UNIVERSITY OF NATAL

POSTHARVEST MAINTENANCE OF THE SHELF LIFE AND QUALITY ATTRIBUTES OF BANANA AND PAPAYA

By

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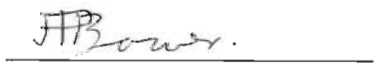
Declaration

I, Biniam M. Ghebreslassie, certify that the research work reported in this thesis, except where otherwise acknowledged, is my own original research and has not been submitted in part or as whole to any other university. The research work was conducted at the University of Natal, Pietermaritzburg.

Signed:- 

Biniam Ghebreslassie

I certify that the above statement is correct.

Signed: - 

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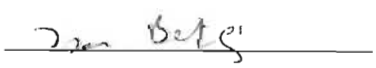
Signed: - 

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General Abstract

Banana and papaya are two commonly grown fruits of the tropical regions of the world. Like most tropical climacteric fruits, these fruits have a short shelf life and deteriorate rapidly after harvest. This becomes more pronounced in tropical areas where cold chain management practices are poor and the ability environmental control is limited. To extend the shelf life with maintained quality of the fruits and to investigate the effect of various postharvest treatments a study was conducted on banana cv. Williams and on papaya cv. Hortus Gold. The treatments included storage in different packaging materials such as micro perforated polypropylene bags (MPB), Micro perforated polypropylene bags with ethylene absorbent (MPB+K), macro perforated polypropylene bags coated with anti mist coating (PP), fruit waxing, gibberellic acid (GA_3) and indole butyric acid (IBA) applications. Comparison was made against untreated fruits. Banana fruits were held at 12, 15 and 22°C, while papaya fruit were held at 5.5, 7, 10 and 22°C. Percentage weight loss (PWL), firmness change, visual colour development and respiration rate were evaluated on a weekly basis during the storage period. Selected quality attributes such as total soluble solids (TSS), pH, total chlorophyll, total carotenoids and organic acids for banana and TSS, pH, titratable acid (TA) and sugar: acid ratio for papaya were analyzed at the end of the storage period. PWL, softening, colour development and respiration rate increased significantly ($P<0.001$), irrespective of treatments, as the storage period and temperature were increased. Banana fruits stored at 12°C and papaya fruits at 7°C had an extended shelf life of eight weeks. On the other hand, both fruits stored at higher (22°C) exhibited lower storage life and rapid deterioration. Despite the effect of storage temperatures, MBP, MPB+K, PP and waxing significantly ($P<0.001$) reduced PWL at all storage temperatures for both fruits, especially as compared to control fruits. None of the packaging materials resulted in development of any off-flavour and/or fungal decay, although fruits displayed an increased respiration rate at higher storage temperatures. Similarly, waxing, GA_3, PP and MPB significantly ($P<0.001$) reduced fruit softening and colour development as compared to control fruits. As a result fruit shelf life was extended. Waxing and GA_3 significantly ($P<0.001$) retarded respiration rate for both fruit types as compared to control fruits. Waxing was the most effective treatment in retarding postharvest physiological changes for both fruits at all storage temperatures. IBA treated banana fruits exhibited significantly ($P<0.001$) reduced PWL and fruit softening as compared to control fruits.

Quality analysis after storage for TSS, pH, organic acids, total chlorophyll and total carotenoids for banana, and analysis of TSS, pH, TA and sugar:acid ratio for papaya showed no measurable differences ($P<0.05$) among treatments. This implies all the treatments, which increased fruit shelf life, also preserved fruit internal quality. It is, therefore, concluded that waxing, GA_3, PP, MPB+K and/or IBA and MPB can be alternative approaches to extend fruit shelf life and maintain fruit quality within the context of low infrastructure and poor storage facilities.

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Table of contents

List of Tables

List of Figures

List of Abbreviations

1-MCP	1-Methylecyclopropane
2,4-D	Dichlorophenoxy Acetic Acid
ABA	Abscisic acid
ACC	1-AminoCycloprpane-1-Carboxylic Acid
ANOVA	Analysis of Variance
AVG	L-2-amino-4-(2-aminoethoxy)-trans-3-butenoicacid,aminovinyl glycene
CA	Controlled Atmosphere
C_2H_4	Ethylene
CHO	Carbohydrate
CI	Chilling Injury
CO_2	Carbon dioxide
CRD	Complete Randomized Design
EFE	Ethylene Forming Enzyme
FAO	Food and Agriculture Organization
GA_3	Gibberellic Acid
H_2SO_4	Sulfuric Acid
HPLC	High Performance Liquid Chromatography
IAA	Indole Acetic Acid
IBA	Indole Butyric Acid
IITA	International Institute of Tropical Agriculture
INIBAP	International Network for the Improvement Banana and Plantain
IRGA	Infra Red Gas Analyzer
$KMnO_4$	Potassium permanganate
LSD	Least Significant Difference
MA	Modified Atmosphere
MAP	Modified Atmosphere Packaging
MPB	Micro Perforated Bags
MPB+K	Micro Perforated Bags with ethylene absorbent
NaOH	Sodium Hydroxide
O_2	Oxygen

PCP	Pre Climacteric Period
PG	Polygalacturonase
PGR	Plant Growth Regulators
PME	Pectin Methyl Esterase
PP	Polypropelene bag coated with anti-mist coating
PWL	Percentage Weight Loss
RH	Relative Humidity
SAM	S-adenosylmethionine
TA	Titratable Acid
TBZ	Thiabendazole
TCA	Tricarboxylic Acid (pathway)
TSS	Total Soluble Solid
WVPD	Water Vapour Pressure Deficit

Chapter 1: General Introduction

1. General Introduction

Hunger has been a reality ever since *Homo sapiens* first walked on earth. The Food and Agricultural Organization (FAO), Rome, in its report in the early, 1980s, agriculture towards 2000, indicated that food production would have to be doubled in the last two decades of the twentieth century to avoid global famine (Salunkhe and Kadom, 1995). The chief concern of the modern world is "enough and nutritious food for all" (Salunkhe and Desai, 1984b). Despite the remarkable progress made in increasing food production at the global level, approximately half of the population in the third world does not have access to adequate food supplies (FAO, 1989). This problem becomes more realistic to consumers in the developing countries where more than 80% of the disposable income is spent on food, and where there is lack of processing and storage facilities and, hence, food supply is limited to seasonal production and climatic variability. There are many reasons for this, one of which is food loss occurring during the postharvest and marketing period (FAO, 1989). The problem of population food imbalance can obviously be solved either by increasing food supplies or by limiting population growth. However, this demands significant amounts of money and time (Salunkhe and Desai, 1984a). One other alternative, but most of the time overlooked solution, is to control or reduce the food loss that occurs throughout the food production, harvesting and marketing process (postharvest loss). Although the scientific and technological revolution has created unlimited opportunities for production of sufficient and/or the right type of food for all mankind, not enough attention has been given to one of the vital aspects of the food problem, namely, "the conservation of food that is already produced" (Salunkhe and Desai, 1984b). Hence, these authors recommended that coordinated efforts are essential to bring food and people to a reasonable balance.

Postharvest food losses include very diverse food categories ranging from durable cereal grain products to perishable fruits, vegetables, dairy, animal, and marine products (Toma *et al.*, 1990). According to Kays (1991) the major concern with handling perishable crops is the maintenance of products condition. Therefore, it is necessary to consider not only the nature of the plant material, but also the technological and economic aspects associated with getting the material to the consumer. Informed opinion now suggests (Wills *et al.*, 1998) that increased emphasis should be placed on conservation after harvest, rather than endeavouring to further boost crop

production, as this would appear to offer better return for the available resources of labour, energy and capital. Postharvest deterioration is related to the botany, physiology, and biochemistry as well as growth and development of the crop. According to Kader (1992a) losses in quality and quantity affect horticultural crops between harvest and consumption. These are very dramatic, ranging from 5 to 25% and 20 to 50% in developed and undeveloped countries, respectively. This means one quarter, if not half, of what has been produced never reaches the consumer for whom it was grown. The reduction of the losses in a systematic way requires knowledge of postharvest physiology, and applied technology in handling. This and the appreciation of the biological limitations represent storage potential (Paull, 1994).

According to Salunkhe *et al.* (1991) fruits with increasing recognition of their value in the human diet are gaining commercial importance. Since fresh or processed fruit forms an important part of our diet, there is an ever-increasing demand, at least in the Western society, for both, improved quality and extended variety of fruit available (Tucker, 1993). This is also of great importance in the nutrition of less-developed countries. Such products are essential for man as far as food nutrition is concerned. In most of the less developed countries, food inadequacies relate not only to the quantity (caloric need), but also the quality (nutrition) (Salunkhe and Desai, 1984b). The nutritional self-sufficiency of the rural world demands vast increase in the quality of fruits, which are excellent sources of the essential dietary nutrients, vitamins, and minerals. In line with that, perishable fruits are one of the most important of all food categories in combating the so-called malnutrition diseases of the developing world. Adequate consumption of fruits helps prevent or guard against nutrition-related diseases (Beri- Beri, Xeropthalemia, and others) (Toma *et al.*, 1990). Apart from this fruits can add flavour, colour, and variety to the otherwise monotonous diet.

Fruits, unlike seeds, fleshy roots, tubers, and bulbs, which are morphologically and physiologically adapted to maintain the tissue in the dormant state, are vulnerable to environmental conditions. As fresh fruits are living tissues they will deteriorate relatively rapidly following harvest (Kader, 1992a). Because of their generally high value, seasonality and great perishability, fruit are the most common commodities to be given special postharvest treatments (Harris, 1988). Hence, it becomes very important to minimize losses by the application of available technology, and by undertaking new research in this vital area (Salunkhe and Desai,

1984b). Banana and papaya are some of the commonly grown tropical fruits characterized by short shelf-life after harvest.

Banana, belonging to the family of *Musacea,* is a large herb with a pseudostem built up from leaf sheaths. The primary center of origin is thought to be Malesia (Malaysia, Indonesia, the Philippines, Borneo and Papua New Guinea) (Nakasone and Paull, 1998). Most of the cultivars of the edible banana are triploid and are derived from *M. acuminata* (A), and *M. balbisiana* (B) (Turner, 1997). Banana is one of the most important crops grown, with a global annual production of about 45 million metric ton (FAO, 1990). After citrus, banana is the most important fruit in world trade. Similarly, it is an important food crop in the humid forest and mid-altitude agroecologies of Sub-Saharan Africa (Ferris, 1998), especially in some parts of East, Central, and West Africa. It is a useful source of vitamin A, C (ascorbic acid), and B_6 and has about twice the concentration of potassium (K) compared with other ripe fruits (Crane and Balerdi, 1998). Moreover, the flavour, texture, convenience, ease of eating and nutritional value has made it very popular throughout the developed world (Nakasone and Paull, 1998).

Ferris (1998) explained that preharvest crop management of banana has a direct effect on postharvest characteristics. A high quality crop gives the best postharvest performance. According to John and Marchal (1995) the postharvest life of banana has three stages. These authors defined these stages as the pre-climacteric or green life, the ripening stage and the senescence phase, during which metabolism slows down and the quality of the fruit declines. Growth ceases when the fruit is fully mature and the ripening phase is initiated. Bananas are climacteric fruit with a short shelf-life of 6-10 days at ambient temperature and are susceptible to chilling injury (CI) if stored at a temperature below 10°C. Developing methods to delay ripening would therefore be of immense value in prolonging the shelf-life and increasing the international marketing potential. Postharvest research is concerned with maintaining crop quality until the crop reaches the consumer. This is particularly important for perishable crops that deteriorate rapidly (Ferris, 1998). Seymour (1993) indicated that research on banana has given details of the sequence of metabolism that occurs during ripening and that future work needs to elucidate control mechanisms in postharvest loss of banana that ranges between 20 – 80% (Appendix A).

Papaya (*Carica papaya)* belongs to the family *Caricaceae.* It is a native of tropical America and has spread all over the tropical and warmer subtropical parts of the world. It is a smaller unbranched, soft-wooded tree with latex vessels throughout the plant. Papaya is grown mainly for fresh consumption or for production of the proteolitic enzyme papain from the fruit latex (Salunkhe and Kadam, 1995). It is a popular breakfast food and serves to make fruit salads and many desserts. The fruit contains about 10% sugar, considerable amount of vitamin A, some vitamin C, and is considered a mild laxative (Samson, 1980). The plant is usually dioecious with either male or female flowers; however, hermaphrodite (bisexual) trees also occur. Like many other climacteric fruits papaya has a short postharvest life. According to Salunkhe and Desai (1984b) the postharvest loss of fresh papaya has been estimated to vary from 40 – 100% under varying climatic conditions. The high figures of these postharvest losses underscore the importance of careful handling and quality maintenance after harvest (Wang, 1999).

This study was devoted to maintaining quality and hence increasing shelf-life of fruits to meet food requirements in developing countries. An extension of the shelf-life of banana and papaya was, therefore, attempted by various treatments. A means of extending the period for which these fruits remain attractive and appealing to the consumer, within the context of poor infrastructure, lack of storage facilities, such as cold chain and limited environmental control of tropical conditions, could be of considerable benefit to both, growers and consumers. Quality evaluation after storage was also part of the study.

The objectives of the study were as follows:

1. To investigate the effects of various treatments, including waxing, different micro-perforated bags with or without ethylene (C_2H_4) absorbent, polypropylene bags coated with anti-mist coating and growth regulators: gibberellic acid (GA_3) and Indolebuytric acid (IBA) on the storage life and subsequent ripening characteristics of banana and papaya compared against untreated control fruits.
2. To evaluate the quality of the fruits after storage, mainly through the following quality parameters: Total soluble solids (TSS), pH, change in total chlorophyll, carotenoids, and organic acids in banana and TSS, titratable acid (TA), pH, and sugar:acid ratio in papaya.
3. To recommend to the growers, packhouses and distributors, especially in developing countries which treatments to use to delay ripening and enhance shelf-life.

Chapter 2: Literature Review

2.1 Postharvest Loss

Postharvest research has been traditionally divided in to postharvest physiology and an empirical evaluation of handling and storage conditions on quality (shelf-life evaluation) (Shewfelt, 1986). Postharvest physiology is concerned with plants or plant parts that are handled and marketed in the living state (Kays, 1991). Fruits like banana and papaya are living organs subjected to continuous change after harvesting (Marchal, 1998, Shewfelt, 1986), hence a considerable amount of the harvested produce is lost after harvest if handling is improper. Quality decline of fruits through high respiration rate, high metabolic activity, moisture loss, softening, yellowing, postharvest decay and/or loss of flavour and nutritional value leads to significant postharvest loss (Wang, 1999). These losses, which, if avoided could positively improve the nutritional status of the poor population dwelling both in cities and rural areas of developing countries (Salunkhe and Desai, 1984b). Unfortunately, much of the harvested plant material never reaches the consumption or processing point of utilization but is discarded (Kays, 1991). As a consequence, the producers are deprived of a large income on one hand and nutritive food to the consumers on the other. Hence the handling, storage and marketing of plant products needs great attention, so as to ensure the produce that required large input of labour, material, and capital reaches the consumer with minimal loss (Mattoo and Handa, 2001). Postharvest loss is any change in quality or quantity of a product after harvest that prevents or alters its intended use or decreases its value.

2.2 Factors Affecting Postharvest Loss

Factors that affect the postharvest losses of perishable fruits like banana and papaya vary widely and become more complex as the system of marketing becomes more complex (FAO, 1989). Moreover, according to Toma *et al.* (1990) perishable produce differs in physical and biochemical characteristics from durable produce, so the mechanisms and causative factors of loss are quite different. The ripening of banana fruit is a complex process, which is strongly affected by preharvest and postharvest factors (Lebibet *et al.*, 1995). Like other horticultural crops banana and papaya are high in water content and thus subject to desiccation and mechanical injury, which leads to all stress and attack by bacteria, fungi and other pathological breakdown (Kader, 1992a). The high water content is linked to high perishability and a short

shelf-life (Tucker, 1993). It is this perishability, and inherent short shelf-life, that presents the greatest problem to the successful transport and marketing of these fresh fruits. Factors assumed to affect fruit shelf-life and thus cause postharvest loss of fresh banana and papaya can be categorized as follows:

2.2.1 Preharvest Factors

In considering final quality, it is important to begin with preharvest fruit growth and harvest maturity. Unfortunately, in studying the effect of conditions of storage and transportation on the commercial life of fruits, far too little, and often no attention, has been given to the preharvest factors (Frenkel *et al.*, 1975). Preharvest practices set the maximum value for the quality of the fruit. Salunkhe and Desai (1984a) have mentioned that although soil type cannot be changed in an established orchard, the contribution the soil makes to the attributes of the developing fruit, which affects its postharvest physiology, is greatly influenced by almost every preharvest cultural practice. The main preharvest factors that influence postharvest performances of the fruits are genetic (cultivar) and environmental (temperature, light, water, nutrient supply, maturity at harvesting and application of agricultural chemicals).

2.2.1.1 Cultivar

Although cultural practices and climate are important preharvest factors that affect postharvest performance of the fruits, relatively little attention has been given to cultivar effects on postharvest responses at a metabolic level (Watkins and Pritts, 2001). Different cultivars have different degrees of resistance to different stresses and to environmental conditions in general. Accordingly, the compositions of the fruit in relation to flavour, texture, colour, nutritional value, and quality varies considerably, even if the preharvest treatment of the plant was the same. Crane and Balerdi (1998) reported that banana cultivars vary greatly in plant and fruit size, plant morphology, fruit quality, disease and insect resistance. A significant effect of cultivar on the shelf-life and quality (Seberry and Harris, 1998), weight loss and ripening of banana fruit has been reported by Burdon *et al.* (1993). Field observations suggested that cultivars with a *Musa balbisiana* genome are more drought resistant than those containing only the *M. acuminata* genome (Simmonds, 1966). Furthermore, most cultivars of banana grow well in the tropics; the dwarf Cavendish, however, grow also well in some subtropical areas without

any effect to the quality, because these cultivars are relatively frost resistant compared to the others (Salunkhe and Desai, 1984a). The flavour of pear fruit in the ripened condition appears to be largely determined by genetic constitution and is less influenced by environment (Quamme and Gray, 1985). Similarly, the CI sensitivity, shelf-life, physiological disorder and disease tolerance of citrus fruits after harvest varies depending upon the cultivar (Pantastico *et al.*, 1975). These become some of the many examples that explain how cultivar affects fruit quality and ultimate shelf-life.

2.2.1.2 Environmental Factors

Environmental variables directly influence every aspect of plant growth and development. Climatic factors including temperature, light, and soil water, have been considered to a lesser extent because they are more difficult to control experimentally (Watkins and Pritts, 2001). Although the climate is difficult to control, knowledge of how it affects postharvest quality is useful in predicting postharvest problems and determining a market policy (Pantastico, 1975). Climatic factors have a profound effect on plant phenotype and determine the extent to which genotypic potential is expressed (Schaffer and Andersen, 1994). Length of storage, respiration, transpiration, chemical composition, external appearance, anatomical structures, decay, taste qualities, and other postharvest behaviours and characteristics partly reflect the cultural and environmental conditions to which the produce is exposed (Pantastico, 1975). The main climatic factors that affect fruit quality are discussed below.

a. Light: As Rom (1996) stated, light has several important functions in tree fruit, including photosynthesis, photomorphogenesis, phototropism and photoperiodism. The duration, intensity and quality of light affect fruit colour, sugar accumulation, antioxidant concentrations, and thus quality of the fruit at harvest (Pantastico, 1975). Crane and Balerdi (1998) mentioned that in subtropical areas, banana requires full sunlight, because excessively shaded plants are stunted and produce small and poor quality fruits. It is also reported by Samson (1980), that the best quality papaya fruit, which is determined largely by sugar content, is harvested from plants grown under full sunlight. As a result fruit grown in the inner portion of large tree canopies is usually affected by low light interception, and hence has small size and low quality. Moreover, fruits grown under light levels less than 70-80% full sunlight exhibited reduced colour and soluble solids (Rom, 1996).

b. Temperature: For most fruits, the higher the temperature during the growing period, the earlier the harvesting time. Temperature effects related very closely to cell division, probably the most critical time for fruit development. High temperature during the vegetative stage hastens growth and reproductive maturity, however, an excessive growth rate can result in misshapen produce and unpleasant flavour in most fruits (Shewfelt and Prussia, 1993). Exposure of banana fruit to high temperature (>30°C) before and after harvesting causes mixed ripeness and, if a temperature of 40-45°C is experienced during the period of just before and after flower emergence, ripe fruit breakdown will be observed (Lagerwall, 1997). According to Rom (1996) the concentration of secondary metabolites in fruits such as colour and flavour can be reduced under high temperature mainly because of the reduced carbohydrate (CHO) produced, and a higher percentage of CHO breakdowns to maintain respiration rates at high temperature. Apart from this, temperature is the driving force for the transpiration of plants and hence affects the uptake of nutrients, which later affects the quality of the fruit. On the other hand, Lagerwall (1997) indicated that low temperature, especially during flower initiation inside the pseudostem of the banana plant, causes "November dump" (small and malformed fruit). It was, also, reported by Marler (1994) that papaya fruit flavour tends to be insipid if fruit maturation occurs during periods when temperatures are above freezing but below optimum (21-33°C).

c. Water supply: According, to Srikul and Turner (1995) preharvest water deficit reduces green life of the fruit much more than it reduces growth. Insufficient water supply can reduce fruit enlargement and ultimately can lead to wilting, which is a serious defect in most fruits (Shewfelt and Prussia, 1993). Fruit of banana plants which experienced high water deficit, turn a yellowish green colour and die prematurely (Ke, 1979). Moreover, moisture stress is the major cause of nutrient deficiency as it interrupts the root uptake and leads to localized deficiency in fruits. FAO (1989) reported that optimum application of water is essential as too much may increase the tendency of fruit decay and low supply leads to low juice and thick skin for most fruits.

d. Nutrients: Lack of plant nutrients in the soil can seriously affect the quality of fresh produce at harvest (FAO, 1989). There are numerous reports of the relationship between mineral composition of the fruit and postharvest disorders involving a number of elements. The nutrient composition of the harvested product is strongly influenced by the nutrition of the parent plant (Kays, 1991). Plant nutrients affect energy flow in the plants since they are essential for photosynthesis and for regulating energy metabolism and CHO transport (Shewfelt and Prussia,

1993). Calcium (Ca) is one of the essential nutrients that play a pivotal role in fruit quality and hence postharvest storage life. It is the main constituent of the cell wall and all membranes. Many of the physiological disorders occurring in fruit and vegetables are related to a Ca content of the tissue (Bangerth, 1974). Adequate fruit Ca enhances storage life and helps stored fruit resist a range of breakdown conditions including internal breakdown, low temperature breakdown, water core, lenticel's breakdown, as well as bitter pit (Weir and Cresswell, 1993). According to Kays (1991) and Salunkhe and Desai (1984a) fruits high in Ca have a low respiration rate and a longer potential storage life than does a low-Ca fruit. Nitrogen (N) fertilization has been reported to affect quality of various fruits and vegetables. Although N is essential for plant growth, too high level can promote excessive vegetative growth, delay fruit ripening and lead to soft, poorly coloured fruit, with poor storage qualities (Weir and Cresswell, 1993). Increased application of N reduces green life of banana fruits. This is correlated to the effect of N to cause an elevated respiration rate (Kays (1991). It is also reported that excessive N application to papaya results in softer fruit and accumulation of nitrate, which is a problem when fruits are canned (Lindsay and Brian, 1982). Potassium (K) is another important nutrient for the formation of proteins, CHO and fats and for the functioning of chlorophyll and several enzymes (Weir and Cresswell, 1993). It affects and is required for fruit filling (size and weight) of banana and hence bunch mass. Lack of K brings about poor development, abnormal ripening and high respiration rate. Yield and quality can be affected by K deficiency even before leaf symptoms are seen (Weir and Cresswell, 1993). Fruits from K deficient plants are often small, poorly coloured and taste insipid. Boron (B) is needed for proper pollination and the development of viable seeds, which in turn affects the normal development of fruit (Weir and Cresswell, 1993). According to Wojcik *et al.* (1999) B is necessary for high yield and quality of apple fruit. It plays a similar role to Ca in plant nutrition, which makes it essential for quality factors such as skin strength, fruit firmness and storage life. B deficiency can lead to lumpiness in papaya fruit (FAO, 1989) hard and misshapen, and contain brown, gummy discolouration in the albedo of citrus fruits (Pantastico, 1975). B usually stimulates pollen tube growth and Zinc appears to improve fruit set and quality in most fruits.

e. Maturity and harvesting time: Maturation is the stage of development leading to the attainment of physiological or horticultural maturity (Kader, 1999). Harvesting of fruit at an appropriate stage is important from a postharvest shelf-life and quality point of view (Pantastico, 1975; Shewfelt, 1986). Maturity at harvest is an important factor that determines the subsequent

storage life, ripening and final fruit quality (Kader, 1994; Mustaffa *et al.*, 1998; Techawongstien, 1999). These authors also added that fruit of each cultivar picked either too early or too late in the season are more susceptible to physiological disorders and have a shorter storage life than those picked at the correct stage. Wilson *et al.* (1999) reported that commodities to be stored should be harvested at optimum maturity, because storage life may be reduced if they are immature or over mature. Similarly, Pantastico *et al.* (1975) reported that prematurely harvested bananas, mangoes, or tomatoes ripen unsatisfactorily, although storage period may be longer. Hence, harvesting of the fruit at the correct time is essential, as it can physiologically influence the postharvest performance of the fruit. It is also reported that fruits harvested during cool nights or periods are less susceptible to CI after harvest than those harvested during a hot day (Swarts, 1992). It is therefore desirable to harvest commodities " in the cool time of the days " and allow it to stand in the orchard or field in open boxes during part of the night to cool or hydro-cool before loading onto trucks (Salunkhe *et al.*, 1991).

f. Agricultural chemicals: agricultural chemicals can be categorized in two groups (FAO, 1989):

- *Pesticides and herbicides*: pesticides and herbicides are used as sprays or soil application to control weeds, diseases and pests. They are dangerous because they can damage produce by producing spray burns if used incorrectly, and they can leave poisonous residues on the produce after harvest, which in turn affects the shelf-life and quality.
- *Growth regulator chemicals*: Plant growth regulators (PGRs) are used in the field mainly to improve the marketability of the produce through control of fruit set and production of uniform ripening. Marcell and Clijisters (1978) reported that PGRs could modify the uptake, translocation and concentration of minerals in the plant. According to Balasubramaniam and Agnew (1990) application of GA_3 at the beginning of stage three of fruit growth significantly improved postharvest handling and storage quality of cherries. Moreover Lurier *et al.* (1997) reported that preharvest treatment of nectarine with GA_3 could both delay ripening on the tree and improve their quality in controlled atmosphere (CA) storage. If PGRs are used at the right time and in the right amount they provide the grower with an additional means of plant manipulation. However, "since very small amounts of PGRs will cause significant changes in the plant, it is essential that we understand the interactions between environmental condition, plant growth and development, and plant growth regulations" (Seeley, 1981).

It can, therefore, be inferred that almost 50%, if not higher, of the postharvest quality of produce is affected by preharvest activities. Hence, the grower must make every effort to control his orchard practices to ensure the production of fruits of high potential storage quality.

2.2.2 Postharvest Factors

Harvested fruits are still living organisms continuing to respire and lose water. They, therefore, suffer detrimental changes after harvest (Burdon, 1997; Kader, 1992a). Rapid and uncontrolled ripening after harvest is a cause that triggers utilization of energy reserves through respiration, changes in biochemical composition, changes in texture associated with both water loss and biochemical change and increased C_2H_4 production (Burdon, 1997). Nakasone and Paull (1998) further stated that postharvest life terminate because of physiological, mechanical and pathological stress with associated symptoms, such as excessive water loss, bruising, skin scald, failure to ripen, and decay. The rate at which changes occur in harvested fruits may be influenced by range of factors including biological (internal) and environmental (external).

2.2.2.1 Biological (Internal) Factors

Biological factors are endogenous factors that affect postharvest condition of the fruit. They can include the respiration rate, C_2H_4 production, compositional change and/or metabolic activity, transpiration, moisture loss and physiological breakdown. Improper handling and storage of fruits after harvest further enhances biological postharvest loss.

a. Respiration: Several workers (Kader, 1992a; FAO, 1989; Wilson *et al.*, 1999) defined respiration as the process by which stored organic material (CHO, protein, and fats) are broken down into simple end products with a release of energy. Increased respiration means loss of stored food reserves leading to speeding up of senescence processes, reduce food value for the consumer, loss of flavour, especially sweetness and loss of salable dry weight and rapid deterioration (Kader, 1992a; Wilson *et al.*, 1999) it also causes temperature to rise up and rate of growth of pathogens are accelerated (Kays, 1991). Fruits can be classified into two groups, depending on their respiration pattern, as climacteric and non-climacteric (Table 2.1). The former can be defined as fruits which can be ripened after harvest in response to C_2H_4 and have a peak in respiration rate during ripening and, in the latter case, fruit ripening is protracted and the attainment of the ripened state is not essentially associated with a significant increase in

respiration and/or C_2H_4 production (Harris, 1988; Sivalingam and Charles, 1995; Thompson, 1996). In other words the ripening of non-climacteric fruit is considered to be C_2H_4 independent process and little is known of regulatory mechanisms underlying the biochemical changes (Leliévre *et al.*, 1997). In climacteric fruits however, ripening is associated with an increase in production of C_2H_4 and C_2H_4 treatment can accelerate ripening (Burg and Burg, 1962; Salunkhe and Desai, 1984a).

Table 2.1 Classification of fruits and fruit vegetables based on climacteric and non-climacteric ripening patterns

	CLIMACTERIC	NON-CLIMACTERIC
TEMPERATE FRUIT	Apple Pear Peach Apricot Plum	Cherry Grape Strawberry
'VEGETABLE' FRUIT	Melon Tomato Watermelon	Cucumber
COMMON TROPICAL AND SUBTROPICAL FRUIT	Avocado Banana Mango Papaya Fig Guava Passion fruit Persimmon	Orange Grapefruit Lemon Lime Olive Pineapple Litchi
LESS COMMON TROPICAL AND SUBTROPICAL FRUIT	Cherimoya Soursop Breadfruit Jackfruit Mamey apple Sapote	Cashew apple Java plum Other Eugenia sp

Adapted from Harris (1988)

The study of factors influencing the ripening of fruits is hampered not only by the irregular distribution of applied materials through the bulky fruit structure, but also by the lack of uniform behaviour among different climacteric fruits (Venderell, 1985). Climacteric fruits, exemplified by papaya, banana and avocado, undergo upsurge in respiration and C_2H_4 evolution concomitant with ripening (Frenkel *et al.*, 1975). Furthermore, banana and papaya, being both climacteric fruits, exhibit a ripening respiratory pattern involving the processes of pre-climacteric,

climacteric rise, climacteric peak and post-climacteric periods in the production of carbon dioxide (CO_2) and oxygen (O_2) (Sivalingam and Charles, 1995). In banana, for instance, within a couple of days, a respiratory rate of about 20 in the hard green fruit may rise to about 125 at climacteric peak and then fall to about 100 ml CO_2 $kg^{-1}hr^{-1}$ as ripening advances (Kotecha and Desai, 1995). It is important to note that in general, fruits with the highest respiration rates, such as banana and avocado, tend to ripen most rapidly and hence are most perishable (Tucker, 1993) therefore, the need to lower the respiration rate is more critical (Shewfelt, 1986). The rate and period of banana respiration depends upon environmental conditions such as temperature, RH and C_2H_4 concentration (John and Marchal, 1995). Several workers reported that decreasing the storage temperature of papaya fruit above the critical limit for CI, reduced respiration rate, C_2H_4 production (Lam, 1990) and ripening (Lazan *et al.*, 1993). The respiration rate of stored product can be used as an indicator for adjusting the storage conditions to maximize the longevity of the commodity. Controlling or modifying these factors to regulate respiration as a possible target is, therefore, essential to ensure the safety and quality of the produce.

b. Ethylene (C_2H_4): Fruit ripening is a coordinated series of biochemical changes that renders the fruit attractive to eat. This process is under genetic regulation, but plant hormones play an essential role (Vendrell and Palomer, 1997). Ripening in climacteric fruits is accompanied by evolution of C_2H_4. This implies the involvement of C_2H_4 in the ripening process (Burg and Burg, 1962). C_2H_4 is a colourless gas with a faint sweetish smell that is the naturally produced ripening hormone of most fruits (Jobling 2000), and also involves in the regulation of many aspects of growth development and senescence (Kader, 1992a; Yang *et al.*, 1986). C_2H_4 is essential for the initiation of fruit ripening, although other factors are also believed to be involved (Salunkhe and Desai, 1984a). All fruits produce C_2H_4, but climacteric fruits are characterized by an enormous increase in production (John and Marchal, 1995; Kays, 1991) as well as increase in respiration rate (Jobling, 2000). C_2H_4 is synthesized from the sulfur-containing amino acid methionine. Methionine is first converted to S-adenosyl methionine (SAM) and then to the 4-carbon compounds, 1-amino-cyclpropane-1-carboxylic acid (ACC) via the enzyme ACC synthase. ACC has two fates, one is the conversion to malonyl ACC (which is an inactive endproduct) and the other is the conversion to C_2H_4 via the enzyme ACC oxidase (Ethylene Forming Enzyme, EFE) (Kader, 1992a). It was first shown in apple tissue to be biosynthesized by the following pathway (John and Marchal, 1995)

Methionine → S-adenosylmethionine (SAM) $\xrightarrow{\text{ACC synthase}}$ 1-aminocyclopropane-1-carboxylic acid (ACC) $\xrightarrow{\text{EFE}}$ Ethylene.

The excess production of C_2H_4, above the threshold level, brings about changes in fruit physiology from maturation to ripening. Knee (1984) and Lau *et al.* (1985) have examined the change in ACC content during the ripening of apple. Hoffman and Yang (1980) also reported that during the onset of ripening the ACC content, the C_2H_4 production and the capacity to produce C_2H_4 are greatly enhanced, indicating an increase in the amount or activity of ACC synthase and EFE. The authors added that the ACC content in mature, but unripe apple was very low (about 0.1 $nmolg^{-1}$) but a massive increase occurred when vigorous C_2H_4 production commenced. It has been well documented that the concentration of C_2H_4 required to induce ripening of pre-climacteric fruit varies with different species. Banana, for instance, may require as little as 0.1 μlL^{-1}. Therefore, regulation of C_2H_4 biosynthesis and action is very important in agriculture. It has been shown that the production of C_2H_4 can be reduced by decreasing temperature, reducing O_2, increasing CO_2 treating with inhibitors (aminoethoxy-trans-3-butenoicacid, aminovinyl glycene, (AVG), cobalt chloride, and PGRs (GA_3, cytokinins and /or auxin). C_2H_4 can also be removed by using a number of chemical processes like $KMnO_4$, Ozone (O_3) and 1-methylcylcopropene (1-MCP). The application of 1-MCP has been shown to be highly effective inhibitor of C_2H_4 action (Wills *et al.*, 1998) and significantly delayed and suppressed the onset and magnitude of banana fruit respiration and volatile production (Golding *et al.*, 1999). These authors also added that inhibition of C_2H_4 sensitivity with 1-MCP during the early stage of the climacteric produces changes in subsequent ripening behaviour which reflect the disruption of C_2H_4. A substantial retention of firmness and dramatic reduction of superficial scald in apples treated with 1-MCP has been reported by Weis and Bramlage (2002). It has been shown to bind irreversibly to the C_2H_4 receptors and low concentration treatments for few hours confers protection against C_2H_4 sensitive tissues (Wills *et al.*, 1998) and leaves virtually no residues (Huber *et al.*, 2003).

c. Transpiration/Water loss: Water loss is one of the main factors that cause fruit deterioration and reduce the marketability after harvest. Plants lose water by transpiration via their stomatal and other natural openings such as lenticels, cuticles, epidermal cells and/or trichomes (hairs). Fruit transpiration continues after harvest (Salunkhe *et al.*, 1991), but there are no means to replenish the transpired water; hence the fruit continues to deteriorate. Quick cooling is therefore necessary after harvest to preserve the freshness of fruits. Fruits that have lost 5-8% of their fully

turgid initial weight begin to show signs of mass loss and become shriveled (Wilson *et al.*, 1999). Usually, the initial sign is skin wrinkling although skin discolouration can also be the first symptom in some fruits (Nakasone and Paull, 1998). The rate of transpiration, which must be minimized to avoid loss in salable weight, wilting and shriveling of produce, can be controlled by good handling at recommended humidity and temperature (Hardenburg *et al.*, 1980). Postharvest water loss is dependent upon the commodity, cultivar, preharvest conditions, ambient temperature water-vapor pressure deficit (WVPD), wounds, postharvest heat treatment, the presence of coating or wrap (Nakasone and Paull, 1998), RH and air velocity (Wilson *et al.*, 1999). Ryall and Pentzer (1982) reported that the skin of some fruits develop waxes after harvest that gives an attractive appearance and aid in reducing moisture loss after harvest. Apart from this the surface area to volume ratio (Kader, 1992a) and stomatal density (Ferris, 1998) of fruit affects the rate of water loss. As banana fruit ripens water is lost from the peel because of a threefold increase in evaporation rate, and water is believed to flow from the peel to the pulp in response to changes in the osmotic potential (Turner, 1997). These various water losses enhance postharvest fruit deterioration and retard fruit shelf-life. Transpiration is a physical process that can be controlled by applying treatments to the fruits (e.g., waxes and wrapping with plastic films) or by manipulating the environment surrounding the fruit (Kader, 1992a).

d. Compositional changes: Compositional changes include alteration in concentration and composition of CHO, pigments, phenolics, acids, and volatile chemicals associated with flavour (Thompson and Burden, 1995). A large number of physiological and biochemical changes occur during the ripening of fruits. The change in fruit physiology from maturation to ripening is initiated when the cellular quantity of C_2H_4 reaches a threshold level (Hoffman and Yang, 1980). Compositional changes of fruit are usually related to maturation and ripening. During ripening the fruit softens, starch is converted to sugars (banana), the skin colour changes; it loses its astringency (due to tannin polymerization) and develops characteristic flavour (Frenkel *et al.*, 1975; Thompson and Burden, 1995), as well as, there is modification or partial solubilization of the cell wall (Grierson *et al.*, 1981). The relative changes in weight, sugar, chlorophyll, and acidity are common to most fruits, but other parameters such as respiration, flavour, aroma, and carotenoids can vary from commodity to commodity (Burdon, 1997).

Softening: Changes after harvest take place in the cell wall composition and structure resulting in softening of fruit (Ryall and Pentzer, 1982). The decrease in fruit firmness is a general feature that accompanies ripening of both climacteric and non-climacteric fruits (Biale and Young,

1981). The change in firmness can be partly attributed to the starch and sugar changes and the breakdown of pectic substances by pectinase (Anon, 2001a; Babbitt *et al.*, 1973; Biale and Young, 1981; Seymour *et al.*, 1990). In papaya fruit, however, softening of fruit mesocarp and endocarp is due to the activity of cell wall degrading enzymes, not due to starch degradation, as the fruit has not accumulated/manufactured starch during ontogeny (Paull and Chen, 1983). According to Paull (1993) papaya fruit soften to an edible stage in 6-12 days. During compositional change of fruits certain sugars, e.g., galactose and arbinose, are lost from the cell wall (Hobson, 1981; Paull and Chen, 1983; Seymour *et al.*, 1990). This leads to a loss of cell wall strength and inter cell adhesion. During fruit ripening the initiation of polygalacturonase (PG) activity results in a change in pectic substances from water insoluble (protopectin) to soluble pectins (Hobson, 1981). Similar result was reported on banana fruits where changes in pectin structure during ripening indicated that the activity of pectin degrading enzymes such as PG and pectin methyl esterase (PME) increased in cell walls (Seymour, 1993). It has been reported that the central cavity of papaya fruit can develop a negative pressure which is probably associated with changes in flesh gas transfer as it becomes water-soaked due, presumably, to loss of cellular compartmentation (Paull, 1993). It has been demonstrated that there is a close link between firmness of fruit and PG activity in the fruit (Babbitt *et al.*, 1973; Lazan *et al.*, 1990a). Hence, it is the activity of PG that brings about remarkable fruit softening after harvest.

