a network of internal membranes. The rough ribosomes are a region for protein synthesis and their shape is flat. Contrarily, the smooth ribosomes are a section for lipid synthesis and membrane developments and its shape is rather round or tubular. The golgi body is a site for secretion of proteins and polysaccharides. Next, nucleus is a hereditary storage of a cell for chromosome replication, DNA transcription, and protein complex. It has three components: nucleolus for ribosome synthesis, nuclear envelope (double layer membranes protected the nucleus), and chromatin (DNA-protein complex). Next, spherical single membrane micro-bodies consist of peroxisomes and glyoxysomes. Peroxisomes act as a hydrogen peroxide removal, a by-product in photorespiration whereas glyoxysomes facilitate a conversion of fatty acids to sugars as energy during growth of the juvenile plants. Furthermore, mitochondria is an energy production region for plant respiration and chloroplast consists chlorophyll for photosynthesis (Taiz and Zeiger, 2002).



a hereditary information storage

double layer membranes protected the nucleus

DNA-protein complex

ribosome synthesis

Protected membrane of a vacuole

plant defense, cell growth, turgidity (structural support) and protein storage

hydrogen peroxide removal

a filler to the cell and a medium of cell activities

adjacent cell segregation and cell structural support

Plant respiration site

a semi permeable layer surrounded a cell and allowed transportations of solutes.

protect and provide shape as well as turgidity of cells

a location for protein synthesis

a location for lipid synthesis and membrane developments

A site for secretion of proteins and polysaccharides

chlorophyll that is responsible of photosynthesis

a site for gas exchange and diffusion in photosynthesis and respiration

Figure 1: A diagram of a plant cell; a nucleus, a cytoplasm and subcellular organelles and cell walls and middle lamella protected and segregated the cell from other adjacent cells as well as intercellular air space and their functions (Taiz and Zeiger, 2002)

1.7.2 Plant tissue systems

Plant tissue systems consist of three major tissues: dermal, ground and vascular tissues (Table 7).

Dermal tissues. Dermal tissue components are epidermal and periderm *(‘epi’* and ‘*peri’* in Latin mean top and around respectively). Their primary functions are protection and water loss prevention.

Ground tissues. Ground tissues are composed of three components. Firstly, parenchyma plays a role in a tissue filler; support, storage, and secretion. It specialises in aeration, wound repair, and metabolism (*‘pare’* and *‘chyma’* means beside and to fill respectively). Its tissue development changes from round to elongate. Secondly, collenchyma also serves as filler tissues. However, it has thicker cell walls and its shape is more elongated compared to parenchyma. Their functions are to support and ensure integrity of the plant structures. Thirdly, sclerenchyma is made up by thick cell walls *(‘sclera’* means hard) with elongated shapes. It comprises of cellulose, hemicellulose and lignin and its functions are to strengthen and support plant tissue system.

Vascular tissues. Xylem and phloem compose the vascular tissue, and they act as a plant translocation. Xylem transports water and minerals in a whole plant system (‘*xylem*’ means wood). Meanwhile, phloem transports photosynthesis products (such as sugars) and solutes of plants (*‘phloem’* means bark) (Taiz and Zeiger, 2002; Atwell et al., 1999).

Table 7: Plant tissues system, components, and functions

Tissue types	Components	Functions
Dermal	Epidermis Periderm	Protection and water loss prevention
Ground	Parenchyma Collenchyma Sclerenchyma	Support, storage, and secretion, specialized functions
Vascular	Xylem Phloem	Transportation of photosynthesis products and other solutes

1.7.3 Cell wall structures and roles

Ground tissues strengthen plant cell structures. The cell wall structural components influences the changes during ripening of fresh fruit and vegetables (Hopkins and Huner, 2009; Taiz and Zeiger, 2002). The structure and synthesis of cell walls must be learned so that quality degradation of the fresh produce can be understood. The wall structures comprise the primary and secondary cells, and middle lamella.

Plant cell walls. Four major components compose plant cell walls: cellulose, matrix polysaccharides (pectins and hemicellulose), structural proteins and lignin. Firstly, the cellulose makes up 25% of the cell walls. It provides strength to the cell tissues and exists as a crystalline structure, resistant to water and enzymatic attack. The cellulose retains the rigidity and resistance to fracture. Secondly, the first matrix polysaccharides (hemicelluloses) are also made up by 25% of the cell walls. It ensures segregations of cellulose micro fibril and retains elasticity and stretching. The second matrix polysaccharides (pectins) contribute 35% of the cell wall components. Pectin is hydrated gel acting like filler between cellulose and hemicellulose and associates with the porosity level of the primary cell structure. Its abilities of elasticity and stretching provide mechanical strengths to the cells. In brief, matrix polysaccharides control elasticity of cells. Thirdly, structural proteins represent 1 to 8% plant cell walls. They provide additional strength to the wall. The first three cell wall components compose primary cell walls. Primary cell walls continuously undergo constant growth and vary in shapes. Meanwhile, the fourth component of plant cell walls is lignin. It adds mechanical strength and toughness to the walls. Lignin exists vastly as a component of secondary cell walls. The secondary cell walls are developed when the cell growth stops or reaches maximum capacity (Ruiz-May and Rose, 2013; Cosgrove and Jarvis, 2012; Hopkins and Huner, 2009; Taiz and Zeiger, 2002; Northcote, 1972).

Middle lamella. Middle lamella is the thin walls and also consists of pectin substances. They glue neighbouring cells together. This adhesion provides mechanical strength to the intercellular cell walls. Middle lamella is also sensitive to changes and become an indicator during maturation, ripening/ softening, and storage (Negi and Handa, 2008; Taiz and Zeiger, 2002; Van Buren, 1979).

1.7.4 Primary cell walls and synthesis

Synthesis of each component of the primary cell walls is discussed in this subsection (Table 8 and Figure 2).

Cellulose. Cellulose is composed of (1→4)-linked β-D glucans and these glucans comprises of D-glucose. A ribbon of the glucan chains are held together by noncovalent bonding. The size of the ribbon is about 4 nm width. This bond causes cellulose to be insoluble, chemically stable, and resistant to chemical and enzymatic encounter. Cellulose synthesis starts at cytosol where glucose and fructose polymers are synthesised by the sucrose syntheses. Sucrose synthase slice the glucose and fructose chains. As a result, both glucose and fructose polymers now exist independently. Glucose polymers then are synthesized further by cellulose syntheses in a particle rosette (highly structures/ large, ordered protein located in plasma membrane) (Ruiz-May and Rose, 2013; Negi and Handa, 2008; Paliyath and Murr, 2008; Taiz and Zeiger, 2002).

Matrix polysaccharides. Hemicelluloses and pectins are another major part of cell wall components for polysaccharides matrix polymers. They are hydrated, non-crystallisation network and heterogeneous. They coat around cellulose micro fibrils so that the structures do not collapse to each other. Hemicellulose size is 50 – 500 nm and it consists of xyloglucans and arabinoxylan chains. They can be dissolved by strong alkali such as in NaOH. Meanwhile, pectins can exist as acidic sugars which form as galacturonic acid and Homogalacturonan or polygalacturunan acid. Other pectin components are neutral sugars, coming from the sugars of rhamnose, galactose and arabinose (Ruiz-May and Rose, 2013; Negi and Handa, 2008; Paliyath and Murr, 2008; Taiz and Zeiger, 2002).

Structural proteins. Structural proteins add extra strength to the cell walls and they cross-link with the polysaccharides polymer matrix. Structural protein becomes more insoluble towards maturity and ripening (Ruiz-May and Rose, 2013; Negi and Handa, 2008; Paliyath and Murr, 2008; Taiz and Zeiger, 2002).

Intercellular air spaces. The modifications of the cell walls give an impact to intercellular air space. It is reported that the volume of the air space decreases as the cell wall disassembly increases, indication of softening of the cell tissues (ripening process) (Knee, 2002; Knee, 1993; Hatfield and Knee, 1988).

Table 8: Structural components of plant cell walls (Taiz and Zeiger, 2002)

Components	Elements
Cellulose	Microfibrils of (1→4)β-D-glucan
Matric polysaccharides	
Pectins	Homogalacturonan Rhamnogalacturonan Arabinan Galactan
Hemicelluloses	Xyloglucan Xylan Glucomannan Arabinoxylan Callose (1→3)β-D-glucan (1→3,1→4)β-D-glucan [grasses only]
Lignin	Polymer of phenyl-propanoid groups
Structural proteins	Predominant amino acid composition

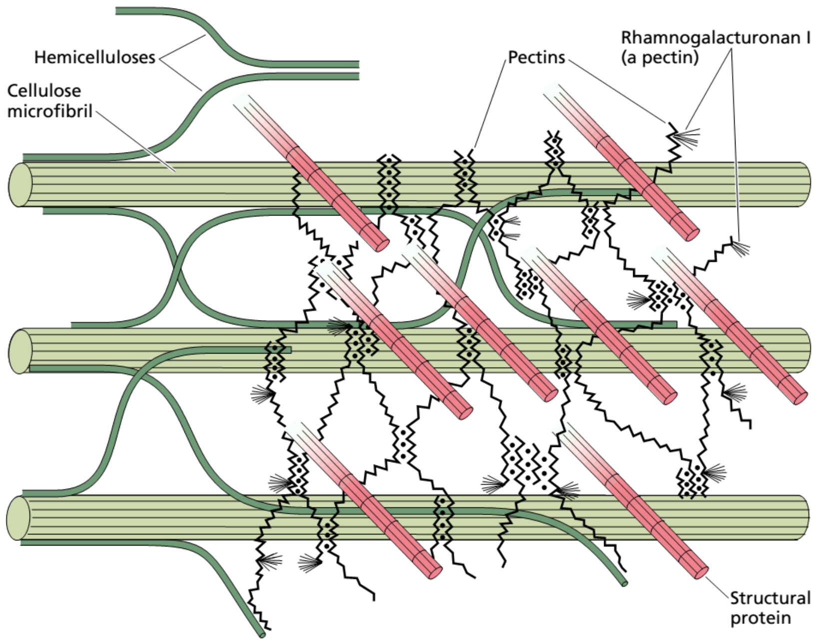


Figure 2: Schematic diagram of the possible arrangement of structural components of the primary cell walls. Cellulose microfibrils are strengthened by hemicelluloses. The hemicelluloses would be crossed-linked to other microfibrils. Pectins inter-wrap the microfibrils while the structural proteins cross-link to the microfibrils (from Brett and Waldron (1990) in Taiz and Zeiger (2002)).

1.8 Maturity and ripening in fruit and vegetables

Changes in physical properties, such as firmness, are influenced by biological properties of mature or ripening fruit and vegetables. Maturity is defined as a stage that fruit or vegetables have reached their growth and development based on their specific maturity index. The maturity index is a set of quality measurements for a specific produce to verify that the produce are mature and fit to be harvested. Meanwhile, ripening of the produce is defined as the internal structural cells changes after harvest and postharvest towards their senescence and it is subjected to the minimum acceptable quality determined by customers. Common features of the quality components of the fresh produce are discussed in section 1.5 (Brummell, 2010; OECD, 2005; Kader, 1999). Some examples of indices in maturity determination for fruit and vegetables are as follows: (1) a growth and development calendar of a particular produce that is elapsed days from full bloom to harvest, (2) physical appearances such as surface morphology and structure, size, shape, solidity, specific gravity and colour, (3) textural properties such as firmness and tenderness and (4) compositional factors such as start, sugar, acid, juice, oil and/or internal ethylene contents (Reid, 2002; Kader, 1999).

Ripening process may begin when fruit and vegetables have completed their maturity stage or after postharvest. During ripening, the produce changes physically and biologically and these changes are complex. Some of the main changes are (1) starch-sugar conversion, where degradation of carbohydrate component and accumulation of sugar that brings the sweetness in the fruit and vegetables, (2) softening in flesh due to textural properties changes, (3) modification of intercellular air space volume, (4) colour changes, (5) release of aromatic volatile compounds, and (6) organic acid formations that also influence the flavour. After these processes have occurred, senescence will take place (Ruiz-May and Rose, 2013; Andrade Júnior and Andrade, 2012; Brummell, 2010; Negi and Handa, 2008; Nunes, 2008; Paliyath and Murr, 2008; Grotte et al., 2007; Vicente et al. 2007; Brummell, 2006; Knee, 1993; Taylor et al., 1993; Woodmansee et al., 1959).

The first three major changes frequently characterise the ripening processes in fruit and vegetables. This is because they associate with the softening of the produce and affect the structure of the cellular cells that lead to their mechanical changes in parenchyma cells. The following subsection elaborates the three changes of cell structures: (1) the conversion of starch-sugar compositions in the cells influencing the loss of turgor of the flesh, (2) softening being resulted by the textural changes due to the loss of cell wall rigidity and (3) the loss of cell-cell adhesion of middle lamella (Brummell, 2010; Abbott, 2004; Waldron et al., 2003; Kader, 1999).

1.8.1 Loss of turgor

Unripe plant tissue structures are filled with starch (carbohydrate compositions) that influences firmness of the fruit and vegetables. However, as the starch starts to degrade and sugar accumulates, parenchyma cells and the intercellular air spaces are flooded with fluid (viscoelastic medium). The conversion of starch-sugar compositions occurs as follows (Figure 3). The starch is hydrolysed by α and β-Amylases and α-Glucosidase. The α-Amylase hydrolyses the starch compositions α -1, 4-linked glucose residues to glucose, one of the sugar contents in ripe fruit and vegetables. Meanwhile, the β-Amylase hydrolyses the glucan chain to maltose. Later on, the maltose is hydrolysed to glucose by the α-Glucosidase, maltase enzymes. The change of the compositions causes softening of the cell tissues, lead to the losses of turgor and reduce the flesh firmness (Negi and Handa, 2008; Taiz and Zeiger, 2002).

1.8.2 Loss of cell wall rigidity

Cell wall disassembly is another influencing modification of textural properties that causes the softening of the flesh and then the decreasing in firmness. Mainly, three modifications are mainly involved in the primary cell wall degradation: pectin and matrix glycan solubilisation, methylesterification, and depolymerisation (Figure 3).

(A) Pectin and matrix glycan solubilisation. Pectin solubility occurs when either pectin galactan or arabinan loses its side chain, or pectic galactan or arabinan disappear and/ or another pectin molecule is developed. The increasing of pectin solubility increases the swelling of cell wall. As a result, the size of cell wall porosity increases. Further, this porosity change generates more enzyme mediating degradation. The porosity causes the cell wall easily to be sliced open easily and become more hydrophilic. Hydrophilic environment further assists the enzymes to access their substrate. Thus, more polysaccharide matrices are disassembled. Pectin solubility modification affects the viscoelastic of the cell wall (effect of solid-liquid composition) (Ruiz-May and Rose, 2013; Cosgrove and Jarvis, 2012; Brummell, 2010; Goulao and Oliveira, 2008; Negi and Handa, 2008; Paliyath and Murr, 2008; Vicente et al., 2007; Brummell, 2006; Waldron et al., 2003; Taiz and Zeiger, 2002; Rose and Bennett, 1999; Brett and Waldron, 1990; Van Buren, 1979).

(B) Pectin and matrix glycan depolymerisation. Depolymerisation of pectins and glucan matrix causes the decrease in cell wall rigidity. This change causes the cell walls swelling due to the increasing of pectin solubility. Consequently, the cells fill with liquid-solid mixture (Ruiz-May and Rose, 2013; Cosgrove and Jarvis, 2012; Brummell, 2010; Vicente et al., 2007; Brummell, 2006; Harker et al., 1997; Rose and Bennett, 1999; Van Buren, 1979; Keegstra et al., 1973; Northcote, 1972).

(D) Pectin methylesterification. Pectin methylestification is also reduced during softening. This change affects the rigidity of peptic network, properties of cell walls and alteration of diffusion and enzymes activities within the wall spaces.

(E) Stress-relaxation and creep*.* Recently, studies have been investigating on another possible process of the cell wall degradations. To expand/ elongate cellular cells after maturity stage, the cell walls must lose their rigidity. Wall loosening proteins focussing on expansins are claimed to be responsible on this cell wall acidification (degradations). The acidification causes hydrogen ion coming out from plasma membrane. This modification leads to the wall stress, and causes the turgor pressure of the cells to increase. To counter-balance this stress, the polymer matrix, xyloglucans, lose their chains (yet, not cut). Thus, they are no longer able to coat the cellulose microfibrils. As a result, the microfibrils slip and creep to each other. Consequently, the surface area increases and the physical stress is reduced (Cosgrove and Jarvis, 2012; Negi and Handa, 2008; Taiz and Zeiger, 2002).

1.8.3 Loss of cell-cell adhesion

The collapse of middle lamella is another responsible reason for the cell tissues losing their firmness. Middle lamella maintains the structures, positions, and stability of cell tissues, by adhering neighbourhood cells. Middle lamella is made up of pectin substances. In unripe plants, the middle lamella is more intact because the pectin substance exists as insoluble water compound and this compound is referred as proto-pectin. The detachment of the middle lamella starts when acid catalysis hydrolyses 1-4 glucotoronic acid and methyl ester by splitting the glycolic acid (Figure 3).

As a result, the former pectin substances become water soluble and are known as pectin. Due to the hydrolysis, the middle lamella losses the adhesion and starts to loss the strength. Hence, the cell structures collapse due to the structural instability (Hong et al., 2013; Waldron et al., 2003; Van Buren, 1979; Keegstra et al., 1973; Northcote, 1972; Woodmansee et al., 1959).

The starch-sugar conversion, the cell wall disassembly and the collapse of middle lamella affect the cell volumes in the cell tissue composition and subsequently influence the turgor pressure level. The correlation between the turgor pressure and the changes of the volumes corresponds to the degree of elasticity of the tissue. This elasticity is the rigidity of the tissue samples that associate the firmness of fruit flesh (Taiz and Zeiger, 2002; Pattee, 1985).

Table 9: Examples of maturity indices for designated fruit and vegetables (Reid, 2002)

Indices	Examples of fruit and vegetables
Elapsed days from full bloom to harvest	Apples, pears
Physical appearances such as	
surface morphology and structure,	Gloss of some fruit
size, shape,	All fruit and vegetables
solidity,	Lettuce, cabbage, Brussel sprouts
specific gravity and	Cherries, watermelons, potatoes
colour (external and internal)	All fruit and vegetables
Textural properties such as	
firmness and	Apples, pears, stone fruit
tenderness	Peas
Compositional factors such as	
starch, sugar,	Apples, pears,
acid,	Citrus, papaya, melons, kiwifruit
juice,	Citrus fruit
oil and /or	Avocados
internal ethylene contents	Apples, pears

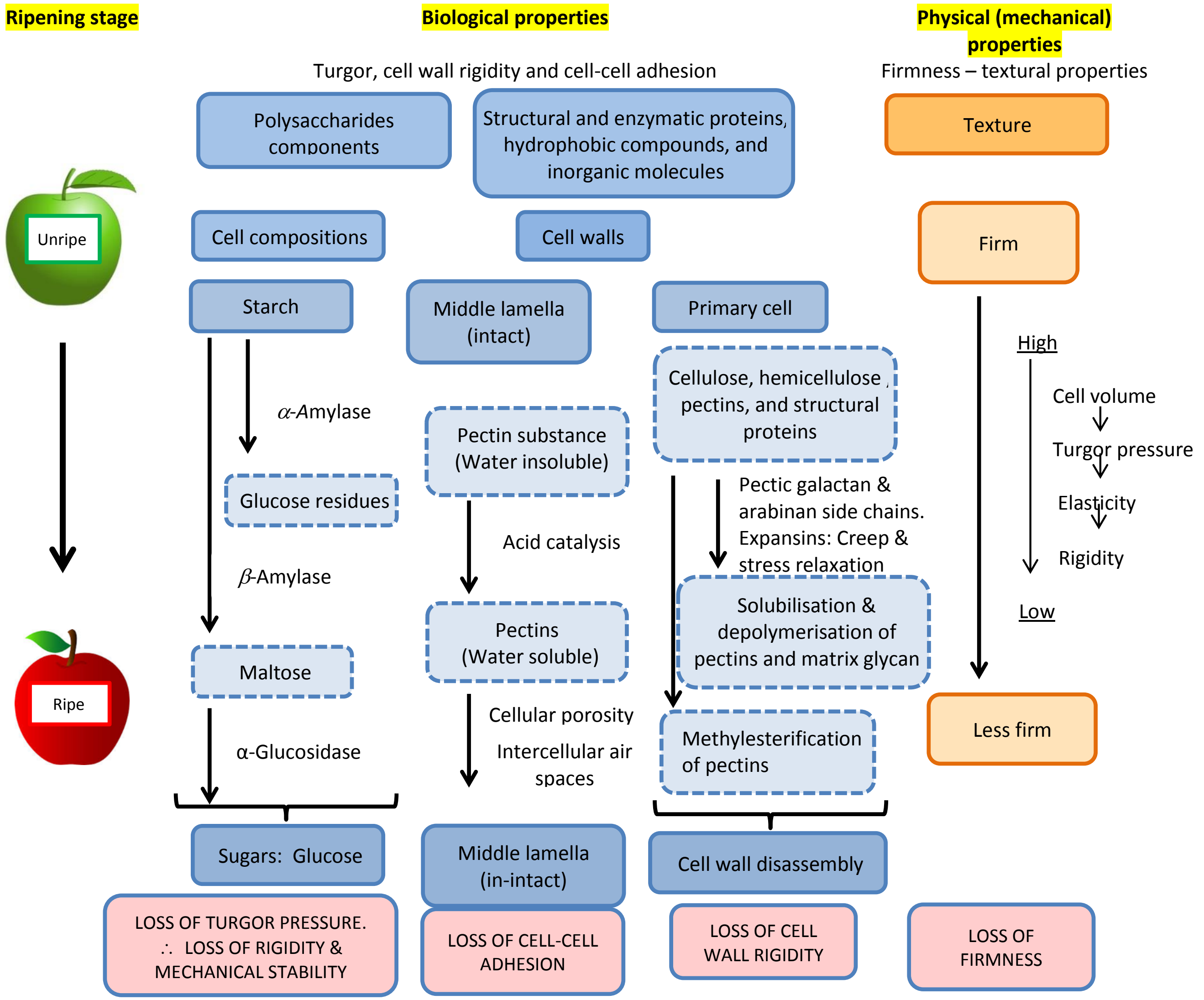


Figure 3: Interrelationship between biological and mechanical (physical) properties during ripening stages

1.8.4 Gas change during ripening: Apples as an example of the changes of cell structure during maturity and ripening

A study of Tukey and Young (1942) on the cell tissue structure changes of *'McIntosh'* from 1 month before full bloom to 4 months after bloom (ripe), demonstrates that the texture of the fruit is associated with its maturity and ripening stages. Six development stages of three different parts of the cell structures (1. Fleshy pericarp, 2. Pith, and 3. Cortex; in Figure 4) are selected and displayed in Table 10 to illustrate changes in size and shape of developing cells. At the stage of 1 month before full bloom and full bloom, cells of the fleshy pericarp, pith, and cortex are relatively round and small. Then, during the first and second months after full bloom, the sizes of the cells enlarge, and their round cells and intercellular air spaces become prominent. As the apple reached its ripe stages, the cells elongate radially. Similar findings of Skene (1962) in Jackson (2003) are reported on the changes of shape of apples' flesh tissue including the change of intercellular air spaces from Day 0 to Day 105 for 'Brownlees Russet' apple and 'Cox's Orange Pippin'. During the pre and postharvest stages, the cells have changed more rounded to elongate. Spots of intercellular air space also are detected. Moreover, the cells are more intact to their neighbours from Day 0 to Day 29. However, the cells detach from each other after Day 47 and the disassembly becomes more obvious until Day 105. These changes are more obvious in collenchyma cells (collenchyma's role is to support and ensure integrity of the plant structures) and in the intercellular air space due to the softening effect during ripening stage.

Apples show anisotropy effects in their cell structure arrangements. Shapes and sizes of the structures from a cross section are non-homogenous, depending on the location of the flesh, whether it is near the skin or the core of the fruit. The cells near the skin are smaller and rounder than the cells far from the skin. The cells get more elongated as the flesh goes deeper to the core of the fruit. Sizes of the air intercellular also changes during the ripening. Contrarily, the cell structures from the radial section of an apple are more homogeneously arranged in shape and size (Reeve, 1953).

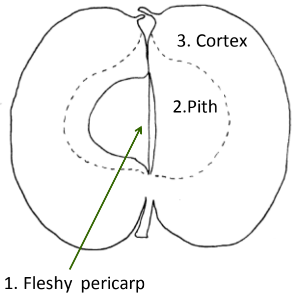
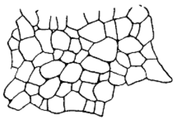
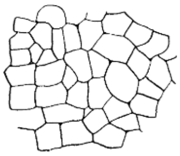
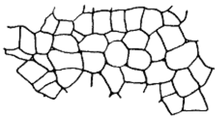
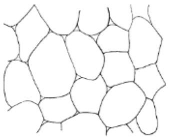
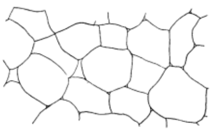
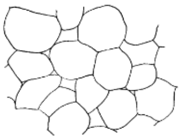
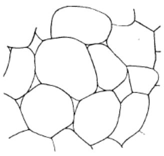
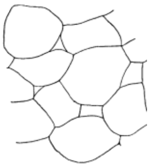
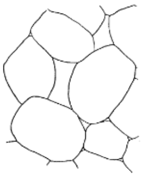
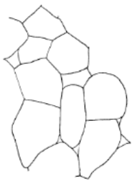
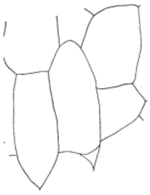
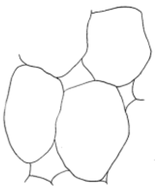
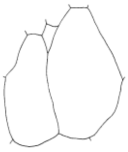
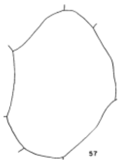
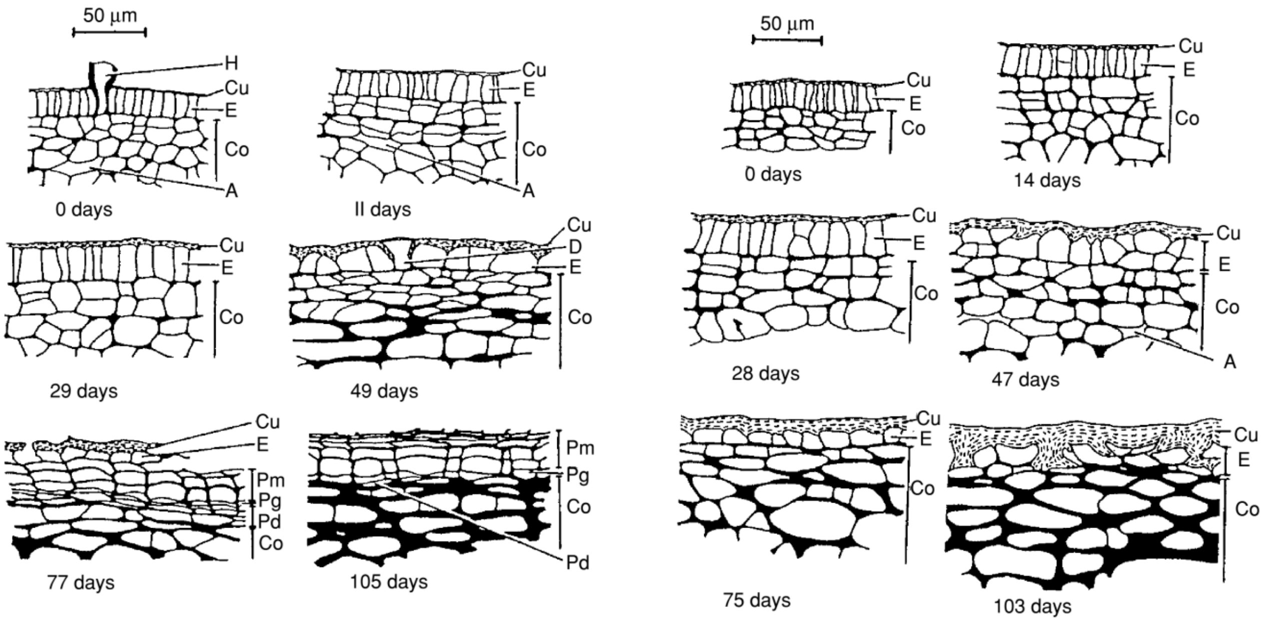


Figure 4: Axial cut section (along the axis) of an apple; position of 1. Fleshy pericarp, 2. Pith and 3. Cortex

Table 10: Six development stages of a *'McIntosh'* apple; 1 month before full bloom; full bloom; 1 month after full bloom; 2 months; 3 months; 4 months (ripe) (Tukey and Young, 1942)

Development stages of an apple	Cellular changes in *'McIntosh'*		
	1. fleshy pericarp	2. pith	3. cortex
1 month before full bloom			
Full bloom			
1 month after full bloom			
2 months			
3 months			
4 months (ripe)			



A. ‘*Brownlees Russet*' apple

B. '*Cox's Orange Pippin*’

Figure 5: Radial cut sections of A. ‘Brownlees Russet' apple and B. 'Cox's Orange Pippin’ showing the changes of shapes of the fruit flesh cells (H:hair base; Cu:cuticle; E:epidermis; Co:collenchyma or hypodermis; A:airspace; Pm:phellem (cork); Pg,phellogen (cork cambium); Pd,phelloderm (inside cork cambium) (from Skene (1962) in Jackson (2003)

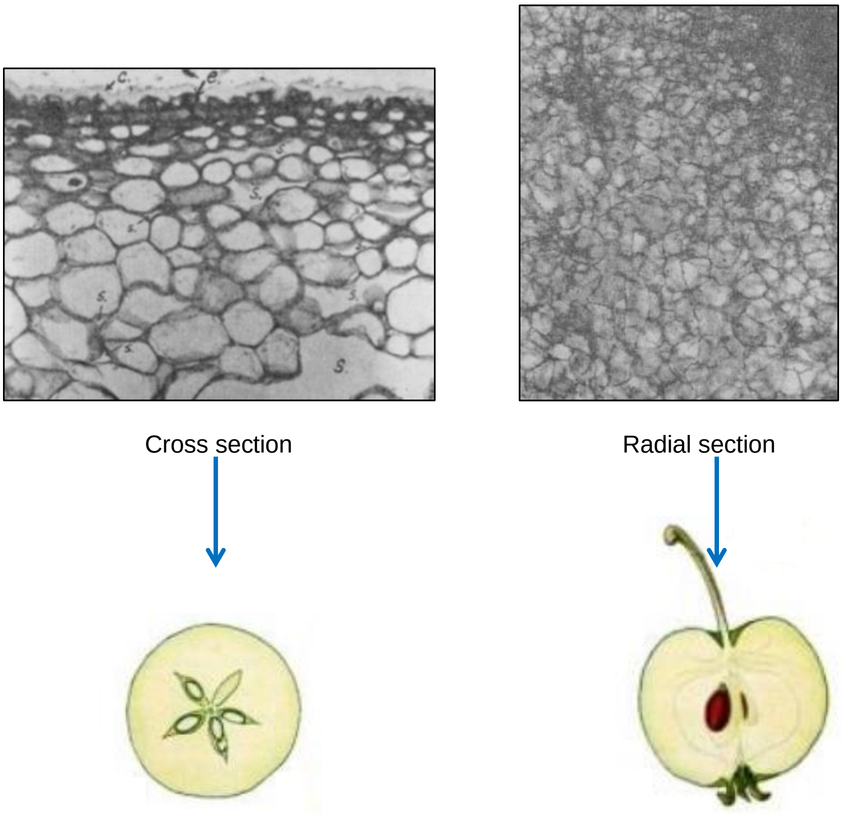


Figure 6 : an apple structure cut crossly (x150, c-cuticle, e-epidermis, s-intercellular space) and radially (15microns) under microscope (Reeve, 1953)

1.9 Instrumental destructive and non-destructive techniques of evaluation of quality of fruit and vegetables

Food stakeholders have implemented various techniques in evaluation on the quality of fresh fruit and vegetables. Most of the techniques are destructive. However, non-destructive techniques are few. Table 11 is a summary of major categories of the destructive and non-destructive techniques used in determining the textural properties for foods (fruit and vegetables). Three technologies (electromagnetic, electrochemical, and mechanical) comprise these techniques followed by characterisations, applications, and examples of instruments for each technique. A progressive trend has been observed towards a combination of several destructive and non-destructive techniques to analyse fresh produce quality (Zou et al., 2016; Zou and Zhao, 2015; Nicolaï et al., 2014; Zdunek et al., 2014; Alfatni et al., 2013; Chen and Opara, 2013; Schmilovitch and Mizrach, 2013; Parker et al., 2010).

Mechanical technology has been widely used to assess the textural properties of fruit and vegetables. Firmness is commonly one of important quality measurements that is related to maturity, ripeness, or internal defects. The storage quality can be determined after postharvest throughout their supply chain transactions. Thus, the firmness measurement can be the main option to verify the optimum quality during storage of fresh fruit and vegetables (García-Ramos et al., 2005). Nevertheless, firmness of fruit is commonly evaluated by destructive measurements (mechanical technologies). The measurements require puncturing, slicing or cross sectioning a product to inspect the interior quality. As a result, no quality measurement can be conducted twice on the same sample during postharvest shelf life (no repeatability and reproducibility). Consequently, statistical data from the destructive techniques may contribute to large uncertainty ranges of quality (Alfatni et al., 2013; Fawole and Opara, 2013; Abbott, 1999; Wallace, 1946).

Studies have shown that non-destructive measurement techniques have been an increasing trend for evaluation of postharvest quality and shelf life of fresh fruit and vegetables together with destructive techniques. Examples of non-destructive techniques based on textural properties parameters of selected produce are presented in Table 12 with the given references (Zou et al., 2016; Zou and Zhao, 2015; Zdunek, 2013; Kilcast, 2013). Apart from the list in the table, other studies also discuss on the similar non-destructive measurements for the produce (Zou et al., 2016; Nicolaï et al., 2014a; Zdunek et al., 2014; Alander et al., 2013; Lee and Cho, 2013; Awad et al., 2012; Diezma and Ruiz-Altisent, 2012; Nourain, 2012; Cubero et al., 2011). These literatures show that the non-destructive technologies are feasible but they have not yet widely been explored.

Ultrasound techniques provide a suitable option in assessments of quality for fruit and vegetables as opposed to the other non-destructive techniques. Ultrasound techniques offer rapid and accurate online sensors, cost and energy effective methods (economical), and simple operations. In contrast, optical techniques (such as machine vision, Vis/NIR and Hyperspectral Imaging Detection) are expensive, lengthy operations and require highly trained and skilled employees due to complicated data analyses and interpretations. Bio-sensor techniques (such as electrical e-nose) show limited detection sensitivity. Instruments and operations using radiation techniques are also costly (Aboonajmi and Faridi, 2016; Aboonajmi et al., 2015; Zou and Zhao, 2015; Alfatni et al., 2013; Schmilovitch and Mizrach, 2013).

Table 11: Destructive and non-destructive techniques used in evaluating fresh fruit and vegetable quality

Technologies	Techniques	Characterisation	Application	To detect, e.g.	Instruments	References
Electromagnetic	Optical	Optical properties Reflectance Transmittance Absorbance Scatter of light Ultra violet –Visible (UV-Vis) Infra-Red (IR) Far Infra-Red (T-Rays) Near Infra-Red (NIR) Colour Chemical bonds	NIR region Water Carbohydrates – starch ,soluble solid, acids Fats -oil protein Visible range Chlorophylls, carotenoids, anthocyanin and other coloured compounds	Bruises Chilling injuries Scald Decay lesion	Colorimeter Spectrometer, spectrophoto-meters	(Sánchez et al., 2013; Zou et al., 2010; Valente et al., 2009; Moros et al., 2006) Ragni et al., 2010; Lallu and Burdon, 2007; Burdon et al., 2002
	Florescence and	Excitation of high energy light (short wavelength) Relaxation of low energy light (longer wavelength)	Maturity Chlorophyll florescence -photosynthetic activity in plant leaves -degradation of chlorophyll in fruit/vegetables		Chlorophyll fluorescence	(Cen et al., 2013; Ragni et al., 2010; Montefiori et al., 2009; Silva et al., 2007)
	Delayed Light Emissions (DLE)		Thykoloid membrane Electron transport Proton pumping of	Chilling injury Stress response	Pulse amplitude modulated (PAM)	(Montefiori et al., 2009)

Technologies	Techniques	Characterisation	Application	To detect, e.g.	Instruments	References
			ATPase pH gradients			
Electromagnetic	X-Ray	Intensity of energy Incident energy Absorption coefficient Density of product Sample thickness	Anatomical and physiological changes Internal disorders	Cell breakdown Water distribution and binding Decay Insect infestation *Apple* Cork spot Bitter pit, Water core and brown core *citrus* blossom end decline membranous stain black rot seed germination freeze damage *potato* hollow heart bruises black heart	2 dimensional radiography (line scan) X-ray computed tomography (CT)	(Aguilera and Stanley, 1999)
	MRI (Magnetic Resonance and Magnetic Resonance	Magnetic moment of nuclei RF	Biological state of tissues Ratio of bound water	Internal structure Bruising	MRI	(Aguilera and Stanley, 1999)

Technologies	Techniques	Characterisation	Application	To detect, e.g.	Instruments	References
	Image)		to free water			
Electrochemical	Electronic nose	*Aromatic and non-aromatic volatiles* Ethylene, Ethyl esters, acetaldehyde, Ethanol, Acetate esters *Electrical conductivity*	Product's pleasing aroma	Ripening	Electrical sniffer	Irudayaraj and Reh, 2007
Mechanical Related to textural properties	Quasi static Force/ Deformation	*Firmness* Puncture Compression Shear test Elastic property (Young Modulus)	Skin starts to rupture	Chilling injury Maturity Storage-ability Shelf life	Penetrometer testers Cornell firmness tester Texture Analyser	(Artés-Hernández et al., 2007) (Mohsenin, 1977)
	"Finger" technique (compression)					
	Bio yield detection					
	Impact	Force/time Force/frequency spectrum	Impact history during handling, packing and transport	Firmness Bruise resistance	Impact firmness tester Probe impact sensor	(De Ketelaere et al., 2006; Diezma-Iglesias et al., 2006; Shmulevich et al., 2003)

Technologies	Techniques	Characterisation	Application	To detect, e.g.	Instruments	References
	Sonic (acoustic) vibration	Wave propagation velocity attenuation reflection	To evaluate tissue /biological properties of horticulture		Acoustic Enveloped Detector (AED)	(Taniwaki and Kohyama, 2012; Costa et al., 2011; Van Vliet and Primo-Martín, 2011; Arimi et al., 2010; Zdunek, 2010; Salvador et al., 2009; Povey, 2006; Varela et al., 2006; Chen et al., 2005; Duizer, 2004; Fillion and Kilcast, 2002)
	Ultrasonic vibration (acoustic impulse resonance) Velocity of sound transmitting in samples	*Sonic (acoustic)* Excited frequency vibrate amplitude peaks elasticity internal friction shape Size Density Stiffness coefficient		Firmness Ripeness	PUNDIT device Fast Fourier transformation (FFT) signal analysers Laser Doppler vibrometer	(Terasaki et al., 2013; Iwatani et al., 2011; Taniwaki et al., 2010; Taniwaki and Sakurai, 2008; Terasaki et al., 2006; Sakurai et al., 2005)
		ultrasonic vibration transmitted reflected refracted/ diffracted when the waves bump with materials	To evaluate tissue /biological properties of horticulture	bruises	Acoustic Enveloped Detector (AED) PUNDIT device Fast Fourier Transformation (FFT) signal analysers Laser Doppler vibrometer	(Al-Haq et al., 2006; Sugiyama et al., 2005; Muramatsu et al., 1997; Muramatsu and Sakurai, 1996)

Table 12: Non-destructive techniques based on textural properties parameters of selected fruit and vegetables

Technique	Textural measured parameters	Examples of fruit and vegetables	References
Quasi-static force–deformation	Firmness	Fruit and vegetables	(Chen and Opara, 2013)
Impact response	Firmness, mealiness	Apple, kiwifruit and peach	
'Finger' method (compression)	Indentation force	Catfish filet	
Bioyield detection	Bioyield detection	Apple	
Acoustic vibration			
Acoustic impulse resonance	Firmness Texture index	Cabbage Pear, apple	
Laser Doppler	Flesh firmness	Persimmon and peach	
Ultrasonic			
Velocity of sound	Elastic properties Firmness	Kiwifruit	
Video analysis	Rigor mortis	Sturgeon Apple, pear and mandarin	
Nuclear magnetic Resonance (NMR)	Firmness, mealiness	Pear	
Magnetic resonance imaging (MRI)	Softening/Firmness	Kiwifruit	
Waveguide spectroscopy	Firmness	Apple	
Fluorescence	Mealiness	Cucumber	
Visible/near/ mid-infrared spectroscopy	Mealiness, firmness Tenderness	Beef	
		Apple and peach	
Hyperspectral scattering technique	Firmness	Fruit	
Time-resolved (domain) reflectance spectroscopy	Firmness		
		Beef	
Raman spectroscopy	Tenderness	Apple	
Light back scattering images	Firmness		
Machine Vision Online	Mechanical damage Bruise detection	Apples, mushroom, tomato and strawberry	(Zou and Zhao, 2015)
NIR Spectroscopy	Internal defect	Carrot and onion	
	Firmness, internal defect	Apple	

Technique	Textural measured parameters	Examples of fruit and vegetables	References
Hyperspectral imaging	Firmness,	Apples, blueberry and peach,	
	bruise detection	Tomato, pear, mushroom and kiwifruit	
	Pit detection	Cherry	
	Canker	Citrus	
Ultrasound	Firmness	Fruit and vegetables	
	Ripeness	Plums and tomatoes	
	Maturity	Potatoes and Melons	
Sensor combination system			
• An acoustic sensor, a firmness tester, a miniature near- infrared (NIR) spectrometer, and an online hyperspectral scattering system	Firmness	Apples	
• Acoustic impulse resonance frequency sensor and miniaturized VIS/NIR spectrometer Partial least square	Firmness	Apples	
• Machine vision system, NIR spectrophotometer and electric nose	Firmness	Apples	
Other Non-destructive Measurement Technologies			(Kilcast, 2013)
X-rays	Internal defect (hollow heart)	Potatoes	
Raman spectroscopy	Maturity	Tomatoes, potatoes and mangoes	
Nuclear Magnetic Resonance (NMR)	Structure	Apples	
	Fruit quality	Apples, peaches and oranges	

1.10 Ultrasonic techniques in evaluation of fresh fruit and vegetables

Literature has shown rapid growth of research interest on a correlation between food texture and sound. Ultrasound measurement is one of the non-destructive techniques used in selected fruit and vegetables. Ultrasound can be generated at high or low intensity. At high intensity, the ultrasound wave can alter textural properties of a tested medium permanently. Therefore, it is destructive technique. It has been applied in ultrasonic cleaning processes, drilling and emulsifications. Contrarily, at lower frequency, characteristics of ultrasound are influenced by changes of textural properties in a tested medium (along the time). As results, the ultrasound set at low frequency offers non-destructive technique for a tested object (Mizrach, 2011; Povey, 2007; Valero et al., 2007; Sinclair, 2001; McClements, 2005; Self et al., 1992; Dickstein et al., 1990). Ultrasound at a designated low frequency is potentially used as online sensor for evaluation the quality of fruit and vegetables due to its non-destructive measurement, and cost and time effectiveness.

Changes to acoustic wave propagation in the produce may demonstrate firmness and internal quality due to the changes of textural properties during postharvest shelf life. Acoustic propagation is influenced by the changes in the elastic and mechanical properties of materials. The wave propagation is characterised by the material and phase properties of a medium. It also resolves the inconsistency of mechanical test of destructive measurement techniques due to its repeatable and robust features. This non-destructive measurement offers an alternative method (1) as an on-line sensor, (2) simple operations, and (3) cost and time effectiveness due to advances in technology (Aboonajmi and Faridi, 2016; Zou et al., 2016; Aboonajmi et al., 2015; Liu and Feng, 2014; Zou and Zhao, 2015; Nourain, 2012; Awad et al., 2012; Mizrach, 2011; Rastogi, 2011; Duizer, 2004; Cartwright, 1998; Povey, 1998b). Studies have shown a correlation between ultrasonic propagation and ripening quality of fresh fruit and vegetables based on firmness during storage (Mizrach, 2008b; Mizrach, 2007; Bechar et al., 2005; Mizrach, 2004; Mizrach, 2000; Mizrach et al., 1996). This is speculated to be associated with changes in the internal structural properties of the produce during storage (Mizrach, 2007; Flitsanov et al., 2000; Mizrach et al., 2000; Mizrach et al., 1996). Some investigations on the textural measurements by using ultrasound techniques on the selected produce are listed in Table 13. In brief, these studies have showed that ultrasonic propagation parameters are correlated to the internal textural changes in the produce and the testing can be alternative non-destructive measurement of quality for fresh fruit and vegetables.

This ultrasonic technique measurement offers high accuracy and precision in evaluation of tissue quality of fresh fruit and vegetables. The wave is powerful to travel through the produce yet adequately gentle to prevent any destruction to their delicate tissues (Baysal and Demirdoven, 2011; Povey, 2000; Povey and McClements, 1988). Recently, ultrasound techniques have been applied in fresh fruit and vegetables for their quality evaluation during pre-harvest and postharvest periods. The ultrasonic parameters have been studied in some varieties of fresh fruit and vegetables. These measurements aim to look the quality indicators of maturity of the produce such as firmness, ripeness and shelf life (Zou et al., 2016; Nicolaï et al., 2014; Zdunek et al., 2014; Diezma and Ruiz-Altisent, 2012; Mizrach, 2011; Figura and Teixeira, 2007a; Figura and Teixeira, 2007b).

Nevertheless, the application of ultrasound technique of textural quality evaluation of selected fresh fruit and vegetables has not been yet extensively investigated and the range variation of quality indicators is different from one variety of the products to another. Even, studies on the protocol for the development of ultrasonic technique for textural assessment and assessment of ripeness in the fresh produce have not well documented. Furthermore, the research has conducted on only small numbers of varieties of fruit and vegetables (Mizrach, 2011; Zou et al., 2016; Zou and Zhao, 2015; Diezma and Ruiz-Altisent, 2012).

The statistical data from the existing conventional instrumental measurements of textural properties of fresh fruit and vegetables by the food industries are beneficial. However, the drastic demand on fruit and vegetables globally requires adequate selections on the instrumental measurements so that their tested parameters can be correlated to the textural properties and the quality attribute of interests of the produce. Acoustic and optical technologies have been showed in a prominent trend in the latest food industries, due to their potential feasibility on ‘on-line sensor’. These possible on-line methods are suggested to be combined together with the traditional instruments for food textural quality measurements. Consequently, a correlation between the measured parameters and the textural properties of the samples is more presentable to interpret the status of the food quality (Zou et al., 2016; Aboonajmi et al., 2015; Zou and Zhao, 2015; Nicolaï et al., 2014; Takizawa et al., 2014; Chen and Opara, 2013;

Bourne, 2002). Based on the reviews, ultrasonic techniques can be alternative to assess the ripeness and internal quality (textural properties) of fresh fruit and vegetables.

An interdisciplinary research can be further conducted on a combination of multiple areas of study on quality of fruit and vegetables by using ultrasonic propagation parameters, physical and biological property measurements. The literatures imply that challenges in food handling are recognised along the fresh fruit and vegetable chain. The followings are proposed to resolve the challenges: to (1) correctly assess ripeness and monitor the quality of food properties; (2) to combine technique in correlating physiochemical, physical, biological and acoustical properties to gain objective measurement of quality.

Studies show that that the ultrasonic propagation parameters at low intensity (velocity, attenuation and impedance) can potentially measure the outcome of the quality attributes of interest (quality level of fruit and vegetables) through the physical properties (elastic moduli, density and microstructure). The correlation of the dependent and independent variables were moderated by the biological properties (tissue turgor pressure, cell wall properties and cell-to-cell bonding as well as anatomy) of these fruit and vegetables. The biological properties influence the strength of the relationship (Zou et al., 2016; Zou and Zhao, 2015; Nicolaï et al., 2014; Zdunek et al., 2014; Ruiz-Altisent et al., 2010; Self et al., 1992). Self and his research team (1992) proposed a conceptual framework for non-destructive ultrasonic technique and its interrelationship of physical and biological properties of fruit and vegetables including their potential measured parameters. In summary, it has been learned that the interdisciplinary research of these three fields for fruit and vegetables has not widely been explored.

