EVALUATION OF THE EFFICACY OF COOLBOT™ COLD STORAGE TECHNOLOGY TO PRESERVE QUALITY AND EXTEND SHELF LIFE OF MANGO FRUITS

By

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A56/70389/2013

A thesis submitted in partial fulfillment of the requirement for the award of a Master of Science degree in Horticulture of the University of Nairobi

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DECLARATION

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DEDICATION

With love, gratitude and appreciation, I dedicate this work to my mum Dr. Lydia Marundu, my brothers Stanley and Edward, and sisters Monicah and Evelyn for their prayers, always being very supportive and encouraging during the entire period.

ACKNOWLEDGEMENT

To the Most High God, I honor and glorify His name for the knowledge, blessings and grace He has bestowed upon me throughout my education up to this level.

I sincerely express gratitude to my supervisor, Dr. Jane Ambuko, for her mentorship and tremendous support throughout this work. I am also grateful to my other supervisors Prof. Margaret Hutchinson and Dr. Willis Owino for support and guidance. My great appreciation to the University of Nairobi for the scholarship award that facilitated my study. I wish to thank the United States Agency for International Development (USAID) for financing the research work through a grant (No. 13-KFI-C-G-0918) awarded to Dr. Jane Ambuko under the Kenya Feed the Future Innovation Engine (KFIE). I have sincere appreciation to Jomo Kenyatta University of Agriculture and Technology administration, for allowing me use their facilities to conduct my laboratory work. Specifically, I acknowledge Mr. Karanja, Mr. Votha, Mr. David and Madam Jessica for their invaluable support and technical guidance.

Special thanks to the Kawala small- scale horticultural farmers through their chairlady madam Teresia Benjamin who allowed me work in their premises and Alice Ndinda for the support she accorded me during the study.

My very special gratitude to my mum and siblings for the sacrifice and support you gave that has enabled me get to this point. Dear friends (Mable, Jacinta and Annah), God bless you for being an encouragement and inspiration.

Lastly, I would like to extend my appreciation to all who contributed in diverse ways to the completion of this study.

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LIST OF ABBREVIATIONS AND ACRONYMS

ACP Agriculture and Consumer Protection

AVG Aminoethoxyvinylglycine

STS Silver thiosulphate

CAS Controlled Atmosphere Storage

CCM Cold Chain Management

FAO Food and Agriculture Organization

FPEAK Fresh Product Exporters Association of Kenya

GCCA Global Cold Chain Alliance

HCDA Horticultural Crops Development Authority

HLPE High Level Panel of Experts

IIRR International Institute for Rural Reconstruction

ITA International Trade Administration

KAA Kenya Airport Authority

KARI Kenya Research Agricultural Institute

KFIE Kenya Feed the Future Innovation Engine

KMUTT King Mongkut's University of Technology Thonburi

MAP Modified Atmosphere Packaging

1-MCP 1-Methylcyclopropane

RSA Research Solutions Africa

USAID United States Agency for International Development

WFLO World Food Logistics Organization

ZEBC Zero Energy Brick Cooler

ABSTRACT

Poor cold chain management is one of the main causes of postharvest losses (>50%) reported in most vegetable and fruit value chains. Therefore maintenance of low temperature for horticultural commodities is critical in postharvest handling of these perishable crops. However, conventional cold rooms are expensive and unaffordable for majority of the horticultural smallholder farmers. This has necessitated research in alternative low-cost storage systems. The CoolbotTM technology is one such technology that has been used effectively in other countries and various commodities. Transfer of the technology to Kenya requires extensive research to validate its efficacy under local conditions. The present study was conducted to establish efficacy of CoolbotTM technology to lower and maintain cold temperatures during storage of mango fruits. The study also sought to evaluate the synergistic effect of cold storage and modified atmosphere packaging (MAP) using Activebag® in preserving quality and extending shelf life of mango fruits. The study site was Makueni County and it was conducted over 2 seasons, between 2014 and 2015. A Coolbot™ cold room was constructed from locally available materials including structural insulated panels made of polystyrene to provide insulation; an LG air conditioner (24,000 BTU) and the CoolbotTM. Temperature probes were strategically positioned in the cold room and ambient room to monitor temperature changes during the storage period. Temperature changes in the cold room were monitored after every one hour until the preset temperature (10±1°C) was attained. The CoolbotTM cooling efficacy studies were conducted using 'Apple' mango variety harvested at physiological maturity stage and stored in the cold room and ambient room... Three fruits were sampled randomly from the two storage conditions at regular intervals to compare progression in ripening based on changes in respiration rate, peel/flesh color and firmness. A replicate experiment was conducted using 'Apple' and 'Ngowe' mango varieties. The fruits were then either packed in Active bags or left unpacked. Random sampling of three fruits was conducted after every three days to measure ripening progression and change in quality attributes. Parameters measured include respiration rate, ethylene evolution, cumulative weight loss, peel/flesh color and firmness, total soluble solids, total titratable acidity, soluble sugars (fructose, glucose and sucrose), Vitamin C and β-carotene. Results show that CoolbotTM effectively lowered and maintained temperature at 10±1°C throughout the storage period whereas the ambient room temperature fluctuated between 25 – 28°C. Cold-stored fruits had an extended shelf life of 35 days (compared to 12 days for fruits in ambient room) as evidenced by slower ripening-related changes. Apple' mangoes in the ambient room reached climacteric peak (53.9 ml/Kg/Hr) earlier on day 12 compared to the delayed one observed on day 35 for cold stored fruits., the synergistic effect of cold storage and MAP in preservation of quality and extension of shelf life of 'Apple' and 'Ngowe' mango varieties was demonstrated. In 'Apple' mango, the 35 days shelf life under cold storage was extended to 40 days under cold storage+MAP. Respiration, ethylene evolution, color changes and softening rate were all significantly reduced under cold storage, with or without MAP. Changes in all the fruit quality attributes including vitamin C, beta-carotene and sugars were significantly slowed down under cold storage. Cold-stored 'Apple' mangoes that were packed in Active bag® retained higher Vitamin C content (59.77 mg/100ml) at the end of storage period compared to unpacked fruits (51.8 mg/100ml). Overall, cold storage and MAP slowed down ripening and senescence as shown in the results. The findings confirm efficacy of the CoolbotTM technology to lower and maintain low temperatures in an insulated storage room thereby preserving quality and extending shelf life of mango fruits. The technology can therefore be promoted as a low-cost alternative cold storage option for adoption by smallholder mango farmers.

Key words: CoolbotTM, cold storage, cold chain, postharvest losses, MAP

CHAPTER 1

1.0. INTRODUCTION

1.1. BACKGROUND INFORMATION

The horticultural sub-sector is the fastest growing in agriculture tallying closely behind tourism and tea, with a growth of 15% to 20% per annum in Kenya (FPEAK, 2015). Within the subsector, flower production has contributed to about 50% while fruits and vegetables take up the other 50% of the total earnings from horticulture (HCDA, 2012).

The sub-sector faces several challenges, themajor one being high post-harvest losses. The major contributor to the post harvest losses is temperature (Vorster *et al.*, 1990). It is estimated that the rate of deterioration of perishables increases two to three-fold with every 10°C increase in temperature (Kader, 2005). Cold chain management is therefore key in reduction of the experienced high post harvest losses.

Cold chain management must start at harvest taking into consideration time of day when the crops are harvested, initial shading before transportation to farm store or removal of field heat prior to cold storage or refrigerated transport (Kathryn and James, 2004). Temperature abuse right from the farm predisposes the produce to faster deterioration in subsequent postharvest handling stages (Isaac *et al.*, 2015). Among the smallholder farmers, temperature abuse is attributed to various factors. One of the factors is lack of knowledge of on good harvest and postharvest handling practices that contribute to cold chain management. Cold chain abuse could also be attributed to the high cost of cold storage facilities such as conventional cold rooms and refrigerated trucks for post-harvest handling of highly perishable horticultural produce (FAO, 2011).

In Kenya, most of the cold chain facilities are owned by privately owned by exporting firms while for domestic market, a few small firms own poorly designed cold rooms which

lead to breaks in the cold chain resulting to product damage (ITA, 2016). The privately owned cold storage facilities are mainly concentrated within Nairobi with an estimated capacity of 1790 tons per day (Global hort., 2010). In the public sector, HCDA owns 8 cold storage facilities with a capacity of 205 tons per day while KAA has 2 cold storage facilities with a capacity of 510 tons per day.

Besides the challenges at production and those associated with post-harvest handling of highly perishable horticultural commodities, market access is a problem for the majority of small holder farmers. Traceability of produce and stringent market standards including CODEX and Sanitary & Phytosanitary standards (Ami and Green, 2006; Robach, 2011) are some of the factors that hinder small holder farmers from accessing export market due to lack of technologies and resources needed for compliance (Das, 2008). Maximum Residue Levels on produce has also led to barring a majority of smallholder farmers from accessing the EU market since the pesticides used leave more than acceptable amounts of residue on produce.

1.2. PROBLEM STATEMENT

One of the key challenges facing horticultural famers in Kenya is high post-harvest losses estimated to be 40 – 50% (KARI, 2012). The losses in the mango value chain are estimated to be 40% (Adeniji, 2010). Poor cold chain management is one of the factors contributing to the losses among the small-holder farmers (Kitinoja, 2002). Temperature plays a central role in post-harvest handling of perishables since it affects most of the enzymatic reactions in harvested produce causing an increase in the rate of deteriorative processes (e.g. microbial growth, respiration, water loss and growth and development) by 2-3 fold for every 10°C rise in temperature (KMUTT, 2007).

Cooling horticultural products immediately after harvest and maintaining the cold chain along the supply chain is key in slowing down the deteriorative processes and thus extend their shelf life (Kitinoja, 2013). The cost of conventional cold rooms used by

commercial operators is out of reach for small holder farmers (HCDA, 2009). As a result, smallholder farmers growing perishable commodities such as mango are forced to sell them immediately they are ready for harvesting. This exposes them to exploitation by middlemen who buy the fruits at very low prices ranging from Kshs. 3-5 (Ambuko, 2016). This situation calls for alternative cold storage options which are appropriate and affordable for smallholder farmers. One such technology is the CoolbotTM which is an innovative technology that has been used successfully in other countries as a cold storage alternative.