Pigmentation: The plant world is dominated by the colour green, which is the result of the presence of the chlorophyll pigments (Kays, 1991). For many fruits the first sign of ripening is the disappearance of the green colour with degradation of chlorophyll. It is one of the common symptoms of senescence in harvested produce (Yamauchi and Watada, 1991). Terblanche (1999) reported that colour break is a stage of maturity where the green fruit turns to yellow or orange. In general colour changes are associated with ripening and represent a key attribute, along with texture, for the determination of eating quality (Seymour, 1993). Many products undergo significant changes in their pigment composition during both, the pre and postharvest period, particularly in the final stage of maturation and ripening. Although the trigger for chlorophyll degradation is not yet clear, many believe that there is an involvement of both enzymes and chemical reaction. Chlorophyllase breaks down the chlorophyll and this causes the expression of other pigments, which are already present in the fruit. A maximum activity of chlorophyllase was observed in apples and banana at the time of the climacteric. The second important pigment is carotenoid, which synthesized concomitant to ripening in most fruits (but

not in banana). Carotenoids are a large group of pigments associated with chlorophyll in the chloroplasts and are also found in the chromoplast (Kays, 1991). They are divided in to two subgroups: the carotenes and their oxygenated derivatives, the xanthophylls. Carotenoids are stable compounds and thus remain intact in the tissue even when excessive senescence has occurred (Wills *et al.*, 1998). The change in colour of ripening banana fruit is associated with the breakdown of chlorophyll with the already presented carotenoids level remaining relatively constant (Mattoo *et al.*, 1975; Seymour, 1985 cited by John and Marchal, 1995). However, an increase in carotenoid level was reported during ripening of papaya fruit and pepper (de Guevara *et al.*, 1996). The principal agents responsible for the chlorophyll degradation are pH change (mainly due to leakage of organic acids from the vacuole), chlorophyll oxidative system, chloropyllase (Wills *et al.*, 1998) light, temperature (Kays, 1991) and cultivar (Robbins and Moore, 1990). Within the natural environment both, high and low temperatures have been implicated in destruction of chlorophyll (Hendry *et al.*, 1987). Similarly, several growth regulators have been shown to have a significant effect on the pigmentation of some harvested produce. Cytokinins and GA_3 were reported to retain chlorophyll (Kays, 1991). PGRs by retarding fruit ripening and senescence process in fruits maintain the green colour for a longer period (Barkai-Golan, 2001).

Carbohydrates (CHO): CHO are the most abundant and widely distributed food components in nature. They can be classified as monosaccharides, oligosaccharides, and polysaccharides. The largest quantitative change associated with fruit ripening is usually the breakdown of CHO polymers, especially the near total conversion of starch to sugars (Hubbard *et al.*, 1990; García and Lajolo, 1988; Wills *et al.*, 1998). Starch granules and pectins could have a structural function in the cell wall (John and Marchal, 1995) as a result fruit softening starts after these chemicals are changed. The average starch of banana can be 22-25% the fresh weight in the pre-climacteric phase and less than 1% in the climacteric phase (Chang and Hwang, 1990; Hubbard *et al.*, 1990). This transformation of starch to sucrose during ripening of banana involves several enzymes and more than one pathway (Cordenunsi and Lajolo, 1995). Garcia and Lajolo (1988) detected three α- and four β-amylase and α-1,4-glucosidase and α-1,6-glucosidase activities in all stage of banana fruit ripening. Similarly respiration rate of the produce is also another factor for the transformation. Many workers reported that the accumulation of sucrose in the fruit preceding the increase of glucose and fructose as the starch content decreases is concomitant

with ripening. Starch-sugar transformation rate can therefore be used as good indication of fruit ripening.

Organic acids: The pattern of organic acids changes in fruits is concomitant with ripening. The major chemical compounds found are ester of aliphatic alcohols and short-chain fatty acids (Salunkhe *et al.*, 1991). In most fruits an increase in sugar content and decrease in acidity is evident during ripening (Akamine and Goo, 1971; Illeperuma and Jayasuriya, 2002). Acid can be considered as a reserve source of energy to the fruit, and would therefore be expected to decline during the greater metabolic activity that occurs with ripening (Wills *et al.*, 1998). However, in case of banana, the highest level of acid is attained when the fruit is fully ripe. The sugar and acid content have a marked influence on the sensory quality of the fruit (Ackermann *et al.*, 1992). The main acids in banana are citric and malic and/or oxalic acid (Inaba and Nakamura, 1988) although malic acid has been identified as the main acid. The level of malic and citric acids normally increases during ripening to a certain level (Wills *et al.*, 1998), whereas oxalic acid is metabolized and decreased. In papaya the predominating acids are malic and citric acids, the presence of tartaric, malonic, fumaric and succinic acids was also noted (Sankat and Maharaj, 1997). The level of organic acids in a fruit can markedly affect its taste. Fig 2.1 summarizes the general compositional changes during the fruit ripening process.

e. Physiological disorders: According to Sankat and Maharaj (1997), a physiological disorder refers to breakdown of tissue that is not caused by invasion of pathogens or mechanical damage. It is usually a result of undesirable temperature exposure after harvest. Deterioration of fresh commodities can result from physiological breakdown due to natural ripening processes, water loss, and temperature injury (Wilson *et al.*, 1999). Kader (1992a) reported that freezing, chilling, and heat injuries are the three main physiological disorders that cause postharvest loss. The rate and degree of disorder varies depending on the environmental conditions, nature of the fruit and harvesting time. CI is a major physiological disorder that occurs in many plants and plant products as a result of their exposure to low but non-freezing temperatures (Jackman *et al.*, 1990; Salunkhe *et al.*, 1991; Sankat and Maharaj, 1997). CI mainly injures the peel, killing certain cells in banana (Hardenburg, 1975). Usually this injury becomes problematic in tropical fruits like banana and papaya when stored at low temperatures (Salunkhe *et al.*, 1991). Symptoms of CI are often apparent only after removal of fruits from chilling temperature (Jackman *et al.*, 1990). Generally, effect and incidence of CI increase proportionately with the decrease in storage temperature and an increase in length of storage period.

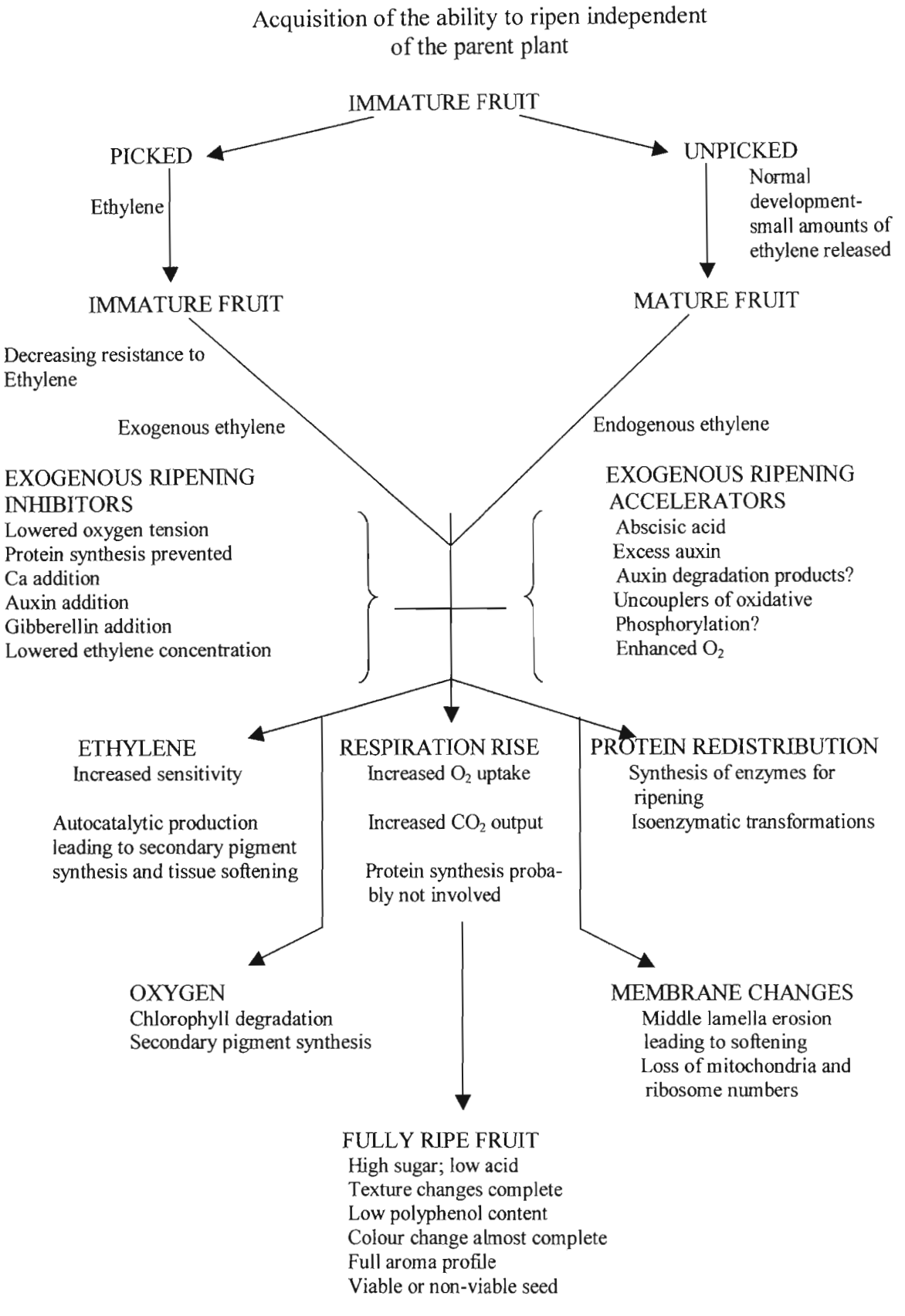


Fig 2.1 The tentative mechanism for ripening of climacteric fruit (Adapted from Hobson, 1979).

2.2.2.2 Environmental Factors

The environmental condition to which the harvested fruit is exposed affects the storage potential and shelf-life of the fruit considerably. Of the different factors that affect the shelf-life of produce after harvest temperature, exogenous C_2H_4, RH, and atmospheric composition are the most common.

a. Temperature: Temperature is the environmental factor that mostly influences the deterioration rate of harvested commodities (Kader, 1992a; Kays, 1991; Shewfelt, 1986; Wilson *et al.*, 1999). Higher temperatures after harvest means quicker spoilage and greater waste. Fresh products exposed to extremes of heat or cold sustain serious physiological damage leading to rapid deterioration (Wilson *et al.*, 1999). An increase in temperature increases the depletion of energy reserves necessary for normal cellular activity, with resultant loss of water and breakdown caused by moulds and other rots causing decay (Harris, 1988) and hence short shelf-life (Fig 2.2). For each increase of 10°C above optimum there is a two to three fold increase in the deterioration rate (Kader, 1992a). Cooling the produce after harvest is therefore very important to extend the shelf-life by slowing down the rate of breakdown and thus maintaining the produce quality. Physiological studies of banana, for instance showed that storage life decreases as external temperature increases over the range from 15 to 35°C. Similarly, shelf-life of papaya fruit at an ambient tropical environment (25-30°C) was only seven days (Maharaj, 1988). On the other hand, Satyan *et al.* (1992b) reported that the shelf-life of banana fruits significantly doubled as the storage temperature dropped from 30 to 13°C. Lebibet *et al.* (1995) reported that banana fruit at 13°C resulted in lower respiration rates, but full ripening with a good colour and excellent flavour. However, precaution must be taken as certain fruits exhibit damage when stored at a temperature lower than optimum. Especially tropical fruits like banana and papaya, having evolved in warmer climates, cannot tolerate low temperature during storage. Banana shows symptoms of CI at a temperature below 12°C, while papaya can be stored at temperatures as low as 7°C (Table 2.2), although Medlicott (2001) reported that storage of papaya below 10°C results in CI. According to Paull (1993) papaya fruits at the phase of colour changing could be stored at temperature as low as 7°C for less than 14 days and would ripen normally. Kader (2000ab) reported that the optimum storage temperature of banana and papaya is 13-14°C and 7-13°C, respectively. There are, however, reports that indicate papaya fruit kept in refrigerated storage are susceptible to fungal decay (Sankat and Maharaj, 1997).

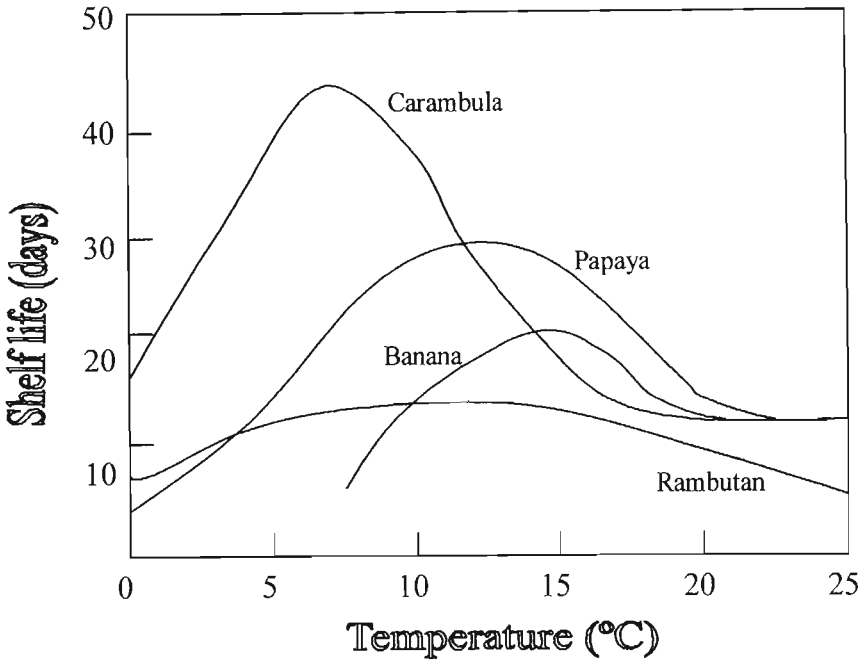


Fig 2.2 The relationship between shelf-life and storage temperature of fruits. (Adapted from Nakasone and Paull, 1998).

Table 2.2 Susceptibility of fruits and vegetables to chilling injury at low but non-freezing temperatures

Commodity	Approximate lowest safe temperature °C	Chilling injury symptoms
Avocados	5-13	Grey discoloration of flesh
Bananas (green/ripe)	12-14	Dull, gray-brown skin colour
Cucumbers	7	Pitting water-soaked spots, decay
Grapefruit	10	Brown scald, piking, watery breakdown
Lemons	13-15	Pitting, membrane stain, red blotch
Limes	7-10	Pitting
Mangoes	10-13	Grey skin scald, uneven ripening
Melons: Honeydew	7-10	Pitting failure to ripen, decay
Watermelon	5	Pitting, biker flavour
Okra	7	Discoloration, water-soaked areas, piking
Oranges	7	Pitting brown stain, watery breakdown
Papaya	7	Pitting failure to ripen, off-flavour, decay
Pineapples	7-10	Dull green colour, poor flavour
Potatoes	4	Internal discoloration, sweetening
Pumpkins	10	Decay
Sweet peppers	7	Pitting *Alternaria* rot
Sweet potato	13	Internal discoloration, piking, decay
Tomatoes: Mature green	13	Water-soaked softening, decay

Adapted from Lutz and Hardenburg (1966)

b. Exogenous ethylene (C_2H_4): While C_2H_4 is invaluable due to its ability to initiate the ripening process in several fruits, it is not always beneficial, especially in terms of postharvest shelf-life as it accelerates the ageing processes and decreases the quality of the product (Jobling, 2000). Ferris (1998) reported that C_2H_4 determines the time between harvest and senescence. The author also added that exogenous C_2H_4 application would shorten the pre-climacteric period; at high concentration, C_2H_4 causes rapid initiation of the climacteric respiratory response and accelerates ripening. When C_2H_4 is applied to a climacteric fruit, at a concentration of 0.1-1.0 ppm, for 12 hrs or more, ripening starts (Reid, 1992). The main source of C_2H_4 during storage or marketing is usually from other ripening fruits in the storage. It can also be emitted from exhaust gas of petrol combustion engine vehicles or forklift trucks around the fruits (Jobling, 2000) all produce C_2H_4 in amounts large enough to cause damage to the C_2H_4 sensitive fruits.

c. Relative humidity (RH): RH is the second most important environmental factor after temperature in the shelf-life of fruits (Shewfelt, 1986). It is the amount of water vapour present in the air, relative to the maximum amount of water vapour that can be held in the air, at a given temperature, saturated air being 100% RH. The rate at which water is lost from the plant part depends on the difference between the water vapour pressure inside the plant and the pressure of water vapour in the air (FAO, 1989). Hence ambient water vapour affects the rate of water loss from the produce. Ferris (1998) reported that banana stored at an ambient RH of 95-100% loses little or no moisture, and ripening period is extended. The optimum RH of banana and papaya is 90-95 % (Kader, 2000ab). Increasing the humidity in storage is correct theoretically, but very high RH (>95%) enhances fungal disease attack to the produce, therefore correct compromise is important. Low RH environments reduce the spread of most decay organisms at the cost of increased transpiration and, thus, increased moisture loss (Shewfelt, 1986). Produce can be protected from lower RH by using various types of permeable polyethylene bags and films (Salunkhe *et al.*, 1991).

d. Atmospheric composition: Naturally the atmospheric composition around the fruit is composed of 78% N_2, 21% O_2 and 0.03% CO_2. Modification of these proportions can affect the shelf-life of the fruit significantly. The composition of the gaseous atmosphere to which postharvest product are exposed can markedly influence both respiratory and general metabolic rate of the commodity (Kader, 1992a; Kays, 1991). Increased CO_2 and decreased O_2 level

inhibits respiration rate (Phan *et al.*, 1975) and suppresses C_2H_4 production (Lazan *et al.*, 1990a) and thus delay fruit ripening (Hardenburg *et al.*, 1980; Shewfelt, 1986). Furthermore, CA with high CO_2 inhibits breakdown of pectic substances and retains fruit texture and firmness for longer periods, and retention of flavour may be improved (Salunkhe and Desai, 1984a). Striking the balance between O_2 and CO_2 is critical and an optimum ratio must be developed for each specific fruit. Optimum CA storage of banana and papaya are as follows: 2-5 % O_2 and 2-5 % CO_2 for banana and 3-5% O_2 and 5-8 %CO_2 for papaya (Kader, 2000ab). Care must be taken during storage as poor ventilation of produce leads to the accumulation of CO_2 around the produce. This in turn will cause bad flavours, internal breakdown, failure to ripen and other abnormal physiological conditions (FAO, 1989). On the other hand, if O_2 is too low (excessively depleted), anaerobic respiration may occur. Exposing banana to <1% O_2 and /or >7% CO_2, for instance, may cause undesirable texture and flavour (Kader, 2000a), whilst O_2 below 2% and/or CO_2 level above 8% causes off flavour development and uneven ripening in papaya (Kader, 2000b).

e. Mechanical / Pathological damage: Mechanical injuries causing bruising lead to water loss, fungal infection and stimulate respiration and C_2H_4 production leading to loss in quality (Kader, 1992a; Phan *et al.*, 1975). Bruise and mechanical damage not only detract from the good appearance of the fruit, but are also good avenues for the entrance of decay organisms (Hardenburg *et al.*, 1980). Disease attack of fruits after harvest represents one of the severe sources of postharvest loss (Swinburne, 1983). After harvest, most fruits lose the ability to repair damaged peels and become a means of pathogen entry. Fruits characterized by high moisture content and relatively soft peel, are highly susceptible to mechanical damage throughout their postharvest period (Burdon, 1997). Papayas, for instance, are more susceptible to mechanical damage than other fruits because of their soft texture and moisture content (Salunkhe and Desai, 1984a). Hence, decay pathogens can easily enter if proper handling is not undertaken. Banana and papaya are attacked by a variety of surface and internal postharvest fungi and bacteria, which include *Colletotrichum sp., Mycosphaerella sp., Phomopsis sp., Alternaria sp., Fusarium sp., Asschochyta sp.* (Coates *et al.*, 1995; Jones and Wade, 1995; Swarts, 1985; Venter, 1993). PGRs have been used to control fruit decay after harvest. According to Barkai-Golan (2001), PGRs may, similar to low temperatures, suppress decay development indirectly, by retarding ripening and senescence processes in fruits and maintain the natural resistance of young tissue. This author added that GA_3 application after harvest delay the senescence and softening of rind

of Navel oranges. Moreover, Ben-Yehoshua (1995) indicated that dipping lemon fruits in GA_3 (50 or 100 ppm) or 2,4-dichlorophenoxyacetic acid (2,4-D) of 200 ppm prior to storage retards the decrease in the antifungal citral compound in the flavedo and results in reduced fruit decay.

2.3 Loss Reduction Techniques

An understanding of the control of ripening is important because it has implications for fruit storage and quality. Different methods and chemicals have been used for control and reduction of postharvest loss. Chemicals are applied to the crops after harvest to control microorganisms and pest infestations, to correct nutrient imbalances in the crop, which may shorten their storage life or cause physiological disorders, and to prevent sprouting and re-growth of crops. Apart from this, supplemental treatments like CA, modified atmosphere (MA), irradiation, film packaging, waxing and chemicals are among the most common (Wang, 1999). The detrimental effect of using CA in storage of fruit and chilling injuries caused by exposing fruit to temperature extremes (Charles and Tung, 1973) on the one hand and lack of storage facilities in developing countries on the other, however, have resulted in an increased realization of the need for alternate methods of regulating ripening and senescence of fruits. The outlook for this is promising because there are certain chemicals that are capable of delaying ripening, while others accelerate the ripening, if needed. The following are some of the commonly used postharvest loss minimization techniques, which may be used.

2.3.1 Plant Growth Regulators (PGRs)

Understanding the role of these PGRs in the regulation of fruit ripening still represents a major challenge to the postharvest physiologist. Information on the effect of these compounds applied postharvest is extensive only for C_2H_4 (Wills *et al.*, 1998). However, recent advances in the knowledge of the biosynthesis and action of C_2H_4 now provide a more rational basis on which to interpret the effect of certain types of exogenously applied PGRs (Sharples and Johnson, 1986). Naturally occurring plant growth substances, their chemical analogs and antagonists, most certainly play a role in growth and development, and therefore must in some way influence the development of ripening capacity (Dilley, 1969; Vendrell and Palomer, 1997). Similarly, Frenkel *et al.* (1975) considered the concept of fruit ripening as a directed phase of plant development, and suggested an involvement of growth substances in the coordination and expression of the process. The effects of these compounds on tissue physiology, however, are

usually complex, making generalizations difficult (Shewfelt, 1986). Although their mode of action is not well known, ABA, auxins, cytokinin and GA_3 may play an important role. It has been reported that PGRs can affect the activity of cell wall degrading enzymes such as PG, PME, cellulase, β-1,3-glucanase and amylase (Babbitt *et al.*, 1973). As they occur naturally, they might be expected to receive consumer acceptance as potential food additives (Wills *et al.*, 1998).

2.3.1.1 Auxin

Auxins are growth substances that are produced in significant quantities in the upper growth region of plants. According to Frenkel (1975) the role of endogenous auxin in C_2H_4 biosynthesis is not fully understood, but there is some evidence that IAA may be the ripening inhibitor in apple and pears, which some workers claim to be present in the tree. The method of application and the resultant distribution of auxin in the tissue may influence the effect on ripening. It was found that vacuum–infiltration of banana fruit slices with 2,4-D or IAA delayed the onset of ripening, although C_2H_4 production was stimulated (Vendrell, 1969). The effect of IAA and 2,4-D at concentrations of 0.01, 0.1 and 1.0 mM on softening, degreening, C_2H_4 and CO_2 evolution were studied. Softening and degreening were inhibited increasingly in response to increasing concentrations of IAA (Babbitt *et al.*, 1973) in tomato and (Frenkel and Dyck, 1973; Vendrell and Palomer, 1997) in different climacteric fruits. Similarly, 2,4-D is believed to prevent abscission of the stem end button of the citrus fruits which if it removed or abscised results in easy penetration of fungus like *Diplodia* and *Alternaria* that cause stem end or core rot in the fruit (Wills *et al.*, 1998). Contrasting results obtained by different types of application of auxin (dipping or spraying versus vacuum infiltration) can be explained by limited penetration of auxin into the tissue when applied by dipping (Vendrell and Palomer, 1997). Not only the application method but also the developmental stages of the fruit at the time of application (Dilley, 1969) and concentration (Babbitt *et al.*, 1973) have a pronounced effect upon response. Recent studies support the previous suggestion that auxin functions as an inhibitory factor in fruit ripening. However, the use of auxin is limited since, in high concentration, it might accelerate C_2H_4 production and thus enhance the ageing process (Barkai-Golan, 2001).

2.3.1.2 Gibberellic Acid (GA_3)

Postharvest dipping of banana in aqueous solutions, of GA_3 delays colour and aroma development, retard chlorophyll degradation, ascorbic acid decrease and the decline in activity of α-amylase and peroxidase (Dilley, 1969; Khader, 1992; Salunkhe *et al.*, 1991; Vendrell and Palomer, 1997). Similarly, Desai and Deshpande (1978) and Kotecha and Desai (1995) reported that an increase in firmness, starch, cellulose, and hemicellulose contents was observed when banana was treated with GA_3 or kinetins. Postharvest treatment of papaya fruit with GA_3, vitamin K, silver nitrate and cobalt chloride were found to extend shelf-life without any adverse effect on palatability (Mehta *et al.*, 1986). According to Khader (1992), a significant delay in the ripening of mango fruit was observed when GA_3 was applied with or without vapourgard. The effect of GA_3 on ripening is indicated by a lowering of respiration rate, retarded climacteric and delayed colour change (Barkai-Golan, 2001; George and Marriott, 1985; Rao and Chundawat, 1990; Salunkhe *et al.*, 1991). Banana fruits treated with GA_3 exhibited delayed ripening measured by the hydrolysis of starch and subsequent production of sugars (Rao and Chundawat, 1990). Furthermore, improved storage has been reported by postharvest GA_3 dip of banana in 500 ppm (Salunkhe and Desai, 1984a) and the sub-tropical pear cultivar "Le Conte" in 100 ppm (Visai *et al.*, 1980 cited by Sharples and Johnson, 1986). It is also reported that GA_3 acts against ABA, which promotes senescence. Generally speaking, GA_3 and C_2H_4 have opposing effects; GA_3 defers senescence in leaves and fruits, whereas C_2H_4 enhances the development of senescence. In line with this GA_3 was found to suppress PG activity with increased cellulase activity in tomato (Babbitt *et al.*, 1973). All the above reports suggested that there is a potential to decrease ripening by using GA_3.

2.3.1.3 Cytokinins

Another crop or plant hormone believed to be a retardant of fruit ripening is cytokinin. Senescence in leaves can generally be delayed by cytokinin treatment and fruit ripening can be regarded as a senescence phenomenon (Vendrell and Palomer, 1997) and thus in several instances kinetins have been advocated as anti-senescence (Rao and Chundawat, 1990). Cytokinin retards chlorophyll degradation by maintaining high protein in the treated tissue (Salunkhe *et al.*, 1991). It was observed that a high level of accumulated endogenous cytokinin delays fruit ripening and the level of this compound declines as ripening proceeds. Exogenous

application of cytokinin can reduce aging in plants and improves the shelf-life of fruit. The activities of catalase and peroxidase in banana fruits were retarded after application of kinetins hence fruit shelf-life increased (Rao and Chundawat, 1990). It has been reported by Dilley (1969) that benzyl adenine retards the rate of ripening of apricots when applied as a postharvest treatment. This effect is in agreement with the kinetin-induced delay of senescence observed in many excised tissues.

In general, PGRs may be of special importance mainly for fruits that cannot be stored at low temperature because of their cold sensitivity, as well as in developing countries where cold storage cannot be applied (Barkai-Golan, 2001).

2.3.2 Potassium Permanganate ($KMnO_4$)

Apart from the application of growth regulators other methods are used which modify the concentration or biosynthesis of C_2H_4. Exogenous application of C_2H_4 induces fruit ripening by promoting the synthesis of various enzymes, respiration and production of C_2H_4 in banana (Rao and Chundawat, 1990). These authors, therefore, reasoned that ripening would be delayed when the ambient levels of C_2H_4 in the fruit package is reduced by insertion of suitable absorbent. C_2H_4 in the storage atmosphere can be oxidized to CO_2 and H_2O using a range of chemical agents (Wills *et al.*, 1998). $KMnO_4$ is quite effective in reducing in-package C_2H_4 levels (Chamara *et al.*, 2000; Fuchs and Temkin-Gorodeiski, 1971; Hewett *et al.*, 1989). The use of $KMnO_4$ absorbed on aluminum silica or vermiculite in the bags prolonged the postharvest life of the different fruits sealed in polyethylene bags (0.1 mm thickness) (Yahia, 1998). It was reported earlier that the ripening time of banana is extended by five weeks after treatment of fruits with $KMnO_4$ (Kotecha and Desai, 1995; Salunkhe and Desai, 1984a) and persimmons fruit could be stored for four months with out losing appreciable quality (Pekmezci *et al.*, 1997) it also prevents injury from high levels of CO_2 or depletion of O_2 of banana fruit (Liu, 1970). Similarly, bananas stored in film bags containing $KMnO_4$ were found to be firmer than control fruits and hence the storage life could be extended for another two weeks (Salunkhe *et al.*, 1991; Scott and Gandanegara, 1974). To ensure efficient destruction of C_2H_4, a large surface area of $KMnO_4$ is achieved by coating or impregnating an inert inorganic porous support, such as active aluminia carrier (Al_2O_3) or expanded mica, with saturated solution of $KMnO_4$ (Thompson, 1996; Wills *et al.*, 1998). Proprietary products such as "ethysorb" and "purafil" are available commercially

which utilize the ability of $KMnO_4$ and can be placed inside the store or even inside the actual package containing the crop to oxidize C_2H_4 (Reid, 1992). The author also added that many porous and readily permeable to gas materials have been used to manufacture permanganate absorbers, including vermiculite, pumice, and brick. Since it is non-volatile, $KMnO_4$ can be physically separated from produce, thus eliminate the risk of chemical injury (Wills *et al.*, 1998). However, high humidity in the storage is limitation to the longevity of $KMnO_4$ absorbents as it also reacts with water.

2.3.3 Waxing

Another alternative method of extending the postharvest shelf-life of certain fresh fruits, without the use of refrigeration, is waxing (Salunkhe and Desai, 1984a). Fruits dipped or sprayed after harvest with waxing materials shown improved appearance or a delay in deterioration and hence increased the shelf-life of the fruit. Surface coatings have been used extensively on bulky commodities to modify internal atmosphere composition and thereby delay ripening (Banks, 1984; Banks *et al.*, 1997; Smith *et al.*, 1987) and reduce water loss (Hagenmaier and Baker, 1993). In nature, a protective wax shield covers most plants, flowers and fruits (Akamine *et al.*, 1975; Anon, 2001b) and forms an important interface between a plant and its environment (Jenks and Ashworth, 1999). Plant waxes are extremely important during the postharvest storage and marketing of the plant products in that they function by limiting the water loss from the tissue (Durand *et al.*, 1984; Meheriuk and Porrit, 1972; Sankat and Maharaj, 1997) and impeding invasion of pathogens (Kays, 1991). Furthermore, thin, edible wax coating gives fruit an attractive glossy sheen to the consumer (Sankat and Maharaj, 1997). Application of waxes to fruits was developed after the observation of slower water loss from the commodities that have a waxy skin. In most fruits, however, the repeated washing after harvest to remove and clean off dirt removes the natural wax and renders the remaining of this naturally protective compound ineffective for protection (Akamine *et al.*, 1975; Anon, 2001b). Hence, artificial waxes are applied to replace the lost natural waxes. While there is admittedly a cosmetic effect, the principal function of the waxes is to prevent and retard shriveling and dehydration of the fruit (Anon, 2001b). According to Amarante and Banks (2001) the coating material of wax could be made of lipids, resins, polysaccharides or proteins. Lipid components of coating include natural waxes such as carnauba wax, candelilla wax, rice bran wax, and bees wax; petroleum-based waxes such as paraffin and polyethylene wax; vegetable oil (corn, soybean, or palm); and oleic.

Resins are represented by shellac, wood rosin, and coumarone indene (Amarante and Banks, 2001; Mitra and Baldwin, 1997). However, fruits coated with resin have been reported to develop whitening of the skin due to condensation that develops when they are brought from cold storage to ambient temperature (Hagenmaier and Baker, 1994). Polysaccharide-based coatings have been extensively studied for their selective permeability to O_2 and CO_2, resulting in modified internal atmosphere composition and delayed ripening for mango fruits (Mitra and Baldwin, 1997) and for many fruits (Amarante and Banks, 2001). Proteins have been less investigated as film formers than lipids and polysaccharides. Carnauba waxes are extracted from palm leaves while candellia being from *Euphorbia spp* (Jenks and Ashworth, 1999). According to Thompson (1996) the composition of the coating material should be a water-soluble high polymer material such as a polysaccharide, hydrophobic and non-volatile liquids such as natural waxes (carnauba wax). Water-wax emulsion waxes are safer to use than solvent waxes, which are highly flammable (Akamine *et al.*, 1975). Water-wax emulsion waxes can be used without the necessity of drying the fruit prior to application. The thickness of coating material when applied to the produce significantly alters the permeability of the skin to gases in such a way that the permeability to oxygen is considerably reduced while affecting CO_2 to a lesser degree (Hagenmaier and baker, 1993; Salunkhe and Desai, 1984a). The wax film must, therefore, be thin or else gas exchange may be overly hindered, causing anaerobiosis and associated quality loss such as the production of off flavour (Wills *et al.*, 1998). Waxes are either brushed, sprayed, fogged or foamed onto produce, or produce is conveyed through tank of wax emulsion. Kotecha and Desai (1995) reported that Topsin-plus-wax emulsion treated banana fruits had significantly lower values of pulp:peel ratio, TSS, total sugars, TA, and total yellow pigments and significantly higher values of starch, ascorbic acid, and total chlorophyll, indicating that the ripening process was retarded when compared to control fruits. Similarly colour development (Paull and Chen, 1989; Sankat and Maharaj, 1997) and ripening (Krishnamurthy and Kushalappa, 1985) of papaya fruit was retarded by waxing. Skin coating of banana fruits with waxol (12%) significantly reduced weight loss and respiration rate and the occurrence of respiratory climacteric was delayed (Rao and Chundawat, 1990). Similarly, reduced in internal browning symptoms of chilling injury, reduced weight loss and improved fruit appearance was observed after waxing pineapple fruits (Paull, 1997). No adverse effects on sensory quality were detected when 0.75% Tal Pro-long (a mixture of sucrose esters of fatty acids and carbxymethylecellulose) was used for mango fruits, but coated fruits showed a slower decrease in TA and ascorbic acid as well as retarded softening and carotenogenesis (loss of green colour)

(Mitra and Baldwin, 1997). Tal Pro-long coating also significantly altered the permeability of banana skin to gases (Banks, 1984) and retarded ripening and extended shelf-life of papaya fruits (Baldwin *et al.*, 1992). It was also reported that waxing helps to inhibit mold growth if applied with fungicide. Many commodities such as cucumber, tomato, passion fruit, pepper, banana, apple, papaya and some root crops are now being waxed to reduce water loss by 30-50% (Wills *et al.*, 1989). However, not all waxed produce will respond favourably. The storage life of waxed breadfruit was significantly shortened, especially in cold storage (Akamine *et al.*, 1975). Waxing does not improve the quality of any inferior fruit, but with good handling helps to maintain the quality.

2.3.4 Packaging

Another method by which the deterioration of banana and papaya can be minimized is the use of plastic packaging. Modified Atmosphere Packaging (MAP) can be employed to improve products shelf-life (Hintlian and Hotchkiss, 1986). MAP refers to the development of a MA around the fruit through the use of permeable polymeric films (Yahia, 1998). Several researchers have investigated the potential for the sealed polymeric films to generate a favourable MA within the package environment (Ben-Yehoshua, 1985; Cameron *et al.*, 1989). The main function of packaging is to minimize postharvest loss and increase the shelf-life of fruit mainly by controlling the water loss (Hardenburg, 1975; Jobling, 2001). Sankat and Maharaj (1997) reported that plastic film wraps were more effective than waxing in reducing water loss from papaya fruits. Moreover, fruit waxing reduced weight loss by 14-40%, while plastic shrink-wraps reduced it by about 90% (Paull and Chen, 1989). The shelf-life as measured by appearance, firmness, shrinkage, weight loss and other keeping quality has been doubled for many fruits (Ben-Yehoshua, 1985) and for banana (Krishnamurthy and Kushalappa, 1985) by seal packaging. Scott *et al.* (1971) also reported that polyethylene bag packaging delayed ripening, restricted weight loss and resulted in considerable reduction of mechanical injury during transport and storage. A delay in ripening of mature green Nigerian banana was also observed when fruits were dipped in fungicidal suspension held at room temperature in white polyethylene bags (Kotecha and Desai, 1995; Salunkhe and Desai, 1984a) and retarded yellowing was exhibited (Fuchs and Temkin-Gorodeiski, 1971). A shelf-life similar to that obtained with refrigeration could be achieved at ambient temperature by packing fruits in sealed

polyethylene bags (Satyan *et al.*, 1992a; Satyan *et al.*, 1992b). Moreover, the authors added that the average storage life was increased two to three times when banana bunches were held in sealed polyethylene tubes and was further increased three to four times when an C_2H_4 absorbent was included in the bags. A substantial reduction in internal O_2 and a concomitant decrease in internal C_2H_4 concentration appeared to be instrumental in delaying the ripening of sealed fruits (Lazan *et al.*, 1990a). Different types of films have been tested, including polyolefins, several polyethylene types of low and high density (LDPE and HDPE, respectively) and polypropylene (Ben-Yehoshua, 1985), but relatively few have been used mainly due to their permeability (Kader, 1992b). Micro perforated polyethylene plastic bags with different thickness are some of the commonly used fruit packaging materials. These are less expensive and their ease of application makes the technique more affordable even for subsistence farmers (Satyan *et al.*, 1992a). However, only little commercial success has been reported for the use of this procedure because of the difficulty in controlling the O_2 and CO_2 variation with in the package (Barkai-Golan, 2001). Despite the long list of potential benefits and advantages of MA for tropical fruits, there are several potential problems and hazards (Yahia, 1998). This can be related to their perforation size and density. Such sealed packages must be perforated or opened before marketing (Turter, 1992) to allow ripening and prevent injury from high levels of CO_2, or depleted O_2. Similarly, different thickness of the same type of films or different conditions (temperature and RH) surrounding the package resulted in different permeability and therefore in different package atmosphere (Yahia, 1998). The author added many factors must be considered when trying to develop MAP system, including type, thickness, and method of fabrication of film; packaging size; temperature; humidity; length of storage; type, quantity, and physiological stage of fruit; and tolerance of each fruit to the different gases (O_2, CO_2, C_2H_4). An ideal film must let more CO_2 exit than it lets O_2 enter to avoid accumulation of CO_2 (Kader, 1992b). Several polymers used in film formulation meet this criterion (Table 2.3). Ripening of fruits or yellowing of leafy vegetables may occur rapidly within the packaging if not perforated. The drastic decrease in the O_2 level, combined with the increase CO_2 level, may lead to anaerobic respiration, fermentation and development of an off flavour, especially in unperforated bags (Cantwell and Reid, 1993; Jobling, 2001). Inadequate atmosphere can aggravate or initiate physiological disorders, fermentation and increase microbial growth (Yahia, 1998). This becomes more problematic when fruits are transferred or stored at higher storage temperatures. Storage of mango fruits in polyethylene bags inhibited normal fruit colour development and causes off flavour making the fruit unacceptable (Illeperuma and Jayasuriya,

2002; Satyan, *et al.*, 1992a). For this reason Yahia (1998) recommended films used for tropical fruits should be characterized by a relatively higher permeability to gases and to water vapour. Perforation of bags therefore counterbalances the enhanced respiration rate, depletion of O_2 and mould development. Similarly, inclusion of C_2H_4 scavengers overcomes the problem mainly by reducing C_2H_4 concentration, which enhances respiration rate and hence O_2 depletion. Use of film bags impregnated with anti-microbial compounds that actively retard the development of harmful microorganisms during storage has also been recommended by Barkai-Golan (2001).