Table 13: Applications of ultrasonic techniques on selected fruit and vegetables

Examples of fruit and vegetables	Textural property measurements	Ultrasonic instruments	Frequency, kHz	Ultrasonic parameters		Reference
				Velocity, m/s	Attenuation, dB/mm	
Avocado *‘Ettinger”*	Shelf life, Flesh firmness	Ultrasonic pulser-receiver (Krautkramer Model USL33)	50	200 – 400	2.5–5.0 (0 to 300 hours)	(Mizrach et al., 1996)
			50	-	2.5–5.0 (0 to 30 days)	(Flitsanov et al., 2000)
Avocado *’Fuerte’*	Flesh firmness	Ultrasonic pulser-receiver (Krautkramer Model USL33)	50	105 – 450	-	(Mizrach and Flitsanov, 1995)
	Flesh firmness, ripeness	Ultrasonic pulser-receiver (PUNDIT, CNS Electronics Ltd.)	37	270 – 350	-	(Self et al., 1994)
Avocado *‘Ettinger” and ‘Fuerte’*	Maturity	Ultrasonic pulser-receiver (Krautkramer Model USL33)	50	-	4.5 – 3.0 (July to Nov 1996)	(Mizrach et al., 1999)
Carrots *‘Daucus carota L. cv. Tamino’*	Flesh structural changes during storage	Ultrasonic pulser-receiver (PUNDIT, CNS Electronics Ltd.)	37	396 – 406	1.2 – 1.5 (0 – 19 days)	(Nielsen et al., 1998)
Korean apples *‘Malus pumila, cv. Sansa’*	Flesh firmness	Ultrasonic pulser-receiver PUNDIT 6 (CNS FARNELL Inc., US)	100	200 - 100 (0 – 25 days)	1.2 – 1.5	(Kim et al., 2009)

Examples of fruit and vegetables	Textural property measurements	Ultrasonic instruments	Frequency, kHz	Ultrasonic parameters		Reference
				Velocity, m/s	Attenuation, dB/mm	
Oranges *'Navelina'* *'Ortanique'*	Peel firmness	A harmonic wave function generator (Agilent model 33220A, Agilent Technologies Canada, Mississauga, ON, USA)	40	120 – 200 (0 – 70 days)	-	(Jiménez et al., 2012)
Orange *'Lane-Late', 'Valencia-Late', 'Fortune', 'Ortaniqu'e, 'Nave- lina' and 'Salustiana'*	Peel firmness	A harmonic wave function generator (Sony AFG320)	200	Varies ~ 100 – 200 (0 – 15 days)	Varies ~ 2.0 – 4.0 (for ambient and chamber condition)	(Camarena and Martínez-Mora, 2006)
Plum '*Royal Z'*		Ultrasonic pulser-receiver (Krautkramer Model USL33)	50	-	~ 3.5 – 2.5 (0 – 70 days)	(Mizrach, 2004)
Tomatoes *'Lycopersicon esculentum Mill. cv. 870'*	Firmness	Ultrasonic pulser-receiver (Krautkramer Model USL33)	50	-	~ 3.8 – 2.8 (0 – 9 days)	(Mizrach, 2007)

1.11 Conceptual research framework and aim of the research

A summary of the scope of the research based on the literature review is displayed in Figure 7 with the highlighted areas in the respective boxes. Meanwhile, Figure 8 shows an integrated conceptual framework of the research to describe ripening and internal quality of fresh fruit and vegetables. The integrated approach was a combination of measurements of ultrasonic propagation parameters, mechanical and biological properties. Next, the theoretical principal and hypotheses of the correlations among these three properties were outlined in Figure 9. Therefore, the Research Objectives (RO), Research Questions (RQ) and hypothesis (RH) had been formulated for this research as follows:

RO 1	To develop and optimise a methodology using the PUNDIT Plus Transducer system measurement as an alternative to non-destructive techniques for the assessment of ripeness in apples during storage
RQ 1	How can the PUNDIT Plus Transducer system measurement be characterised and optimised so that ripening stages and internal quality can be assessed?
RO 2	To investigate the correlation between the ripening stages and the anisotropy in apples during storage, and the ultrasonic velocity techniques together with firmness and sugar content measurements
RQ 2	How do (1) the changes in ripeness and (2) anisotropy in apples during storage affect the ultrasonic velocity, together with firmness and sugar content measurements, and (3) the correlation among those quality measurements using the different testing?
RO 3	To investigate the relationship between Brown Heart (BH) in swede and the velocity of sound, together with firmness measured by a puncture testing.
RQ 3	How does BH in swede affect the velocity of sound and firmness measurements?
RH	If the changes of ripening and internal flesh quality influence the ultrasonic velocity together with firmness and sugar content measurements, the ultrasonic technique is feasible as alternative non-destructive measurements for the assessment of ripeness in the apples and detection of brown heart in swedes.

GENERAL TITLE

AREA OF RESEARCH:
(Scope)
Sub issue 1:
Food quality attributes

Sub issue 2:
Sensory attributes

Sub issue 3:
Textural properties

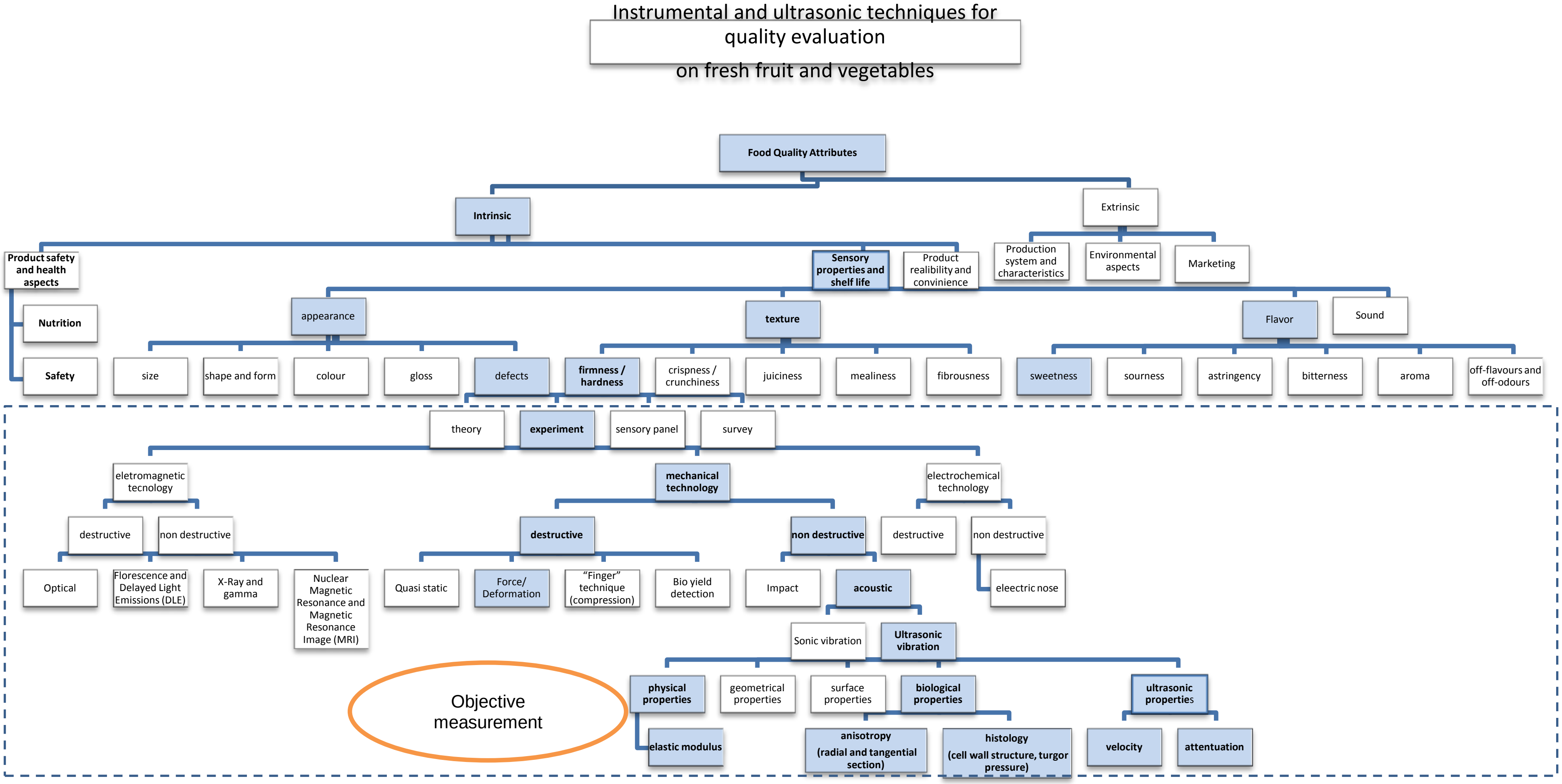
SUB-AREA:
Firmness

METHODOLOGY:
(Techniques)

GAP:
A problem to be solved

ACHIEVEMENT:
(Results)
Quality properties and characteristic parameter

- Correlation between ultrasonic velocity, firmness and sugar content; and ripeness/ internal quality of fresh fruit and vegetables



Thesis:

Journal / conference / poster 1: Development / Application of ultrasonic technique for quality evaluation of fresh fruit and vegetables

Journal / conference / poster 2: Comparative instrumental analysis between ultrasonic and mechanical techniques of firmness for quality in fresh fruit and vegetables.

Figure 7: My research scope. The highlighted areas discussed in the introduction.

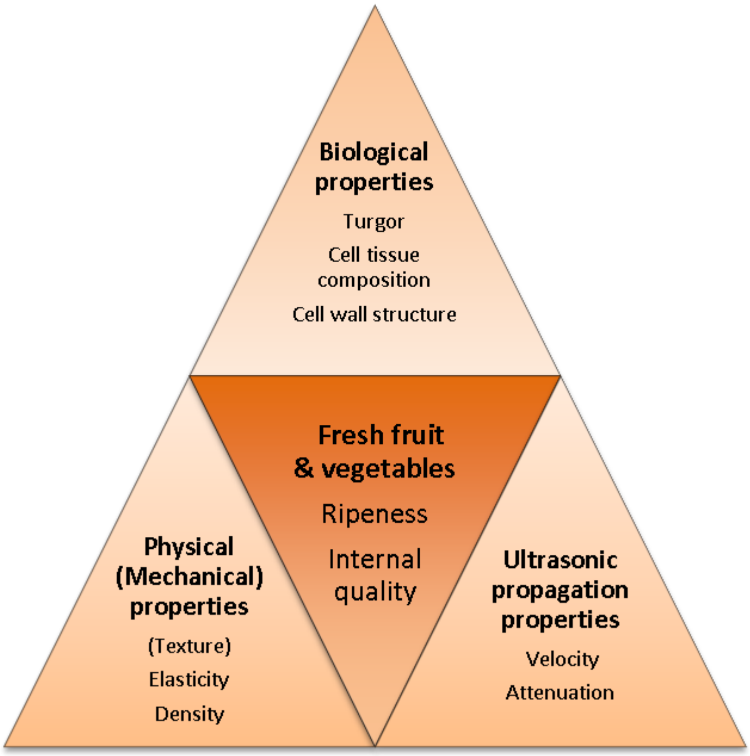


Figure 8: **An integrated conceptual framework of my research on fresh fruit and vegetables quality (selected parameters): Ultrasonic propagation parameters, mechanical and biological properties**

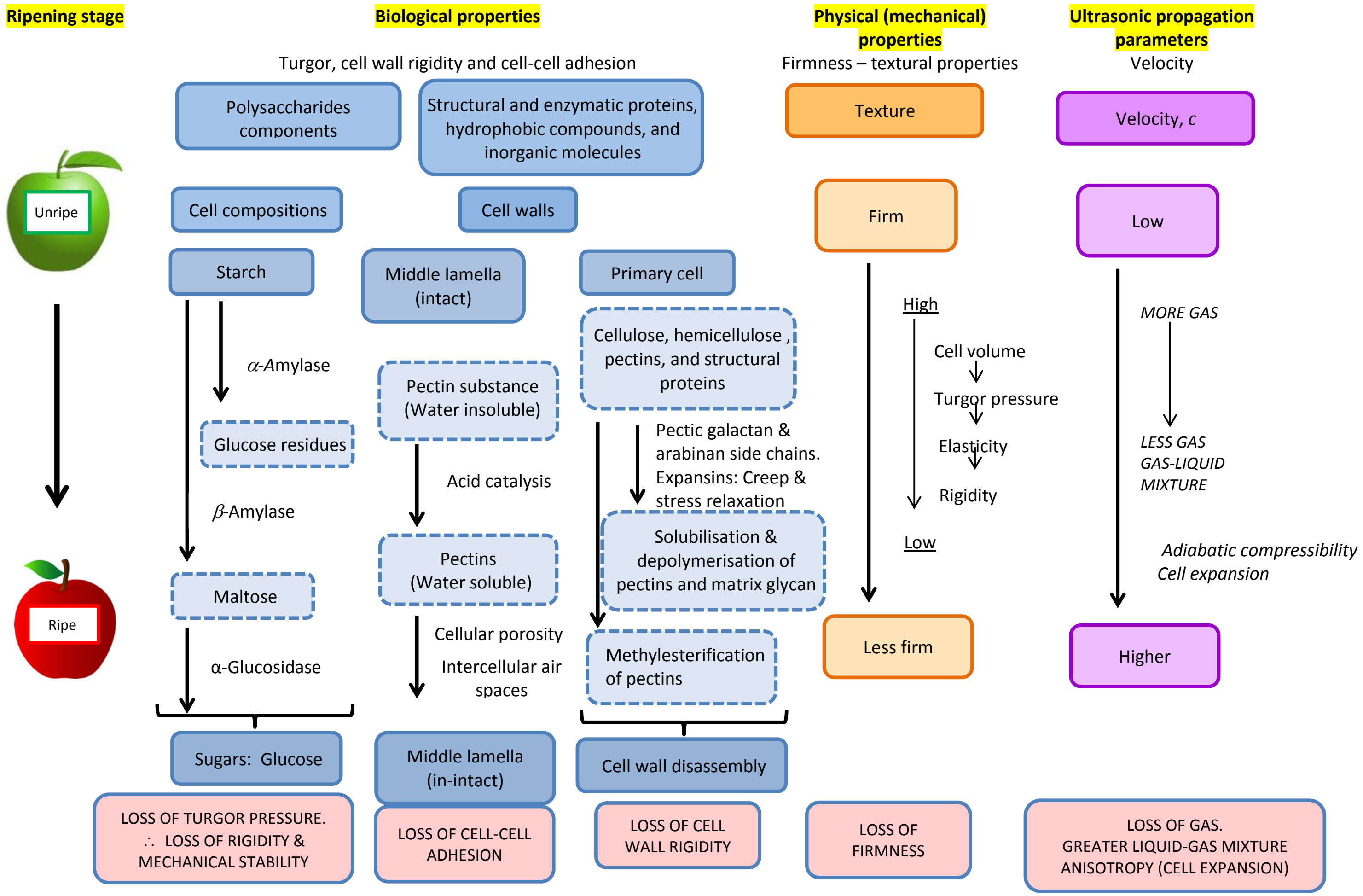


Figure 9: Relationships between biological, mechanical/ physical and ultrasonic propagation properties (velocity) to describe ripening and internal quality:

Chapter 2
Materials and methods

The thesis research questions raised in Chapter 1 generated investigations of this research. Chapter Two provides materials and methods of three parts of the investigations.

The first part of the research was the development of a PUNDIT Plus System by using the apples. It aimed to: (1) optimise and characterise the system and generate the procedure protocol, (2) establish systematic errors on the ultrasonic measurement variabilities and (3) investigate an effect of anisotropy of apple parenchyma on ultrasonic velocity (the preliminary experiment).

The second part of the research involved an evaluation of the storage quality of envy apples as an indication of fruit ripeness by using the non-destructive ultrasonic measurement together with three destructive measurements (compression, puncture, and sugar content tests). Changes in ultrasonic velocity, firmness, and sugar content were monitored during the fruit storage as the measured parameters. The evaluation on the fruit ripeness quality was determined by two investigations to test: (1) if there was an effect of anisotropy of apple parenchyma on the ultrasonic velocity based on the direction of measurements, maturity levels and storage temperatures and (2) if there was a correlation between ultrasonic, firmness, and sugar content in the ripening fruits during storage. Principal Component Analysis (PCA) was used in data analysis section to establish a correlation between the ultrasonic propagation parameter (velocity of sound through an apple), the firmness (compression and puncture tests), and sugar content.

The third part of the research was the experimental work on swedes carried by Dr. Jin Chu under the direction of Dr. Melvin Homes using the methodology developed by Mohd Shah and described in this chapter. It referred to detection of BH in swedes by using ultrasound in parallel to the visual and firmness tests. BH is an internal defect where the core of swede flesh floods with water mixtures and the flesh turns brown. The brownness appears in circular to oval spots of few millimetres to centimetres in size. Uniform light yellowish flesh is graded as an acceptable quality for a healthy swede. Consequently, BH affects the saleability of the vegetables. One of the factors causing the development of the internal browning is a nutritional deficiency associated with boron imbalance in soil (Teagasc, 2010; Dixon, 2007; Golob et al., 2002; Dermott and Trinder, 1947). The study aimed to an investigation to test if non-destructive ultrasonic velocity measurement was able to differentiate between healthy and defected swedes.

Materials used in the research were apples (‘*Pink Lady*’, ‘*Royal Gala*’, and ‘*Envy’*) and swedes. Their descriptions of cultivars, scientific names and hybrid parentage of the studied samples are listed in Table 14. The apples and swedes were studied for ultrasonic propagation parameters travelled inside the tested mediums by using non-destructive ultrasound means. The ultrasonic propagation parameters were compared with physical parameters of the produce based on the non-destructive and destructive methods.

Table 14: A description of cultivars, scientific names, and hybrid parentage of the studied samples

Samples	*Cultivar	Scientific name	*Hybrid of	References
‘*Pink Lady*’	a trade mark cultivar of ‘*Cripps Pink*’	*Malus domestica 'Cripps Pink'*	‘*Lady Williams*’ *x* ‘*Golden Delicious*’	(Cripps et al., 1993)
‘Royal Gala’	a trade mark cultivar of ‘*Gala*’	*Malus domestica ‘Gala’*	‘*Kidd's Orange Red’ x* ‘*Golden Delicious*’	(The National Fruit Collection, 2015; Mckenzie, 1974)
‘*Envy*’	a trade mark of '*Scilate*'	*Malus domestica ‘Scilate’*	‘*Royal Gala*’ *x* ‘*Braeburn*’	(Brown and Maloney, 2009)
Swedes or Rutabagas	a root vegetable	‘*Brassica napobrassica’*	Believed to be a cross breeds between turnips (*Brassica rapa var. napobrassica*) *x* wild cabbages (*Brassica oleracea*)	(Benedict et al., 2013; Undersander et al., 1992)

*Cultivar is a plant that is raised based on their selected characteristics. These selected characteristics are distinguishable, homogenous, consistent, and replicable. The raise must be replicable. A hybrid plant is a product by crossbreeding of other two grouped plants (Brickell et al., 2009; Brickell, 1999)

The sampling, measurement procedures, data collection, and statistical data analysis of the experiments were also detailed. The three experiments were conducted in the laboratories of School of Food Science and Nutrition, University of Leeds, UK.

2.1 ‘*Pink Lady*’, ‘*Royal Gala*’ and ‘*Envy*’: Characterisation and optimisation of ultrasonic technique measurements

Seven apples for each group were bought at a supermarket in Leeds, UK in February 2014 and they represented ripeness stage at the end of the fruit supply chain. Ripeness of the produce is defined as changes of the internal structural cells after harvest towards their senescence that are subjected to the minimum acceptable quality by customers (ENZAFOODS, 2014; Brummell, 2010; OECD, 2005; Kader, 1999). Each apple was labelled with number from 1 to 7 on the peel. The fruit was marked with A1 at the stem and A2 at the calyx for axial measurements and from R1 to R10 approximately 30° apart for radial measurements. The positions of the measurements are illustrated in Figure 10. Five measurements were repeated from each position of A1 and A2 of the fruit that brought to total 10 measurements for the axial direction. Meanwhile, 10 measurements were taken for the radial direction.

Figure 11 defines the axial direction for ultrasound propagation between the calyx and the stem of the fruit and the radial direction of propagation perpendicular to the calyx-stem axis of the fruit. A portion of the cells represented in a rectangular box of Figure 11 is expanded in Figure 12 to depict of the direction of the ultrasound measurement relative to cell columns and the intercellular air space arrangement of the apple. This shows that the axial direction of propagation is across the axis of the cell columns and intercellular air spaces of the apple. Conversely, the radial direction is propagation along the axis of the cell columns and intercellular air spaces of the apple.

The choice of the direction of measurement was related to anisotropy of an apple cell structure and the axial and radial directions related to apple fracture and its cell structures based on the fracture strengthens level during the biting of the fruit (Khan and Vincent, 1993a; Khan and Vincent, 1993b).

The apple parenchyma (one of the plant tissues) is arranged in a column or manner perpendicular direction to the calyx-stem axis. The arrangement of the cells in column patterns is related to the cells’ morphological growth by cell progressive enlargements, which is more elongated from the core to the skin especially during the maturity and ripening of the fruit. The parenchyma cells are adjacent to intercellular air space. The increase of the size and the elongated shape of these cell columns also increase the intercellular air spaces in the apple tissue. The orientation of the cell columns and intercellular air spaces reveals the structural anisotropy and heterogeneity of an apple (Taiz and Zeiger, 2002; Self et al., 1994; Khan and Vincent, 1993a; Khan and Vincent, 1990; Reeve, 1953).

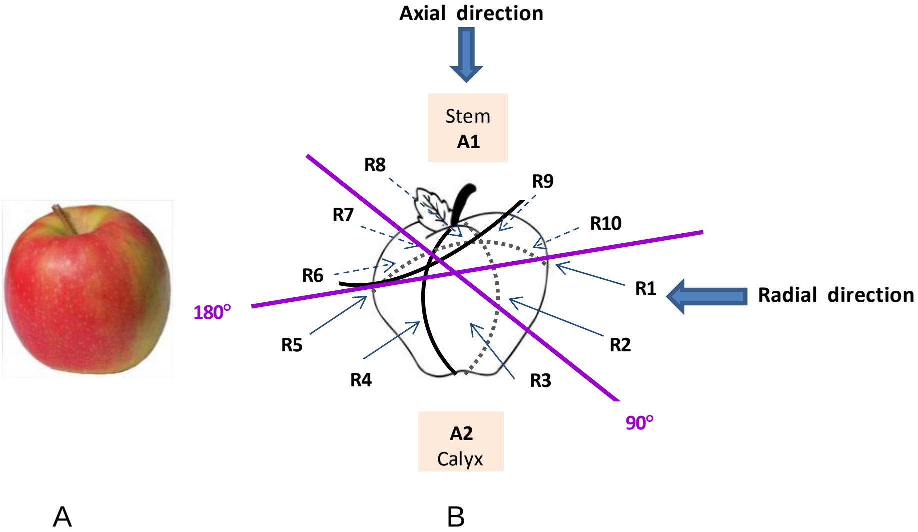


Figure 10: A. Pink Lady Apple. B. Pictorial display of positions of for apple measurements; Axial (A1 and A2) and radial (R1 to R10 in approximately 30° apart) measurements.

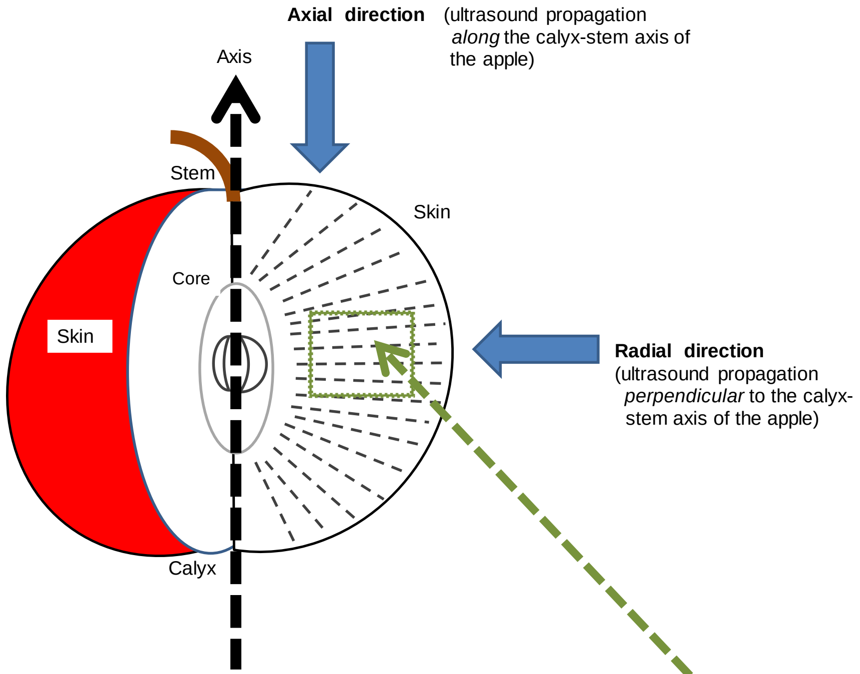


Figure 11: A diagrammatic representation of a whole apple, its orientation of cell structure and two directions of measurements (not to scale) adapted from (Khan and Vincent, 1993b)

- The dashed lines showed the direction of an arrangement of the cell columns and intercellular air space;
- The dashed arrow facing the north defines the calyx-stem axis as the reference axis for the measurements;
- The solid arrow shows the direction of the axial and radial

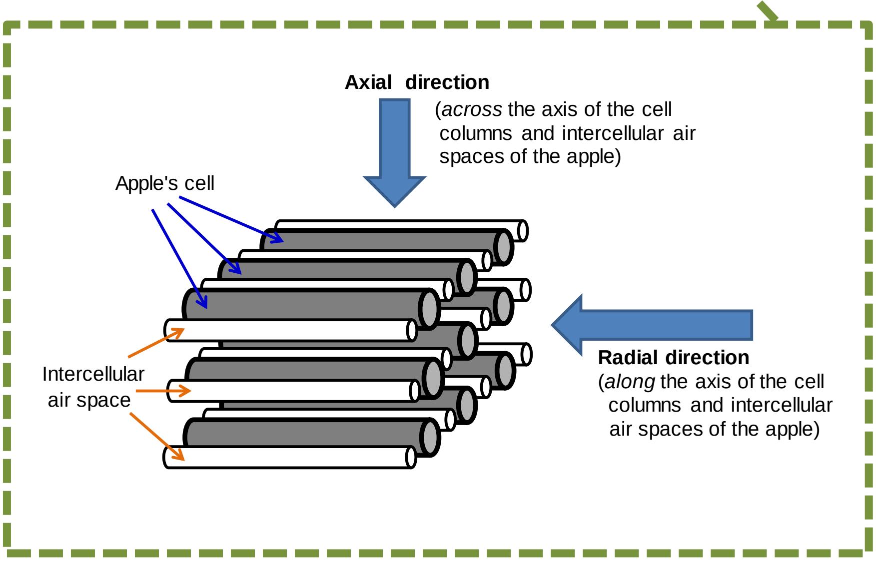


Figure 12: A diagrammatic representation of the arrangement of the cell columns and intercellular air space; and radial and axial directions of measurements of an apple (not to scale) adapted from Khan and Vincent, 1993a

- The dark columns represented the apple's cells.
- The white columns represented the intercellular air space.
- The solid arrow shows the direction of the axial and radial measurements.

The behaviour of the ultrasound propagation through a medium is correlated to mechanical properties. The mechanical properties are influenced by the biological properties of the fruit tissue (Self et al., 1992). Therefore, these two orientations of the apples were hypothesized to have an effect on the ultrasonic and mechanical properties due the changes of biological properties of the fruit samples as they were ripening. However, no experimental study has been conducted yet to test the hypothesis.

2.1.1 Principal of sound (Ultrasound)

2.1.1.1 Mechanical wave and disturbance

Sound is a mechanical wave. Using air as an example, sound is produced when a system in an equilibrium state is disturbed and this disturbance travels as a wave through a medium (air in this case). The disturbance towards the dormant air particles causes displacements of these particles. However, the air particles do not travel. It is rather that the region of the closer particles to the disturbance produces compressed towards each other and increases pressure (compression) whereas the segment of the farther particles to the disturbance is less compressed and less pressure (rarefaction). Next, the pressure of the compressed region is released to the neighbouring segment (rarefaction region) due to a tendency of the system to remain in its equilibrium state. This disturbance creates a chain of mechanical wave transported energy through the air. This travelling wave is detected by ear drum and is interpreted by brain as a sound (Ensminger and Bond, 2012; Young et al., 2012; Halliday, 2011; Giordano, 2010; Kuttruff, 2007).

2.1.1.2 Particle displacement and pressure variation

The disturbance occurred in the medium is associated with particle displacement and pressure variation during the wave propagation. The parallel direction of the compressed and rarefaction particles along the propagated wave indicate that sound is a longitudinal wave. The compressed particles produce a high-pressure zone while the expanded particles produce a low-pressure zone. The displacements and the pressure variations of the two sections of the particles are repetitive and they are described as oscillation motions. The mechanisms on how the sound wave works is illustrated by using water wave as analogy, as shown in Figure 13 (Povey, 1997).

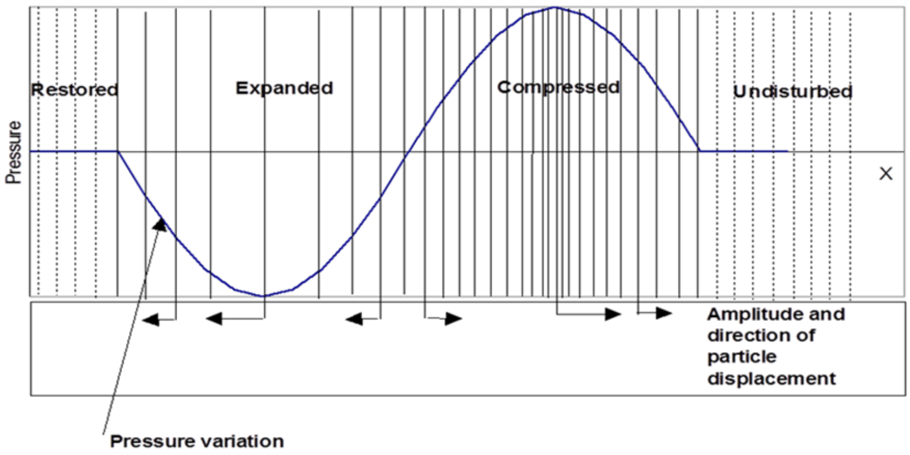


Figure 13: Analogy of sound waves to water waves: Particle position (amplitude), particle displacement and spatial pressure variation as dependent variables against position (x) for a single cycle, sinusoidal, plane-traveling wave in a fluid medium (Adapted from Pierce,1981, in Povey, 1997)

2.1.1.3 Elastic waves

The deformation of the particles experiencing compression and rarefaction motions shows elastic wave property of a material medium during the disturbance of the propagation of wave. Elastic property refers to a tendency of a solid medium to return its original shape and size when it is deformed (restorative force). This wave was the focus of the research. The deformation of an elastic medium is called isotropic when it is independent to direction of a measurement. Contrarily, the dependent deformation of an elastic medium to a direction of measurement is called anisotropic.

Elastic waves comprise bulk and surface waves. Bulk waves propagate inside a material and are independent to its shape. Conversely, surface waves propagate near to surface of a material. Bulk waves can be further characterised by two waves (longitudinal and transverse waves). As mentioned in earlier paragraph, sound is an example of longitudinal waves. Longitudinal waves are which the deformation of particles and pressure variation are parallel or along the direction of wave propagation (Figure 13). Longitudinal wave can travel through solid, liquid, or gas medium. In contrast, transverse waves are which the deformation of particles and pressure variation are perpendicular to the direction of wave propagation. Transverse wave can only travel through solid, not liquid or gas. This is because the displacement of particles in solid in transverse waves causes the material to be bent and restored to its original position (reversible). Contrarily, the displacement of particles in liquid and gas do not happen because both medium flows (irreversible). Meanwhile, transverse waves are associated with a shearing (Ensminger and Bond, 2012; Young et al., 2012; Halliday, 2011; Giordano, 2010; Kuttruff, 2007; Lempriere, 2002; Povey, 1997).

2.1.1.4 Characteristics of sound wave

Basic types of wave. Wave can be pulse and/ or periodic waves. A pulse is a limited or short signal (disturbance) of finite duration (one time). The strength of the pulse is characterised by amplitude. The amplitude is the maximum amplitude (displacement) from the equilibrium position of a sine wave or signal. Meanwhile, the periodic wave is repetitive identical pulses (repetitive pulses) and is characterised by amplitude, wavelength, frequency and period. The following sub-section discusses on these characteristics (Halliday, 2011; Crowell, 2006; Lempriere, 2002; Sinclair, 2001).

Wavelength λ and angular wave number *k*. The pressure wave has a relationship among pressure, distance and wavelength (Figure 14). λ is distance dependent (displacement). It is measured the highest or lowest pressure amplitudes or the distance of two points between one cycle and the next cycle of the oscillating wave in unit of meter m. The number of cycles in a unit distance can be obtained by inverting the wavelength $1/\lambda$. Subsequently, an oscillation of wave is generally expressed by a simple harmonic of sine wave. The position of the two parallel points of the λ moves as the wave moves. A specific wavelength at a specific distance in a sine wave is referred as its angular wave number k. A sine wave completes its cycle by 2π rad. Thus, one complete angular wave number is k multiplied by λ that equals to 2π rad ($k\lambda = 2\pi$). Therefore,

$$k = \frac{2\pi}{\lambda}, \qquad \text{Equation 1}$$

where k is wave number in unit rad per meter, 2π is radian in one cycle and λ is wavelength (Halliday, 2011; Povey, 2007; Lempriere, 2002; Povey, 1997).

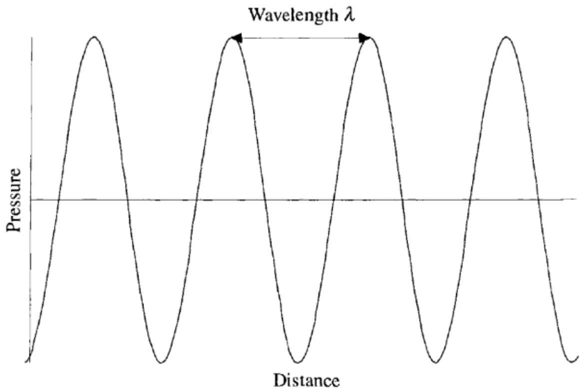


Figure 14: A pressure wave has a relationship among pressure, wavelength and distance (Povey, 1997)

Frequency f, Period T, Angular frequency ω and Phase ϕ. The pressure wave also has a correlation among pressure, wavelength λ and frequency f (Figure 15). **Frequency f** is number of oscillations (repetitive pulses) per unit time (rate of oscillation) in Hertz Hz. A period of time *T* taken for one oscillation over a certain point can be obtained (a pulse) by inverting the frequency $\frac{1}{f}$. Its unit is unit second s. A wave oscillating at a constant angular rate is called a simple harmonic oscillation of a wave (typically in sine wave). The oscillation is represented by magnitude *a(t)* as a function of time *t* (Equation 2). Angular frequency ω is a specific frequency at a specific time in a sine wave,

$$a(t) = A \sin \omega t, \qquad \text{Equation 2}$$

where *t* is time at a certain angular rotation, *A* is amplitude, and ω is the angular frequency. A sine wave completes its cycle by *2π radian*. Thus, one complete angular frequency is ω multiplied by *T* that equals to *2π radian* ($\omega\lambda = 2\pi$). Therefore,

$$\omega = \frac{2\pi}{T} = 2\pi f, \qquad \text{Equation 3}$$

where ω *is* angular frequency in unit radian per second*, 2π* is radian in one cycle and λ is wavelength. Next, a specific rotational angle at a specific time (such as A and B) is referred to its **phase shift ϕ** given by $\phi = 2\pi f t$ (Figure 16). ϕ is also correlated to the angular frequency ω with a given formula of $\phi = \omega\, t$. Therefore, the phase shift has a relationship of formulas,

$$\phi = 2\pi f t = \omega t \qquad \text{Equation 4}$$

(Halliday, 2011; Povey, 2007; Lempriere, 2002; Povey, 1997).

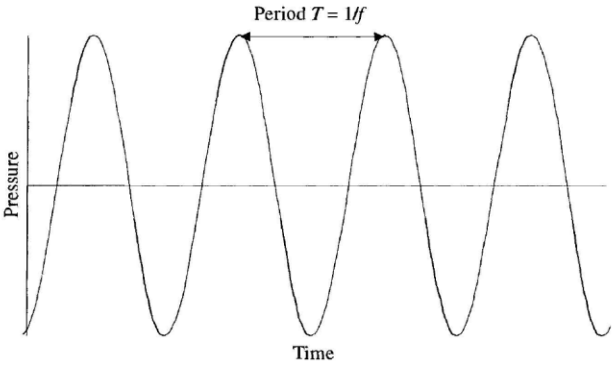


Figure 15: The pressure wave also has a correlation among pressure, wavelength λ and Frequency f (Povey, 1997).

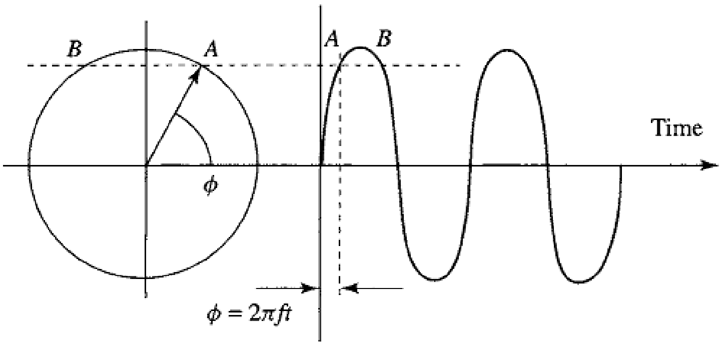


Figure 16: Relationship among phase ϕ, angular frequency f and time t during a rotation 2π *radian* of steady oscillation (Lempriere, 2002)

2.1.1.5 Ultrasonic propagation parameters

Application on ultrasound expands understanding on molecular structure of the medium been studied (foods) (Povey, 2007; McClements, 2005; Povey, 2000; Povey, 1998b; McClements and Gunasekaran, 1997; Povey and McClements, 1988). In the case of fresh fruit and vegetables, this application offers a non-destructive technique in quality evaluation of the produce such as in aspects of maturity, ripening, storing condition and shelf life. The development of ultrasonic technique in fruit and vegetables can be an alternative technique to the existing destructive methods by food industries. Low intensity of ultrasonic frequency is the interesting frequency in this research because the wave propagation does not destroy the textural properties of a tested medium (non-destructive technique). The properties influence the characteristic of propagated ultrasonic wave. Therefore, the information is useful in quality evaluation of fruit and vegetables.

Ultrasonic velocity, attenuation and acoustical impedance are common ultrasonic propagation parameters been used in textural property measurements on fruit and vegetables.

(1) Ultrasonic velocity

Characterisations of the oscillating wave depend on its wavelength and frequency. Velocity c or wave speed is dependent to wavelength λ and frequency f with a formula of,

$$c = f\lambda, \text{ (known frequency)} \qquad \text{Equation 5}$$

or

$$c = \frac{d}{\Delta t}, \qquad \text{Equation 6}$$

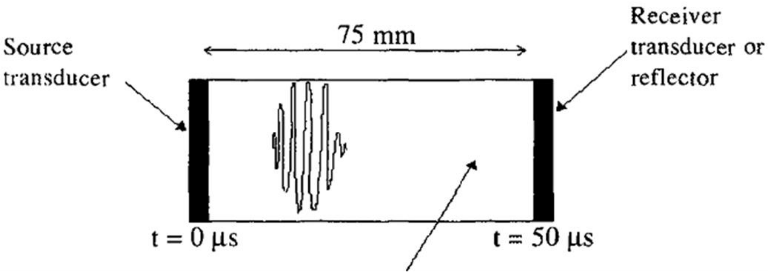
where time Δt is the time between excitation pulse and pulse arrival for a wave to propagate through a medium (distance d) (Povey, 2007; Povey, 2000; McClements and Gunasekaran, 1997).

Wave behaviour of ultrasonic velocity is very useful to access information about the quality stage of fresh fruit and vegetables along their supply chain. An analogy between a pressure wave motions is drawn as in a row of parallel planes and plant cell tissue structures. The plant cells are surrounded by the cell walls that come in different shapes, sizes, and volumes depending on the stage of development of the plants. When ultrasonic pulse triggers the measured plant's skin and through the cell tissues, the ultrasonic energy is transferred from one cell wall to one another. This energy motion also undergoes a series of cycles of restoring, expanding, compressing, and un-disturbing movements. The distance is the length of the ultrasonic pathway of the plant measured between entrance and exit of a contacted distance of the plant's skin (fruit and vegetables). Meanwhile, the time taken for the ultrasonic wave passed through the studied medium is the value measured in the measurements. The longitudinal pressure wave behaves liked an elastic material, because the energy is restored, expanded, compressed and back to undisturbed condition. This behaviour is also true to the plant's cell wall. Hence, this behaviour shows that the ultrasonic testing is a non-destructive technique (Taiz and Zeiger, 2002; McClements and Gunasekaran, 1997; Povey, 1997; Self et al., 1992).

The textural changes during the apple ripening were evaluated based on measurement of ultrasonic velocity from the time of flight and the path distance of the ultrasonic propagating wave through the apple. The velocity of speed measurement of known distance of the propagated wave is calculated by the distance between two transducers d over the time between excitation pulse and pulse arrival Δt for the propagated wave through the tested medium (Equation 6).

The diagram in Figure 17 shows an example of the measurement. The distance d of the two transducers is 75 mm and the time between excitation pulse and pulse arrival Δt is 50 $\mu\mathrm{s}$. Therefore, the velocity is 1500 $\mathrm{m/s}$ (Povey, 1997).

Ultrasonic propagation parameters (such as velocity by using low intensity ultrasound) are influenced by the tested material. In case of fruit and vegetables, their cellular cells of fruit and vegetables are undergone chemical and physical changes through their growth, development, maturity and ripeness and senescence. The cells of the unripe produce consist of solid (starch) and gas composition. Later, as the produce starts to ripen, the cells are flooded with liquid (mixture of starch-sugar composition) and lesser gas volume. As a result, velocity is feasible to give useful information about those changes. It is possible to evaluate the ripeness or the internal defect of the produce after the postharvest.



Velocity in sample equals distance/time = $d/\Delta t$,

for example, 75 $\mathrm{mm}/$50 $\mu\mathrm{s}$ = 1500 $\mathrm{m/s}$ for a single transit

Figure 17: The principles of velocity of speed measurement based on an acoustic pulse-echo apparatus by using an oscilloscope trace (Povey, 1997)

(2) Attenuation

Attenuation is another characterisation of ultrasonic wave propagation. It is influenced by structures of material and an interaction of propagated wave.

Structures of material. In general, most plant's cell structures are non-uniform due to the structure changes by developments, maturity, ripening and quality degradation (senescence). These progresses show that the food material is not completely elastic.

Sound wave carries energy. In the ultrasonic applications, the energy propagates from one cell wall to another (kinetic energy) in the case of plants' cells. Due to the internal friction between the cells, the kinetic energy is converted to heat, referred as absorption. The conversion of the energy influences the strength of the propagated wave speed. In addition, changes in the structures of cell walls during plant development, maturity and ripening (due to non-elastic medium, for example, viscoelastic) also distort the speed of the ultrasonic wave propagation, called dispersion (Lempriere, 2002; Povey, 1998b; McClements and Gunasekaran, 1997; Povey and McClements, 1988).

Interaction with propagated wave. The ultrasonic wave changes its direction of movement as it propagates through the studied material referred as scattering. Hence, the direction change causes lesser receiving wave been detected.

The wave amplitude decreases due to its interaction with the medium (for example, changes in the cell structures of a plant influence the wave propagation characteristics). The change of the signal amplitude as a function of distance called attenuation. Attenuation is expressed by the logarithmic decay in the pressure wave travelled through the studied medium with the ratio of magnitudes of amplitude changes (from the initial amplitude A_0 to the changed amplitude A) in decibels units as the following equation,

$$A = 20 \log_{10}\left(\frac{A}{A_0}\right), \qquad \text{Equation 7}$$

where the factor of 2 is acoustic characterised power such that $\log(A^2) = 2\log(A)$. Then $[2\log(A)] \times 10$ is a factor of 10 from bels to decibels which produces the factor 20. Attenuation is also expressed in Neper unit. 8.685 dB is equal to 1 Neper. Attenuation coefficient α takes place with the following equation,

$$A = A_0 e^{-\alpha x}, \qquad \text{Equation 8}$$

where A is the initial amplitude and A_0 is the changed amplitude. α is the attenuation coeffient and x is the distance travelled by the ultrasonic wave through a medium. This equation is a Beer's law equation (Povey, 2007; Lempriere, 2002; McClements and Gunasekaran, 1997). The diagram of the attenuated wave is illustrated in Figure 18.

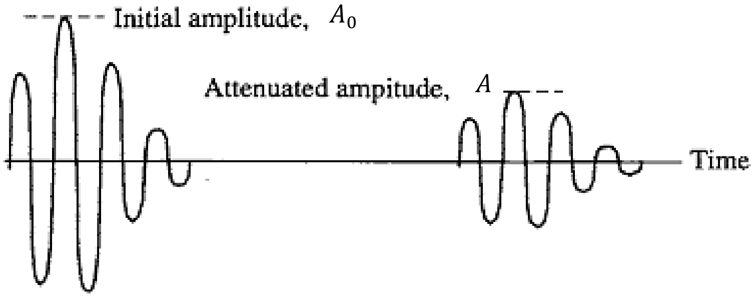


Figure 18: Diagram of an example of an attenuated wave (Lempriere, 2002)

(3) Acoustical Impedance Z

Acoustical impedance is the third ultrasonic propagation parameter. It occurs when the ultrasonic wave transmits through difference mediums that do not share the same properties. This parameter provides information of the amount of ultrasonic wave been reflected from its surface. The changes of the materials affect this parameter. The impedance is defined as,

$$Z = \frac{\Delta p}{\xi'} = \rho \frac{\omega}{k} = \rho v \,, \qquad \text{Equation 9}$$

where Δp is ratio of the acoustic excess pressure, ξ' is particle velocity, ρ is density of the mediums dan ω is angular frequency (Povey, 2007; Lempriere, 2002; McClements and Gunasekaran, 1997).

2.1.1.6 Adiabatic compressibility

Elasticity longitudinal wave propagates through solid, liquid, and gas. Velocity is calculated by square root of Bulk modulus B over the density of the tested medium (Equation 10). As the fruit and vegetables soften, their cellular cells consist of a mixture of the three compositions (solid, liquid, and gas). Therefore, the velocity of the compression wave in the mixtures depends on the adiabatic compressibility and density of the material. The adiabatic compressibility is obtained by inversion of B,

$$c = \sqrt{\frac{B}{\rho}} = \sqrt{\frac{1}{a\rho}} \qquad \text{Equation 10}$$

where B is the Bulk modulus, a is the adiabatic compressibility and ρ is the density. This is the **Wood equation** describing the compression wave propagated through a pure material (a single phase).

In reality, materials have more than one phase. Therefore, the **Urick equation** is a modified the Wood equation for two phase system (dispersed phase) by replacing the a and ρ with the following expressions,

$$a = a_0 = \emptyset a_2 + (1-\emptyset)a_1 \qquad \rho = \rho_0 = \emptyset\rho_2 + (1-\emptyset)\,\rho_1 \qquad \text{Equation 11}$$

where $\emptyset$ is the dispersed phase volume fraction and a_0 and ρ_0 are the volume average values and their subscripts a_1, a_2 and ρ_1, ρ_2 refer to the constituent phases (Povey 1997).

2.1.2 Experimental setup of a PUNDIT Plus System measurements

The experimental setup of the instruments for an ultrasonic velocity testing included a PUNDIT (Portable Ultrasonic Non-destructive Digital Indicating Tester) Plus device and its two transducers with 0.025 m radius (transmitting and receiving Transducers), a LeCroy Wave Surfer 44x oscilloscope, and a computer (Scope Explorer software - optional) (Figure 19).