The Coolbot[™] technology has been used in Uganda to extend shelf life of onions (Saran *et al.*, 2012), in Honduras for cooling flowers and enhancing their vase life and in India to cool and reduce rate of deterioration in horticultural commodities like potatoes, tomatoes, turnips and beans (Kitinoja, 2011). The Coolbot[™] cold room can be modified based on the locally available materials. To introduce and promote this technology for the benefit of smallholder farmers in Kenya, there is need to adapt it (build from locally available materials) and test it under local conditions.

1.3. JUSTIFICATION

Cold chain management is important in slowing down the deterioration processes that take place in harvested commodities (Senthilkumar, 2015) hence extending shelf life and the marketing period (Borompichaichartkul *et al.*, 2009). With affordable low cost cold storage facilities, small holder farmers will not be forced to sell their produce immediately after harvest during peak season and can bulk their produce which will increase their bargaining power hence getting better prices. The CoolBotTM system is a viable option for small holder farmers who can't afford the expensive conventional cold rooms since it's cheaper. It also saves up to 60% of the electricity bill hence small holder farmers organized in a group can benefit from it (Dubey, 2016). The technology has not been tested in Kenya to evaluate its efficacy to lower storage temperature and extend the shelf life of perishable commodities such as mango. This study is therefore critical to rolling out the CoolbotTM technology in Kenya. The findings of the study will be used as factual evidence in promotion of the technology among smallholder farmers, traders and other stakeholders in the horticultural sub-sector.

1.4. OBJECTIVES

Overall objective

To investigate the efficacy of a new innovative low cost low-cost cold storage technology, CoolbotTM, to extend shelf life and preserve quality of mango fruits and their response to a combination of cold storage and modified atmosphere packaging.

Specific objectives

- To evaluate the efficacy of the Coolbot[™] to lower and maintain cold temperatures during storage of mango fruits.
- 2. To evaluate the effect of the CoolBotTM and modified atmosphere packaging to preserve quality and extend shelf life of mango fruits.

HYPOTHESES

- The Coolbot[™] gadget will not lower and maintain cold temperatures in an insulated room during storage of mango fruits.
- 2. A combination of CoolBotTM cold storage and modified atmosphere packaging has no effect on quality and shelf life of mango fruits.

CHAPTER 2

2.0. LITERATURE REVIEW

2.1. AN OVERVIEW OF THE CHALLENGES FACED IN HORTICULTURE SUB-SECTOR IN KENYA

Despite the growth experienced over years in horticulture sub-sector, it has faced challenges that have caused dwindling in production (Figure 1.1) (Mutiso, 2014). In 2008 the turnover in quantity exported was 423.129 MT (valued at 73.737 B) which dropped to 360.474MT (valued at 71.597B) in 2009. In 2010, produce exported rose to 403.026 MT (valued at 114.59B) though it dropped in 2011 to 382.638 MT. Another drop was experienced in 2012 in value (87.713776B) without a significant drop in quantity (from 380.84MT in 2011 valued at 91.229703B to 380.42 MT in 2012). A 30% increase in volume and 8.8% increase in value were experienced in 2013 in comparison to 2012 and there is still great potential for improvement in the coming years (HCDA, 2013). The majority of small holder famers sell their produce to the local market. 95% of the produced horticultural commodities are sold at the domestic market with only 5% getting to the export market (RSA, 2015).

Horticultural exports are among the top foreign exchange earners for the country (National Horticulture Policy, 2012). However, the earnings can be increased if the total potential within the sector is fully exploited. The unexploited potential is due to the fact that the sector is dominated by small scale farmers who are not able to overcome the challenges that are facing the industry. The smallholder farmers are faced with challenges at each stage of the supply chain; from input supply to marketing and consumption.

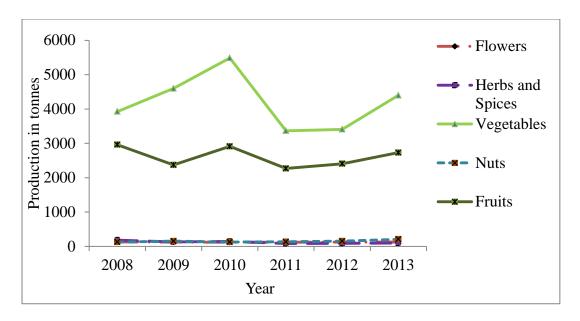


Figure 2. 1. Production trend of various horticultural commodities in Kenya from 2008-2013 SOURCE: Mutiso, 2014

During production, the high cost of inputs like quality planting materials, fertilizers and pesticides has discouraged small holder farmers from using them. This has resulted to slow crop growth and low yields of poor quality (KARI, 2012). After harvest, the key challenges that small holder farmers face are attributed to high perishability of horticultural commodities (due to inadequate efficient storage facilities like cold rooms at the market place (for domestic market) and the stringent market standards (for export market) (Atanda, 2011).

2.2. POST-HARVEST LOSSES IN PERISHABLE HORTICULTURAL COMMODITIES

Post-harvest losses is one of the greatest challenge experienced by small holder farmers. According to the latest reports, it is estimated that 1.3 billion tons (or 30%) of the food produced for human consumption is lost or wasted along the food supply chain (FAO, 2011). High losses are experienced by small holder horticultural farmers due to high perishability of their commodities. This is evident since of the volume reported by FAO, 44% of the losses are in fruits and vegetables only. Sometimes the losses in fruits and vegetables

can be as high as 100% depending on the commodity, season and region amongst other factors (Phiri, 2010). The losses cited in these reports are quantitative and therefore easily measurable. However, significant qualitative losses occur in fruits and vegetables after harvest (FAO, 2014).

Globally, 50% of fruits vegetables and root crops are lost in a year (Kiaya, 2014) while about 55% of fruits and vegetables are lost p.a. in developing countries where postharvest handling technologies are limited (Jaspreet et al., 2014). In Africa, 30-40% of the harvested perishable commodities are lost per annum. The range increases in Sub-Saharan Africa where 30-80% of the harvested horticultural produce is lost annually (Kitinoja, 2013). In Kenya, the losses in horticultural products are estimated to be between 20-50% (HCDA, 2012) though little emphasis has been placed on postharvest losses which present a knowledge gap for scientific research. AVRDC estimates losses in vegetables to be between 9-25% due to inadequate facilities for postharvest handling (Weinberg et al., 2008) while in Kenya the vegetable losses are at 50% (Aseno-Okyere, 2012). Post harvest losses in bananas are estimated to be up to 50% (Adeniji et al., 2010) and between 18-45% (Kitinoja and Cantwell, 2010) which is attributed to storing them in unfavorable conditions and not handling them carefully (Mashau et al., 2012). Mango post harvest losses have been on the increase especially during post harvest handling. KARI recorded 40-45% losses during the post-harvest handling stage along the value chain (KARI, 2012). According to Gathambiri et al. (2009) mango losses are more than 45% of the total production whose occurrence is attributed to poor postharvest handling and oversupply during the peak season.

2.2. FACTORS CAUSING HIGH POST HARVEST LOSSES

To reduce the high postharvest losses reported in fruits and vegetables, proper handling and reduction of the time between harvest and the retail or consumption stage is critical. This can be done by obtaining adequate knowledge of factors that hasten deterioration rate and how they can be mitigated (Kitinoja, 2013). There are various factors, in all stages of the value chain that affect keeping quality and shelf life of harvested produce (Jobling, 2002). These factors can be broadly classified as: pre-harvest factors, harvest factors and post-harvest factors.

2.2.1. Pre-harvest factors

Pre-harvest production practices while the crop is still growing have implications on its keeping quality after harvest (Isaac *et al.*, 2015).

2.2.1.1. Mineral nutrients and water supply

A crop provided with optimal nutrients according to its requirements throughout the growing season will give a produce that show optimal quality throughout the value chain after harvest. Excess or deficiency in essential mineral nutrient is detrimental and affects quality of fruits (Kader, 2002). Nutrient and water oversupply may lead to cracking of fruits like tomatoes and bananas creating entry point for disease causing microorganisms (Goletti, 2003). In mango fruits excess nitrogen application causes the fruits to remain abnormally green even during ripening. Imbalance of calcium and potash leads to fruit disorders like jelly seed (Cracknell *et al.*, 2004). Jelly seed is a physiological disorder in mangoes where the pulp (mesocarp) near the seed breaks down becoming mushy and off flavored. In severe cases, the tissue forms hollow cavities and is discolored. This disorder can lead to 50% loss of the harvested produce (Oosthyuse, 2003). The most susceptible mango variety to jelly seed disorder is "Van Dyke" (Cracknell *et al.*, 2004).

2.2.1.2. Choice of cultivar

Choice of cultivar affects the shelf life of the harvested produce. According to Yashoda *et al.* (2006) textural softness of mango fruits vary from one variety to another and affects shelf life. A study by Ouma (2015) showed that 'Apple' mango fruits had a higher firmness (both peel and flesh firmness) with extended shelf life compared to Ngowe mango

fruits. Rate of respiration during ripening also affects post harvest quality and shelf life (Razzaq *et al.*, 2013). Ouma (2015) showed that Apple mango fruits had lower rate of respiration consequently extending shelf life compared to Ngowe mango fruits that exhibited high respiration rate during ripening.

2.2.1.3. Agro-ecological conditions

Environmental conditions during production affect shelf life of harvested horticultural produce (Hewett *et al.*, 2006). They not only affect fruit growth and development by altering accumulation of water, dry matter and biochemical compounds but also fruit behavior during storage (Lechaudel and Joas, 2006). A study on the effect of agro-ecological zone on post harvest quality and shelf life of mango fruits showed that fruits produced in a low potential area characterized by low rainfall and high light intensity had longer shelf life compared to fruits produced in high potential area where with high rainfall and low light intensity (Kemunto, 2013).