Table 2.3: Permeability of Films Available for Packaging fresh Produce.

	Permeabilities (cc/m^2/mil/day at 1 atm)		
Film type	**CO_2**	**O_2**	**CO_2:O_2 ratio**
Polyethylene low density	7,700-77,000	3,900-13,000	2.0-5.9
Polyvinylchloride	4,263-8,138	620-2,248	3.6-6.9
Polypropylene	7,700-21,000	1,300-6,400	3.3-5.9
Polystrene	10,000-26,000	2,600-7,700	3.4-3.8
Saran	52-150	8-26	5.8-6.5
Polyester	180-390	52-130	3.0-3.5

Adapted from Kader (1992b)

Chapter 3: Studies on the Shelf-Life and Quality Attributes of Banana Fruits as Affected by Modified Atmosphere Packaging

3.1 Abstract

Banana fruit is one of the common tropical climacteric fruits identified by rapid deterioration after harvest. To maintain postharvest quality and increase shelf-life of the fruit, green mature banana (cv. Williams) fruits were wrapped postharvest using micro-perforated polypropylene bags (MPB), micro-perforated polypropylene bags with ethylene absorbent (MPB+K), and macro-perforated polypropylene bags coated with anti-mist coating (PP). Fruits were stored either at 12, 15 or 22°C. Percentage weight loss (PWL), visual colour change, change in firmness and respiration rate of the fruits were recorded weekly during the storage period. Total soluble solids (TSS), pH, total chlorophyll and total carotenoid concentrations as well as organic acid concentrations were analysed at the end of the storage period. Based on the colour change and softening days to ripen was determined. Storage temperatures affected shelf-life of fruits as measured by softening and colour development. As a result fruits were stored for upto eight, six and three weeks at 12, 15 and 22°C, respectively. MPB and MPB+K significantly ($P<0.001$) reduced PWL, colour development and softening as compared to control fruits, although these treatments resulted in an increased respiration rate at 22°C. PP on the other hand showed a reduced respiration rate at all storage temperatures and retarded PWL, softening and colour development as compared to control fruits, thus shelf-life was extended accordingly. PWL, colour development, softening, and respiration rate increased concomitant to ripening. PP at all storage temperatures as well as MPB and MPB+K at 12 and 15°C seem to overcome fruit deterioration and perishabilty after harvest. Analysis of the quality parameters (TSS, pH, total chlorophyll and carotenoids and organic acids) after storage showed no measurable difference at ($P<0.05$) among treatments at all storage temperatures, which indicates that application of these treatments will extend fruit shelf-life without affecting the quality of the fruit. It can, therefore, be concluded that PP, MPB+K and/or MPB can be used as alternative storage methods in areas where the cold chain facility and distribution systems are poor.

Key words: Banana, firmness, MAP, percentage weight loss, respiration rate, skin colour.

3.2 Introduction

Like many tropical climacteric fruits banana exhibits considerable loss after harvest in the tropical environment. The fruit represents a major non-grain starch staple in the humid tropics where it grows in low input systems (Burdon *et al.*, 1993). Although it has traditionally been a dietary staple in many countries, it has, until recently, been relatively neglected by most policy makers and research institutes (Olorunda, 2000). Part of this neglect can be attributed to the high postharvest losses coupled with difficulties in marketing and processing of the highly perishable commodity. According to Satyan *et al.* (1992a) banana fruits are produced throughout the year, but are only available in quantity for part of the year. This reveals the need to optimise storage action to extend the fruits availability. However, in the tropical environment under which the fruit is produced, storage is usually not practised due to lack of facilities and infrastructures. As a result, bananas have a shelf-life of one to ten days (Ferris, 1997). High ambient temperature, poor handling and lack of knowledge results in massive crop losses after harvest, because of which it becomes over-ripe by the time it reaches its ultimate destination (Olorunda and Aworh, 1984). The difficulties associated with the short storage life of the fruit are worsened by poor marketing systems of developing countries (Ferris, 1997). It is, therefore essential to establish methods, which delay ripening and extend the shelf-life without affecting fruit quality within the context of the conditions under which fruit is produced and marketed. Previous work on the use of polyethylene bags with and without ethylene absorbent to extend the shelf-life of banana under refrigerated conditions has been reported by Chamara *et al.* (2000) and Satyan *et al.* (1992b). It has also been reported by Ferris (1997), that storing banana fruits in polyethylene bags at 20°C reduced weight loss and less damage to the fruit and ripening was delayed by upto six days. However, high build-up of CO_2 inside the polyethylene bags was evident which ultimately limited storage life of the fruits. Similarly, Yantarasri *et al.* (1995) reported that unperforated bags used for seal-packaging of mango were no sufficiently permeable to O_2 exchange required for the proper ripening and colour development of the fruit. These packages were merely to create a modified atmosphere while avoiding anaerobic conditions. Similarly, fruit decay was enhanced by polyethylene film wrapping of mango (Miller *et al.*, 1986) strawberries (Nunes *et al.*, 1998) and development of off-flavour made fruits unacceptable (Chaplin *et al.*, 1982). For this reason different packaging materials such as micro-perforated polypropylene bags with and without ethylene absorbent and polypropylene bags coated with

anti-mist coating are used in this study. Several other workers (George and Marriott, 1985; Lebibet *et al.*, 1995; Seberry and Harris, 1998) investigated various ways of reducing postharvest losses and increasing fruit shelf-life, but not only were they unsuccessful within the production context, but there is also still a need for low-cost storage together with quality maintenance of this crop in poorly developed countries.

This study was carried out to evaluate and observe the effect of packaging materials with or without ethylene absorbent to counteract rapid fruit deterioration within the context of conditions likely to be experienced in many poorly resourced areas.

3.3 Materials and Methods

Freshly harvested banana fruits were obtained from a Komatipoort commercial farm, Mpumalanga, South Africa. On arrival at the laboratory in the Horticultural Science Department, University of Natal, fruits were washed with tap water and dipped in a fungicide solution containing 670g/1000L (w/v) of Magnate sulphate® (Makhteshim-Agan, Israel). After drying, fruits were randomised and subjected to the following treatments: Control (untreated), micro-perforated polypropylene bags with 9 μm perforation size (MPB), micro-perforated polypropylene bags with ethylene absorbent and 9 μm perforation size (MPB+K), and polypropylene bags coated with anti-mist coating with macro perforation of eight holes of 1 cm diameter each (PP). The thickness of all bags was 35 μm. Eighteen fruits were allocated to each treatment, i.e., six replications and three fruits in each replication. Fruits were sealed in the bags and stored at 12, 15 or 22°C. Data was collected every week for percentage weight loss (PWL), firmness, colour change and respiration rate. Fruits were allowed to ripen naturally until the table ripe-stage (6- Loesecke scale) and time taken to ripen was noted. After fruits were fully ripened, a portion of fruit rind was cut and stored in deep-freezer (-20°C) for further analysis of total chlorophyll and carotenoid concentrations. Similarly, a portion of the pulp was cut longitudinal (from end to end) to minimize variation in sugar concentration (Garner *et al.*, 2001) and used for analysis of TSS, pH and organic acids concentrations. Data were subjected to variance analysis (ANOVA) using the GenStat® 5th edition statistical analysis software (VSN, 2001). Treatments were compared at $P<0.05$ according to Fisher's protected LSD Test.

3.3.1 Percentage Weight Loss (PWL): Each fruit was weighed before any treatment was

applied. At weekly intervals fruits were reweighed and PWL calculated. The difference in weight was expressed as percentage weight loss calculated as follows:

$$PWL = \frac{Initial\ weight - actual\ weight}{initial\ weight} * 100$$

3.3.2 Firmness Change: Fruit firmness was measured at the beginning of the experiment before any treatment was applied. At weekly intervals fruit firmness was determined as measured by a fruit firmness tester (Mechanical densimeter, FL, USA) (Fig 3.1) with a reading ranging from 0-100, the higher the reading the firmer the fruit and vice versa. Data was taken from two different sides of the fruit and the mean result calculated.

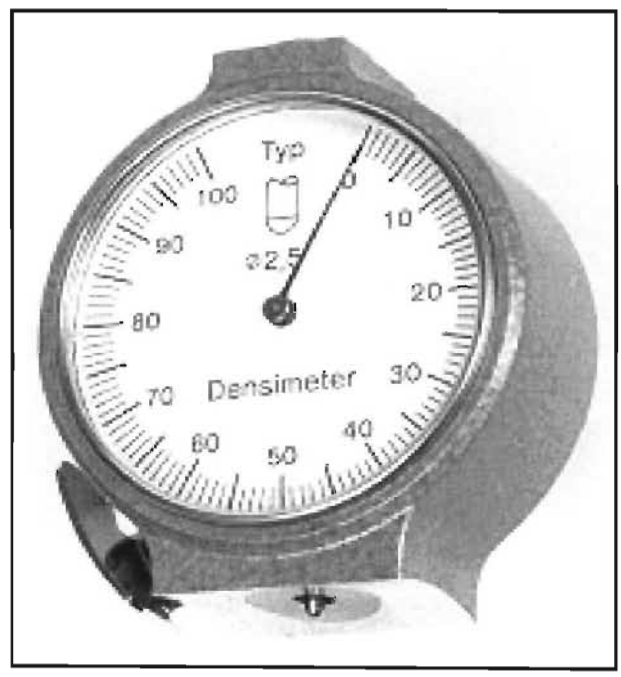


Fig 3.1Mechanical densimeter used to measure fruit firmness change during storage.

3.3.3 Skin Colour Change: Colour change of the fruits was scored numerically using a standard banana-ripening chart (Fig 3.2) with colour plates ranging from 1-7 as described by von Loesecke (1949), where: 1-green; 2- green with yellow tracks; 3- more green than yellow; 4-more yellow than green; 5-yellow with green tips; 6-all yellow and 7- yellow flecked with black spots. A mean colour score was calculated for each sample of fruits within the treatments.

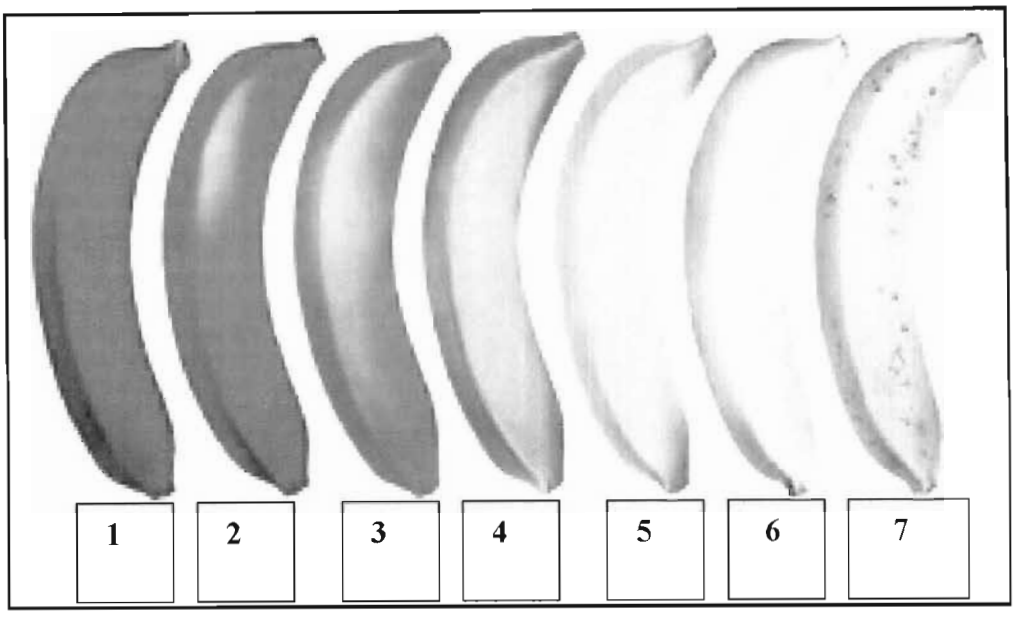


Fig 3.2 Banana colour development during ripening used to score colour change. Where 1=green, 2= green with yellow tracks, 3= more green than yellow, 4 = more yellow than green, 5 = yellow with green tips, 6 = all yellow, 7 = yellow flecks with black spots

3.3.4 Respiration Rate: On a weekly basis the respiration rate of each fruit was determined as CO_2 release using an Infra Red Gas Analyzer (IRGA) (Fig 3.3). Each fruit was placed in a separate enclosed jar of 3 L volume. Initially the ambient CO_2 inside the jar was monitored and the increase in CO_2 concentration in to the container was determined by placing the fruit in to the jar for 10 min. The actual CO_2 release was determined by taking the difference between CO_2 after 10 min and ambient concentrations. The CO_2 released by the fruit was calculated as $mlkg^{-1}hr^{-1}$ adjusting for volume in the containers using the previously determined fruit volume. For detailed calculation see Appendix 2.

3.3.5 Pigment Determination: For the determination of total chlorophyll and carotenoids 1 g of fresh weight of banana peel was finely ground with liquid nitrogen and blended with 10 mL of 100% methanol. The sample was then placed on ice in the dark for 10 min before it was homogenized using an ultra-turax® (IKA, Germany), 2 x 1 min bursts, waiting 5 min between bursts and centrifuged using (Sc-158T, Tawain, R.O.C.) for 5 min at 3000 rpm. The supernatant was decanted and placed in the spectrophotometer (Anthelie Advanced, Secoman, France) to read the absorbance at 470, 652, and 665 nm wavelengths. The concentration of total chlorophyll and carotenoids were calculated using the Wellburn (1994) equation. Results were expressed as μgg^{-1} fresh weight. For detailed calculations see Appendix 3.

Fig 3.3 Measurement of respiration rate by Infra Red Gas Analyser (IRGA)

3.3.6 Total Soluble Solids (TSS): As it was difficult to extract juice from banana fruits, method was adapted according to Illeperuma and Jayasuriya (2002). TSS was determined from finely ground fresh (5 g) sample, homogenized with 5 mL distilled water using an ultra-turax® (IKA, Germany) for 2 min and the pooled fraction was centrifuged using (Sorvall RC 5C Plus, CA, USA) at 6000**g** for 10 min. The supernatant was decanted and TSS was measured using a calibrated hand held digital refractometer (PR-101, 0-45%, ATAGO CO., Ltd, Japan).

3.3.7 pH: The remaining supernatant was diluted to 80 mL with distilled water according to Illeperuma and Jayasuriya (2002) and pH measured with a (Corning 430, NY, USA) pH meter.

3.3.8 Organic Acids: Five gram fresh weight of fruit was taken from a frozen sample. The sample was then blended with 95% ethanol for 4 min at maximum speed with an ultra-turax homogenizer (Pérez *et al.*, 1997). The homogenate was centrifuged using (Sorvall RC 5C Plus, CA, USA) at 27000**g** for 10 min and the residue was washed twice with 80% ethanol. The supernatants were combined and adjusted to 5 mLg^{-1} of fresh weight (FW) with 80% ethanol. For the organic acid separation, 10 mL of sample from the ethanolic extract was taken and evaporated in the dark for four to five hrs to dryness with speed vacuum evaporator (Savant, Sc200, NY, USA). The dry residue was redisolved in 1 mL of 0.2 N H_2SO_4 and 0.05% disodium

ethylene diamine tetra acetate (EDTA). All the fruit sample concentrates were deeply pigmented and would thus severely reduce analytical column life if they were to be injected directly onto the high performance liquid chromatography (HPLC) system (Richmond *et al.*, 1981). Therefore, the sample was loaded onto a C_{18} Sep-Pak cartridge (isolute C_{18}, EC, Mid Glamorgan, UK) and eluted with upto 4 mL of the above solution. Before each sample was injected into the HPLC for analysis it was filtered through a 0.45 μm nylon filter to further ensure removal of any particulate impurities that might be present.

Standards: To determine the retention times of the analytes, standard solutions were prepared individually for each of the main organic acids found in the fruit (malic and citric) from analytical reagent grade materials (Sigma Chemical Co., St. Louis, MO, USA). For calibration, a set of mixed organic acid standards (malic and citric acids) was prepared.

HPLC Conditions: Organic acids were analysed in a PerkinElmer (Wellesley, MA, USA) equipped with Diode Array Detector (DAD) (series, 200) and fitted with a PerkinElmer Pump (series, 200) and auto sampler (series, 200). Data were processed by means of a Network chromatography interface (NCI 900) and Totalchrom workstation software computing system (Waters, Milford, MA, USA). Isocratic separations of the compounds were made on a stainless steel (Waters Spherisorb 5 μm ODS2 4.6 x 250 mm) column. The mobile phase utilized for the elution consisted of a filtered (0.22 μm nylon) and degassed solution of $NH_4H_2PO_4$ (15 gL^{-1}), which was prepared with analytical reagent grade and adjusted to pH of 2.1 with phosphoric acid. Elution was monitored using a DAD UV detector set at 210 nm. The mobile phase flow rate was 1 mL min^{-1} with an injection volume of 20 μL. The concentration of each sample was calculated by comparison of retention times and peak areas to the areas obtained using the standard acid solutions with known concentrations. A set of calibration standards was run at the beginning and end of each batch of (10 –15) samples. Concentrations were expressed as percentage of fresh weight. For detailed calculations see Appendix 4.

3.4 Results and Discussions

3.4.1 Percentage Weight Loss (PWL)

Rapid weight loss after harvest is one of the common problems of most tropical fruits. Especially, if the fruit is to be sold by weight, such as banana (Lebibet *et al.*, 1995), additional

economic losses are likely. Storage temperature exerted a significant effect on fruit PWL and hence shelf-life. Fruits stored at 12°C showed less weight loss compared to both, 15 and 22°C. This can be related to the effect of temperature on fruit transpiration, and is in agreement with results found by Lebibet *et al.* (1995), where higher storage temperatures increase the banana fruits water loss and hence increase weight loss. However, fruits stored in the MPB and MPB+K packaging had significantly reduced ($P<0.001$) PWL at all storage temperatures as compared to the other treatments, especially the control (Fig 3.4). These results were similar to the reports of Ferris (1997), Miller *et al.* (1983) and Pekmezic *et al.* (1997) for banana, mango and persimmons, respectively. A substantial reduction in mango and avocado fruit weight loss was reported by Gonzalez *et al.* (1990) under modified atmosphere packaging (MAP). There was not any visible fungal development or/and water condensation inside the bags, unlike the reports of Satyan *et al.* (1992a) where banana bunch stalk rot development occurred inside the polyethylene bags. This may be due to type of packaging material used in this experiment. This further can be related to the effect of the perforation in combination to the anti-mist coating. It is therefore important to note the advantage of these packaging materials especially at higher storage temperatures. Similar to MPB, PP also performed better weight loss prevention throughout the storage period. There was significantly ($P<0.001$) reduced PWL as compared to control fruits at all storage temperatures. On average fruits lost 5.65, 5.52 and 3.64% after 52, 42 and 21 days at 12, 15 and 22°C, respectively. Control fruits exhibited highest weight loss. As a result, fruits exhibited shrivelled appearance and stored for only five, five and two weeks at 12, 15 and 22°C, respectively (Tables 3.1-3.3). The high weight loss of control fruits was more evident as the storage period was extended. This explains the rapid water loss from harvested fruits in tropical environment (Burdon *et al.*, 1993). A similar result was reported by Sanz *et al.* (1999) in strawberries.

The beneficial effect of MPB and PP can be utilized in tropical environments where fruits suffer from excessive water loss postharvest. To avoid off-flavour and fermentation, however, the thickness and porosity of the packaging must be appropriate to the physiological requirement of the fruit (John and Marchal, 1995).

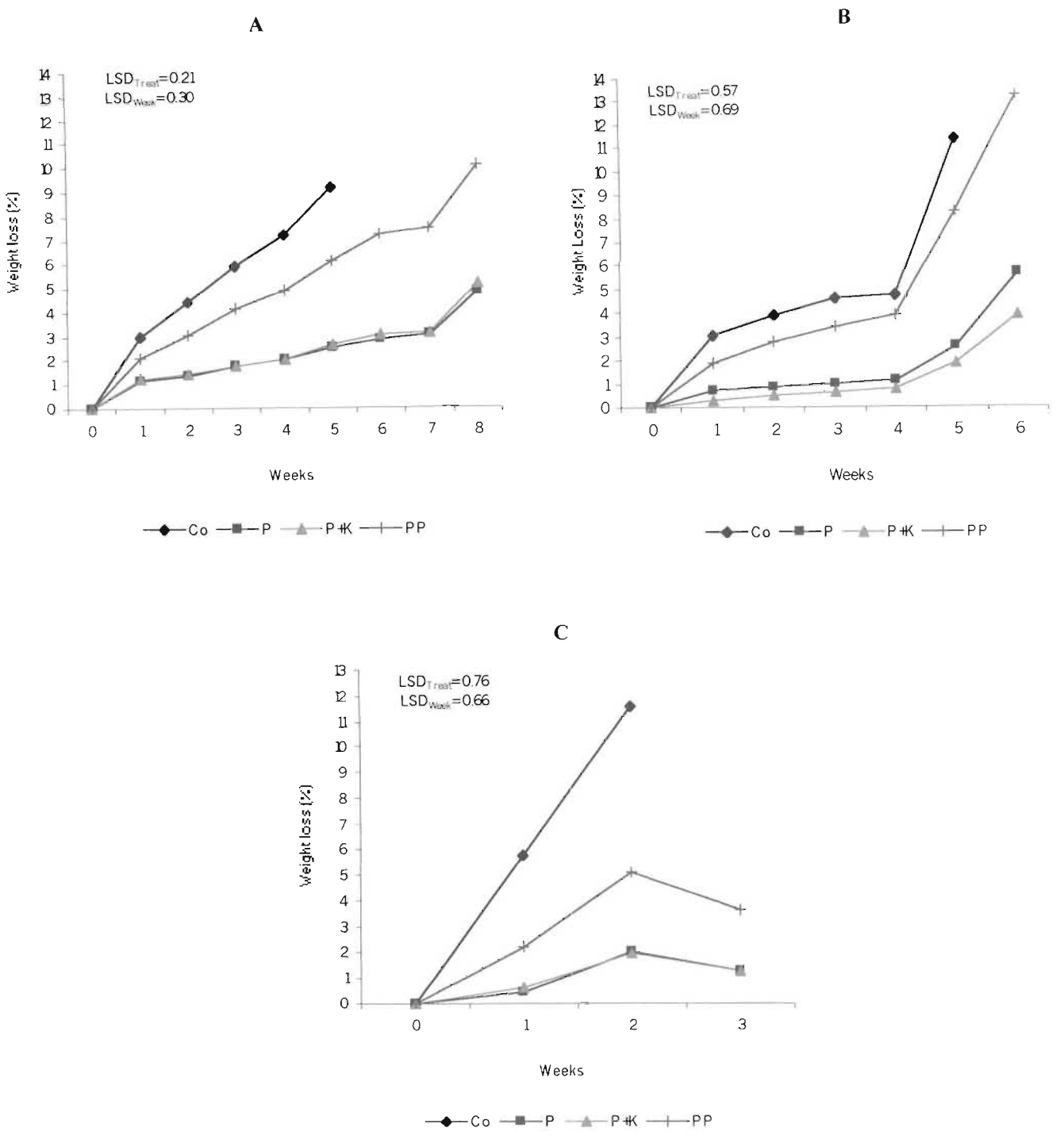


Fig 3.4 Percentage weight loss of banana fruits during storage as affected by packaging materials at 12°C (A), at 15°C (B) and at 22°C (C). For all figures, data points are means of six replicates. Least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and time (LSD_{Week}) are shown. Weight losses for each fruit were monitored to the table ripe stage (scale 6). Note data collection for control fruits terminated shortly. Co=control, P= Micro-perforated bags, P+K= Micro-perforated bags with ethylene absorbent and PP= Polypropylene bags.

Table 3.1 Physico-Chemical Characteristics of Banana Fruits Stored at 12°C as Affected by Different Postharvest Treatments

Treatments	Shelf-life (days)*	TSS (°Brix)	pH	Malic (%)	Citric (%)
Initial	-	1.53^{a}	5.16^{b}	0.021^{a}	0.016^{a}
Control	35	10.23^{b}	4.75^{a}	0.036^{bc}	0.056^{c}
MPB	50	10.23^{b}	4.76^{a}	0.034^{b}	0.059^{c}
MPB+K	52	10.30^{b}	4.77^{a}	0.042^{c}	0.060^{c}
PP	52	10.89^{b}	4.79^{a}	0.041^{bc}	0.046^{b}
LSD		0.86	0.10	0.007	0.007
P value		NS	NS	0.001	0.001

Table 3.2 Physico-Chemical Characteristics of Banana Fruits Stored at 15°C as Affected by Different Postharvest Treatments

Treatments	Shelf-life (days)*	TSS (°Brix)	pH	Malic (%)	Citric (%)
Initial	-	1.53^{a}	5.16^{b}	0.021^{a}	0.016^{a}
Control	35	10.97^{c}	4.71^{a}	0.053^{b}	0.054^{c}
MPB	42	10.53^{bc}	4.80^{a}	0.051^{b}	0.037^{b}
MPB+K	42	10.43^{bc}	4.71^{a}	0.049^{b}	0.039^{b}
PP	42	9.73^{b}	4.76^{a}	0.052^{b}	0.041^{b}
LSD		0.86	0.10	0.007	0.007
P value		NS	NS	0.001	0.001

Table 3.3 Physico-Chemical Characteristics of Banana Fruits Stored at 22°C as Affected by Different Postharvest Treatments

Treatments	Shelf-life (days)*	TSS (°Brix)	pH	Malic (%)	Citric (%)
Initial	-	1.53^{a}	5.16^{b}	0.021^{a}	0.016^{a}
Control	14	9.90^{b}	4.76^{a}	0.038^{b}	0.032^{b}
MPB	19	9.97^{b}	4.74^{a}	0.052^{c}	0.046^{c}
MPB+K	20	9.90^{b}	4.74^{a}	0.051^{c}	0.044^{c}
PP	21	9.93^{b}	4.75^{a}	0.058^{c}	0.042^{c}
LSD		0.86	0.10	0.007	0.007
P value		NS	NS	0.001	0.001

Means in each columns followed by the same letter are not significantly different at 5% level.
NS= Not Significant.
*** Days were determined when the fruit reaches table ripe stage (Scale -6)**

3.4.2 Firmness Change

A decrease of fruit firmness is one of the general features that accompanies ripening of both, climacteric and non-climacteric fruits (Biale and Young, 1981). Softening is one of the most significant quality alterations consistently associated with the ripening of fleshy fruits (Kays, 1991). Results from this experiment showed that all fruits irrespective of treatment, decreased in firmness with an extension of the storage period or an increase in temperature (Fig. 3.5). This is in general agreement with the results reported for kwifruit by Agar *et al.* (1999) and for banana by Zhang *et al.* (1993). These authors noted that fruit softening increases as the storage temperature and time is extended. Furthermore, the treatments resulted in different fruit firmness depending on the storage temperature. MPB, PP and MPB+K significantly ($P<0.001$) decreased the rate of softening as compared with control fruits. An elevated CO_2 concentration around the fruit was reported to maintain initial firmness or slows down softening rate in tomato (Ait-Oubahou, 1999) apples (Geesen and Smith, 1989) and strawberries (García *et al.*, 1998; Rosen and Kader, 1989). Similarly, Talhouk *et al.* (1999) reported a delayed shrivelling of loquat fruit by at least two weeks under 5 and 12°C storage temperatures. This retardation in firmness may be related to a decreased activity of cell wall degrading enzymes and thus a slowed alteration in the composition of cell walls (Lebibet *et al.*, 1995) and/or reduction in middle lamella cohesion (Dadzie, 1998). MPB did not retard fruit firmness changes at room temperature. The effect of temperature influenced fruit softening more than the treatments. Fruits packed in MPB+K, however, did not soften as rapidly as other treatments, especially near the end of shelf-life. This result agrees with previous results for mango by Illeperuma and Jayasuriya (2002) and for banana by Satyan *et al.* (1992a). Control fruits exhibited rapid softening at all storage temperatures. The use of an ethylene scrubber resulted in a consistently better fruit firmness than the control. The retardation of softening is important to reduce mechanical damage and to maintain fruit quality (Smith *et al.*, 1990). Softening is of great practical importance in the food industry to maintain texture characteristics of fruits (Castaldo *et al.*, 1989). MPB, MPB+K and PP, by maintaining fruit firmness and /or retarding fruit softening, might assist in minimizing fruit damage and deterioration, especially in rural tropical regions with poor fruit handling practices.

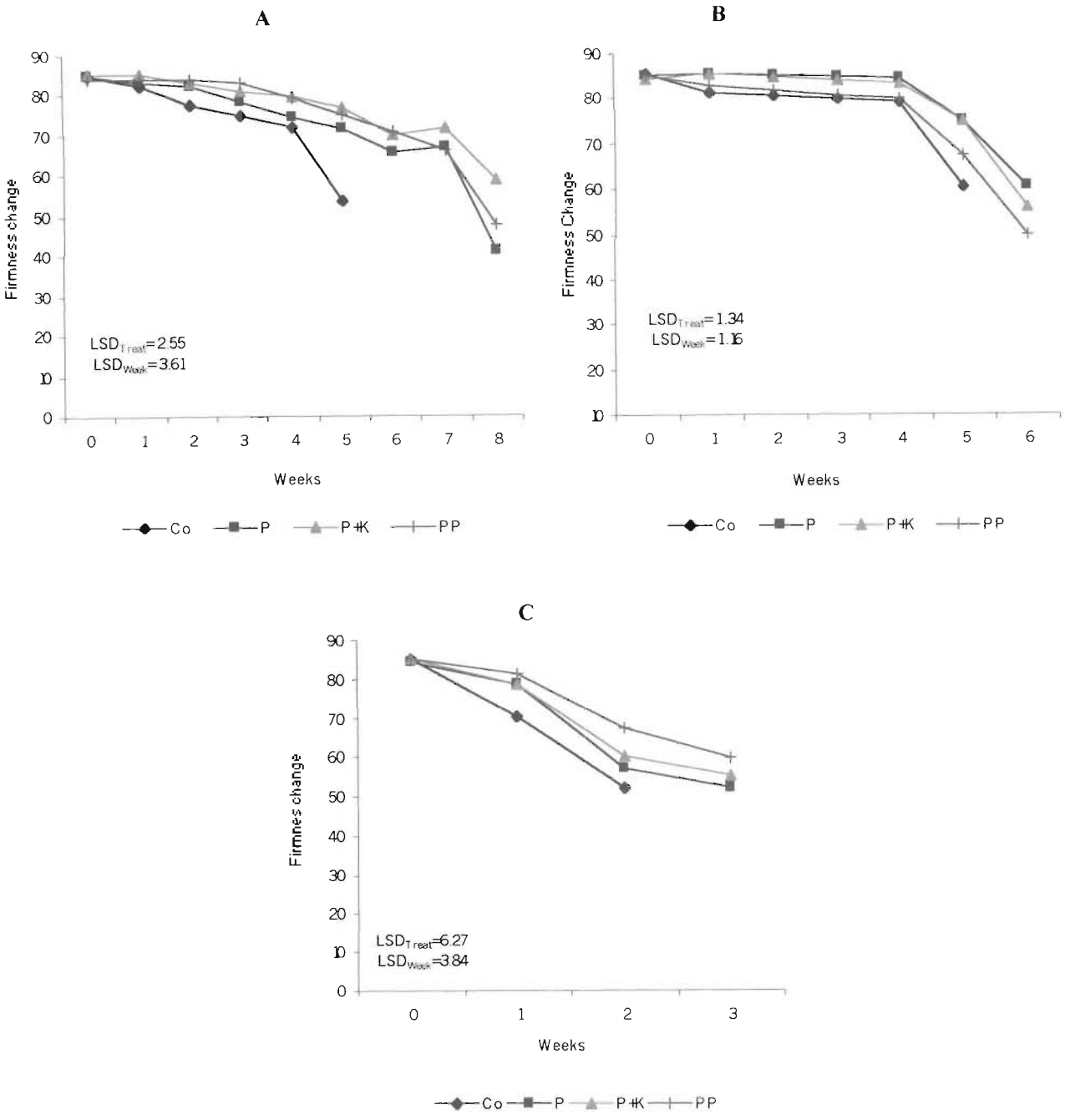


Fig 3.5 Firmness changes of banana fruits during storage as affected by storage conditions at 12^{o}C (A), at 15^{o}C (B) and at 22^{o}C (C). For all figures, data points are means of six replicates. Least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and time (LSD_{Week}) are shown. Firmness changes for each fruit were monitored to the table ripe stage (scale 6). Note data collection for control fruits terminated shortly. Co=control, P= Micro-perforated bags, P+K= Micro-perforated bags with ethylene absorbent and PP= Polypropylene bags.

3.4.3 Skin Colour Change

The ripening rate of fruit can be measured by colour change during storage. It is also one of the apparent parameters used to determine fruit quality and shelf-life. It is crucial in purchasing decisions, especially if the product is wrapped and cannot be touched or smelled (García *et al.*, 1998). In this experiment fruit colour development differ depending on storage temperature and treatment. Increased fruit colour change from the initial stage was, however, evident for all fruits irrespective of treatment. This is related to the natural ripening process of fruits, which is accompanied by colour development. The disappearance or loss of green colour and corresponding increase in peel yellowing are obvious signs of banana ripening (Dadzie, 1998). Storage temperature of fruits had considerable impact on the colour development (Fig 3.6) and, hence, shelf-life (Tables 3.1-3.3). Fruits at 12°C exhibited intense colour development after eight weeks while fruits at 15 and 22°C exhibited after six and three weeks, respectively. This result might be a good indicator of how higher ambient temperature in tropical regions affects fruit colour in particular, and storage life and quality of the fruit in general. This is in agreement to Lebibet *et al.* (1995) and Proft *et al.* (1998) who reported rapid colour change was evident in banana fruit at higher temperature. PP and MPB+K exhibited significantly ($P<0.001$) retarded colour change as compared to control fruits at all storage temperatures (Fig 3.6), however, MPB showed a trend of increased colour development at 22°C. Full colour development was obtained in four to five weeks for the control, while fruits stored in MPB, MPB+K and PP reached the maximum of the colour scale after seven to eight weeks at 12°C (Fig 3.7). Macnish *et al.* (1997) reported that packaging of mango fruits in container (1.2 L) delayed fruit ripening and colour development. Similarly, Ferris (1997) reported that banana fruits in polyethylene bags remained green for longer as compared to untreated fruits. This may be related to the effect of MA created by the packaging, which retard enzymatic activity (chlorophyllase) responsible for colour development. This further can be related to the reports by Krishnamurthy and Kushalapa (1985) that CO_2 build-up inside the bag retarded colour development in banana and lycopene development in tomato (Ait-Oubahou, 1999). Moreover Sanz *et al.* (1999) found a retarded redness in strawberries during storage at 20% CO_2 and or a very low O_2 level, although García *et al.* (1998) reported increased red skin colour of strawberry for fruits packed in polyethylene bags. Over all, PP and MPB+K were found to be promising approaches to retard colour change even at higher storage temperature, which can thus be used under tropical conditions.

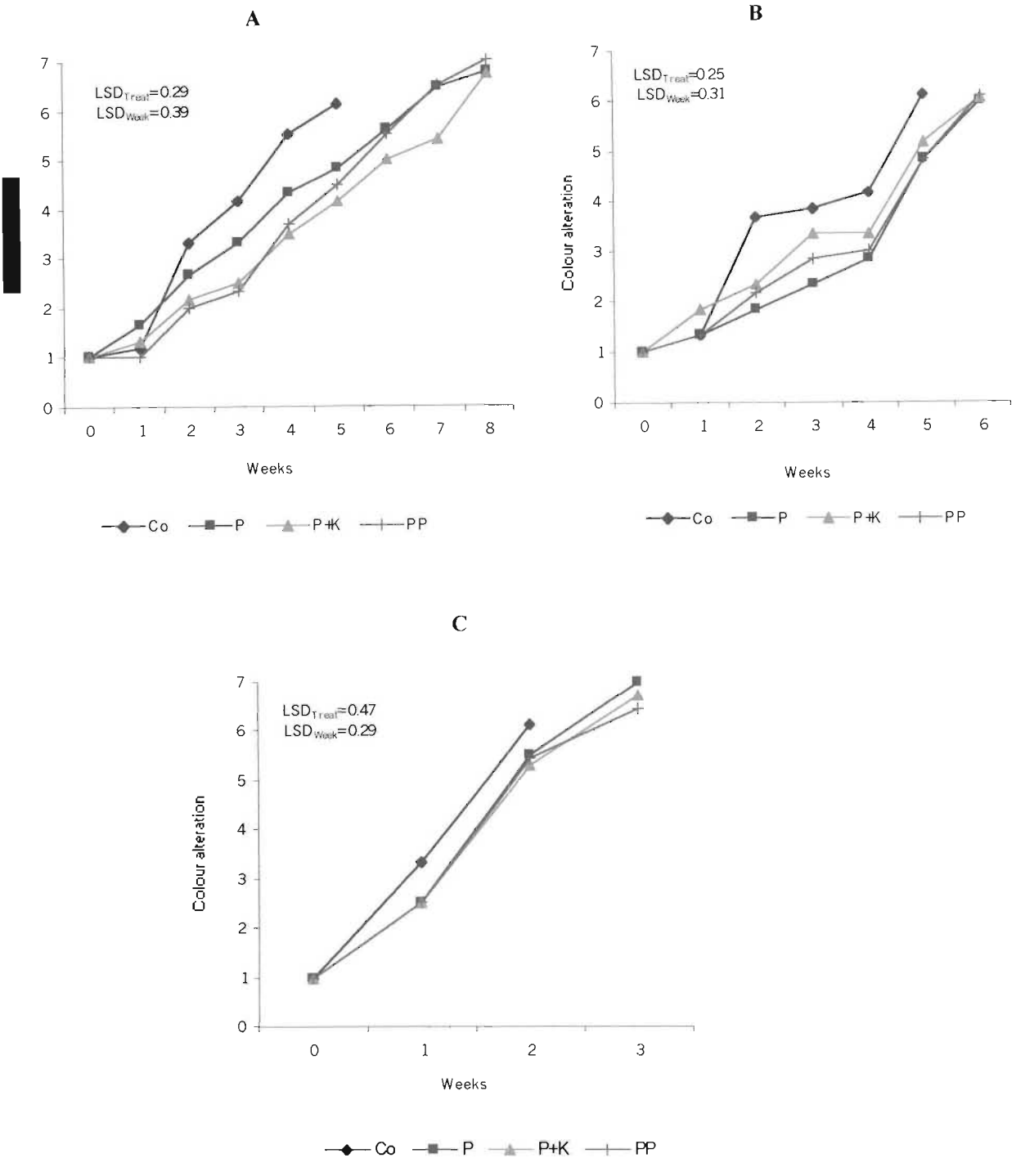


Fig 3.6 Colour changes of banana fruits during storage as affected by storage conditions at 12^{0}C (A) at 15^{0}C (B) and at 22^{0}C (C). For all figures, data points are means of six replicates. Least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and time (LSD_{Week}) are shown. Colour alteration for each fruit was monitored to the table ripe stage (scale 6). Note data collection for control fruits terminated shortly. Co=control, P= Micro-perforated bags, P+K= Micro-perforated bags with ethylene absorbent and PP= Polypropylene bags.