The PUNDIT device was the source of ultrasonic pulse and it displayed a digital numerical reading of the time of flight on its screen. The device is an ultrasound pulse generator (CNS Farnell Electronics Ltd, 61-63 Holmes Road, London, NW5 3AL), and transmitting and receiving transducers. The first transducer is driven by a high voltage, 1kV pulse, creating an oscillating pressure wave, with around 39 kHz as a centre frequency. Pulse repetition rate is 10 Hz. The transmitting transducer works like a piston producing a chain of longitudinal (also called compression waves) through the medium. The 1kV pulse impacts on the piezoelectric ceramic transducers. Each pulse from the pulsed source excites the resonant frequency of the transducer. The pulse will change its rest state (restored) from compression to rarefaction state where the molecules in the compression area produce higher pressure than they in the rarefaction area. The repetitive cycle of restoring, expand and compress generates a series of pulses, where a particle position (amplitude), particle displacement and spatial pressure variation plays an important role in influencing the behaviour of the ultrasonic waves. The electrical energy from the transmitting transducer was converted to the mechanical energy when the pulse propagated through the tested medium. The wave behaviour is influenced by the medium. Next, the mechanical energy was reverted to electrical energy via the signals received by the receiving transducer (Povey, 1997). Therefore, it was hypothesized that the change of textural properties of the apple during storage changed the characteristics of the ultrasonic propagation wave that passed through the fruit without damaging the internal cell structures. Then, the ultrasonic propagation parameters measured in the experiment can be correlated to the fruit ripening quality.

Subsequently, the electrical connections from the device to the pair of ultrasonic transducers and to the oscilloscope are displayed in Figure 19.1 using a drawing of the back panel of the device showing its back panel for the illustrations of the wire connections. A cable from the 'TX' (transmitter output) was connected to the transmitting transducers, and a cable from the receiving transducers was fixed to the 'RX' (Receiver) respectively. After that, the device was plugged to the oscilloscope (Figure 19.3). A cable from the X of the PUNDIT device was fixed to the Channel 1 (C1 - axis X deflection speed or time-based) of the oscilloscope and a cable from the TRIG (triggering) was attached to the Channel 2 (C2 - Trigger circuitry) respectively.

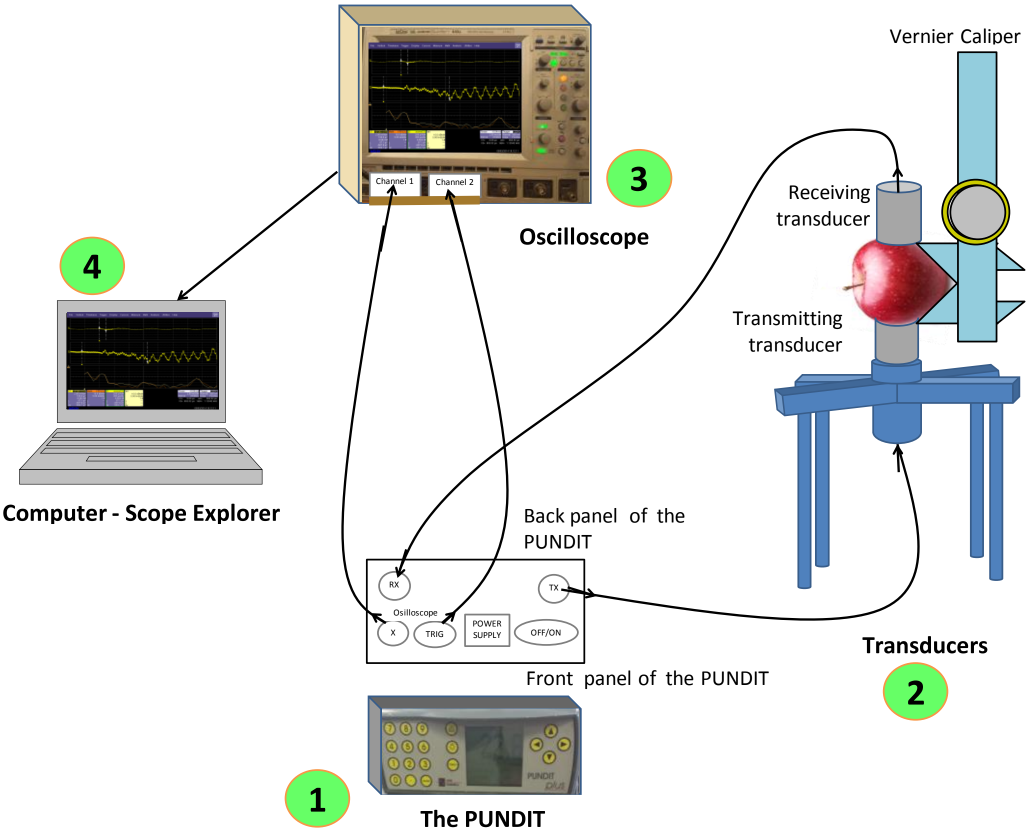


Figure 19: Schematic diagram of the experimental setup of the apparatus and electronic equipment for a PUNDIT Plus System of an apple

The whole apple was measured by placing it in between the pair of the transducers (Figure 19.2). The distance of an acoustical path of the fruit between the transmitting and receiving transducers was measured with a Vernier calliper (±0.1 mm). Based on the principles of the measurement of the velocity of sound, the following explains the steps in extracting the information on the signal and data acquisitions and data processing of the first wave arrival time of the propagated wave through the apples in the experiments (Figure 19). The arrival time was chosen based on visual inspection of the oscilloscope trace (Figure 19A). Very frequently the time of flight determined in this manner differed markedly from the time display on the PUNDIT devices. This is due to the complex relation between frequency and velocity of sound (called dispersion) which is evident in the difference between the low frequency component in the oscilloscope trace and the higher frequency component (Figure 19B) which gives a better resolved time of flight. This is due to the complex structure of the fruit which encourages the propagation of different modes of sound (e.g. through the gas bodies and through the rigid solid cells) at different speeds. The transducer is nominally 39 kHz generated a wide range of frequencies (Figure 20D) ranging between 1 kHz and 39 kHz. The choice of transducer frequency was based on a compromise between the need to the low frequency in order to get sufficient signal through the acoustically highly attenuating apple (attenuation increases rapidly with frequency) and the fact that time resolution improves as frequency increases, increasing the accuracy of the sound velocity measurement. The availability of commercially produced transducers which operated with the PUNDIT equipment also limited device.

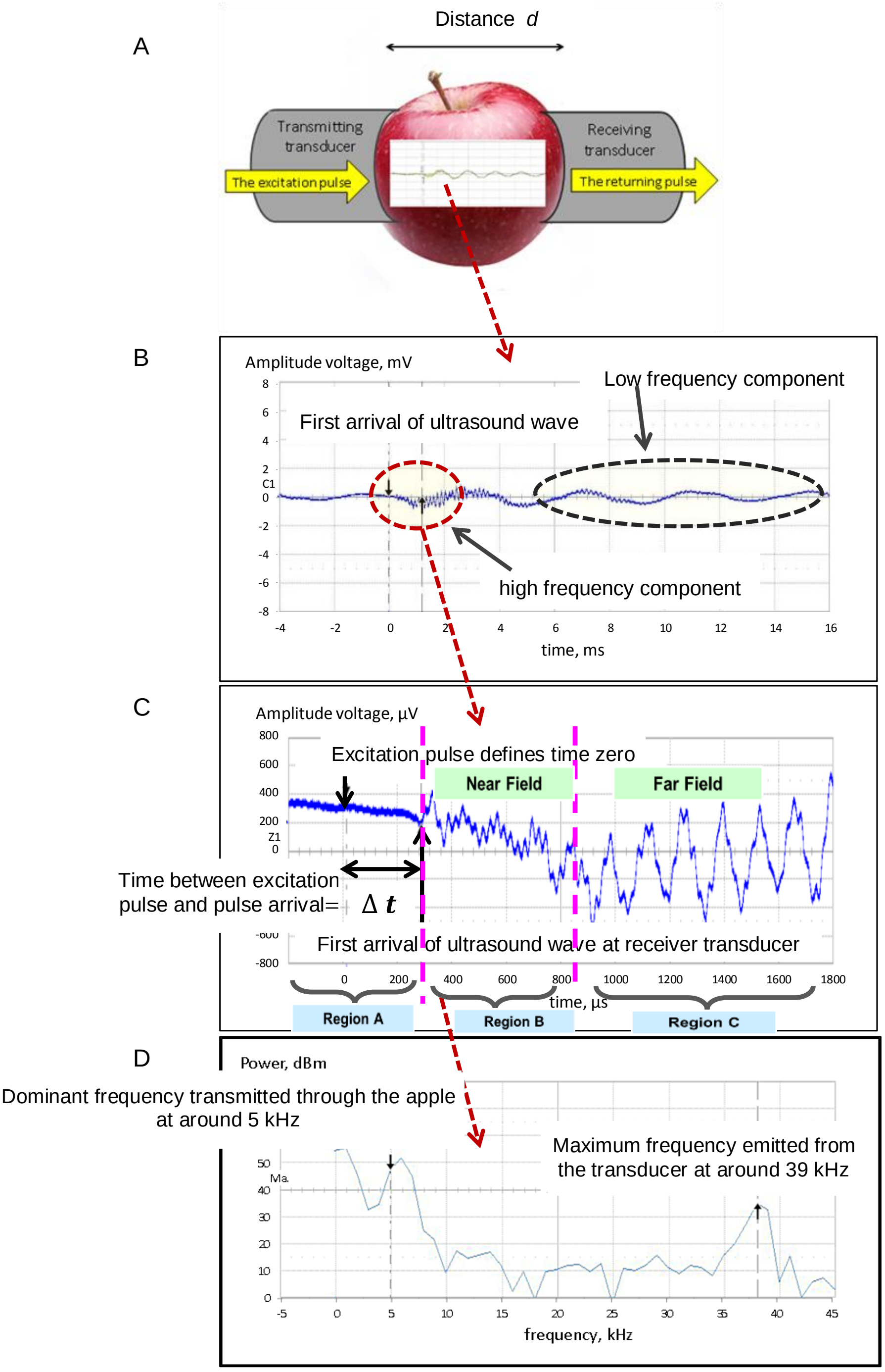


Figure 20 : A. Diagram of the pulse-echo acoustic time of flight measurement of an apple, B. Screen shot of a waveform showing the detected oscilloscopic trace of the ultrasound pulse propagated through an apple, dark blue trace in Channel 1 (C1). The left hand cursor positioned at the start of ultrasonic waveform; C. The longitudinal acoustic waveform in the highlighted region in B was expanded, bright blue trace in in Channel 2 (Z1), describing the three frequency components; Region A, Region B and Region C at the selected window; and D. Describing the frequency component, the corresponding Fast Fourier Transform, light blue in Ma.

2.1.2.1 Digital oscilloscope

A LeCroy Wave Surfer 44 x oscilloscope displayed signal amplitudes through waveforms (Figure 19.3). In the time domain from those waveforms, the time of the first ultrasonic arrival wave was determined and recorded. Spectra in the frequency domain were also displayed on the screen.

(1) Time domain analysis

The time of the ultrasonic wave travelled through the acoustical distance was obtained based on the oscilloscope trace. The oscilloscope provided this value by amplifying, measuring and interpreting the time of flight.

The procedure of time-based setup for the acquisition of a trace from the oscilloscope is described in the following steps (Figure 20). The waveform acquired after an apple was placed in between the two transducers of the ultrasonic testing system is displayed on Channel 1 (C1) (Figure 20A). The signals are displayed (Figure 20B) as Cartesian coordinates with the vertical axis as voltage (Y-axis) and the horizontal axis as a function of time (X-axis). The ultrasonic pulse (a waveform) was examined by the time-based generator as scan or deflection speed in X-axis. The time at the beginning coincided with the PUNDIT device trigger signifying the start of the pulse. Later, the receiving transducer captured the received ultrasonic pulsed waveforms, and the captures are displayed as a waveform on the oscilloscope. The graticule was set to a scale of 2 ms per division for the flight of time and 2 mV per division for the amplitude voltage in C1. The frequency of the main pulse was approximately 39 kHz. The time base was selected by choosing the ‘Cursors’ on the menu bar and choosing the ‘Horizontal time’ the drop-down menu. The dialogue appeared in the horizontal time base.

However, at this stage, the pulses were too narrow to see and it was difficult to select the time where the ultrasonic wave started. For this reason, the waveform was further intensified (Figure 20C). The waveform C1 was zoomed by selecting the highlighted region of C1 which is expanded and represented as trace Z1 (Zoomed channel). The left hand cursor was at the time zero, which coincides with the start of the excitation pulse and the zoom is presented on the scope screen in the bottom right hand corner. As a result, a zoomed channel (Z1) was activated for Channel 1 (time base) scale per division (scale/div). Optimum settings were determined for the voltage of the vertical scale (Y-axis) in μV per division and the time base of the horizontal scale (X-axis) in μs per division.

Therefore, the time of flight was determined visually based on the detection of an appearance of the pulse trace on the distinct change of the appearance (shape) of the waveform, the indicator of the first arrival of ultrasound. The step was done by adjusting the right hand cursor (Hickman, 1995; Maplin Electronics Ltd., n.d.).

(2) Frequency domain analysis: Frequency spectrum

The second experimental value required in this experiment was the spectral amplitude in the frequency domain before entering the pulse gate, within the pulse and leaving the pulse trace. The spectra were analysed using Fast Fourier Transform (FFT) signals. The frequency spectrum is represented as Cartesian coordinates with the spectral amplitude variations on the Y-axis in –dB/mm per division against frequency on the x-axis in kHz per division. Optimum settings were determined for the Y-axis and the X-axis. The period of the dominant frequency emitted from the transducer was approximately 39 kHz. The cursor was changed from the FFT signal to C1 (time base) in the next sequence. Note that the transducer also produced an audible 'click' when pulsed and this acoustic output showed up in the acoustic region at 5.2 kHz as an arrival with roughly 10x the power (20 dBm) of the ultrasound.

2.1.2.2 Scope Explorer (Optional)

A LeCroy Scope Explorer was able to run on a separate PC and was used to save waveforms in a picture format (bmp) and it can remotely control the command of the oscilloscope (Figure 19.4).

2.1.2.3 Percentage of relative humidity and temperature meter

Percentage of Relative Humidity (% RH) and temperature in the laboratory were monitored using humidity and temperature meter (Precision GOLD, model N18FR). The humidity accuracy is ± 3.5% RH (at 25°C, 5% to 95% RH) and the air temperature accuracy is ± 2.5°C (Maplin Electronics Ltd., n.d.).

2.1.2.4 Calculation of ultrasonic velocity *c*

In this experiment, the velocity was calculated by the distance of the ultrasonic wave propagated over the time of flight through the tested medium (Equation 12). A value of time (*t*) taken by the ultrasonic wave was determined by the first arrival of the ultrasonic waveform in the oscilloscope and/ or the PUNDIT screen (whichever applicable) in units of microseconds ($\mu\mathrm{s}$),

$$c = \frac{d}{t}$$ Equation 12

where c is an ultrasonic wave velocity expressed in meter per second ($\mathrm{m/s}$) as a function of d that is a distance travelled in millimetre (mm) over t that is a time of flight in milliseconds (ms) (Povey, 1997; McClements, 1995).

2.1.3 Method development of the PUNDIT Plus System

Characterisation and optimisation of the PUNDIT Plus System of measurements were conducted to ensure the performance of the test was fit to use for ultrasonic measurements. The instrument procedures were similar to the section 2.1.1. The details of the aspect of the characterisation and optimisation were described as the followings:

2.1.3.1 Sound field measurement of a transducer

This study was to determine a sound field of acoustic beam intensity of the transducer and to confirm that the field was within the sample. This is important because the ultrasonic measurement is associated with the acoustic intensity of a transducer travelling through the studied material and the intensity is related to the sound field of the transducer. The sound field of a transducer consists of two regions: near field and far field (Figure 21). Near field *N* is the first segment in front of the transmitting transducer which it starts with a series of maximum and minimum amplitudes and ends at the last maximum amplitude. Far field *F* is the second segment that goes beyond the focal point. The focal length is approximately equivalent to the *N* (natural focus) for unfocussed transducers. The *N* is calculated as follows,

$$\mathrm{N} = \frac{D^2 f}{4c} = \frac{D^2}{4\lambda} = \frac{r^2}{\lambda},$$ Equation 13

where *D* is a diameter of the transducer (m), f is the frequency, *c* is a sound velocity in the investigated medium (m/s), λ is a wavelength and r is a radius.

The signal amplitude vigorously varies in time and distance in the *N* due to the interference effects. Therefore, the determination on the time of flight is challenging. Nevertheless, the time of flight can be measured in the *N* if the signal amplitude can be distinguished (Olympus, 2006; Povey, 1997). In the apple and swede experiments, the signal amplitude of the first arrival of the ultrasonic wave was clearly detected on their waveforms. Consequently, the time of flight was determined in the *N*.

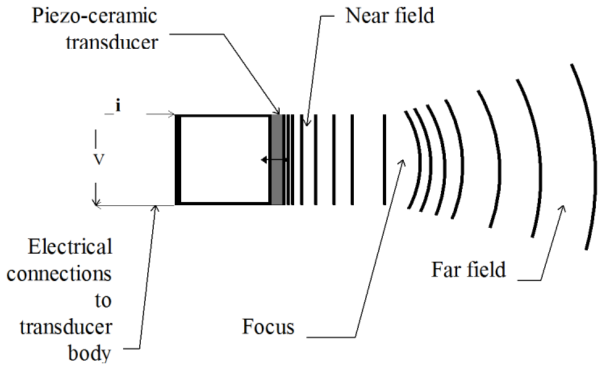


Figure 21: Illustration of the sound field of a transducer: Near and Far fields (Povey, 1997)

2.1.3.2 Waveform analysis in the time domain

This study was conducted to determine the expected waveform pattern and determine the location of the first arrival of the ultrasonic pulse from the waveform in the oscilloscope display. The determination was important because the first arrival of the ultrasonic pulse was the time of flight that was used to compute the ultrasonic velocity in the experiment.

A post-study was also carried out to investigate the reliability of the transit time display from the PUNDIT device.

2.1.3.3 Spectral analysis in the frequency domain using the FFT signal

(1) Regions of the waveform of the FFT signal

The purpose of the study on the waveform of the frequency FFT was to verify the optimum frequency of the working transducer passing through the fruit. Three different regions of the waveform of the frequency were investigated: (1) before entering the pulse gate, (2) within the ultrasonic pulse and (3) after the pulse trace. The regions were based on the chosen time domain signal in C1 and they were amplified in Z1. The oscilloscope parameters were set as the following: PRF usually 50 Hz, number of 100 averages and the zoom scale of 200 μV for the amplitude power and 200 μs for the ultrasonic transit time.

(2) Impact of the pulse repetition frequency (PRF)

The study of an impact of Pulse Repetition Frequency (PRF) on spectral analysis aimed to further investigate an optimum frequency spectrum at the working frequency of the transducer. Firstly, the optimum PRF is to ensure that the last region of the first pulse has completely died away before the second pulse comes so that the two pulses do not coincide with each other. Secondly, the optimisation can also reduce the background noise. Background noise refers to undesired echo and random noise that can overlap and interfere with the signal of interest. The sources of this interference can come from the electrical and environmental noises. As a result, selections on a proper region of ultrasonic signal are important to analyse an FFT signal of a waveform by the windowing. In addition, the decreasing PRF pushes up the received amplitude (Lempriere, 2002; Povey, 1997).

The studied was performed by reducing the PRF from 50 Hz (parameter setup previously) to 10 Hz, and 1 Hz. All the three spectra were compared and lastly the optimum setting PRF was decided.

(3) Natural frequency

This study aimed to characterise the behaviour of PUNDIT measurement using an apple as a sample to understand the interference of the device's audible buzz focussing on frequency components below 5 kHz. The receiving transducer detected environmental electromagnetic and acoustic disturbances, in addition to the interested transmitted ultrasound signal. The audible buzz sound was transmitted through an apple and it was detected easily in in the frequency spectrum. As a result, these signals may be misinterpreted during frequency spectra analysis. Therefore, the audible buzz may generate inaccurate data, because the low acoustic frequencies transmit much more readily through the sample.

(4) Insertion loss measurement of frequency signal attenuation

A study of an insertion loss measurement of a frequency signal aimed to understand the attenuation of the frequency when the ultrasonic pulse was transmitted inside the tested medium. It was hypothesized that the amplitude signal of the transmitted pulse decreased while propagating through the apple due to the changes of the fruit cell structures during ripening. The physical effects, which contribute to this insertion loss effect, were discussed in Section

1.8. Maturity and ripening in fruit and vegetables (loss of turgor, cell wall rigidity and cell-cell adhesion, and gas change during ripening).

Apart from the attenuation of the frequency amplitude signal of the medium, insertion loss may also be affected by reflection/ transmission at the two-transducer faces due to the path length from the piezo electric disk to the wear plate of the transducers prior to a contact with the apple (offset). The offset of the ultrasonic path distance was discussed in section 2.1.5.1.

The insertion loss measurement was obtained by a subtraction of two frequency spectra. Firstly, the amplitude of the highest frequency in air (without an apple) was recorded. The highest frequency was referred the frequency of the transmitting transducer at approximately 39 kHz. Secondly, the whole apple was inserted in between the transducers and the amplitude at 39 kHz was recorded. Finally, the frequency in air was subtracted by the frequency in the fruit and the subtracted value was the insertion loss from the frequency signal attenuation of the tested apple. The insertion loss of frequency was expressed as a signal voltage (in the unit of dBm). Insertion loss differs from signal attenuation in that no account is taken of signal path length and signal diffraction.

2.1.4 Verifying the measurement setup accuracy with the velocity of sound in air

The aim of the measurement of the velocity of sound in air was to verify accuracy of the PUNDIT Plus System. The verification was conducted by measuring the time of flight taken for the ultrasonic pulse propagated through the designated distance of ambient air between the two transducers (without an apple). This verification was carried out prior to the PUNDIT Plus System of measurements.

2.1.5 Experimental errors

This section introduces the experimental errors that can affect the accuracy and precision during the experiment. However, the errors can be estimated and accounted for. The sources of errors were identified as the followings.

2.1.5.1 Offset of the ultrasonic path distance

This experiment was to investigate the offset of the ultrasonic path distance of the PUNDIT device if exist to compensate the measurement error. The PUNDIT devices have been used for ultrasonic measurements for a wide range of materials due to its non-destructive technique. However, because no single transducer is manufactured identically, a transducer has its unique ultrasonic characteristics. A pulse offset of the device is one of measurement errors occurred because of the inherent characteristic of the transducers. It refers to electric and mechanical delay (if any) because of the path length from the piezo electric disk to the wear plate of the transducer prior to a contact with a tested medium (Figure 22).

The experiment was carried out by changing ultrasonic distance and recording the time of flight by using air path and slices of an apple.

1. Air path: 10 readings of the ultrasonic path lengths and 10 readings of the time of flight were obtained by moving the distance of the two transducers for the air offset investigation.
2. Slices of an apple: Five ultrasonic path lengths and times of flights were obtained by slicing an apple into five slices. The slices were labelled as S1, S2, S3, S4 and S5 and they were put back in the following manners for the measurements of the ultrasonic path distances and times:
    i. S1;
    ii. S1 and S2;
    iii. S1, S2 and S3;
    iv. S1, S2, S3, and S4; and
    v. S1, S2, S3, S4 and S5,

The graph of the ultrasonic path length against the time of flights was developed. A linear of regression was used to find The Y intercept. The y –intercept indicated the offset of the transducer for an ultrasonic measurement.

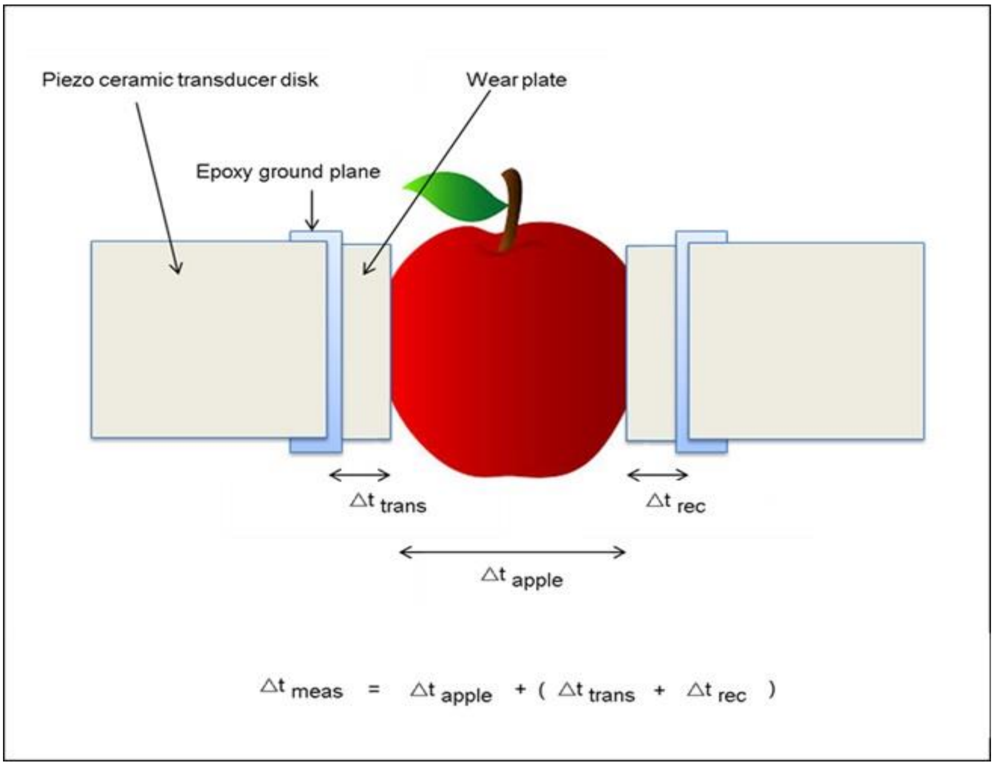


Figure 22: Diagram illustrating a pulse offset due to the path length of the electrical and mechanical delay influenced the time of flight during measurement (such as air or an apple) (Δt_{trans} is time difference of transmitting transducer, Δt_{rec} is time difference of receiving transducer and Δt_{meas} is time difference of a measurement.)

2.1.5.2 Bulk wave propagation

A study of bulk wave propagation was investigated to confirm that the ultrasonic wave travelled inside the measured apple throughout the experiment rather than around its surface. The investigation was conducted by computing the *t*-Test for the ultrasonic velocity of the apple slices in section 2.1.5.1. The *t*-Test was conducted to check any significant differences in velocity between the whole apple and the accumulated numbers of the apple slices. The hypothesis was that if the velocities between the whole apple and the apple slices were not significantly different, then, the ultrasonic wave had propagated inside the fruit.

2.1.5.3 Attenuation of an ultrasonic pulse signal

A study of the attenuation of an ultrasonic pulse signal passing through an apple was also investigated. The investigation aimed to confirm that the signal amplitude decreases exponentially against the ultrasonic path distance (Povey 1997). The investigation was a continuation of section 2.1.5.1 by recording the maximum amplitude signal height in –dBm at 39 KHz and the ultrasonic path distance based on the numbers of the fruit slices. Next, a graph of the maximum signal amplitude against the number of the apple slices was developed. It was hypothesized that the graph was exponentially curved. If this hypothesis was correct, the attenuation of the ultrasonic signal amplitude through the apple can be substantiated. However, this method does not account for diffraction errors.

2.1.5.4 Variability in shapes and the angles (directions) of measurements of apples

The followings are the source of variabilities that aimed to be tested if the variability in the shape and the angles of measurements can contribute errors in the PUNDIT Plus System of measurements. The experiment was conducted as in section 2.1 (‘*Pink Lady’*, ‘*Royal Gala’* and ‘*Envy’*) and 2.1.1.

2.1.5.5 Shape and angles of radial measurements

Surface appearances of the fruit axial and radial positions were not similar although they were opposite each other. The shape and the angles of radial measurements of the fruit influenced the distance of the ultrasonic wave path length travelled through the fruit. The measurements were based on the height (from a stem to a calyx for axial measurement) and the width (from side to side for radial measurement).

(1) Variability of time of flight of the first arrival of ultrasonic wave

Determination of the time of flight on the ultrasonic waveform the oscilloscope screen can be varied for each velocity measurement.

(2) Variability of axial and radial velocity measurements: Preliminary study on effect of anisotropy of apples

An effect on anisotropy of apples on ultrasonic velocity was preliminarily studied to test if the axial and radial measurements showed significant differences for apples at the end of their ripening stage prior to reaching consumers.

2.2 ‘Envy’ apples: Assessment of ripening and anisotropy in apples

The experiment was conducted from November 2014 to January 2015. Two boxes of 99 ‘Envy’ apples consisting of 49 M and 49 MM fruit were received from a fruit supplier in Kent, UK via an express courier service. The fruit were harvested in October 2014, based on the fruit maturity index for harvest guidelines. Both groups comprised apples of different sizes and shapes. The mature ‘Envy’ apples were the harvested fruit that meet a specification of maturity requirements of harvest parameter tests prior to their harvest time (ENZAFOODS, 2014). Meanwhile, the more mature ‘Envy’ apples were the fruit at more advanced maturity levels, based on their dry matter content. However, the rest of the harvest parameter tests were within the specifications.

The fruit were labelled as ‘M’ for the mature fruit and ‘MM’ for the more mature fruit, and they were given a number from 1 to 49 on their marked peels. The samples were also marked with A1 at the stem and A2 at the calyx for axial measurements and R1 at 90° and R2 at 180° for radial measurements. The axial direction was defined as the ultrasound propagation *across* the axis of the cell columns and intercellular air spaces of the fruit. In contrast, the radial direction was defined as the ultrasound propagation *along* the axis of the cell columns and intercellular air spaces of the fruit (Figure 23). These marks ensured that the sample was measured at the same point of the intended tests throughout the repeated measurements.

The fruit from 1 to 7 of the M and MM groups were held at ambient temperature (approximately 20°C) and served as the control group. Meanwhile, the fruit from 8 to 49 of both groups were stored in a chiller at 4°C.

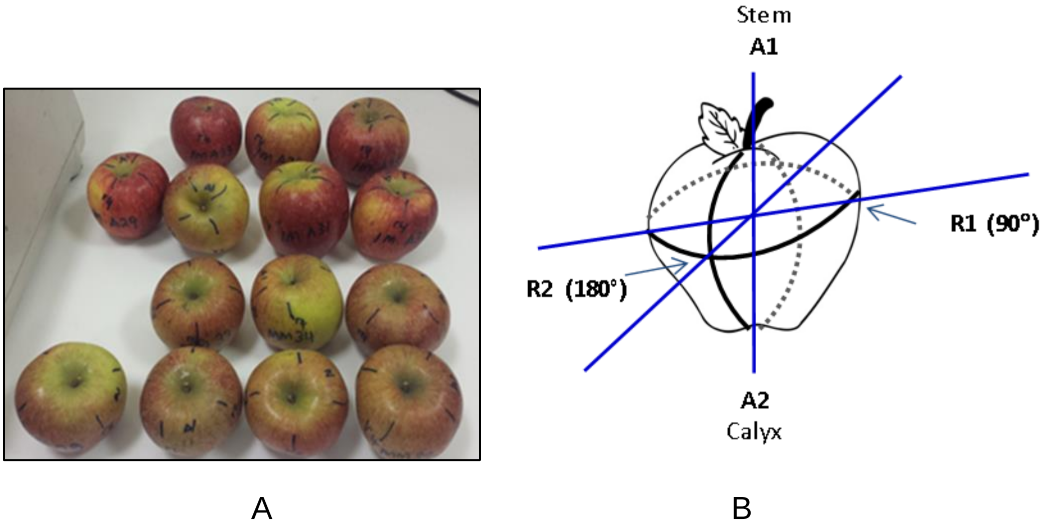


Figure 23: Envy Apples. B. Pictorial display of positions of axial (A1 and A2) and radial (R1 and R2) measurements for an ‘Envy’ apple experiment.

Figure 24 shows five tests conducted for the 98 ‘Envy’ apples for every 2 weeks along the 10-weeks-storage period:

1. ultrasonic velocity test for an intact apple by PUNDIT Plus transducer system,
2. a compression test and a penetration Test by a Texture Analyser (TA),
3. sugar content test by a refractometer and
4. another ultrasonic velocity test by Ultrasonic Velocity Meter (UVM) for apple puree.

The first two measurements were non-destructive testing (NDT) whereas the remaining four measurements were destructive testing (DT). The measurement procedures for those tests are detailed in the following section.

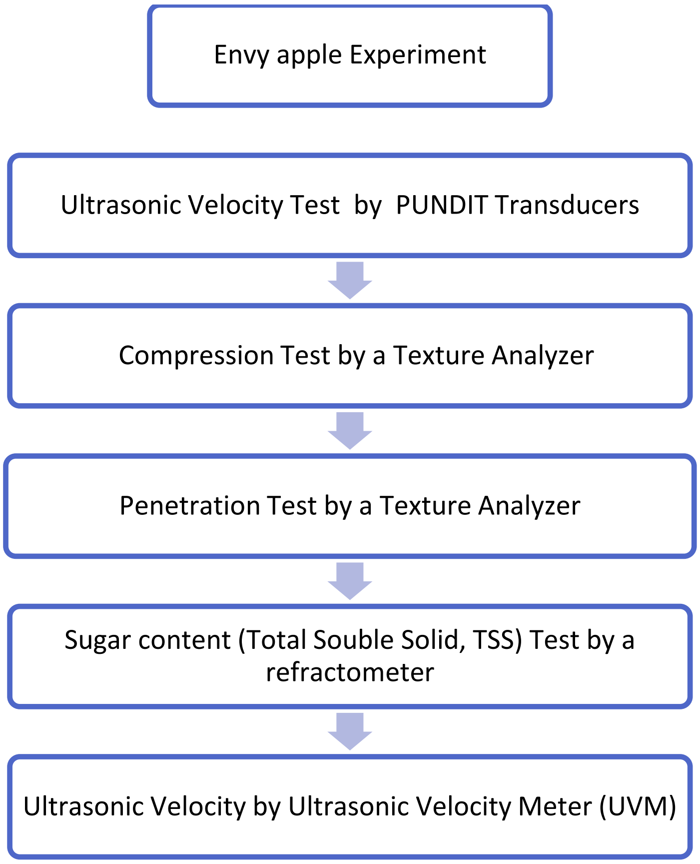


Figure 24: Flow chart of the ‘Envy’ apple experiment

2.2.1 Ultrasonic velocity measurement of an intact apple by a PUNDIT Plus System

The apple velocity was measured axially and radially by using the PUNDIT Plus System to study a correlation of the changes of the ultrasonic propagation parameter to the fruit ripeness during storage. The measurement procedures were similar to the section 2.1.1 and were repeated for samples M1 – 49 and MM1 - 49. Seven apples were measured in every two weeks and an average was calculated.

2.2.2 Insertion loss measurement of amplitude frequency signal

The value difference of the amplitude between the frequency spectrum in air (without an apple) and in an apple at the 39 kHz was calculated to indicate an insertion loss due to attenuation of the apple measurement. The procedures were similar to the section 2.1.3.3 (4) (Insertion loss measurement of frequency signal).

2.2.3 Texture Analysis by a compression test

Firmness of the fruit was measured non-destructively (both axially and radially) to investigate the fruit ripening changes over 8 weeks storage of 4°C. A measurement of force/displacement indicated the elasticity of the samples. The elasticity was explained by the modulus of deformability (Young’s modulus of elasticity) of the tested material (Bourne, 2002). The measurement was obtained using a Stable Microsystems Texture Analyser TA XT Plus (Stable Micro Systems, Surrey, U.K.) with texture analysis software (Texture Exponent 32) (Figure 25). A flat 75-mm diameter aluminium plate (SMS P/75) and a 5-kg load cell were used. The set-up speeds were at 0.04 mm/s (pre-test), 0.04 mm/s (test), and 0.40 mm/s (post-test). Distance of the compression was 0.200 mm and trigger force was 0.1N (adapted from Kim et al., 2009; Varela et al., 2007; Kim et al., 2006; Al-Haq et al., 2004; Alvarez et al., 2002). The modified setting parameters were to prevent irreversible deformation of the fruit, so that the next test (a puncture test) can be carried out properly. Therefore, no flesh rupture was performed onto the sample. The loaded force was sufficient to recognize the change of firmness during storage. The linear region of the curve was confirmed by investigating a set of the force and displacement measurements of the compression to demonstrate the elasticity of the fruit.

The fruit was measured at the marked points (Figure 23B). Firstly, the fruit with its specific marked point of measurement (A1 and A2, R1 and R2) was placed on the centre between the base plate of the TA and the flat compression plate (Figure 26). Secondly, the flat compression plate was brought in contact with the sample operated as the TA setting parameters. Thirdly, the measurement was repeated for all respective samples that were scheduled for that particular period. The test was performed in the laboratory with the temperature between 16°C and 20°C and the percentage relative humidity between 40% and 50%.

The graph of force versus time was automatically generated and appeared on the Texture Exponent (Figure 27). The firmness was expressed by work of compression being applied onto apple represented by the area under the load-deformation curve with a unit of *Ns*. The calculated areas under the curve for the scheduled sampled were recorded. After that, the two directions of measurements were averaged and standard deviation and standard error were determined. Note: The compression test was introduced in Week 2 of the experiment.

Figure 25: Stable Microsystems Texture Analyser TA XT Plus (Stable Micro Systems, Surrey, U.K.) with texture analysis software (Texture Exponent 32)

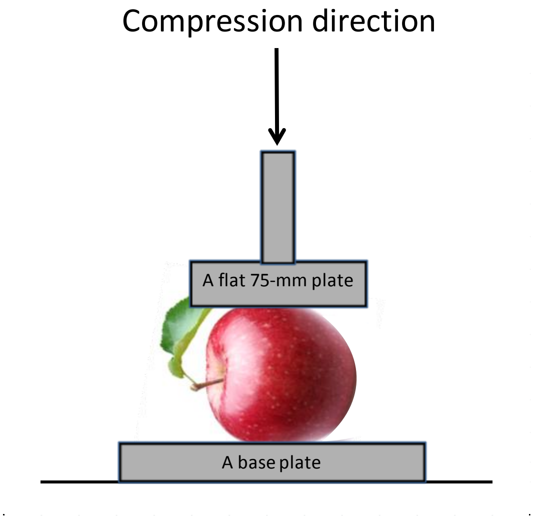


Figure 26: An example of a schematic diagram of the experimental setup of a compression test using a Texture Analyser

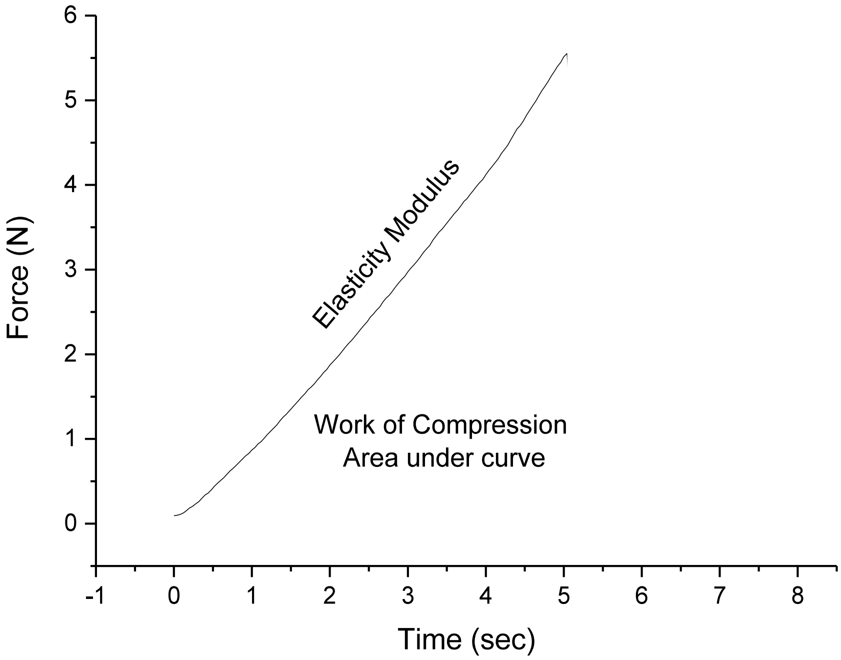


Figure 27: A typical force-deformation curve for an apple under compression (75-mm flat plate, test speed: 0.04mm/s, Distance of the compression: 0.200 mm, trigger force: 0.1N)

2.2.4 Texture Analysis by a puncture test

Firmness of the fruit was assessed destructively by a puncture test, axially and radially during 8 weeks storage at 4°C based on the maximum puncture force by using a Stable Microsystems Texture Analyser TA XT Plus, the TA instrument in section 2.2.3. A 5 mm diameter stainless steel punch cylindrical probe (SMS TA-55) was used. The set-up test speeds were at 1.5 mm/s (pre-test), 1.5 mm/s (test), and 10.0 mm/s (post-test). The puncture distance was 5.0 mm and the trigger force was 0.245N.

The fruit was measured at the marked points (Figure 23B). Firstly, the skin of the apple was sliced by using an apple peeler at its specific location (A1 and A2, R1 and R2). The position of the peeler blade was properly aligned with the location of the measurement to ensure the slicing uniformity to reveal the flesh of the fruit (OECD, 2005). Secondly, the fruit was positioned between the lower base of the TA and the cylindrical probe (Figure 28). The probe punctured the fruit until the puncture reached the set up distance. Thirdly, the measurement was repeated for all samples that were scheduled for that particular week. The test was performed in the laboratory with the temperature between 16°C and 20°C and the percentage relative humidity between 40% and 50%.

A maximum puncture force was expressed in Newton (*N*). Figure 29 is typical force-deformation curve for an apple puncture test displayed on the Texture Exponent. The forces for the designated samples were recorded. After that, the two maximum forces of the axial and radial measurements were averaged and standard deviation and standard error were determined. Note: The compression test was introduced in Week 2 of the experiment.

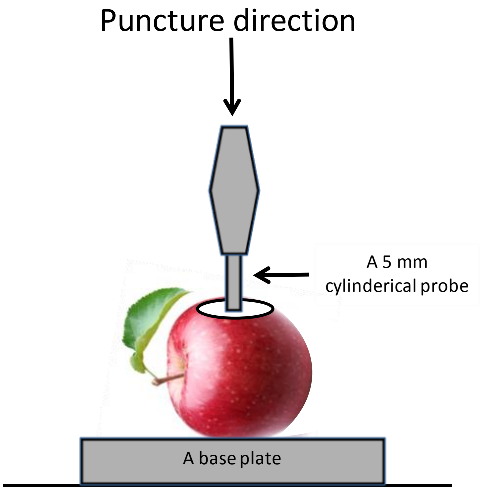


Figure 28: Schematic diagram of the experimental setup of a puncture test by using a Texture Analyser

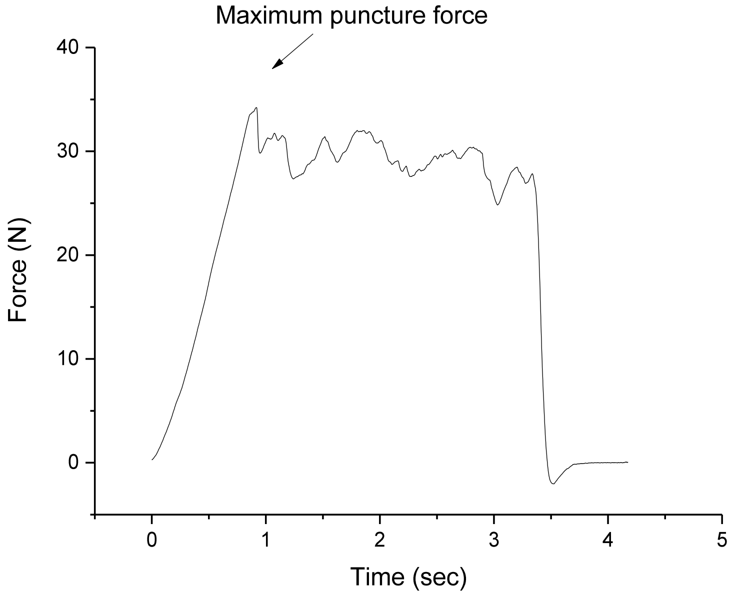


Figure 29: A typical force-deformation curve for flesh apple puncture test (5 mm diameter punch cylindrical probe, test speed: 1.50 mm/s, puncture distance: 5.000 mm, trigger force: 0.245 N)

2.2.5 Sugar content measurement by a hand refractometer

Sugar content was measured to investigate the sweetness changes of the fruit ripening quality during storage in order to describe the Total Soluble Solid (TSS) in apples. The changes are associated with a conversion from starch to sugars during ripening (Kader, 2002a; Taiz and Zeiger, 2002). The measurements were sampled from an axial and radial location of the fruit. It was measured to investigate the fruit ripening change during 8 weeks of storage at 4°C based on the percentage of Brix reading. Brix scale (degrees Brix, °Bx) is to measure the percent sugar and TSS. One degree Brix is equal to 1 gram of sucrose in 100 grams of solution (Guardabassi and Goldemberg, 2014; Ball, 2006). A refractometer measuring uses a principle of the Brix scale where reflection of light of the concentration of sugars produces an image. When lights from a light source transmitted between a prism and the sugar solution, some of the lights are absorbed into the solution while some other portions are reflected from the solution. The reflected lights produce an image. This image depends on the refractive index (a critical angle of total reflection). The boundary position of the image defines the concentration of the sugar solution and it is determined by the light detectors of the refractometer (Kahre, 1997).

A B+S handheld refractometer (0 to 30% Brix) was used (Bellingham and Stanley Ltd. Kent, England) with 0.2% graduations (Figure 30). Two main steps were performed in using the hand refractometer. Firstly, the instrument was verified by distilled water reading prior to the actual measurement. The distilled water was placed on the prism surface. The prism cover was closed and the refractometer was held towards the light to read the °Brix. The reading was obtained by looking through the eyepiece lens on the other side of the instrument and adjusting the knob at the end of the lens until a clear image appearance was obtained. The refractometer was fit to use when the reading of the distilled water indicated at zero. Secondly, the sampling and measurement were followed after the verification. The sample was cut at the designated positions from the location at which the puncture test had been carried out. The portion of the slices was squeezed into juice by using a garlic press and the juice was positioned on the prism plate. The reading was recorded. The sugar content readings of the two positions were averaged and standard deviation and standard error were determined (adapted from OECD, 2005).

The test was performed in the laboratory with the temperature between 16°C and 20°C and the percentage relative humidity between 40% and 50%. The test was repeated for the remaining direction of measurement for all designated samples of the particular week.

Figure 30: A B+S handheld refractometer (0 to 30 % Brix) with 0.2% graduations.

2.2.6 Ultrasonic velocity measurement in apple puree by using Ultrasonic Velocity Meter Test (UVM)

Velocity of the fruit puree was measured to investigate the different velocities between the two ultrasonic propagation mediums (intact apple and apple puree) during the fruit ripening due to the change of internal cell structures. The measurements were conducted by using a Cygnus UVM1 Ultrasonic Velocity Meter (UVM). Gases in the fruit puree had been removed prior to the measurement see Section 2.2.6.1 below. Moreover, an unripe apple has lesser intercellular air gases compared to the ripening apple.

The ripening apple flooded with a mixture of gas-water gasses (Zdunek, 2013; Zdunek et al., 2010; Khan and Vincent, 1990; Reeve, 1953; Bain and Robertson, 1951). The mixture can be related to the conversion of starch to sugar towards senescence (Kader, 2002a; Taiz and Zeiger, 2002). This change of gas volume may influence the velocity characteristics in the fruit. The degasified apple puree was designated as a control to represent unripe apple conditions that all gases was removed from the apple.

2.2.6.1 Apple puree preparation

Apple puree was prepared by blending the remaining portion of the apple of the sugar content measurement using a grinder, Spice Grinder ZX789/ZX809X (James Martin, Kent, England). Next, the puree was transferred into a container. Bubbles were observed indicating gas existence in the sample at this stage (Figure 31A). Therefore, the sample was degasified using a vacuum device for about one hour for gas removal to avoid the UVM reading interference (Figure 31C). Later, the samples were heated at about 70°C for about another 2 hours for further degasification by using a water bath (Grant, SUB Aqua 18 Plus). Finally, the samples were cooled at the ambient temperature (Figure 31B).