2.2.2. Harvest factors

2.2.2.1. Harvest maturity

Maturity at harvest affects longevity and quality of a commodity (Shaiq *et al.*, 2014). Harvest of immature or over ripe fruits is one of the factors that contribute to high post harvest losses (Ingle *et al.*, 2000). Fruits that are left to ripen on the tree usually have shorter shelf life as compared to those that are harvested when partially ripe but mature (Dargie *et al.*, 2013 and Moneruzzaman *et al.*, 2008). The short shelf life is attributed to high enzymatic and physiological activities (e.g. ethylene evolution and respiration rate) in the advanced maturity fruits (Arumugam and Vadivel, 2013). However they have better eating quality in comparison to those harvested at physiological maturity stage as shown in a study on mangoes (Ouma, 2015). Often, small holder farmers are forced to harvest immature fruits and sell them for fear of their produce being stolen while in the farm or to attend to an

emergency. Such fruits don't develop good eating quality once they are ripe and are susceptible to damages and high rate of water loss which leads to shriveling and loss of weight (Kader, 2008). Most traders prefer produce at physiological maturity stage so that it can withstand post harvest handling during long distance shipping but this has an effect on quality hence the need to strike a balance between shelf life and quality of produce (Kadzere *et al.*, 2006; Sivakumar *et al.*, 2011).

2.2.2.2. Time of harvest

Time of the day of harvesting horticultural produce significantly affects the amount of heat absorbed by the produce which in turn affects its shelf life since temperature enhances the deteriorative processes (Chopra *et al.*, 2003) and the increases the amount of effort required in lowering product temperature (Kader, 2002). Harvesting fruits early in the day when there is low field heat ensures the fruit has low internal fruit temperature hence requiring little amount of time and energy to cool it to the desired safe optimum storage temperature (Samtani and Kushad, 2015; Kader and Rolle, 2004). The fruit's low internal temperature translates to reduced enzymatic degradation and slower rates of physiological processes such as ethylene evolution and respiration rate consequently extending shelf life (Borompichaichartkul *et al.*, 2009). In mango fruits harvesting early in the morning enhances latex retention which promotes peel firmness and reduces susceptibility to anthracnose (Karunanayake *et al.*, 2014).

2.2.2.3. Harvesting method

Harvesting method determines the extent of physical injuries induced on the fruits and vegetables which become entry points for pathogens, increase water loss of the produce and accelerate the rate of ethylene evolution all of which are deteriorative processes that contribute to the short shelf life (Kader, 2002). The two main harvesting methods are hand picking and mechanized harvesting. Mechanical injuries break the skin which is a protective

layer of the fruits which causes increase in normal physiological changes hence leading to depletion of food and water reserve in the fruit. The broken skins are wounds on the fruits which act as entry point for microorganisms. This accelerates rate of pathogen establishment on the fruits which promotes rotting and reduced shelf life (Bachmann and Earles, 2000).

Injuries also increase the fruit's rate of transpiration, promoting loss of water and Vitamin C which is highly volatile (Kader, 2002). In fruits and vegetables, multiple handling during harvesting increases damages (entry point for microorganisms, increases water loss and high rates of ethylene evolution) on the produce reducing their shelf life (FAO, 2013).

2.2.3. Post harvest factors

There are various post harvest handling practices that affect post harvest longevity of horticultural product and can either increase or reduce losses (FAO, 2014). The postharvest component can be divided into the following handling stages.

2.2.3.1. Grading and sorting

Grading and sorting is conducted after harvesting, where injured/ bruised, diseased and blemished fruits are separated to promote extended shelf life for the unaffected ones (FAO, 2011). The good quality fruits are then batched based on traits like size, maturity stage and ripeness (Asghar et al., 2012). Injured fruits and ripening ones have increased enzymatic activities and physiological processes like ethylene evolution and respiration rate hence shorter shelf life and ethylene produced can cause ripening of all the fruits in the storage area (Miller, 2003). Diseased fruits may spread the inoculums to uninfected ones reducing the whole batch's shelf life (Nafussi *et al.*, 2001).

2.2.3.2. Sanitization

This involves cleaning the produce with treated water to disinfect and remove microorganisms that would otherwise contribute to pathological deterioration (Kader and Rolle, 2004). Most farmers don't clean their produce after harvest due to lack of knowledge

on the importance of such a practice and its implication on post-harvest losses (NRI, 2014). Some of the sanitation practices used in fruits includes dipping the fruits in hot water 50°C for 20 minutes to avoid anthracnose (Mirshekari *et al.*, 2012). Sanitizing agents such as chlorine and acetic acid have also been used successfully to disinfect mango fruits (Ouma, 2015). Addition of post-harvest fungicides such as imazalil and thiobendazole has been used effectively to disinfect citrus fruits. Since microorganisms play a role in post-harvest losses by enhancing deteriorative rate, their elimination promotes extended shelf life (Kader and Rolle, 2004).

2.2.3.3. Packing and packaging

Packing and packaging are important tools for minimizing losses in perishable horticultural commodities by extending their shelf life (FAO, 2011). They promote both air flow within the storage area which helps during the cooling process to avoid heat buildup and ease of produce handling (Kader and Rolle, 2004). Fibre board and plastic containers are the commonly used packaging containers with packaging accessories like wraps and cushioning pads to immobilize the produce (Kader and Rolle, 2004) and it shields the produce from mechanical damages i.e. cuts and bruises due to abrasion during transporting (Karacay and Ayhan, 2009). Packaging accessories like wraps also provides a vapor transmission barrier reducing rate of water loss which if not countered, may result to shriveled produce not appealing to consumers and lower shelf life (Diaz-Mula *et al.*, 2011) while providing convenient units for marketing. Use of poor quality packaging material may lead to high post harvest losses (FAO, 2011).

2.2.3.4. Transportation

Transport of perishable horticultural commodities from the production site to the consumer has an aspect of time and may cause great losses if there are delays at any stage along the supply chain (FAO, 2014). Since nutritional qualities of commodities diminish with

nutrients hence not beneficial to the consumer (Idah *et al.*, 2007). Vehicles used to transport horticultural commodities are also placed under other uses and this introduces foreign organisms like insects and microorganisms which feed on the produce or cause diseases respectively hence reducing the commodity's shelf life (FAO, 2013). Most of these vehicles are unrefrigerated so the high temperatures inside cause faster deterioration of produce being transported. The seasonal rough roads are impassable especially during the rainy seasons and this has also led to great losses in horticultural produce. Mostly the vehicles get stuck, delivering the produce after a delayed period of time (Rolle, 2006). During loading and unloading, mechanical injuries are made on the fruits by the loaders that sometimes step and sit on them as they arrange (Miller, 2003) and are aggravated by poor packing for transport where produce is just dumped directly into the trucks (FAO, 2014). Mixing different produce into the same load during transportation affects shelf life especially where the mixture contains high ethylene producing (e.g. passion fruits) and ethylene sensitive commodities like mangoes (Kader and Rolle, 2004).

2.3. POSTHARVEST MANAGEMENT TECHNOLOGIES FOR PERISHABLE COMMODITIES

2.3.1. 1-Methylcyclopropene (1-MCP)

1-MCP is a gas (at atmospheric temperature and pressure) that has a similar structural formula as that aminocyclopropanecarboxylate oxidase hence its ability to compete successfully for binding sites to inhibit the deteriorative effects of ethylene (Hofman *et al.*, 2001). It acts by preventing ethylene from binding by occupying ethylene receptors since it has ten times greater affinity for the receptors than that of ethylene (Sisler and Serek, 1997). It also inhibits expression of 1-aminocyclopropane-1-carboxylic acid (ACC) and aminocyclopropanecarboxylate oxidase (ACO) which are important enzymes in ethylene

biosynthesis (Blankenship and Dole, 2003). The compound, in comparison to other possible solutions to ethylene like Silver thiosulphate (STS) and aminoethoxyvinylglycine (AVG), has been found to be more effective in ethylene management and non- toxic (US Environmental Protection Agency). Silver thiosulphate is harmful to the environment due to the silver element which is a potent pollutant (Abdi *et al.*, 1998). 1-MCP has successfully been applied to delay ripening in apples (Fan *et al.*, 1999), avocado (Feng *et al.*, 2000), bananas (Harris *et al.*, 2000), and mangoes (Githiga *et al.*, 2012; Kemunto *et al.*, 2013).

2.3.2. Waxing

This is a practice that is carried out on most horticultural commodities like citrus fruits, cucumbers and apples to reduce rate of water loss and maintain the commodity's turgor (Atanda, 2011). Water loss causes shriveling and loss of saleable weight which are signs of deterioration (Banks *et al.*, 1997). Weight loss is known to increase the rate of ripening and susceptibility to diseases (Joyce *et al.*, 1995). Therefore, reducing rate of water loss will extend the commodity's shelf life. Waxing also enhances the appearance of a commodity by giving it a gloss and making it shinier than the non-waxed commodities (Atanda, 2011). The technique has been successfully applied to reduce water loss and extend shelf life in avocado (Banks *et al.*, 1997), mango (Baldwin *et al.*, 1999), melons (Cong *et al.*, 2007) and tangerine (Hassan *et al.*, 2014).

2.3.3. Modified Atmosphere Packaging

MAP is a technology that aims at altering the storage atmospheric conditions of a produce by use of a polymeric film. The major alterations are the gaseous composition and amount of water vapor in the package which slows down deteriorative processes that take place in harvested horticultural commodities (Sandhya, 2010). Altered gaseous composition of O₂ and CO₂ retards the rate of respiration, inhibit biosynthesis of ethylene and degradation of chlorophyll (Valero and Serrano, 2010). Low O₂ levels promote slow rate of the fruit peel

color change from green to yellow/ purple (depending on the variety) due to reduced rate of chlorophyll degradation and delay in biosynthesis of colour pigments like anthocyanin and carotenoids (Artes *et al.*, 2006). Water loss, as is the case without packaging, causes shriveling of fruits. MAP is a technology that mitigates water loss due to high water vapor in the bag (Siddiqui and Dhua, 2009). MAP has extended tomatoes' shelf life up to 21 days since they remain turgid longer and colour change was observed to be slower, though off flavors developed in films that had low gas permeability since there was accumulation of high levels of CO₂ (Majidi *et al.*, 2014). Shelf life was extended in apple (Moodley *et al.*, 2002) while in strawberry, packaging reduced weight loss and fruit firmness (attributed to reduced water loss) resulting to good keeping quality (Jouki and Kazaei, 2012). In mango fruits though it has not been commercialized as to be used during shipping of the fruits. However, Yuen *et al.* (1993) showed poor chlorophyll degradation in packaged fruits with retention of green colour which affected marketability since it is not appealing to the consumers.

2.4. COLD CHAIN MANAGEMENT IN PERISHABLE COMMODITIES FOR REDUCED POST-HARVEST LOSSES

At harvest, horticultural commodities are alive and continue with the metabolic processes such as respiration and transpiration. For fruits which are harvested at physiological maturity, ripening proceeds after harvest. These processes require water, nutrients and energy reserves all of which are cut off at harvest (Alfred and Paul, 2013).