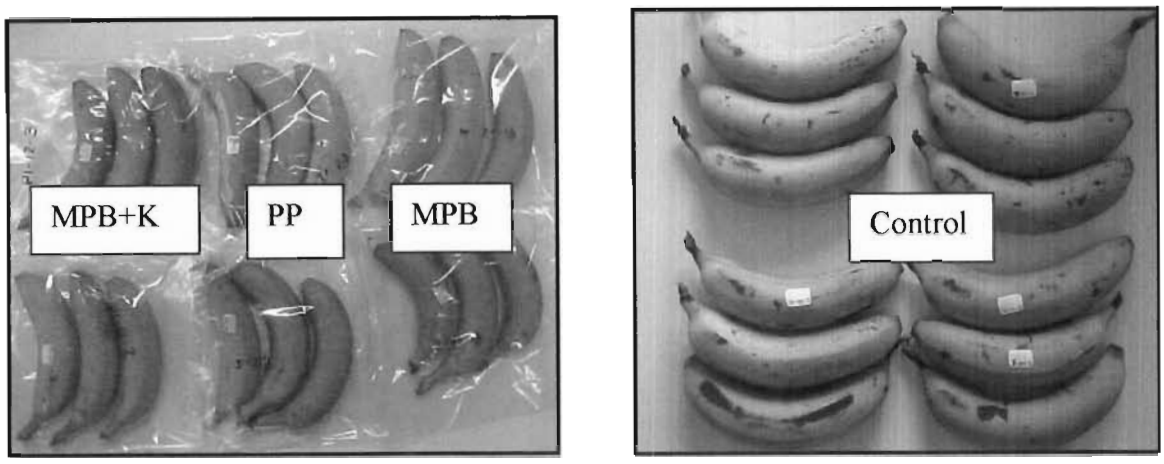


Fig 3.7 Fruit colour development as affected by various packaging materials after 28 days storage at 12°C.

3.4.4 Respiration Rate

The respiration rate of produce is an excellent indicator of metabolic activity of tissue and thus is a useful guide to the potential storage life of the produce (Wills *et al.*, 1998). Fruit cannot replenish the energy source consumed during respiration. The higher respiration rate after harvest is thus a good index of fruit deterioration, perishability and shelf-life. In general, storage temperatures and treatments influenced rate of respiration. A high respiration rate was exhibited in short period of time by fruits stored at the higher storage (22°C) temperature as compared to the other temperatures (Fig. 3.8 and 3.9). As a result fruit shelf-life was reduced considerably. Similar results were reported for guava by Osman and Ayub (1998) and for banana by Zhang *et al.* (1993). Increased CO_2 and C_2H_4 contents were found three or four days earlier at 30°C than at 20°C (Zhang *et al.*, 1993). Banana fruits stored at 12°C exhibited a lower respiration rate and thus increased shelf-life upto eight weeks as compared to only six weeks at 15°C and three weeks at 22°C. Although a considerable increase in fruit respiration rate concomitant to ripening was evident for all treatments, control fruits exhibited significantly higher ($P<0.001$) respiration as compared to the other treatments at 15°C and a trend of increase at 12°C (Fig 3.8). Similarly, MPB followed by MPB+K exhibited significantly higher rate at 22°C (Fig 3.9). The higher respiration rate of MPB and MPB+K at higher temperature may be related to the exposure of fruits to different (high O_2 and low CO_2) atmosphere during the time when the fruits were removed out of the bags for the determination of respiration rate under high ambient temperature. On the other hand, PP retarded respiration rate at all storage temperatures. Thus

fruit shelf-life was increased accordingly (Tables 3.1-3.3). It was reported by Sanz *et al.* (1999) that gas exchange for the same overall perforation surface area affected by diameter of perforation, which could infer a perforation edge effect on gas exchange. When fruit respiration does not match film permeability, CO_2 will increase and O_2 decrease with fruit fermentation to follow (Ait-Oubahou, 1999). As a result the apparent beneficial effect of MAP can be negated. From the results of the CO_2 evolution studies it would appear that polyethylene bags without or with only small perforations does not improve fruit shelf-life, but may decrease fruit quality and shelf-life. This is mainly because imperforated bags cause excessive depletion of O_2 in the bags and hence anaerobic respiration. Variable results have been reported for MA mainly due to the use of different plastic types, thickness, and perforations, and different storage conditions. However, when considering the other results obtained in this experiment, it would seem that all the MAP provide advantages over the control with PP being more effective than the others, especially at higher storage temperature.

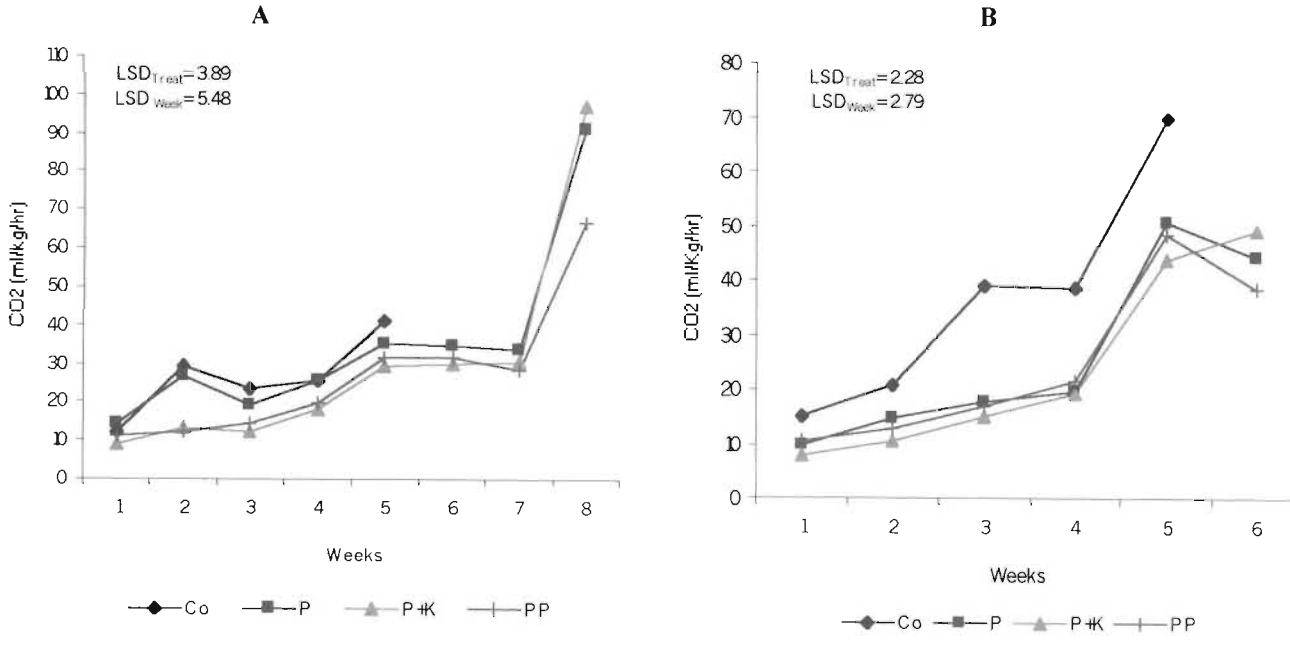


Fig 3.8 Respiration rates of banana fruits during storage as affected by storage conditions at 12°C (A) and at 15°C (B). For all figures, data points are means of six replicates. Least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and time (LSD_{Week}) are shown. Respiration rate for each fruit was monitored to the table ripe stage (scale 6). Note data collection for control fruits terminated shortly. Co=control, P= Micro-perforated bags, P+K= Micro-perforated bags with ethylene absorbent and PP= Polypropylene bags.

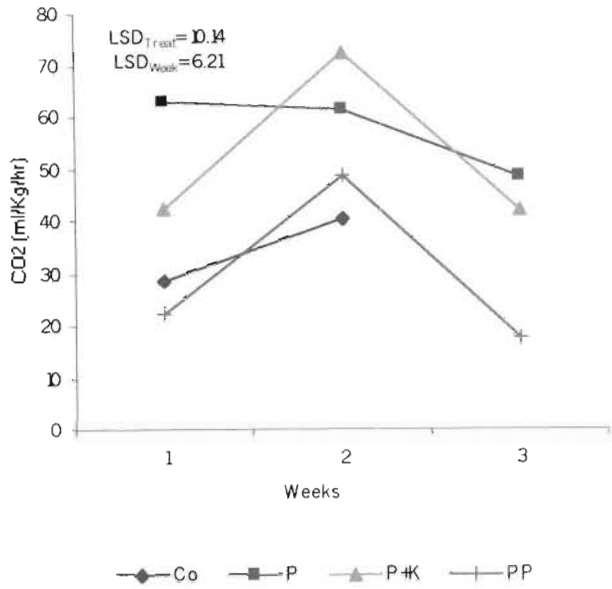


Fig 3.9 Respiration rates of banana fruits during storage as affected by storage conditions at 22°C. Data points are means of six replicates. Least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and time (LSD_{Week}) are shown. Respiration rate for each fruit was monitored to the table ripe stage (scale 6). Note data collection for control fruits terminated shortly. Co=control, P= Micro-perforated bags, P+K= Micro-perforated bags with ethylene absorbent and PP= Polypropylene bags.

3.4.5 Pigment Determination

3.4.5.1 Total Chlorophyll: The physiological status of fruit can be assessed by chlorophyll measurement. As senescence and ripening of fruits is frequently accompanied by the loss or degradation of chlorophyll. Results from this experiment showed that all the fruits irrespective of treatment exhibited reduced chlorophyll concentrations during storage. This chlorophyll degradation was more evident as the storage period was extended. As a result, fruits at 12°C exhibited a higher loss after eight weeks storage than those at 15°C after six weeks. Turner (1997) reported the rate of chlorophyll degradation in the banana follows an optimum response with temperature. Dadzie (1998) and Deli *et al.* (1996) reported that the chlorophyll content decreased slowly with ripening. There was, however, no measurable differences ($P<0.05$) in total chlorophyll concentration between treatments after different periods of storage (Fig 3.10 A). No significant difference among treatments infers the applicability of the treatments without any detrimental effect to the external visual quality of the fruit.

3.4.5.2 Total Carotenoids: The numerous studies on carotenoid pigments highlight their attractive properties (Míngueze-Mosquera *et al.*, 2000). Carotenoids are large group of pigments associated with the chlorophyll in the chloroplast and are found in the chromoplast (Kays, 1991). Analysis of the concentration of carotenoids both before storage and after the fruit ripened, showed no significant difference ($P<0.05$) (Fig 3.10 B). However, control fruits at 12°C exhibited a trend of lowered concentration. This result revealed that carotenoids in banana fruit presented even at the green stage of the fruit. This is supported by the previous reports by Robinson (1996) and Thompson and Burden (1995) where they stated that the peel colour changes of banana from dark green to bright yellow is a process of unmasking the already present carotenoids. No significant difference between treatments give a good indication of the applicability of these packaging materials to extend shelf-life without effect to the quality including at higher ambient temperatures, which might be used in tropical environment.

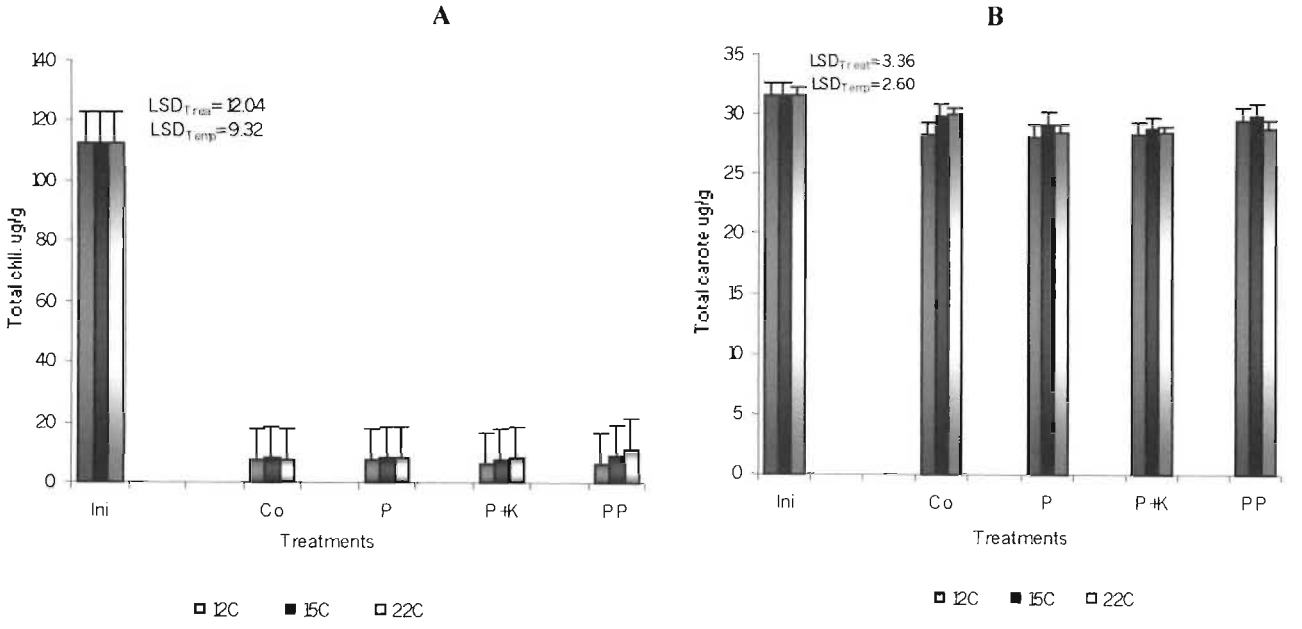


Fig 3.10 Total chlorophyll (A) and total carotenoid (B) concentrations of banana fruits peel before storage and after full ripe stage as affected by different treatments and storage temperatures. For all figure, data points are means of six replicates. The standard error mean of difference (+SED) and least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and storage temperature (LSD_{Temp}) are shown. Ini= Initial Co=control, P= Micro-perforated bags, P+K= Micro-perforated bags with ethylene absorbent, PP= Polypropylene bag.

3.4.6 Total Soluble Sugars (TSS)

Measuring the TSS is commonly used as indicator of internal eating fruit quality after harvest. One of the most evident changes that occur during banana ripening is the conversion of starch to sugar (Chang and Hwang, 1990). In this experiment most fruits exhibited significantly increased ($P<0.05$) TSS as compared to their initial concentration. A similar result was reported for banana fruits earlier by Chamara *et al.* (2000) and Wills *et al.* (1998). Control fruits exhibited significantly higher concentrations at 15°C as compared to PP. There was no, however, any measurable difference among the rest treatments at all storage temperatures (Tables 3.1-3.3). Yantarasri *et al.* (1995) similarly reported for mango fruits where they found no significant difference in the increase of TSS among various treatments of packaging materials with different perforations. It is, therefore, concluded that the packaging types did not adversely affect sugar content of the fruit, which can be used as a possible means to extend fruits shelf-life with limited effect to the quality of the fruit.

3.4.7 pH

The pH value of the samples exhibited a significantly ($P<0.001$) reduced pattern as the fruit ripened. This was evident for all treatments and at all storage temperatures. Similar reduced fruit pH in banana was reported by Chamara *et al.* (2000) and Robinson (1996). On the other hand, an increased pH value in banana was reported by Kruger *et al.* (1996). Inconsistent fruit pH during ripening was, also, reported in banana fruits by Mustaffa *et al.* (1998). Although all fruits exhibited significantly reduced fruit pH as compared to the initial value, there was, however, no significant difference ($P<0.05$) between treatments at various storage temperatures (Tables 3.1-3.3). This result once more indicates the possibility of storing fruits for longer period in MPB, MPB+K and PP without defects to the quality.

3.4.8 Organic Acids

According to Wills *et al.* (1998), apart from their biochemical importance, organic acids contribute greatly to taste with a balance of sugars and acids giving rise to the desirable taste of specific produce. Furthermore, they are also the second contributors (after sugars) to the soluble solids of fruits (Cordenunsi *et al.*, 2002). Results from this experiment revealed that organic acid increased during fruit ripening regardless of treatments and storage temperatures. A considerable

increase in acidity during ripening of banana fruit has previously been reported by Robinson (1996); Shanmugavelu *et al.* (1992) and Turner (1997).

3.4.8.1 Malic Acid: An increased concentration of malic acid was evident for all fruits as compared to the pre-storage value. All treatments and storage temperatures exhibited a significant increase ($P<0.001$) compared to the initial content. It has been reported earlier by Wills *et al.* (1983) that malic acid in banana fruits increases during ripening. Several other workers reported similar increased organic acids during ripening for banana (John and Marchal, 1995; Robinson, 1996; Turner, 1997). MPB+K at 12°C exhibited significantly higher malic acid than MPB. Similarly, control fruits at 22°C exhibited reduced value than the other treatments. With the rest of the treatments, however, there was no significant difference ($P<0.05$) in their concentrations at all storage temperatures (Tables 3.1-3.3). It can, therefore, be concluded that the different packaging materials increase fruit shelf-life via reducing PWL, softening and colour development, with a limited effect to the fruit eating quality.

3.4.8.2 Citric Acid: The concentration of citric acid during ripening followed the same trend as malic acid. All fruits increased significantly ($P<0.001$) in citric acidity concentration during ripening. This can be related to the ripening process of the fruits. A similar increase was reported by Alique and Oliveira (1994) in cherimoya, although Yonea *et al.* (1990) reported decreasing citric acid concentration during mango ripening. On average, fruits at 22°C showed a lower concentration of the citric acid as compared to the other storage temperatures (Tables 3.1-3.3). Control fruits exhibited a significantly lower concentration of malic and citric acids at 22°C. Despite the increase in acidity with ripening, the increase in sugars is greater and the sugar:acid ratio, which is an important component of flavour, increases with ripening (Turner, 1997). It can therefore be inferred from the above results that by extending fruit shelf-life with the PP, MPB and MPB+K fruit quality remained unaffected.

3.5 Conclusion

The maintenance of banana fruit quality and shelf-life is critically important in a tropical environment where excessive postharvest losses occur. Climacteric fruits, including banana, exhibit rapid and high fruit deterioration and perishability after harvest. Excessive PWL, softening, colour change and respiration rate are some of the major factors that cause fruit

deterioration. MAP would significantly retard the rate of changes for the above-mentioned attributes, and thus increase fruit shelf-life. This was evident for all storage temperatures, although MPB and MPB+K increase respiration rate at increased storage temperatures. Quality evaluation (TSS, pH, total chlorophyll and carotenoids, malic and citric acid) after storage showed no differences among treatments at all storage temperatures. These results suggested that MAP of banana fruits using micro-perforated bags coated with anti-mist reduce postharvest loss and without affecting quality of the fruit. Turner (1997) stated in his report that in the next few years, modified atmosphere will be used more widely to manage supply and improve quality at the market, particularly where controlled temperature facilities are expensive. The findings of the current study agree with the above hypothesis where the three packaging methods used are found to be viable alternative approaches to prolong postharvest shelf-life of banana even at higher storage temperatures of tropical environment lacking in effective cold chain management. These packaging materials will also not cause any increased decay or off-flavour development and thus can minimize postharvest loss and deterioration after harvest. Further study, especially with the effect of these packaging materials to the other quality parameters such as ascorbic acid, reduced sugars, polyphenolic oxides of fruits would be very helpful to establish the appropriate type of packaging material for best fruit quality.

Chapter 4: Effect of Plant Growth Regulators (PGRs) and Waxing on Shelf-life and Quality Attributes of Banana Fruits

4.1 Abstract

Short shelf-life and high perishabilty of banana fruit is one of the limiting factors for production and sales of this commodity in many countries. The effect of gibberellic acid (GA_3), indole butyric acid (IBA), and waxing on the shelf-life and selected quality attributes of banana (*Musa spp*) cv. Williams were evaluated. Fruits were stored at three different temperatures (12, 15, and 22°C). Percentage weight loss (PWL), firmness change, visual colour change, and respiration rate were measured weekly during the storage period. Total soluble solids (TSS), pH, and organic acid concentrations of the pulp as well as chlorophyll and carotenoid concentrations of fruit rind were determined at the beginning and at the end when fruit fully ripe. PWL, softening, colour change, and respiration rate increased irrespective of PGRs application or waxing as the storage temperature and storage period increased. However, under all storage temperatures waxing, GA_3 and IBA application significantly reduced ($P<0.001$) the parameters recorded as compared to control fruits resulting in an extended shelf-life. Waxing was the most effective followed by GA_3. Chlorophyll concentration of rind tissue and pH of pulp tissue declined significantly ($P<0.001$) throughout the storage period in all fruits, while rind carotenoids remained constant. There was, however, no significant difference ($P<0.05$) among treatments for all the quality attributes after storage, which implies these treatments not only extend shelf-life but also preserve the quality of the fruit. It is therefore concluded that waxing, IBA and GA_3 can be used as alternative methods to extend shelf-life and maintain fruit quality subjected to poor infrastructure, especially the cold chain.

Keywords: GA_3, IBA, percentage weight loss (PWL), respiration rate, waxing.

4.2 Introduction

Banana is an important staple crop supplying up to 25% of the carbohydrate requirement for approximately 70 million people in the humid zone of Sub-Saharan Africa (Gauhl *et al.*, 1998; Ferris, 1997), Asia, and Central and South America (George and Marriott, 1985). Despite its economic and social importance, postharvest losses are high due to poor handling and lack of knowledge. Especially fruits harvested during the warm summer months tend to have poor quality after ripening (Swarts, 1992). Olorunda and Aworh (1984) reported that poor postharvest handling practices and lack of storage facilities resulted in a substantial amount of fruit loss. As a result fruits never reach to the consumer to whom they were grown. The increased attention given to postharvest handling in recent years has come from the realization that poor practices can cause large losses of produce and potential income where considerable inputs of labour, material and capital have been made (Wills *et al.*, 1998). The high perishability and short shelf-life of banana is one of the limiting factors in production of this crop in many countries. The combination of high product perishability, fruit exposure to high ambient temperature, a slow marketing system, and poor marketing conditions leads to considerable fruit losses (Ferris, 1997). It is therefore very important to investigate means to extend the shelf-life and reduce postharvest losses of the fruit within the context of the conditions under which fruit is produced and marketed in countries of the above mentioned regions. Several workers (George and Marriott, 1985; Lebibet *et al.*, 1995; Liadó and Domínguez, 1998; Olorunda, 2000; Proft *et al.*, 1998; Seberry and Harris, 1998) have investigated systems for improvement of the situation, but not all successful within the production context probably due to insufficient storage facilities in the production areas. In addition, many banana and plantain improvement programs are being undertaken by different organizations, especially the International Network for the Improvement of Banana and Plantain (INIBAP), and the International Institute of Tropical Agriculture (IITA) to solve postharvest problems of the fruit. Although different workers reported various ways of reducing postharvest loss, methods to improve the storage life must be economically viable in tropical environments where there is lack of storage facilities and poor infrastructure. The quality of the fruit after extended storage should also be taken into consideration. Delayed fruit ripening and extended shelf-life of banana was reported by Lebibet *et al.* (1998) at lower storage temperature (13°C). On the other hand, high storage temperature caused abnormal ripening of the fruit (Proft *et al.*, 1998; Zhang *et al.*, 1993). Fruit surface coating with waxing (Tal-prolong®) reduced ripening and extended shelf-life up to eight (Olorunda and Aworh, 1984) or

six days (Krishnamurthy and Kushalappa, 1985) as determined by colour development. Ferris (1997) reported delayed ripening of banana by dipping the commodity into GA_3 where high atmospheric humidity was present, but no effect was found under low humidity. Many other researchers reported positive effects of PGRs on shelf-life extension of fruits (Babbitt *et al.*, 1973; Khader, 1992; Purgatto *et al.*, 2001). However, there is very little information concerning the quality of the fruit after extended storage, although Krishnamurthy and Kushalappa (1985) reported that quality of Prolong® treated fruits had an extended shelf-life, but were of inferior quality.

This study was conducted to examine the response of banana fruit to certain storage treatments at different storage temperatures in order to establish their effect on postharvest shelf-life and quality of the fruit within the context of poor infrastructure and storage facilities. Quality evaluation after storage was also a mandate of this study so as to assess fruit quality after extending the shelf-life.

4.3 Materials and Methods

Freshly harvested banana fruits were obtained from a Komatipoort commercial farm, Mpumalanga, South Africa. Fruits were picked from a uniform population. On arrival at the laboratory in the Horticultural Science Department, University of Natal, fruits were washed with tap water and dipped in a fungicide solution containing 670g/1000L (w/v) of Magnate sulphate® (Makhteshim-Agan, Israel). After being dried, fruits were randomized to the following treatments: control (untreated), GA_3, waxing (citrus carnauba), or IBA. Eighteen fruits were allocated to each treatment, i.e., six replications of three fruits each. Fruits that were allocated to GA_3 and IBA were dipped into aqueous solutions of 100 ppm GA_3 or IBA, respectively for 30 min. The waxing treatment was applied by lightly waxing fruits with a cloth dipped in the citrus carnauba wax, and allowed to dry for 30 min. Fruits were then stored either at 12, 15 or 22°C. PWL, firmness change, colour change, and respiration rate were measured weekly during storage period. Fruits were allowed to ripen naturally until the table ripe-stage (6- von Loesecke scale) von Loesecke (1949) and time taken to ripen was noted. After fruits were fully ripened, a portion of fruit rind was taken and stored in a deep freezer (-20°C) for further analysis of total chlorophyll and carotenoid concentrations. Similarly, a portion of the fruit pulp was cut longitudinal (to minimize variation in sugar concentration) for the analysis of TSS, pH, and

organic acids. All the data were subjected to variance analysis (ANOVA) using GenStat® 5th edition statistical analysis software (VSN, 2001). Treatments were compared for significance at $P<0.05$ according to Fisher's protected LSD Test.

4.3.1 Percentage Weight Loss (PWL): Each fruit was weighed before any treatment was applied. At weekly intervals fruits were reweighed and PWL calculated. The difference in weight was expressed as percentage weight loss calculated as follows:

$$PWL = \frac{Initial\ weight - actual\ weight}{initial\ weight} * 100$$

4.3.2 Firmness Change: Fruit firmness was measured at the beginning of the experiment before any treatment was applied. At weekly intervals fruit firmness was determined as measured by fruit firmness tester (Mechanical densimeter, FL, USA) (Fig 3.1) with a reading ranging from 0-100, the higher the reading the firmer the fruit and vice versa. Data was taken from two different sides of the fruit and the mean result calculated.

4.3.3 Skin Colour Change: External fruit colour change was scored numerically using a standard banana-ripening chart (Fig 3.2) with colour plates ranging from 1-7 as described by von Loesecke (1949). Where: 1-green; 2- green with yellow tracks; 3- more green than yellow; 4-more yellow than green; 5-yellow with green tips; 6-all yellow and 7- yellow flecked with black spots. A mean colour score was calculated for each sample of fruits.

4.3.4 Respiration Rate: On a weekly basis the respiration rate of each fruit was determined in terms of CO_2 release using an Infra Red Gas Analyzer (IRGA). Each fruit was placed in a separate enclosed jar of 3 L volume after the ambient CO_2 was measured. The CO_2 release was monitored by connecting the respiration jar to the IRGA 10 min after placing the fruit in the jar. The actual CO_2 was calculated by taking the difference between CO_2 after 10 min and ambient CO_2. The CO_2 released by the fruit was expressed as $mlkg^{-1}hr^{-1}$ adjusting for volume of the containers using the previously determined fruit volume. For detailed calculation see Appendix 2.

4.3.5 Pigment Determination: For the determination of total chlorophyll and carotenoids 1 g of fresh weight of banana peel was finely ground with liquid nitrogen and blended with 10 mL of 100% methanol. The sample was then placed on ice in the dark for 10 min before it was homogenized using an ultra-turrax® (IKA, Germany), 2 x 1 min bursts, waiting 5 min between bursts and centrifuged using (Sc-158T, Taiwan, R.O.C.) for 5 min at 3000 rpm. The supernatant

was decanted and placed in the spectrophotometer (Anthelie Advanced, Secoman, France) to read the absorbance at 470, 652, and 665 nm wavelengths. The concentration of total chlorophyll and carotenoids were calculated using the Wellburn (1994) equation. For detailed calculations see Appendix 3.

4.3.6 Total Soluble Solids (TSS): As extraction of juice from banana fruits was difficult, TSS determination was adapted according to Illeperuma and Jayasuriya (2002). Finely ground fresh (5 g) tissue was homogenized with 5 mL distilled water for 2 min using an ultra-turax® (IKA, Germany). The pooled fraction was centrifuged using (Sorvall RC 5C Plus, CA, USA) at 6000**g** for 10 min. The supernatant was decanted and TSS was measured using a calibrated hand held digital refractometer (PR-101, 0-45%, ATAGO CO, Ltd, Japan).

4.3.7 pH: The remaining supernatant was diluted to 80 mL with distilled water according to Illeperuma and Jayasuriya (2002) and the pH measured with a (Corning 430, NY, USA) pH meter.

4.3.8 Organic Acids: Five gram fruit fresh weight was taken from a frozen sample and blended with 95% ethanol for 4 min at a maximum speed with an ultra-turax homogenizer (Pérez *et al.*, 1997). The homogenate was centrifuged using (Sorvall RC 5C Plus, CA, USA) at 27000**g** for 10 min and the residue was washed twice with 80% ethanol. The supernatants were combined and adjusted to 5 mLg^{-1} of fresh weight (FW) with 80% ethanol. For organic acid separation 10 mL of sample from the ethanolic extract was taken and evaporated in the dark for four to five hrs to dryness with vacuum evaporator (Savant, Sc200, NY, USA). The dry residue was redisolved in 1 mL of 0.2 N H_2SO_4 and 0.05% disodium ethylene diamine tetra acetate (EDTA). All the fruit sample concentrates were deeply pigmented and would thus severely reduce analytical column life if they were to be injected directly onto the high performance liquid chromatography (HPLC) system (Richmond *et al.*, 1981). Therefore the sample was loaded onto a C_{18} Sep-Pak cartridge (isolute C_{18}, EC, Mid Glamorgan, UK) and eluted with up to 4 mL of the above solution. Total organic acids were contained in the elute. Before each sample was injected into the HPLC for analysis it was filtered through a 0.45 μm nylon filter to further ensure removal of any particulate impurities that might be present.

Standards: To determine the retention times of the analytes, standard solutions were prepared individually for each of the main organic acids found in the fruit (malic and citric) from

analytical reagent grade materials (Sigma Chemical Co., St. Louis, MO, USA). For calibration, a set of mixed organic acid standards (malic and citric acids) was prepared.

HPLC Conditions: Organic acids were analysed in a PerkinElmer (Wellesley, MA, USA) equipped with a Diode Array Detector (DAD) (series, 200) and fitted with a PerkinElmer Pump (series, 200) and Auto sampler (series, 200). Data were processed by means of a Network chromatography interface (NCI 900) and Totalchrom workstation software computing system (Waters, Milford, MA, USA). Isocratic separations of the compounds were made on a stainless steel (Waters Spherisorb 5 μm ODS2 4.6 x 250 mm) column. The mobile phase utilized for the elution consisted of a filtered (0.22 μm nylon) and degassed solution of $NH_4H_2PO_4$ (15 gL^{-1}), which was prepared with analytical reagent grade and adjusted to pH of 2.1 with phosphoric acid. Elution was monitored using a DAD UV detector set at 210 nm. The mobile phase flow rate was 1 mL min^{-1} with an injection volume of 20 μL. The concentration of each sample was calculated by comparison of retention times and peak areas to the areas obtained using the standard acid solutions with known concentrations. A set of calibration standards was run at the beginning and end of each batch of (10–15) samples. Concentrations were expressed as percentage of fresh weight. For detailed calculations see Appendix 4.

4.4 Results and Discussions

4.4.1 Percentage Weight Loss (PWL)

Fruit weight loss and rapid deterioration during storage are the major factors determining fruit quality and shelf-life. All fruits, irrespective of treatment, exhibited considerable PWL during the storage period, especially at high storage temperature. This weight loss was more pronounced as the storage period was extended and when fruits were transferred to room temperature for ripening after seven and five weeks storage at 12 and 15°C, respectively (Fig 4.1). This experiment shows that fruit weight loss has an inverse relationship with fruit shelf-life and quality. More rapid and higher weight loss was noted from fruits stored at 22°C as compared to the other storage temperatures and thus fruits were stored for a shorter period of three weeks. This can be related to the effect of temperature on fruit transpiration and evaporation, and hence higher weight loss of fruits. This is in agreement with Lebibet *et al.* (1995) that higher storage temperature increased the capacity of banana fruits to loose water and hence to increased weight

loss. Despite this, waxing significantly ($P<0.001$) reduced PWL at all storage temperatures as compared to the other treatments, especially as compared with control fruits (Fig 4.1). As a result waxing extended the fruit shelf-life up to 56, 42 and 24 days at 12, 15 and 22°C, respectively, while the shelf-life of control being 35, 35 and 14 days, respectively (Tables 4.1-4.3). Similar results were reported by several workers for banana (Banks, 1984), for various fruits (Drake, 1997; Techawongstien, 1999), for avocado (Durand *et al.*, 1984), and for mango (Lazan *et al.*, 1990b). This reduced weight loss can be ascribed to the effect of wax covering gas exchange sites of the fruit, and modification of the atmosphere around the fruit. It has, also been reported by Amarante and Banks (2001) and Banks *et al.* (1997), that edible surface coatings are applied to fruits to improve cosmetic features of the fruit and to reduce deterioration by suppressing water loss or by achieving MA benefits. This is in close agreement to the result obtained from this experiment. Burdon *et al.* (1993) previously reported that the removal of epicuticular wax from the fruit surface increased the amount of water loss from the fruit. Waxing might, therefore, be an alternative means of preserving fruits for an extended time mainly via reducing fruit water loss, which is one of the major fruit deterioration factors in a tropical environment. Similar to waxing GA_3 and IBA also resulted in reduced PWL. GA_3 at 12°C and IBA at 12 and 15°C exhibited significantly lower PWL ($P<0.001$) than control fruits and showed a trend of decreased rate at the other storage temperatures. Postharvest treatment of banana (Stover and Simmonds, 1987) and mango (Khader, 1992) with GA_3 extended the pre-climacteric period and hence shelf-life. Similarly, Purgatto *et al.* (2001) reported that auxin extended ripening and decrease the rate of senescence of banana fruit mainly by counteracting the effect of ethylene within the fruit tissue. In addition, Vendrell and Palomer (1997) reported both, GA_3 and IAA inhibit ripening in most climacteric fruits mainly by impeding the activity of ethylene which triggers ripening. These growth regulators can also be an alternative technique to control and reduce fruit water loss after harvest.

Techawongstien (1999) reported that exposure of fruits to undesirable temperature can result in many physiological disorders; freezing injury, chilling injury (CI) and/or heat injury and that the sensitivity of these disorders is highly influenced by both, temperature and relative humidity. From Fig 4.1 it can be inferred that the higher the ambient temperature to which fruits are exposed the more significant the effect on fruit weigh loss in particular, and shelf-life in general. Modifying the atmosphere surrounding the fruit by waxing and postharvest application of GA_3

and IBA to retard ripening, therefore, become possible alternative methods of shelf-life enhancement, especially in tropical environments.