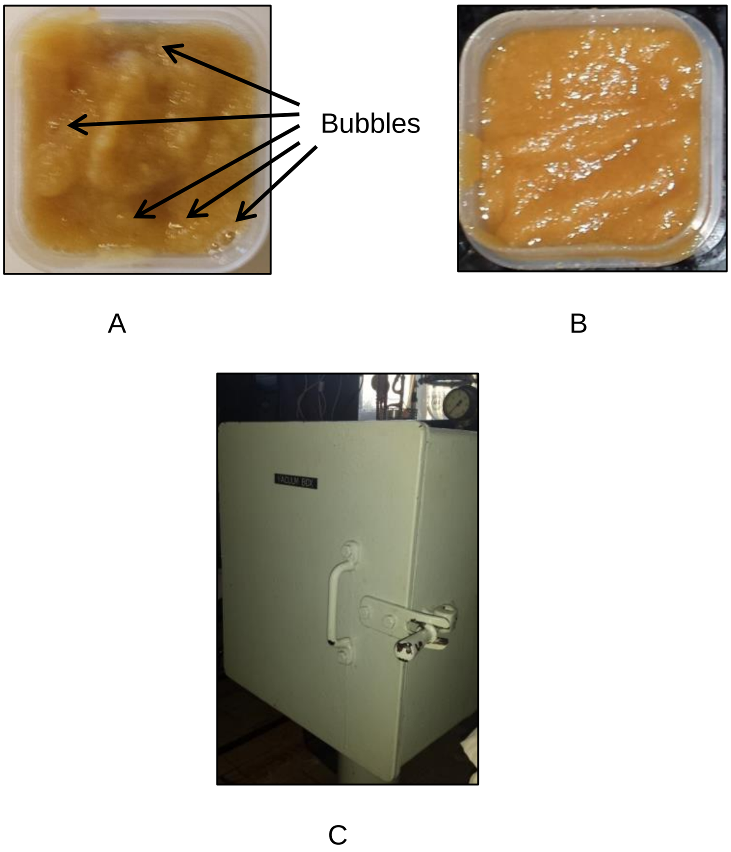


Figure 31: A. Apple puree with air bubbles; B. Degasified apple puree; and C. Degasification using a vacuum device.

2.2.6.2 UVM instrumental set up and apple puree measurement

The experimental setup consisted of a measurement cell attached to a Lead Zirconate Titanate (PZT) acoustic transducer and a Platinum Resistance Thermometer (PRT), a Cygnus UVM ultrasound velocity meter (Cygnus Instruments, Dorchester, UK), a computer (UVM1 Analyser Software), a water bath (Grant W28), magnetic stirrer plate and a refrigerated cooling unit, Grant Model CZ2 (Grant Instruments (Cambridge) Ltd. Barrington Cambridge, England) (Figure 32). A 75.5 mm-height-cylindrical measurement cell with a 40 mm-internal diameter was attached by a PZT transducer with a 10mm diameter. The transducer generates an ultrasonic pulse at a frequency of approximately 2.25 MHz that propagates towards the other side of the cell wall and bounces back to the same transducer. Apart from the transducers, the measurement cell also is connected by a PRT to measure the temperature of the sample with an accuracy of ± 0.2°C (Povey, 1998a).

In the following stage, the fruit puree was poured into the cell. Next, the measurement cell was closed with a lid and immersed on the magnetic stirrer plate in the water bath at 20°C together with the refrigerated cooler to maintain the temperature (Figure 32.1). PVC rounded floating balls covered surface area of the water to maximize the water temperature stability (Figure 33). The sample was continuously stirred by the magnetic stirrer. Meanwhile, the measurement cell was connected to the UVM electronic device to interpret the time of flight, temperature and to calculated sound velocity as the communication system (Figure 32.2). The device also displays a digital reading of the time of flight and temperature on the screen.

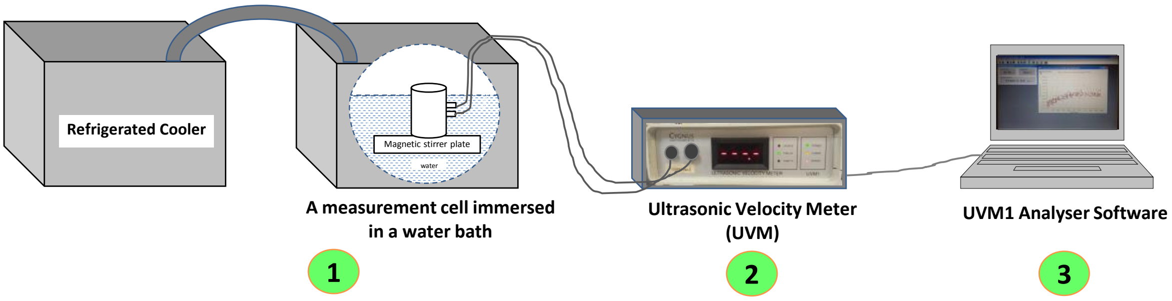


Figure 32: Schematic diagram of the experimental setup of the apparatus and electronic equipment of an Ultrasonic Velocity Meter (UVM) for apple puree

Figure 33: Picture from the top of the apple puree in a Cygnus UVM1 Ultrasonic Velocity Meter (UVM) cell measurement immersed in the water bath

2.2.6.3 Data acquisition

Reading for the time of flight, temperature and calculated sound velocity was generated and displayed on the computer via the Cygnus UVM1 Analyser Software in a UVM1 Form. A graph of ultrasonic velocity (m/s) and time (µs) are also produced (Cartwright, 1998; Povey, 1998a). The data were stored in Microsoft Excel form. The UVM system was stopped when the velocity curve on the graph was consistent. The measurements were repeated for the next designated sample.

2.2.7 Calculation of estimations of the uncertainty of measurement

The measured variables involved in the 'Envy' apple experiment were collected and the expanded uncertainty U_e of the measurement of the tests were calculated and reported with 95% confidence limit. The values were based on the computation of an individual uncertainty value from the sources of uncertainty u of each test and the combined uncertainty U_c. The u, U_c, and U_e were calculated using the following formulas in the respectively discussion.

2.2.7.1 Specification the measurand

The relationship between the measurands and the variables of the tests were specified (Table 16). For the PUNDIT Plus System, the measured parameters were time of flight, ultrasonic path and anisotropy of an apple, (axial and radial measurements) and the measured property for this test was ultrasonic velocity. For the compression test, the measured parameters were the area under curve and the anisotropy of the fruit and the firmness (skin and flesh elasticity) was its measured property. For the puncture test, the measured parameters were the maximum puncture force and the anisotropy of the fruit and the measured property was its flesh firmness. For the sugar content test, the measured parameters were °Brix and anisotropy of the fruit and the sugar level was its measured property. For the UVM test, the measured parameters were time of flight and ultrasonic path and the measured property for this test was ultrasonic velocity.

2.2.7.2 Identification and quantification of the sources of uncertainty, *u*

Based on the measurands, the following uncertainty contributions, and u, were identified and quantified following the respective equations (Table 16) (Barwick and Ellison, 2000; AOAC, 2007; NDT, 2010; Tesileanu and Niculita, 2013):

(1) Precision uncertainty *u(P)*

The estimation of uncertainty of the method precision *u(P*) was analysed by using the repeatability measurements of the measured properties of each test for Weeks 0, 2, 4, 6, and 8. The value of the uncertainty of the precision study was the pooled relative standard deviation RSD_{pool} by using Equation 14 and Equation 15 respectively,

$$RSD \ = \frac{s}{\bar{x}} \qquad \textbf{Equation 14}$$

where *s* is the standard deviation of the measurements for the designated week, and $\bar{x}$ is the mean measurement.

$$RSD_{pool} \ = \sqrt{\left[\frac{{RSD_1}^2 \text{ x } (n_1 - 1) + {RSD_2}^2 \text{ x } (n_2 - 1) + \cdots}{(n_1 - 1) + (n_2 - 1) + \cdots}\right]} \qquad \textbf{Equation 15}$$

where RSD is the relative standard deviation of the measurements for the designated week, and $n_1, n_2, \ldots$ is the number of replicates of the analysis involved in the experiments.

(2) Accuracy uncertainty *u(A)*

The estimation of uncertainty of the method accuracy *u(A)* was conducted by using the measurements of:

1. The speed of sound in air based on the published data (Ford, 1970) for the ultrasonic velocity in the PUNDIT Plus System,
2. The certified reference materials (CRM) of 5kg reference weight for firmness in the compression and puncture tests by using the Texture Analyser,
3. Verifications of refractometer reading by using distilled water and 5% pure sucrose for the sugar content test, and
4. Verified with an external digital thermometer Accuracy practices were followed by ensuring that the UVM cell was immersed completely in a water bath and the sample was stirred as well as only Platinum Resistance Thermometry was used throughout the experiment for the ultrasonic velocity in the UVM test.

The value of the uncertainty of the accuracy study was the mean recovery $\bar{x}_{rec}$ and the uncertainty in the accuracy by using Equation 16 and Equation 17 respectively,

$$\bar{x}_{rec} = \frac{\bar{x}_{meas}}{x_{ref}}$$ **Equation 16**

where $\bar{x}_{meas}$ is the mean of replicate measurement, and x_{ref} is the expected/ given value of the reference,

$$u\,(\mathrm{A}) \;=\; \bar{x}_{rec} \;\mathrm{x}\; \sqrt{\left[\frac{s^2_{meas}}{n \;\mathrm{x}\; \bar{x}^2_{meas}}\right] + \left[\frac{u\,(x_{ref})}{x_{ref}}\right]^2}$$ **Equation 17**

where s^2_{meas} is the standard deviation of the mean of replicate measurement (Equation 16), n is the number of replicates, $u\,(x_{ref})$ is the given uncertainty or 95% confidence interval and x_{ref} is the expected/ given value of the reference.

(3) Instruments' specifications uncertainty *u(I)*

The estimation of uncertainty of the instruments' specifications *u(I)* was calculated based on the information given by the manufacturers or the manuals by using Equation 15,

$$u(I) \;=\; \frac{Given\;uncertainty}{1.96\;(95\%\;Confidence)}$$ **Equation 18**

where $Given\;uncertainty$ is the value of uncertainty stated by the manufacturer or the manual and $1.96\;(95\%\;confidence)$ is the value of the normal distribution for the level of given confidence interval.

2.2.7.3 Calculation of the combined uncertainty, *U$_c$*

The measurement of the tests were influenced by the parameter precision, accuracy and instruments' specifications which each has uncertainty *u(P), u(A)* and *u(I)* respectively. The combined uncertainty, U_c was calculated by using Equation 19,

$$Uc = \sqrt{u(P)^2 + u(A)^2 + u(I)^2}$$ **Equation 19**

where $u(P)$ is the uncertainty of the precision study, $u(A)$ is the uncertainty of the accuracy study, and $u(I)$ is the uncertainty of the instruments' specification study

2.2.7.4 Calculation and report of the expanded uncertainty, U_e

The expanded uncertainty, $U_{e,}$ was calculated by multiplying the combined uncertainty with coverage of 2 (at 95% confidence level) given in Equation 20. The U_e for each test was stated as "Measurement of (*measured property of the test*) was *Result* (unit) ± U_e (unit), where the reported uncertainty is based on a standard uncertainty multiplied by a coverage factor k = 2, providing a level of confidence of approximately 95%",

$$Ue = U_c \text{ x } 2, \quad 95\% \text{ confidence limit}$$ **Equation 20**

where U_c is the combined uncertainty and 2 is a value at 95% confidence limit.

Table 15: Summary of the steps in estimation of the measurement of uncertainty

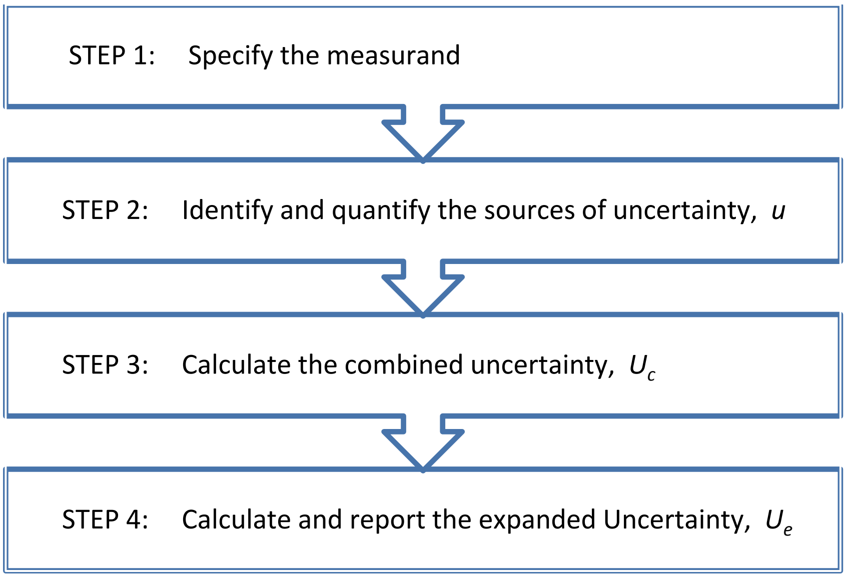
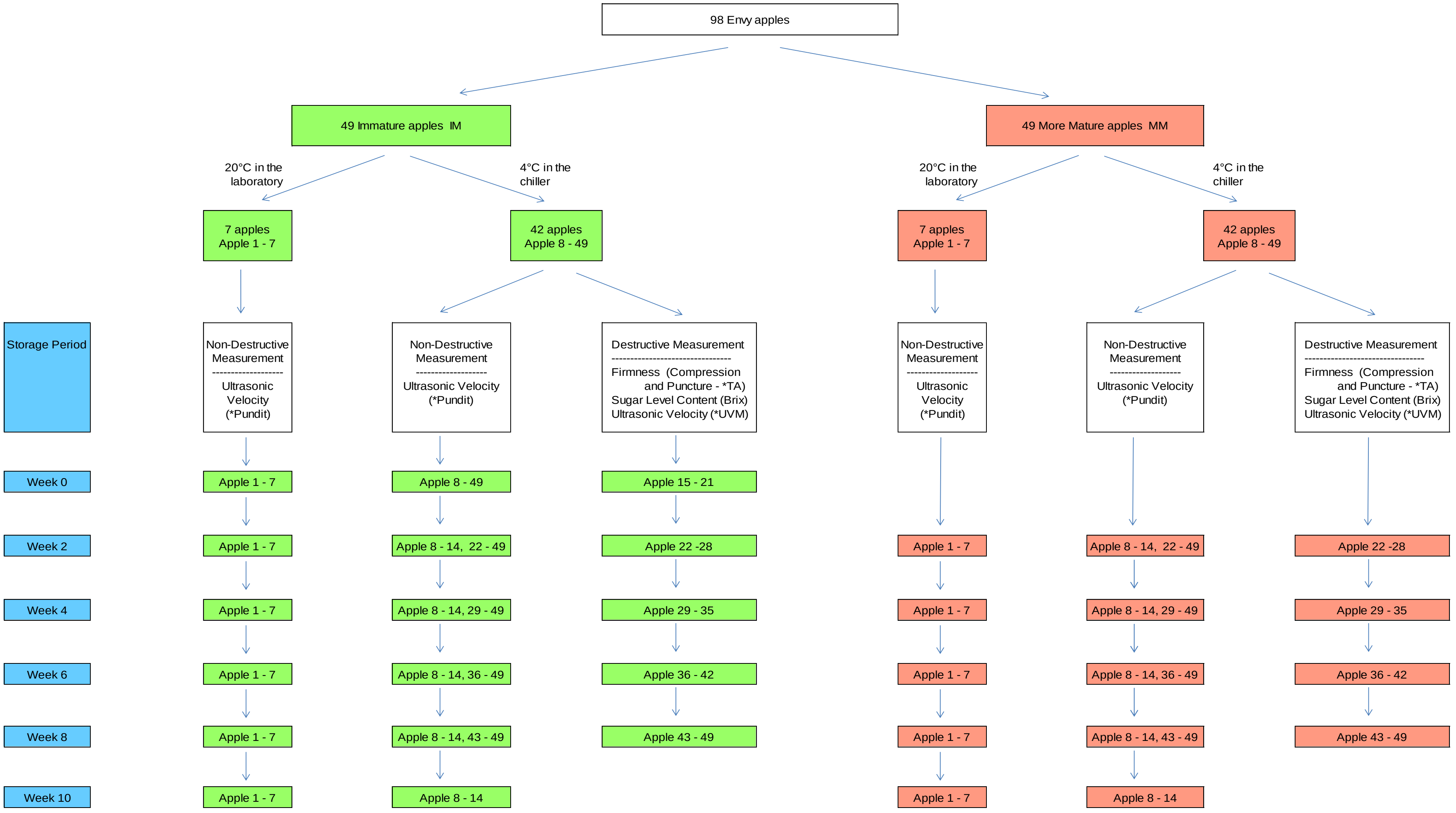


Table 16: Sources of uncertainty in calculating the estimation of the measurement of uncertainty for an ’Envy’ apple experiment

Test	Type of test	Non-destructive (NDT) / Destructive test (DT)	Instrument used	Variables			Sources of uncertainty		
				Measured parameters		Measured properties	Accuracy *u(A)* Verification of the instrument for fit for use	Precision *u(P)* Repeatability/ Reproducibility	Instrument Uncertainty *u(I)* Manufacturers’ specifications
1	Pundit Plus System	NDT	Pundit device	Time of flight, ultrasonic path	Anisotropy of an apple, Axial and Radial measurements	Ultrasonic velocity	✓ Verified against sound velocity in air	✓ Repeatability of 7 to 49 apples	✓ PUNDIT device, Oscilloscope, Thermometer, Humidity, Vanier calliper
2	Compression test	NDT	Texture Analyser (TA)	Area under curve	Anisotropy of an apple, Axial and Radial measurements	Firmness (skin and flesh elasticity)	✓ Verified at 5.000kg using Reference Standard weight	✓ Repeatability of 7 apples	✓ Force
3	Puncture test	DT	Texture Analyser (TA)	Maximum puncture force	Anisotropy of an apple, Axial and Radial measurements	Flesh firmness	✓ Verified at 5.000kg using Reference Standard weight	✓ Repeatability of 7 apples	✓ Force
4	Sugar level content or Total Soluble Solid test	DT	Hand Refracto-meter	% Sugar content level	Anisotropy of an apple, Axial and Radial measurements	Sugar level content	✓ Verified with distilled water and 5% sucrose readings	✓ Repeatability of 7 apples	✓ Refractometer
5	Ultrasonic Velocity test	DT	Ultrasonic Velocity Meter (UVM)	Time of flight, ultrasonic path	Apple puree	Ultrasonic velocity	✓ Verified with an external digital thermometer	✓ Repeatability of 7 apples	✓ UVM, Water bath

2.2.8 Summary of the Envy apple experimental design

The summary of the non-destructive and destructive measurements for the ‘Envy’ apple is outlined in Figure 34.



* Remarks: Pundit : PUNDIT Plustransducer system UVM : Ultrasonic Velocity Meter TA : Texture Analyser (compression and puncture tests were started on Week 2.)

Figure 34: Design of Envy apple experiment

2.2.9 Data analysis

The results were analysed by using Microsoft Excel (Version 2010, IBM Corporation, Chicago USA), IBM SPSS Statistics, Version 22 (IBM Corporation, Chicago USA) and MATLAB (Version R2013a, the Math Works Inc., MA, and USA). All statistical data analysis was based on p-value <0.05 (two-tailed).

2.2.9.1 Descriptive statistical analysis

The descriptive statistics provided the means, standard deviation (SD), standard errors (SE) for all variables in the experiments. This analysis was to observe a preliminary trend of the measured parameter in the tests.

2.2.9.2 *t*-test (Comparing two means)

Dependent and independent-means *t*-tests were carried out by comparing two means of axial and radial measurements of the measured variables onto the apples. The analysis was conducted to check if differences in the following parameters were significant:

1. Anisotropy measurements: Axial and radial directions
2. Maturity levels: M and MM fruit
3. Storage temperature setting: 4°C and 20°C

2.2.9.3 Pearson’s Correlation

A Pearson’s correlation coefficient (Bivariate correlation) was computed to assess the relationship between two variables in this experiment. It was to check the strength of a relationship of two variables in the following measurements:

1. Ultrasonic velocity and firmness by the compression test

2. Ultrasonic velocity and firmness by the puncture test

3. Ultrasonic velocity and the sugar content

4. Ultrasonic velocities between an intact apple and apple puree

2.2.9.4 Principle Component Analysis PCA

Principal Component Analysis (PCA) was conducted to study the trend of a set of the designated variables (ultrasonic velocity, firmness through compression and puncture forces and sugar content) by using linear factors, among the studied variables in the experiment (Field, 2005). The analysis tests the degree to which quality changes assessed by ultrasonic propagation (velocity), physical/ mechanical (firmness through compression and puncture forces) and flavour (sugar content) parameters are associated with the apple ripening during storage due to the fruit senescence.

2.3 Swedes: Detection of Brown Heart

The experimental work was conducted from May to June 2015. About 100 swedes from the BH batch were delivered by a swede supplier in Wellington, UK and were stored at the laboratory cold storage at 4°C until testing. 65 defective BH vegetables were examined from the batch. Meanwhile, seven healthy swedes were bought from supermarkets in Leeds. These measurements were conducted by Dr. Chu under the supervision of Dr. Holmes using the methodology partly developed by Mohd Shah. Figure 35 shows (A) the internal flesh of the defective BH (yellowish flesh with random brown spots) and (B) healthy (yellowish flesh without any brown spot) of the swedes. This swede study focussed on:

(1) a laboratory-developed BH Index,
(2) categorisation on BH severity levels for samples based on a visual inspection,
(3) ultrasonic velocity measurements by a PUNDIT Plus system testing, and
(4) Texture Profile Analysis (TPA) of flesh firmness based on a puncture test measured by a Texture Analyser (TA)

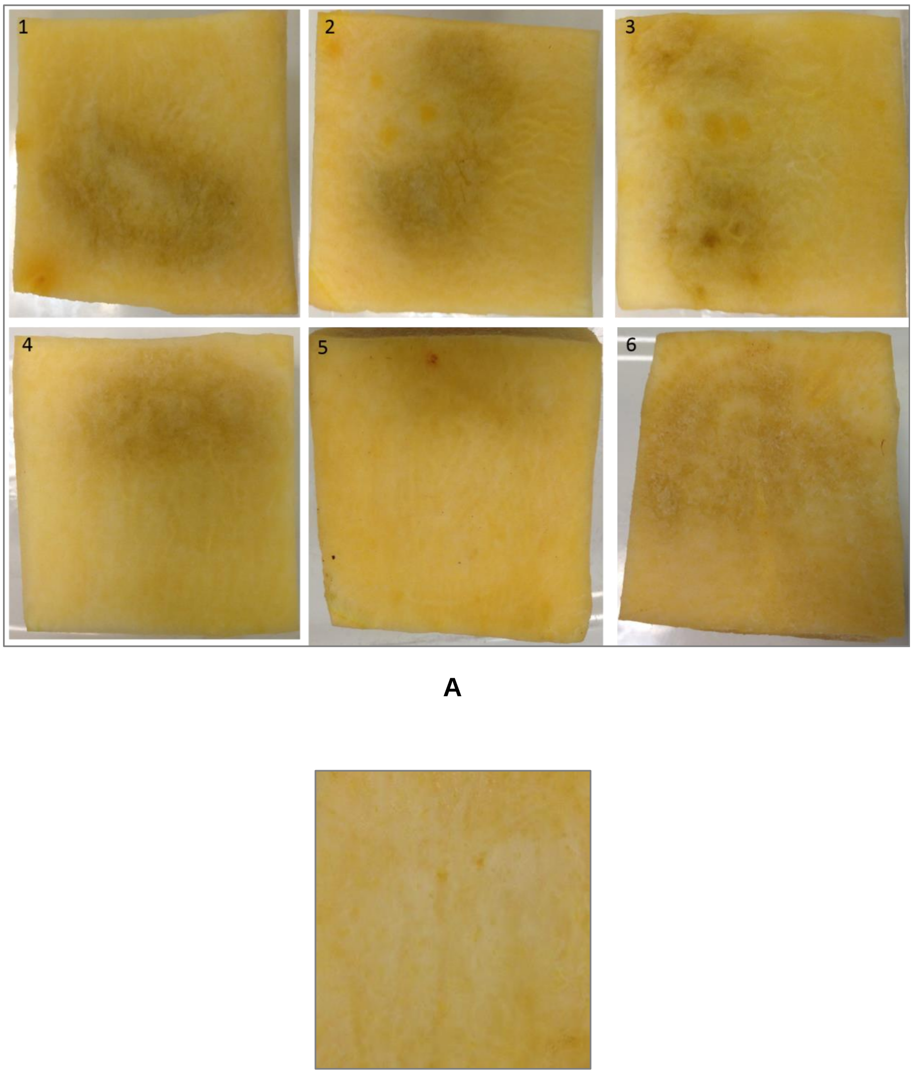


Figure 35: Cross-sections taken from the internal swede samples: (A) examples 1 to 6 showing the appearance of BH swede flesh randomly selected from the batch (yellowish flesh with spatial variations of brown spots; and (B) healthy swede (yellowish flesh without any brown spot). The swedes in the photos were about 50mm of cross cut section.

2.3.1 Ultrasonic velocity measurement of swedes by a PUNDIT Plus Transducer system measurement

The ultrasonic velocity of swede was determined by the PUNDIT Plus system measurements axially to investigate a correlation between the velocity and BH. The measurement procedures were similar to the section

2.1.1 and were repeated for samples B1 – B87. Four repeat measurements were taken in each swede and an average was calculated. After the velocity measurements had been obtained, the swede samples were cross-sectioned and visually inspected for the presence of BH (the severe BH levels based on the laboratory-develop BHI).

2.3.2 Laboratory-developed BHI and visual inspections

A laboratory-developed BH Index (BHI) measurement methodology was prepared by Dr. Chu for the determination of the BH severity in swede. Ten categories of the BH indicated the severe brownness and the size of the defective regions.

2.3.3 Texture Analysis by a puncture test

A measurement of force/displacement was obtained by Dr. Chu using the instrumental method described in the sub-section 2.2.4 with the following variations. A 2 mm diameter stainless steel punch cylindrical probe was used. The set-up test speeds were 2.0 mm/s (pre-test), 1.0 mm/s (test), and 10.0 mm/s (post-test). The puncture distance was 10.0 mm and the trigger force was 0.049 N. The skin of the fruit was sliced radially by using an apple peeler at the marked points. The BH was cut out into a proper dimension and the cut portion was measured. The measurement was conducted for all respective swedes B1 to B20. The graph of force versus time was generated and a maximum puncture force was expressed in Newton (N).

2.3.4 Data analysis

The results were analysed using Excel (Version 2010) and IBM SPSS Statistics (Version 22, IBM Corporation, Chicago USA). All statistical data analysis was based on *p*-value < 0.05 (two-tailed) as follows:

2.3.4.1 Descriptive statistical analysis

The descriptive statistics analysis was conducted to observe a preliminary trend of the measured parameters. The mean values, standard deviation (SD) and the standard errors (SE) were calculated.

2.3.4.2 Pearson's Correlation

The graph was plotted to assess a correlation between the ultrasonic velocity, the BH in swede and flesh firmness by the best fitted regression line and the Pearson Correlation.

Chapter 3
Results and Discussions

Chapter 3 reports and discusses the findings the three parts of the experiments using the methods provided in Chapter 2. The collected experimental data were analysed in association with three raised research questions in this thesis:

(1) How can the PUNDIT Plus Transducer system measurement be characterised and optimised so that ripening stages and internal quality can be assessed?

(2) How do (i) the changes in ripeness and (ii) anisotropy in apples during storage affect the ultrasonic velocity, together with firmness and sugar content measurements, and (iii) the correlation among those quality measurements using the different testing?

(3) How does BH in swede affect the velocity of sound and firmness measurements?

The PUNDIT Plus system techniques to measure the ultrasonic velocity through a sample, the texture analyser to measure the firmness via the compression and puncture tests, and a hand-held refractometer to measure sugar content were used to conduct the three part of the experiments in this study. The ultrasonic measurement techniques were developed, optimised, and characterised to be used together with firmness and sugar content parameters in the apple and swede experiments. These experiments were designed to ascertain (1) if the PUNDIT Plus system techniques (via the ultrasonic velocity) can be utilised to assess the ripening in apples and to detect the Brown Heart in swedes and (2) if a correlation among those measurements was significant.

3.1 PART 1: Characterisation and optimisation of PUNDIT Plus Transducer measurements

In this part, the PUNDIT Plus Transducer system measurement was characterised and optimised prior to the designated apple and swede experiments. The following necessitated checking during the PUNDIT measurement: (1) Sound fields and a focal length of the ultrasonic transducer, (2) Waveform analysis in the time domain, (3) Regions of the waveform of the frequency FFT, (4) Natural frequency, (5) Insertion loss measurement of amplitude frequency signal, (6) Verifying the measurement setup accuracy and (7) Experimental errors in ultrasonic velocity measurement.

3.1.1 N of the ultrasonic transducer

The calculated N distance for the transducer was approximately 100 mm for the axial and radial measurements. The calculation was the followings,

$$\mathrm{N} = \frac{D^2 f}{4c} = \frac{(0.05)^2 \times 39000}{4 \times 281.7}$$

$$= 0.087 \approx 100$$

where, the diameter of the transducer, D was 0.05 m, the frequency, f was 39 kHz that is equal to 39000 Hz and the sound velocity of the axial measurement is 281.7 $\mathrm{m/s}$ for the “*Envy*” apples (the average time-of-flight value was used in this sound velocity measurement). The value was equivalent to the estimated focal length of the natural focus of the working transducer throughout the experiments. The general diameter of an apple was about 50-80 mm, which was the distance of ultrasonic wave propagated through the fruit. This result means that the N zone was larger than the dimension of the fruit. Therefore, this finding verified that the wave travelled wholly through the tested medium and the measurement was taken within the focal length.

Meanwhile, the wavelength was approximately 7 mm. The value means that the wavelength of sound and the distribution of the propagated pulse were much smaller than the dimension of the fruit.

$$\lambda = \frac{c}{f} = \frac{281.7}{39000} = 0.00722 = 7$$

3.1.2 Waveform analysis in the time domain

The pulse-echo sensitivity was determined by the first arrival of ultrasonic wave from a traced waveform that was based on a relationship of the amplitude between the triggered pulse set at zero and the first delay time signal through the apple (Figure 20A). The traced waveform of the radial measurement of the apple was identified in the oscilloscope screen in C1 (Figure 20B). The circled portion of the waveform at the high frequency component was expanded in Z1 (Figure 20C) and frequency components were represented for the expanded waveform (Figure 20D).

Three distinct components were observed on the voltage against time plot, represented as: **Region A**, **Region B** and **Region C** in Figure 20C. Firstly, in **Region A**, an excitation pulse was triggered from the transmitting transducer and it was set as the starting time, 0 μs. Secondly, the first arrival of the pulse was seen in **Region B**. The left hand cursor on Z1 was placed to coincide with the signal trigger, defining zero time. The right hand cursor was placed at the first slope of the arriving signal. The time difference between the delayed and started times was determined as the flight time of the pulse (272.8 μs). The time of the first arrival pulse was the highlighted delayed time. The characteristic of the waveform indicating as the region of interest was used as a guide throughout the experiment. Meanwhile, the frequency composition of the pulse altered between **Region B** and **Region C** and the transition coincided with the transition from near to far field. However, it was not clear if the coincidence occurred. The magnitude of the time travelled is dependent on the acoustical characteristics of the transducer and the apple tissues, as well as the acoustical distance (Povey, 1997). Thirdly, the frequency component in **Region C** began at 892.52 μs, where it occurred at low frequency. Here, the waveform started to be distorted. It was observed that the PUNDIT device displayed the reading of the time of flight in this region.

Subsequently, the frequency spectrum in Figure 20D described the frequency component of the expanded waveform. No ultrasound pulse was received In Region A. However, a dominant low frequency component was detected between 0 to 10 kHz which was synchronous with the emission pulse. The short burst of the amplitude of the power at approximately 39 kHz indicated the maximum frequency of the transducer. The result confirmed that the 39 kHz was the working frequency of the transducer throughout the experiment.

The post-study investigating a tendency for the PUNDIT instrument to display an incorrect transit time for the apple experiment was discussed, based on Figure 36 (Long, 2000) and Figure 37 (Povey, 1997). Figure 36 displays a principle of PUNDIT measurement of Long's study, based on a detection of the 1st negative arriving signal (cycle) above the PUNDIT device's threshold crossing value (250 µV in this case) with the 54 kHz transmitting transducer. The transducer is excited by a 500 V pulse, generating an ultrasound pulse which propagates through a 100 mm thick medium and then is collected by the receiving transducer. Next, the first received pulse is averaged by an oscilloscope. An example trace is shown in purple and labelled a(t) in Figure 36. Subsequently, the signal amplitude of the PUNDIT device is reduced to half of the original value by reducing the amplitude of the exciting pulse and the resultant signal is displayed in blue and labelled a(t)/2. This first half negative arriving pulse is detected at the later time $t_{a(t)/2}$ because the device has set up a specific threshold value at 250 µV. This time shift is considered to be within the acceptable uncertainty range for the commercial PUNDIT device. However, if the first half negative amplitude is below threshold value, the device recognizes its first arriving signal at the 2nd negative amplitude instead. As a result, this detection by the reading directly from the PUNDIT display generates incorrect data (Long, 2000).

This point is illustrated in a different way in Figure 37. In this figure, the detection threshold is gradually increased, creating a jump in timing of the order of the period of the wave. Clearly, large timing errors can be created, particularly for pulses with a relatively large number of cycles. Therefore, this is why the reading from the PUNDIT display for ultrasonic time of flight is likely to be less accurate compared to the oscilloscope measurement, (although more convenient, economical and fairly acceptable in some practices). This is an example of artefacts (a systematic error that generates consistent, reproducible, incorrect data).

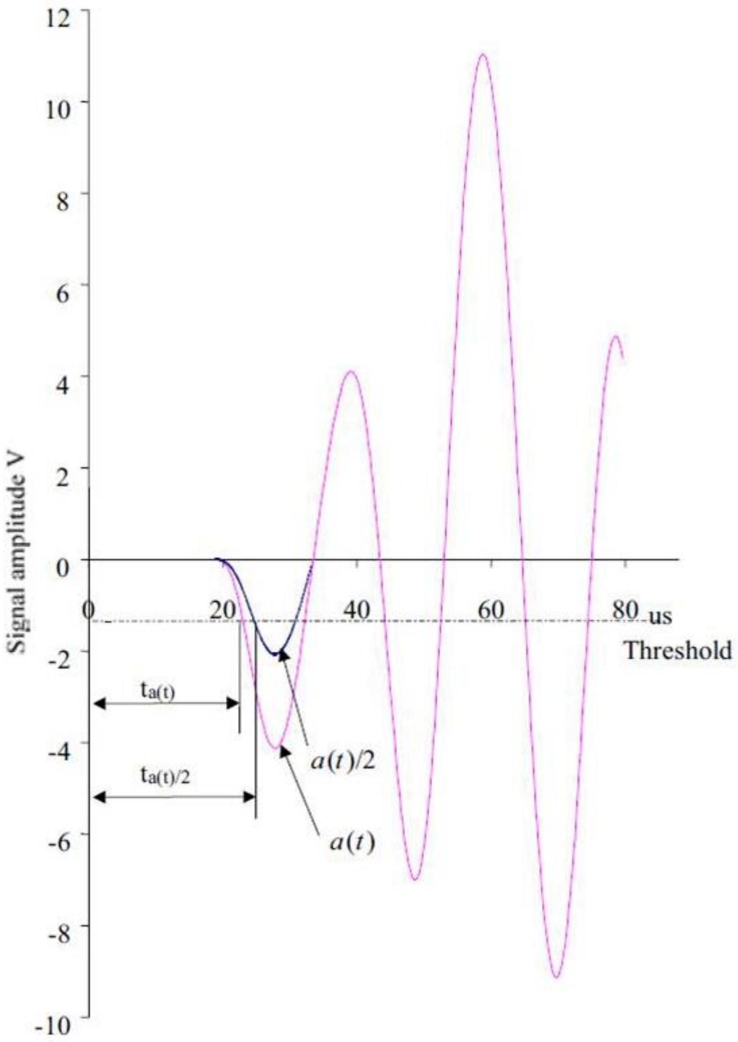


Figure 36: An ultrasonic pulse detected after passing through 100mm thick aluminium (Long, 2000). The example trace is shown coloured magenta and labelled a(t). Following the reduction of the signal amplitude of the PUNDIT device to half of the original value by reducing the amplitude of the exciting pulse, the resultant signal is displayed in blue and labelled a(t)/2. The trigger point in both cases is on the negative slope of the pulse.

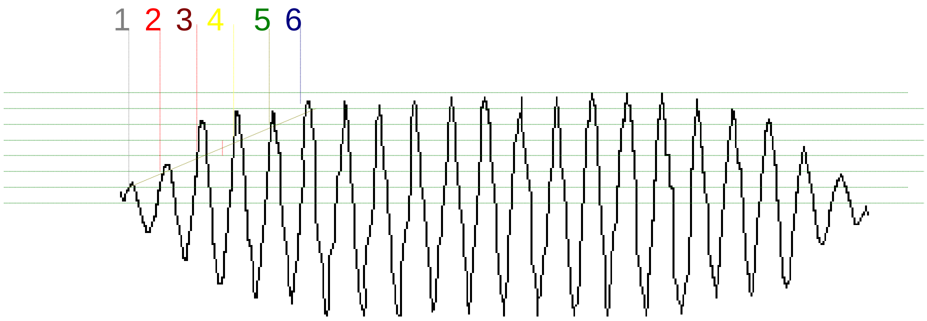


Figure 37: Illustration of the impact of changing detection threshold on a detected ultrasound pulse showing the generation of timing errors of integer multiplications of the period of the detected wave (Povey, 1997). In this figure the trigger point is chosen as a positive slope, rather than the negative slope used in the illustration of Figure 36.

3.1.3 Spectral analysis in the frequency domain using the FFT signal

3.1.3.1 Regions of the waveform of the frequency FFT signal

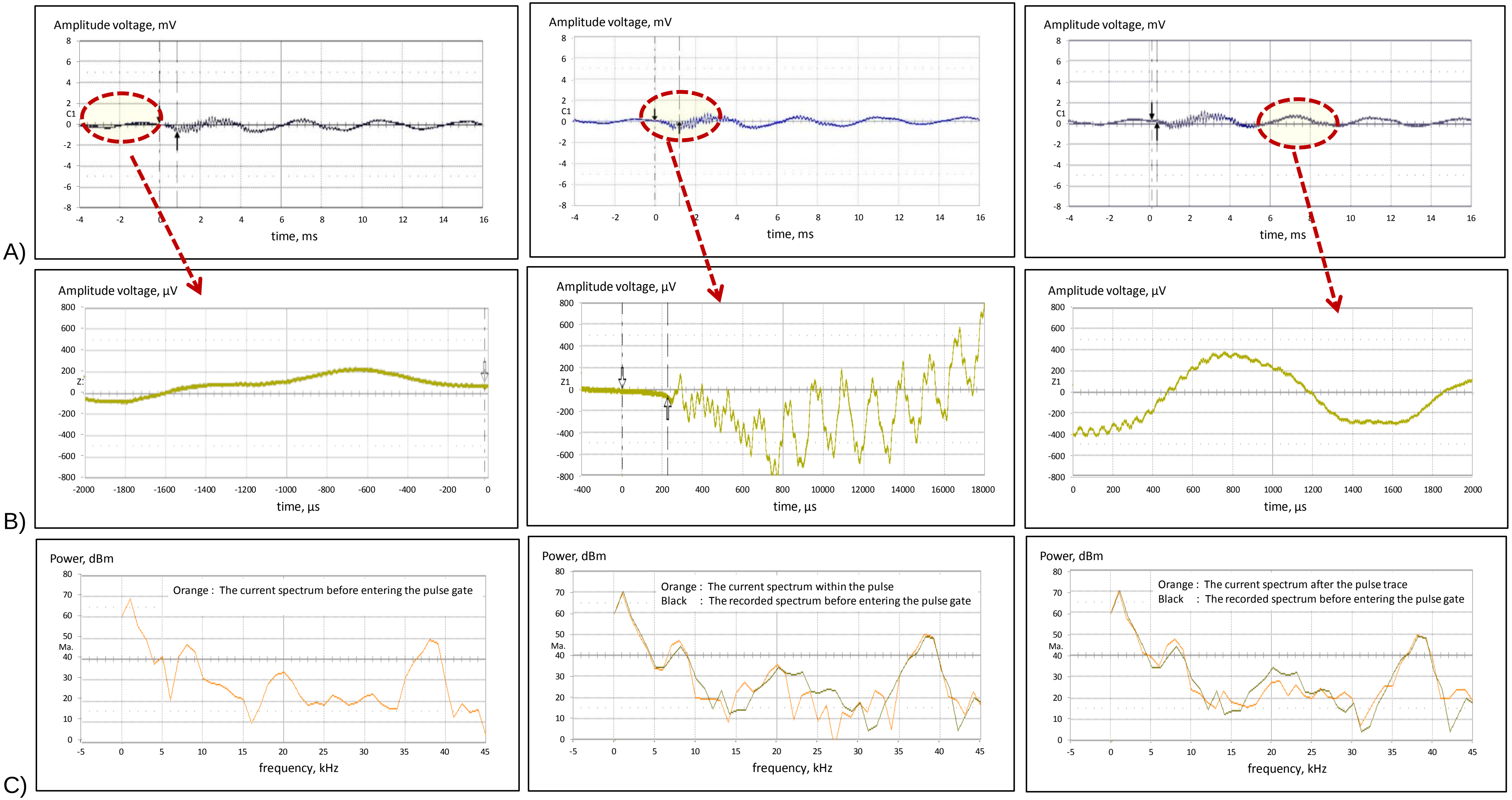
This section showed the findings of three different regions of the waveform of the frequency FFT signal: (1) before entering the pulse gate, (2) within the ultrasonic pulse and (3) after the pulse trace. They were based on the following oscilloscope parameter variables: PRF usually 50 Hz, number of 100 averages and the zoom scale of 200 μV for the amplitude powers and 200 μs for the ultrasonic transit times. An apple was used in between the two transducers.

A plot of frequency amplitude spectra (Ma., math function traces of an oscilloscope using FFT, power spectrum) of each region of the waveform was depicted in the third row of Figure 38(C), based on the chosen time domain signal in C1. This selected region of the waveform was amplified in Z1. The frequency spectra of the three region of the pulse trace were in Figure 38(C) 1, 2 and 3 respectively.

The amplitudes of the frequency spectra for the three pulse trace regions did not show much visually significant to one another. These insignificant spectra may have been because the first arrival and second pulse coincided with each other when the PRF was set at 50 Hz in this experiment. At the

50 Hz, an electromechanical effect and electro pulse interfere with one another. In addition, background noise possibly occurs.

Therefore, another post-study was conducted to further investigate an impact and an optimum setting of PRF on spectral analysis in the frequency domain. The results of this investigation are discussed in the next section with an introduction of experimental artefact in this study.



1. Before entering the pulse gate
2. Within the ultrasonic pulse
3. After the pulse trace

Figure 38: Row A) a region selected in a waveform in C1 before, within, and after the pulse traces; Row B) the highlighted region of C1 is expanded in Z1; Row C) Frequency of of Fast Fourier Transform (FFT) signals

3.1.3.2 PRF and its impact

The study of Pulse Repetition Frequency (PRF) was to select an optimum PRF value for the PUNDIT measurements and ensure the ultrasonic pulse had completely died away prior to the second pulse arrival. The results of comparison of impact of the PRF of 50, 25, 20, 10, and 1 Hz were reported by using air in between the two transducers. The plots of frequency amplitude spectra were visually evaluated every time the level of the PRF was changed. Figure 39 displays the comparison of five frequency spectra of the investigated PRF: (A) before entering the pulse gate, (B) within the ultrasonic pulse and (C) after the pulse trace. The results of the spectral amplitudes showed that sufficient low PRF at 10 Hz (in a light green spectrum) was the optimum setting for the measurement system. It ensured that the pulse signal had died away before the next pulse began.

On the other hand, the high PRF has tendency to produce the trailing edges of a former pulse to coincide with the leading edge of the latter signal. This interference can cause a false interpretation in an envelope of the waveform. Therefore, this was another example of artefacts (a systematic error that generates consistent, reproducible, incorrect data). Selections on a proper region of ultrasonic signal (windowing) are important to analyse an FFT signal of a waveform (Lempriere, 2002). In addition, decreasing PRF pushes up the amplitude power. It also reduces the background noise (such as an acoustic ‘buzzing’ sound, undesired echo, and random noise coming from the electrical or mechanical noise that can overlap and interfere with the region of interest (Lempriere, 2002; Povey, 1997).

Hence, the results of the spectral amplitudes supported the assumption that the decrease of the PRF from 50 to 10 Hz pushed up the amplitude power at 39 kHz. This value was applied in the experiments. In addition, the setting ensured that the coherence of the pulse sequences was consistent during the measurements.

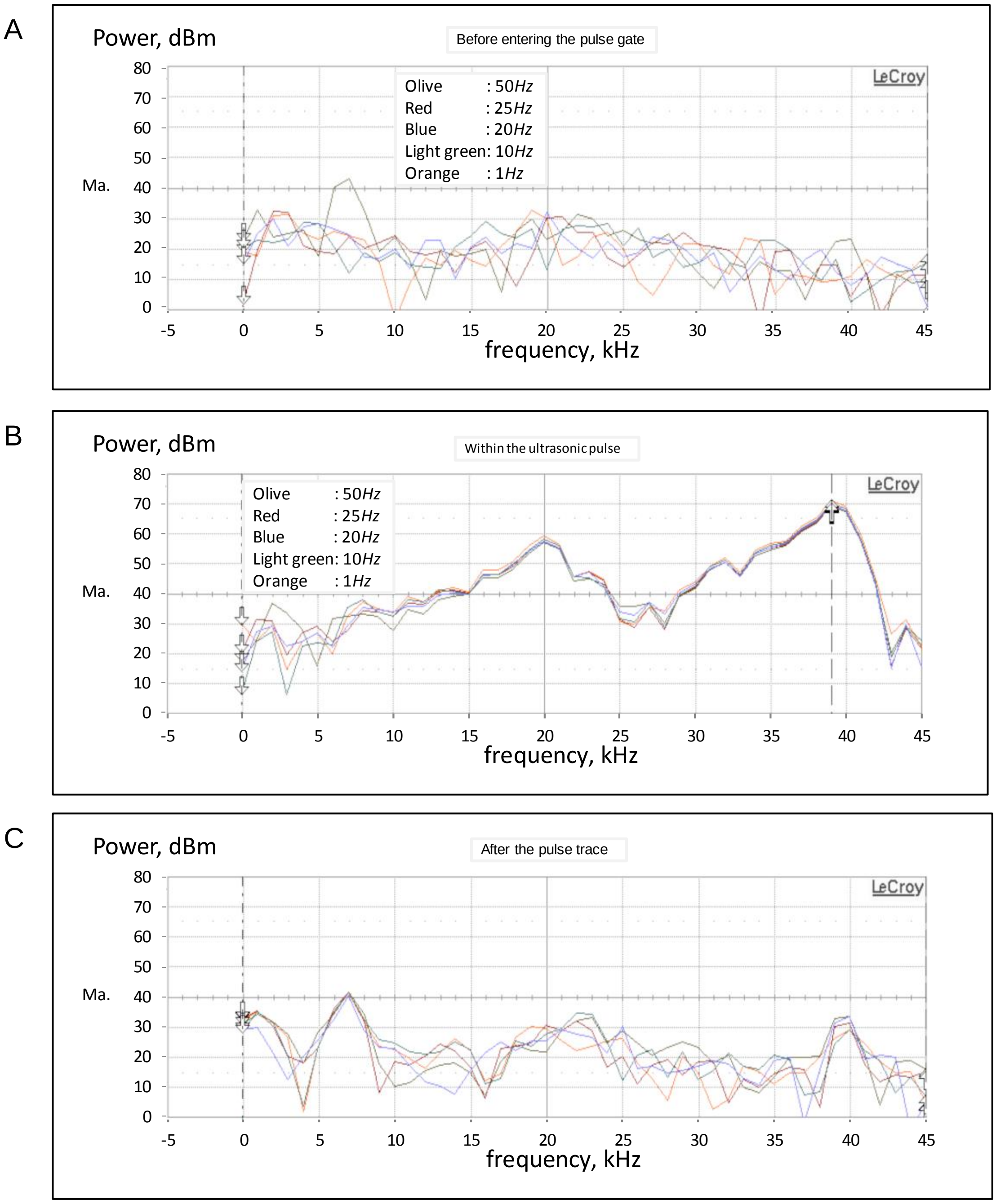


Figure 39: An impact of the PRF: Comparison among the five frequency spectra of the pulse repetition frequencies (50 Hz in dark green; 25 Hz in Red, 20 Hz in blue, 10Hz in light green and 1 Hz in orange): (A) before entering the pulse gate, (B) within the ultrasonic pulse and (C) after the pulse trace

The possibility of an impact of PRF on spectral analysis and time domain signal analysis are discussed in the following sub-section:

At a high PRF, such as 50 Hz, the time for one frequency (rate of oscillation) takes 0.02 s or 20,000 µs. This rate of oscillation occurs in rapid speed, where the first pulsed tone bursts and produces an initial wave. Then, the second pulse follows as an echo. This signal produced from high frequency disappears quickly. The trailing edge of a first pulse of the frequency overlapps with the second arriving pulse (Figure 40A). This behaviour affects the accuracy of averaging because an accumulative series of the trailing edges coincides with the leading edge of the ‘echoes’ and can generate large phase shifts and change in amplitude. This interference can cause a false interpretation in an envelope of the waveform. Therefore, this is an artefact. In contrast, a signal generated by low frequency dies away slowly (Figure 40B). The signal has died away before the next pulse begins. Consequently, no trailing edge occurs between and among signals (Povey, 1997).