Other deteriorative processes in harvested produce like ethylene evolution also hastens the rate of ripening and softening hence the fruits become unappealing to consumers. Colour changes occur as fruits are de-greened during synthesis of carotenoids to give the fruit a consumer appealing colour. Water loss from the produce causes them to shrivel (fruits) or wilt (vegetables) after the cells have lost their turgidity (Deirdre, 2015).

Central to these deteriorative processes is temperature (Vorster *et al.*, 1990). It is estimated that the rate of deterioration of perishables increases two to three-fold with every 10°C increase in temperature (Kader, 2005) as shown in the table below:

Table 2. 1. Effect of temperature on shelf life of fresh foods

Fresh product	Storage potential			
Fresh green	1 month at 0°C	2 weeks at 10°C	1 week at 10°C	< 2 days at 30°C
vegetables				
Potatoes	5-10 months at	< 2 months at	< 1 month at	< 2 weeks at
	4-12°C	22°C	32°C	42°C
Mangoes	2-3 weeks at	1 week at 23°C	4 days at 33°C	2 days at 43°C
	13°C			
Apples	3-6 months at	2 months at	1 month at 20°C	A few weeks at
	1°C	10°C		30°C

Source: Kitinoja, 2013.

$Q_{10}^* =$ Rate of deterioration at temperature T + 10 °C

Rate of deterioration at temperature T

A study conducted on sweet cherries that were stored at different temperatures (0°C, 5°C, 10°C and 20°C) and their respiration rates taken showed that respiration rate increased with an increase in temperature (Carlos *et al.*, 1993). A study on asparagus and peach by Jobling (2008) gave similar results as shown in the figure below.

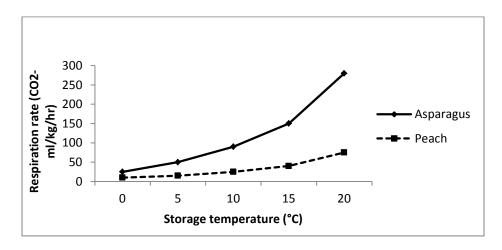


Figure 2. 2. The relationship between respiration rate and temperature rise during storage of peaches and asparagus.

Source: Jobling, 2008.

Therefore, controlling product temperature and reducing the amount of time that a product is at sub-optimal temperature is key to maintaining the quality, improving shelf life and extending marketing period and ultimately reducing postharvest losses (Kassim *et al.*, 2013).

2.4.1. Temperature control in harvested horticultural produce

Temperature abuse is one of the major causes of the very high losses experienced in horticultural production (FAO, 2014). It starts right from harvesting to when the product is being marketed and reaching the ultimate consumer (Nunes *et al.*, 2003). The rate of deterioration has been estimated to increase by two or threefold with every 10°C increase in temperature within the optimum range for the commodity in question and so temperature abuse is critical and detrimental to all horticultural products (Kader and Rolle, 2004). Its detrimental effects on the shelf life of horticultural products is attributed to increased water loss, respiration rate, softening, ethylene evolution and other physiological processes which increase rate of deterioration of the fruits (Chopra *et al.*, 2003; Alfred *et al.*, 2013).

Post-harvest cooling and maintaining a cold chain is thus the most effective way to curb effects of high temperature on perishable horticultural commodities and by maintaining low temperatures conducive for a certain commodity, increase shelf life will be realized (Bachmann, 2000).

Table 2. 2. Theoretical relationship between temperature, respiration rate and deterioration rate of a non-chilling sensitive fresh commodity.

Temperature °C	Assumed Q10	Relative velocity	Relative shelf	Loss per day
		of deterioration	life	(%)
0	-	1.0	100	1
10	3.0	3.0	33	3
20	2.5	7.5	13	8
30	2.0	15.0	7	14
40	1.5	22.5	4	25

Developed from data available in USDA Handbook 66 (Kitinoja, 2013)

In developed countries, temperature management is characterized by sophisticated management system and infrastructure (Hodges *et al.*, 2010). They have continued to improve on the efficiency of their cold chain technologies which explains the low postharvest losses of 5-25% (Opiyo, 2012). Logistics companies and suppliers have invested in cold chains by in pursuit of accessing local markets and opening temperature controlled operations which have helped in strengthening cold chains and prolonging the product's shelf life (Rolle, 2006). They also complement their cold storage with other post-harvest technologies like MAP and waxing hence extending the shelf life of their produce, consequently getting longer marketing period (FAO, 2014).

Developing countries' cold chain system is characterized by poor storage facilities (FAO, 2011) poorly developed and inefficient infrastructure which has caused the highest amount of deterioration in harvested produce over the years (Kitinoja, 2010). Most small scale farmers are operating below the poverty line and are unable to obtain the highly sophisticated facilities to maintain a cold chain at the farm level. A study conducted in Latin America and Caribbean countries showed poor cold chain management to be the main reason for post-harvest losses (FAO., 2015). 2.4.2. Cold Chain Management

A cold chain refers to an uninterrupted series of activities from the production point to the consumer which maintain a given temperature range (Ilic and Vukosavljevic, 2012). Any hiccup along the cold chain may reduce its efficiency and lower the produce's shelf life unlike where the cold chain is maintained fully along the supply chain. A research conducted on importance of cold chain showed that fruit quality is severely reduced by breaking the cold chain even for a short duration (Blakey *et al.*, 2011).

Knowing the damaging effects of high temperature on horticultural commodities necessitates clear and efficient cold chains at all stages of the supply chain so as to impede the challenge of post-harvest losses due to temperature abuse. The other component is

cooling during holding which occurs after pre-cooling, during other activities like sorting and grading, packaging, transportation and even during marketing the produce, optimal temperature needs to be maintained (Ginsberg, 1985).

Maintenance of cold chain is therefore very necessary so that we can preserve the commodities at their best possible quality (Kader, 2005). However, there are hitches that cause farmers and traders not to maintain the cold chain like high cost of purchase, installation and operation of the requisite infrastructure. Most small scale farmers do not have on-farm cold storage facilities or even temporary shades for their produce (Kitinoja *et al.*, 2003).

Cooling horticultural commodities has been shown to reduce respiration rate, transpiration rate, ethylene evolution to slow ripening, decrease activity of microorganisms and reduce changes like browning, loss of texture, flavor and nutrients. All this aims at delaying senescence and lengthening the product's shelf life (Kitinoja, 2013). Generally, cold chain management allows for maintenance of physical and biochemical properties of produce which are desirable to consumers. This is nutritionally important since the nutrients in the produce will be maintained at high levels if they are not exposed to high temperatures.

Cold chain management also helps in regulating the market prices of products especially during peak season when there is so much of the specific product in the market such that it causes market glut. With a well-managed cold chain, farmers are able to store some of their produce for extended periods of time and avoid selling them at very low prices which is necessitated by fear of losing the produce due to poor storage facilities (Umali-Deininger and Sur, 2006). They then can sell the products when the supply is low and the market prices are relatively high. Consequently, both farmers and traders will benefit by selling the products at reasonable prices and since they were well stored, they will be of good quality hence having a better value of money (IBRD, 2011). It is also important since it helps

farmers, traders and manufactures avoid the high wastage that occurs when there is poor maintenance of the cold chain. Avoiding losses in turn results to increased income and thus better livelihoods for all involved (IBRD, 2011). Generally, low temperature maintenance (optimum for the specific commodity) at all stages along the supply chain will result in great reduction of losses and the produce's quality will be maintained at high levels which is the goal of anyone dealing with horticultural products.

The consumers will also benefit by having the products readily available at all times even during the off season period for such commodities hence giving them fresh and nutritious fruits and vegetables among other commodities that can be stored therein (Mazibuko and Oladele, 2012).2.4.2.1. Post-harvest handling practices that affect cold chain

Use of cold chain has been in use since the 1950s with aim of prolonging shelf life of horticultural commodities but it has not been fully adopted in most of the developing countries, Kenya being one of them (Kitinoja, 2013). Some of the practices that farmers in developing countries can adopt to ensure their produce has reduced chances of succumbing to the high temperatures in the surrounding include the following:

2.4.2.1.1. Timely harvesting

Time of harvest affects the amount of field heat that accumulates in the produce. Harvesting during the hot hours of the day causes the produce to accumulate a lot of heat in it. The more the heat a produce picks from its surroundings, the more energy and time it requires to cool it down to manageable temperatures (Cantwell, 1998). Harvesting should be done during the cool hours of the morning when the temperatures are still low or in the evening.

2.4.2.1.2. Temporary shades after harvest

Temporary shades are important in protecting harvested produce from exposure to direct sunlight thus preventing its warming up and heat accumulation (Kitinoja, 2013).

Exposure of perishable horticultural produce to direct sunlight reduces quality and shelf life (Ahmad and Siddiqui, 2016). Most of the small scale farmers don't have temporary shades for holding their harvested producing before transportation to the store/market takes place. This leads to heat build-up in the produce, thus promoting high respiration, weight loss and other deteriorative processes resulting to shorter shelf life (Saran *et al.*, 2012).

2.4.2.1.3. Pre-cooling of harvested produce

For successful storage of any horticultural commodity, pre-cooling has to be done immediately after harvest which is one of the components of a cold chain system (IIR, 2009). Its importance is to remove field heat from the produce before other handling operations are conducted so as to relieve the product of heat that could lead to faster deterioration if it stays longer in the commodity (Cantwell, 1998). The longer the commodity stays heated, the higher the rate of deterioration hence shorter shelf life (Kader, 2002). Pre-cooling enhances shelf life by suppressing enzymatic degradation, reducing respiration rate and ethylene evolution, slowing down water loss and inhibiting growth of disease causing microorganisms on the produce (Mohammed and Brecht, 2014). Produce that has not been pre-cooled immediately after harvest, deteriorates faster even after being stored at low optimum temperature. Pre-cooling is also essential in reducing the load on the cold storage facility hence little amount of energy will be required to cool the produce to required temperatures (KMUTT, 2007). Pre-cooling can be done by sprinkling water on the harvested commodities if they are not sensitive to water e.g. fruits like mangoes, apples and oranges (hydro-cooling), forced air cooling where cold air is pushed through produce containers so as to take away the heat and by room cooling.