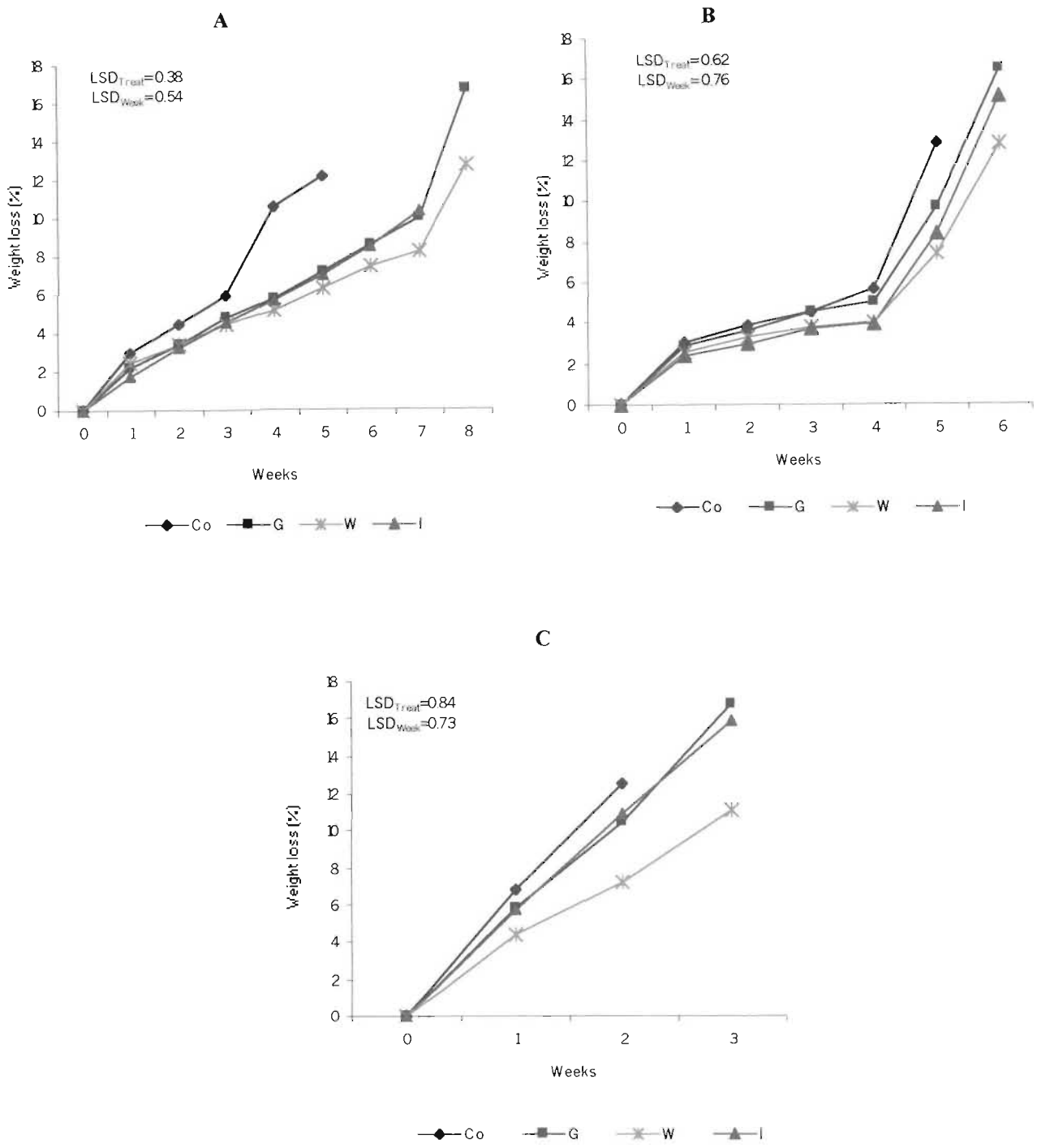


Fig 4.1 Percentage weight loss of banana fruits during storage as affected by storage conditions at 12^{0}C (A), at 15^{0}C (B) and at 22^{0}C (C). For all figures, data points are means of six replicates. Least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and time (LSD_{Week}) are shown. Weight losses for each fruit were monitored to the table ripe stage (scale 6). Note data collection for control fruits terminated shortly. Co=control, G= GA_3, W=waxing, I= IBA

Table 4.1 Physico-Chemical Characteristics of Banana Fruits Stored at 12°C as Affected by Different Postharvest Treatments

Treatments	Shelf-life (days)*	TSS (°Brix)	pH	Malic (%)	Citric (%)
Initial	-	1.53[a]	5.16[a]	0.021[a]	0.016[a]
Control	35	10.23[b]	4.75[b]	0.036[b]	0.056[b]
GA_3	50	10.60[b]	4.76[b]	0.041[b]	0.053[b]
IBA	48	10.80[b]	4.77[b]	0.037[b]	0.052[b]
Waxing	56	10.30[b]	4.72[b]	0.033[ab]	0.057[b]
LSD		0.81	0.11	0.013	0.01
P value		NS	NS	0.007	0.008

Table 4.2 Physico-Chemical Characteristics of Banana Fruits Stored at 15°C as Affected by Different Postharvest Treatments

Treatments	Shelf-life (days)*	TSS (°Brix)	pH	Malic (%)	Citric (%)
Initial	-	1.53[a]	5.16[a]	0.021[a]	0.016[a]
Control	35	10.97[c]	4.71[b]	0.053[b]	0.054[b]
GA_3	41	10.53[bc]	4.71[b]	0.048[b]	0.052[b]
IBA	38	10.03[b]	4.77[b]	0.040[b]	0.046[b]
Waxing	42	10.27[bc]	4.67[b]	0.041[b]	0.055[b]
LSD		0.81	0.11	0.013	0.01
P value		NS	NS	0.007	0.008

Table 4.3 Physico-Chemical Characteristics of Banana Fruits Stored at 22°C as Affected by Different Postharvest Treatments

Treatments	Shelf-life (days)*	TSS (°Brix)	pH	Malic (%)	Citric (%)
Initial	-	1.53[a]	5.16[a]	0.021[a]	0.016[a]
Control	14	9.90[b]	4.76[b]	0.038[b]	0.032[bc]
GA_3	22	10.33[bc]	4.73[b]	0.029[ab]	0.031[b]
IBA	21	10.97[c]	4.73[b]	0.057[c]	0.048[d]
Waxing	24	10.33[bc]	4.75[b]	0.035[b]	0.042[cd]
LSD		0.81	0.11	0.013	0.010
P value		NS	NS	0.007	0.008

Means in each columns of all tables followed by the same letter are not significantly different at 5% level. NS= Not Significant.

*** Days were determined when the fruit reaches table ripe stage (Scale -6)**

4.4.2 Firmness Change

As fruits ripen they soften or decrease in firmness, largely because pectins, comprising the middle lamella of cell walls, are solubilised (Wills *et al.*, 1998). In many fruits, firmness is related to the stage of developmental maturity (Aung *et al.*, 1996). Fruit firmness decreased during ripening regardless of treatments. This was more pronounced at higher storage temperatures. Consequently, fruits at higher storage temperature maintained acceptable condition only for a maximum of three weeks (Fig 4.2 C). Despite the general decrease of firmness, waxing, GA_3 and IBA significantly ($P<0.001$) retarded fruit softening as compared to control fruits, especially at higher temperature. Waxing yielded better results than the others (Fig 4.2). This is in agreement with the previous report in avocado by Drake (1997) and Durand *et al.* (1984) as well as in apples by Meheriuk and Porrit (1972). The authors reported that waxing reduces both, fruit weight loss and softening. This phenomenon might be related to the effect of an increased endogenous CO_2 level, as the respired CO_2 can't escape easily through the stomata when the fruits are waxed. This in turn will retard fruit softening. In addition, according to Zagory (1998) the elevated CO_2 suppresses plant tissue sensitivity to the effect of the ripening hormones. This indicates that waxing might be commercially used to extend fruit shelf-life in tropical countries where there is lack of storage facilities. Similarly, GA_3 and IBA application exhibited promising results especially as compared to control untreated fruits. This might be related to the effect of these growth regulators to retard the ripening process. According to Desai and Deshpande (1978) the retarded fruit softening by GA_3 is related to its effect on degradation of polymers like starch, cellulose and hemicelluloses. Similarly, delayed starch hydrolysis by IAA treated banana slices were reported recently by Purgatto (2001), although a contrasting result was found by Desai and Deshpande (1978) that IAA enhanced fruit softening. This result may have been related to poor penetration of auxin, which may vary depending on the concentration and application method (vacuum or dip). With proper application at optimal concentrations GA_3 and IBA may also be viable treatments to extend banana shelf-life. The progressive increase in softening of the tissue together with a change in colour of the skin or flesh are some of the easily recognizable changes that accompany ripening in climacteric fruits (Hobson, 1981). Fruit softening is also an important attribute that leads to fruit deterioration and consumer rejection. 'Green ripe" in banana is a good example of the effect of softening on fruit quality. The fruit softens while the colour still remains green. Waxing, GA_3 and IBA can be used as alternative methods to retard fruit softening in particular and increase fruit shelf-life in

general. This is especially so in poorly resourced regions of tropical countries where the fruit commonly grows.

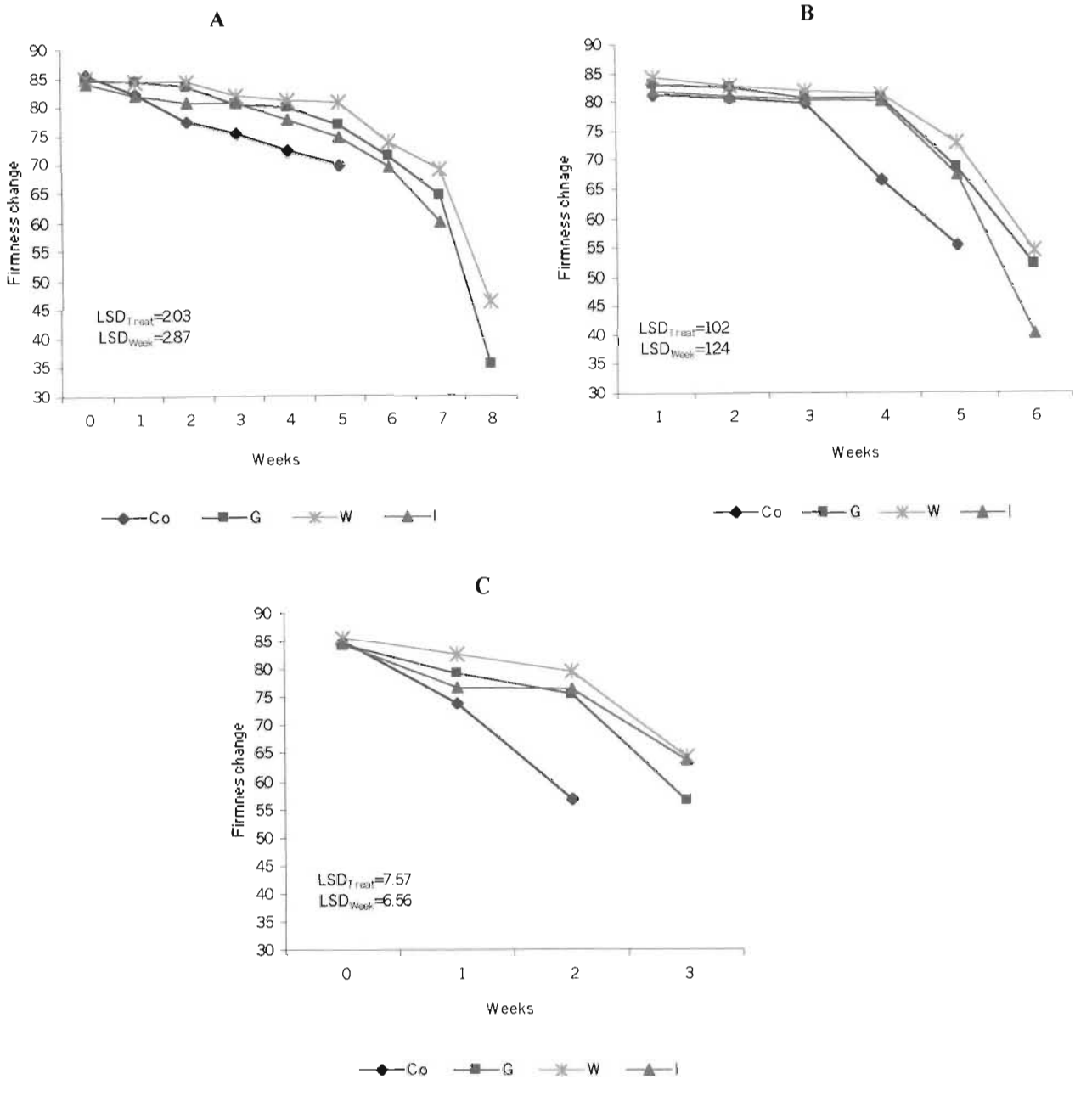


Fig 4.2 Firmness changes of banana fruits during storage as affected by storage conditions at 12^{o}C (A), at 15^{o}C (B) and at 22^{o}C (C). For all figures, data points are means of six replicates. Least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and time (LSD_{Week}) are shown. Firmness changes for each fruit were monitored to the table ripe stage (scale 6). Note data collection for control fruits terminated shortly. Co=control G= GA_3, W=waxing, I= IBA

4.4.3 Skin Colour Change

Fruit skin colour is routinely used by both, market operators and consumers, to determine fruit ripeness (Dadzie, 1998; Wills *et al.*, 1989) and quality. Banana undergoes significant textural and colour transformation as the ripening process progresses (Chen and Ramaswamy, 2002). In this experiment increased fruit colour from the initial stage was evident in all fruits irrespective of treatment and storage temperature (Fig 4.3). The storage temperature had an impact on colour development of all fruits and, hence, shelf-life. More intense colour development was exhibited after eight weeks at 12°C as compared to six and three weeks at 15 and 22°C, respectively. Similar report by Chen and Ramaswamy (2002), Lebibet *et al.* (1995) and Proft *et al.* (1998) described that rapid colour change was evident in banana fruit at higher temperature, although a contrasting result was reported by Terblanche (1999) in citrus fruits where he found more fruit colour change at lower (18°C) than at higher (24°C) temperature. Waxing as well as GA_3 and IBA application retarded colour development significantly ($P<0.001$) and the shelf-life was extended as compared to the control fruits at all storage temperatures (Fig. 4.3 and 4.4). Similar results were reported by Banks (1984) for waxing, Babbitt *et al.* (1973) as well as Vendrell and Palomer (1997) for GA_3 and IAA, where all these treatments retarded colour development. Colour scale 6 was reached after 35 days storage at 12°C in control fruits (Fig 4.5), whereas it took 56, 50 and 48 days to reach the same mark in fruits treated with waxing, GA_3 and IBA respectively (Table 4.1). The effect of waxing may be related to the high build up of CO_2 within the fruit, which retards fruit ripening and, hence, colour development. The lack of colour change after application of GA_3, may be related to a decreased chlorophylase activity as reported by Khader (1992) for mango and Vendrell and Palomer (1997) for tomato. Moreover, Vendrell and Palomer (1997) reported that auxin suppresses the expression of developmental genes, like those for colour development. However, contrasting results on colour development of banana fruits after IAA treatment were reported by Desai and Deshpande (1978). The authors found that IAA application considerably increases the fruit colour rating according to the von Losecke's colour chart. This variable result of the effect of auxin on fruit ripening in general and in colour development in particular, can again be explained by limited penetration ability of the treatment into the tissue when applied by dipping or if a low concentration is used. Fruit colour development is an essential parameter used to evaluate the rate of fruit ripening and, hence, shelf-life. Waxing, GA_3 and /or IBA are therefore a promising technique to retard colour development during fruit storage period even at higher storage temperatures, which reveals the

potential of these treatments in a tropical environment. A postharvest wax treatment and dip-treatment of fruits with GA_3 and IBA to supplement and possibly increase the endogenous GA_3 and auxin concentration to overcome rapid colour development might become a recommended postharvest treatments in areas where there is lack of storage facilities to retard fruit ripening.

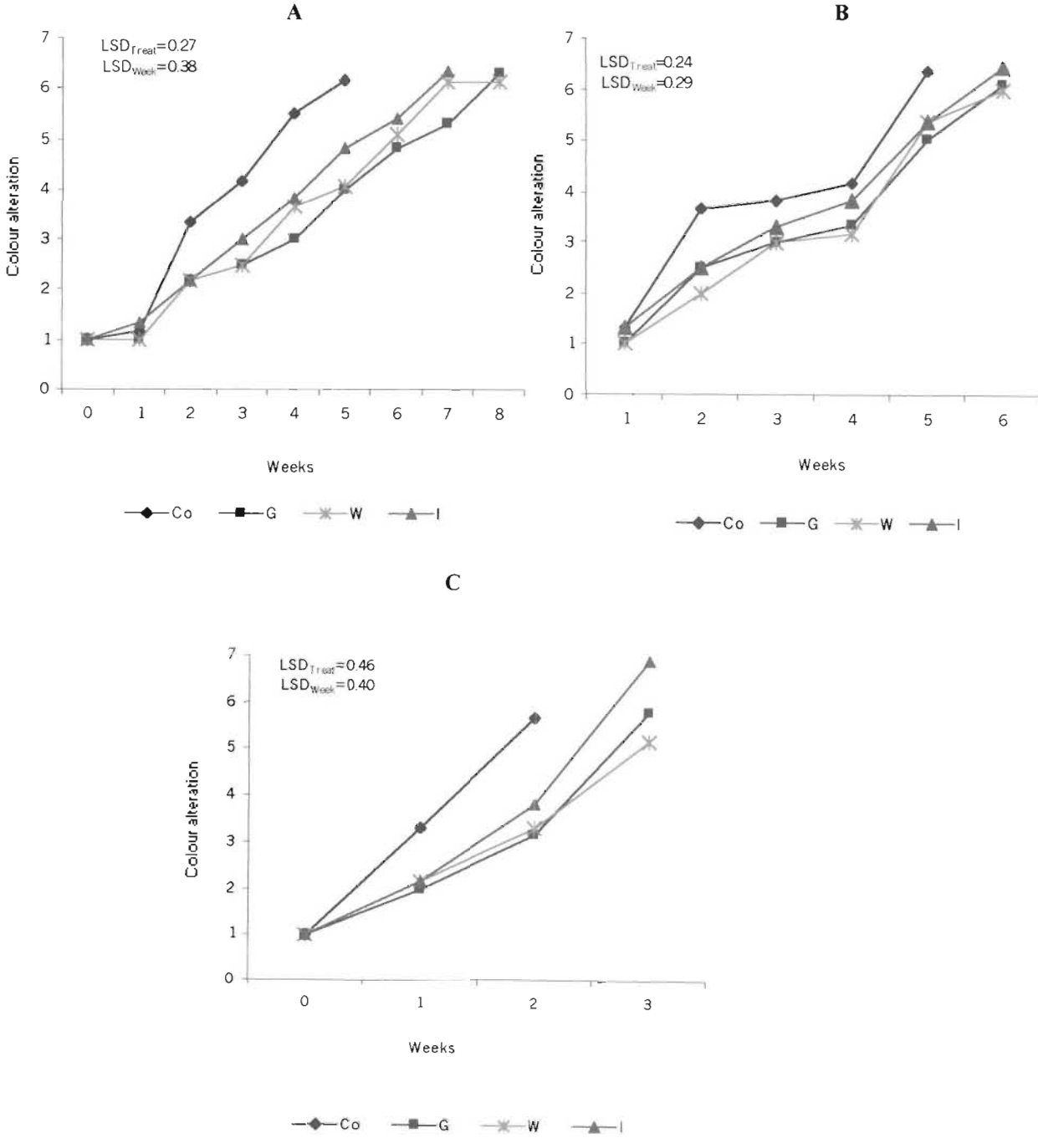


Fig 4.3 Colour changes of banana fruits during storage as affected by storage conditions at 12°C (A), at 15°C (B) and at 22°C (C). For all figures, data points are means of six replicates. Least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and time (LSD_{Week}) are shown. Colour alteration for each fruit was monitored to the table ripe stage (scale 6). Note data collection for control fruits terminated shortly. Co=control, G= GA_3, W=waxing, I= IBA

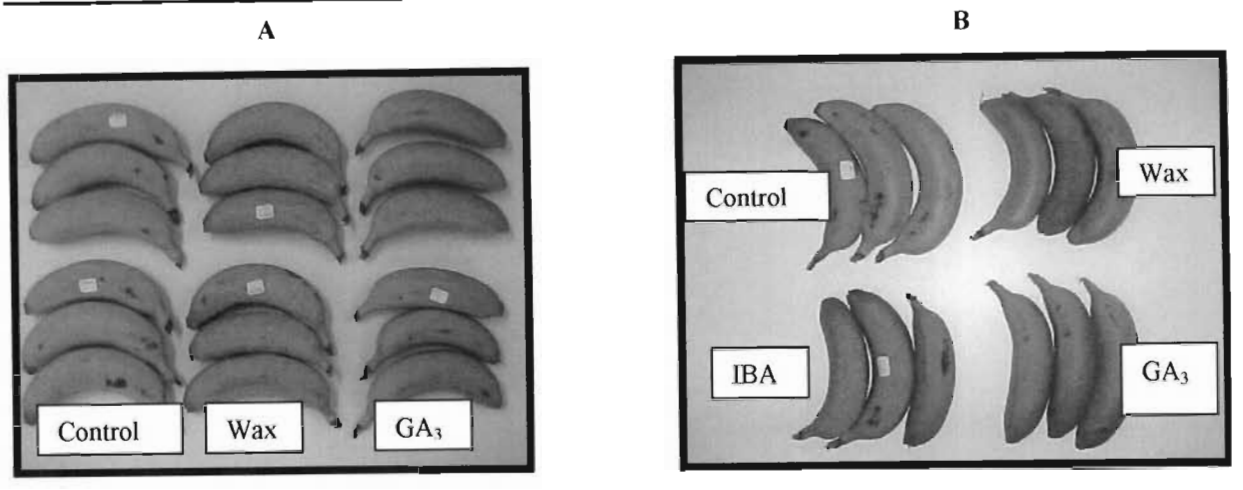


Fig 4.4 Fruit colour development as affected by different treatments and storage temperatures. After two weeks storage at 12°C (A) and after one week storage at 22°C (B). Note fruits waxed or treated with either GA_3 or IBA maintain green colour of the fruit.

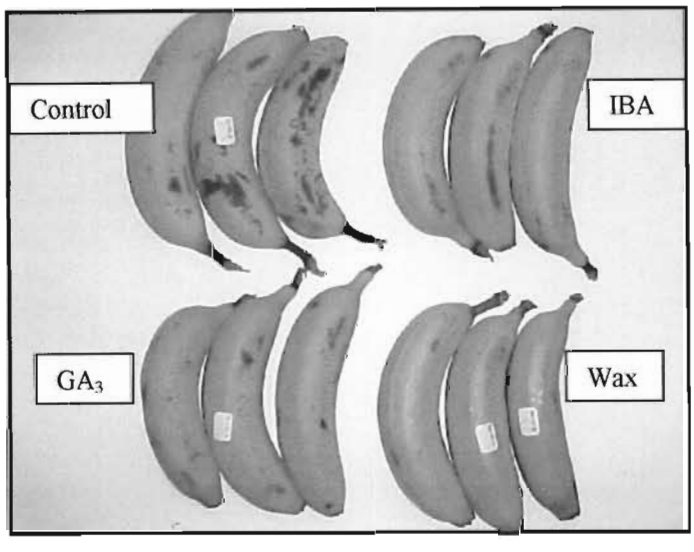


Fig 4.5 Fruit Colour development as affected by different treatments at 12°C after five weeks storage. Note that control fruits started to bruise.

4.4.4 Respiration Rate

Fruit respiration rate is a major factor determining fruit quality after harvest and shelf-life. In general, there is an inverse relationship between respiration rate and postharvest life of fresh commodities. The storage temperatures significantly affected respiration rate and shelf-life of the fruits. Temperature was the most important postharvest factor affecting fruit respiration rate (Saltviet, 2002). On average fruits at 12°C exhibited a lower respiration rate after eight weeks

storage period as compared to the fruits at 15 and 22°C after six and three weeks storage, respectively. This may be ascribed to the effect of storage temperatures on the metabolic activity of the fruit and hence the respiration rate. A similar result was reported by Proft *et al.* (1998) where storage temperature strongly stimulates a higher release of CO_2 from banana fruits. Irrespective of the effect of temperature on the respiration rate, waxing, GA_3 and IBA significantly reduced ($P<0.001$) the respiration rate, although waxing was most effective. This was evident at all storage temperatures (Fig 4.6). On the other hand control fruits had a significantly higher respiration rate at all storage temperatures and hence short shelf-life. This increased respiration rate of control fruits indicates the effect of temperature on fruits in the higher tropical environment. A decreased respiration rate due to waxing was previously reported for avocado by Bender *et al.* (1993), for apples by Hagenmaire and Baker (1993) as well as Meheriuk and Porrit (1972) and for banana by John and Marchal (1995). This may be related principally to alterations in the permeability characteristics of the fruit skin. A wax coating forms an additional barrier through which the gas must permeate. According to John and Marchal (1995) coating extends the storage life of banana and plantain by slowing down gaseous exchange between fruits and the atmosphere thus delaying the onset of the climacteric phase. Previously, it was also, reported by Babbitt *et al.* (1973) for tomato and Vendrell and Palomer (1997) for other climacteric fruits that GA_3 and auxin retard ripening mainly by acting antagonistically to the effect of ethylene, which increases fruit respiration rate. More recently Purgatto *et al.* (2001) reported that auxin overcomes the effect of ethylene as a result IAA markedly retarded fruit respiration rate as compared to control, untreated fruits. On the other hand, contrasting result of increased respiration was reported by Shanmugavelu *et al.* (1992) after application of 2,4-D for banana fruits with a concentration of 500 ppm and above.

An increased respiration rate after harvest is one the major factors that causes fruit deterioration, which results in reduced shelf-life of most tropical fruits. Investigating methods to reduce or retard respiration rate after harvest is therefore very important to extend fruit shelf-life. The treatments of waxing, GA_3 and IBA were all found to decrease the respiration rate at all storage temperatures. This implies that these treatments might be possible alternatives in regions where storage infrastructure is lacking.

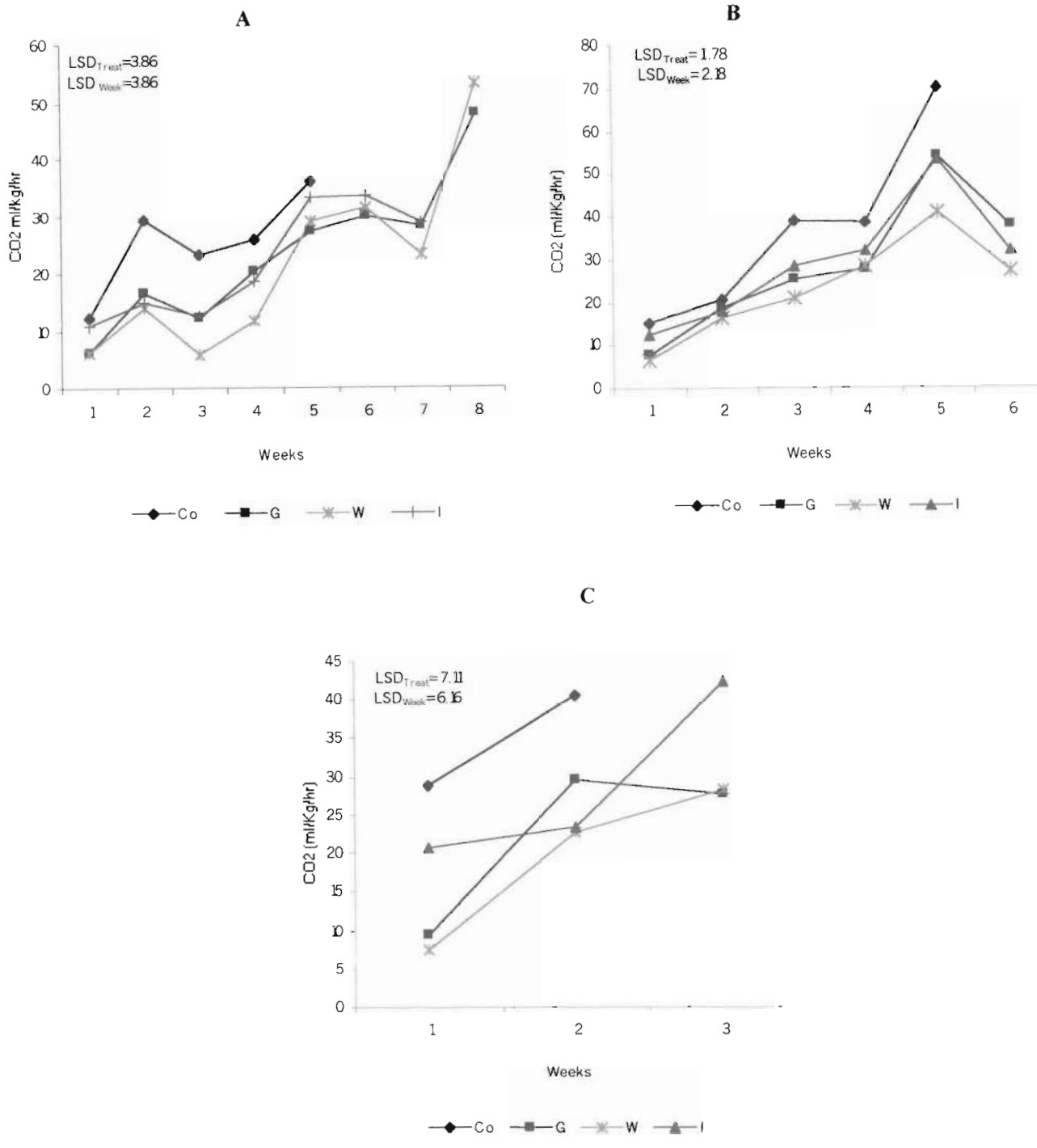


Fig 4.6 Respiration rate of banana fruits during storage as affected by storage conditions at 12°C (A), at 15°C (B) and at 22°C (C). For all figures, data points are means of six replicates. Least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and time (LSD_{Week}) are shown. Respiration rate for each fruit was monitored to the table ripe stage (scale 6). Note data collection for control fruits terminated shortly. Co=control, G= GA_3, W=waxing, I= IBA

4.4.5 Pigment Determination

4.4.5.1 Total Chlorophyll: Some of the major compositional changes in fruits during ripening are chlorophyll degradation and synthesis of other pigments. Decomposition of chlorophyll may, in many cases, be quite rapid and dramatic, which is part of the chloroplast - chromoplast transition (Kays, 1991). After eight, six and three weeks storage of fruits at 12, 15 and 22°C, respectively, fruits showed no significant difference ($P<0.05$) in total chlorophyll concentration among all treatments, although all treatments exhibited rapid degradation of chlorophyll during the storage period (Fig.4.7). Dadzie (1998) for banana and Deli *et al.* (1996) for pepper reported that the chlorophyll content of fruits decreased slowly with ripening. Although there was no significant difference between treatments, waxing and GA_3 exhibited a trend of higher concentration at all storage temperatures. This may be related to the effect of these treatments in retarding fruit ripening as the chlorophyll concentration decreased with fruit ripening. Chlorophyll concentration measurement is an objective method to evaluate colour change in fruits during storage and ripening. All the treatments that resulted in an extended storage life did not affect the process of chlorophyll degradation, but slowed the rate of degradation. This might be useful during extended storage period to slow down rapid colour disappearance.

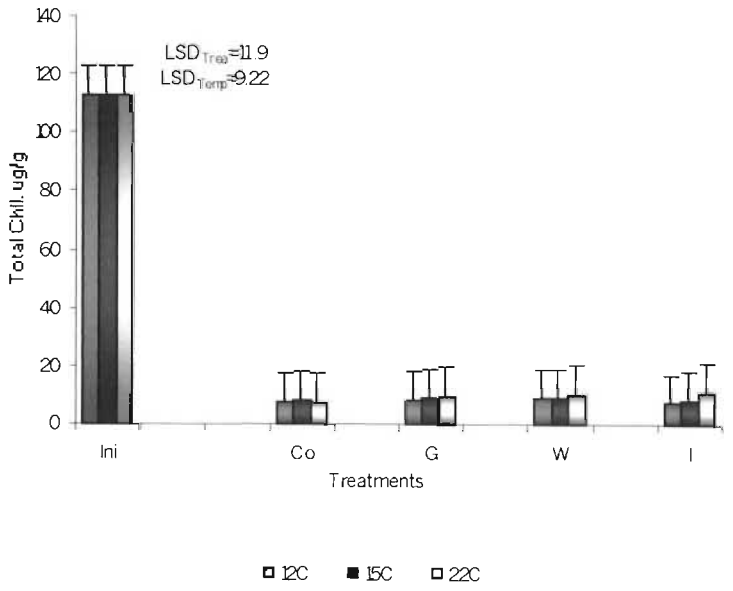


Fig 4.7 Total chlorophyll concentrations of banana fruit peel before storage and at full ripe stage as affected by different treatments and storage temperatures. Data points are means of six replicates. Standard error mean of difference (+SED) and Least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and time (LSD_{Week}) are shown. Ini= initial, Co=control, G= GA_3, W=waxing and I= IBA.

4.4.5.2 Total Carotenoids: Results from this study showed that there was no significant difference ($P<0.05$) among treatments and storage temperatures, although there was a trend to increased value at 22°C (Fig 4.8). There was also no measurable difference in the total carotenoid concentrations between the initial and when the fruit reaches table ripe stage after storage. It has been reported earlier (Robinson, 1996; Thompson and Burden, 1995) that during banana ripening, peel colour yellowing is due to chlorophyll degradation and gradual unmasking of the already existing carotenoid pigments. This implies carotenoids in banana fruits are not synthesised concomitant to chlorophyll degradation as in some other fruits, but are only revealed as chlorophyll is degraded (John and Marchal, 1995). This is in contrast to the results reported by Deli *et al.* (1996) and Mínguez-Mosquera *et al.* (1994) in paprika where a considerable increase in carotenoid content was evident concomitant to ripening. This study revealed the potential feasibility of extending fruit shelf-life without affecting fruit colour quality attributes. It can therefore be inferred that waxing, GA_3 and even IBA can extend the shelf-life of the fruit with its preserved quality both at low and high storage temperature.

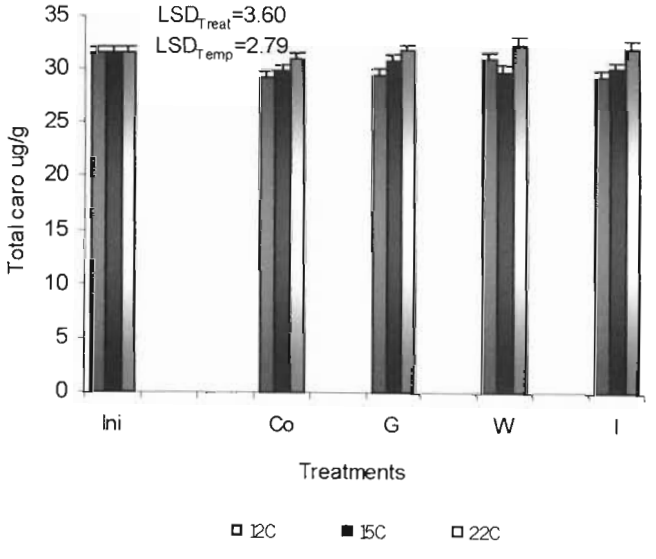


Fig 4.8 Total carotenoid concentrations of banana fruit peel before storage and at full ripe stage as affected by different treatments and storage temperatures. Data points are means of six replicates. Standard error mean of difference (+SED) and Least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and time (LSD_{Week}) are shown. Ini= initial, Co=control, G= GA_3, W=waxing and I= IBA.

4.4.6 Total Soluble Solid (TSS)

Fruit TSS has been routinely used to assess fruit quality. Sugars are the main soluble solid component in most ripe fruits including bananas (Gauhl *et al.*, 1998). Starch-sugar conversion

during ripening is one of the most evident changes in climacteric fruits such as banana (Chang and Hwang, 1990). In this experiment all the treatments resulted significantly ($P<0.05$) increased TSS as the fruit ripened. TSS reached its maximum level as the fruit reached its full ripe stage (Table 4.1-4.3). This is in agreement with previous reports by Mustaffa *et al.* (1998) for banana and Illeperuma and Jayasuriya (2002) for mango. This increase in TSS alteration of fruits may be related to the general physiochemical change of fruit concomitant to ripening, as the starch of the fruit transformed to simple sugars during ripening. Different treatments at different storage temperatures exerted an effect on the fruit TSS content. Control fruits at 15 and fruits treated with IBA at 22°C exhibited significantly increased TSS from each other (Tables 4.2-4.2). However, there was no significant difference between the rests of treatments at all storage temperatures. This reveals waxing and GA_3 can be used to extend fruit shelf-life, while still preserving the quality of the fruit, especially with respect to TSS.

4.4.7 pH

Fruit pH decreased in all treatments and storage temperatures during the storage period. A similar reduction in fruit pH in banana was reported previously by Robinson (1996). On the other hand contrasting result was reported for mango by Illeperuma and Jayasuriya (2002). Irregular changes of fruit pH during ripening were also reported for banana fruits by Mustaffa *et al.* (1998). Despite this significantly reduction ($P<0.001$) of fruit pH during storage, there was no significant difference ($P<0.05$) among all the treatments and storage temperatures (Tables 4.1-4.3). Therefore, extending fruit shelf-life by waxing, GA_3 or IBA application does not affect this aspect of fruit quality. This implies the application potential of these treatments to extend shelf-life of fruits in areas where there is lack of storage facilities.

4.4.8 Organic Acids

Fruit sugar and acid concentration ratio has a marked influence on the sensory quality of fruits (Ackermann *et al.*, 1992). Like sugars, organic acids contribute to fruit flavour. Results from this experiment revealed that organic acids increased significantly ($P<0.001$) during fruit ripening irrespective of treatment at all storage temperatures. Several workers reported similar increase organic acids during ripening in banana (Inaba and Nakamura, 1988; John and Marchal, 1995) in cherimoya (Alique and Oliveira, 1994) and in onion bulb (Salama *et al.*, 1990).

4.4.8.1 Malic Acid: The malic acid concentration increased in all fruits during ripening process, but control fruits and IBA treated fruits showed a trend of higher values at the end of ripening as compared to the others at 15 and 22°C, respectively (Tables 4.1-4.3). This might be related to the ripening effect of the fruits. It has been reported earlier by Wills *et al.* (1983) that malic acid in banana fruits increased during ripening. The malic acid concentration at 15 and 22°C was noted to be higher than that of fruits stored at 12°C. This may further be related to the effect of temperature in increasing fruit ripening, and thus increased malic acid. This is supported by the report of Salama *et al.* (1990) where there was a significant influence of storage temperature on sugar and acid concentrations. However, there was no appreciable difference among the rest of the treatments and storage temperatures. This is another good indication of these treatments (waxing, GA_3 and/or IBA) to be used as alternative methods to extend fruit shelf-life with limited quality defects.

4.4.8.2 Citric Acid: The concentration of citric acid during the fruit ripening period followed the same profile as the malic acid ones. Fruits of all treatments showed increase in acidity during ripening, regardless of the treatment. Similar results were reported in cherimoya by Alique and Oliveira (1994). On average fruits stored at 22°C had lower acid concentrations than other storage temperatures. However, there was no significant difference in citric acid concentration between treatments (Tables 4.1-4.3). Theses results further confirms that fruit quality is not affected by an extended storage period. Waxing, GA_3 and IBA appear to extend shelf-life with no detrimental effect to the acid quality, even at higher storage temperatures.

4.5 Conclusion

Postharvest management of fruits requires a knowledge of the desired storage duration and commodity response to storage conditions, as well as quality specifications for the ultimate product (Shewfelt, 1986). Banana is one of the commonly grown climacteric tropical fruits. Climacteric fruits exhibit rapid deterioration after harvest mainly due to excessive weight loss and high respiration rate. This becomes very severe in a tropical environment with no or insufficient storage and cold chain facilities. Fruit deterioration is not, however, only determined by weight loss and respiration rate. Higher softening rates and colour change are also other attributes routinely used to evaluate fruit deterioration rate and, hence, shelf-life. It was noted from the current study that all fruits showed increased water loss, softening, colour development and respiration rate concomitant to ripening at all storage temperatures. Waxing, GA_3 and IBA,

however, can reduce almost all the above-mentioned parameters even at higher storage temperatures. Through minimizing fruit water loss, softening, respiration rate and/or colour development of fruits, these treatments can be used as alternative approaches to solve the common problems of tropical fruits, and especially banana. Quality evaluation of TSS, pH, total chlorophyll, carotenoids and organic acids after storage resulted no measurable difference among treatments at all storage temperatures. This implies that fruit shelf-life can be extended without any significant effect on fruit quality.

While decreased temperature and a managed cold chain will clearly enhance shelf-life, this work has shown that considerable benefit can be gained through single pack house procedure such as waxing and PGRs application, even where a poor cold chain infrastructure exists.