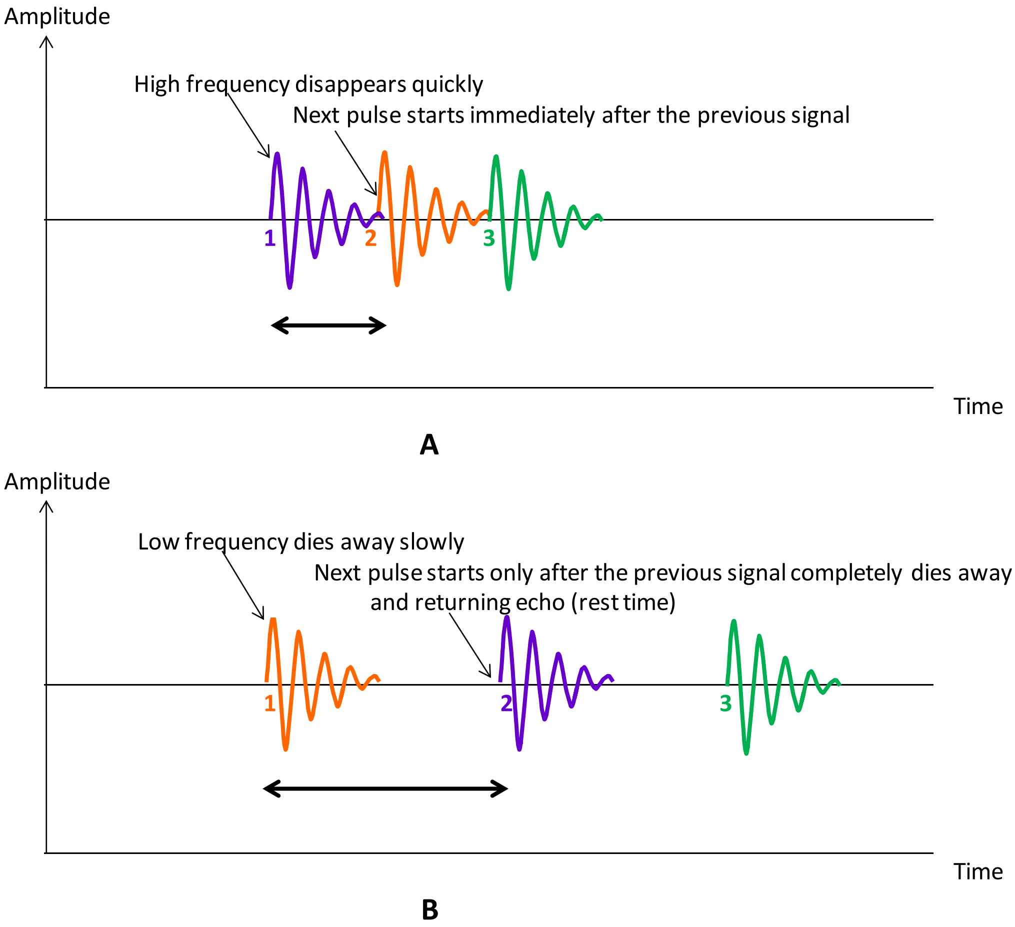


Figure 40: (A) High frequency dies away more quickly compared to (B) low frequency that dies away more slowly.

3.1.4 Natural frequency

This study aimed to characterise the behaviour of PUNDIT measurement using air as a sample to understand the interference of the PUNDIT's audible buzz, focussing on frequency components below 50 kHz. Figure 41 provides an example of environmental electromagnetic and acoustic interferences. The time domain (blue) and frequency domain (blue and grey) were represented the two replicates. At approximately 2 kHz, the highest signal amplitude was observed (electromagnetic interference). At the same time, other instruments were also operated in the laboratory and they can contribute to the audible sound production (environmental noise). Slight tapping on the table, closing door, or whistling disturbed the frequency signal amplitude between 2 and 20 kHz. Meanwhile, no environmental activity was at approximately 39 kHz (the working frequency of the transducer). Therefore, the characteristics of natural frequency were recognised.

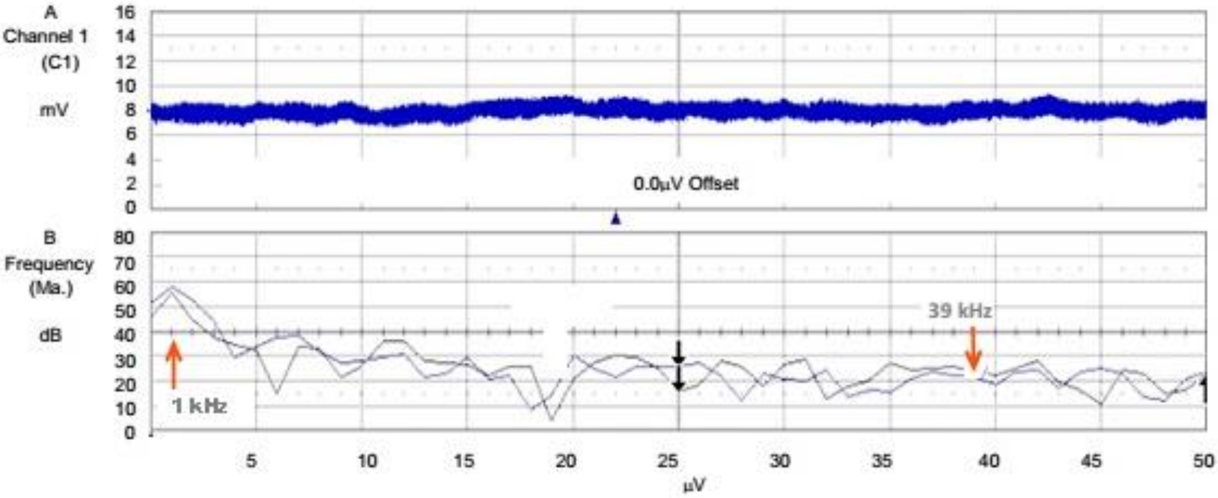


Figure 41: A. Time domain signal (C1) detected with the pulser switched off. B. A large 2 kHz (electromagnetic interference) component can be seen in the frequency domain (Ma). 39 kHz is the frequency of interest (Both frequencies displayed in blue (former) and black (latter) in the frequency domain were two replicates).

3.1.5 Waveform averaging value

The study of the waveform averaging value without a sample was to minimise those spurious random signals (for example electrical or environmental effects). The signals were not correlating with the pulse. However, they appeared in the frequency domain. Figure 42 presents (A) the time domain and frequency domain signals for (B) 20 and (C) 40 sweeps. The frequencies of before (former, in black spectrum) and after the adjusted sweep numbers (in blue spectrum) were stated on the figure with the respective arrow. Averaging at 40 sweeps (C) reduced the interfering environmental noise and improved compared at 20 sweeps (B). Thus, 40 sweeps was the optimum setting for the experiments. This study demonstrated that a proper selection of the waveform averaging value could optimise performances of the frequency spectra amplitude.

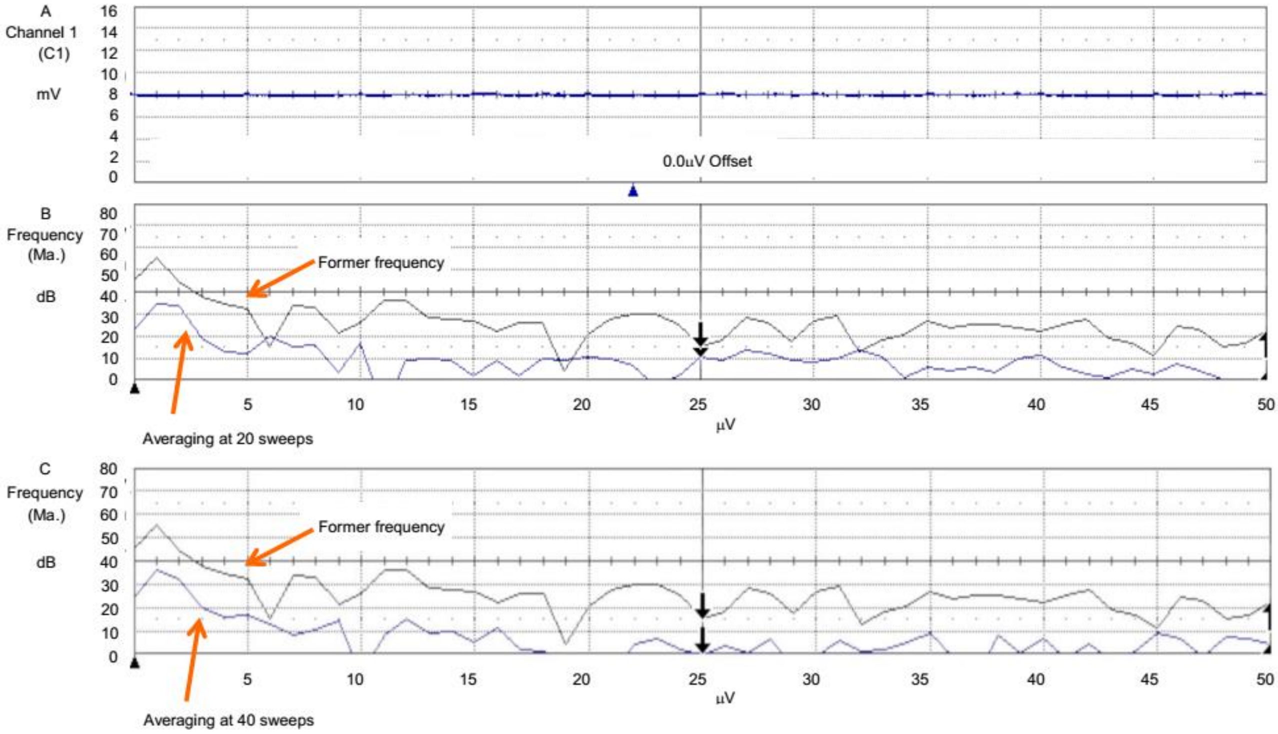


Figure 42: Proper selection of averaging level can affect the frequency spectra analysis: A. Screen shot of a waveform; (B) Averaging at 40 sweeps produced more distinct change in the signal amplitude than that (B) at 20 sweeps (the two spectra in plot B and C displayed in blue are the former recorded frequency, both spectra are identical. Meanwhile, the frequency displayed in black was the changed averaging sweeps.).

3.1.6 Insertion loss measurement of amplitude frequency signal: Comparison of two frequency spectra

Insertion loss measurement of frequency signal amplitude was studied to understand the attenuation of the signal when the ultrasonic pulse propagated through the sample. Figure 43 depicts two frequency spectra in with an apple (the red plot) and without an apple (the blue plot). The low frequency labelled around 5 kHz and the high frequency labelled around 39 kHz were the focussed frequencies for this study.

3.1.6.1 Frequency spectrum in air (without an apple)

The highest signal amplitude was detected at 39 kHz for the frequency spectra in air (the blue plot) and this frequency was the region of interest of the measurement. Relatively high signal amplitudes were also detected below 20 kHz (audible sound ranges) such as around 5 kHz. The characteristics of the regions have been acknowledged for the ultrasonic measurements.

3.1.6.2 Frequency spectrum with apple

The amplitude frequency signal at 39 kHz with an apple decreased (the red plot) compared to the measurement in air (without an apple). Yet, the power spectrum at the low frequency (5 kHz) of measurement with an apple was slightly lower than its former frequency in air (without an apple). This behaviour indicated that the apple acted as a sound filter. The ultrasonic pulses were attenuated when passing through the fruit cell structures. As attenuation increases, the corresponding component in the power spectrum in a FFT signal decreased.

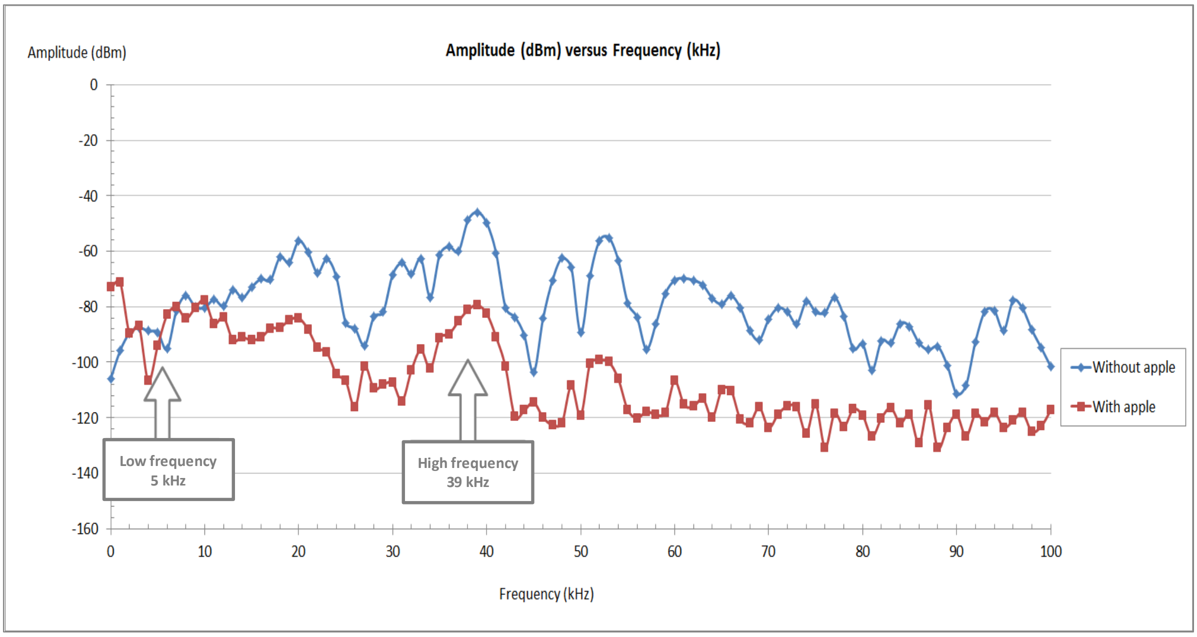


Figure 43: Behaviour of transducers, illustrated by two frequency spectra with and without an apple (the red and blue frequency spectra respectively), focusing on the high frequency around 39 kHz and the low frequency around 5 kHz

3.1.7 Verifying the measurement setup accuracy with the velocity of sound in air

The setup accuracy of the PUNDIT device and the oscilloscope measurement was verified by the velocity of sound in air. Based on the independent *t*-test result, at 95% confidence interval, no significant difference in the mean velocity of sound in air showed between the PUNDIT device (M = 344.7 m/s, SE = 0.3, N = 26) and the oscilloscope measurements (M = 343.7 m/s, SE = 0.2, N = 26), $t(50) = -1.63$, $p < 0.05$, two-tailed. These two velocity values closely agreed with the reported speed of sound in air value of 343 m/s provided by Sinclair (2001) and Ford (1970). Therefore, this finding confirmed the accuracy of the setup of the PUNDIT Plus System.

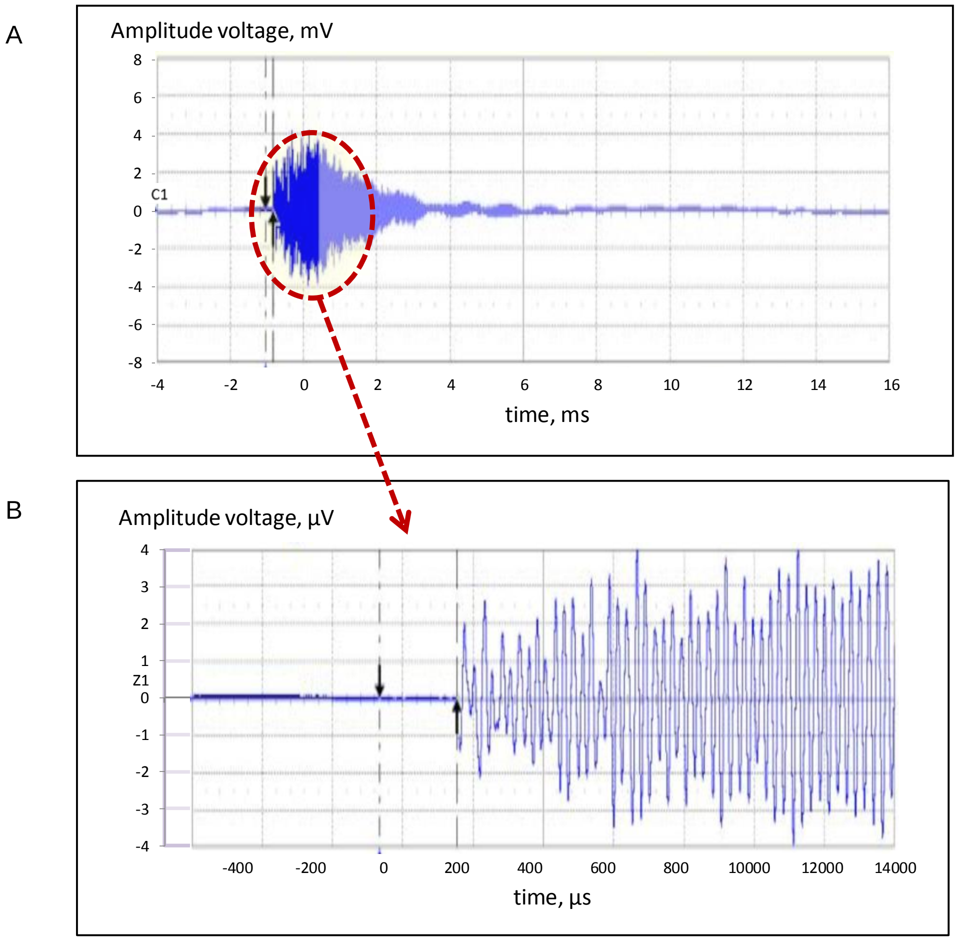


Figure 44: An example of the waveforms of velocity of sound in air for C1 plotted against time and Z1 plotted against time. The waveform in the C1 was zoomed at 1.00 mV and 200 µs to give the expanded waveform Z1.

3.1.8 Experimental errors in ultrasonic velocity measurement

This section introduces how experimental errors can affect accuracy and precision during the experiment. However, they can be estimated and accounted for. The sources of errors identified in this study were as follows.

3.1.8.1 Offset of the ultrasonic path distance

Air as a medium of the ultrasonic wave propagation. This experiment was to identify the offset of the ultrasonic path distance of the working transducers by using air path to compensate for the measurement error. The linear regression in Figure 45 shows a positive relationship between the measured ultrasonic path distance in mm (——) and the time of flight in μs by using air measurements. The linear expression $y = 0.3446x - 0.0773$ indicates that the correlation strength between the two variables was very strong, based on the coefficient correlation, $R^2 = 1$. The y-intercept of -0.0773 represents the offset value of the ultrasonic path distance while the slope *m* represented the speed of sound in air. The offset value was relatively small.

Next, in Figure 45, a corrected ultrasonic path distance with a linear regression $y = 0.3446x$ was constructed (- - - -), based on a subtraction of the measured distance from the offset value -0.0773. The corrected and measured path distance lines overlapped with each other because the offset value was very small. Therefore, the finding showed that the offset value for the ultrasonic distance was considerably negligible for the working transducer based on air as a medium of the ultrasonic wave propagation.

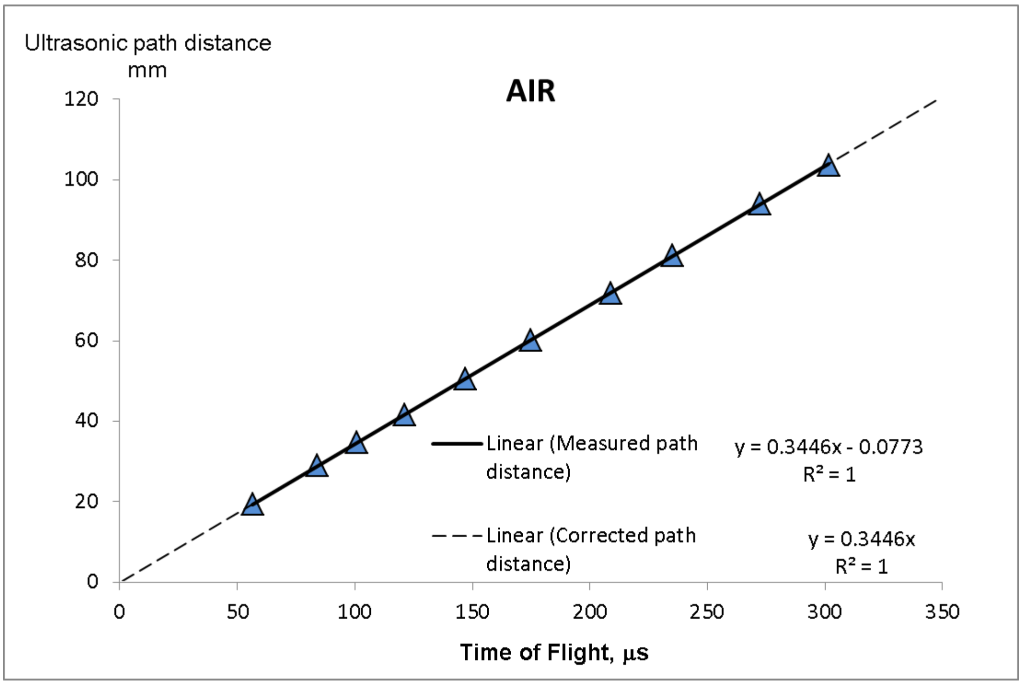


Figure 45: Ultrasonic distance offset of the working transducer based on air as a medium of the wave propagation. The offset value ~ 0.1 mm from the y-intercept of the regression line (——) of the path length (▲) was relatively small, thus the value was negligible. The corrected path distance line (- - - -) **overlapped with the former line.**

Apples as a medium of the ultrasonic wave propagation. The similar experiment was also conducted on *'Envy', 'Pink Lady'* and *'Royal Gala'* to investigate the offset of the ultrasonic path distance of the transducers by using an apple as a tested medium to compensate for the measurement error. The linear regression in Figure 46 shows a positive relationship between the ultrasonic path length in mm and the time of flight in μs by using the apple measurements. The linear expressions of the fruit indicate that the correlation strength between the two variables was also very strong based on the coefficient correlation R^2 = 1. The *y*-intercept of the regression lines between 0.0 and 0.3 mm represents the offset value of the ultrasonic path distance with the slope *m* representing the speed of sound in the apple. These offset values were also fairly small. In addition, the offset values for these apples were varied and thus suggested that the offset investigation should be determined on each sample type prior to the ultrasonic experiment.

Next, in Figure 46, a corrected ultrasonic path distance with a linear regression was constructed (- - - -) for each apple, based on a subtraction of the measured distance from the offset value respectively. Some of the corrected and measured path distance lines also overlapped with each other due of the small offset values. As a result, the finding indicated that the offset value for the ultrasonic distance was negligible for the working transducer, based on the apple as a medium.

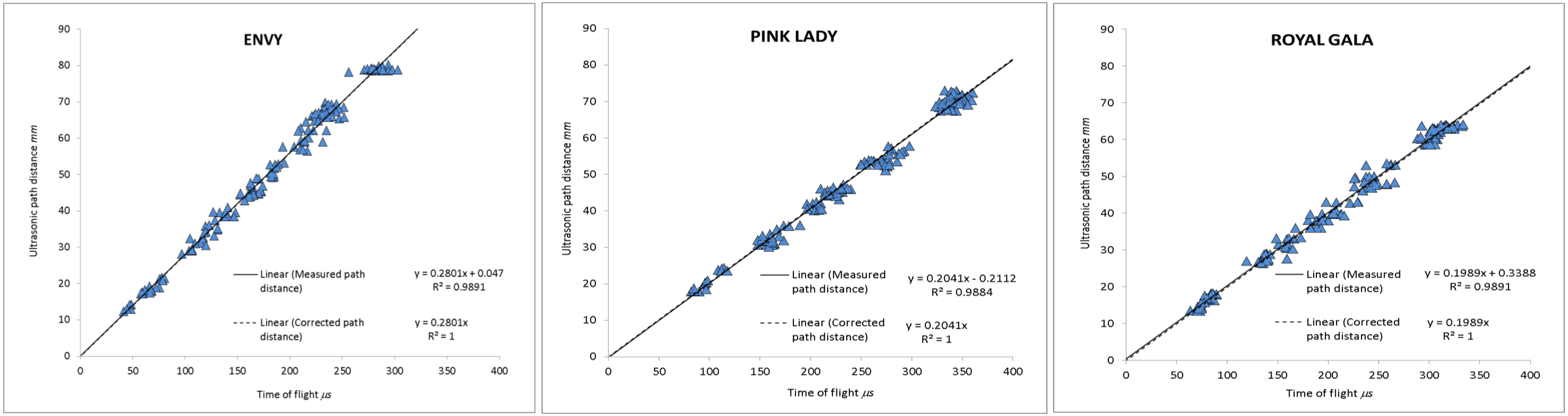


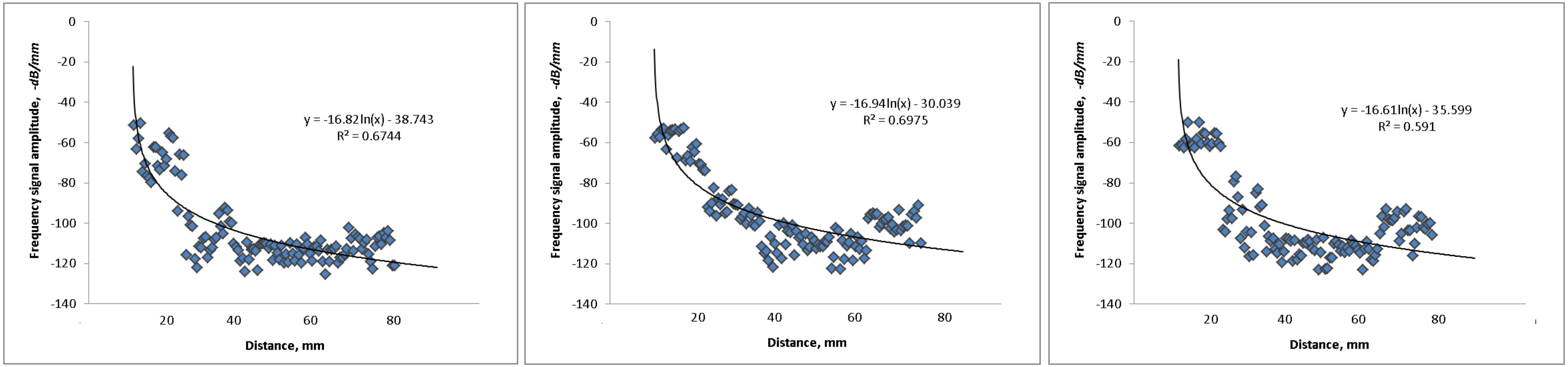
Figure 46: Ultrasonic distance offset of the working transducer based on an apple as a tested medium (three different apples, five fruit of each apple, five sets of apple slices, and five measurements of each slice set). The offset values between 0.0 and 0.3 mm from the *y*-intercept of the regression line of the path length (▲) was relatively small. Thus, they were negligible. The corrected path distance lines (- - - -) overlapped with the former line, inferring small offset values.

3.1.8.2 Ultrasonic pulse as bulk waves

A study of bulk waves was investigated to confirm that the ultrasonic wave was propagated inside the fruit, rather than as surface waves throughout the experiment. An experiment was performed in which the velocity of sound in the whole fruit and its five sliced portions was measured and compared. The findings showed that no significant difference among those measurements (within experimental error), $p < 0.05$ by using the two-way ANOVA (F (5,432) = 2.04, p = 0.07). The finding confirmed that the ultrasonic wave was propagated inside the measured fruit as bulk waves.

3.1.8.3 A study of attenuation of an ultrasonic pulse by using sliced apples

The attenuation of an ultrasonic pulse signal passing through an apple was investigated to confirm that the signal amplitude decreases exponentially against the ultrasonic path distance. The investigation was conducted by changing ultrasonic path lengths, and by recording the frequency in –dB/mm at 39 kHz (the frequency of the working transducer), using apple slices. The findings showed that the graphs were exponentially curved. This behaviour demonstrated the decreasing frequency signal amplitudes against the distance of the ultrasonic wave propagation. Therefore, the attenuation aspect of the ultrasonic signal amplitude through the sample was determined.



A. ‘*Envy*’

B. ‘*Royal Gala*’

C. ‘*Pink Lady*’

Figure 47: The attenuation of an ultrasonic pulse signal passing through the apple slices: the signal amplitude decreases exponentially as the ultrasonic path distance increases: A. ‘*Envy*’, B. ‘*Royal Gala*’ and C. ‘*Pink Lady*’, based on signal height of frequency

3.1.8.4 Variability in the apple dimensions, flight time of a pulse, and axial-radial of velocity measurements

The two-way ANOVA analysis was conducted on *'Envy'*, '*Pink Lady'* and '*Royal Gala*' apples to investigate the effect of the following variables performed in replicate measurements: (1) the dimensions of apples (the stem-calyx and vice versa positions) for axial measurements, and the angle positions from 0° to 360° around an apple (with approximately 30° in between two angles) for radial measurements; (2) the time of flight of the first arrival of ultrasonic wave for the axial and radial measurements and (3) the axial and radial directions of measurements as a preliminary study on apple cell anisotropy effect prior to the second part of the experiment in this project.

The results showed that the shapes and dimensions of the apples did not affect the ultrasonic velocity measurements. However, the measurements were influenced by the direction of the measurements (axial and radial). The mean values, the standard deviations and the standard deviations for the variables are displayed in Table 17. The findings of the two-way ANOVA indicated that the *p*-values for the three samples (*n* = 10) of *'Envy'*, '*Pink Lady'* and '*Royal Gala*' dimensions (0.5177), the axial-radial velocities (0.000), and the interaction between the fruit dimensions and velocities (0.8514). The first *p*-value indicated that the variability in dimensions of those apples did not affect the ultrasonic velocity measurements (*F* (4,270) = 0.81, *p* = 0.5177). However, the second *p*-value represented that the anisotropy in apples influenced the axial-radial velocity (*F* (5,299) = 1568.57, *p* = 0.0000). The third *p*-value showed no evidence of an interaction effect between dimensions of the apples and axial-radial velocity measurements.

These findings were supported by a multiple comparison test. Firstly, the *p*-values (>0.60) were large (between fruit pairs based on the dimensions). These values confirmed that the variability in the fruit dimensions did not influence the velocity measurements, based on indiscriminate velocity group means across all sample pairs. Secondly, the *p*-values between axial-radial pairs of the three apples verified that differences in the ultrasonic velocity between the axial and radial directions of the measurements were significant. The finding also indicated that the axial velocity was higher than the radial velocity. Hence, these preliminary findings demonstrated that the apple anisotropy did affect the ultrasonic velocity measurements for the apple ripening stages at the end of the supply chain (supermarket).

Table 17: Mean values and Standard Errors *SE* of dimension of apples, time of flight and ultrasonic velocity for ‘*Envy’, ‘Pink Lady’* and *‘Royal Gala’*

		‘Envy’			*‘Pink Lady’*			*‘Royal Gala’*		
		Mean	SD	SE	Mean	SD	SE	Mean	SD	SE
Dimensions of apples, mm	Axial	72.3	5.8	0.3	69.5	1.7	0.2	67.4	1.9	0.2
	Radial	70.1	2.9	0.2	70.3	1.3	0.2	69.4	2.0	0.2
Time of flight, μs	Axial	257.1	21.7	0.7	345.5	13.5	0.5	336.9	12.1	0.5
	Radial	305.0	14.4	0.5	383.9	7.9	0.4	387.7	13.9	0.5
Ultrasonic velocity, m/s	Axial	281.7	10.5	0.5	201.5	6.8	0.4	200.0	4.3	0.3
	Radial	229.9	7.4	0.4	183.1	4.1	0.3	179.2	5.1	0.3

Remarks: SD: Standard deviations, SE : Standard Errors, *N* = 5, *n* = 10

The means of the axial and radial ultrasonic velocities, 281.7 m/s and 229.9 m/s respectively, were above than those velocities at 114 m/s for ‘*Golden Delicious*’, 147 m/s for ‘*Washington Red*’ at 37 kHz and at 185 m/s for the unknown apples at 50 kHz reported by Self et al. (1992). Note: Different types of apple have different velocity ranges. The Self et al. (1992) study also did not focus the velocity differences between axial and radial measurements. Besides, the ripening stage of the measured apples was not reported: whether the stage was at the beginning of the harvest, the middle, or the end of the supply chain. The location of the sampling potentially influences changes of the measured ultrasonic velocity through the apples’ tissues. This is because ultrasonic velocity (ultrasonic properties) are associated with the firmness (physical properties) of the fresh fruit and vegetables (Schmilovitch and Mizrach, 2013; Mizrach, 2008b; Mizrach, 2008a; Self et al., 1992). The association is influenced by the change of the texture during the development and growth, maturity and ripening stages of the apples (Reeve, 1970; Reeve, 1953; Tukey and Young, 1942). The results suggested that anisotropy of apples was related to the textural property changes during the apple postharvest life.

3.2 PART 2: Assessment of ripeness in apples during storage

The experimental data on assessment of ripeness in apples during storage is discussed in this section. The first aim of this investigation was to test if the feasibility of the non-destructive technique of ultrasonic velocity in assessment of ripeness in apples during storage by studying an effect of anisotropy on ultrasonic velocity. The second aim was to establish a correlation between ultrasonic velocity (non-destructive techniques) and firmness measured by a compression and puncture test, and sugar content (destructive techniques). The third aim was to demonstrate the correlation by using the linear combinations of the measured variables in the experiment by Principal Component Analysis (PCA).

3.2.1 Effect of anisotropy of apple parenchyma on ultrasonic velocity measurements

Ultrasonic velocity was measured based on the time of flight over the wave propagation distance as an indicator of the ripening change of the fruit. Four observations were identified as general trends of the velocity curves between the axial and radial velocity measurements of the fruit, $p < 0.05$ (Figure 48A to D). Firstly, the radial velocity was higher than the axial velocity of the M and MM fruit from weeks 0 to 6. Second, after week 6, the axial velocity dominantly accelerated during storage, whereas the radial velocity remained stable. Thirdly, the findings showed that a convergence in the velocities occurred between the axial and radial measurements when approaching week 8 of the fruit storage. Fourthly, after week 8, the axial velocity was higher than the radial velocity.

Those findings above are elaborated. The mean axial and radial velocities were compared based on the fruit maturity levels (M and MM fruit) and storage temperatures (4°C and 20°C). Linear regressions described the patterns between the parameters, and *t*-tests were used to express the significance of the mean values between the variables.

3.2.1.1 Axial-radial velocities in the same maturity groups

For the first maturity group (the M fruit), the mean ultrasonic velocities of the fruit stored at 4°C varied from 209.0 to 277.9 m/s for the axial and 256.6 to 273.6 m/s for the radial measurements (Figure 48A). The values revealed that the radial velocity was higher compared to axial velocity from weeks 0 to week 6. However, the axial velocity changed dramatically compared to the radial velocity that remained stable during storage. The linear expression showed a strong positive relationship between the velocities and the storage time for the axial measurements ($R^2 = 0.72$), whereas no association was found for the radial measurements ($R^2 = 0.01$). Interestingly, the curves of the axial-radial velocities converged after week 8. The axial velocity exceeded the radial velocity after the convergence.

Similar patterns were observed in the M fruit stored at 20°C (Figure 48C). The mean ultrasonic velocities varied from 220.8 to 274.0 m/s for the axial measurements and 258.6 to 272.2 m/s for the radial measurements. The linear expression showed a strong positive relationship between the velocities against the storage time for the axial measurements ($R^2 = 0.86$), whereas no association was found for the radial measurements ($R^2 = 0.00$). The results of the 4°C and 20°C stored fruit indicated that the ultrasonic velocities were significantly different between the axial and radial directions of the measurements for the M fruit ($p < 0.05$).

For the second maturity group (the MM fruit), the mean ultrasonic velocities of the fruit stored at 4°C varied between 256.9 to 289.0 m/s for the axial and 262.2 to 297.7 m/s for the radial measurements (Figure 48B). The linear regression showed a moderate positive association for the mean axial velocities ($R^2 = 0.48$), and a weak negative association for the mean radial velocities against storage time ($R^2 = 0.24$). Similarly, the axial and radial velocity curves of the MM fruit converged at approximately week 8. The results of the MM fruit stored at 20°C also showed similar velocity trends. A significant velocity difference was identified between the axial ($R^2 = 0.80$, strong relationship) and radial ($R^2 = 0.06$, weak relationship) measurements in Figure 48D.

The discussions on the results are the followings. Firstly, the radial velocity higher than the axial velocity from weeks 0 to week 6 might be associated with textural changes and orientations of the apple cell structures during storage. Reeve (1953), Reeve (1970), Aguilera and Stanley (1999) and Taiz and Zeiger (2002) state that the textural changes and orientations of fruit cell structures indicate fruit ripeness. During ripening, the flesh parenchyma cells change from rounded to elongated shape in the radial direction and toward the core or the pit of an apple due to depletion of starch. Once the sugar starts to accumulate, the intercellular air space volume decreases. This causes collapses of the cells tissue structures due to the loss of turgidity of the middle lamella. These behaviour traits are possibly due to senescence. Furthermore, Povey (1997) and Self et al. (1992) highlight that ultrasonic wave travels slower in a gaseous medium, compared to liquid and solid. This is because the velocity of the wave depends on medium properties of adiabatic compressibility and density. Therefore, the changes from gaseous to gas-water mixture in the apple cells during ripening in the experiment demonstrated the effect of ripening on the velocity measurements.

A study by Khan and Vincent (1993a) also showed that arrangements of the cell columns of radial and axial orientations does affect mechanical measurements of an apple. It reveals that the axial direction requires 40% more force than a radial direction does by using a mechanical penetration test. The fruit flesh in the axial direction is harder to crack through as the force necessary to fracture cell tissues across the cell columns. Contrarily, the flesh fractures easier from the radial direction, when the penetration is parallel to the cell columns. From the findings of the study, it can be suggested that the dramatic changes of the axial velocities compared to those of the radial velocities in the apples is associated with the cell textural change, the softening effect in the fruit texture during storage.

The studies conducted by Mizrach et al. (1996) and Mizrach (2008b) reveals the decreasing–increasing curve patterns of the ultrasonic velocity in avocados stored for 7 and 12 days at 20°C (two separate investigations). It has been speculated that the change of the velocity is associated with the changes of the biological properties of the avocado along the storage time that the ultrasonic velocity measurements in the avocado was conducted by using an angled-pointed transducer and the fruit are assessed only once in the middle of radial direction (different ultrasonic techniques and orientation of the measurements). Another study on the effect of elasticity of Baobab fruit on ultrasonic velocity reported by Phadke et al. (2012) shows that the velocity of less porous baobab is higher than more porous ones. It demonstrates that the ultrasonic velocity depends on the density and the properties of the material. Note: The study did not indicate the fruit maturity stage and it did not focus on the effect the orientation of the fruit measurements on ultrasonic velocity.

Secondly, the convergence of the axial-radial velocities can be explained by the textural changes of cell tissue structure of an apple during ripening due to senescence. A study by Tukey and Young (1942) discovers the changes of flesh parenchyma cells of '*McIntosh*' apples from rounded to elongated shapes in the radial direction during pre-harvest (growth and development), harvest (maturity) and postharvest (ripening). The shape of the cells is round and the size of intercellular air spaces is relatively small at pre-harvest. After that, the cell shape is increasingly elongated radially, and the intercellular air space size are grown bigger during storage. Therefore, the axial-radial ultrasonic velocity convergence in the *'Envy'* apples was suggested due to the cell structure changes have approached their optimum elongation period after postharvest (after week 8 of storage).

Thirdly, after week 8, the axial velocity became higher than the radial velocity. This finding was in line with the study of the axial velocity for *'Envy', 'Pink Lady'* and *'Royal' Gala'* apples in the preliminary experiment (Section 3.1.8.4). These samples were at the end of their ripening stage (from a supermarket: at the end of the fruit supply chain). This trend implied that the M and MM 'Envy' apples were also approaching their ripening stages after Week 8. These findings confirmed that the anisotropy of apple parenchyma and ripening stages during storage influenced the axial and radial velocity measurements.

3.2.1.2 Axial-radial velocities between the M and MM fruit stored at 4°C and 20°C

The velocities between the M and MM fruit were compared. Firstly, the axial and radial velocities of the MM fruit were found to be higher than the M fruit (Figure 48A to D). Secondly, the convergences of the axial-radial velocities of the MM fruit occurred faster before week 8, compared to the M fruit that occurred after week 8.

The followings are the discussions on the results. Firstly, the axial and radial velocities of the MM fruit were higher than the M fruit. These findings can be influenced by the different ripening stages between the two maturity groups. Studies conducted by Harker and Hallett (1992) and Ahmed and Labavitch (1980) that the MM cells are more affected by the dramatic changes of the internal properties: mixtures of solid and fluid, cell wall compositions, and conversions of starch to sugars. These changes of the internal properties possibly influenced the velocity measurements in apples.

Another study reported by (Contreras et al. 1992) shows a similar effect of sugar concentrations on the ultrasonic velocity. The finding reveals that the velocity increases linearly from 1480 to 1650 m/s with the increasing concentration of glucose, fructose and sucrose (pure sugars) at 20°C. This explains why the ultrasonic velocity of MM apples was higher than that of M apples in this experiment. The MM apples accumulated more sugars and these sugars influence the characterisation of the ultrasonic velocity, propagated through the cells due to the crystallisation effect of the soluble sugars.

Secondly, the convergence of the axial-radial ultrasonic velocities was faster in the MM apples than that in the M fruit. This finding implied that cell tissue structures for the MM apples were less compact due to less gaseous body, compared to the M apples. It was supported by the studies of (Reeve, 1970; Reeve, 1953; Tukey and Young, 1942) that shows the structures of the ripe apple parenchyma cells are less dense and the intercellular air spaces have been diminished (getting smaller), compared to those unripe cells.

Strikingly, these findings on the effect of ripeness in apple on the ultrasonic velocity measurements (due the biological property changes in the fruit parenchyma) well agreed with the study conducted by Povey (1997) on the changes of gas bodies in air-mixture. This agreement was based on the study of the effect of degasification in apple puree on ultrasonic velocity (demonstration of assessment on ripeness in apple) that will be presented and discussed in the next section.

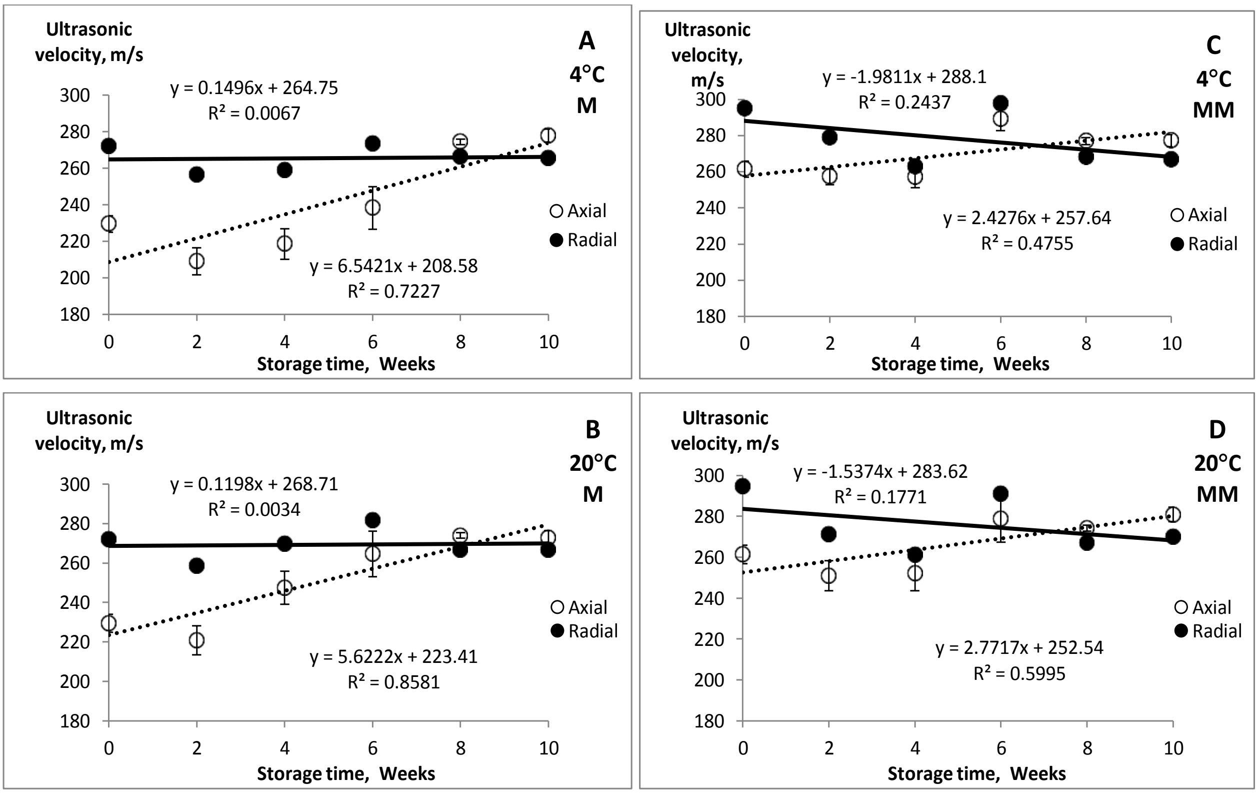


Figure 48: Effect of storage time, anisotropy and harvest maturity levels on ultrasonic velocity for 'Envy' apples during ripening of 10 weeks : (A) M and (B) MM apples stored at 4°C; and (C) M and (D) MM apples stored at 20°C (room temperature as a control group). Symbols represent mean values of ultrasonic velocity of axial (○) and radial (●) measurements. They were measured in every two weeks at room temperature (20°C) while the dashed (axial) and solid (radial) lines represent a least-square fit. Each point represents the mean value of 7 to 49 apples, with two measurements of each direction per fruit. Vertical lines represent the standard error bars of 95% confidence intervals (Some error bars cannot be observed because they are overlapping with the symbols) with *p*-value<0.05 (Mohd Shah et al., 2016).

3.2.2 Effect of degasification in apple puree on ultrasonic velocity (Demonstration of assessment on ripeness in apple)

Ultrasonic velocity was also measured based on the time of flight and the pulse-propagated distance through the degassed apple puree. The mean velocity showed an increasing trend from week 2 to 8 for the M and MM fruit stored at 4°C stored (Figure 49). The mean velocity varied from 1527.5 to 1543.3 m/s and 1520.8 to 1547.0 m/s for the M and MM puree respectively. The ultrasonic velocity in apple puree (around 1500 m/s) was nearly six times of the velocity in an intact apple (around 250 m/s).

These findings demonstrated the effect of gas bodies in apple on the ultrasonic velocity measurements and they were supported by Povey's study (1997), a theoretical prediction of the effect of volume fraction of air in water on the velocity of sound in air-water mixtures at 20°C (Figure 50). The internal changes in gas-liquid composition of the apple parenchyma cells during storage suggest a correlation with the changes in the volume fraction of air in water. The air in the fruit puree (■) was less (degasification) than that of the intact apple (□). The velocity increased as the decreasing of air during the eight weeks of storage suggested due to the ripening effect (senescence) that the gas bodies in the intact apple cell decreased as the fruit ripened. During the ripening, the cells have undergone changes from starch to sugar constituents that caused the cells to flood with more liquid and less gases. At the same time, the changes in the cell walls, and collapse of the middle lamella adjoining the neighbouring cells, also influenced the gas space volume. In addition, the findings of the increasing velocity with the increasing of the apple ripening also agreed with a study of Contreras and the research (1992) that shows the velocity in sugar solutions increases with increasing concentrations of glucose, fructose and sucrose from 1480 to 1650 m/s at 20°C. Studies by Povey (2000), Cartwright (1998) and Povey (1997) demonstrates the similar trend. The finding suggested that the velocity measurements could be an indicator of ripeness assessment in apples due to textural changes in the fruit.

Thus, these findings demonstrated that the velocity measurement can differentiate between the M and MM apples and the non-destructive ultrasonic testing techniques can assess the ripeness in apples during storage.