2.4.2.1.4. Transportation

Refrigerated trucks mostly used by commercial farmers are expensive initially when purchasing and costly to operate. However, the small scale farmers should have a well

considered means of transport to avoid temperature abuse, e.g. when using a pickup or a lorry the top should be well covered so that the produce is not exposed to direct rays of sunlight. Once in the market, temperature abuse still continues where the retailers expose their produce to direct sunlight hence making it absorb more heat. Some don't have shades where they can protect the produce from sun's heat during sale. Shades should be erected to prevent the produce from high water loss which results to withering due to heat from the sun which is detrimental since it increases the rate of deterioration leading to high losses (Wilson *et al.*, 2011).

2.4.2.2. Cooling options for harvested perishable produce

2.4.2.2.1. Hydro-cooling

This is done by cooling a warm produce by sprinkling chilled water on the produce. It is not only more efficient and faster (15 times) than air, but also increases relative humidity around the produce hence reducing the rate of water loss (Borompichaichartkul, 2009). However, the method can only be used on commodities that are not water sensitive e.g. mangoes, apples, carrots, cucumbers and oranges. The two major ways of hydro-cooling are the immersion where the produce is place in the water and the shower type where the produce has water sprinkled on it mechanically (El-Ramady *et al.*, 2015). Since the water comes into contact with the produce, it should be sanitized to avoid microbial growth (Kader, 2004).

2.4.2.2. Forced air cooling

It is achieved by passing cold air through vents in the storage containers that cools the produce as it picks up heat (El-Ramady *et al.*, 2015). However, it leads to high water loss due to reduced relative humidity around the produce. Its efficiency is 75-90 times higher than that of room cooling (Borompichaichartkul, 2009). The method has been used successfully in cooling products like, avocado, melons, cucumber, coconuts, banana and mangoes (Kader and Rolle, 2004).

2.4.2.2.3. Room cooling

In this option, produce is packed in a cold room and cold air allowed to flow through. It is a very slow method but energy efficient and can't be used for products that require rapid cooling like strawberry but has been successfully applied in storage of less perishable products like onions, potatoes and citrus (Kitinoja *et al.*, 2010). It is mostly used for products that are sensitive to chilling injury (Kitinoja *et al.*, 2011).

2.4.2.3. Technologies for cold storage of harvested perishable produce

2.4.2.3.1. Conventional cold rooms

The conventional cold room has two major components which are an insulated room and a refrigeration system. The refrigeration system's two main components are the evaporator unit which is inside the cold room and the compressor unit which is outside the cold room. These components are connected using a refrigeration piping (Elisa Carter, 2016). The cooling is facilitated by a refrigerant and controlled by thermostat which ensures the temperature is maintained near the desired set point. To get a conventional cold room in place, one requires more than 10,000 USD (https://www.alibaba.com/showroom/cold-room-price.html).

2.4.2.3.2. Low cost alternatives to conventional rooms

The technological options for small holder farmers are the low-cost cold storage (LCCS) methods, being emphasized for use to lower and or maintaining temperature at low levels during storage. Once adopted, the LCCS technologies will allow for storage of horticultural products at a lower cost and this will increase farmers' income becoming more economically empowered (IBRD, 2011).

2.4.2.3.2.1. Evaporative Cooling Technologies

The charcoal cooler and zero-energy brick cooler use the principle of evaporative cooling. The charcoal cooler is made by building a structure whose walls are filled up with

charcoal held in by wire netting while the ZEBC is made of a double wall of bricks and the space in between filled with sand (Das and Chandra, 2001). The pads (charcoal and bricks) are wetted by a constant supply of water. As warm dry air passes through the wetted pads, water evaporates taking with it heat from the environment within the chamber hence cooling the air around the product, consequently cooling the product itself (Basediya *et al.*, 2013). The coolers should be placed in a shade and the area should be preferably windy or with good ventilation such that water vapor can easily be swept away from the product and this will hasten the rate of cooling of the product stored therein (Ngoni, 2000). Produce stored in the cool chambers has shown reduced weight loss, better firmness and reduced deterioration rate (Bhatnagar *et al.*, 1990).

A study conducted in Isiolo, Kenya (Wayua *et al.*, 2012) for milk storage revealed that charcoal cooler had efficiency of 74.2 to 86.7% during hottest time of the day cooling the product to 10.5°C below the ambient temperatures. The effect of cooling could be felt up to the fourth day after removal from charcoal cooler (Younan, 2004). In Rwanda, temperature in the charcoal cooler where passion fruits were store was 7°C lower in comparison to the temperatures outside the storage chamber. Site selection is important in setting up an evaporative cooler. The area should have adequate water to ensure the charcoal is wet at all times for it to be effective (Ngoni, 2000).

The ZEBC has been found to maintain relatively low temperature and high humidity in comparison to the outside environmental factors (Islam *et al.*, 2012). Ensuring there is adequate moisture on the walls and the ground is the most effective way of promoting the cooling process within the device and also maintaining high relative humidity. The cooler has successfully extended shelf life of potatoes, tomatoes, eggplants, mangoes, bananas and spinach by 3 to 15 days as compared to products stored at ambient conditions in Orissa (Kalpana *et al.*, 2010). In India, a 20% reduction in losses and an increase in market value of

vegetables by 20% from \$ 1.00 to \$ 1.20 were realized with an extended shelf life of 5-6 days (Susanta, 2009). In Ghana, the temperatures within the cooler were lower (22°C to 27°C) as compared to the ambient temperatures (26°C to 33°C). Cabbages stored within the cooler were found to be of better physical quality as compared to those that were stored at ambient temperature (Saran *et al.*, 2012).

The advantages of using the two mentioned evaporative coolers is that they don't require high level of technological knowhow to construct and one can use the locally available materials to come up with either of the coolers which makes them affordable (Basediya *et al.*, 2013). No electrical energy is needed to operate either of the coolers hence they provide better solutions for the small holder farmers who reside in areas without power supply (Singh and Satapathy, 2006). The great challenge however is availability of water which is mostly a scarce resource especially in areas where the technologies are being implemented. For effective cooling of the products in storage, water has to be present throughout (Kouchakzadeh *et al.*, 2013).

2.4.2.3.2.2. The CoolbotTM Technology

The CoolbotTM (Figure 2.1) is a controller for standard air conditioner which works by manipulating it (air conditioner) to cool the storage room to the desired temperature ranging from 0°C to 18°C depending on the optimum temperature range for stored commodity. The air conditioner is turned into an air compressor that detects the presence of so much heat in the environment and continues to run so as to reduce the temperatures. Even at very low temperatures there is no build-up of ice on the evaporator coil which may cut back airflow and impair the cooling process. Without the CoolbotTM, the air conditioner alone would go as low as 18°C but not lower than that (Kitinoja *et al.*, 2010).

The CoolbotTM has multiple sensors and one of them acts as a regulator. When the fins are about to freeze, the sensor makes the air conditioner dormant so that it can stop

lowering the temperatures until freezing of the fins. The danger in frozen fins is that temperatures would so low as cause injury on the product (Dubey, 2016). It also has a microcontroller that allows the system to work even in very hot areas. The amount of electricity used is very little, especially if the storage room is insulated, as compared to when using the normal conventional cold rooms.

The Coolbot[™] cold storage technology is a cheaper alternative for small scale farmers since operation costs are very low due to the fact that it doesn't break up easily (Dubey, 2013). Apart from being environmental friendly by having low electricity consumption, the Coolbot[™] system also has very low carbon emissions. Therefore, it will not be detrimental to the users nor have negative externalities to the environment (Dubey, 2016).

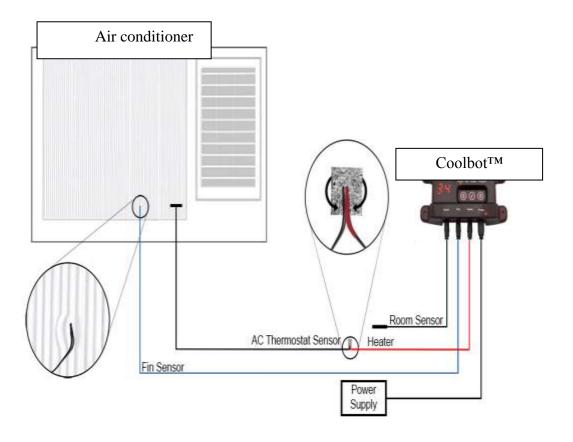


Figure 2. 3. The CoolbotTM gadget and air conditioner

The Coolbot[™] also is more advantageous compared to using the normal coolers where one is required to use a number of fans. The multiple fans, in pursuit of lowering the temperature will dry out the product so quickly causing it to wither (e.g. vegetables) or shrivel in case of fruits which also hastens deterioration rate. While using the Coolbot[™] fitted with an air conditioner, only one fan is used and this reduces the chances of drying out the products. Therefore, with the Coolbot[™] the temperatures will just be kept low without affecting the products' quality negatively (http://www.storeitcold.com/advantages.html accessed on 12th June 2014).

The CoolbotTM has disadvantages though they can't deter one from using it for the advantages overweigh the disadvantages. It takes relatively longer time to cool down and the rate of cooling slows as the temperatures drop. For example, it takes about 20 minutes to drop

the temperature from 30°C to 7°C and then 30 minutes to drop from 7°C to 5°C. However, this would not be so detrimental since most horticultural products can get chilling injury if they are exposed to temperatures above freezing point but below 5°C. The system doesn't function well at a temperature of 2°C or below. This is an advantage in disguise because the farmers wouldn't want the temperatures to be too low as to get to 0°C since freezing injury may occur on the produce.

The temperatures will remain high if the cold room is opened several times, e.g. more than 6 times in an hour and it will also use up more electricity than if the storage room was left closed for longer hours. So it can only be used in a place that is not so busy as to prompt the store to be opened frequently. This is conducive for farmers because they don't have to go into the store every now and then.

CoolbotTM cold storage technology has been successfully used in Ghana for storage of onions which was done in comparison with the traditional storage shed. There was a reduction in losses by 25% from 30% to 5% and stored onions were sold a few months after the production period during the off-season, point at which they had a value of \$1.5 more than the value when there was a market glut (Saran *et al.*, 2012). It was also tested in Amity University where freshly harvested vegetables (chili, eggplants and okra) were stored in a CoolbotTM fitted cold room. Temperatures below 10°C were recorded inside the storage room while in the ambient room there were great fluctuations of temperatures which ranged from 42°C-45°C. After the storage period, the vegetables were still firm, marketable and fresh (Dubey, 2011). A challenge encountered was fluctuations in electricity during storage period but it was arrested by having a generator as a back-up. Though the pay-back period was long (1 year), its useful life was far beyond the pay-back period and could be used for some more years even after break-even point (Saran *et al.*, 2012).