Chapter 5: Effect of GA_3, Waxing and Micro-Perforated Bag on Shelf-Life and Selected Quality Attributes of Papaya Fruit

5.1 Abstract

Like most tropical climacteric fruits papaya is characterized by a relatively short shelf-life. To extend the shelf-life and to investigate the effect of various postharvest treatments on shelf-life and quality parameters a study was conducted on papaya (*Carica papaya*) cv. Hortus Gold. The treatments included gibberellic acid (GA_3), waxing and micro-perforated bags (MPB) as compared to control fruits. Fruits were held at four different temperatures (5.5, 7, 10 or 22°C). Percentage weight loss (PWL), firmness, skin colour change and respiration rate were evaluated on a weekly basis during the storage period. Selected quality attributes such as total soluble solids (TSS), titratable acids (TA), pH and sugar:acid ratio were also measured after storage. Based on the colour change and softening days to ripen was determined. PWL, softening, yellowing and respiration rate increased irrespective of treatments as the storage time and temperature was increased. MPB and waxing significantly ($P<0.001$) reduced PWL and softening compared to the control fruits. This was consistent for all storage temperatures. GA_3 resulted in significantly lower ($P<0.001$) PWL compared to the control at 7, 10 and 22°C, but not at 5.5°C. Moreover, waxing and GA_3 treatments significantly reduced colour development and the respiration rate at all storage temperatures, although an increased respiration rate was evident in MPB at 5.5 and 22°C. However, there was no off-flavour and/or fungal development. Fruits at 7 and 10°C exhibited an increased shelf-life up to eight and six weeks, respectively, while fruits at 5.5°C showed chilling injury, which resulted in an increase in respiration rate and PWL. Similarly, fruits at 22°C showed a rapid respiratory climacteric after two weeks of storage and deteriorated very rapidly. Waxing and GA_3 increased the shelf-life at all storage temperatures compared to the control. There was no measurable difference among treatments in their TSS, TA, pH and sugar:acid ratio at all storage temperatures. This indicates the applicability of these treatments to extend shelf-life without affecting the quality. Waxing, GA_3 and MPB are, therefore, promising means of controlling papaya postharvest loss and, hence, increase shelf-life with no significant effect to the quality within the conditions of poor infrastructure and poor storage facilities.

Key words: GA_3, micro-perforated bags, papaya and waxing.

5.2 Introduction

Cultivation of papaya has spread from tropical Central America, where it originated, to all tropical and warmer subtropical areas of the world. Despite its popularity, little attention has been given to postharvest maintenance and handling, especially in poorly resourced countries. Papaya fruits have a poor quality outturn and high postharvest losses if harvesting, postharvest treatments and handling techniques are inadequate and inappropriate (Medlicott, 2001). Fruits stored under ambient tropical conditions have a maximum storage life of seven days (Maharaj, 1988). This short shelf-life results in poor fruit quality, that limits the availability of the fruit in local markets and export to distant markets. A major part of postharvest research is devoted to reducing respiration and other metabolic processes associated with quality reduction by manipulating the external environment (Saltveit, 2002). Most of the postharvest losses occur during the rapid and/or uncontrolled ripening of the fruit after harvest. These losses can assume a considerable economic and social importance, especially in tropical regions, which include a large proportion of developing countries (Wills *et al.*, 1998) and which suffer from lack of adequate storage facilities. Understanding the biological and environmental factors involved in fruit deterioration and use of postharvest technology procedures are, therefore, important to delay senescence and maintain the best possible quality (Kader, 1992a). Postharvest studies of papaya have been conducted in various areas and different workers reported different outcomes. It has been reported previously that lower storage temperatures retard fruit respiration rate (An and Paull, 1990), softening (Lam, 1990) and ripening (An and Paull, 1990; Lazan *et al.*, 1993) by reducing weight loss and thus increase shelf-life. However, this becomes difficult to apply in most of the tropical areas where cold chain becomes difficult to maintain. Different packaging material systems were found to improve postharvest life of papaya fruits. According to Lazan *et al.* (1990a) fruit weight loss was reduced considerably by packaging in polyethylene bags as compared to non-sealed control fruits. Similarly, Paull and Chen (1989) reported delayed water loss, softening and colour development by plastic wrapping and waxing. However, high build-up of CO_2, development of off-flavour and saprophytic fungi has been reported inside the polyethylene bags, as the bags were highly impermeable (Chaplin *et al.*, 1982; Miller *et al.*, 1986; Nunes *et al.*, 1998; Thompson and Lee, 1971). A potential for extension of storage life by PGR application was also reported for mango by Parikh *et al.* (1990). However, most of the reports were conducted in developed countries with good storage chains and facilities, and aimed at export. Moreover, Ferris (1997) added that GA_3 is only effective in retarding ripening at high

humidity but not at low humidity storage conditions. Similarly, for all the above mentioned storage methods very little information is available concerning the quality of the fruit after storage.

This experiment was conducted to investigate and evaluate the effect of micro-perforated polypropylene material, postharvest GA_3 application, and waxing on the postharvest maintenance and shelf-life of papaya cv. Hortus Gold within the context of a poor infrastructure.

5.3 Materials and Methods

Papaya fruits (*Carica papaya* cv. Hortus Gold) were obtained from Ukulinga, the University of Natal experimental farm in Pietermaritzburg, KwaZulu-Natal, South Africa. For this study freshly harvested, clean, bright, firm and bruise free fruits were used. On arrival at the laboratory of the Horticultural Science Department, University of Natal, fruits were washed with tap water and dipped in a fungicide solution containing 670g/1000L (w/v) of Magnate sulphate® (Makhteshim-Agan, Israel). After drying in air, fruits were randomized to the following treatments with five replications to each treatment: Control (untreated), GA_3, Waxing (citrus carnauba), or micro-perforated bags with 9 µm perforation and coated with anti-mist (MPB). For GA_3 fruits were dipped into an aqueous solution of 100 ppm for 30 min. For the wax treatments, fruits were lightly waxed with citrus carnauba (to allow gas exchange) using a cloth, and allowed to dry. Fruits were then stored at 5.5, 7, 10 or 22°C while at the same time data for PWL, firmness, colour change, and respiration rate were recorded weekly. Fruits were allowed to ripen at the designated temperature naturally until the visually table ripe stage (scale 6) (Fig 5.1) and fruits moved to room temperature for final ripening. Time taken to ripen was noted and used to determine the shelf-life. After fruits were fully ripened, a portion of the fruit pulp was cut longitudinal (from end to end) to minimize within fruit variation in sugar concentrations (Garner *et al.*, 2001) and stored in a deep freezer for further analysis of TSS, pH, TA and sugar:acid ratio. All data were subjected to analysis of variance (ANOVA) using the GenStat® 5th edition (VSN, 2001). Treatments were compared at $P<0.05$ according to Fisher's protected LSD Test.

5.3.1 Percentage Weight Loss (PWL): Each fruit was weighed prior to storage and before any treatment was applied. At weekly intervals fruits were reweighed and PWL calculated. The difference in weight was expressed as percentage weight loss and calculated as follows:

$$PWL = \frac{Initial\ weight - actual\ weight}{initial\ weight} * 100$$

5.3.2 Firmness Change: Firmness of the fruit was measured using firmness tester (Mechanical densimeter, FL, USA) (Fig 3.1) with a reading ranging from 0-100 (the higher the reading the firmer the fruit and vice versa). Data was collected at weekly intervals starting from the day of harvest. Data was taken from two different sides of the fruit and the mean result was calculated.

5.3.3 Skin Colour Change: Colour development of the fruits was scored numerically using standard of papaya ripening chart (Fig 5.1) adopted from banana charts (von Loesecke, 1949). The colour plates ranging from 1-7 where: 1=green; 2= green with yellow tracks; 3= more green than yellow; 4=more yellow than green; 5=yellow with green tips, 6= all yellow and 7= full yellow with some black spots.

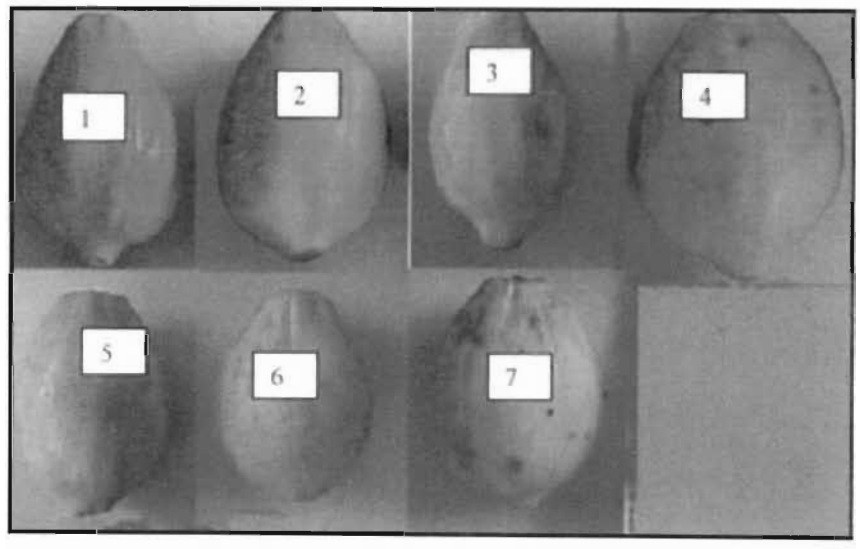


Fig 5.1 Suggested papaya fruit colour development chart adopted from banana colour development chart by von Loesecke (1949).

5.3.4 Respiration Rate: The respiration rate of each fruit was determined at weekly interval in terms of CO_2 release using an Infra Red Gas Analyzer (IRGA) (Fig 3.3). The ambient CO_2 was measured before each fruit was placed in a separate enclosed jar of 7 L volume. The CO_2 release was monitored 10 min after placing the fruit in the jar by connecting the respiration jar to the IRGA. The actual CO_2 release was determined by taking the difference between ambient CO_2 concentrations and CO_2 release after 10 min. The CO_2 release by the fruit was calculated as

$mLkg^{-1}hr^{-1}$ adjusting for volume in the containers using the previously determined fruit volume. For details of calculations see Appendix 2.

5.3.5 Total Soluble Solids (TSS): The TSS was determined from a finely ground fresh (5 g) sample, homogenized with 5 mL distilled water using an ultra-turax® (IKA, Germany) according to Illeperuma and Jayasuriya (2002). The homogenate was then centrifuged using (Sorvall RC 5C Plus, CA, USA) at 6000**g** for 10 min. The TSS of the supernatant was measured using a hand held digital refractometer (PR-101, 0-45%, ATAGO, CO, Ltd Japan).

5.3.6 pH: The remaining supernatant was diluted to 80 mL with distilled water according to Illeperuma and Jayasuriya (2002) and pH measured with a (Corning 430, NY, USA) pH meter.

5.3.7 Titratable Acid (TA): The diluted supernatant was titrated against 0.1N NaOH until pH of 8.1-8.3 (Illeperuma and Jayasuriya, 2002). The amount of 0.1N NaOH used for titration then recorded to calculate titratable acids of the fruit. For acid calculation see Appendix 5. TA was expressed as percentage malic acids, which is the predominant acid in papaya (Sankat and Maharaj, 1997).

5.3.8 Sugar:Acid Ratio: This ratio was calculated as TSS/TA.

5.4 Results and Discussions

5.4.1 Percentage Weight Loss (PWL)

Water loss, apart from its physiological consequences, is a loss of saleable weight and thus is a direct loss to the producer (Wills *et al.*, 1989). Increased weekly PWL of fruits was evident for all fruits as the storage period and temperature increased (Fig 5.2). Most of the water loss (as measured by weight loss) was probably due to fruit transpiration. Highest and most rapid weight loss was noted from fruits stored at 22°C, which may be associated with the effect of temperature on transpiration rate. Although this result agrees with the previous result by Aung *et al.* (1996), the current experiment reveals that the effect of the treatments overcome this detrimental effect of temperature with regard to weight loss. MPB showed significantly ($P<0.001$) reduced PWL as compared to the other treatments, especially compared to control fruits. This was evident at all storage temperatures, including higher temperature (22°C). Several workers reported similar results for papaya (Chen and Paull, 1986; Lazan *et al.*, 1990a), for tomato and mushroom

(Hobson and Burton, 1989), for mango (Miller *et al.*, 1983; Singh and Janes, 2001) and for many other fruits (Wang, 1999). The authors reported that polyethylene packaging markedly reduced fruit weight loss. This reduced weight loss due to sealed MPB can be ascribed to the effect of modified atmosphere (MA) around the fruit on the transpiration or evaporation from the fruit surface. Maintaining the micro- atmosphere of the produce brings about saturation thus transpiration loss is minimized and there is no shrinkage or shrivelling to the produce (Salunkhe *et al.*, 1991; Wills *et al.*, 1998). Although all the above workers reported reduced PWL of produce packed in polyethylene bags, high build- up of water vapour was evident inside the bags leading to mould development and fruit decay, especially at higher storage temperatures. To avoid this problem in this experiment micro-perforated bags coated with anti-mist were used. Thus, there was no build-up of water vapour inside the bag as it was designed with small perforations (9 μm) to allow some gas exchange (Yantarasri, *et al.*, 1995). MPB were also effective in reducing water loss at lower storage temperatures of 5.5°C, however this did not protect the occurrence of chilling injury. It has been reported by Wills *et al.* (1998) that chilling injury might cause high water loss due to surface pitting. Although all fruits at 5.5°C suffered chilling injury especially when transferred to room temperature for ripening, MPB exhibited the lowest ($P<0.001$) weight loss. As a result, fruits could be stored for 50, 45, 38, 20 days at 7, 5.5, 10 and 22°C, respectively. Waxing also reduced PWL at all storage temperatures (Fig 5.2) especially at higher storage temperature (Fig 5.3). This is in general agreement with previous results by Chen and Paull (1986) and Sankat and Maharaj (1997) for papaya and Khader (1992) for mango. This can be related to the effect of waxing on fruit permeability to retard rapid fruit transpiration rate and water loss. Sankat and Maharaj (1997) and Meheriuk and Porrit (1972) added in that waxing not only reduced water loss (and, hence, shrivelling), but also served a twofold purpose by improving the fruit appearance. Waxing reduced weight loss, both at lower and higher storage temperatures, which reveals its applicability in various environmental conditions including the tropics. This treatment, therefore, is a possible means to reduce fruit deterioration and hence increase shelf-life. Many workers (Paull and Chen, 1989; Thompson and Lee, 1971) reported that untreated fruits lose water rapidly during storage, hence deteriorate rapidly. This report held true in this experiment as well. Therefore, the maximal storage period of control fruit was 43, 41, 29 and 15 days as compared to waxing with a storage period of 48, 55, 42 and 21 days at 5.5, 7, 10 and 22°C, respectively (Tables 5.1-5.4). This increased weight loss of control fruit was pronounced as the fruit transferred to room temperatures for ripening.

Similarly, GA_3 application resulted in most cases in retarded PWL compared to control fruits. It significantly ($P<0.001$) reduced weight loss at 7, 10 and 22°C as compared to control, but at 5.5°C an effect was only seen when the fruits were transferred to room temperature after seven weeks storage in the cold room. This is in agreement with previous results of Khader (1992) in mango fruits. The author reported that GA_3 exhibited a significant delay in ripening as evidenced by colour, aroma and other biochemical changes in constituents measured during storage. Although the role of GA_3 on PWL has not been investigated its effect of reducing PWL at higher storage temperatures implies the potential feasibility of this treatment as an alternative method in tropical environments.

Although improper storage or environmental temperatures can have detrimental effects on both, fruit quality and fruit shelf-life, appropriate handling and treatments pre or post storage will minimise these negative effects. It was reported by Arjona *et al.* (1992), Aung *et al.* (1996) and Pekmezci *et al.* (1997) that higher storage temperatures increase weight loss and thus decrease shelf-life. However, from the results presented in Fig 5.2 and Fig 5.3 it can be inferred that MPB, waxing and/or GA_3 application have the potential to prevent fruit weight loss and hence increase shelf-life both, at high and low storage temperatures.

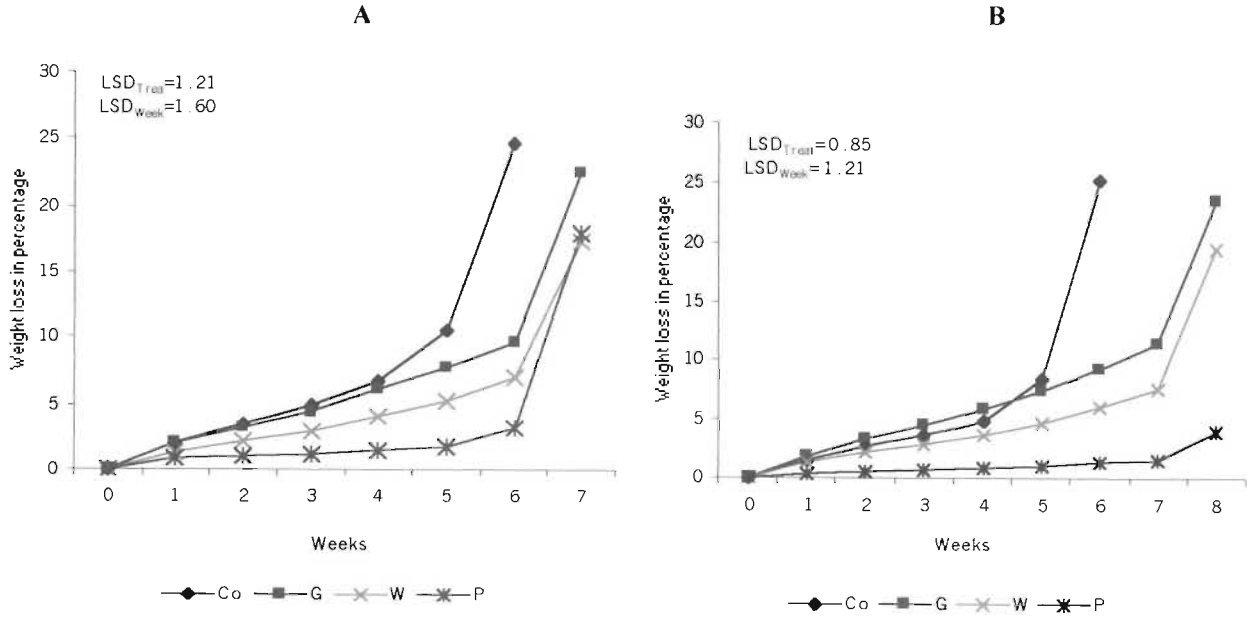


Fig 5.2 Percentage weight loss of papaya fruits during storage as affected by pre-storage treatments and storage temperatures; at 5.5°C (A) and at 7°C (B). For all figures, data points are means of five replicates. Least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and time (LSD_{Week}) are shown. Weight losses for each fruit were monitored to the table ripe stage (scale 6). Note data collection for control fruits terminated shortly. Co=control, G= GA_3, W=waxing and P= micro perforate polypropylene bag.

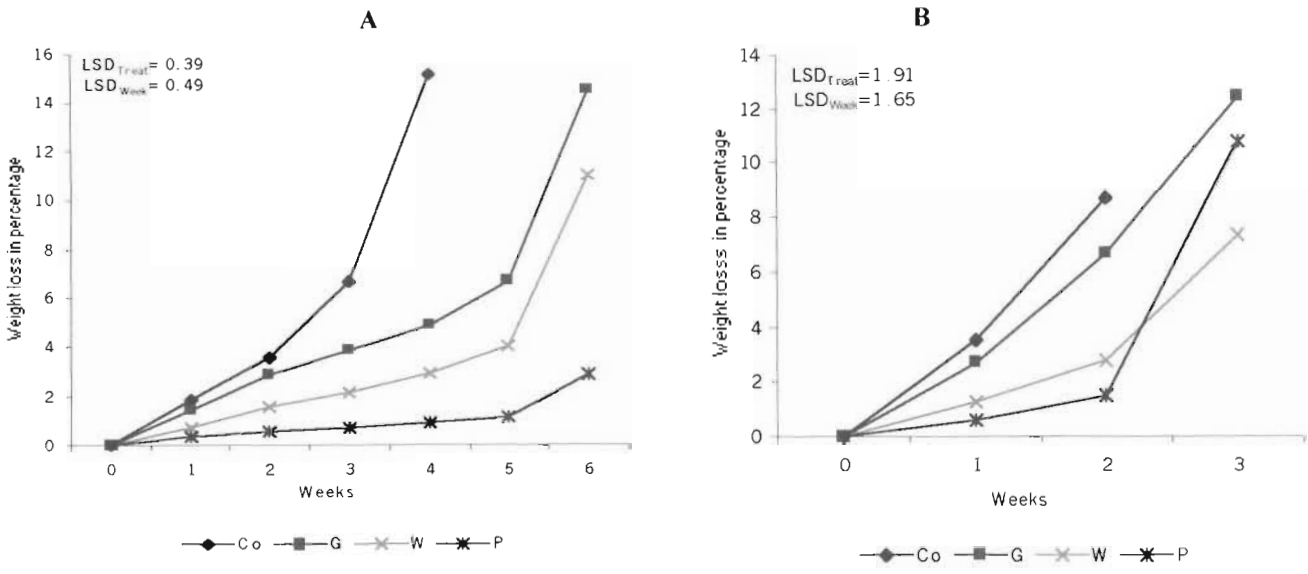


Fig 5.3 Percentage weight loss of papaya fruits during storage as affected by pre-storage treatments and storage temperatures; at 10°C (A) and at 22°C (B). Data points are means of five replicates. Least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and time (LSD_{Week}) are shown. Weight losses for each fruit were monitored to the table ripe stage (scale 6). Note data collection for control fruits terminated shortly. Co=control, G= GA_3, W=waxing and P= micro perforate polypropylene bag.

5.2 Firmness Change

Softening is one of the most significant quality alterations consistently associated with ripening (Aung *et al.*, 1996; Kays, 1991). Fruit quality of papaya is often assessed for the fresh and processing market by the change of softness of the fruit (Chan *et al.*, 1981). Hence, control of fruit softening is important to reduce mechanical damage and to maintain fruit quality (Smith *et al.*, 1990). All fruits from this experiment exhibited reduced firmness irrespective of treatments as the storage period was extended. This is supported by findings of Aung *et al.* (1996), Harker *et al.* (1997) and Lazan *et al.* (1993). The authors reported that papaya fruit, regardless of storage treatment experience not only extensive softening but also extensive depolymerization of wall pectins when reaching full ripe or full yellow stage. The implication of a close link between firmness and PG as well as firmness and PME activity have been previously reported by Castaldo *et al.* (1989), Kays (1991) and Lazan *et al.* (1993). Although for all fruits, softening was evident concomitant to ripening there was, however, some variability between the treatments. Waxing showed good results maintaining fruit firmness, particularly at higher temperatures. Compared to control fruits it significantly ($P<0.004$) reduced fruit softening at 7, 10, and 22°C (Fig 5.4) and thus fruit shelf-life was extended (Tables 5.1-5.4). Similar results were reported for papaya by Baldwin *et al.* (1992) and Chen and Paull (1986); for avocado by

Durand *et al.* (1984) and for apples by Meheriuk and Porrit (1972) where the authors found reduced fruit softening after waxing. Fruit softening was most rapid at the higher storage temperature of 22°C. This is in general agreement with the report of An and Paull (1990) in papaya and Agar *et al.* (1999) in kiwi fruits. Hobson, (1987) also found that storage temperatures had a significant effect on both, number of days to ripen and the firmness of tomato when ripe. Nevertheless waxing consistently reduced the rate of softening (at all storage temperatures), such that it could effectively be used in tropical environments with poor storage facilities. Fruit firmness showed no significant difference ($P<0.05$) at 5.5°C. However, waxing still showed a trend toward reduced fruit softening (Fig 5.4a). The increased firmness of other treatments, including control fruit at 5.5°C may be related to high water loss at 5.5°C which caused fruit wilting and hence, tough skin texture. This can be supported by the reports of Amarante and Banks (2001) who found that wilting toughens the flesh and causes a higher firmness reading, not truly reflecting the stage of ripening. On the other hand, according to Harker *et al.* (1997) this reduced softening at lower temperature could be due to retarded metabolic activity of the fruit as lower temperature inhibits a wide range of metabolic process including those associated with fruit softening and the deterioration of various textural attributes. MPB was the second most effective treatment limiting fruit softening during ripening. It significantly ($P<0.001$) reduced fruit softening at 7 and 10°C and showed a trend toward reduced fruit softening at 22°C, especially as compared to control fruits (Fig 5.4). A similar result was reported by Chen and Paull (1986) that papaya fruits stored in MAP were less shriveled compared to control fruits. This may be related to the MA created which retard softening. This is further ascribed to the accumulation of CO_2 concentration inside the bag, which retard metabolic activity associated with fruit softening (García *et al.*, 1998; Rosen and Kader, 1989). Previous results by Lazan *et al.* (1993) in papayas, confirm that sealed packaging reduces fruit colour development and softening. Ben-Yehoshua (1985) ascribed the delay in softening of waxed and wrapped fruits to the effect of the treatments to prevent water loss. On the other hand, GA_3 showed inconsistent results among the different storage temperatures. It showed of retarded softening at 5.5 and 7°C as compared to the control fruits. These results were in agreement with Desai and Deshpande (1978) and Khader (1992) who reported retarded fruit softening after GA_3 application.

Proper application of waxing (limited thickness to allow some gas exchange and to avoid off-flavour development), GA_3 and/or MPB might, therefore, be alternative postharvest applications to retard fruit softening.

Table 5.1 Average Physico-Chemical Change of Papaya Fruit after Storage at 5.5°C as Affected by Different Postharvest Treatments

Treatments	Shelf-life (Days)*	TSS (°Brix)	pH	TA (% malic)	Sugar:acid ratio
Control	43	7.60^a	5.39^a	1.32^a	6.15^a
GA_3	48	7.50^a	5.31^a	1.35^a	7.18^a
MPB	45	7.79^a	5.06^a	1.73^a	4.82^a
Wax	48	6.49^a	5.16^a	1.80^a	3.85^a
LSD	-	1.68	0.39	0.72	3.44
P value	-	NS	NS	0.035	NS

Table 5.2 Average Physico-Chemical Change of Papaya Fruit after Storage at 7°C as Affected by Different Postharvest Treatments

Treatments	Shelf-life (Days)*	TSS (°Brix)	pH	TA (% malic)	Sugar:acid ratio
Control	41	9.17^{ab}	4.56^a	2.58^b	3.85^a
GA_3	53	7.70^a	4.86^a	2.42^b	3.21^a
MPB	50	9.40^b	4.36^a	2.48^b	3.81^a
Wax	55	7.57^a	4.95^a	1.69^a	4.48^a
LSD	-	1.68	0.39	0.72	3.44
P value	-	NS	NS	0.035	NS

Table 5.3 Average Physico-Chemical Change of Papaya Fruit after Storage at 10°C as Affected by Different Postharvest Treatments

Treatments	Shelf-life (Days)*	TSS (°Brix)	pH	TA (% malic)	Sugar:acid ratio
Control	29	8.23^a	4.36^a	1.43^a	5.61^a
GA_3	42	7.87^a	4.48^a	2.12^{ab}	4.39^a
MPB	38	7.60^a	4.62^a	2.57^b	4.17^a
Wax	42	9.20^a	4.74^a	2.51^b	4.53^a
LSD	-	1.68	0.39	0.72	3.44
P value	-	NS	NS	0.035	NS

Table 5.4 Average Physico-Chemical Change of Papaya Fruit after Storage at 22°C as Affected by Different Postharvest Treatments

Treatments	Shelf-life (Days)*	TSS (°Brix)	pH	TA (% malic)	Sugar:acid ratio
Control	15	8.20^{ab}	4.70^a	2.13^a	4.06^a
GA_3	21	7.13^a	4.85^a	1.90^a	3.77^a
MPB	20	9.70^b	4.48^a	2.16^a	4.84^a
Wax	21	6.93^a	4.93^a	1.81^a	3.97^a
LSD	-	1.68	0.39	0.72	3.44
P value	-	NS	NS	0.035	NS

Means in each columns of all tables followed by the same letter are not significantly different at 5% level. NS= Not Significant.

*** Days were determined when the fruit reaches table ripe stage (Scale -6)**

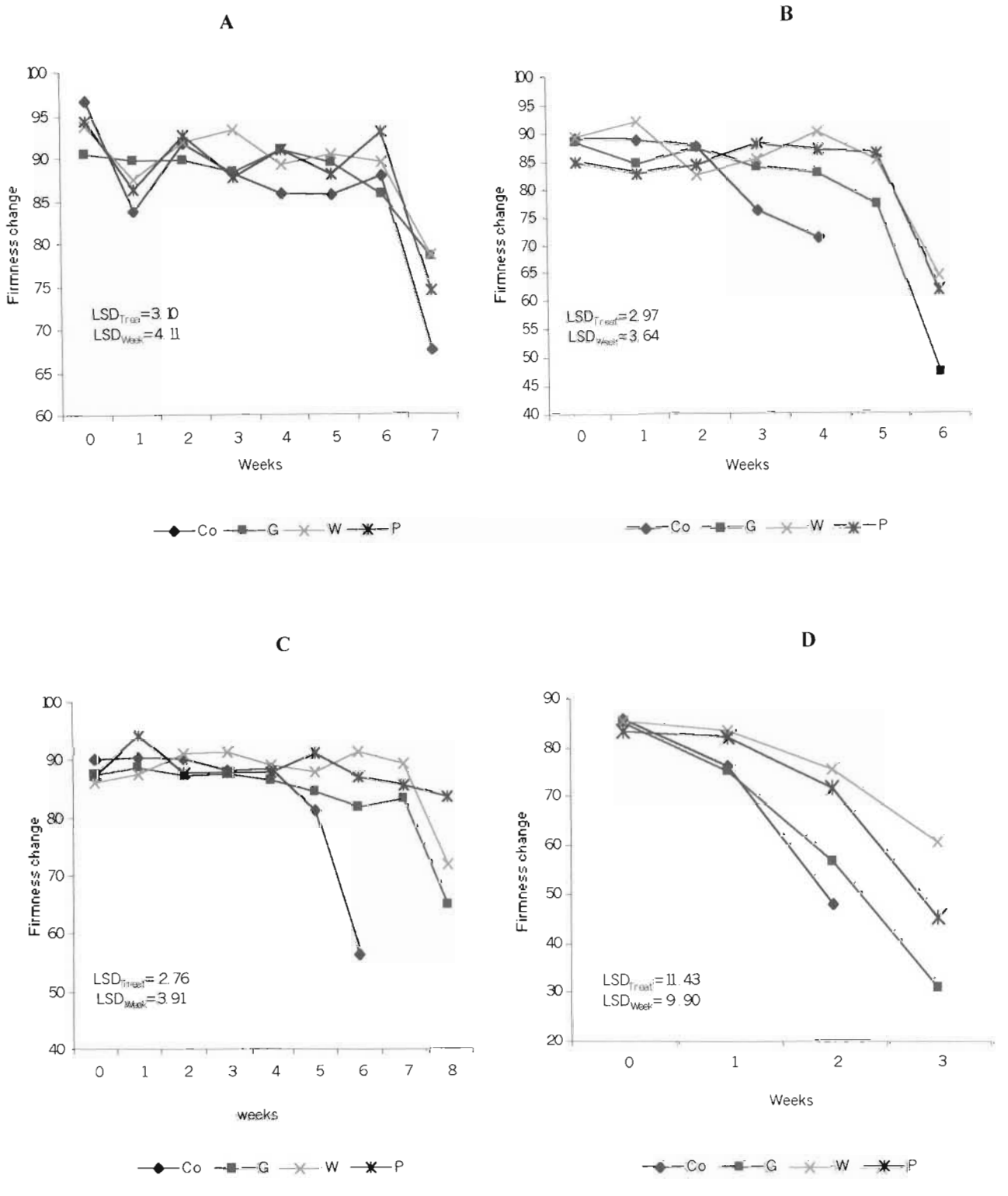


Fig 5.4 Firmness changes of papaya fruits during storage as affected by pre-storage treatments and storage temperatures; at 5.5°C (A), at 7°C (B), at 10°C (C) and at 22°C (D) For all figures, data points are means of five replicates. Least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and time (LSD_{Week}) are shown. Firmness changes for each fruit were monitored to the table ripe stage (scale 6). Note data collection for control fruits terminated shortly. Co=control, G= GA_3, W=waxing and P= micro-perforated bag.

5.4.3 Skin Colour Change

The external appearance of fruits, particularly their colour, is of prime importance when considering product quality (de Guevara *et al.*, 1996). Colour development intensified as storage time was extended in all fruits, irrespective of treatment. Colour development was pronounced as storage temperature was increased. These observations are in general agreement with An and Paull (1990) who reported the rate of skin yellowing of papaya showed a linear relationship to the increased ripening temperature. Control fruits exhibited significantly ($P<0.001$) more rapid colour development at all storage temperatures and, as a result, fruit could only be stored for a shorter period (Tables 5.1-5.4). Waxing and GA_3 as well as MPB, however, retarded colour change significantly as compared to control fruits. Waxing retarded colour change better than other treatments at all storage temperatures (Fig 5.5 and 5.7). This agrees with a previous report by Olorando (2000) that fruit surface coating delayed colour development by up to eight days. Paull and Chen (1989) and Sankat and Maharaj (1997) also reported that papaya fruits coated with polyethylene paraffin wax exhibited a slow colour development than control fruits. Retardation of colour development by waxing can be ascribed to its effect on limiting fruit skin permeability to CO_2 and O_2. Fuchs and Temkin-Gorodeiski (1971) have demonstrated that a high concentration of CO_2 around the fruit interferes with the colour development of banana fruit. In our experiment fruits held at higher storage temperature exhibited more rapid colour change on a short period of time. Hence, fruits were stored for a maximum (waxing and GA_3) of three weeks. Wills and Widjanarko (1995) reported that ripening time as expressed by colour change, was reduced at 25 and 30 as compared to 20°C. In this experiment waxing still retarded colour development even at higher storage temperature. Therefore, waxing has a positive effect on retardation colour change at higher storage temperature, which indicates the application potential of the treatment for poorly resourced tropical environments. Similarly, the positive effect of GA_3 (Fig 5.6 and Fig 5.7) is supported by previous results for banana by Desai and Deshpande (1978) and Salunkhe *et al.* (1991), for mango by Khader (1992) and for other climacteric fruits by Vendrell and Palomer (1997). These authors reported that GA_3 significantly retards fruit skin colour development as compared to control fruits. This agrees with an earlier report that a GA_3 dip at 200 ppm extended the shelf-life of mango fruit (Khader *et al.*, 1988) by retarding ascorbic acid decrease and chlorophyll degradation and decreasing enzyme activity. Moreover, this can be ascribed to the effect of GA_3 to act against fruit senescence, which is characterized by colour development. Similarly, Lazan *et al.* (1990a), Paull and Chen (1989) and Sankat and Maharaj (1997) reported that MA packaging retards papaya fruit colour development.

Macnish *et al.* (1997) and Yantarasir *et al.* (1995) also reported that mango fruits in MA packaging with small perforations show inhibited carotenoid synthesis. This report agrees with the current result (Fig 5.7). Hence, modification of O_2 and CO_2 concentration in the atmosphere within and around the fruit by either waxing or MPB and treatment of fruits with GA_3 may be possible techniques to extend shelf-life by retarding colour development.

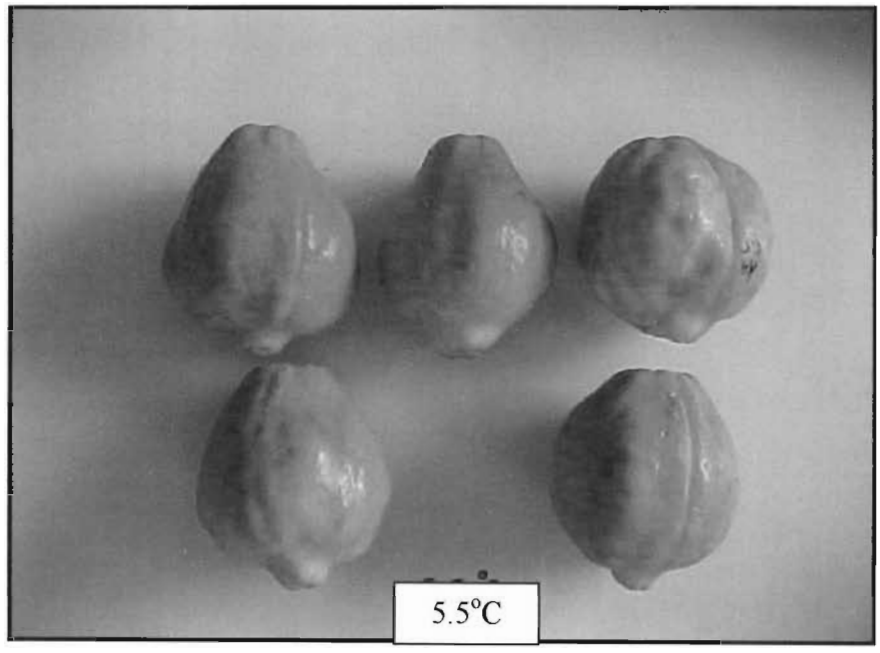


Fig 5.5 Effect of waxing on fruit colour development at 5.5°C after three weeks.

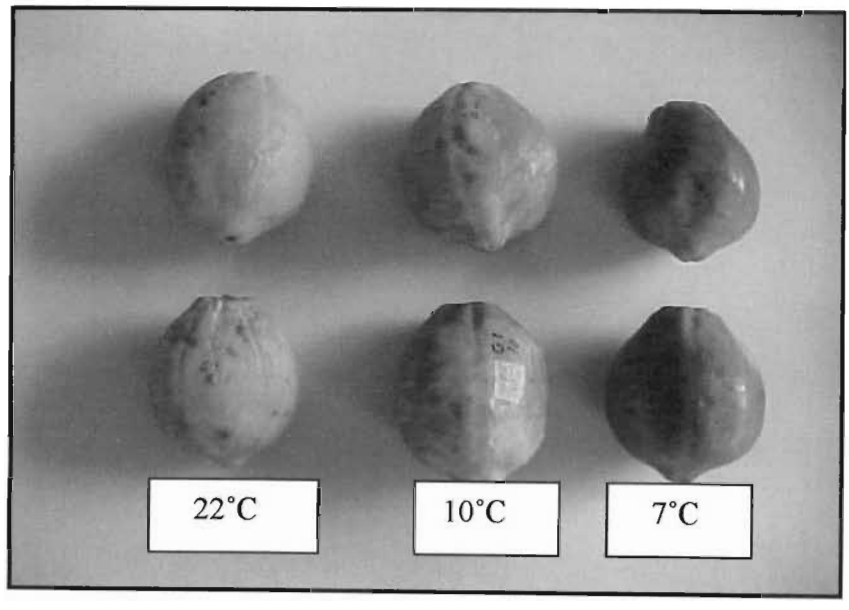


Fig 5.6 Effect of GA_3 treatment on fruit colour development at different storage temperatures after three weeks storage.