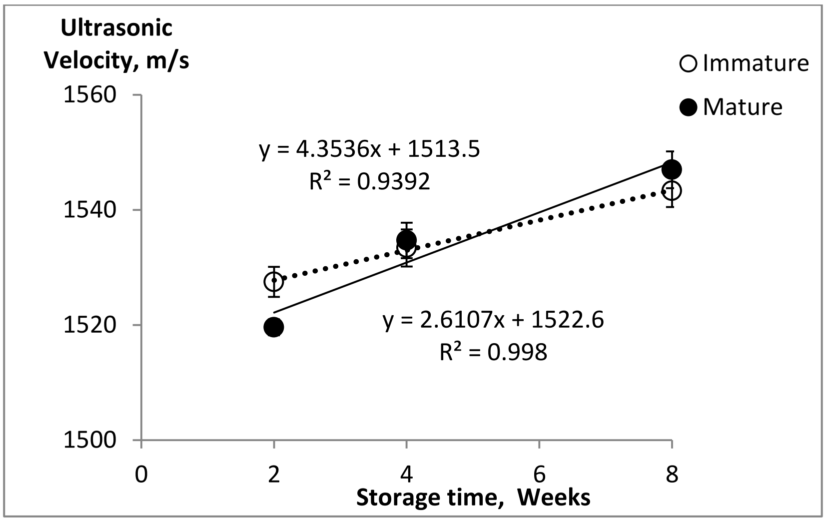


Figure 49: Mean ultrasonic velocities of the M (○) and MM (●) 'Envy' apples by using the Ultrasonic Velocity Meter test (UVM) for weeks 2 to 8 of storages at 4°C. Each point represents the mean value of three to seven apples respectively. The standard error bars of 95% confident intervals.

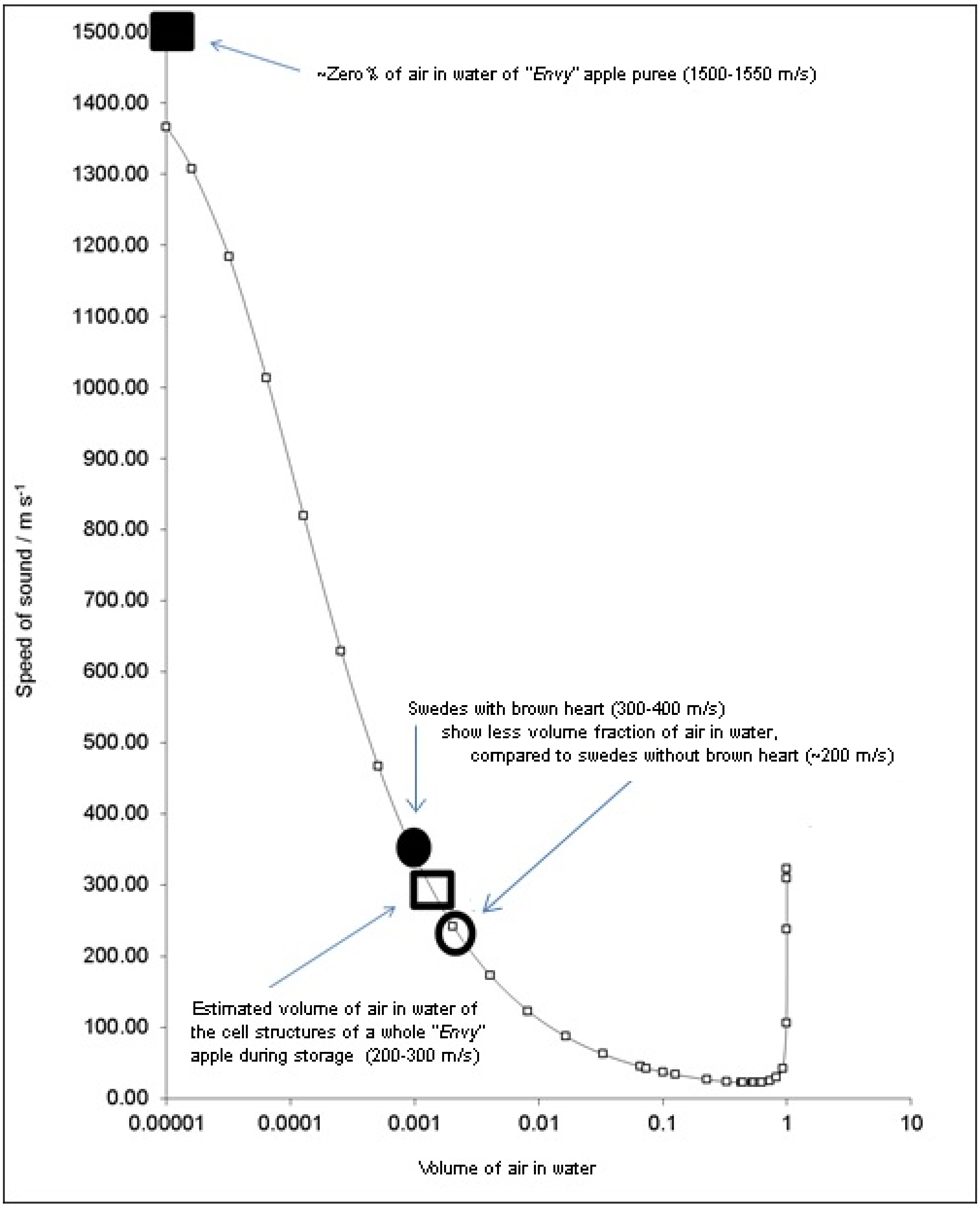


Figure 50: A theoretical prediction of effect of fraction by volume of air in water on velocity of sound in air-water mixtures at 20°C (From Povey 1997, with permission). The velocity in the apple puree (■) was almost six times higher than that of the intact apples (☐). The decreasing of % air as the velocity increased during the 10 week-storage as an indication that the parenchyma cells were undergone internal composition change due to ripening effect (senescence) (Mohd Shah et al., 2016).

3.2.3 Effect of anisotropy of apple parenchyma on insertion loss measurements of amplitude frequency signal.

The change in attenuation of a frequency signal was investigated by a study of insertion loss measurements of amplitude frequency signal. The insertion loss measurements were obtained by a value difference in the signal amplitudes between the frequency spectrum with and without an apple at approximately 5 kHz and 39 kHz (Figure 51).

The insertion loss around 5 kHz for the M group, (Figure 51A and C respectively) decreased more visibly compared with the MM group (Figure 51B and D respectively) (stored at 4°C and 20°C). Meanwhile, the decreasing trend at 39 kHz was more visible in the MM group (Figure 51G and H respectively) (stored at 4°C and 20°C). This trend implied that the insertion loss measurements correlated with the fruit ripeness during storage. Consequently, these findings suggested that the change in the attenuation measured by the insertion loss of the frequency signal amplitudes was influenced by the maturity levels during storage.

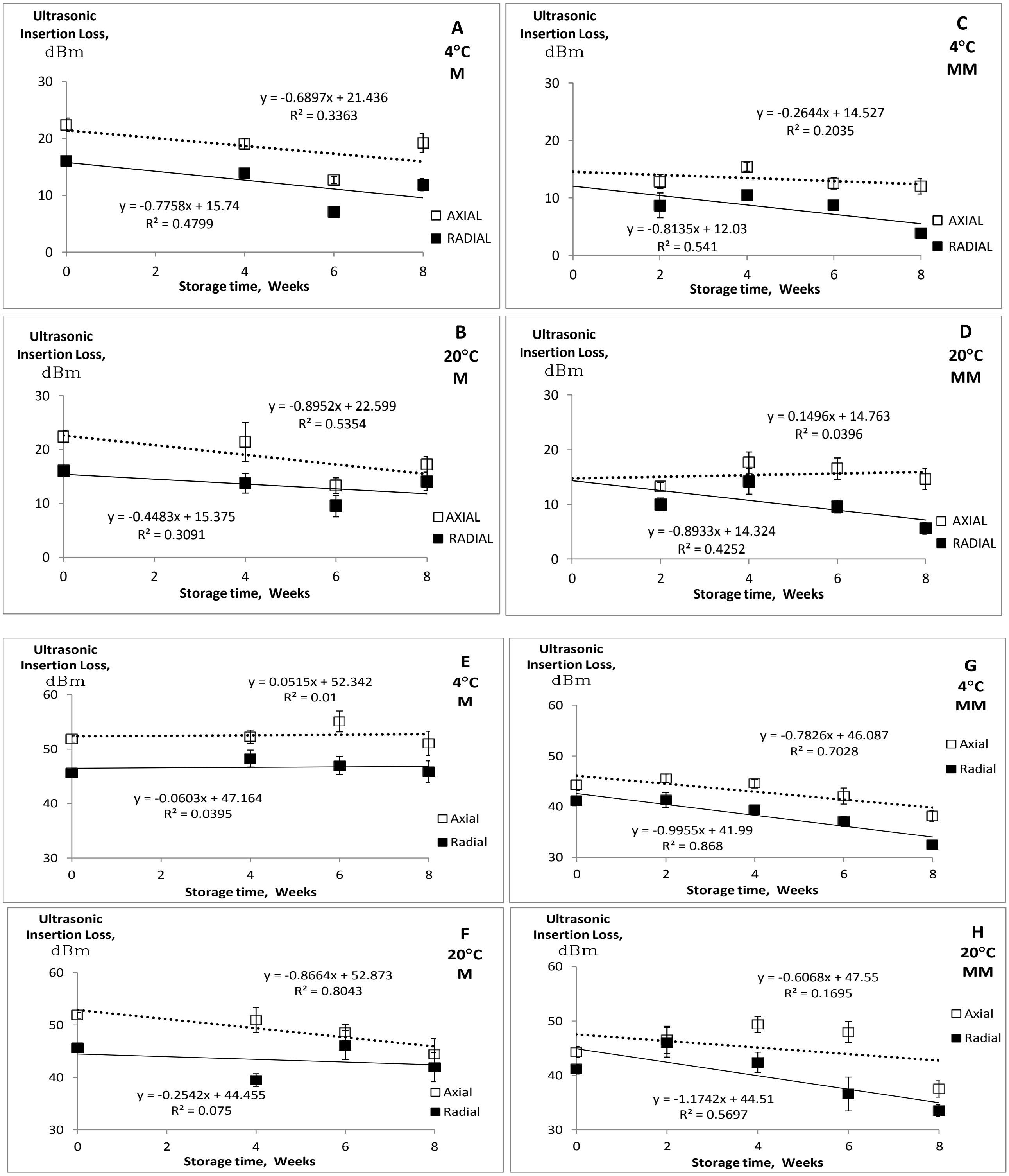


Figure 51: Effect of storage time, anisotropy and harvest maturity levels on insertion loss at 5 kHz (A – D) and 39 kHz (E – H) for 'Envy' apples during ripening of 8 weeks: M fruit stored in (A and E) 4°C and (B and F) 20°C; and MM fruit stored in 4°C (C and G) and 20°C (D and H) (20° was a control group). Symbols represent mean values of ultrasonic velocity of axial (☐) and radial (■) measurements. They were measured in every two weeks at ambient temperature while the dashed (axial) and solid (radial) lines represent a least-square fit. Each point represents the mean value of 7 to 49 apples, with two measurements of each direction per fruit. Vertical lines represent the standard error bars of 95% confidence intervals (Some error bars cannot be observed because they are overlapping with the symbols) with *p*-value<0.05 (Mohd Shah et al., 2016).

3.2.4 Correlation between ultrasonic velocity, firmness and sugar content

The ripeness in apples during storage was assessed by establishing a correlation between the changes in the velocity of sound and firmness as well as sugar content. The quality parameters were measured by using the PUNDIT Plus system, the compression, a puncture and sugar content tests respectively (axial measurements).

3.2.4.1 Ultrasonic velocity and firmness measured by a compression test

The ultrasonic velocity had a negatively strong relationship with the fruit firmness measured by a compression test during storage (Figure 62A). A linear regression shows a strong relationship between the two parameters, R^2=0.69 (N=7, n=2). Therefore, the correlation between the ultrasonic velocity and the fruit firmness was convincingly strong. The strength of the correlation suggested that the velocity measurement can be alternative techniques in evaluating the fruit ripening during storage. This finding demonstrated that the velocity measured by the PUNDIT Plus test can be used along with apple firmness by using a compression test.

3.2.4.2 Ultrasonic velocity and firmness by a puncture test

The ultrasonic velocity showed a negative relationship with the fruit firmness measured by a puncture test during the storage (Figure 62B). It indicated the velocity increased as the firmness decreased during storage. A linear regression also showed a strong relationship, R^2=0.52 (N=7, n=2). This finding was in agreement with other studies regarding a correlation between velocity and firmness by a puncture test of several fruit (Mizrach, 2011; Kim et al., 2009; Subedi and Walsh, 2009; Mizrach, 2007; Mizrach, 2000; Mizrach et al., 1999; Mizrach et al., 1999). This finding revealed that the velocity measured by the PUNDIT Plus test can be used together with apple firmness by using a puncture test.

3.2.4.3 Ultrasonic velocity and sugar content

The ultrasonic velocity showed no relationship with the fruit sugar content measured by a hand-held refractometer test during the storage (Figure 52C), R^2=0.003 (N=7, n=2). This finding suggested that the velocity measured by the PUNDIT Plus test cannot be correlated with the sugar content measured by the hand-held refractometer. No studies have been found on correlation between ultrasonic velocity and sugar content related to assessment of ripeness of the fruit.

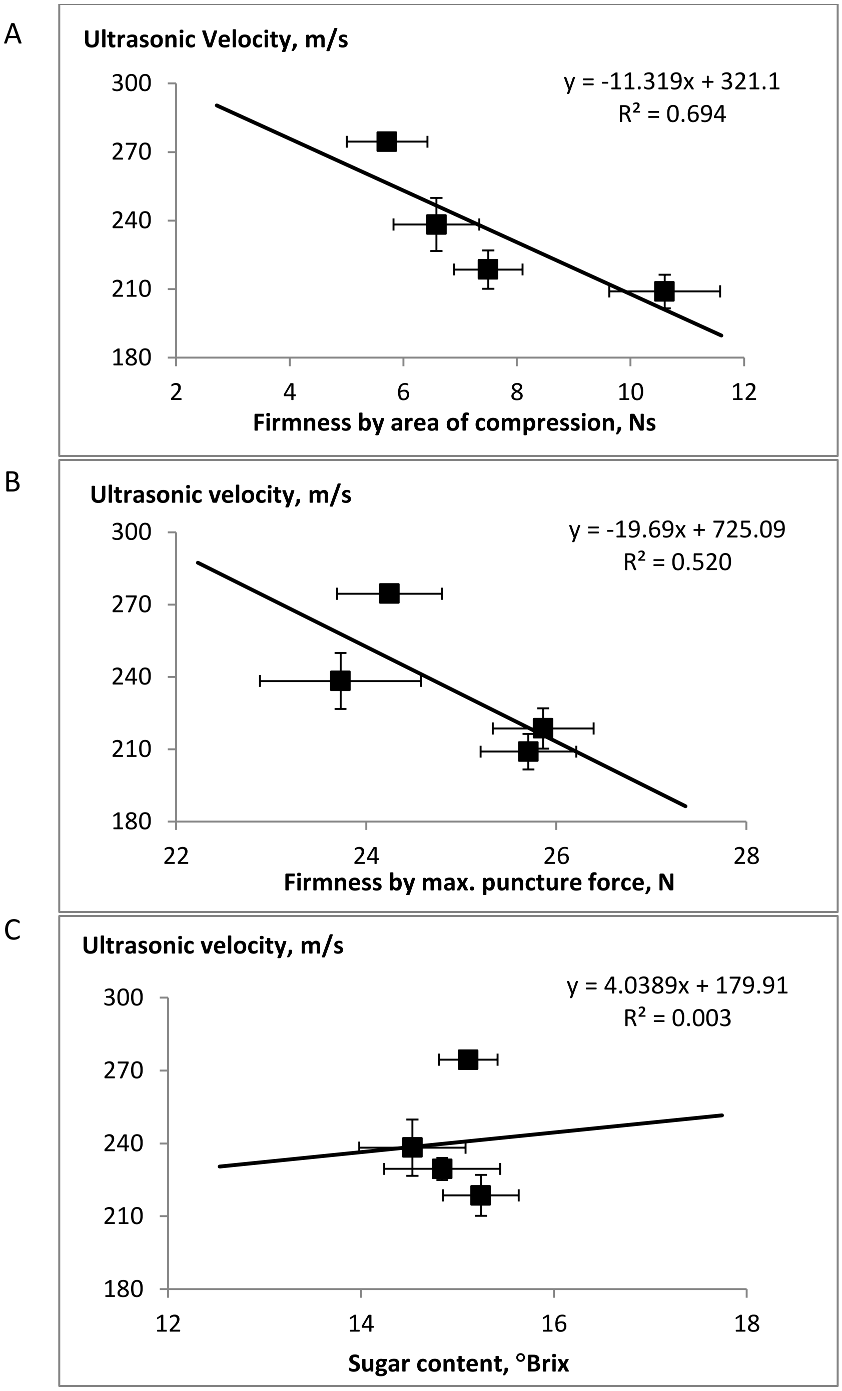


Figure 52: Correlations between the mean ultrasonic velocity using a PUNDIT Plus test and the mean firmness using a compression and puncture tests, and sugar content (axial measurements) during 8-week-storage of *'Envy'* apples. Vertical and horizontal lines represent the standard error bars of 95% confidence intervals with *p*-value < 0.05 (One of the error bars for the velocity cannot be observed because it was overlapping with its symbol). The data points are the average of seven apples with two measurements.

3.2.5 Principal Component Analysis (PCA): Correlation among the measured quality variables

The 24 quality measurement variables of the '*Envy*' apples were further analysed by using PCA of the correlation matrix to test if ripeness of apples can be discriminated based on (1) the storage durations, (2) the maturity levels and (3) the orientations of velocity measurements, a correlation between ripeness of the fruit and the storage monitoring quality measurements would be demonstrated.

3.2.5.1 Correlation between ripeness in apples and storage durations, maturity levels and anisotropy of velocity measurements

Ripeness and storage durations. The results showed that the discrimination of fruit ripeness based on the storage durations explained 55.9% of the total variance (Figure 53A). The value consisted of the first three principal components (PCs): PC1: 28.1%, PC2: 14.9% and PC3: 12.9%. These three PCs explained over half of the variability in the PCA data.

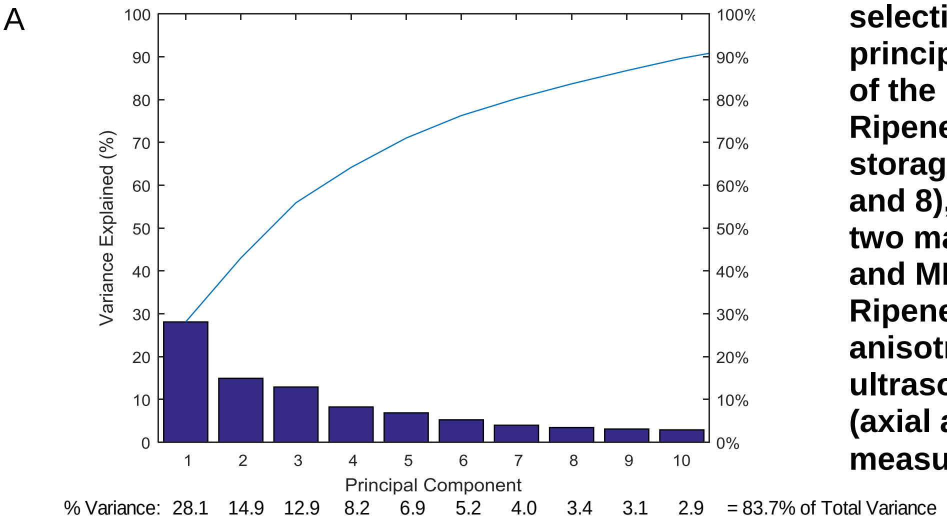
The samples projected on PC1 in a score plot (Figure 54A) were discriminated into four week storage groups (weeks 2, 4, 6 and 8) indicating the ripeness stages. Meanwhile, two groups of the data, labelled weeks 6 and 8, were clearly distinguished by PC2 confirming the prominent ripening stage occurred between these weeks. This discrimination demonstrated that the ripeness in apple was evidently indicated by the measured variables of the testing techniques in the experiment and had a correlation with the storage durations.

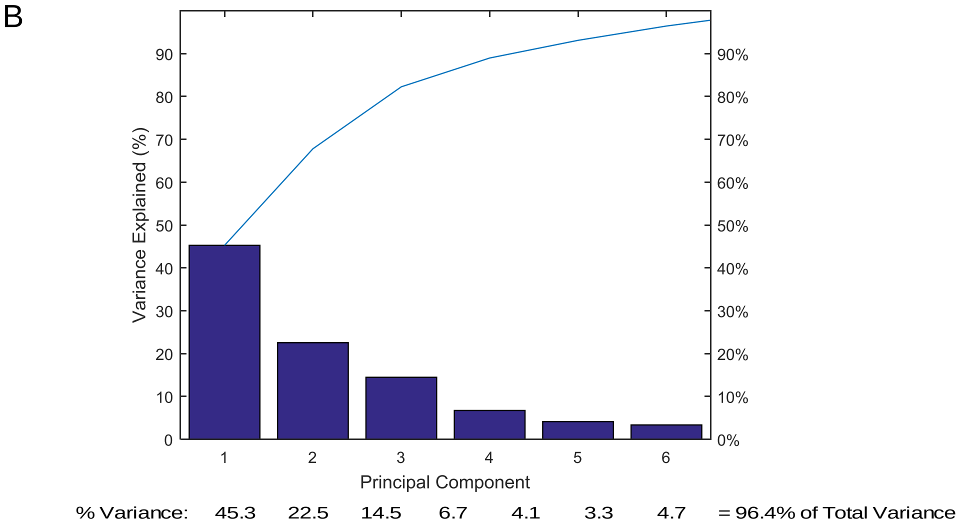
Ripeness and maturity levels. The data were also analysed to test if the maturity levels can be differentiated to indicate apple ripeness. The first three PCs explained 82.3% of the total variance (PC1: 45.3 %, PC2: 22.5% and PC3: 14.5%) (Figure 53B). This total percentage of variance represented over more than three quarters of the variability in the data associated with the maturity levels and ripeness of the fruit.

Subsequently, in Figure 54B, the samples were clearly divided into the M and MM groups. The two discriminations imply that the maturity levels can be associated with the ripeness in the apples. Once again, two clusters of samples for weeks 6 and 8 groups were clearly separated by PC2, indicating a visible change in the fruit ripeness. This segregation demonstrated that the ripeness in apple was also indicated by the the measured quality variables and correlated with the maturity levels.

Ripeness and orientations of velocity measurements. Feasibility of the ultrasonic technique in assessment of ripeness based on orientations of velocity measurements was explained by the first three PCs with 87.7% of the total variance (PC1: 46.6%, PC2: 24.5% and PC3: 16.6%) (Figure 53C). Again, these PCs described more than three quarters of the variability of the data, implying a high correlation between ripeness and the velocity measurements.

Next, the data in Figure 54C were segregated into two groups (axial and radial velocity measurements) along the PC1 axis. The data in the dotted circle (labelled for the fruit in week 8) refers to the convergence of the axial and radial velocities occurring between weeks 6 and 8. The discrimination between the axial and radial velocity measurements suggested that the ripeness in apples can be determined by the ultrasonic testing techniques.





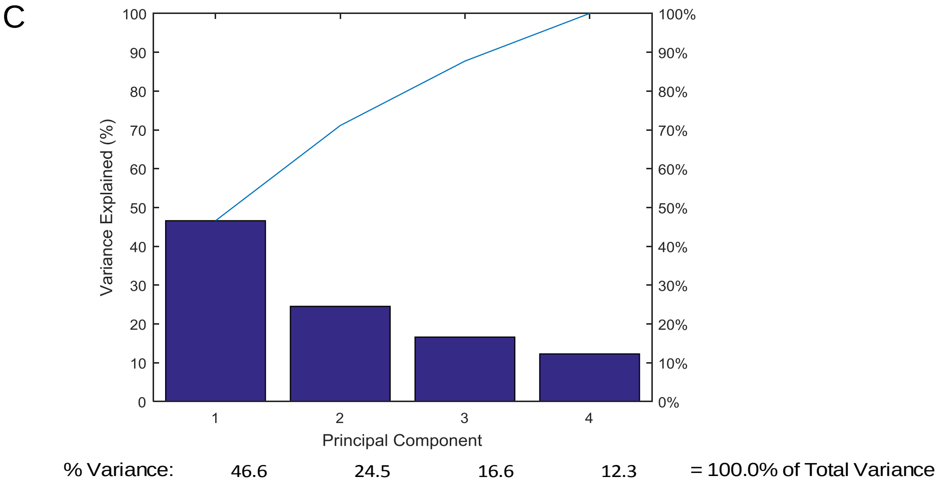


Figure 53: Scree plots for selecting numbers of principal components of the *'Envy'* apples: A. Ripeness and weeks of storage (weeks 2, 4, 6 and 8), B. Ripeness and two maturity levels (M and MM fruit); and C. Ripeness and anisotropy of apples on ultrasonic velocity (axial and radial measurements)

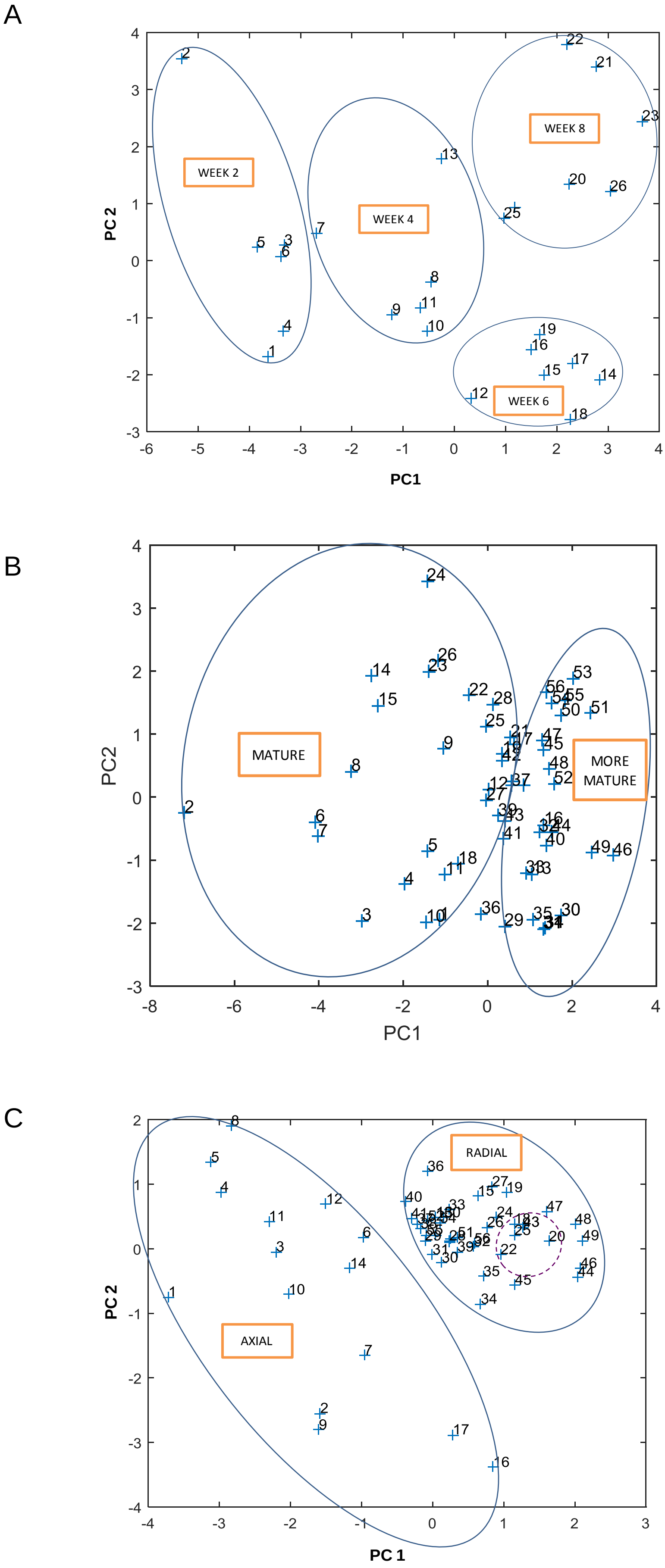


Figure 54: 2-D score plot for PC1 against PC2 of *'Envy'* apple experiment. A correlation between:

A. Ripeness and weeks of storage (weeks 2, 4, 6 and 8);

B. Ripeness and two maturity levels (M and MM fruit); and

C. Ripeness and anisotropy of apples on ultrasonic velocity (axial and radial measurements)

3.2.5.2 Discrimination among the measured quality variables

Ripeness and storage durations. The ultrasonic velocity was clearly discriminated in the negative region from the remaining variables in the positive region by PC1, in the 2-D loading plot for an association between the apple ripeness and weeks of storage (Figure 55A). The firmness measured by the puncture test and the sugar content were in the similar region on the plot. This trend revealed that the two quality variables gave similar information on ripeness in apples. The trend suggested that the either of these parameters could be excluded from the fruit quality measurements. This cluster was also found between the firmness by the compression test and the measurement of insertion loss of the frequency signal amplitudes. Thus, time and cost of assessment of ripeness in apples could be reduced, if some of measured quality variables are eliminated from the quality evaluation.

Concurrently, PC2 on the vertical axis distinguishes the axial ultrasonic velocities from the radial velocities in the positive and negative clusters respectively. The ultrasonic velocity was segregated from the other techniques and it was affected by the anisotropy of the fruit despite of the remaining measurements that were less influenced by the anisotropy. This finding demonstrated the use of non–destructive ultrasonic velocity testing as an alternative technique in assessing the ripeness of apples.

Ripeness and maturity levels. The discrimination of the ultrasonic velocity from the rest of the tests on PC1 implied that this quality measurement had a strong relationship with the fruit ripeness. Therefore, the ultrasonic velocity can be used to assess fruit ripeness (Figure 55B). In addition, the clear segregation between the axial and radial velocities on PC2 suggested that anisotropy of apple parenchyma influenced the velocity measurements. This was in agreement with the results (Section 3.2.1). Hence, the velocity measurements corresponding to the axial-radial measurements were feasible alternative to destructive testing in assessment of ripeness in apples.

PC2 demonstrates that firmness measured by the compression test can be isolated from the rest of the measured variables. This indicated that the compression test was adequate in determining fruit ripeness. Subsequently, firmness measured by the puncture test and sugar content by the Brix test cannot be distinguished in the evaluation of apple ripeness. These findings implied that the quality assessments from either of these two tests were sufficient to determine apple ripeness. Hence, the first two PCs reveal that the ripening in apples associated with their maturity levels.

Ripeness and orientations of velocity measurements. The velocities for the M and MM fruit were discriminated into two different regions on PC1 and PC2, based on the anisotropy of velocity measurements (Figure 55C). Thus, the axial-radial ultrasonic measurements distinguished the ripeness in apples during storage.

In conclusion, the findings of Part 1 of the project confirmed the correlation between ripeness and apple storage quality changes, measured by using ultrasonic propagation (velocity), mechanical/ physical (firmness) and flavour (sugar content) parameters and these parameters as alternative indicators for assessment of ripeness in apples.

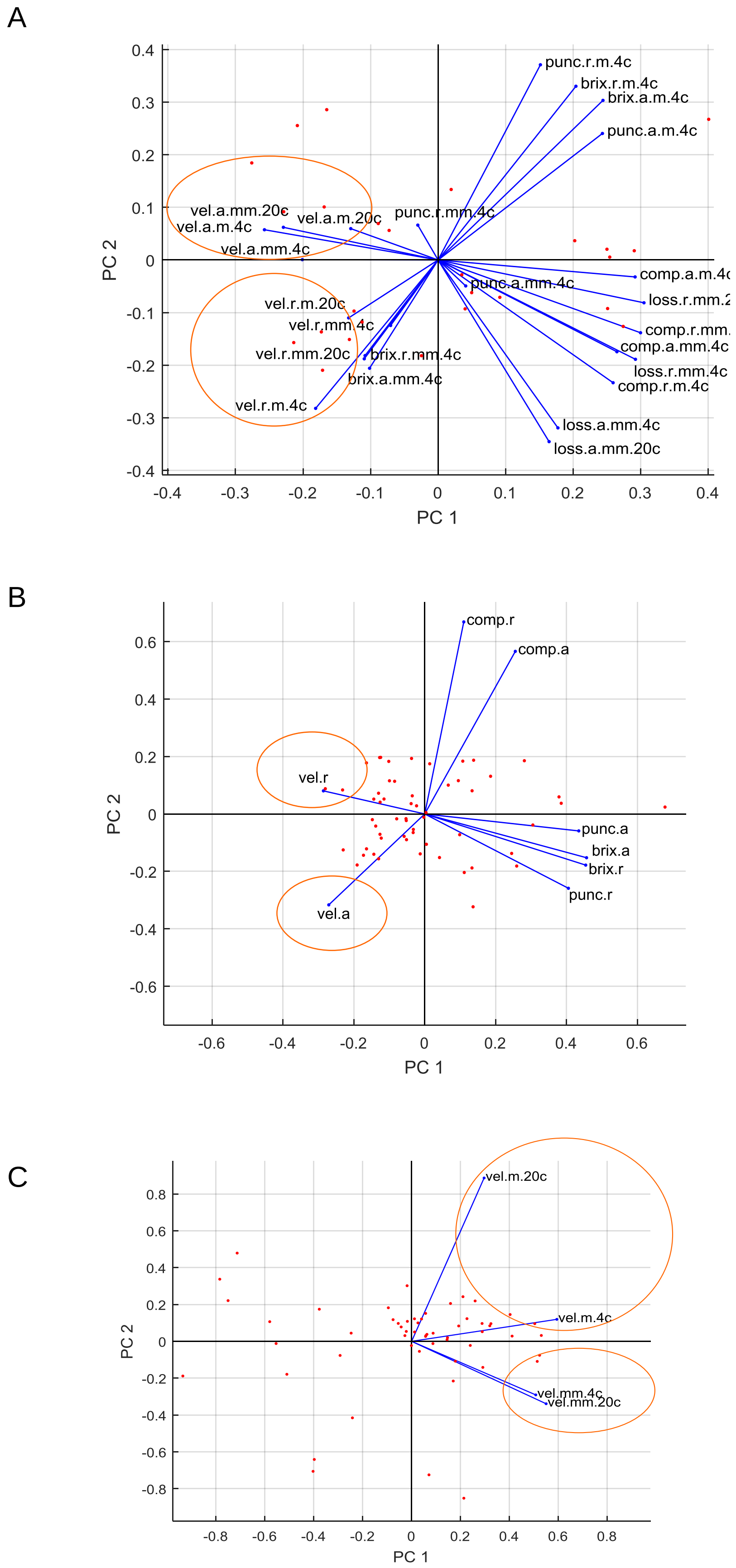


Figure 55: 2-D loading plot for PC1 against PC2 of *'Envy'* apple experiment. Discrimination among the 24 measured variables between: A. Ripeness and weeks of storage (Weeks 2, 4, 6 and 8); B. Ripeness and two maturity levels (M and MM fruit); and C. Ripeness and anisotropy of apples on ultrasonic velocity (axial and radial measurements)

3.2.6 Schematic overview of the findings of assessment of ripeness in *‘Envy’* apples experiment

A schematic overview of the findings of this study (assessment of ripeness in ‘Envy’ apples experiment) (Figure 56) illustrates the integrated approach of the relationship between an ultrasonic measurement (velocity) and physical property (firmness) mediated by biological properties (cell compositions and cell wall properties).

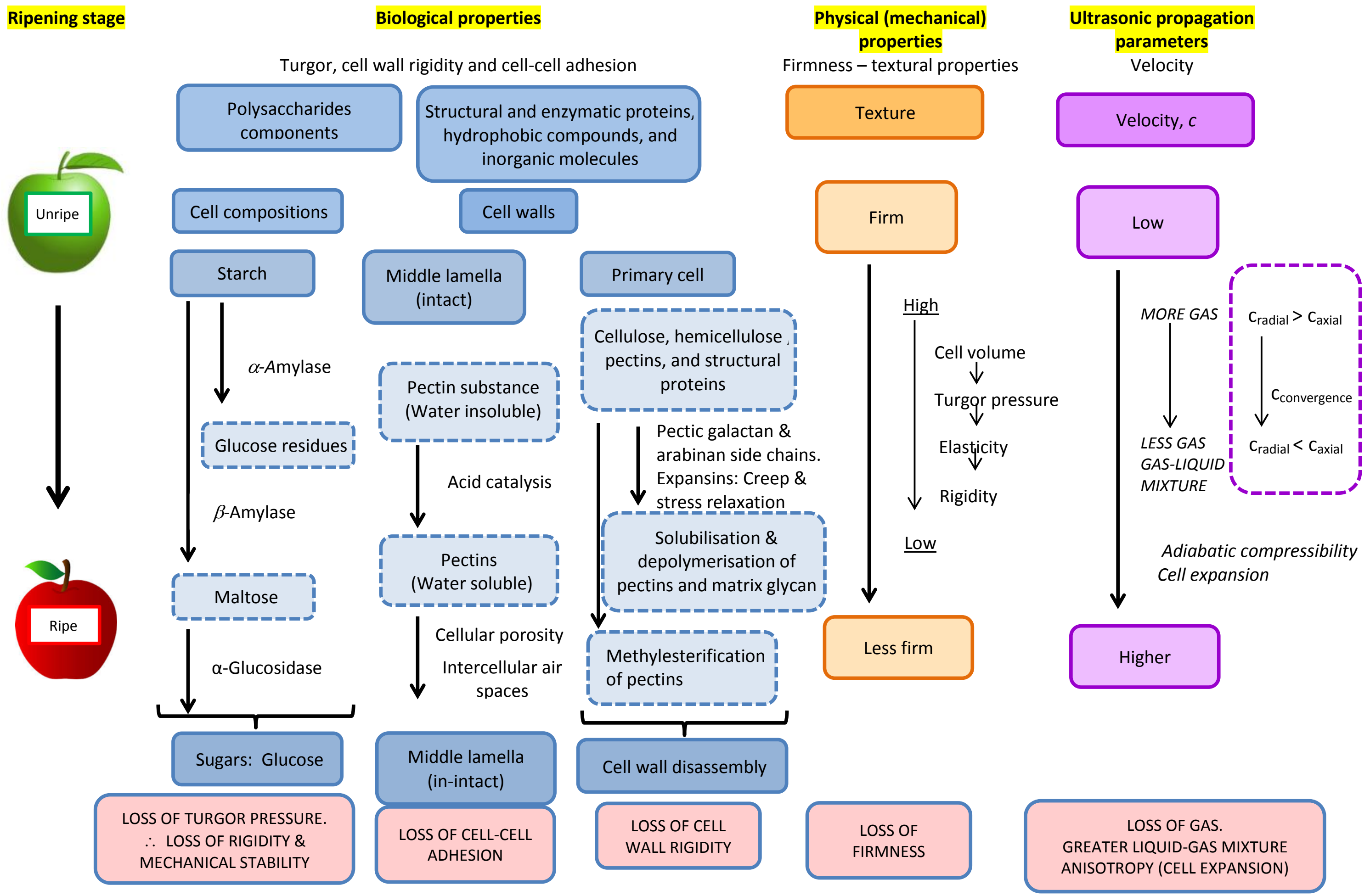


Figure 56: A schematic overview of the findings of assessment of ripeness in ‘Envy’ apples experiments: the integrated approach of the relationship between an ultrasonic measurement (velocity) and physical property (firmness) mediated by biological properties (cell compositions and cell wall properties).

3.2.7 Effect of apple anisotropy on firmness and sugar content

3.2.7.1 Firmness measured by a compression test

Ripeness of the apples was determined by the firmness. It was measured by the compression test based on the work of area under the load-formation curve. This data represented the elasticity of the apples that was associated with the fruit ripening of fruit. Thus, preliminarily, a study was conducted to confirm that the compression test of the apple experiment was in the elasticity region of the curve. The result showed that the curve (Figure 57) of force against the displacement of the flesh of the apple (with the skin) was linear (R^2=0.997). This linear curve confirmed that of the compression test in this research was conducted in the elasticity region

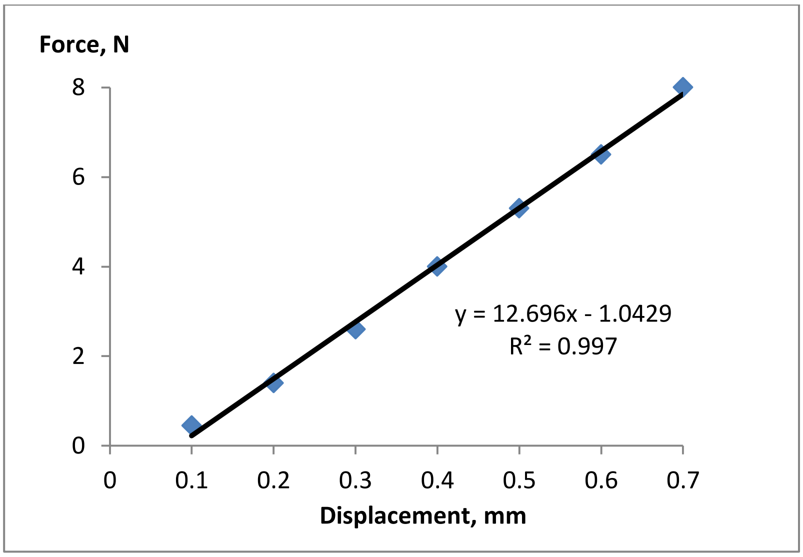


Figure 57: Linear curve of the force N against the displacement mm of the flesh of the apple (with the skin) confirming the working elasticity region of the compression test

The fruit firmness based on the axial-radial compression measurements showed the decreasing trend of firmness during storage. For the M fruit, the mean value of the work of compression decreased from 10.6 to 5.7 Ns and from 11.1 to 6.6 Ns for the axial and radial measurements respectively (Figure 58 (1) A and B). Meanwhile, for the MM fruit, the mean values decreased from 8.05 to 4.4 Ns and from 10.8 to 5.2 Ns for the axial and radial measurements respectively. The compression testing also revealed that the effect of anisotropy was more prominent in the MM group significantly at weeks 2 and 4 compared to the M fruit.

The decrease in the firmness with storage duration can be explained by the changes in turgor level over the cell volumes based on the following studies. Rose et al. (2003), Taiz and Zeiger (2002), Atwell et al. (1999) and Tyree and Jarvis (1982) highlights that this change due to the decrease in rigidity of the apple cell walls. The cell wall rigidity is dependent on the mechanical properties and it represents the elasticity of the fruit flesh. This elasticity is related to the change in turgor pressure levels over the change of cell volumes. During ripening, softening of the cell tissues is a result of the degradation of the middle lamella during ripening. Moreover, the changes in enzyme-hydrolysis in the cell walls also contributes to changes in the tissue cells (leading to the collapse of the walls and generate the softening of the fruit). Therefore, the loss of firmness in the fruit can be due to the loss of the cells mechanical supports due to middle lamella, intercellular air space, and cell wall degradations. Furthermore, Tyree and Jarvis (1982) states that the tissue cells consist of a mixture of solid and liquid as well as gas (viscoelastic properties). This viscoelastic property is correlated to the turgor pressure of the cell walls. Their studies show that the elasticity of the cell wall is decreased proportionally to the decrease in relative cell volume. The decreasing trend in the firmness agreed with the interrelationship between the volume and the turgor of the fruit cells indicating of the softening of the fruit flesh of the Tyree and Jarvis' finding.

3.2.7.2 Firmness measured by a puncture test

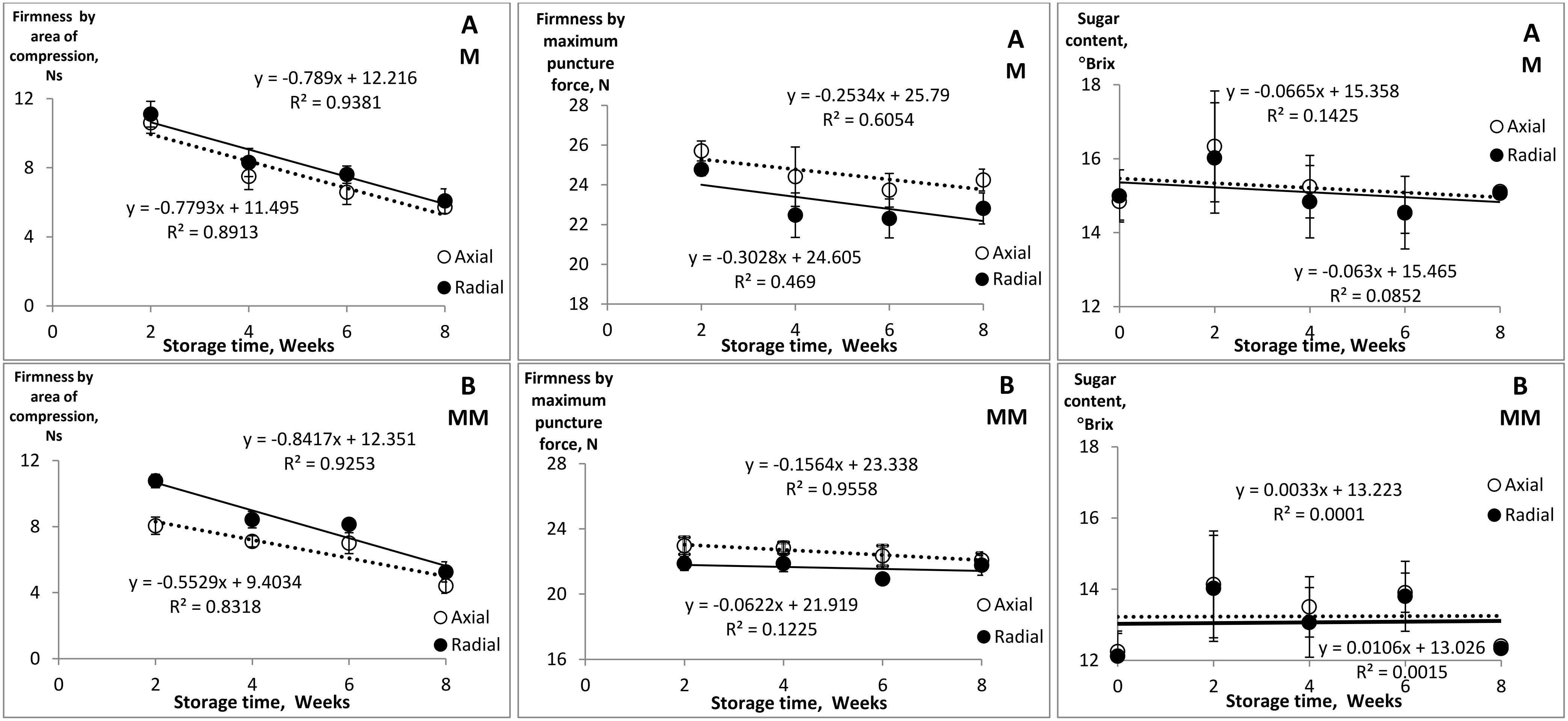
Firmness (an indicator for the ripening of the apples) was measured by the maximum puncture force. For the M fruit, the mean firmness of the axial and radial measurements varied from 25.7 to 23.7 N and 25.1 to 22.1 N respectively (Figure 58 (2) A and B). For MM fruit, the mean firmness of the axial and radial measurement was in the ranges of 22.8 to 22.1 N and 21.9 to 20.9 N respectively. A puncture test showed that the firmness of the axial measurement was higher than that of the radial measurement at week 2 for both maturity groups. In addition, the M fruit were firmer, compared to the MM fruit for the axial measurements at week 2, $p < 0.05$.

The finding at week 2 was consistent with the other measurement techniques (different values of the axial and radial firmness in the same fruit). Khan and Vincent (1993a) report that the axial force is 40% higher than the radial force in four types of apples: Bramley Cox, Gloster, Norfolk

Beefing, and Rock Pippin due to the axial and radial position of cellular structures in the parenchyma. The apple cell tissues elongate radially like columns in which intracellular air spaces are between them. Hence, when the fruit flesh is fractured radially, the penetration goes through the columns parallel to the intracellular air spaces. Conversely, when the flesh is fractured axially, the penetration must break through these column structures perpendicularly, meaning more force is required. As a result, the fruit flesh is fractured more easily from the radial direction compared to the axial direction.

3.2.7.3 Effect of apple anisotropy on sugar content.

The sugar content was measured based on the °Brix values. The change of °Brix values was an indicator of the change in ripening of the fruit along the axial and radial measurements. Not unexpectedly, the Brix test was insensitive to the effect of anisotropy on the sugar content for both M and MM apples during storage, $p < 0.05$ (Figure 58 (3) A and B). For the M fruit, the ranges of variation of the mean sugar content was 14.8 – 16.4% and 14.5 – 16.0% for the axial and radial measurements. Meanwhile, for the MM fruit, the mean sugar contents of the axial and radial measurements were in the range of 12.3 – 14.1% and 12.1 – 13.9% for the axial and radial measurements.