Full installation of a Coolbot[™] cold room of 1 tonne capacity costs about 3,500 USD since its cooling system is a standard air conditioner costing about 1000 USD and the Coolbot[™] gadget of about 300 USD (John, 2012). Using locally available materials in its construction further reduces the cost of obtaining a Coolbot[™] cold room. On the other hand, installing a conventional cold room of the same capacity requires about 10,000 USD which is more than twice the cost of installing a Coolbot[™] cold room. This is due to sophisticated refrigeration system used in the conventional cold room making it expensive beyond the reach of small holder famers (Alexiades *et al.*, 2014). The cost of Coolbot[™] cold room installation can further be reduced by using locally available materials in place of the expensive conventional materials.

CHAPTER 3

3.0. MATERIALS AND METHODS

3.1. EXPERIMENTAL SITE

The study was conducted in Makueni county, Nzaui district, Kawala location at Jasho village. Makueni County is semi-arid with low potential classified as AEZ (V) (Kamau and Mativo, 2014). It lies at an altitude of 450m above the sea receiving not more than 550mm of rainfall per annum which is distributed in two seasons. Mean annual temperature ranges from 26°C to 35°C. Despite the harsh conditions, Makueni County produces some of the best mangoes for export in terms of eating and keeping quality (KALRO, 2015).

3.2. EXPERIMENTAL MATERIALS

3.2.1. Test fruit samples

Two popular mango fruit varieties, "Apple" and "Ngowe" were harvested from commercial farms in Makueni County at physiological maturity stage. Chronological maturity index used was based on days after bloom i.e. Apple at 101 days after bloom and Ngowe at 90 days after bloom. Other subjective maturity indices used include flesh colour (most of the fruits' flesh, white just turning yellow at the seed) and shoulder orientation (shoulders at the same level with the point of attachment). The fruits were harvested at this stage so as to strike a balance between shelf life and fruit quality (to ensure no compromise on either keeping or eating quality).

3.2.2. The CoolbotTM cold room

The Coolbot[™] cold room (Figure 3.1) is made up of three components; an insulated room, air conditioner and the Coolbot[™] gadget. The wall of the 16m² (4m by 4m) insulated room was built using 200 mm thick structural insulated panels made of polystyrene. Polystyrene is a great sound and temperature insulation material that is also water proof. The expanded type of polystyrene used has an R-value of R-5.5. R-value is a measure of the

ability of an insulation material to block heat. Some of the commonly used insulation materials are fibre glass, rock wool and vermiculite which have R-values of 4.3, 4.0 and 2.13. The materials with lower R-value are not as good insulators as those with higher R-values. The walls were then plastered and smoothened using cement and then painted with white paint. A 24,000 BTU LG air conditioner was installed in the insulated room then connected to the Coolbot unit that was sourced from Store-it-Cold LLC, USA. A normal air conditioner working independently would lower the temperatures in a room to a minimum of 18°C. Below 18°C, ice builds on the air conditioner's evaporator coils and would need thawing for the cooling effect to continue. The CoolbotTM is an electronic gadget which acts as a control unit (thermostat) that manipulates the air conditioner to work harder. The CoolbotTM control unit surpasses the thermostat of the air conditioner. Therefore the desired room temperature (depending on the produce) can be set in the CoolbotTM controller. Temperatures as low as 0°C can be set on the CoolbotTM control unit without ice forming on the evaporator coils of the air conditioner. In this study, the temperature on CoolbotTM control unit was pre-set at 10°C which is the lowest safe temperature for mango fruits. The insulation serves to maintain low temperatures in the room by slowing transfer of heat (by conduction and radiation) from outside into the room hence external environmental conditions do not interfere with the low temperatures in the cold room. To reduce power consumption, the room remained closed at all times and was only accessed when necessary so as to avoid entry of hotter air from outside. In addition, only pre-cooled fruits were allowed into the cold room ensuring a reduced heat load.

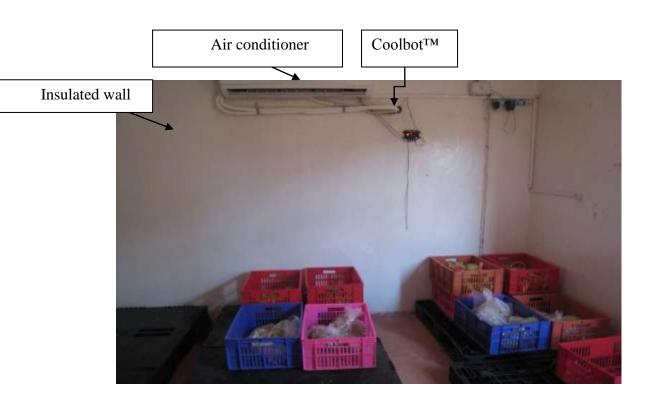


Figure 3. 1. The Coolbot[™] cold room with crates of mango fruits.

3.3. EXPERIMENTAL DESIGN

The mango fruits ("Apple" and "Ngowe" at physiological maturity stage) were harvested by handpicking during the early morning hours to minimize heat load. The fruits were packed into crates lined with moist papers to cushion them from mechanical damage and reduce field heat then transported to the storage area. They were sorted for uniformity and the injured/blemished ones discarded. They were then washed with cold water and 5% acetic acid to disinfect and pre-cool then left to air dry. The fruits were randomly separated into two batches and stored either in the CoolbotTM cold room (temperature preset at $10\pm2^{\circ}$ C) or at ambient temperature conditions ($24^{\circ}-35^{\circ}$ C).

After evaluating the efficacy of the CoolbotTM gadget to lower room temperature during the storage period, a second experiment was set up to evaluate the efficacy to preserve mango fruit quality during the storage period. Further, the synergistic effect of cold storage and modified atmosphere packaging using Activebag® packaging to extend the shelf life and preserve quality of mango fruits was evaluated. In this case the fruits were further batched into two lots which were either packaged using Activebag® or left unpackaged and stored at

ambient room conditions or in the CoolbotTM cold room. The experimental design used was a Completely Randomized Design with a factorial arrangement of treatments.

3.4. EVALUATION OF THE EFFICACY OF THE COOLBOT™ TO LOWER AND MAINTAIN SET TEMPERATURE DURING STORAGE OF MANGO FRUITS.

3.4.1. Monitoring of temperature changes in the CoolbotTM Cold Room and ambient room

Temperature probes sourced from Amiran Kenya LTD were used to monitor temperature changes in the cold room and the ambient room during the storage period. These were not tampered with until the end of storage period when the data recorded in the sensors was retrieved. They were also used to monitor temperature changes in the cold room after every one hour till the preset conditions were achieved and temperatures stabilized at $10\pm1^{\circ}$ C.

3.4.2. Monitoring of internal temperature changes of the stored mango fruits

Tips of the temperature probes were plunged into selected fruits stored in the CoolbotTM cold room and at ambient room conditions. This was done to measure temperature changes inside the fruit during the storage period.

3.5. EVALUATION OF EFFICACY OF THE COOLBOT™ COLD STORAGE TO SLOW DOWN RIPENING OF STORED FRUITS

In the two different storage conditions, ripening progress was compared by sampling three 'Apple' mango fruits from each treatment after every three days for the first 15 days and thereafter every five days for measurement of ripening-related changes. The parameters measured include respiration rate, peel/flesh color and firmness changes.

3.5.1. Changes in respiration

Three 'Apple' mango fruits from the two storage conditions were sampled randomly after every three days for the first 15 days in storage and at intervals of five days thereafter. They were then incubated for two hours at room temperature in 5775 ml plastic jars that had

covers fitted with a self-sealing rubber septum for gas sampling. With the aid of an airtight syringe, gas samples were taken from the headspace gas and injected into gas chromatograph (Models GC-8A, Shimadzu Corp., Kyoto, Japan). Respiration rate was expressed as amount of carbon dioxide gas produced in ml/Kg/Hr at standard atmospheric pressure and calculated as shown below:

 CO_2 production rate ($\mu l/g/h$) =k x 1/r x h x (v-w)/t/w

Where:

 $K \text{=} calibration \quad value \quad (\mu L \quad equivalent \quad to \quad 1 \quad cm \quad peak \quad height \quad on \quad gas$ chromatography)

R=volume of gas injected for sample (ml)

H=peak height (cm)

V=volume of incubation container (ml)

W=weight of sample (g)

T= incubation time (h)

3.5.2. Changes in peel and flesh firmness

The three sampled 'Apple' mango fruits from each treatment had their peel firmness measured at three different spots on an intact fruit while for flesh firmness measurements were taken from three peeled spots. Peel and pulp firmness were measured using a penetrometer (Model CR-100D, Sun Scientific Co. Ltd, Japan) fitted with a 5mm probe. The probe was allowed to penetrate the peel or flesh to a depth of 1.5cm and the corresponding force required to penetrate this depth was determined. Firmness was then expressed as Newton (N) according to Jiang *et al.* (1999).

3.5.3. Changes in peel and flesh colour

Peel and pulp color were measured for three randomly sampled fruits from the two storage conditions using a Minolta color meter (Model CR-200, Osaka, Japan) which was

calibrated with a white and black standard tile. Color coordinates were obtained i.e. L*, a* and b* then the hue angle (h°) calculated by converting the a* and b*according to McLellan *et al.* (1995) as shown:

3.6. EVALUATION OF THE EFFICACY OF THE COOLBOT™ COLD ROOM TO PRESERVE MANGO FRUIT QUALITY, WITH AND WITHOUT MODIFIED ATMOSPHERE PACKAGING

In the second experiment, 'Apple' and 'Ngowe' mango varieties were used to evaluate the efficacy of the Coolbot™ cold room to preserve quality during storage. The fruits were harvested and handled as described in section 3.3 above. In the Coolbot™ cold room, mango fruits for both varieties were either packed or left unpacked in an innovative modified atmosphere packaging (MAP) bag called Activebag® which was sourced from Amiran Kenya Ltd. From each treatment, three fruits were randomly sampled after every three days for the first 15 days and thereafter every five days for measurement of ripening-related changes. The parameters measured include respiration rate, ethylene evolution, cumulative weight loss, peel/flesh color and firmness changes, total soluble solids (TSS) and total titratable acidity (TTA). Changes in quality attributes during storage including soluble sugars, vitamin C and beta carotene were also determined.