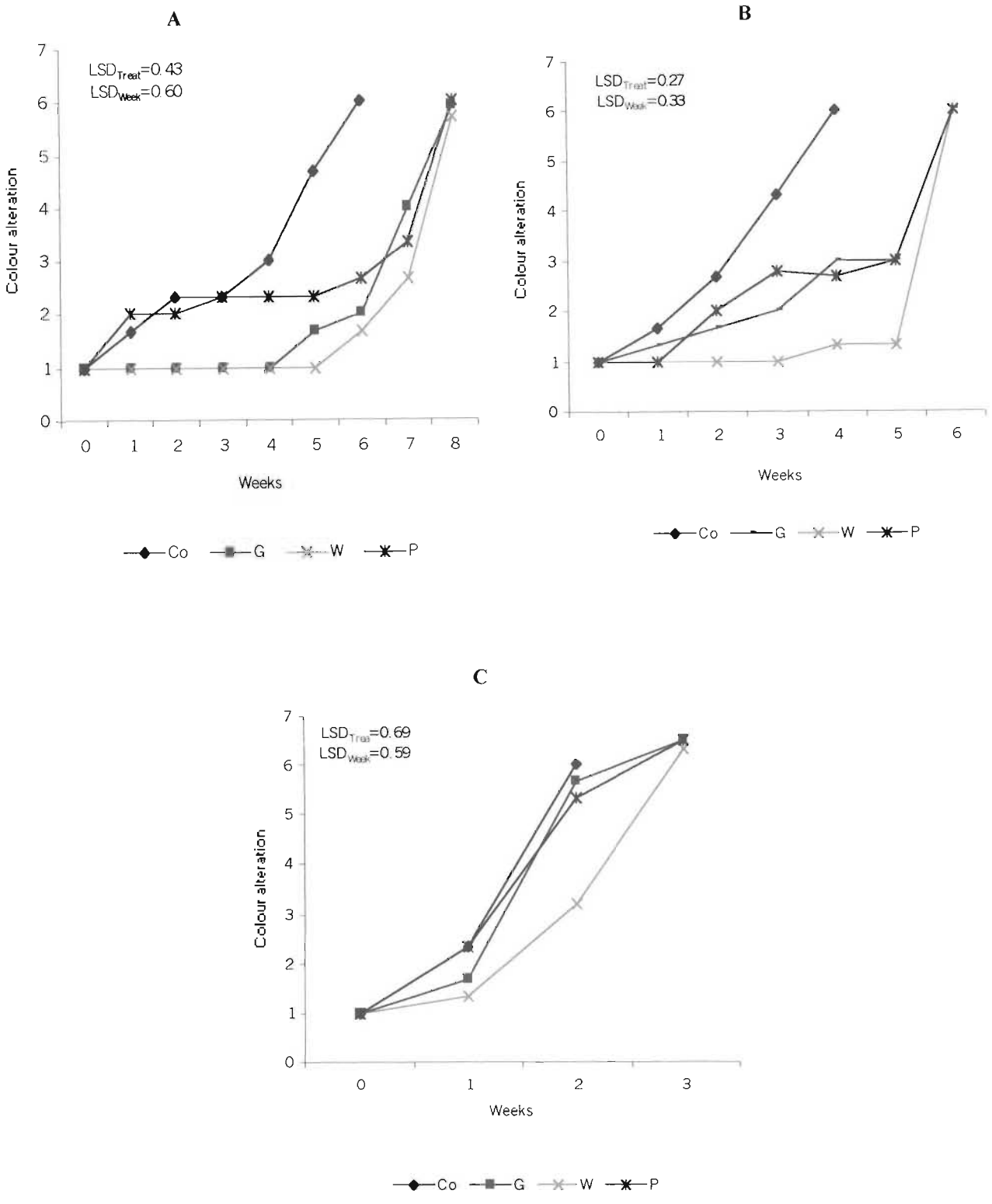


Fig 5.7 Colour changes of papaya fruits during storage as affected by pre-storage treatments and storage temperatures; at 7°C (A) at 10°C (B) and at 22°C (C). For all figures, data points are means of five replicates. Least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and time (LSD_{Week}) are shown. Colour alteration for each fruit was monitored to the table ripe stage (scale 6). Note data collection for control fruits terminated shortly Co=control, G= GA_3, W=waxing and P= micro perforate bags.

5.4.4 Respiration Rate

The climacteric pattern of ethylene production and respiration can be used as a means to measure the physiological status or degree of fruit ripening (Strydom, 1991) and fruit deterioration (Wilson *et al.*, 1999). The rate of respiration for climacteric fruits affected to a various degree by different storage temperatures. Respiration rate increases as the storage temperature and period increases. This was also true in this experiment. Rapid respiration rate was noted at higher storage temperature (Fig 5.7). Subsequently, fruits deteriorated rapidly and, as a result, only stored for a maximum of three weeks (Tables 5.1-5.4). Similar results were reported for plantains by George and Marriott (1985) and Salunkhe *et al.* (1991) and for papaya by Yahia *et al.* (1992). Waxing and GA_3 significantly reduced ($P<0.001$) respiration rate at all storage temperatures. It was noted that all the waxed fruits exhibited a lower respiration rate and the least colour and firmness change. Waxing was effective at extending the shelf-life even at higher temperature (22°C). Waxed fruits exhibited a respiratory climacteric after 15 days at 22°C, 35 days at 10°C, 42 days at 5.5°C and 50 days at 7°C only when fruits were transferred to room temperature for final ripening, whereas control fruits showed respiratory climacteric after 7, 21 and 35 days at 22, 10, 7 and 5.5°C, respectively (Fig 5.8). A similar result was reported recently by Amarante and Banks (2001) noting that coating delayed ripening of fruits at higher temperature as compared to lower temperature storage. Waxing restricts O_2 diffusion into the fruit (Durand *et al.*, 1984) and exerts its effect on skin permeance by blocking a greater or lesser proportion of the pores on the fruit surface (Amarante and Banks, 2001). This in turn limits fruit respiration rate. Waxing thus has the potential to extend the storage life of fruits at higher temperatures in a tropical environment. The positive effect of GA_3 also agrees with previous results for papaya (Mehta *et al.*, 1986) and for tomato (Babbitt *et al.*, 1973). The shelf-life extension by GA_3 can be attributed to a decrease in respiration rate as a result of less succinate and malate dehydrogenase activity associated with the TCA cycle (Mehta *et al.*, 1986). Similarly, Salunkhe *et al.* (1991) reported that GA_3 markedly retards colour change, the climacteric peak and the respiration rate of both, tomato and banana. This reduction of ripening and the increase in shelf-life is related to the inverse relationship of respiration rate and shelf-life (Paull and Chen, 1989). On the other hand, MPB exhibited significantly higher ($P<0.001$) rate of respiration at 10 and 22°C storage temperatures as compared to the other treatments. This agrees with a previous report for papaya by Lazan *et al.* (1990a) and for strawberry by García *et al.* (1998) and may be attributed to the differential permeability of the bags. The film provided a barrier to normal atmospheric conditions and as a result fruits within the film presumably exposed to lower O_2 and higher CO_2

than the ambient atmosphere. It has been reported by Zagory (1998) that an elevated CO_2 suppresses plant tissue sensitivity to the effect of the ripening hormone ethylene, which results in an increased respiration rate. However, when the fruits were removed from the bag (for measurement of respiration), the fruits started to respire rapidly after they were exposed to ambient higher O_2 concentration. This might be the cause for the increased measurement of respiration rate of fruits in MPB. Although the increased CO_2 level inside the bag retards respiration rate, CO_2 concentration beyond the tolerable level, however, might cause undesirable damage to the fruit. Polyethylene bags apart from their ability to reduce water loss may cause high build-up of CO_2 inside the bag, which can result in off-flavour and decay. According to Tan and Ali (1989) high build-up of CO_2 inside the bag leads to cellular disorganization and subsequent tissue collapse manifesting itself as CO_2 injury. Interestingly, there was no any off-flavour development, fruit decay and/or CO_2 injury to the fruits in this experiment. This may be related to the kind of packaging used. The micro-porous and micro-perforated films allow much more rapid gas exchange than other plastic films (Zagory, 1998). Increasing or decreasing the number and size of holes can regulate the O_2/CO_2 ratio within the bags. It has been reported that relatively increased hole size and number provides better ventilation and gas exchange, however, this may be at the expense of excessive water loss. It was also reported (Illeperuma and Jayasuriya, 2002) that inclusion of granular charcoal could be effective to prevent the accumulation of CO_2 beyond the maximum tolerable level. If the high build-up of CO_2 inside the MPB can be reduced by any means, such packaging could become the most effective method to store fruits for an extended period.

From the above results it can be inferred that waxing and GA_3 might overcome the detrimental effect of a tropical environment on fruit respiration rate in particular and to shelf-life in general. Therefore, they possess the potential as alternative methods of fruit treatment in a tropical environment with low or poor storage facilities.

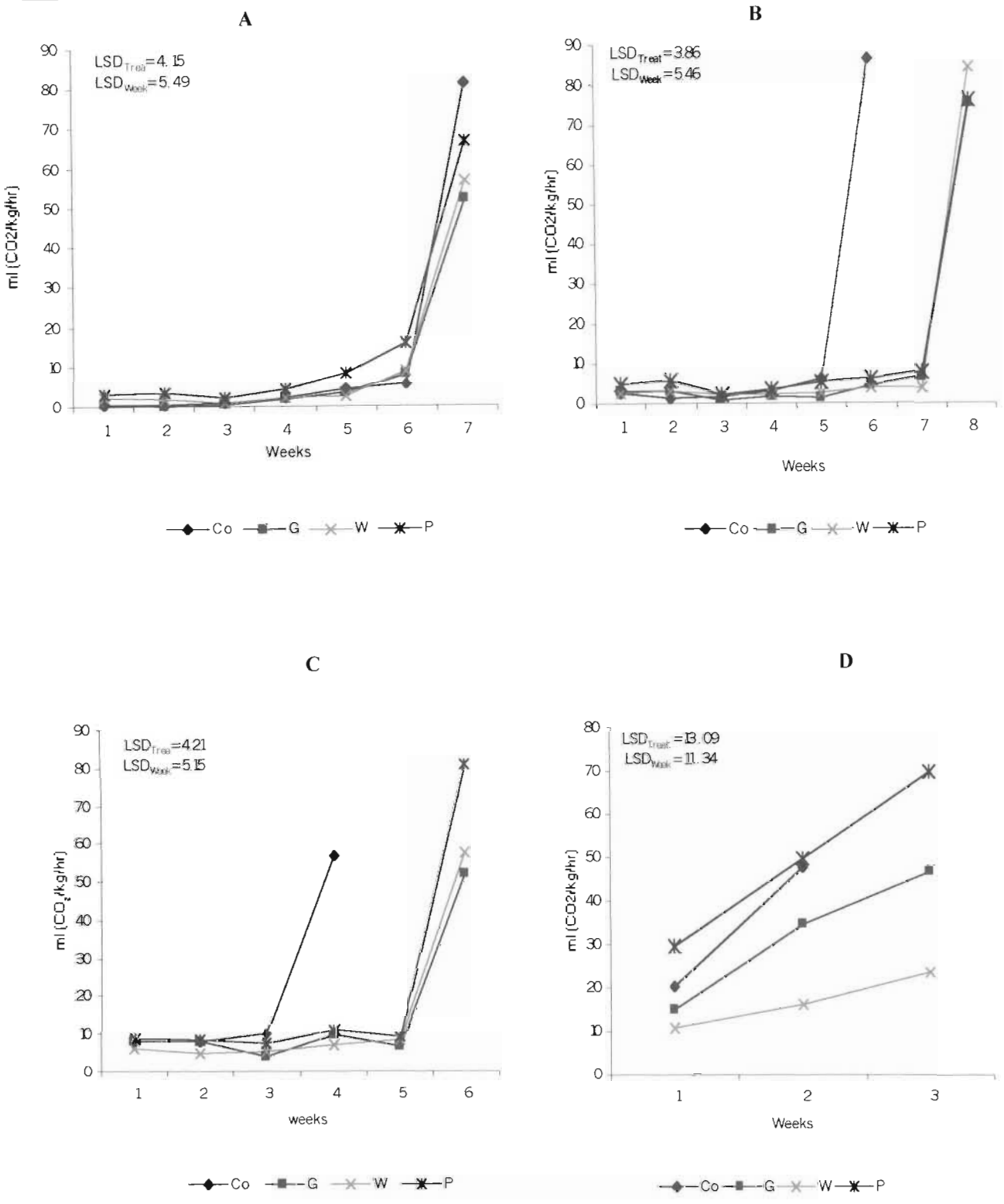


Fig 5.8 Respiration rates of papaya fruits during storage as affected by pre- storage treatments and storage temperatures; at 5.5°C (A), at 7°C (B), at 10°C (C) and at 22°C (D). For all figures, data points are means of five replicates. Least significant difference (LSD) at 5% level for treatments (LSD_{Treat}) and time (LSD_{Week}) are shown. Respiration rate for each fruit was monitored to the table ripe stage (scale 6). Note data collection for control fruits terminated shortly. Co=control, G=GA_3, W=waxing and P= micro-perforated bag

5.4.5 Total Soluble Solid (TSS): There was no significant difference ($P<0.05$) among treatments and storage temperatures for fruit TSS, although waxing and GA_3, in most cases, showed a trend to a lower value (Tables 5.1-5.4). As the ripening process is retarded by GA_3 (Desai and Deshpande, 1978; Khader, 1992) and waxing (Paull and Chen, 1989) so is the TSS level of the fruits. On the other hand, control fruits and MPB exhibited a trend toward an increased TSS value. This was evident at all storage temperatures. This increased level may be related to increased water loss and respiration rate, which may lead to accelerated ripening and deterioration (Ben-Yehoshua, 1985). Similar to these results, no significant difference in TSS was reported for mango for various storage treatments by Yantarasri *et al.* (1995) and for banana by Chamara *et al.* (2000). Although, storage temperature affected fruit TSS content, no trend was visible. The lowest TSS value was determined at 5.5°C, followed by 22°C. On the other hand, fruits at 7°C followed by 10°C exhibited relatively higher TSS values (Tables 5.1-5.4). From this result it can be inferred that there was no direct or inverse relationship between TSS and storage temperatures as whole. Therefore, the shelf-life increase either by waxing, GA_3, and/or MPB did not have any significant effect on the fruit TSS quality.

5.4.6 pH: The pH value of all fruits displayed an opposite pattern to the TSS value, but there was no significant difference ($P<0.05$) among treatments. This was evident at all storage temperatures. Lazan *et al.* (1990a) reported for papaya and Chamara *et al.* (2000) for banana that there were no measurable difference among treatments after fruits were stored for an extended period using various seal-packaging. The change of tissue pH in this experiment appeared to be affected by the storage temperature rather than treatments. Fruits at 5.5 and 22°C exhibited a trend of increased pH content as compared to the 7 and 10°C storage temperatures (Tables 5.1-5.4). Waxing, GA_3 and MPB might thus be used to extend the shelf-life without any quality defect to the fruit.

5.4.7 Titratable Acids (TA): Similar to the other quality parameters the TA value of fruits showed no significant difference ($P<0.05$) among treatments. This was evident at all storage temperatures (Tables 5.1-5.4). Similarly, Chamara *et al.* (2000) reported no significant difference between MA treated and control banana fruits after ripening. The TA value was more affected by temperature than by treatments. Fruits at 5.5°C exhibited relatively low TA value as compared to the other temperatures. This may be attributed to the effect of temperature on the general physico-chemical activities of ripening fruit (Wang, 1999). An increased TA content during

ripening of papaya fruit was reported by Paull (1993) as well as Wills and Widjanarko (1995). This may be associated, partly to the increase in free galacturonic acid during ripening. On the other hand, Kays (1991) and Wills *et al.* (1989) reported that during ripening the TA of most fruits decrease as the acids are used in respiration or converted to sugars. Similarly, Sankat and Maharaj (1997) reported that non-volatile organic acids formed the major portion (about 80-90%) of total acid in fruits; and the total acid concentration decreases as the fruit ripens. This makes it difficult to use TA as a sole parameter to evaluate fruit ripening. However, as there was no significant difference between treatments there is still a potential of using waxing, GA_3 and MPB as storage techniques to extend shelf-life with limited effect to the quality.

5.4.8 Sugar:Acid Ratio: There was no significant difference ($P<0.05$) between treatments at all storage temperatures for the sugar:acid ratio. Changes in sugar:acid ratio of all sample fruits showed almost a similar pattern to TSS with the same profile corresponding to treatments. Fruits at 5.5°C exhibited a trend towards higher sugar:acid ratio as compared to the other storage temperatures. This was related to the lower TA value of the fruits, as TA is inversely related to the sugar:acid ratio.

5.5 Conclusion

There is a need for technological advances in methods for extending the shelf-life of perishable fruits (Hintlian and Hotchikiss, 1986). Papaya, like most tropical fruits, suffers from the detrimental effect of surrounding temperature affecting the shelf-life considerably. This becomes more pronounced in areas with poor storage facilities, which implies the need for an intervention after harvest to overcome the problem. Although postharvest techniques cannot improve the quality of the fruit, good handling helps to manipulate and maintain initial quality in the best possible manner until consumption (Wang, 1999).

This experiment helped to investigate and identify possible techniques and methods for postharvest maintenance of fruit metabolic activity and thus shelf-life, particularly techniques suited to poor resourced regions. Waxing, GA_3 and MPB reduced postharvest deterioration at all storage conditions. Therefore, these treatments can provide valuable means to retard PWL, softening, colour development and respiration rate (except MPB). Thus all these treatments are recommended as alternative methods to reduce postharvest deterioration of papaya fruit, particularly at higher ambient temperatures. Application of different packaging material, notably

micro-perforated bags coated with anti-mist, reduced the high build-up of CO_2 inside the bags. This type of packaging material overcomes the common drawback of polyethylene bags to cause high build-up of CO_2. No pathological decay development and/or off-flavour symptoms commonly known as CO_2 injury were observed. It is, therefore, advisable that MPB to be used as a postharvest treatment to extend fruit shelf-life. Determination of certain quality parameters (TSS, TA, pH and sugar:acid ratio) after storage showed no significant difference among treatments at any of the storage temperatures. This indicates the feasibility of using waxing, GA_3 and MPB to extend fruit shelf-life with no significant effect to fruit quality.

Chapter 6: General Conclusions and Recommendations

6.1 Conclusions

Banana and papaya are popular and commonly grown fruits of the tropics. In spite of their importance, higher ambient temperature, under which the fruits are grown, makes their shelf life limited. In areas where there is lack of storage facilities and poor cold chain maintenance a considerable amount of the produce is lost through deterioration. Postharvest loss of banana and papaya in developing countries varies from 20-80% and 40-100%, respectively, depending on the cultivar and environmental conditions. This postharvest loss becomes more pronounced by the poor distribution and marketing system in tropical countries. In this study different methods were used to extend the shelf life and reduce postharvest loss of the fruits in a manner suitable for use in tropical environmental conditions where poor storage and distribution facilities exist.

High storage temperature in all the experiments resulted in a higher fruit deterioration rate and caused reduced storage life. This was evident in both banana and papaya, which confirmed the detrimental effect of high ambient temperature to the shelf life of tropical fruits. Waxing and GA_3 were among the best treatments. The increased percentage weight loss, softening, colour development and respiration rate, which are the main causes of fruit deterioration, can be minimised by postharvest treatment of waxing and GA_3. These treatments appear to offer some prospects for reducing storage and distribution losses of banana and papaya in developing tropical countries where more complex postharvest storage is impractical. Postharvest treatment of banana fruits with IBA is also a promising alternative as a means to retard excessive loss of the fruit after harvest, mainly in areas where there are poor infrastructures and limited environmental controls.

Modified atmosphere packaging of both banana and papaya in micro perforated polypropylene bags extended the shelf life for both fruits. Inclusion of an ethylene absorbent in the bag would further increase fruit shelf life. Interestingly these packaging materials can extend fruit shelf life without fungal or off flavour development. Micro perforated polypropylene bags are considered to be a low-cost storage technique to help subsistent farmers in developing countries where they can be used as an alternative to refrigeration.

Evaluation of quality parameters after storage indicated waxing, GA_3, IBA and MAP might be used to preserve fruit quality. The results suggested that these treatments exerted no significant effect on the quality of the fruit, which further indicated that these treatments not only prolong the shelf life of the fruit, but also preserve fruit quality for an extended storage period, which brings additional advantage to their application.

In general, waxing, GA_3, and all the packaging materials used in this study can be used effectively as postharvest treatments in poorly resourced regions of the tropics where there is lack of storage facilities such as cold rooms or as an alternative when the existing cold room facilities become out of use for some reasons.

6.2 Recommendations

Based on the study conducted the following recommendations can be outlined:

- Introducing the use of waxing, synthetic growth regulators GA_3 and IBA and micro perforated bag packaging to poorly resourced farmers of developing countries as postharvest treatments, is recommended.
- Most of the existing packaging materials are made for the general purpose of packaging freshly harvested fruits or vegetables. It is highly recommended to design and prepare fruit specific packaging materials depending on the respiration pattern of the fruit. This is mainly important for climacteric fruits to avoid high accumulation of CO_2 inside the bags, as airtight sealed fruits would ultimately suffer from lack of O_2 resulting in anaerobic respiration and fermentation.
- Further research on the effect of the treatments on the ethylene production, reduced sugars, ascorbic acids and pectin enzyme activities as related to fruit physiological changes are recommended. This will further provide more information to confidently recommend the effect of the treatments on fruit quality and ripening behaviour.
- Further studies on the applicability of these treatments to other tropical and highly perishable fruits should be conducted to investigate their overall functions as postharvest treatments.

- Although IBA showed promising results with the specific concentration used in this study, further investigation should be conducted with different application methods and concentrations. This will help to identify the best possible concentration and application method of the treatment to achieve better results.

- Universally applicable methods of reducing postharvest loss are not practical, mainly because postharvest loss involves more than just technical issues. Fruits grown under different conditions have different structural and physiological features, which enable them to react differently to postharvest handling. It is, therefore, recommended to consider the preharvest aspects under which the crops were grown as related to postharvest handling. Preharvest crop management has a direct effect on postharvest characteristics and quality. Only high quality fruit can result in best postharvest performance.

References

Ackermann, J., Fischer, M., and Amadò, R. (1992). Changes in Sugar, Acids, and Amino Acids during Ripening and Storage of Apples (Cv. Glockenapfel). Journal of Agricultural and Food Chemistry **40**:1131-1134

Agar, I.T., Massantini, R., Hess-Pierce, B., and Kader, A.A. (1999). Postharvest CO_2 and Ethylene Production and Quality Maintenance of Fresh-Cut Kiwifruit Slice. Journal of Food Science **64**(3):433-440

Ait-Oubahou, A. (1999). Modified Atmosphere Packaging of Tomato. In: Postharvest Losses of Perishable Horticultural Crops in the Mediterranean Region. Gerasopoulos, D. (ed) Vol.42. CIHEAM-Options Mediterraneennes, Chania, Greece. pp103-113

Akamine, E.K., and Goo, T. (1971). Relationship between Surface Colour Development and Total Soluble Solids in Papaya. HortScience **6**(6):567-568

Akamine, E.K., Kitagawa, H., Subramanyam, H., and Long, P.G. (1975). Packing House Operations. In: Postharvest Physiology, Handling and Utilization of Tropical and Subtropical Fruits and Vegetables. Pantastico, Er. B. (ed) AVI Publishing Co. Westport Connecticut. pp267-282

Alique, R., and Oliveira, G.S. (1994). Changes in Sugars and Organic Acids in Cherimoya (*Annona cherimola* Mill.) Fruit under Controlled-Atmosphere Storage. Journal of Agricultural and Food Chemistry **42**:799-803

Amarante, C., and Banks, N.H. (2001). Postharvest Physiology and Quality of Coated Fruits and Vegetables. Horticultural Review **26**:161-238

An Jo-Feng, and Paull, R. E. (1990). Storage Temperature and Ethylene Influence on Ripening of Papaya Fruit. Journal of the American Society for Horticultural Science **115**(6):949-953

Anon, (2001a). What Happens during the Ripening of Fruits. [Internet] Oregon State University, Corvallis, USA. Available from http://osu.orst.edu/food-resource/faq/ripening.html [accessed 12 November 2001]

Anon, (2001b). The Truth about Waxing. In: Facts [Internet] BC Tree Fruits Limited, USA. Available from http://www.bctree.com/faq. [Accessed 16 November, 2001]

Arjona, H.E., Matta, F.B., and Garner, J.O. (1992). Temperature and Storage Time Affect Quality of Yellow Passion Fruit. HortScience **27**(7):808-810

Aung, L.H., Harris, C.M., Rij, R.E., and Brown, J.W. (1996). Postharvest Storage Temperature and Film Wrap Effects on Quality of Chayote, *Sechium edule* Sw. Journal of Horticultural Science **71**(2):297-304

Babbitt, J.K., Powers, M. J., and Patterson, M.E. (1973). Effects of Growth Regulators on Cellulase, Polygalacturonase, Respiration, Colour, and Texture of Ripening Tomatoes. Journal of the American Society for Horticultural Science **98**(1):77-81

Balasubramaniam, R., and Agnew, R. (1990). The use of Gibberellic Acid to Improve Postharvest Handling and Storage Quality of Cherries. Horticultural Research publication. Marlborough Research Center, New Zealand

Baldwin, E., Nisperos-Carriedo, M., and Campbell, C. (1992). Extending Storage Life of Papaya with Edible Coating. HortScience **27**(6):679 (Abstract)

Bangerth, F. (1974). Second Discussion Meeting on Bitter Pit in Apples. Acta Horticulturae **45**: 43-52

Banks, N.H. (1984). Some Effect of TAL Pro-Long Coating on Ripening Banana. Journal of Experimental Botany **35**(150):127-137

Banks, N.H., Cutting, J.G.M., and Nicholson, S.E. (1997). Approaches to Optimizing Surface Coating for Fruits and Vegetables. New Zealand Journal of Crops and Horticultural Science **25**:261-272

Barkai-Golan, R. (2001). Postharvest Disease of Fruits and Vegetables: Development and Control. (1st ed.). Elsevier Science B.V., Amsterdam, The Netherlands.

Bender, R.J., Brecht, J.K., Sargent, S.A., Navarro, J.C., and Campbell, C.A. (1993). Ripening Initiation and Storage Performance of Avocados Treated with an Edible-Film Coating. Acta Horticulturae **343**:184186

Ben-Yehoshua, S. (1985). Individual Seal-Packaging of Fruits and Vegetables in Plastic Film – A New Postharvest Technique. HortScience **20** (1):32-37

Ben-Yehoshua, S., Rodovo, V., Fang, D.Q., and Kim, J.J. (1995). Performed Antifungal Compounds of Citrus Fruit: Effect of Postharvest Treatments with Heat and Growth Regulators. Journal of Agriculture and Food Chemistry **43**:1062-1066

Biale, J., and Yaung, R.E. (1981). Respiration and Ripening in Fruits Retrospect and Prospect. In: Recent Advances in the Biochemistry of Fruits and Vegetables. Friend, J., and Rhodes, M. J. C. (ed) Academic Press, London. pp1-39

Burdon, J.N. (1997). Postharvest Handling of Tropical and SubTropical Fruit for Export. In: Postharvest Physiology and Storage of Tropical and Subtropical Fruits. Mitra, S. (ed) CAB International, London. pp 1-18

Burdon, J.N., Moore, K.G., and Wainwright, H. (1993). Postharvest Water Loss of Plantain and Cooking Banana Fruits. Acta Horticulturae **343**:307-308

Burg, S. P., and Burg, E. A. (1962). Role of Ethylene in Fruit Ripening. Plant physiology **37**:179-189

Cameron, A., Boylan-Pett, W., and Lee, J.L. (1989). Modeling Oxygen Concentrations within Sealed Packages of Tomato Fruits. Journal Food Science **54**(6):1413-1416

Cantwell, M.I., and Reid, M.S. (1993). Postharvest Physiology and Handling of Fresh Culinary Herbs (Rev.). Journal of Herbs, Spices and Medical Plants **3**:96-127

Castaldo, D., Quagliuolo, L., Servillo, L., Balestrieri, C., and Giovane, A. (1989). Isolation and Characterization of Pectin Methylesterase from Apple Fruit. Journal of Food Science **54**(3):653-655, 673

Chamara, D., Illeperuma, K., Galappatty, P., Theja, and Sarananda, K.H. (2000). Modified Atmosphere Packaging of 'Kolikuttu' Banana at Low Temperature. Journal of Horticultural Science and Biotechnology **75**(1):92-96

Chan, H.T., Tam, Jr.S.Y.T., and Seo, S.T. (1981). Papaya Polygalacturonase and its Role in Thermally Injured Ripening Fruit. Journal of Food Science **46**:190-191, 197

Chang, W.H., and Hwang, Y.J. (1990). Effect of Some Inhibitors on Carbohydrate Content and Related Enzyme Activity during Ripening of Taiwan Northern Banana Fruit. Acta Horticulturae **275**:611-619

Chaplin, G.R., Scott, K.J., and Brown, B.I. (1982). Effect of Storing Mangoes in Polyethylene Bags at Ambient Temperatures. Singapore Journal of Primary Industries **10**:84-88

Charles, R.J., and Tung, M.A. (1973). Physical, Rheological and Chemical Properties of Bananas during Ripening. Journal Food Science **38**:456-459

Chen, C.R., and Ramaswamy, H.S. (2002). Colour and Texture Changes Kinetics in Ripening Bananas. Wiss.U.-Technol **35**:415-419

Chen, N.M., and Paull, R.E. (1986). Development and Prevention of Chilling in Papaya. Journal of the American Society for Horticultural Science **111**(4):639-643

Coates, L.M., Cooke, A.W., and Persely, D.M. (1995). Papayas. In: Postharvest Diseases of Horticultural Produce (Vol II): Tropical Fruits. Beattie, B. B., McGlasson, W. B., and Wade, N. L. (ed). Department of Primary Industries, Qeensland. pp 63-71

Cordenunsi, B.R., and Lajolo, F.M (1995). Starch Breakdown during Banana Ripening: Sucrose Synthase and Sucrose Phosphate Synthase. Journal of Agricultural and Food Chemistry. **43**:347-351

Cordenunsi, B.R., do Nascimento, J.R.O., Genovese, M.I., and Lajolo, F.M (2002). Influence of Cultivar on Quality Parameters and Chemical Composition of Strawberry Fruits Grown in Brazil. Journal of Agricultural and Food Chemistry: **50**:2581-2586

Crane J.H., and Balerdi, C.F. (1998). The Banana in Florida. University of Florida, Institute of Food and Agricultural Science USA.

Dadzie, B.K. (1998). Postharvest Characteristics of Black Sigatoka Resistant Banana, Cooking Banana and Plantain Hybrids. Inibap Technical Guidelines. International Plant genetic Resources Institute, Italy.

de Guevara, R.G-L., Pardo-González, J.E., Varón-Castellanos, R., and Navarro-Albaladejo, F. (1996). Evaluation of Colour during the Ripening of Selected Varieties of Paprika Pepper (*Capsicum annum* L.). Journal of Agricultural and Food Chemistry **44**:2049-2052

Deli, J., Matus, Z., and Tóth, G. (1996). Carotenoid Composition in the Fruits of *Capsicum annuum* Cv. Szentesis Kosszarvú during Ripening. Journal of Agricultural and Food Chemistry **44**:711-716

Desai, B.B., and Deshpande, P.B., (1978). Chemical Control of Ripening in Banana. Physiological Plantarium **44**:238- 240

Dilley, D.R. (1969). Hormonal Control of Fruit Ripening. HortScience **4**(2):111-114

Drake, S. (1997). Fruit Quality as Influenced by Wax Application. 13th Annual Postharvest Conference.

Durand, B.J., Orcan L., Yanko, U., Zauberman, G., and Fuchs Y. (1984). Effect of Waxing on Moisture Loss and Ripening of "Fuerte" Avocado Fruit. HortScience **19**(3):421-423

FAO (1989). Prevention of Postharvest Food Loss Fruits, Vegetables and Root Crops. A Training Manual. Food and Agricultural Organizations of The United Nations, Rome.

FAO (1990). Production Year Book. Food and Agriculture Organization, Rome.

Ferris, R.S.B. (1997). Improving Storage Life of Plantain and Banana. IITA Research Guide 62. Training material Unit, IITA, Ibadan, Nigeria.

Ferris, R.S.B. (1998). Postharvest Physiology of Plantain and Banana. IITA Research Guide 64. Training Materials Unit, IITA, Ibadan, Nigeria.

Frenkel, C. (1975). Role of Auxin in the Hormonal Regulation of Fruit Ripening. Proceedings XIX International Horticultural Congress. pp 2, 103-118

Frenkel, C., and Dyck, R. (1973). Auxin inhibition of Ripening. Plant Physiology **51**:6-9

Frenkel, C., Dyck, R., and Haard, N.F. (1975). Role of Auxin in the Regulation of Fruit Ripening. In: Postharvest Biology and Handling of Fruits and Vegetables. Haard, N., and Salunkhe, D.K. (ed) AVI publishing Company, USA. pp .19-34

Fuchs, Y., and Temkin-Gorodeiski, N. (1971). The Course of Ripening of Banana Fruits Stored in Sealed Polyethylene Bags. Journal of the American Society for Horticultural Science **96**(4):401-403

García, E., and Lajolo, F.M. (1988). Starch Transformation during Banana Ripening: The Amylase and Glucosidase Behaviour. Journal of food science **53**:1181-1186

García, J.M., Medina, R.J., and Olías, J.M. (1998). Quality of Strawberries Automatically Packed in Different Plastic Film. Journal of Food Science **63**(6):1037-1041

Garner, D., Crisosto, C. H., Wiley, P., and Crisosto, G. M. (2001). Measurement of pH and Titratable Acidity. In: Establishing a Quality Control System. Crisoto, C.H. (ed). Pomology Department, UC Davis Kearney Agricultural Center, Parlier, CA.

Gauhl, F., Ferris, S., Pasberg-Gauhl, C., and Lawrence, A. (1998). On-Farm Yield Loss Assessment of Black Sigatoka on Plantain and Banana. IITA Research Guide **67**. Training Program, IITA, Ibadan, Nigeria.

Geeson, J.D., and Smith S.M. (1989). Retardation of Apple Ripening During Distribution by the Use of Modified Atmosphere. Acta Horticulturae **258**:245-253

George, J.B., and Marriott, J. (1985). The Effect of Some Storage Conditions on the Storage Life of Plantain. Acta Horticuturae **158**:439-447

Golding, J.B., Shearer, D., McGlasson, W.B., and Wyllie, S.G. (1999). Relationship between Respiration, Ethylene, and Aroma Production in Ripening Banana. Journal of Agricultural and Food Chemistry **47**:1646-1651

Gonzalez, G., Yahia, E.M., and Higuera, I. (1990). Modified Atmosphere Packaging (MAP) of Mango and Avocado Fruit. Acta Horticulturae **269**:335-344

Grierson, D., Tucker, G.A., and Robertson, N.G. (1981). The Molecular Biology of Ripening. In: Recent Advances in the Biochemistry of Fruits and Vegetables. Friend, J., and Rhodes, M.J.C. (ed) Academic Press, London. pp149-180

Hagenmaier, R.D., and Baker, R.A. (1993). Reduction in Gas Exchange of Citrus Fruit by Wax Coating. Journal of Agricultural and Food Chemistry **41**:283-287

Hagenmaier, R.D., and Baker, R.A. (1994). Wax Microemulsions and Emulsions as Citrus Coating. Journal of Agricultural and Food chemistry **42**:899-902

Hardenburg, R. E. (1975). Part I Principles of Packaging General Considerations. In: Postharvest Physiology, Handling and Utilization of Tropical and Subtropical Fruits and Vegetables. Pantastico, Er.B. (ed) VI Publishing Co.Westport Connecticut. pp 283-302

Hardenburg, R.E., Watada A.E., and Wang C.Y. (1980). The Commercial Storage of Fruits, Vegetables, and Florist and Nursery Stocks. United States Dept. of Agriculture, Handbook. No 66. MD.

Harker, F.R., Redgwal, R.J., Hallett, I.C., and Murry, S.H. (1997). Texture of Fresh Fruit. Horticultural Review **20**:121-224

Harris, S.R. (1988). Production is only Half the Battle: A Training Manual in Fresh Produce Marketing for the Eastern Caribbean. Food and Agriculture Organization of the United Nations, Barbados.

Hendry, G.A., Houghton, J.D., and Brown, S.B. (1987). The Degradation of Chlorophyll - A Biological Enigma. New phytologist **107**:255-302

Hewett, E.W., Banks, N.H., and Dixon, J. (1989). Shelf-Life Extension of Apples with Polymeric Films. Acta Horticulturae **258**:237-243

Hintlian, C.B., and Hotchkiss, J.H. (1986). The Safety of Modified Atmosphere Packaging: A Review. Food Technology **40**(6):70-76

Hobson, G.E. (1979). What Factors are involved in the onset of Ripening in Climacteric Fruits? Current Advances in Plant Science. pp 1-10.

Hobson, G.E. (1981). Enzymes and Texture Changes during Ripening. In: Recent Advances in the Biochemistry of Fruits and Vegetables. Friend J., and Rhodes, M. J. C. (ed) Academic Press, London. pp123-148

Hobson, G.E. (1987). Low Temperature and the Storage of Ripening Tomatoes. Journal of Horticultural Science **62**(1):55-62

Hobson, G., and Burton, K.S. (1989). The Application of Plastic Film Technology to the Preservation of Fresh Horticultural Produce. Professional Horticulture **3**:20-23

Hoffman, N.E., and Yang, S.F. (1980). Changes in 1-Aminocyclopropane 1-carboxylic acid Content in Ripening Fruits in Relation to Their Ethylene Production Rates. Journal of the American Society for Horticultural Science **105**:492-495

Hubbard, N.L., Pharr, D.M., and Huber, S.C. (1990). Role of Sucrose Phosphate Synthase in Sucrose Biosynthesis in Ripening Bananas and Its Relationship to the Respiratory Climacteric. Plant Physiology **94**:201-208

Huber, D., Jeong, J., and Ritenour, M. (2003). Use of 1-Methylcyclopropane (1-MCP) on Tomato and Avocado Fruits: Potential for Enhanced Shelf-Life and Quality Retention. [Internet] University of Florida, USA. Available from http://edis.ifas.ufl.edu/BODY_Hs151 [accessed 12 March 2003]

Illeperuma, C.K., and Jayasuriya, P. (2002). Prolonged Storage of 'Karuthacolomban' Mango by Modified Atmosphere Packaging at Low Temperature. Journal of Horticultural Sciences and Biotechnology **77**(2):153-157

Inaba, A., and Nakamura, R. (1988). Numerical Expression for Estimating the Minimum Ethylene Exposure Time Necessary to Induce Ripening Banana Fruits. Journal of the American Society for Horticultural Science **113**(4):561-564

Jackman, R.L., Marangoni, A.G., and Stanley, D.W. (1990). Measurement of Tomato Fruit Firmness. HortScience **25**(7):781-783

Jenks, M.A., and Ashworth, E.N. (1999). Plant Epicuticular Waxes: Function, Production, and Genetics. Horticultural Review **23**: 1-68

Jobling, J. (2000). Postharvest Ethylene: A Critical Factor in Quality Management. Good Fruit and Vegetables Magazine **11**. Melbourne, Australia.

Jobling, J. (2001). Modified Atmosphere Packaging: Not as Simple as It Seems. Good Fruit and Vegetables Magazine **11**(5). Melbourne, Australia

John, P., and Marchal, (1995). Ripening and Biochemistry of the Fruit. In: Banana and Plantains. Gowen, S. (ed). Chapman and Hall. London pp 434-467

Jones, D.R., and Wade, N.L. (1995). Bananas. In: Postharvest Diseases of Horticultural Produce (Vol II): Tropical fruits. Beattie, B. B., McGlasson, W. B., and Wade, N. L. (ed). Department of Primary Industries, Qeensland. pp 18-31

Kader, A.A. (1992a). Postharvest Biology and Technology. In: Postharvest Technology of Horticultural Crops. (2nded). Kader, A.A. (ed). University of California Division of Agriculture and Natural Resources. Special Publication 3311, Okland CA pp15-20

Kader, A.A. (1992b). Modified Atmosphere during Transport and Storage. In: Postharvest Technology of Horticultural Crops. (2nded). Kader, A.A. (ed). University of California Division of Agriculture and Natural Resources. Special Publication 3311, Okland CA pp85-92

Kader, A.A. (1994). Fruit Maturity, Ripening and Quality Relationships. Perishable Handling Newsletter, Published by University of California, USA.