(1) Firmness by a compression test

(2) Firmness by a puncture test

(3) Sugar level by a Brix test

Figure 58: Effect of anisotropy and maturity levels on firmness by using a compression and punture tests, and sugar content for 'Envy' apples during ripening durations of 8 weeks: (A) M and (B) MM fruit stored at 4°C. Symbols represent mean values of ultrasonic velocity of axial (○) and radial (●) measurements. They were measured every two weeks at ambient temperature while the dashed (axial) and solid (radial) lines represent a least-square fit. Each point represents the mean value of five to seven apples, with two measurements of each direction per fruit. Vertical lines represent the standard error bars of 95% confidence intervals with *p*-value < 0.05.

3.2.8 Calculation of estimations of the uncertainty of measurement

The measured variables involved in the '*Envy*' apple experiment were presented and the expanded uncertainty U_e of the measurement of the tests were calculated and reported with 95% confidence limit in Table 18. The values were based on the computation of the combined uncertainty U_c coming from the uncertainty contributions, u.

The U_e of the measurements of the tests were obtained as the followings:

1. PUNDIT Plus Transducers test : 0.4 m/s
2. Compression test : 0.6 N
3. Puncture test : 0.2 N
4. Sugar content test : 0.2°Brix
5. Ultrasonic Velocity test : 1.0 m/s

Table 18: Calculation of the estimation of the measurements of uncertainty for an '*Envy*' apple experiment

Test	Type of test	Non-destructive (NDT) / Destructive test (DT)	Instrument used	Variables			Sources of uncertainty			Combined Uncertainty U_c	Expanded Uncertainty U_e
				Measured parameters Measured property		Measured property	Accuracy *u(A)* Verification of the instrument for fit for use	Precision *u(P)* Repeatability/ Reproducibility	Instrument Uncertainty *u(I)* Manufacturers' specifications		
1	PUNDIT Plus Transducers test	NDT	PUNDIT Transducers	Time of flight, ultrasonic path	Anisotropy of an apple, Axial and Radial measurement	Ultrasonic velocity,	Verified against sound velocity in air	All procedures were repeated for every two weeks for the same apples	PUNDIT: 0.1 µs Oscilloscope: 1 % Thermometer: 2°C Humidity: 3.5 % Vanier calliper:0.2 mm	0.2	0.4
2	Compression test	NDT	Texture Analyser (TA)	Area under curve	Anisotropy of an apple, Axial and Radial measurement	Firmness (skin and flesh elasticity)	Verified at 5.000kg using Reference Standard weight	All procedures were repeated for every two weeks for the same apples	Force: 0.025 %	0.3	0.6
3	Puncture test	DT	Texture Analyser (TA)	Maximum puncture force	Anisotropy of an apple, Axial and Radial measurement	Flesh firmness	Verified at 5.000kg using Reference Standard weight	All procedures were repeated for every two weeks for the same apples	Force: 0.025 %	0.1	0.2
4	Sugar level content or Total Soluble Solid test	DT	Hand Refracto-meter	Sugar content level	Anisotropy of an apple, Axial and Radial measurement	Sugar level content	Verified with distilled water and 5%of sucrose readings	All procedures were repeated for every two weeks for the same apples	Refractometer: 0.2°Brix	0.1	0.2
5	Ultrasonic Velocity test	DT	Ultrasonic Velocity Meter (UVM)	Ultrasonic velocity	Apple puree	Ultrasonic velocity	Verified with an external digital thermometer	All procedures were repeated for every two weeks for the same apples	UVM: 1m/s Temperature: 0.2°C	0.5	1.0

3.3 PART 3: Detection of BH in Swedes

3.3.1 Laboratory-developed BHI

A set of ten laboratory-developed BHI swede pictures was prepared to standardize the grading in BH measurements (Figure 59). Severity of BH was categorised from 0 to 9 (the least to the most severe BH, based on the affected size and colour).

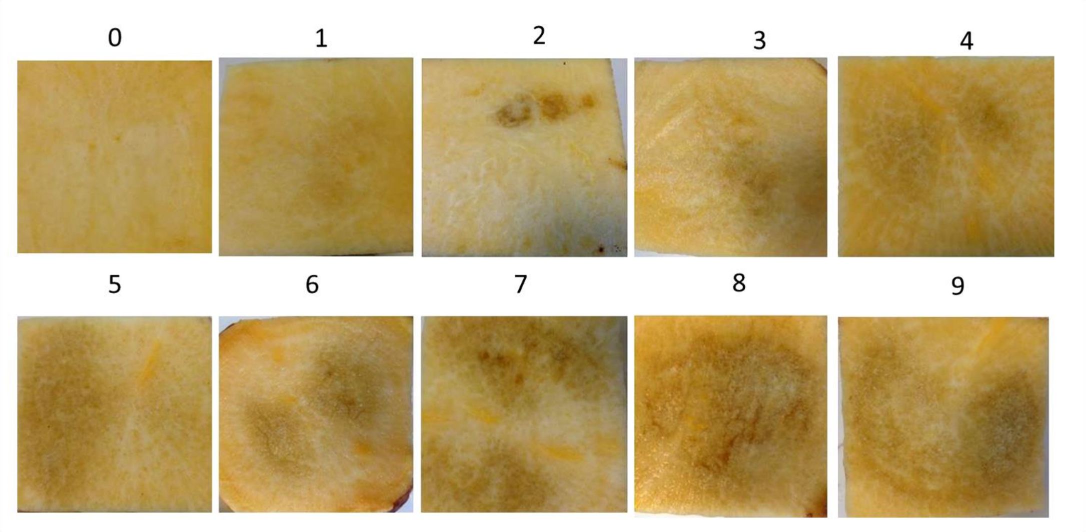


Figure 59: A characteristic of BH conditions in swede in a radial cut section. The scales from 0 to 9 (the least to the most severe BH). The swedes in the photos were about 50mm of cross cut section.

3.3.2 Determination of the BH conditions by a visual inspection

The BH determination of the 20 swedes (B1-B20) was based on the developed BHI. Different categories from 0 to 9 were found in 20 defective BH vegetables (Figure 60). The BH category is shown in red in the figure.

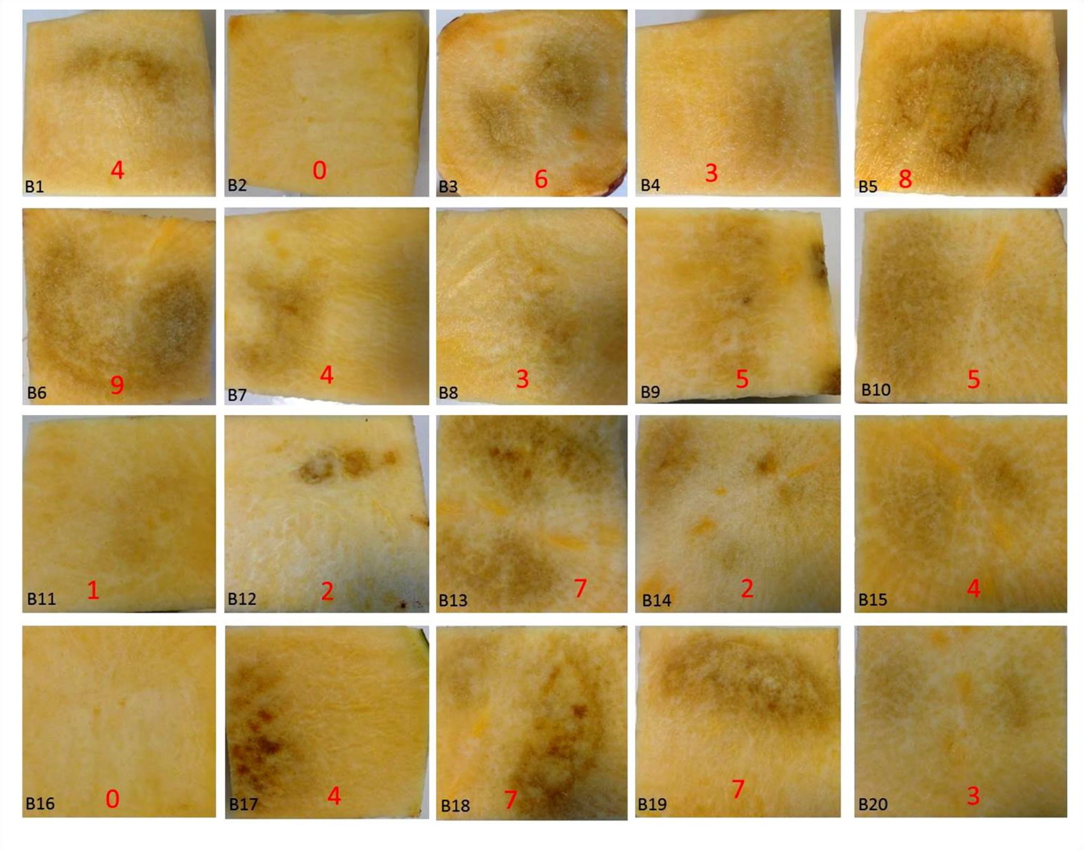


Figure 60: Visual inspection of the BH symptom of 20 swedes, based on the laboratory-developed BHI. The swedes in the photos were about 50mm of cross cut section.

3.3.3 Correlation of BH detection by using the BHI (visual grading) and PUNDIT Plus transducer system measurements

BH determination of the 39 swedes (B1-B39) was conducted based on the laboratory-developed BHI. Different categories from 0 to 9 were found in the 39 defective BH swedes and the categories were recorded in red in the middle of each figure (Figure 61).

The ultrasonic velocity was also measured in another 33 swedes to determine the correlation in BH between the visual grading and ultrasonic PUNDIT Plus transducer system measurement. The results showed that the velocity measurement positively correlated with swede BH degree (Figure 62) with a regression coefficient for a linear fit R^2 of 0.67 and Pearson correlation coefficient of 0.82, $p > 0.05$. The ten independent healthy samples had a mean value of 235 m/s and standard deviation of 17 m/s.

Overall, there was a strong, positive correlation between the measured velocity and the BHI in the swede samples. Increases in BHI correlated with positive increase in the ultrasonic velocity. This correlation confirmed on the effect of BH on ultrasonic velocity in swede. Thus, the study demonstrated that the ultrasonic technique offered an alternative, non-destructive measurement in detection of BH in swede.

Volume fraction of air in water in swede. The effect of BH on velocity was explained by the changes of the air fraction in the swede. The association between ultrasonic velocity and volume of air in water was shown in the vegetable (Figure 50) (Povey, 1997). Higher velocity was found in BH-detected swede compared to the healthier swede in the previously discussion. This observation was theoretically due to the changing air content in the internal vegetable cell tissues. Normal healthy cell membranes and intercellular spaces contain volume fraction of air, about 2%.

The gas-bodies in the defective BH decrease due to the changes in the cell compositions, intercellular airspace, and cell wall disassembly. A study conducted by Dermott and Trinder (1947) also shows that the defective BH parenchyma cell structures are cumulatively flooded with water. This environment affects the characteristics of ultrasound velocity, and indeed, it can be seen in the fraction by volume of air in water on velocity of sound in air-water mixtures at 20°C (Figure 50) (Povey, 1997). Based on the graph, the velocity in BH swede had air in water showing around 0.1%. In contrast, the velocity of healthy swedes had air in water showing higher than 0.1%. These results indicate that velocity was higher when BH was detected in swede, due to the air-water mixture level changes within the cellular structures.

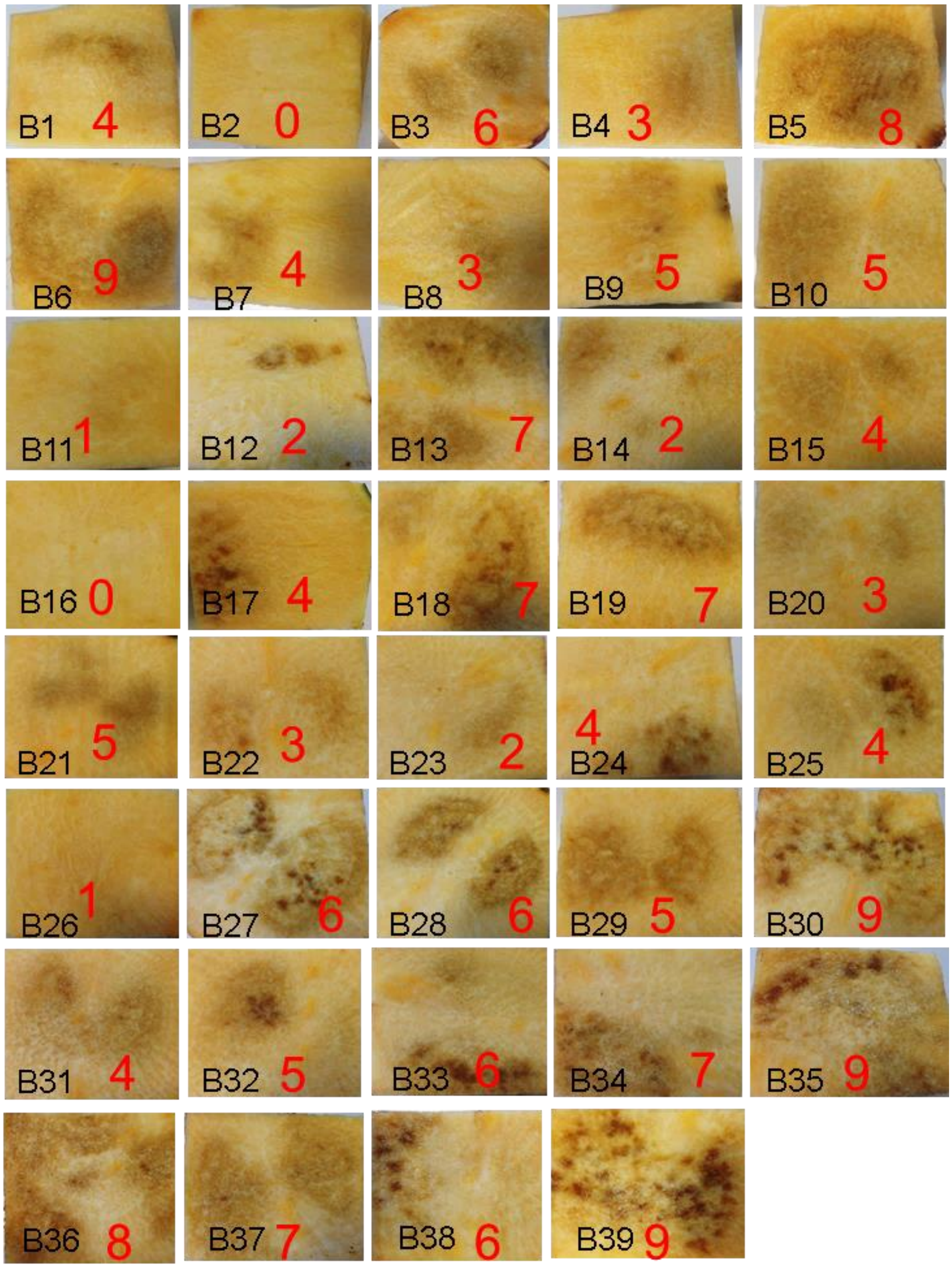


Figure 61: Examples of the visual inspection of BH symptom in measured swede based on the laboratory-developed BHI (B1-B39 swedes). The value in red is the assigned BH level, from 0 to 9 scale.

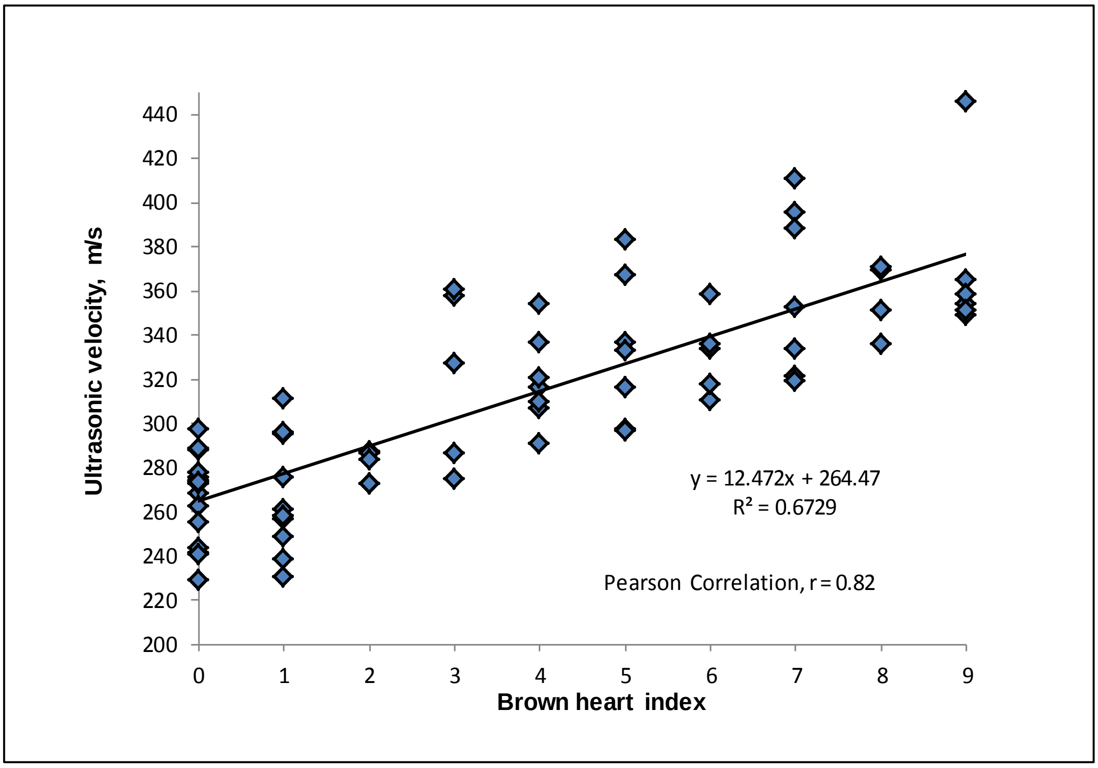


Figure 62: A strong positive relationship between ultrasound velocity measurements and the severe BH in swede for *n* = 72 swedes. Each point represents the average of four repeat measurements.

3.3.4 Quality control chart of BH based on the velocity: Discrimination between defective BH and healthy swede

A quality control chart of ultrasound velocity was developed, based on the seven healthy swedes and the 39-suspected BH samples (Figure 63). The chart confirmed that the velocity measurement within the swedes detected and discriminated the defective BH from the healthy swedes in the sample set. The values of mean, 1 standard deviation (1SD) and 2 standard deviations (2SD) of the velocities measured from the ten healthy swedes were represented by long and short-dashed and dotted lines respectively. The velocity in the defective BH swede was higher compared with the healthy swedes. Only four out of the 39 BH swede (10%) fell into the 2SD quality control chart ranges for healthy swede. This finding showed that the non-destructive PUNDIT Plus test can detect BH in swede with 90% of the reproducibility and repeatability.

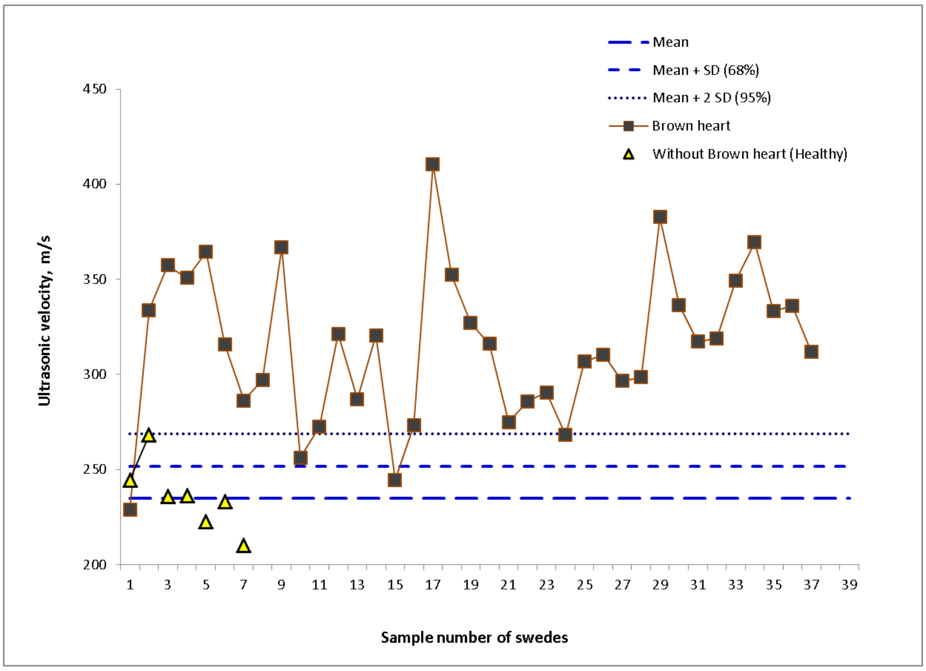


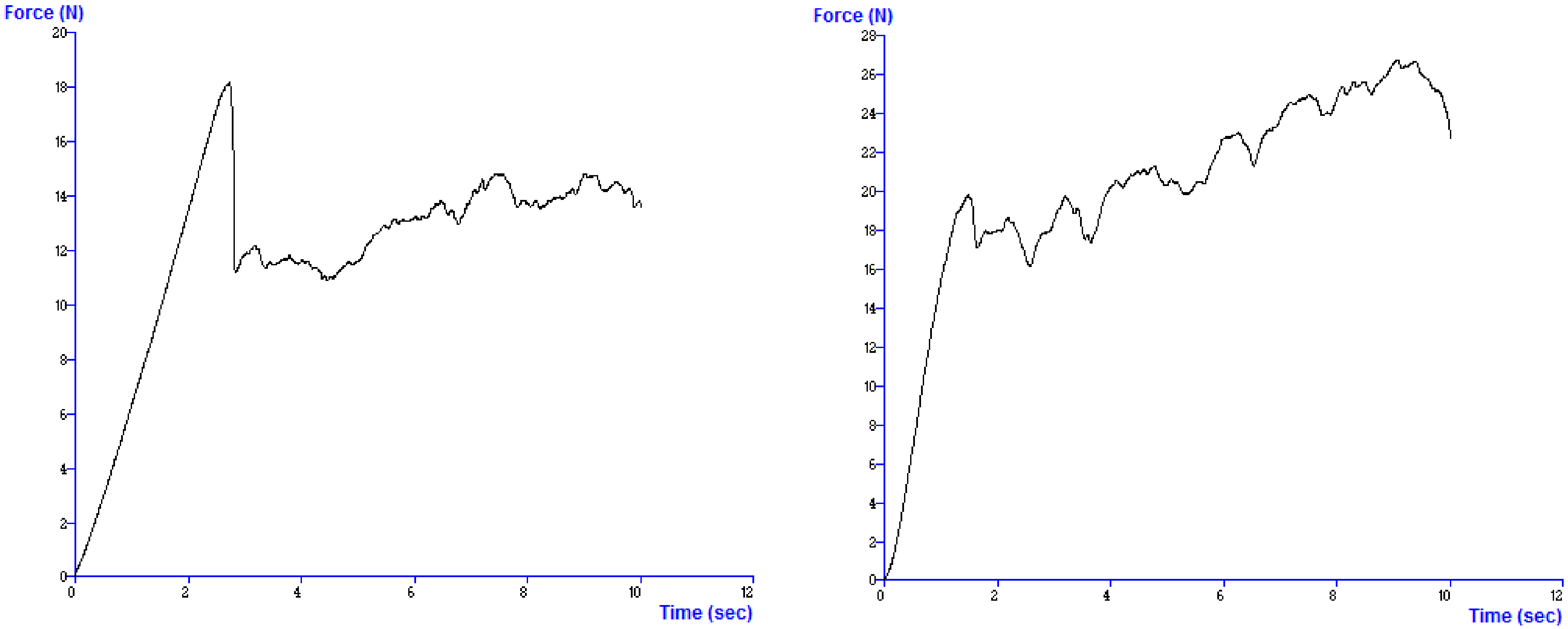
Figure 63: The ultrasonic velocity in BH swedes (■) and the healthy swedes (△). The quality control chart for the velocities in healthy swedes was represented by the long dashed line (———) for the mean value, short dashed line (- - - - -) for the mean value + the 1 Standard Deviation (1SD) and dotted lines (··················) for the mean value + the 2 Standard Deviations (2SD). Four points of the detective BH Swede fell within the quality control chart.

3.3.5 Texture Profile Analysis (TPA)

TPA curves. The force-time curve in the puncture test showed different trends between BH and healthy swedes (Figure 64). The maximum force in the healthy swede curve (A), represented the vegetable firmness, increased sharply, called as a '*bio-yield point*' (the first fracture of the vegetable flesh during the puncture test) and it diminished dramatically after the point. Conversely, the force in the BH swede curve was less distinct compared to the healthy swede. In fact, the force after the first peak increased as time increased. The observation referred to the visual assessment and no statistical data was conducted on the samples.

The healthy and BH curves were similar to the force-time curves discussed in the theory of the puncture test in Bourne (2002). The textural changes in fresh vegetables such as softening and crispiness are due to the pectin degradation and moisture and turgor losses. These textural changes lead to the decreasing in flesh elasticity. The demonstration of the elasticity differences in the cell tissues of the vegetables were observed in the two TPA force-time curves. The cell tissue of the healthy swede was more compact and contained less fluid, contributing to the harder cells to be compressed and fractured compared to the defective BH swede. This finding was in line with Dermott and Trinder (1947), indicating that the cell tissues in the BH swede are flooded with a mixture of gas-water causing a more fluid environment. The more fluid in the medium, the less compressible it will be. This characteristic can be traced from the BH curve, where the force consistently rises even after the first puncture peak.

Correlation between velocity and firmness in swede. Despite of the different curve between the BH and healthy swedes from the TPA, no direct relationship was found between the velocity and firmness (by the puncture test) of BH in swede, $p < 0.05$ (Figure 65). A linear regression showed a week association between the two variables ($R^2 = 0.06$). The possible explanation of this finding was due to the limitation of assumption of correlation between textural properties and physical properties of food. Bourne (2002) stressed that numbers of textural properties represent food physical properties. However, it is not necessary that all physical property changes can be correlated to the textural properties.



A: Healthy swede

B: BH swede

Figure 64: Texture Profile Analysis (TPA) force-time curves, A: Healthy swede; B: BH swede, of a puncture test from a texture analyser for swede samples at pre-speed: 2.00 mm/s, test-speed: 1.00 mm/s and post-speed: 10.00 mm/s

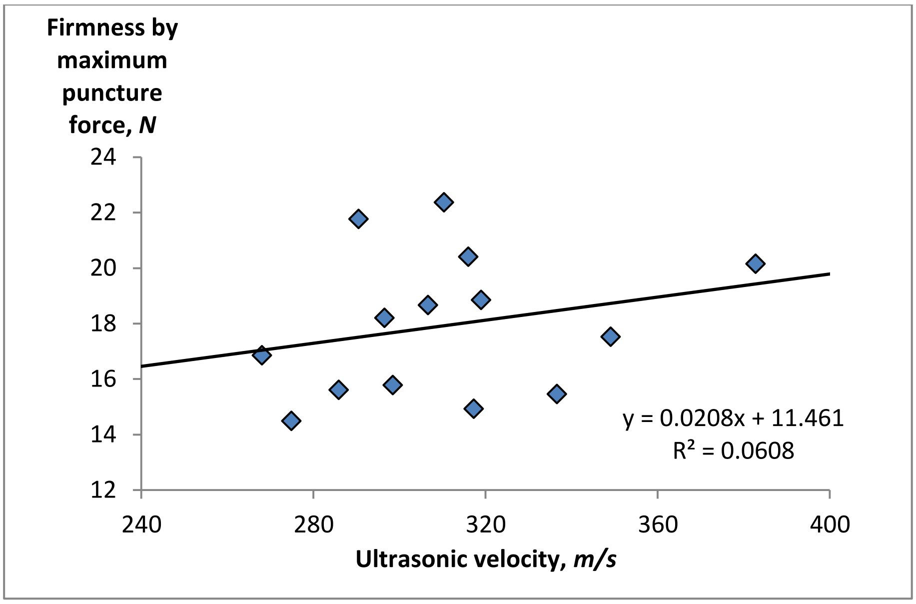


Figure 65: Relationship between firmness by a puncture test and ultrasonic velocity in swede (*N*=14)

3.4 Confirmation of Research Questions (RQ) and Research Hypothesis (RH)

The findings from the apple and swede experiments confirmed the RQ and RH as follows:

1. The PUNDIT Plus Transducer system measurement had been characterised and optimised. The results showed that the ultrasonic measurement technique is fit-for-purpose for the experiments in this research.
2. Changes in ripeness and (2) anisotropy in apples during storage showed an effect on the velocity of sound (with firmness and sugar content measurements). There was a correlation between velocity measurements and the firmness of the fruit. Velocity measurements were also influenced by the anisotropy of the fruit cells.
3. BH in swede showed an effect on the velocity of sound measurements and firmness through the Texture Profile Analysis (TPA). There was a correlation between the BH and ultrasonic velocity in swede.

It can be concluded that the changes in ripeness in apples and internal flesh quality of swedes influenced the ultrasonic velocity measurements (together with firmness and sugar content). Therefore, the ultrasonic technique can be utilised as a feasible alternative to non-destructive testing due to its offer of useful data for interpretations of the apple ripening during storage and the detection of BH in swedes.

Chapter 4
Conclusions

4.1 Summary of the thesis

Chapter 1 discussed on the current challenging scenario associated with the quality of the fruit and vegetables and the preliminary discussion raised the statement of the problem. Based the substantiated literature reviews, the research scope was determined (Figure 7): (1) food quality, (2) food quality attributes and costumers' perception on quality, (3) textural quality attributes for fruit and vegetables,(4) instrumental destructive and non-destructive techniques of evaluation of quality of fruit and vegetables, and (5) ultrasonic techniques specifically. The research aimed to investigate the utilisation of ultrasonic technique measurements in assessments of ripeness in apples and detection of brown heart in swedes, together firmness, and sugar content tests.

Chapter 2 elaborated the materials and the method development of the ultrasonic techniques, compression, puncture, and sugar content testing for data collection in answering the research questions (based on the problem statement and literature reviews in Chapter 1). It comprised three parts of the investigations: PART 1: Characterisation and optimisation: Method development of the PUNDIT Plus System measurement; PART 2: 'Envy' apples: Assessment of ripening and anisotropy in apples; and PART 3: Detection of Brown Heart.

Chapter 3 presented and discussed about the finding of the data in Chapter 2. It dedicated answers to the research questions and confirmation on the hypothesis of the research.

4.1.1 Assessment of ripening and anisotropy in apples

Firmness is one of the most important quality attribute indicators of fresh fruit and vegetables perceived by customers. It is challenging to minimize the quality degradation of the produce due to their high perishable products. Most of the instruments measuring the firmness utilise destructive techniques. Some non-destructive measurement techniques are implemented by food industry and ultrasonic measurements are one of the alternatives. However, studies on the feasibility of ultrasonic techniques to measure the textural properties of the fresh fruit and vegetables have not well explored and documented.

This experiment of the study was carried out by using (1) a PUNDIT Plus System testing to measure the ultrasonic velocity of sound through an apple, (2) a texture analyser to measure the fruit firmness through compression and puncture tests, and (3) a hand-held refractometer to measure sugar content.

The aim of this part of the study was (1) to assess the ripeness of ‘Envy’ apples during the eight-weeks-storage by using ultrasonic measurement techniques (velocity of sound), together with firmness and sugar content measurements, and (2) to establish a correlation between the ultrasonic velocity, firmness, and sugar content.

The study was to answer the following research questions: *How do (1) the changes in ripeness and (2) anisotropy in apples during storage affect the ultrasonic velocity, together with firmness and sugar content measurements, and (3) the correlation among those quality measurements using the different testing?*

The ultrasonic measurement techniques by a PUNDIT Plus System was characterised and optimised and the method was fit for purpose to measure ultrasonic velocity of sound through apples.

The study reported six major findings. Firstly, anisotropy on apple cell tissues affects the ultrasonic propagation velocity as a function of the fruit ripeness during storage. Secondly, the changes of the velocity of the axial measurements increased more dramatically, compared to the velocity of the radial measurements during storage (the radial measurements were higher than the axial measurements, yet the changes of the measurements were rather stable). Thirdly, convergence of the axial and radial measurements occurred after eight-weeks-storage. Fourthly, the velocity was higher in ripening apples compare to unripe fruit that agreed with the findings in other research. The changes of air-liquid volume fraction in apple cells influenced the velocity measurements during storage. The changes of the air-liquid volume fraction were associated with the changes of cell compositions (conversion of starch to sugar) and structures (degradation of primary cell walls and middle lamella). Fifthly, the ultrasonic velocity had a strong negative correlation with the firmness via the compression and puncture tests, and a weak positive correlation with the sugar content via the hand-held refractometer test. Sixth, the PCA discriminated the ripeness in apples based on ultrasonic velocity measurements, maturity levels, and storage durations (with the first three PCs explained 87.7%, 82.3% and 55.9% of the total variance respectively of the total variance explained). Based on these findings, the research questions for the second part of the research had been answered: The ultrasound velocity was influenced by maturity levels, ripening stages and directions of measurements (anisotropy) of apples during storage.

4.1.2 Detection of Brown Heart in swede

Brown heart (BH) is an internal defect in swedes that possibly due to Boron deficiency. The BH can only be assessed prior to the vegetable harvest time by using a visual inspection at orchards. Detection of BH will cause the produce unable to be marketed and the swedes in the defective area must be destroyed. As a result, the BH detection in swede causes farmers loss their income, postharvest losses and waste, and possible shortage of supplies.

Moreover, the detection of this disorder is challenging due to its random spatial variation patterns, sizes, and shapes and the measurements of detection of BH in swede are conducted destructively (Swedes are cut opened and visually inspected) on considerably small number of samples. Consequently, interpretation of the data from this destructive assessment is less representative for the actual distribution of the population. Alternatively, ultrasonic measurement techniques offer non-destructive, simple, fast, and economical methods for the detection of BH in swede.

The work involved the PUNDIT Plus System testing and the texture analyser (a puncture test) used in the apple experiment to measure the ultrasonic velocity of sound through a swede and firmness respectively. A laboratory developed BH Index was also set up for a visual inspection consisting 10 scales of the severity of the defect. A correlation between the ultrasonic velocity and the severity of the defective BH (by the visual inspection) was established. In addition, the Texture Profile Analysis (TPA) curves for BH and healthy (non-defective BH) swedes were compared.

The aim of the swede study was (1) to detect BH in swede by using ultrasonic measurement techniques (velocity of sound), together with firmness measurements, and (2) to establish a correlation between the ultrasonic velocity and a visual inspected severity of BH in swedes (practised by swede industries).

The study was to answer the following research question: *How does BH in swede affect the velocity of sound and firmness measurements?*

The ultrasonic measurement techniques by a PUNDIT Plus System was characterised and optimised and the method was fit for purpose to measure ultrasonic velocity of sound through swedes.

The study reported three major findings. Firstly, ultrasonic velocity measurements were correlated with the severity of BH in swedes possibly due to the change air-water mixtures in the parenchyma cells. Secondly, ultrasonic velocity measurements also discriminated the defective BH from the healthy swedes. Thirdly, the discrimination of the cells between the two swede groups was supported by the dissimilar appearances on their TPA curves. Based on these results, the research questions for the third part of the research had been answered: The ultrasound velocity varied according to the degree of severity of BH in swedes.

4.2 Practical applications and importance of the research for science and industrial community

The major findings in this research suggested that the ultrasonic measurement technique could be utilised as a fast, economical, non-destructive online sensor to determine the maturity index and ripening stage of apples through a comparison of the axial and radial velocities and to detect the internal disorder-the brown heart linked to boron deficiency in swedes. This research suggested that ultrasonic measurement techniques could be feasible alternative non-destructive testing for quality evaluation of fruit and vegetables.

This research can provide meaningful statistical data for stakeholders in the food industry and they can gain benefits through understanding the trend of quality variation for the fresh fruit and vegetables after the postharvest (measurements using non-destructive ultrasonic measurement techniques).

Farmers, packinghouses, and exporters. Farmers, packing houses and/or exporters would benefit from this research because the quality variation level of the fresh produce can be better understood. The information on the quality changes of ripening can be essential in determination of the shelf life of fresh produce across different batches with confidence (batch variability issue can be entertained). When the handlers realize on the level of the quality variation, they would implement appropriate quality control programs at specific points such as curing time, grading tables, post-treatment, packaging and labelling steps in their packing houses. This estimation of the shelf life is important in arranging destinations for the local and global markets. Note: The ultrasonic techniques can also be utilised in determination of harvest time of fruit and vegetables based on maturity levels of produce. The determination by using this non-destructive technique possibly offers more representative data due repeatability of measurements. By carefully selecting the right time for harvest, prediction of keeping quality of batches of fresh fruits and vegetables at the beginning of postharvest would be improved and higher chance of the intended fruit and vegetables to reach global markets. This chance means higher output and income for farmers and other food handlers.

A discussion on applying the experiment into an online system (actual scale) in a pack-house was initiated for future collaboration among AHDB, Ultrasound group of The University of Leeds and engineers (a companies).

The methodology of the research is also aimed to be applied on other suitable fruit and vegetables (such as durian when I return to Malaysia).

Importers, distributers, and wholesalers. These food handlers would be appreciate and realize their important roles as stakeholders in the midstream of the food supply chain in continuously aiming to maintain the product quality reaching their destinations. The quality of the produce can continuously monitored if necessary at this transaction point, to estimate the quality deterioration levels, by using the ultrasonic measurements.

Retailers. In addition to the significance of this research, retailers who are at the downstream of the food supply chain could become better actors due to the better understanding towards the expectation of quality variation among the fruit and vegetables batches arriving at their supermarkets. Therefore, they would invest time and effort to adapt the best conditions to maintain the quality of the produce based on the estimated shelf life before delivering the goods to consumers.

Consumers. At the end, the product quality meets consumers’ expectation. They get the product within acceptable shelf life with high quality and perhaps the loyalty to the product would be continued.

Agriculture extension officers. Apart from the stakeholders in the food industry, extension programs can be initiated to concentrate on improving areas where the non-destructive measurements are applicable, such as cultivations, global competitive varieties, and farm managements as well as postharvest issues. Hence, collaborations between the government and the food industry can be strengthened (technological and managerial perspectives).

Researchers (Institutes/ Universities). The outcome of this research can extend the techniques and uses of the non-destructive ultrasonic measurements and the statistical data on the evaluation of fresh fruit and vegetable quality. A software program can be developed for a prediction of keeping quality of batches of the produces (at specific transaction points at a specific interval along the food supply chain to predict the range of variation levels).

4.3 Strengths and limitations

Strengths. The followings evaluate the strengths of the research. The research met the designated time blocks/activities of its schedule. The statement of the problem was substantiated with facts and models from credible literature. The three investigations in this research provided further perspective of non-destructive ultrasound techniques in evaluation of fruit and vegetables, due to the limited access and study. Communicating with the actual apple and swede industries had given a chance to get insight of the actual practice of handling the quality of fruit and vegetables.

Limitations: Despite these strengths, this research possessed some limitations.

(1) Limitations of the method

Sample size: The sample size was considerably small (7 to 47 apples for each measurement) due to the capacity and availability of the fruit allocation for the two-months-measurements during the postharvest time. The statistical analysis can be improved so that the significant correlations from the experimental data can be more representative for the population distribution, if larger sample size is used for each measurement.

Frequency of the measurements: The measurements were conducted for every two weeks in the apple experiment due to the constraint of the sample size. The curve trend in the quality parameter plots can be improved, if the frequency of the measurements is increased. Therefore, the quality changes during storage can be better understood.

Literature on the topic: Reviews on the effect of anisotropy of the cell tissues on the ultrasonic propagation parameters were limited due to few studies on this topic. Most studies utilise destructive techniques in measuring firmness of fruit and vegetables. Moreover, limited studies explore ultrasonic techniques on evaluation of quality of fruit and vegetables (and limited samples were investigated). Studies on a correlation between ultrasonic propagation parameters and the textural properties of the fresh produce based on the anisotropy effect of the cell structures have not well documented. Therefore, most the literature in the research was based on the combination and availability of references (studies).

Methods in data collection: Firstly, only five measurement techniques used in this research (a combination of ultrasonic velocity measurements via a PUNDIT Plus system and Ultrasonic Velocity Meter, UVM; firmness via a compression and puncture tests, and sugar content). These measurements covered techniques for ultrasonic propagation parameters and physical properties of apples. A microscopy test (changes in biological properties) was not be included in this study due to the time constraints of the three years-awarded scholarship. If the microscopy test was conducted, it will complete the integrated approach of the conceptual framework of the research. Secondly, the apple investigation was conducted only at the beginning of postharvest (early section of the food supply chain). No quality monitoring was carried out at the rest of transaction points on the supply chain due to the time constraints and common practice by food industry. Therefore, the experimental data of the research focussed in-sight understanding on the quality changes of fresh apples at the beginning of the postharvest shelf life (at the beginning of the supply chain).

(2) Limitations of the researcher

Access: Longer amount of time had been spent on these following activities at the problem definition phase: (1) understanding the problem statement by using the selected literature, (2) designing the integrated approach in investigating the ripening of apples based on the ultrasonic propagation parameter, physical (mechanical) and biological properties of fresh fruit and vegetables; and (3) developing the respective methods to their optimum performance. As a result, less time had been focussed on the data analysis phase for learning, analysing and interpreting the experimental data using new software (MATLAB and OriginPro). This limitation may result the collected data were not analysed potentially from other angles (such as advanced statistical data analysis).

Longitudinal effects: The Government of Malaysia sponsored only three years for the research. Therefore, all the experimental designs, method development, data collection, analysis, and interpretations, writing of manuscripts of papers and thesis were designed to be completed within these three years.

4.4 Recommendations for future research

The research target was to test if non-destructive ultrasonic technique can offer as an alternative to evaluate the quality of fruit and vegetables on –line together with other tests. The followings are recommended for areas of future research.

Holistic quality monitoring throughout the fruit and vegetable supply chain. The investigation can be expanded to the next transaction points along the food supply chain to learn levels of quality degradation of the produce from the harvest time until reaching their consumers. Ideally, this investigation can minimise the quality loss and forecast the shelf life of the produce holistically.

Other fruits and vegetables (different cultivars, varieties). Ultrasonic propagation waves travel uniquely through a medium. Investigations on other fruit and vegetables can be conducted to understand their ripening and internal quality behaviours and to obtain their own statistical data.

Multiple non-destructive and destructive measurements. The data of the trend on variation of quality attributes of interest using multiple destructive and non-destructive instruments may also beneficial to investigate the correlation among these techniques in assessing the quality of fresh fruit and vegetables (Some measurements can be eliminated from the quality assessment, such as if the PCA results are not significant.)

Method developments of other ultrasound techniques. Airborne ultrasound techniques (acoustic microscopy) can be developed to assess the ripening and internal quality of the fresh fruit and vegetables. This non-contact and non-destructive technique utilises sound waves to study the structure and composition of the tested produce.

Consumer behaviours towards quality of fresh fruit and vegetables. It would be beneficial to investigate a relationship between the measurements by the designated instruments and consumer behaviours towards quality of fresh fruit and vegetables.

Multi-disciplinary researches. Studies on a combination of quality, marketability, and economic feasibility on fresh fruit and vegetables can also be considered as a continuation of this research.

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Appendices

Appendix 1: Terminology of texture in other languages

Standardization on the terminology of the textural properties is important in understanding consumers' expectation towards food quality. Most consumers express their quality perception towards textural properties of food through languages. However, this understanding is restricted. The studies reveal that different consumers from different countries refer the vocabulary of texture differently due to their locations, cultures, and origin of languages (Kilcast 2004; Antmann et al. 2011; Chen 2007; Varela et al. 2013).

Table 19 is a summary of different terminologies used by people from the designated countries to describe the textural properties: French, Japanese, Chinese, Finnish, Spanish, and Vietnamese. The details of these terminologies are given in the following appendices. The terminology used in French based on mechanical, flow, surface/ contact, structure and appearance/ shape properties in Appendix 2 (Nishihari et al. 2008). Japanese and Chinese expressions for crispness are displayed in Appendix 2 and Appendix 4 respectively (Szczesniak 1988). The terms related to textural properties in Japanese language are listed in Appendix 3 (Nishihari et al. 2008). 30 fundamental terms and a variety for textural properties in Chinese language are presented in Appendix 5 and Appendix 6 (Nishihari et al. 2008). Meanwhile, Appendix 7 is a list of visual appearances and texture attributes mentioned in France and Vietnam languages (Blancher et al. 2007). The Finnish term in English translations and English terms with multiple Finnish equivalents of texture are given in Appendix 9 and Appendix 10 (Lawless et al. 1997).