3.6.1. Changes in physiological parameters

3.6.1.1. Respiration rate

Random sampling of three fruits was done after every three days for the first 15 days in storage and at intervals of five days thereafter for Apple and Ngowe mango varieties in the

cold room that were either packed in Activebag® or left unpacked. Respiration rate was measured and calculated as described in section 3.5.1 above.

3.6.1.2. Ethylene evolution

Ethylene evolution was measured and calculated as described in section 3.5.1 above using gas chromatograph GC-9A, Shimadzu Corp., Kyoto, Japan.

3.6.1.3. Physiological weight loss (% cumulative)

In both varieties, either packed or unpacked fruits in the cold room, three mango fruits were numbered one to three and weighed on each sampling day using a digital weighing balance (Model Libror AEG-220, Shimadzu Corp. Kyoto, Japan). The initial weight (W1) of the mango fruits at day 0 after harvest was recorded. The new weight of the same fruit (W2) on each sampling day was noted and % cumulative weight loss calculated using the formula below:

% cumulative weight loss = $100 \text{ x (W}_1 - \text{W}_2)/\text{W}_1$

3.6.2. Changes in physical parameters

3.6.2.1. Peel and flesh firmness

Peel and pulp firmness were measured as described in section 3.5.2 above.

3.6.2.2. Peel and flesh colour

Peel and pulp color were measured as described in section 3.5.3 above.

3.6.3. Changes in biochemical quality attributes

3.6.3.1. Total soluble solids

Total soluble solids (TSS) content was determined using an Atago hand refractometer (Model 500, Atago, and Tokyo, Japan). On each sampling day, 3 ml of the fruit juice was extracted from three different fruits and placed on the hand refractometer to obtain the Brix level. The total soluble solid was then expressed as ^oBrix.

3.6.3.2. Total Titratable Acidity

Titration was done to determine the total titratable acidity of the fruits as they ripened.

5ml of the extracted juice was diluted with 25ml of distilled water. 10ml of the diluted juice was titrated with 0.1N Sodium Hydroxide using phenolphthalein as an indicator. The TTA was expressed as % citric acid using the formula;

% Citric acid equivalent = Sample reading (ml)*Dilution factor/sample weight (ml)*Citric acid factor (0.0064)*100

3.6.3.3. Vitamin C (Ascorbic acid) content

Vitamin C content was determined according to AOAC (1996) method. 5mls of the extracted juice was topped up with 10% trichloroacetic acid (TCA) in 100ml volumetric flask. The indicator used (2, 6-dichlophenolindophenol- DCPIP) was then titrated into 10ml of the fruit juice extracted. Ascorbic acid content was calculated as follows:

Ascorbic acid $(mg/100ml) = (A-B) \times C \times 100/S \times (50/5)$

Where,

A = volume in ml of indophenol solution used in the sample.

B = Volume (in ml) of indophenol solution used for the blank.

C = Mass (in mg) of ascorbic acid equivalent to 1 ml of standard indophenol solution.

S = Weight of the sample taken (in ml)

50/5 = total extraction volume/volume of titrated sample

3.6.3.4. Beta-carotene content

Beta-carotene content was determined using a modified chromatographic procedure (Heionen, 1990). 5ml of extracted juice was mixed with 50ml of acetone to extract the carotenoids then filtered using a glass funnel. In a separating funnel 25ml of petroleum ether was used for partitioning to obtain the upper layer which is rich in beta-carotene. Washing was done three times using distilled water to remove acetone residues while keeping the

upper phase. Anhydrous sodium sulphate was added to remove water then the upper phase was stored in sample bottles. The β -carotene content was determined using High Performance Liquid Chromatography (HPLC) (Model LC-10AS, Shimadzu Corp., Kyoto, Japan) and samples read at 450nm. β -carotene content was then calculated as follows:

 β -carotene (mg/100ml) = <u>A*Volume (ml)*104</u>

A^{1%}_{1cm} *sample weight (ml)

Where,

A= Absorbance

Volume = Total volume of extract (25 ml)

 $A^{1\%}_{lcm}$ = Absorption coefficient of β -carotene in Petroleum ether (2592).

3.5.3.5. Main soluble sugars (fructose, sucrose and glucose)

AOAC method (1996) was used to analyze sugars (fructose, sucrose and glucose) in the sampled mango fruits. 5ml of extracted juice was mixed with 50ml distilled water. 2ml of lead acetate was added to the diluted juice and mixed thoroughly. The solution was filtered in 5% anhydrous oxalate and then micro-filtered. Individual sugars were analyzed using a high performance liquid chromatography (HPLC) (Model LC-10AS, Shimadzu Corp., Kyoto, Japan) fitted with a refractive index (RI) detector and running under the following conditions: Oven temperature: 30°C, Flow rate: 0.5-1.0 ml/min, Injection volume: 20 μL and mobile phase: Acetonitrile: water (75:25). Sugars present were identified and their individual concentration calculated using the standards.

3.7. STATISTICAL DATA ANALYSIS

The data collected was subjected to Genstat 15th edition and analyzed using Analysis of Variance (ANOVA). The means were separated by Least Significant Difference (LSD) at P=0.05 using Fisher's protected test.

CHAPTER FOUR

4.0. RESULTS

4.1. FEASIBILITY OF A LOCALLY FABRICATED COOLBOT™ COLD ROOM TO ATTAIN AND MAINTAIN COOL STORAGE TEMPERATURES FOR MANGO FRUITS

4.1.1. Changes in air temperature in the Coolbot™ cold room and ambient room

Initially, temperature in the CoolbotTM cold room was 18° Cwhile that in the ambient room was 27° C. In the cold room, the temperatures were lowered to the pre-set $10\pm1^{\circ}$ C within the first 6 hours (Figure 4.1). The cooling process was rapid at the beginning (4° C/hr) and slowed to 0.5° C/hr towards the 6^{th} hour as temperature conditions in the cold room stabilized at $10\pm1^{\circ}$ C. In the ambient room, the temperatures fluctuated between 25° C- 31° C.

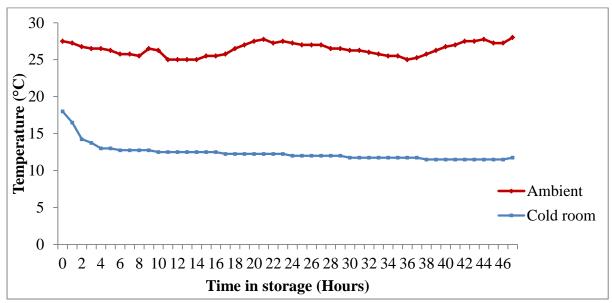


Figure 4.1. Changes in temperature in the CoolbotTM cold room and ambient room during the first 47 hours of storage as collected by the temperature probes

4.1.2. Changes in internal temperature of mango fruits during storage

Temperature changes in the storage area had an effect on the temperature inside the produce. The initial internal temperature in mangoes stored either in the CoolbotTM cold room or at ambient room conditions was 25°C. In the CoolbotTM cold room, fruits were cooled as

shown by the reduction in internal temperature which initially happened at a faster rate of 2°C/hr for the first three hours then the rate slowed down to 1°C/hr till the 7th hour when the temperature stabilized at 10±1°C and was maintained during storage. Internal temperature in fruits stored at ambient room conditions remained significantly high fluctuating between 25°C and 28°C (Figure 4.2).

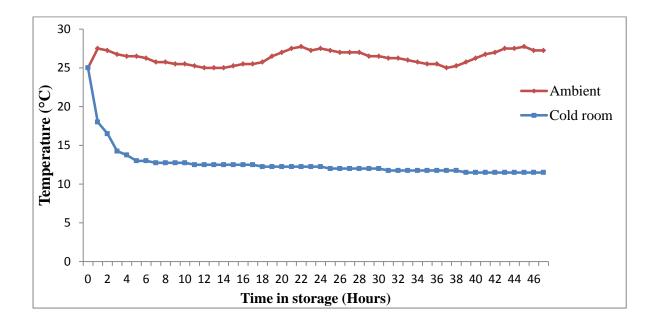


Figure 4.2. Changes in internal temperature of 'Apple' mango fruits stored either in the CoolbotTM cold room and ambient room during the first 47 hours of storageas collected by temperature probes.

4.1.3. Changes in temperature in the Coolbot TM cold room and ambient room conditions during the storage period.

The Coolbot[™] had a significant effect on air temperature in the cold room. Temperatures in the Coolbot[™] cold room dropped from18°C to the pre-set 10±1°C within the first 6 hours on the first day of storage (Figure 4.3). Once the temperatures stabilized, 10±1°C was maintained throughout the storage period. Temperatures in the ambient room fluctuated between 25°C and 31°C during the 40 days storage period.

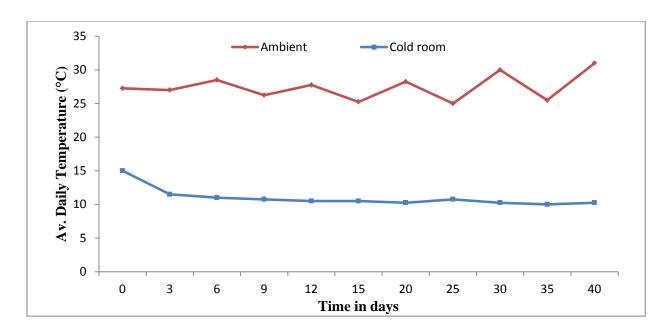


Figure 4.3. Changes in average daily temperature in the Coolbot[™] cold room and ambient room during the 40-day storage period.

4.2. COMPARISON OF RIPENING CHANGES IN FRUITS UNDER THE COOLBOT™ COLD ROOM AND AMBIENT ROOM CONDITIONS

4.2.1. Respiration rate

In storage conditions, respiration rate increased gradually with storage time and as the fruits ripened. Fruits stored in the Coolbot[™] cold room had a significantly (p≤0.05) lower respiration on each sampling day and reached the peak levels several days later than those stored at ambient room conditions (Figure 4.4). In fruits stored at ambient room conditions, respiration rate increased from 32.1 ml/kg/hr to 53.87 ml/kg/hr on 12th day while cold stored fruits had a delayed respiratory peak of 52.6 ml/kg/hr by the end of marketable shelf life on day 35.