Kader, A.A. (1999). Fruit Maturity, Ripening, and Quality Relationship. Acta Horticulturae. **485**:203- 207

Kader, A.A. (2000a). Banana. In: Recommendations for Maintaining Postharvest Quality. Department of Pomology, University of California, Davis, USA

Kader, A.A. (2000b). Papaya. In: Recommendations for Maintaining Postharvest Quality. Department of Pomology, University of California, Davis, USA

Kays, S.J. (1991). Postharvest Physiology of Perishable Plant Products. Nostrand Reinhold, New York. pp 1,229-247

Ke, L.S. (1979). Studies on the Physiological Characteristics of Banana in Taiwan. Effect of Soil Moisture on Physiological Functions and Yield of Banana Plant. Journal of the Agricultural Association of China **108**:11

Knee, M. (1984). 1- Aminocyclopropane-1- Carboxylic Acid Level in Ripening Apple Fruits. Journal of Experimental Botany **35**:1794-1799

Khader, S.E.S.A. (1992). Effect of Gibberellic Acid and Vapour Gard on Ripening, Amylase and Peroxidase Activities and Quality of Mango Fruit during Storage. Journal of Horticultural Science **67**(6):855-860

Khader, S.E.S.A., Singh, B.P., and Khans, S.A. (1988). Effect of GA as a Postharvest Treatment of Mango Fruits. Scientia Horticulturae **36**:261-266

Kotecha, P.M., and Desai, B.B. (1995). Banana. In: Fruit Science and Technology (Salunkhe, D.K., and Kadom, S.S. (ed). Marcel Dekker, Inc. New York. pp 67

Krishnamurthy, S., and Kushalappa, C.G. (1985). Studies on the Shelf-Life and Quality of Robusta Bananas as Affected by Postharvest Treatments. Journal of Horticultural Science **60**(4):549-556

Kruger, F.J., Claassens, V., and Madeleine, L. (1996). Gold-Finger Consumer Survey: Retrospection and Pointers for Future Cultivar Release in South Africa. Banana Growers Association of South Africa. Year Book 1:47-54

Lagerwall, G.B. (1997). Banana (*Musa* AAA; Cavendish Subgroup) Cultivar Density Trials in Three Bioclimatic Groups on the Northern Coast of Kwazulu-Natal. University of Natal, Department. of Horticultural Science, Thesis M.Sc. in Agriculture.

Lam, P.F. (1990). Respiration Rate, Ethylene Production and Skin Colour Change of Papaya at Different Temperatures. Acta Horticulturae **269**:257-267

Lau, O.L., Liu Y., and Yang, S.F. (1985). Effect of Fruit Detachment on Ethylene Biosynthesis and Loss of Fresh Firmness, Skin Colour and Starch in Ripening "Golden Delicious Apple". Submitted to Journal of the American Society for Horticultural Science.

Lazan, H., Ali, M.Z., and Sim, W.C. (1990a). Retardation of Ripening and Development of Water Stress in Papaya Fruit Seal-Packaged with Polyethylene Film. Acta Horticulturae **269**:345-357

Lazan, H., Mohd.Ali, Z., and A.Sani, H. (1990b). Effect of Vapour Gard on Polygalacturonase, Malic Enzyme and Ripening of Harumanis Mango. Acta Horticulturae **269**:359-366

Lazan, H., Ali, Z. M., and Selamat, M.K. (1993). The Underlying Biochemistry of the Effect of Modified Atmosphere and Storage Temperature on Firmness Decrease in Papaya. Acta Horticulturae **343**:141-147

Lebibet, D., Metzidakis, I., and Gerasopoulos, D. (1995). Effect of Storage Temperature on the Ripening Response of Banana (*Musa. sp*) Fruit Growing in the Mild Winter Climate of Crete. Acta Horticulturae **379**:521-526

Leliévre, J.M., Latché, A., Jones, B., Bouzayen, M., and Pech, J.C. (1997). Ethylene and Fruit Ripening. Physiological Plantarium **101**:727-739

Liadó, J.D., and Domínguez, A.M. (1998). The Effect of Peel Abrasion on the Postharvest Physiology and Commercial Life of Banana. Acta Horticulturae **490**:547-553

Lindsay, P., and Brian, C. (1982). Fruit Growing in Warmer Climates. Reed Books Private Ltd., Hong Kong.

Liu, F. (1970). Storage of Banana in Polyethylene Bags with an Ethylene Absorbent. HortScience **5**(1):25-27

Lurie, S., Arie, R.B., and Zilkah, S. (1997). The Ripening and Storage Quality of Nectarine Fruits in Response to Preharvest Application of Gibberellic Acid. Acta Horticulturae **463**: 341-346

Lutz, J.M., and Hardenburg, R.E. (1966). The Commercial Storage of Fruits, Vegetables and Florists and Nursery Storks. Agricultural Handbook No66, USDA, Washington.

Macnish, A.J., Joyce, D.C., and Hetherington, S.E. (1997). Packaging to Reduce Water Loss can Delay Ripening of Mango (*Mangifera indica* L. cv. 'Kensington Pride) Fruit. Australian Journal of Experimental Agriculture **37**:463-467

Maharaj, R. (1988). The Handling and Storage of Papaya (*Carica papaya* L.) under Controlled Condition. M.Sc. Thesis, Department of Chemical Engineering, University of the West Indies, St. Augustine Trinidad.

Marcelle, R., and Clijsters, H. (1978). Effect of Growth Regulators on the Absorption and Distribution of Calcium in Fruits. Acta Horticulturae **80**:353-360

Marchal, J. (1998). An overview of Postharvest Aspects of Banana ISH. Acta Horticulturae **490**:501-506

Marler, T.E. (1994). Papaya. In: Handbook of Environmental Physiology of Fruit Crops. (Vol II). Subtropical Crops. Schafter, B., and Andersen, P.C. (ed). CRC Press Inc. Florida. pp 216

Mattoo, A.K., and Handa, A. (2001). Postharvest Science Toward the Third Millennium. Acta Horticulturae **553**:761-767

Mattoo, A.K., Murata, T., Pantastico, Er.B., Chachin, K., Ogata, K., and Phan, C.T. (1975). Chemical Changes during Ripening and Senescence. In: Postharvest Physiology, Handling and Utilization of Tropical and Subtropical Fruits and Vegetables. Pantastico, Er.B. (ed). AVI Publishing Co. Westport Connecticut. pp 103-125

Medlicott, A. (2001). Postharvest Handling of Papaya. Product Specification and Postharvest Handling for Fruits, Vegetables and Root Crops Exported from the Caribbean.

Mehta, P.M., Shiva, R.S., and Raju, P.S. (1986). Influence of Fruit Ripening Retardant on Succinate and Malate Dehydrogenases in Papaya Fruit with Emphasis on Preservation. Indian Journal of Horticulture **43**:169-173

Meheriuk, M., and Porrit, S.W. (1972). Effect of Waxing on Respiration, Ethylene Production, and other Physical and Chemical Changes in Selected Apple Cultivars. Canadian Journal of Plant Sciences **52**(3):257-259

Miller, W.R., Spalding, D.H., and Hale, P.W.(1986). Film Wrapping Mango at Advancing Stage of Postharvest Ripening. Tropical Science **26**:9-17

Miller, W.R., Hale, P.W., Spalding, D.H., and Davis, P. (1983). Quality and Decay of Mango Fruit Wrapped in Heat-Shrinkable Film. HortScience **18**(6):957-958

Mínguez-Mosquera, M.I., and Hornero-Méndez, D. (1994). Changes in Carotenoids Esterification during the Fruit Ripening of *Capsicum annuum* Cv. Bola. Journal of Agricultural and Food Chemistry **42**:640-644

Mínguez-Mosquera, M.I., Pérez-Gálvez, A., and Garrido-Fernández, J. (2000). Carotenoid Content of the Varieties Jaranda and Jariza (*Capsicum annum* L.) and Response

during the Industrial Slow Drying and Grinding Steps in Paprika Processing. Journal of Agricultural and Food Chemistry **48**(7):2972-2976

Mitra, S.K., and Baldwin, E.A. (1997). Mango. In: Postharvest Physiology and Storage of Tropical and Subtropical Fruits. Mitra, S. (ed). CAB International, London. pp 85-111

Mustaffa, R., Osman, A., Yusof, S., and Mohamed, S. (1998). Physico-Chemical Changes in Cavendish Banana (*Musa Cavendish* L. var Montel) at Different Position within a Bunch during Development and Maturation. Journal of the Science of Food and Agriculture **78**:201-207

Nakasone, H.Y. and Paull, R.E. (1998). Tropical Fruits. CAB International, London.

Nunes, M.C.N., Bercht, J.K., Morias, A.M.M.B., and Sargent, S.A. (1998). Controlling Temperature and Water Loss to Maintain Ascorbic Acid Levels in Strawberries during Postharvest Handling. Journal of Food Science **63**(6):1033-1036

Olorunda, A.O. (2000). Recent Advances in Postharvest Technologies of Banana and Plantain in Africa. Acta Horticulturae **540**:517-527

Olorunda, A.O., and Aworh, O.C. (1984). Effect of Tal-Prolong, a Surface Coating Agent, on the Shelf-Life and Quality Attributes of Plantain. Journal of the Science of Food and Agriculture **35**:573-578

Osman, A., and Ayub, M.N.A. (1998). Effect of Different Postharvest Treatments on the Respiration Patterns of Guava (*Psidium guajava* L.). Acta Horticulturae **464**: 502

Pantastico, Er.B. (1975). Preharvest Factors Affecting Quality and Physiology after Harvest. In: Postharvest Physiology, Handling and Utilization of Tropical and Subtropical Fruits and Vegetables. Pantastico, Er.B. (ed) AVI Publishing Co. Westport Connecticut. pp 25-40

Pantastico, Er.B., Chattopadhyay, T.K., and Subramanyam, H. (1975). Storage and Commercial Storage Operations. In: Postharvest Physiology, Handling and Utilization of Tropical and Sub-tropical Fruits and Vegetables. Pantastico, Er.B. (ed) AVI Publishing Co. Westport Connecticut. pp314-338

Parikh, H.R., Nair, G.M., and Modi, V.V. (1990). Some Structural Changes during Ripening of Mangoes (*Mangifera indica* vr. Alphonso) by Abscisic Acid Treatment. Annals of Botany **65**:121-127

Pariser, E.R., Avensu, E.S., and Bourne, M.C. (1978). Postharvest Food Losses in Developing Countries. National Academy of Science, Washington, DC.

Paull, R.E. (1993). Pineapple and Papaya. In: Biochemistry of Fruit Ripening. Seymour, G., Taylor, J.E., and Tucker, G.A. (ed). Chapman and Hall, London. pp 291-323

Paull, R.E. (1994). Tropical Fruit Physiology and Storage Potential. In: Postharvest Handling of Tropical Fruits (ed). ACIAR Proceeding No **50**, Canberra pp 198- 204

Paull, R.E. (1997).Pineapples. In: Postharvest Physiology and Storage of Tropical and Subtropical Fruits. Mitra, S. (ed). CAB International, London. pp 123-144

Paull, R.E., and Chen, N.J. (1983). Postharvest Variation in Cell wall-Degrading Enzymes of Papaya (*Carica papaya* L.) during Ripening. Plant Physiology **72**:382-385

Paull, R.E., and Chen, N.J. (1989). Waxing and Plastic Wraps Influence Water Loss from Papaya Fruit during Storage and Ripening. Journal of the American Society for Horticultural Science **114**(6):937-942

Pekmezci, M., Erkan, M., and Gübbük, H. (1997). The Effect of Harvesting Time, and Method and Duration of Storage on Quality of "Hachiya" and "Fuyu" Persimmons. Acta Horticulturae **441**:279-286

Pérez, A.G., Olías, R., Espada, J., Olías, J.M., and Sanz, C. (1997). Rapid Determination of Sugars, Nonvolatile Acids, and Ascorbic Acid in Strawberry and other Fruits. Journal of Agriculture and Food Chemistry **45**(9):3545-3549

Phan, C.T., Pantastico, Er.B., Ogata, K., and Chachin, K. (1975). Respiration and Respiratory Climacteric. In: Postharvest Physiology, Handling and Utilization of Tropical and Subtropical Fruits and Vegetables. Pantastico, Er. B. (ed) AVI Publishing Co. Westport Connecticut. pp 86-102

Proft, M.P., Omoaka, P., and Pekke, A.M.(1998). Forced Ripening of Bananas: Evaluation of the after Treatment Temperature. Acta Horticulturae **490**:555-561

Purgatto, E., Lajolo, F.M., do Nascimento, J.R.O., and Cordenunsi, B.R. (2001). Inhibition of β-Amylase Activity, Starch Degradation and Sucrose Formation by Indole-3-Acetic Acid during Banana Ripening. Planta **212**:823-828

Quamme, H.A., and Gray, J.I. (1985). Pear Fruit Quality and Factors that Condition It. In: Evaluation of Quality of Fruits and Vegetables. Pattee, H.E. (ed), AVI Publishing Co. Westport Connecticut. pp 47- 61

Rao, D.V.R., and Chundawat, B.S. (1990). Effect of Certain Ripening Retardants on Postharvest Behaviour of Basrai Banana at Non-Refrigerated Temperature. In: Advances in Horticulture and Forestry (Vol 1). Singh, S.P. (ed), Scientific Publishers, Jodhpur, India. pp 33-48

Reid, M.S. (1992). Ethylene in Postharvest Technology. In: Postharvest Technology of Horticultural Crops. (2nd ed). Kader, A.A. (ed). University of California Division of Agriculture and Natural Resources. Special Publication 3311, Okland CA pp 97-108

Richmond, M.L., Brandao, S.C.C., Gray, J.I., Markakis, P., and Stine, C.M. (1981). Analysis of Simple Sugars and Sorbitol in Fruits by High-Performance Liquid Chromatography. Journal of Agricultural and Food Chemistry. **29**:4-7

Robbins, J.A., and Moore, P.P. (1990). Colour Change in Fresh Red Raspberry Fruit Stored at 0, 4.5, or 20°C. HortScience **25**(12):1623-1624

Robinson, J.C. (1996). Banana and Plantains. CAB International, University Press, Cambridge

Rom, C.R. (1996). Environmental Factors Regulating Growth: Light, Temperature, Water, Nutrient. In: Tree Fruit Physiology Growth and Development. Maib, K.M., Andrews, P.K., Lang, G.A., and Mullinix, K. (ed). Good Fruit Grower, Washington. pp 11- 30

Rosen, J.C., and Kader, A.A. (1989). Postharvest Physiology and Quality Maintenance of Sliced Pear and Strawberry Fruits. Journal of Food Science **54**(3):656-659

Ryall, A.L., and Pentzer, W.T. (1982). Handling, Transportation and Storage of Fruits and Vegetables (2nd ed.) Vol II AVI Publishing Company Inc. Westport Connection.

Salama, A.M., Hicks, J.R., and Nock, J.F. (1990). Sugar and Organic Acid Changes in Stored Onion Bulbs Treated with Maleic Hydrazide. HortScience **25**(12):1625-1628

Saltveit, M.E. (2002). Respiration Metabolism. Mann Laboratory, Department of Vegetable Crops, University of California, Davis, CA.

Salunkhe, D.K., and Desai, B. B. (1984a). Postharvest Biotechnology of Fruits (Vol. I). CRc Press, Inc. Florida.

Salunkhe, D.K., and Desai, B.B. (1984b). Postharvest Biotechnology of Fruits (Vol. II). CRc Press, Inc. Florida. pp1

Salunkhe, D.K., and Kadom, S.S. (1995). Handbook of Fruit Science and Technology: Production, Composition, Storage and Processing. Marcel Dekker, Inc. New York. pp 1, 297

Salunkhe, D.K., Bolin, H.R., and Reddy, N.R. (1991). Storage Processing, and Nutritional Quality of Fruits and Vegetables (2nd ed). Vol. I. CRS Press Inc. Boston.

Samson, J.A. (1980). Tropical Fruits. Longman, Scientific and Technical, London.

Sankat, C.K., and Maharaj, R. (1997). Papaya. In: Postharvest Physiology and Storage of Tropical and Subtropical Fruits. Mitra, S. (ed) CAB International, London. pp167-185

Sanz, C., Pérez, A.G., Olías, R., and Olías, J.M. (1999). Quality of Strawberries Packed with Perforated Polypropylene. Journal of Food Science **64**(4):748-752

Satyan, S., Scott, K.J., and Graham, D. (1992a). Storage of Banana Bunches in Sealed Polyethylene Tubes. Journal of Horticultural Science **67**(2) 283-287

Satyan, S., Scott, K.J., and Best, D.J. (1992b). Effect of Storage Temperature and Modified Atmosphere on Cooking Banana Grown in New South Wales. Tropical Agriculture (Trinidad) **69**(3):263-267

Schaffer, B., and Andersen, P.C. (1994). Introduction. In: Handbook of Environmental Physiology of Fruit Crops. (Vol II). Subtropical Crops. Schafter, B., and Andersen, P.C. (ed). CRC Press Inc. Florida. pp 1-2

Scott, K.J., and Gandanegara, S. (1974). Effect of Temperature on the Storage Life of Banana Held in Polyethylene Bags with Ethylene Absorbent. Tropical Agriculture (Trinidad) **51**(1):23-26

Scott, K.J., Blake, J.R., Strachan, G., Tugwell, L.B., and McGlasson, W.B. (1971). Transport of Banana at Ambient Temperatures using Polyethylene Bags. Tropical Agriculture (Trinidad) **48:**245-254

Seberry, J.A., and Harris, D.R. (1998). Postharvest Evaluation of FHIA-01 and other New Banana Variety for Subtropical Australia. Acta Horticulturae **490**:537-546

Seeley, E.J. (1981). Plant Growth Regulators and a Fruit Tree Physiology. In: Tree Fruit Growth Regulator and Chemical Thinning. Tukey, R.B., and Williams, M.W. (ed). Short Course Proceedings, Washington State University, WA. Pp 46-50

Seymour, G.B. (1985). The Effect of Gases and Temperature on Banana Ripening. Ph.D. Thesis, University of Reading. England.

Seymour, G.B. (1993). Banana. In: Biochemistry of Fruit ripening. Seymour, G., Taylor, J.E., and Tucker, G.A. (ed) Chapman and Hall. London pp 83-106

Seymour, G.B., Tucker, G.A., and Wainwright, H. (1990). Cell Wall Changes in Ripening Fruit. Acta Horticulturae **269**:249-253

Shanmugavelu, K.G., Aravindakshan, K., and Sathiyamoorthy, S. (1992). Banana: Taxonomy, Breeding, and Production Technology. Metropolitan Book Co. Pvt.Ltd. New Delhi.

Sharples, R.O., and Johnson, D.S. (1986). Effect of Some Growth Regulators on the Ripening and Storage Quality of Apples and Pears. Acta Horticulturae **179**:721-727

Shewfelt, R.L. (1986). Postharvest Treatment for Extending the Shelf-Life of Fruits and Vegetables. Food technology **40**(5):70-81

Shewfelt, L.R., and Prussia, E.S. (1993). Postharvest Handling: A Systems Approach Academic Press, Inc. San Diego. pp 64-93

Simmonds, N.W. (1966). Bananas. (2nd ed). Longman London.

Singh, Z., and Janes, J. (2001). Effect of Postharvest Application of Ethephone on Fruit Ripening, Quality and Shelf-Life of Mango under Modified Atmosphere Packaging. Acta Horticulturae **553**:599-601

Sivalingam, P.M., and Charles, S.V. (1995). Biopreservation of Flocal Fruits. SEAMEO-Jasper Fellowship Monography, Series 3., The University of Science Malaysia.

Smith, S., Geeson, J., and Stow, J. (1987). Production of Modified Atmosphere in Deciduous Fruits by the use of Film and Coating. HortScience **22**:772-776

Smith, N.J., Jeger, M.J., Seymour, G.B., and Tucker, G.A. (1990). Cell Wall Changes in Bananas and Plantains. Acta Horticulturae **269**:283-289

Srikul, S., and Turner, D.W. (1995). High N Supply and Soil Water Deficits Change the Rate of Fruit Growth of Bananas (cv. Williams) and Promote Tendency to Ripen. Scientia Horticulturae **62**:165-174

Stover, R.H., and Simmonds, N.W. (1987). Bananas (3rd ed). Longman Scientific and Technical, New York. pp 386-393

Strydom, G.J. (1991). The Effect of Gamma Radiation (^{60}Co) on the Postharvest Physiology of *Musa acuminata* collar cv. Dwarf Cavendish. Ph.D Thesis Department. of Botany, University of Natal, Pietermaritzburg.

Swarts, D.H. (1985). The Postharvest Handling of Papaya, Citrus and Subtropical Fruits Research Nelspruit. Papaya I. pp1

Swarts, D.H. (1992). The Effect of Cooling Rate on the Quality of Bananas Harvested during Hot Conditions. Institute for Tropical and Subtropical Crops, Annual Report 1991-1992 pp 27

Swinburne, T.R., (1983) Quiescent Infection in Postharvest Disease. In: Postharvest Pathology of Fruits and Vegetables. Dennis, C. (ed) Academic Press Inc. London. pp 1-17

Talhouk, S.N., Ghalayini, A., and Toufeili, I. (1999). Effect of Temperature and Polyethylene Wraps on Storage Life of Loquat. In: Postharvest Losses of Perishable Horticultural Crops in the Mediterranean Region. Gerasopoulos, D. (ed) CIHEAM-Options Mediterraneennes, Chania, Greece. pp103-113

Tan, S.C., and Ali, A.M. (1989). The Effect of CO_2 on Enzyme Activity during Storage of "Mas" Banana. In: Managing Postharvest Horticulture in Australia. Beattie, B.B. (ed). NSW Agriculture and Fisheries, Australia.

Techawongstien, S. (1999). Postharvest Technology and Packaging of Vegetables and Fruit Crops. Paper for Training Course on Agricultural Technology for Crop Production at Khon Kaen University, Thailand.

Terblanche, E. (1999). Effect of Temperature on the Colour of Citrus during Degreening. ASAE Annual Meeting Presentation, No 996120, University of Pretoria, South Africa.

Thompson, A.K. (1996). Postharvest Technology of Fruit and Vegetables. Black well Science Ltd, Cambridge.

Thompson, A.K., and Burden, O.J. (1995). Harvesting and Fruit Care. In: Banana and Plantain. Gowen, S. (ed). Chapman and Hall, London. pp 403-433

Thompson, A.K., and Lee, G.R. (1971). Factors Affecting the Storage Behaviour of Papaya Fruit. Journal of Horticultural Science **46**:511-516

Toma, R.B., Fansler, L.T., and Knipe, M.T. (1990). World Food Shortage: The Third Dimension. In: Science of Food and Agriculture **365**:1-5.

Truter, A.B. (1992). Controlled Atmosphere Storage of Banana. Institute for Tropical and Subtropical Crops, Annual Report 1991-1992 pp 27

Tucker, G.A. (1993). Introduction. In: Biochemistry of Fruit Ripening. Seymour, G., Taylor, J.E., and Tucker, G.A. (ed). Chapman and Hall, London. pp 1-43

Turner, D.W. (1997). Banana and Plantains. In: Postharvest Physiology and Storage of Tropical and Subtropical Fruits. Mitra, S. (ed). CAB International, London. pp 47-84

Vendrell, M. (1969). Reversion of Senescence: Effect of 2,4-Dichlorophenoxyacetic Acid and Indole Acetic Acid on Respiration, Ethylene Production, and Ripening of Banana Fruit Slices. Australian Journal of Biological Sciences **22**:601-610

Vendrell, M. (1985). Dual effect of 2,4-D on Ethylene Production and Ripening of Tomato Fruit Tissue. Physiological Plantarium **64**:559-563

Vendrell, M., and Palomer, X. (1997). Hormonal Control of Fruit Ripening in Climacteric Fruits. Acta Horticulturae **463**:325-332

Venter B. (1993). Postharvest Handling of Papayas. Institute for Tropical and Subtropical Crops, Nelspruit. Papaya I. pp1

Visai, C., Trecanni, C. P., and Mezzadir, G. (1980). The Effect of GA_3 and a Calcium Salt on the Occurrence of Internal Browning in Pear, Cv Passe crassane. Revista Della Ortofloro Fruitticoltura. Italiana, **64**:257-264ear

Von Loesecke, H. (1949). Banana. Interscience. New York.

VSN. 2001. GenStat Release 4.2 (5^{th} ed). VSN International Ltd, Lawes Agricultural Trust, Oxford.

Wang, C.Y. (1999). Postharvest Quality Decline, Quality Maintenance and Quality Evaluation. Acta Horticulturae **485**:389-392

Watkins, C.B., and Pritts, M.P. (2001). The Influence of Cultivar on Postharvest Performance of Fruit and Vegetables. Acta Horticulturae **553**:59-63

Weir, R.G., and Cresswell, G.C. (1993). Plant Nutrient Disorder: Temperate and Subtropical Fruit and Nut Crops. NSW Agriculture. INKATA Press. Melbourne, Sydney.

Weis, S.A., and Bramlage, W.J. (2002). 1-MCP: How Useful Can it be on New England Apples? Fruit Notes **67**:5-9

Wellburn, A.R.J.P.P. (1994). The Spectral Determination of Chlorophyll a and b, as well as Total Carotenoids, Using Various Solvents with Spectrophotometer of Different Resolution. Journal of Plant Physiology **144**:307-313

Wills, R.B.H., and Widjanarko, S.B. (1995). Changes in Physiology, Composition and Sensory Characteristics of Australian Papaya during Ripening. Australian Journal of Experimental Agriculture **35**:1173-1176

Wills, R.B.H., Lim, J.S.K., and Greenfield, H. (1983). Changes in Chemical Composition of 'Cavendish' Banana (*Musa acuminata*) during Ripening. Journal of Biochemistry **8**:67-77

Wills, R.B.H., McGlasson, B., Graham, D., and Joyce, D. (1998) Postharvest: An Introduction to the Physiology and Handling of Fruit and Vegetable. (4th ed) CAB International, Adelaide, Australia.

Wills, R.B.H., McGlasson, W.B., Graham, D., Hall, E.G., and Lee, T.H. (1989) Postharvest: An Introduction to the Physiology and Handling of Fruit and Vegetable. (3rd ed) BSP Professional Books, London.

Wilson, L.G., Boyette, M.D., and Estes, E.A. (1999). Quality Maintenance (Part I). In: Postharvest Handling and Cooling of Fresh Fruit, Vegetables, and Flowers for Small Farmers. Horticultural Information Leaflet, **800**, Northern Carolina State University (NCSU).

Wojcik, P., Mika, A., and Cieslinski, G. (1999). Effect of Boron Fertilization on The Storage Ability of Apples (*Malus domestica* Borth). Acta Horticulturae **485**:393-397

Yahia, E.M. (1998). Modified and Controlled Atmosphere for Tropical Fruits. Horticultural Review **22**:123-184

Yamauchi, N., and Watada, A.E. (1991). Regulated Chlorophyll Degradation in Spinach Leaves during Storage. Journal of the American Society for Horticultural Science **116**(1):58-62

Yang, S.F., Liu, Y., and Lau, L.O. (1986). Regulation of Ethylene Biosynthesis in Ripening Apple Fruit. Acta Horticulturae **179**:711-719

Yantarasri, T., Ben-Yehoshua, S., Rodov, V., Kumpuan, W., Uthaibutra, J., and Sornsrivichai, J. (1995). Development of Perforated Modified Atmosphere Package for Mango. Acta Horticulturae **398**:81-88

Yoneya, T., Nip W.K., Wei, P.S., and Cai, T. (1990). Physico-Chemical Parameters of Postharvest Ripened Mangoes from Hawaii. Acta Horticulturae **269**:291-297

Zagory, D. (1998). An Update on Modified Atmosphere Packaging of Fresh Produce. Packaging International. **117**

Zhang, D., Haung, B.Y., and Scott, K.J. (1993). Some Physiological and Biochemical Changes of " Green ripe" Bananas at Relative High Storage Temperatures. Acta Horticulturae **343**:81-85

Appendix 1: Postharvest Losses of Perishable Commodities in Developing Countries.

Commodity	Estimated Loss (%)
Fruits	
Banana	20-80
Papaya	40-100
Avocado	43
Peach, Apricot, and Nectarine	28
Citrus	23-33
Raisins	20-95
Apples	14
Vegetables	
Onion	16-35
Tomatoes	5-50
Plantains	35-100
Cabbage	37
Cauliflower	49
Lettuce	62
Roots and Tubers	
Potatoes	5-40
Sweet Potatoes	35-95
Yams	10-60
Cassava	10-25

(Adapted from Pariser, *et al.*, 1978)

Appendix 2

Calculation for the conversion of ppm to ml $kg^{-1}hr^{-1}$

1ppm = 1mg of solute in 1L of solution

1ppm = μLL^{-1}

1ppm CO_2*(7L - Volume of fruit) = XμL CO_2

Where 7L is the volume of the container

XμL CO_2 /Weight of fruit = YμL CO_2 Kg^{-1}

Fruit respiration rate was determined for 10min. ∴ to change it per hour respiration rate

YμL CO_2 Kg^{-1}*6 = ZμL CO_2 Kg^{-1} hr^{-1}

Final result was expressed in mLCO_2 $kg^{-1}hr^{-1}$ ∴ it was divided to 1000

ZμL CO_2 Kg^{-1} hr^{-1}/1000 = mL CO_2 Kg^{-1} hr^{-1}

Appendix 3

Calculation for the determination of total chlorophyll and carotenoid concentration

$C_a = 16.72A_{665.2} - 9.16A_{652.4}$

$C_b = 34.09A_{652.4} - 15.28A_{665.2}$

$C_{a+b} = 1.44A_{665.2} + 24.93A_{652.4}$

$$C_{x+c} = \frac{1000A_{470} - 1.63\,C_a - 104.96\,C_b}{221}$$

Where: C_a = Chlorophyll a

C_b = Chlorophyll b

C_{a+b} = Total chlorophyll

C_{x+c} =Total carotenoids

A_{470} = Absorbance at 470

$A_{652.4}$ = Absorbance at 652.4

$A_{665.2}$ = Absorbance at 665.2

Total chlorophyll and carotenoids expressed in $\mu g g^{-1}$

Appendix 4

Calculation for the determination of organic acids in the fruits

Method was developed for the calculation of final organic acid concentrations in the fruits after fully ripened.

Solution was prepared from known standards as per the following concentrations.

Malic Acid

Concentrations (mgL^{-1})	Absorbance	Absorbance	Absorbance	Average
115		6843	6563	6703
230	108944	105836	104844	106541
460	245232	241811	243964	243669
920	483034	479817	493597	485483

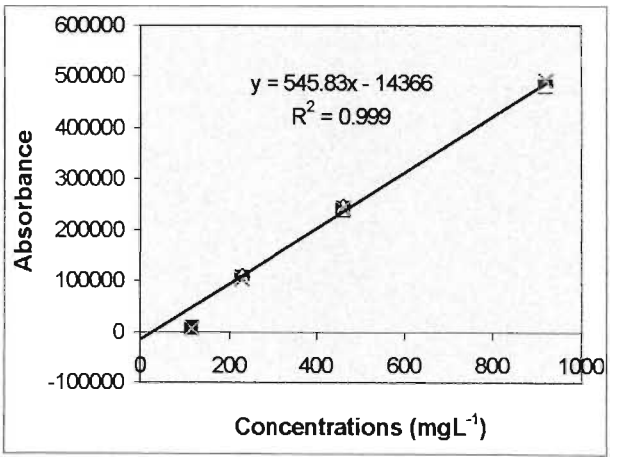


Fig A Regression equation developed to estimate the concentrations of malic acid.

Y= 545.83 x – 14366.....Eq 1

Appendix 4 (Continued)

Citric acid

Concentration (mgL^{-1})	Absorbance	Absorbance	Absorbance	Average
120		4399	3110	3755
240	147960	142301	130477	140246
480	323427	314107	311463	316332
960	631545	619455	627180	626060

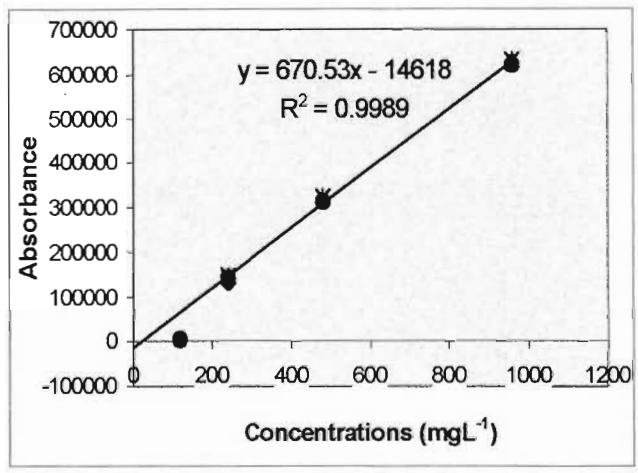


Fig B Regression equation developed to estimate the concentrations of citric acid.

Y= 670.53 X –14618.....Eq 2

Equation 1 and 2 were used to calculate malic and citric acid concentrations, respectively. A chromatogram result of one sample (MPB+K rep 5) obtained from the HPLC after it has been processed is shown in Fig C. Absorbance of malic and citric acid in the chromatogram are shown as 352610.54 and 359782.32 μV*sec, respectively. From the absorbance detected by the HPLC the concentration of each sample fruit was expressed in (mgL^{-1}) and the organic acids were calculated as follows:

Appendix 4 (continued)

Malic acid

$$\text{Concentration } (mgL^{-1}) = \frac{\text{Absorbance}+14366}{545.83}$$

$$= \frac{352610.54 + 14366}{545.83}$$

$$= 672.32\ mgL^{-1}$$

$$= 672.32\ mgL^{-1} * 4/1000\ L$$

$$= 2.7\ mg$$

$$= \frac{100 * (2.7mg/1000)}{5\ g}$$

$$= 0.054\ \%$$

Citric acid

$$\text{Concentration } (mgL^{-1}) = \frac{\text{Absorbance} + 14618}{670.53}$$

$$= \frac{359782.32 + 14618}{670.53}$$

$$= 558.36 mgL^{-1}$$

$$= 558.36\ mgL^{-1} * 4/1000\ L$$

$$= 2.2\ mg$$

$$= \frac{100 * (2.2\ mg/1000)}{5\ g}$$

$$= 0.045\ \%$$

Where: 4 ml is the amount of ml used to extract organic acids

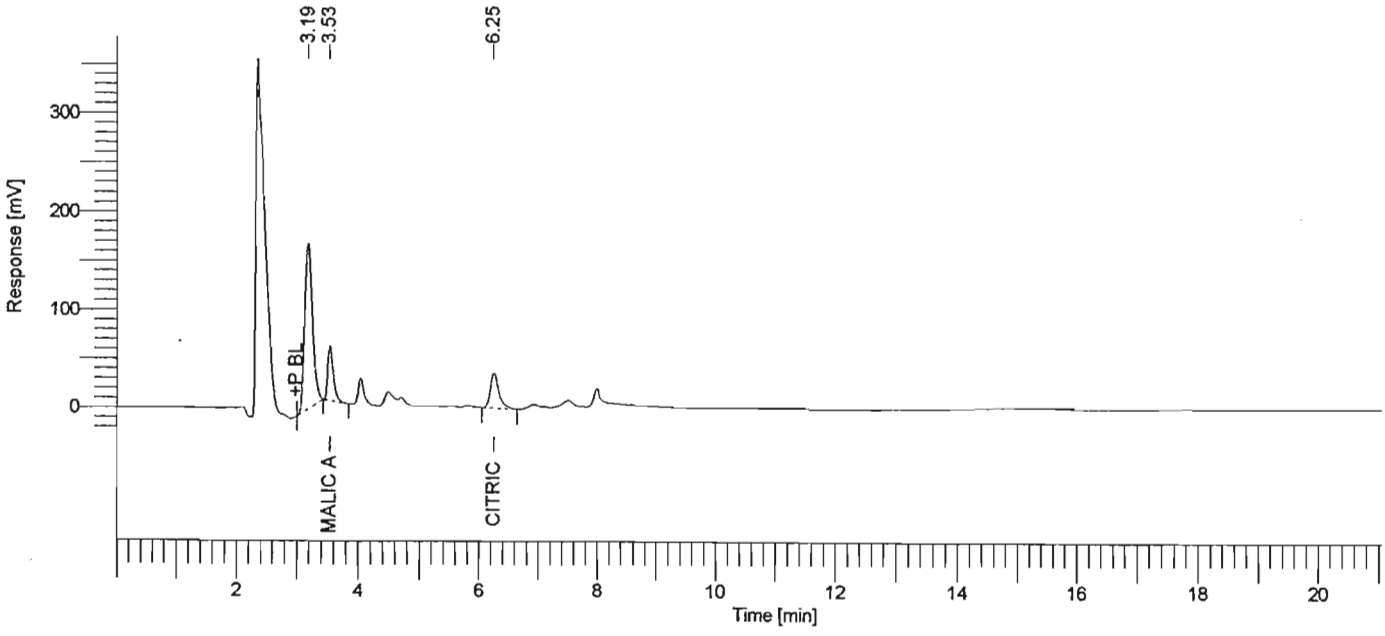
5 g the fresh weight fruit tissue used for the extraction

Appendix 4 (Continued)

Figure C

Software Version	: 6.2.0.0.0:B27	Date	: 1/31/03 4:50:54 PM
Sample Name	: PK5-R	Data Acquisition Time	: 1/22/03 3:42:54 AM
Instrument Name	: PE DAD	Channel	: A
Rack/Vial	: 0/23	Operator	: tcprocess
Sample Amount	: 1.000000	Dilution Factor	: 1.000000
Cycle	: 1		

Result File : C:\PenExe\TcWS\Ver6.2.0\Examples\colin\Biniam\Biniam 3\sample 3008.rst
Sequence File : C:\PenExe\TcWS\Ver6.2.0\Examples\colin\Biniam\Biniam 2\test sequence 2.seq



Organic Acids of Banana Fruit

Peak #	Component Name	Time [min]	Area [uV*sec]	Height [uV]
1		3.187	1481986.18	168961.37
2	Malic Acid	3.534	352610.54	56115.05
3	Citric Acid	6.252	359782.32	36185.65
			2194379.04	261262.06

Missing Component Report
Component Expected Retention (Calibration File)

Appendix 5

Calculation of Titratable Acid

$$\%Acid = \frac{[mls\ NaOH\ Used]x[0.1N\ NaOH]x[milliequivalent\ factor]x[100]}{grams\ of\ Sample}$$

Some of the Common Organic Acids with their Milliequivalent Factors Used to Calculate Titratable Acids Titrated Against 0.1N NaOH

Commodities	Chemical Formula	Predominant Acid	Milliequivalent Factor
Stone fruit, apples, banana, papaya, kiwifruit, Cherries	$H_2C_4H_4O_5$	Malic Acid	0.067
Citrus, banana, papaya, pineapples	$H_3C_6H_5O_7$	Citric Acid	0.064
Banana, papaya	H_2C_2O4	Oxalic Acid	0.045
Grapes, Avocado	$H_2C_4H_4O_6$	Tartaric Acid	0.075
Figs, banana	$HC_2H_3O_2$	Acetic Acid	0.06