Table 19: A Summary of Different Terminology for Textural Properties: French, Japanese, Chinese, Finnish, And Spanish

Languages	Description	Vocabulary expression	References
French	*Properties:* Mechanical Structural Flow Surface/ contact Shape/ appearance	*Number of Words:* 34 48 19 8 31	(Nishihari et al. 2008; Tournier et al. 2007)
Japanese	Crunchiness/ crispiness Other vocabularies of textural properties	*Number of Words:* 13 70 - 445	(Hayakawa et al. 2013; Nishihari et al. 2008; Szczesniak 1988; Yoshikawa et al. 1970)
Chinese	Crunchiness/ crispiness Fundamental terms for textural properties Variety of other words	*Number of Words:* 11 30 92	(Nishihari et al. 2008; Szczesniak 1988)
Finish	A study on Finnish consumers to find the correlation between English and Finnish languages	Estimate of 70 texture and mouth-feel vocabularies Several different Finish words to express ONE English words	(Lawless et al. 1997)
Spanish	Crunchiness/ crispiness	38 % of participants were not able to express word ‘ crunchy’ 17 % of them believed the two words sharing the same definition	(Antmann et al. 2011; Varela et al. 2008; Varela et al. 2013)
Vietnamese	Comparison studies between French and Vietnamese consumers	Vietnamese participants expressed less word for texture and visual appearance of jellies compared to the French participants	(Blancher et al. 2007)

Appendix 2: French Language Related to Textural Properties (Nishihari et al. 2008)

Mechanical Properties

	Translation, example foods
Caoutchouteux	Rubber-like, springy, squid (cuttlefish)
Cassant	Brittle, raw noodles, *gressin*
Craquant	Fracturable, cracking, chips, *Cracotte*
Croquant	Crunchy, short, raw carrot, apple
Croustillant	Crispy, crusty, curly, breakfast cereals
Elastique	Elastic, gelatin candy bears, *surimi*
Flasque	Limp, not stiff, gel, jelly, oysters
Malleable	Pliable, bread paste, pie paste, modeling clay
Masticable	Chewy, masticable, gelified candy, meat
Mou	Soft, very young *gruyère*, bread crumb
Resistant	Resistant, tough meat
Rigide	Rigid, gressin, raw potato or carrot
Souple	Deformable, cooked noodles, surimi, gummy gelatin
Tender	Tender, meat, ripe pear, young cooked carrots
Attendri	Made tender, meat
Dur	Hard, tough, caramel, rice, coffee bean
Ferme	Firm, dry sausage
Flexible	Flexible, gruyère, raw spaghetti
Moelleux	Velvety, soft, bread crumb, chocolate soft cake
Plastique	Plastic, *cancoillotte*
Solide	Solid, *biscotte*, carrot, biscuit, nut
Tartinable	Spreadable, margarine, butter, *Nutella*
Cohésif	Cohesive, yogurt, paste
Compressible	Compressible, bread crumb
Gélatineux	Jelly, egg white
Gélifié	Gelified, *flamby*, fruit jelly
Avec du ressort	Springy, marshmallow
Coriace	Tough, leathery, bad quality meat
Fragile	Fragile, biscuit
Gommeux	Gummy, fruit jelly, cheese
Robuste	Robust, chocolate
Crunch	Crunch, chocolate covered puffed rice
Ramolli	Made soft, soft butter
Raide	Stiff, rigid, raw spaghetti
Pliable	Pliable, flexible, pancake, cooked noodles

Flow Properties

	Translation, example foods
Coulant	Flowing, honey, fruit filling
Epais	Thick, *Béchamel sauce*, concentrated milk
Filant	Thread forming, *natto*, *cancoillotte*, yogurt
Onctueux	Unctuous, *Mont blanc* dessert cream
Fluide	Fluid, liquid honey
Liquide	Liquid, beverage, water
Visqueux	Viscous, ketchup, jam
Couvrant	Covering, chocolate glaze, *Perle de lait yogurt*
Enveloppant	Enveloping, chocolate glaze, cream, dressing
Nappant	Covering surface, chocolate glaze, caramel
Etalable	Spreadable, cheese spread, margarine, *Nutella*
Consistant	Consistent, rice cake, thick honey
Crémeux	Creamy, dessert cream, cream
Fondant	Melting, ice cream, jelly, chocolate
Liquéfié	Liquefied, stirred yogurt, melt ice cream
Sirupeux	Syrupy, syrup, caramel, liquid honey
Dilué	Diluted, diluted syrup, drinking yogurt
Liquoreux	Liqueur-like, sweet, *Moelleux* wine
Court	Short, dessert cream, yogurt

Surface / Contact Properties

	Translation, example foods
Rugueux	Rugose, pear
Adhérent	Adherent, bread paste
Asséché	Drained, dried, stale bread
Collant	Sticky, caramel
Adhésif	Adhesive, soft caramel, packaging
Poisseux	Sticky, cheese, fruits confis
Egratigné	Scratched, bread baguette surface
Gras	Fatty, oily, oil, butter

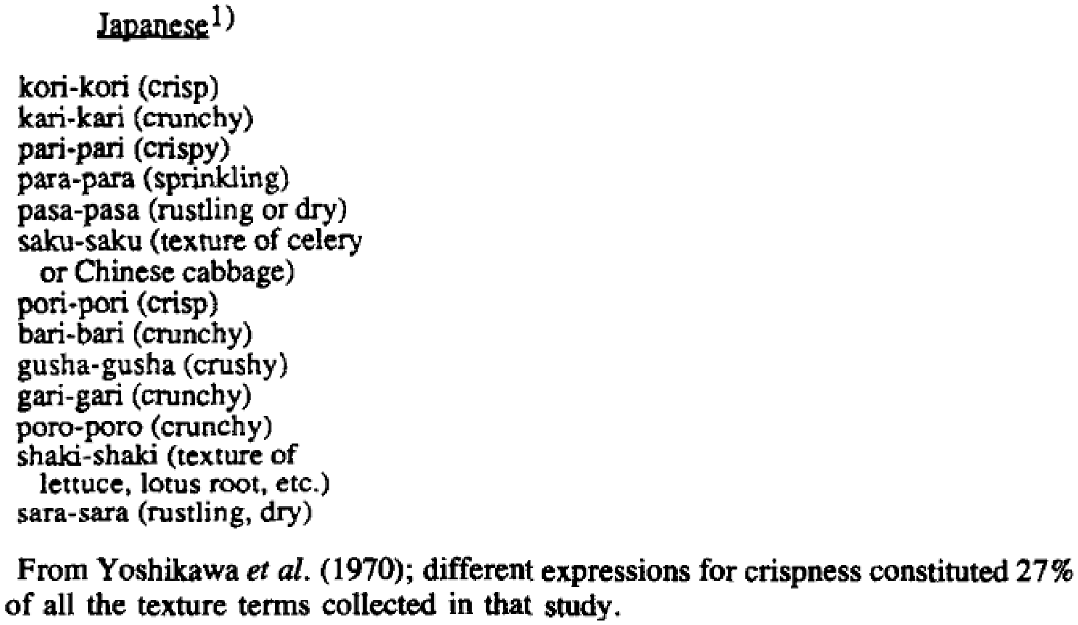
Structure Properties

	Translation, example foods
Aéré	Airy, mousse, sponge cake
Atomisé	Atomized, milk powder
Charnu	Fleshy, peach
Cotonneux	Cottony, candyfloss
Cristallisé	Crystallized, sugar
Feuilleté	Layered, roll and fold paste
Fibreux	Fibrous, rhubarb, asparagus, mango
Floculé	Flocculated, *faisselle*
Fragmenté	Fragmented, *fêta*, *Cantal cheese*
Grumeleux	Clotted, failed dressing
Mousseux	Foaming, chocolate mousse, beer, *Chantilly cream*
Soufflé	Puffed, puffed rice, *cheese soufflé*
Spongieux	Spongy, bread crumb
Aggloméré	Agglomerated, cereal bar, crumble cake paste
Boursouflé	Bloated, swollen, popcorn
Compact	Compact, dense, palet Breton biscuit, cake
Filandreux	Fibrous, low quality meat, French beans, leek
Floconneux	Flaky, oat flakes, dehydrated *Mousline puree*
Poudreux	Powdery, icing sugar, cacao powder
Pulpeux	Pulpy, stewed fruits, orange juice
Exsudant	Exuding, fresh cheese
Farineux	Mealy, some fruits
Granuleux	Granulous, gritty almond paste
Sablé	Sandy, biscuit (*Palet*, *Spritz*)
S'effritant	Crumbling, *crêpe dentelle*
S'émiettant	Crumbling, biscuit, bread
Granulaire	Granulous, fresh cheese, powder
Concassé	Crushed, crushed wheat
Crayeux	Chalky, fêta, *Petit Suisse*
Déchiqueté	Slashed, tuna crumbs
Ecrasé	Mashed, mashed potato
Emietté	Crumbled, crumble
Morcelé	Cut up, faisselle, biscuit
Pulvérulent	Powdery, powder
Tassé	Compacted, *palet breton*, ground coffee
Duveté	Downy, peach
Duveteux	Downy, peach
Eclaté	Split, *Curly*, fruit pieces in a yogurt
Gonflé	Swollen, cheese soufflé
Juteux	Juicy, tomato
Turgescent	Turgid, turnip, fruit
Bulles	Bubbly, fizzy drink, sparkling water
Cellulaire	Cellular, curly, bread
Emincé	Thinly sliced, chicken
Grossier	Coarse, thick, fêta
Mélangé	Mixed, fruited yogurt
Pulvérisé	Powdered, powder
Sablonneux	Sandy, palet, ice cream
Rembourré	Stuffed, cake, cushion edible

Shape / Appearance Properties

	Translation, example foods
Boursouflé	Bloated, swollen, popcorn
Exsudant	Exuding, fresh cheese
Floconneux	Flaky, cloudy beverage
Avachi	Out of shape, worn out, jelly
Effilé	Slender, trenchant, thin sliced almonds
Gonflé	Swollen, cake
Juteux	Juicy, meat
Gaufré	Wafer-like, *wafer biscuite*
Glabre	Smooth, without hair, cherry
Lâche	Loose, cake
Arrondi	Round, gelified candy Skittles type
Globuleux	Globular, jelly
Mouillé	Wet, watery, cake
Aplati	Flattened, pizza paste
Bandé	Bandaged, packaging
Bulbeux	Bulbous, onion, cake
Fin	Fine, thin, sliced vegetable
Flappi	Fagged out, stretched out bread
Froissé	Crumpled, egg custard surface
Imbibé	Imbibed, soaked, Sponge cake with rum
Mince	Thin, stetched out paste
Vrillé	Curled, spiralled, *tortellini*
Acéré	Steely, sharp, crystal
Aigu	Pointed, packaging
Bouffi	Swollen, inflated, pastry
Filiforme	Filiform, sliced vegetable
Fleuri	Flowery, cauliflower
Piqueté	Pitted, marked with little points, bread
Pointu	Pointed, sharp, crystal
Saillant	Protruding, jutting out, dried fruit pieces
Grenu	Full of corn, *semolina*

Appendix 3: Japanese expressions for crispness (Szczesniak 1988)

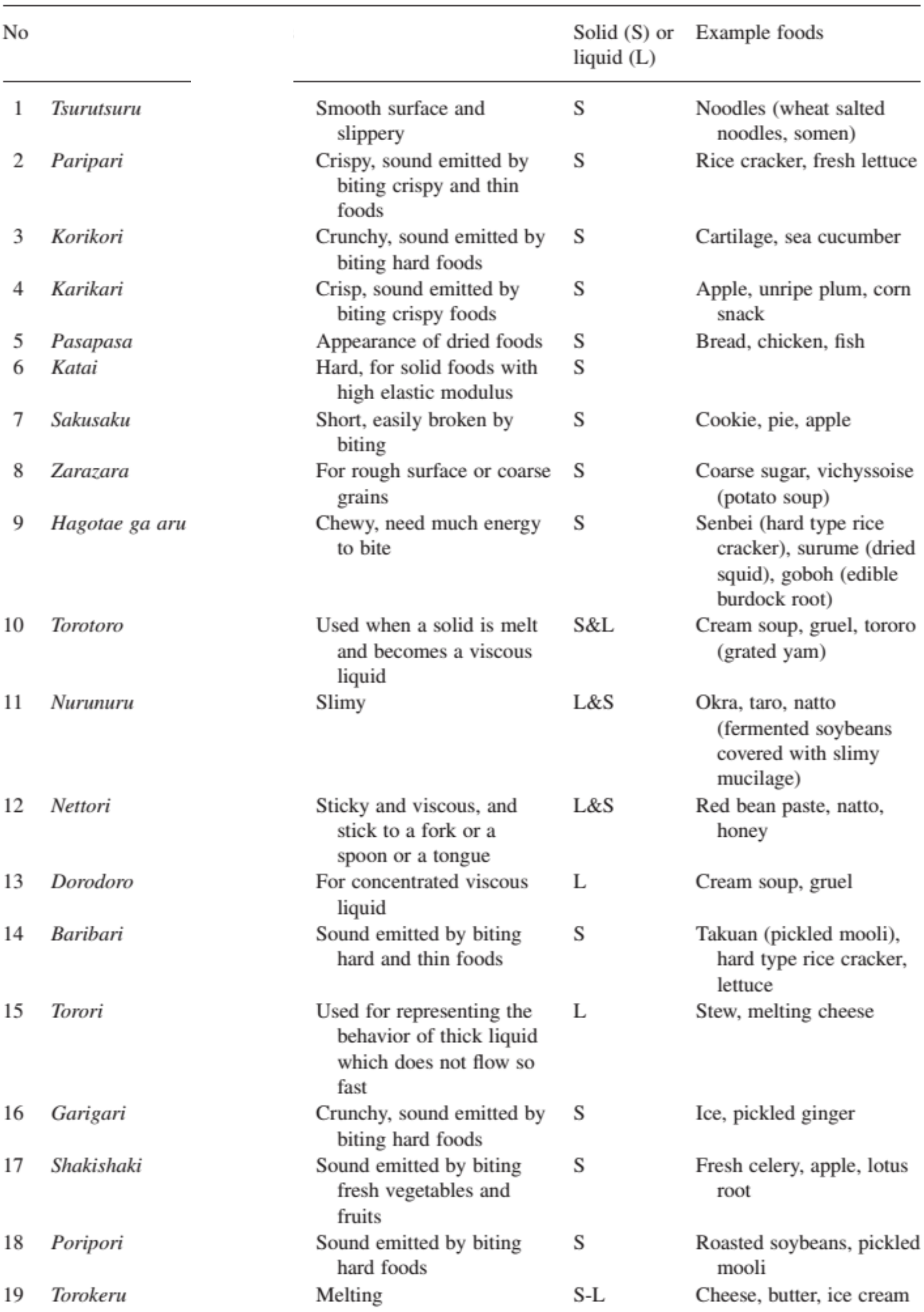


Japanese1)

kori-kori (crisp)
kari-kari (crunchy)
pari-pari (crispy)
para-para (sprinkling)
pasa-pasa (rustling or dry)
saku-saku (texture of celery
or Chinese cabbage)
pori-pori (crisp)
bari-bari (crunchy)
gusha-gusha (crushy)
gari-gari (crunchy)
poro-poro (crunchy)
shaki-shaki (texture of
lettuce, lotus root, etc.)
sara-sara (rustling, dry)

From Yoshikawa *et al.* (1970); different expressions for crispness constituted 27% of all the texture terms collected in that study.

Appendix 4: Terms related to textural properties in Japanese language (Nishihari et al. 2008)



No			Solid (S) or liquid (L)	Example foods
1	*Tsurutsuru*	Smooth surface and slippery	S	Noodles (wheat salted noodles, somen)
2	*Paripari*	Crispy, sound emitted by biting crispy and thin foods	S	Rice cracker, fresh lettuce
3	*Korikori*	Crunchy, sound emitted by biting hard foods	S	Cartilage, sea cucumber
4	*Karikari*	Crisp, sound emitted by biting crispy foods	S	Apple, unripe plum, corn snack
5	*Pasapasa*	Appearance of dried foods	S	Bread, chicken, fish
6	*Katai*	Hard, for solid foods with high elastic modulus	S	
7	*Sakusaku*	Short, easily broken by biting	S	Cookie, pie, apple
8	*Zarazara*	For rough surface or coarse grains	S	Coarse sugar, vichyssoise (potato soup)
9	*Hagotae ga aru*	Chewy, need much energy to bite	S	Senbei (hard type rice cracker), surume (dried squid), goboh (edible burdock root)
10	*Torotoro*	Used when a solid is melt and becomes a viscous liquid	S&L	Cream soup, gruel, tororo (grated yam)
11	*Nurunuru*	Slimy	L&S	Okra, taro, natto (fermented soybeans covered with slimy mucilage)
12	*Nettori*	Sticky and viscous, and stick to a fork or a spoon or a tongue	L&S	Red bean paste, natto, honey
13	*Dorodoro*	For concentrated viscous liquid	L	Cream soup, gruel
14	*Baribari*	Sound emitted by biting hard and thin foods	S	Takuan (pickled mooli), hard type rice cracker, lettuce
15	*Torori*	Used for representing the behavior of thick liquid which does not flow so fast	L	Stew, melting cheese
16	*Garigari*	Crunchy, sound emitted by biting hard foods	S	Ice, pickled ginger
17	*Shakishaki*	Sound emitted by biting fresh vegetables and fruits	S	Fresh celery, apple, lotus root
18	*Poripori*	Sound emitted by biting hard foods	S	Roasted soybeans, pickled mooli
19	*Torokeru*	Melting	S-L	Cheese, butter, ice cream

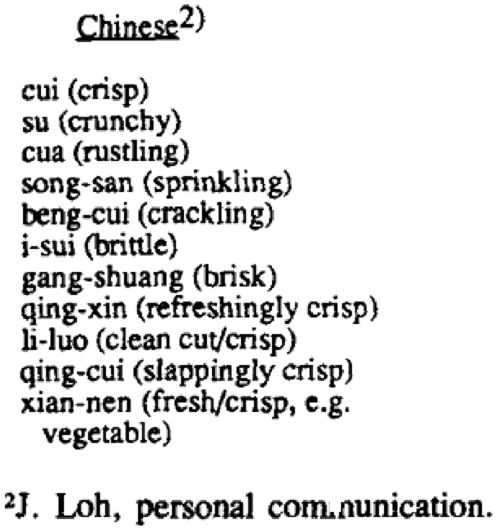
(Continued)

No			Solid (S) or liquid (L)	Example foods
20	*Betabeta*	Sticky and viscous, and stick to a fork or a spoon or a tongue	S&L	Honey, moti (sticky rice cake, totally different from rice pudding, gateau de riz)
21	*Sarasara*	State or behavior for flowing powders or thin liquid	L&S	Powdered milk, sugar, ochazuke (cooked rice in a bowl onto which green tea is poured)
22	*Sharishari*	Sound emitted when hard, thin and juicy foods are broken	S	Sherbet, Japanese pear
23	*Betobeto*	Sticky or greasy, similar to betabeta (20)	S&L	Honey, jam
24	*Poroporo*	Crumbly, easily crumbled	S	Cooked rice ball left after preparation and less sticky
25	*Kouchakoucha*	Sound emitted by chewing	S	Chewing gums
26	*Shittori*	Moist	S	Crumb of white bread, sponge cake
27	*Nebaneba*	Sticky and viscous, and stick to a fork or a spoon or a tongue	S&L	Natto, rice cake, tororo (grated yam)
28	*Puripuri*	State or behavior of elastic gel	S	Sliced raw fish of blowfish or shrimp
29	*Hokuhoku*	Used to represent state of steamed sweet potato which is about to crumble	S	Sweet potato
30	*Shikoshiko*	Chewy, highly elastic but resisting to bite	S	Wheat salted noodles, buckwheat noodles, pasta, octopus
31	*Gorigori*	Sound emitted when hard foods are broken	S	Roasted soybeans
32	*Bosoboso*	Dried and fragile	S	Dried bread, dried rice or fu (Japanese dried food made from wheat gluten)
33	*Funwari*	Fluffy, and soft similar to fukkura (36)	S	Marshmallow
34	*Maroyaka*	Mellow	L&S	Cream soup, fresh cream, yogurt
35	*Sakkuri*	Plain or easily split	S	Cookie, shortbread, pie
36	*Fukkura*	Moonish or light and easily digestible	S	Well expanded bread, steamed bread, marshmallow
37	*Aburappoi*	Oily	L&S	Deep-fried food
38	*Moroi*	Brittle, breaks at a small deformation	S	Soy curd, cookie

(continued)

No				Solid (S) or liquid (L)	Example foods
39	*Creamy*			L&S	Desert cream, cheese
40	*Parapara*		For falling appearance of granular foods, also for a light rain	S	Pilaf, fried rice
41	*Nechanecha*		Sticky and viscous, and stick to a fork or a spoon or a tongue	L&S	Soft candy, caramel, peanut butter
42	*Shitazawari no yoi*		Silky, pleasant feeling on the tongue	S&L	Pudding, jelly, ice cream
43	*Goushagousha*		Sloppy or crushed out of shape	S	
44	*Saratto shita*		Refreshing, less oily and light, dried powder	L&S	Tea, granulated sugar
45	*Boribori*		Sound emitted by biting hard foods	S	Roasted soybeans, hard type rice cracker, pickled radish
46	*Motimoti*		Chewy, highly elastic but resisting to bite	S	Dumpling
47	*Poutipouti*		Sound emitted by biting e.g., herring egg	S	Herring egg
48	*Koshi ga aru*		Body	S	Noodles
49	*Nebari ga aru*		Viscous and sticky	S	Cooked sticky rice, natto
50	*Pourupouru*		Oscillating appearance of jellies	S	Dessert jellies
51	*Juicy*			S	Hamburg steak (beef patty), orange, grapefruit
52	*Shuwashuwa*		Appearance of bubbles ascending	L	Soda, sparkling wine
53	*Jarijari*		Gritty and rough	S	Candy
54	*Mattari*		Thick and viscous for cream-like foods	L&S	Pudding, ice cream, thick Japanese green tea
55	*Katai*		Hard, for gel type foods	S	Desert jelly, pudding, soy curd
56	*Nodogoshi ga yoi*		Easy for swallowing, pleasant feeling during passing through the throat (nodo = throat)	L&S	Noodles, jelly
57	*Tsurun*		Smooth surface and thus slippery	S	Jelly
58	*Kamigotae ga aru*		Chewy, similar to hagotae ga aru (9)	S	Hard type rice cracker, dried squid, French bread (baguette)
59	*Mizuke-no-ooi*		Watery, high water content	S	Orange, watermelon
60	*Kamiyasui*	1	ender, easy to masticate (kamu = masticate, yasui = easy)	S	
61	*Nebai*	1	hick and viscous	L&S	Natto
62	*Kaminikui*	1	ough, difficult to masticate	S	Meat
63	*Tsumetai*	3	old, feeling a low temperature	S&L	Beer, white wine, ice cream, etc.
64	*Sappari shita*	3	lain and less oily	L&S	
65	*Mizuke no nai*	3	ry, less watery	S	Dried fruits
66	*Sawayaka*	3	resh, clean and pleasant	L&S	
67	*Suhtto shita*	3	inty	L&S	Minty candy and drink
68	*Nichanicha*	1	iscous and sticky	S&L	Soft candy, caramel, peanut butter
69	*Sukatto shita*	3	efreshing and pleasant drink	L	Fruit juice
70	*Nechastuku*	1	ticky, similar to nechanecha (41), nichanicha (68)	S	Soft candy, caramel, peanut butter

Appendix 5: Chinese expressions for crispness (Szczesniak 1988)

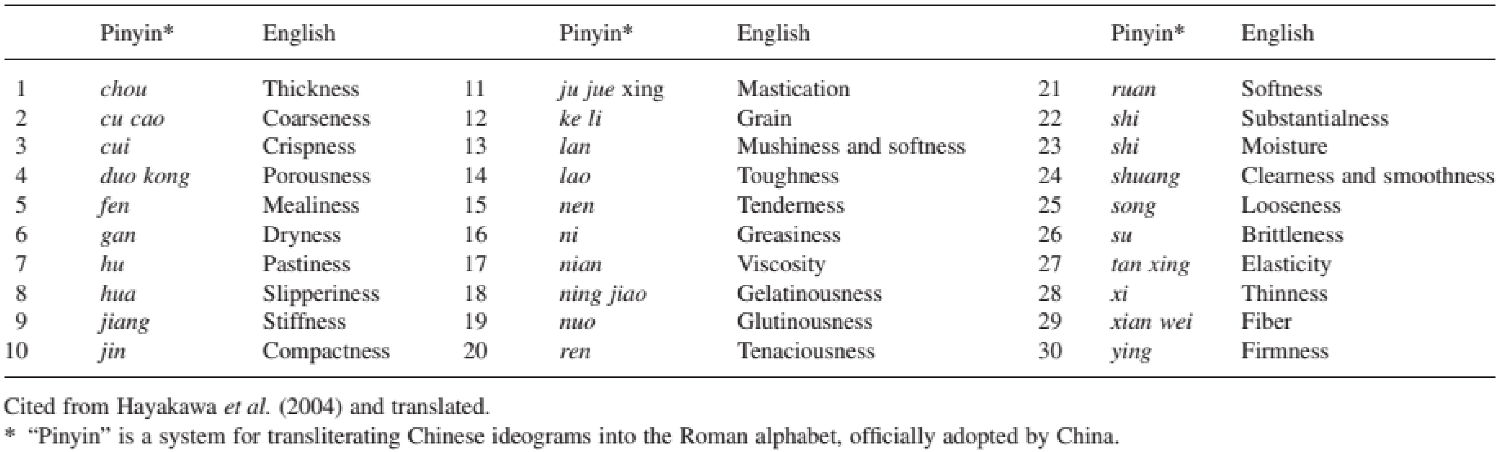


Chinese2)

cui (crisp)
su (crunchy)
cua (rustling)
song-san (sprinkling)
beng-cui (crackling)
i-sui (brittle)
gang-shuang (brisk)
qing-xin (refreshingly crisp)
li-luo (clean cut/crisp)
qing-cui (slappingly crisp)
xian-nen (fresh/crisp, e.g. vegetable)

[2] J. Loh, personal communication.

Appendix 6: 30 fundamental terms for textural properties in Chinese language (Nishihari et al. 2008)



	Pinyin*	English		Pinyin*	English		Pinyin*	English
1	*chou*	Thickness	11	*ju jue* xing	Mastication	21	*ruan*	Softness
2	*cu cao*	Coarseness	12	*ke li*	Grain	22	*shi*	Substantialness
3	*cui*	Crispness	13	*lan*	Mushiness and softness	23	*shi*	Moisture
4	*duo kong*	Porousness	14	*lao*	Toughness	24	*shuang*	Clearness and smoothness
5	*fen*	Mealiness	15	*nen*	Tenderness	25	*song*	Looseness
6	*gan*	Dryness	16	*ni*	Greasiness	26	*su*	Brittleness
7	*hu*	Pastiness	17	*nian*	Viscosity	27	*tan xing*	Elasticity
8	*hua*	Slipperiness	18	*ning jiao*	Gelatinousness	28	*xi*	Thinness
9	*jiang*	Stiffness	19	*nuo*	Glutinousness	29	*xian wei*	Fiber
10	*jin*	Compactness	20	*ren*	Tenaciousness	30	*ying*	Firmness

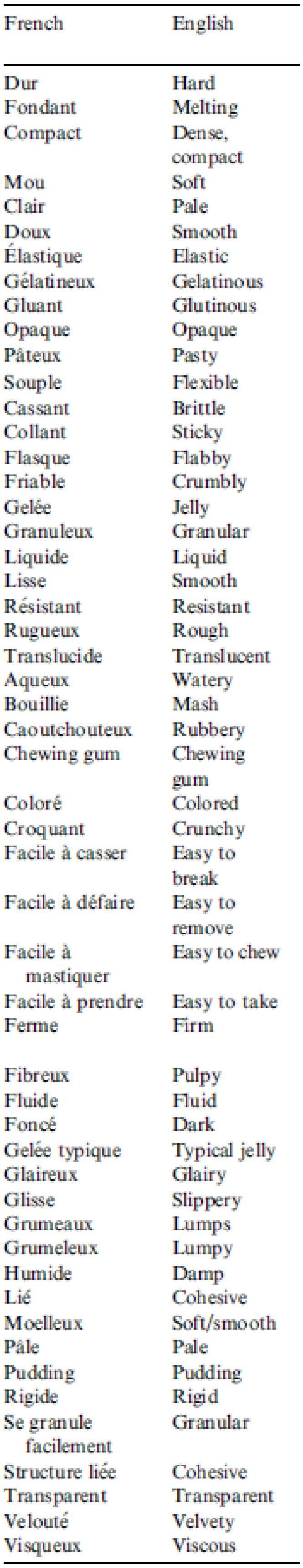
Cited from Hayakawa *et al.* (2004) and translated.

* “Pinyin” is a system for transliterating Chinese ideograms into the Roman alphabet, officially adopted by China.

Appendix 7: Variety of textural properties used in Chinese language (Nishihari et al. 2008)

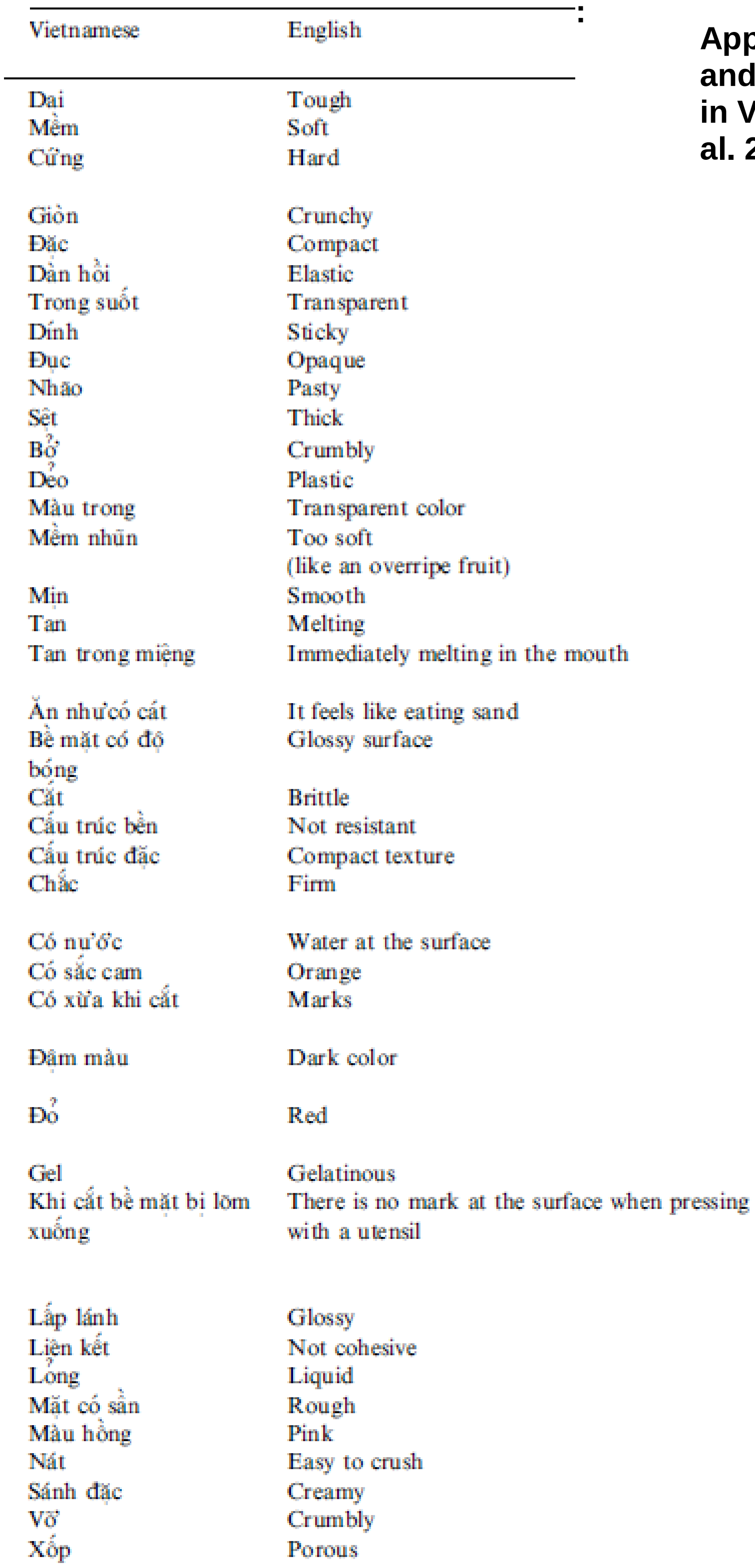
Pinyin*/English translation	Applicable foods	Pinyin/English translation	Applicable foods
bo li-zhuang/glassy	Ice	*peng song*/puffed	Steamed bread
chou/thick	Gruel/Porridge	*ren*/tenacious	Tendon
cu cao/coarse	Coarse wine	*rong mao-zhuang*/villiform	A traditional rice snack
cui/crisp	Crisp pastry	*rou hua*/silkiness	Chocolate spread
cui beng/crunchy	Crunchy biscuits	*rou nen*/soft and tender	Tender bamboo
cui nen/crisp and tender	Fresh bamboo shoot	*rou ren*/flexible	Cotton candy
duo kong-zhuang/multiholed	Overcooked egg thin soup	*rou ruan*/soft and spongy	Egg soup
duo zhi/juicy	Fresh juicy oranges	*ru-zhuang*/milkiness	Milky tea
fa pao-zhuang/foamed	A glass of foaming beer	*ru kou ji hua*/melt immediately	Ice cream
fen zhi/mealy	Boiled potatoes	*ruan*/soft	Warm butter
fen zha/powdered dregs	Dry bread	*ruan gao-zhuang*/ ointment-like	Chinese hawthorn cake
fen-zhuang/powdery	Baking powder	*ruan mian-mian*/sponge like soft	Sponge cake
gan/dry	Pastry	*se kou*/astringent	Diospyros kaki
gan hu/dry-damp	Egg yolk	*sha li-gan(sha li-zhuang)*/ gritty	Sugar
gan song/dry and loose	Common bread	*sha shuang*/gritty and daintily	Ice crystal
gan su/dry and crisp	Biscuit	*shi*/moist	A rich moist fruit-cake
gan ying/dry and hard	Compression biscuits	*shi run*/wet	Water
guo dong-zhuang/jelly	Jellied eels	*shu lan*/thoroughly cooked	Instant noodles overcooked
hai mian-zhuang/spongy	Sponge-pudding	*shuang*/clear and smooth	Bean jelly
hu/pasty	Boiled corn mealjam	*shuang cui*/clear and brittle	Potato slice
hua/slippery	Bean jelly	*shuang kou*/tasty and icy	Icy water/beer
hua liu/sliminess	Noodle	*shui zi-zi*/watery	Watery coffee
hua nen/slippery and tender	Bean curd	*song*/loose	Cake
hua run/smooth and watery	Jelly	*song cui*/loose and crispy	Ladyfinger
hua shuang/slippery and tasty	Ice cream	*song ruan*/loose and soft	Bread
jian ren/tough and firm	Gelatin jelly	*song san*/flaccidity	Rice flour snack
jian ying/hard and solid	The container of juglans regia	*su*/brittle	Brittle biscuit
jiang/stiff	Dehydrated radish	*su lan*/brittle and mushy	Well-cooked pork
jie jing-zhuang/crystalline	Sugar-crystals	*su ruan*/soft and brittle	Vol-au-vent
jie shi/substantial	A substantial steamed bread	*su song*/crisp and loose	Dried meat floss
jing dou/chewy	Noodles	*sui xie-zhuang*/detrital	Broken bits of bread
jin shi/compact	Ham	*xi*/thin	Thin soup, stew, gravy
ke li-gan/grain	Single seed of such a plant	*xi bo)*/thin and rare	Orange juice
lan ruan/mushy and soft	Overcooked noodles	*xi hu*/thin and pasty	Smooth custard
lao/tough	A tough steak	*xi mi*/fine	Fine flour/powder
nen/tender	Pork	*xi nen*/fine and tender	Vermicelli
ni/greasy	Creaminess	*xian wei-gan*/fibrous	Cereals
nian/viscosity	Rice cake	*ying*/firm	Air drying steamed bun
nian chou/gumminess	Chewing gum	*ying cui*/firm and crisp	Hard candy
nian ya/stick to teeth	The feeling of chewing biscuit	*you ju jue xing*/mastication	Tendon
ning jiao-zhuang/gelatinous	Wine gum	*you ni*/fatness	Fat meat
ning xu-zhuang/coagulate floc	Marshmallow	*you su*/oily and brittle	Oilcake
nong hou/dense	Pure juice	*you tan xing*/elasticity	Jelly candy
nuo/glutinous	Glutinous rice	*you wang-wang*/oily	Oily liquid
pao mo-zhuang/foamy	Whipped cream	*zha zhi*/dreg-sensed	Solid particles of wine a beer
peng song/fluffy	Light and fluffy mashed potatoes	*zhi mi*/pycnotic	Ham

Appendix 8: Visual appearance and texture attributes mentioned in France language (Blancher et al. 2007)



French	English
Dur	Hard
Fondant	Melting
Compact	Dense, compact
Mou	Soft
Clair	Pale
Doux	Smooth
Élastique	Elastic
Gélatineux	Gelatinous
Gluant	Glutinous
Opaque	Opaque
Pâteux	Pasty
Souple	Flexible
Cassant	Brittle
Collant	Sticky
Flasque	Flabby
Friable	Crumbly
Gelée	Jelly
Granuleux	Granular
Liquide	Liquid
Lisse	Smooth
Résistant	Resistant
Rugueux	Rough
Translucide	Translucent
Aqueux	Watery
Bouillie	Mash
Caoutchouteux	Rubbery
Chewing gum	Chewing gum
Coloré	Colored
Croquant	Crunchy
Facile à casser	Easy to break
Facile à défaire	Easy to remove
Facile à mastiquer	Easy to chew
Facile à prendre	Easy to take
Ferme	Firm
Fibreux	Pulpy
Fluide	Fluid
Foncé	Dark
Gelée typique	Typical jelly
Glaireux	Glairy
Glisse	Slippery
Grumeaux	Lumps
Grumeleux	Lumpy
Humide	Damp
Lié	Cohesive
Moelleux	Soft/smooth
Pâle	Pale
Pudding	Pudding
Rigide	Rigid
Se granule facilement	Granular
Structure liée	Cohesive
Transparent	Transparent
Velouté	Velvety
Visqueux	Viscous

Appendix 9: Visual appearance and texture attributes mentioned in Vietnam language (Blancher et al. 2007)



Vietnamese	English
Dai	Tough
Mềm	Soft
Cứng	Hard
Giòn	Crunchy
Đặc	Compact
Đàn hồi	Elastic
Trong suốt	Transparent
Dính	Sticky
Đục	Opaque
Nhão	Pasty
Sệt	Thick
Bở	Crumbly
Dẻo	Plastic
Màu trong	Transparent color
Mềm nhũn	Too soft (like an overripe fruit)
Mịn	Smooth
Tan	Melting
Tan trong miệng	Immediately melting in the mouth
Ăn nhưcó cát	It feels like eating sand
Bề mặt có độ bóng	Glossy surface
Cắt	Brittle
Cấu trúc bền	Not resistant
Cấu trúc đặc	Compact texture
Chắc	Firm
Có nước	Water at the surface
Có sắc cam	Orange
Có xửa khi cắt	Marks
Đậm màu	Dark color
Đỏ	Red
Gel	Gelatinous
Khi cắt bề mặt bị lõm xuống	There is no mark at the surface when pressing with a utensil
Lấp lánh	Glossy
Liện kết	Not cohesive
Lỏng	Liquid
Mặt có sần	Rough
Màu hồng	Pink
Nát	Easy to crush
Sánh đặc	Creamy
Vỡ	Crumbly
Xốp	Porous

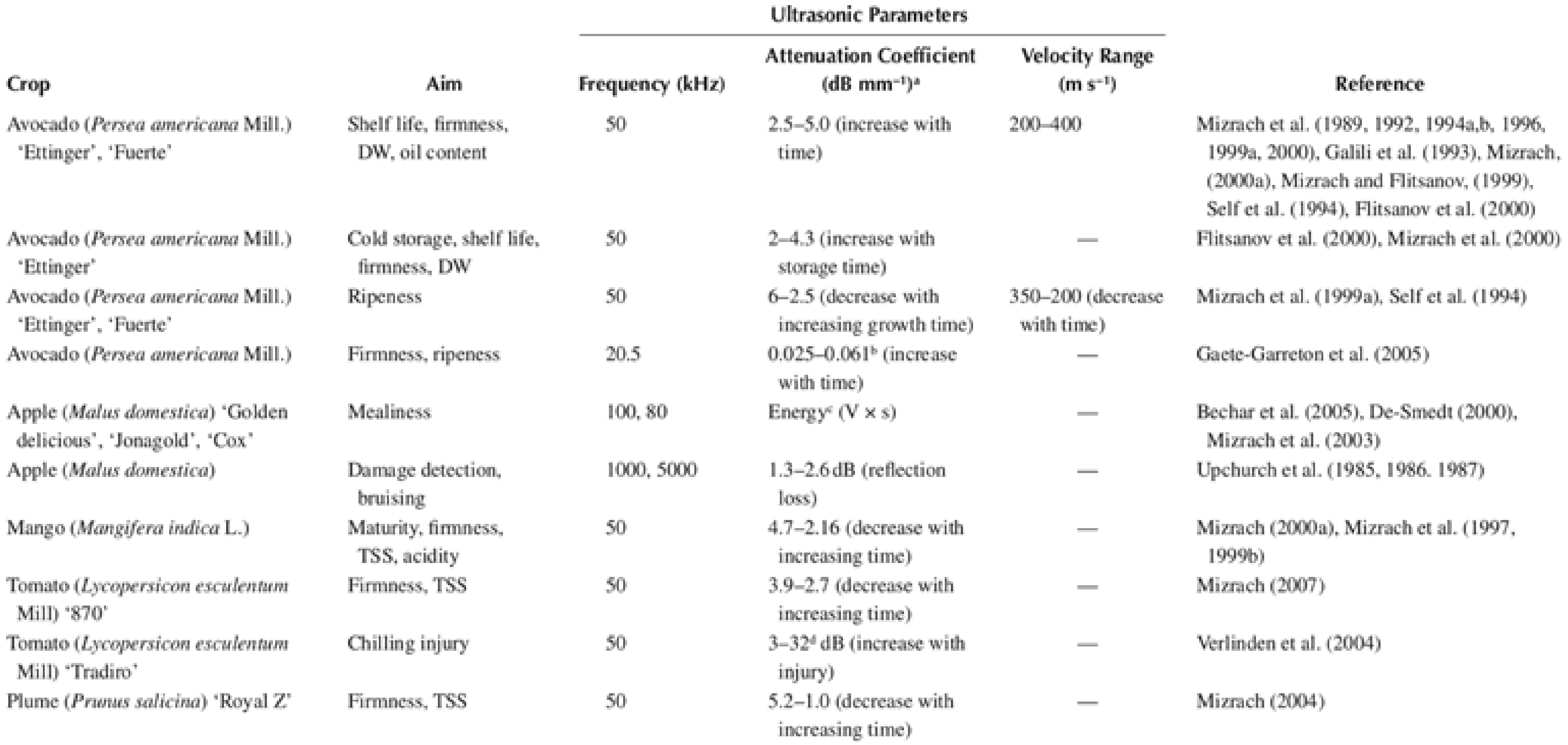
Appendix 10: Texture in Finnish Term (Lawless et al. 1997)

Finnish Term	English Term	Finnish Term	English Term
hauras	brittle	murea	tender
hiekkainen	gritty, sandy	mureneva	crumbly
hienojakoinen	fine	nestemäinen	liquid
hiutaleinen	flaky	notkea	pliable
huokoinen	porous	ohut	thin
hyytelömäinen	gelatinous	öljyinen	oily
ilmava	fluffy	paksu	thick
jähmeä	viscous	pehmeä	soft
jauhoinen	mealy, powdery	pölymäinen	dusty
jäykkä	rigid, stiff	pureskelua vaativa	chewy
joustava	elastic	rakeinen	gritty
juokseva	fluid, thin	rapea	crisp
jyväinen	grainy	rasvainen	greasy, fatty
karkea	coarse, rough	ratiseva	crunchy
kermamainen	creamy	säikeinen	fibrous
kiinteä	solid, firm	sakea	thick
kimmoisa	springy	sihisevä	fizzy
kiteinen	crystalline	siirappimainen	syrupy
kokkareinen	lumpy	sileä	smooth
kokoonpainuva	compressible	sitkeä	tough
koossapysyvä	cohesive	sosemainen	pulpy
kostea	moist	sulava	melting
kova	hard	tahmea	sticky
kuiva	dry	taikinamainen	pasty
kumimainen	rubbery, gummy	taipuisa	flexible
kuohkea	airy	tarttuva	adhesive
kupliva	bubbly	tasainen	smooth
lehtevä	flaky	tiivis	dense
levittyvä	spreadable	vaahtoava	foamy
liimamainen	gooey	vahamainen	waxy
limainen	slimy	venyvä	stretchable, elastic
liukas	slippery	vetelä	runny
löyhä	loose	vetinen	watery
märkä	wet	virtaava	fluid
mehukas	juicy	voidemainen	skin-cream like
muovailtava	ductile, plastic		

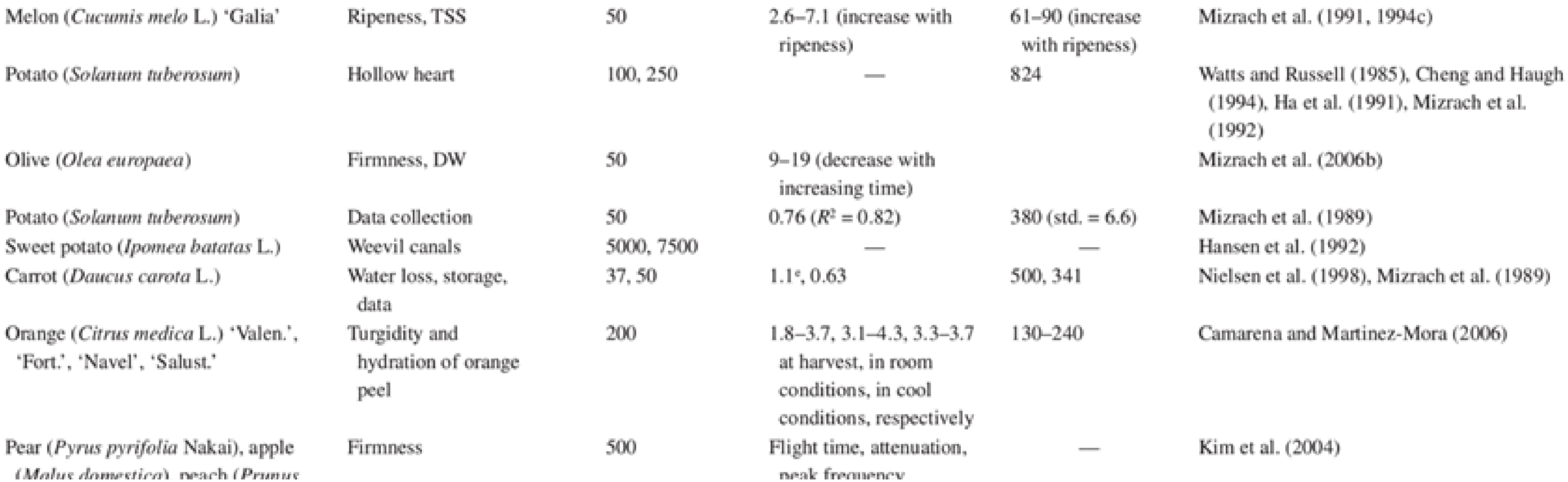
Appendix 11: English terms with multiple Finnish equivalents (Lawless et al. 1997)

Finnish Term	English Term	Nuance
juokseva	fluid, flows, thin	oils, fluids, grease
virtaava	flows	like a river
hiekkainen	gritty, sandy	sandy as in lactose crystals
rakeinen	gritty	larger, sharp-edged particles
tasainen	smooth	throughout the product
sileä	smooth	on the surface
jähmeä	thick, viscous	doesn't flow, unmovable
paksu	thick	in dimensions (wide)
sakea	thick	thick, like a sauce, pureed vegetable soup
lehtevä	flaky	like pastry crust
hiutaleinen	flaky	loose flaky particles in a continuous matrix.
taipuisa	flexible	like a plastic
notkea	flexible, pliable	easy to process, like warm butter

Appendix 12: Summary of Ultrasound Technique Applied to Selected Fresh Horticultural Produce (Mizrach 2011)



Crop	Aim	Ultrasonic Parameters Frequency (kHz)	Attenuation Coefficient (dB mm^{-1})^a	Velocity Range (m s^{-1})	Reference
Avocado (*Persea americana* Mill.) 'Ettinger', 'Fuerte'	Shelf life, firmness, DW, oil content	50	2.5–5.0 (increase with time)	200–400	Mizrach et al. (1989, 1992, 1994a,b, 1996, 1999a, 2000), Galili et al. (1993), Mizrach, (2000a), Mizrach and Flitsanov, (1999), Self et al. (1994), Flitsanov et al. (2000)
Avocado (*Persea americana* Mill.) 'Ettinger'	Cold storage, shelf life, firmness, DW	50	2–4.3 (increase with storage time)	—	Flitsanov et al. (2000), Mizrach et al. (2000)
Avocado (*Persea americana* Mill.) 'Ettinger', 'Fuerte'	Ripeness	50	6–2.5 (decrease with increasing growth time)	350–200 (decrease with time)	Mizrach et al. (1999a), Self et al. (1994)
Avocado (*Persea americana* Mill.)	Firmness, ripeness	20.5	0.025–0.061^b (increase with time)	—	Gaete-Garreton et al. (2005)
Apple (*Malus domestica*) 'Golden delicious', 'Jonagold', 'Cox'	Mealiness	100, 80	Energy^c (V × s)	—	Bechar et al. (2005), De-Smedt (2000), Mizrach et al. (2003)
Apple (*Malus domestica*)	Damage detection, bruising	1000, 5000	1.3–2.6 dB (reflection loss)	—	Upchurch et al. (1985, 1986. 1987)
Mango (*Mangifera indica* L.)	Maturity, firmness, TSS, acidity	50	4.7–2.16 (decrease with increasing time)	—	Mizrach (2000a), Mizrach et al. (1997, 1999b)
Tomato (*Lycopersicon esculentum* Mill) '870'	Firmness, TSS	50	3.9–2.7 (decrease with increasing time)	—	Mizrach (2007)
Tomato (*Lycopersicon esculentum* Mill) 'Tradiro'	Chilling injury	50	3–32^d dB (increase with injury)	—	Verlinden et al. (2004)
Plume (*Prunus salicina*) 'Royal Z'	Firmness, TSS	50	5.2–1.0 (decrease with increasing time)	—	Mizrach (2004)



Melon (*Cucumis melo* L.) 'Galia'	Ripeness, TSS	50	2.6–7.1 (increase with ripeness)	61–90 (increase with ripeness)	Mizrach et al. (1991, 1994c)
Potato (*Solanum tuberosum*)	Hollow heart	100, 250	—	824	Watts and Russell (1985), Cheng and Haugh (1994), Ha et al. (1991), Mizrach et al. (1992)
Olive (*Olea europaea*)	Firmness, DW	50	9–19 (decrease with increasing time)		Mizrach et al. (2006b)
Potato (*Solanum tuberosum*)	Data collection	50	0.76 ($R^2 = 0.82$)	380 (std. = 6.6)	Mizrach et al. (1989)
Sweet potato (*Ipomea batatas* L.)	Weevil canals	5000, 7500	—	—	Hansen et al. (1992)
Carrot (*Daucus carota* L.)	Water loss, storage, data	37, 50	1.1^c, 0.63	500, 341	Nielsen et al. (1998), Mizrach et al. (1989)
Orange (*Citrus medica* L.) 'Valen.', 'Fort.', 'Navel', 'Salust.'	Turgidity and hydration of orange peel	200	1.8–3.7, 3.1–4.3, 3.3–3.7 at harvest, in room conditions, in cool conditions, respectively	130–240	Camarena and Martinez-Mora (2006)
Pear (*Pyrus pyrifolia* Nakai), apple (*Malus domestica*), peach (*Prunus*	Firmness	500	Flight time, attenuation, peak frequency	—	Kim et al. (2004)