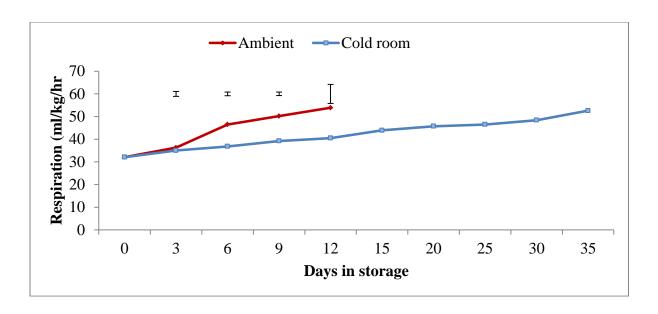


Figure 4.4. Change in respiration rate of 'Apple' mango fruits stored in the cold room and at ambient room conditions. Top bars represent LSD of means ($p \le 0.05$).

4.2.2. Peel firmness

All fruits had reduced peel firmness as ripening progressed despite the storage conditions. Cold storage had a significant (p≤0.05) effect on peel firmness since cold stored fruits retained higher peel firmness during storage compared to fruits in the control whose peel softened at a faster rate (Figure 4.5). In fruits stored at ambient room conditions, peel firmness reduced from 56.8N to 28.1N by the end of marketable shelf life (day 12) compared to cold stored fruits whose firmness was 49.23N on the same sampling day. By the end of marketable shelf life (day 35), peel firmness of cold stored fruits had reduced to 37.43N.

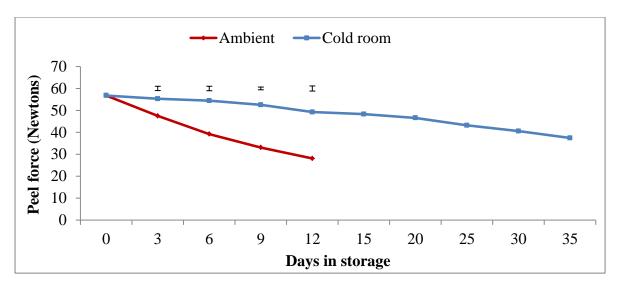


Figure 4.5. Peel firmness of Apple (A) mango fruits stored in cold room and at ambient room conditions. Top bars represent LSD of means ($p \le 0.05$).

4.2.3. Flesh firmness

In both storage conditions, flesh firmness in fruits decreased gradually as ripening progressed. In comparison to fruits at ambient room conditions, cold stored fruits retained significantly ($p \le 0.05$) higher flesh firmness throughout the storage period (Figure 4.6). Fruits stored at ambient room conditions had the highest rate of flesh firmness reduction from 40.9N to 17.57N by day 12 compared to 33.67N for cold-stored fruits on the same day, which reduced to 25.6N by the end of marketable shelf life (day 35).

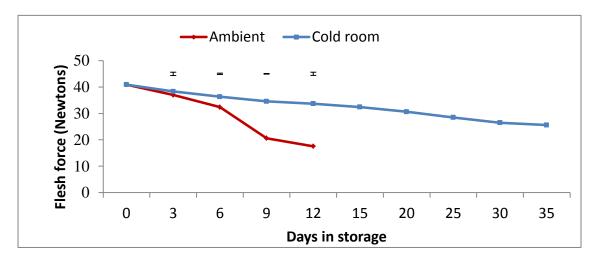


Figure 4.6. Flesh firmness of 'Apple' mango fruits stored in cold room and at ambient room conditions. Top bars represent LSD of means ($p \le 0.05$).

4.2.4. Peel color

Peel colour, expressed as hue angle, decreased gradually in all fruits as they ripened irrespective of the storage conditions they were subjected to (Figure 4.7). There was a significant effect of cold storage on color change from green to yellow. Fruits stored at ambient room conditions had significantly lower hue angles reducing from 103.5° to 62.13° at the end of the fruit's shelf life on day 12. Cold-stored fruits retained significantly (p \leq 0.05) higher hue angles that reduced to 63.6° on day 35.

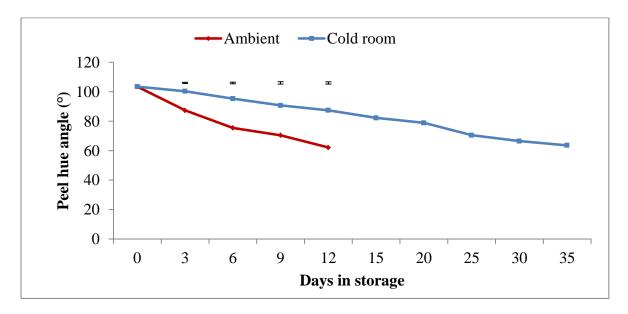


Figure 4.7. Peel colour of 'Apple' mango fruits stored in cold room and at ambient room conditions. Top bars represent LSD of means ($p \le 0.05$)

4.2.5. Flesh colour

Flesh colour (hue angle) reduced as ripening progressed in all fruits irrespective of storage conditions (Figure 4.8). Cold-stored fruits retained significantly (p≤0.05) higher hue angle than fruits stored at ambient room conditions as flesh color changed from cream white to orange. Hue angle in fruits stored at ambient room conditions reduced from 91.5° to 58.53° by the end of storage (day 12) compared to 64.6° in fruits stored in the cold room 23 days later. In cold-stored fruits, color change was slowed down and more color retained at the end of marketable shelf life.

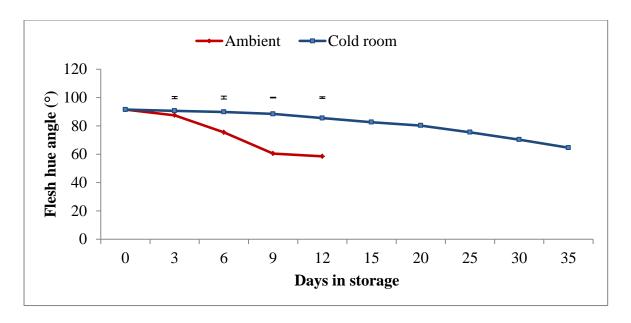


Figure 4.8: Flesh colour of 'Apple' mango fruits stored in cold room and at ambient room conditions. Top bars represent LSD of means ($p \le 0.05$).

4.3. RIPENING AND QUALITY CHANGES IN MANGO FRUITS STORED IN THE COOLBOT™ COLD ROOM OR AT AMBIENT ROOM CONDITIONS, WITH OR WITHOUT MODIFIED ATMOSPHERE PACKAGING

4.3.1. Changes in physiological parameters

4.3.1.1. Respiration rate

In both varieties, respiration rate increased gradually with storage time and as the fruits ripened. Fruits stored in the Coolbot™ cold room had a significantly (p≤0.05) lower rate of CO₂ evolution on each sampling day and reached the peak levels several days later than those stored at ambient room conditions (Figure 4.9). A combination cold storage and MAP using active bags® significantly (p≤0.05) slowed down the respiration rate compared to the cold stored fruits that were not packed. In 'Apple' mango, CO₂ production in unpacked fruits increased from 32.1 ml/kg/hr to 53.87 ml/kg/hr on the 9th day at ambient room conditions while cold stored fruits had a delayed respiratory peak of 52.6 ml/kg/hr observed on day 25. In the cold room, packed fruits had a delayed and significantly smaller peak of 47.1ml/kg/hr that was observed five days later than in unpacked fruits. In 'Ngowe' mango

fruits stored at ambient room conditions, respiration rate increased from 32.27 ml/kg/hr to peak at 60.7 ml/kg/hr (day 6) compared to the cold-stored fruits' 53.1 ml/kg/hr peak which occurred on the 15th day of storage. Packing cold-stored fruits in active bag® resulted in lower respiration rate peaking at 43.6 ml/kg/hr on day 20 compared to 53.1 ml/kg/hr on day 15 in the unpacked fruits.

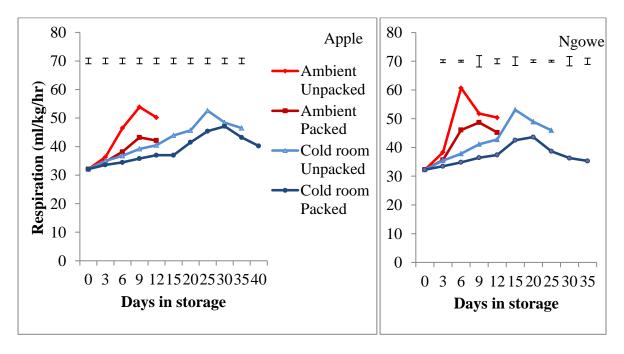


Figure 4.9. Change in respiration rate of 'Apple' and 'Ngowe' mango varieties respectively that were stored in the cold room and at ambient room conditions, either packed in Active bag® or left unpacked. Top bars represent LSD of means (p≤0.05)

4.3.1.2. Ethylene evolution

Ethylene evolution increased gradually in all fruits as they ripened. Cold storage significantly (p≤0.05) affected amount of ethylene produced by the fruits. Though ethylene evolution increased gradually, cold stored fruits produced lower amounts of ethylene compared to those at ambient room conditions on each sampling day. A combination of cold storage and MAP significantly (p≤0.05) lowered the amounts of ethylene produced as compared to cold-stored fruits that were left unpacked (Figure 4.10). In 'Apple' mango fruits, ethylene concentrations were initially 0.07ml/kg/hr which increased to 0.73 ml/kg/hr for fruits stored at ambient

conditions (day 9) and to 0.49 ml/kg/hr for cold stored fruits, 21 days later (day 30). A combination of cold storage and MAP significantly (p≤0.05) lowered and delayed the peak (0.12 ml/kg/hr) observed on day 30. In 'Ngowe' fruits stored at ambient room conditions, ethylene increased from the initial value 0.08 ml/kg/hr to 0.58 ml/kg/hrby day 6 compared to fruits in cold storage whose ethylene levels rose slowly to 0.45 ml/kg/hron day 16. Packed cold-stored fruits had delayed and lowest amount ethylene evolution on each sampling day which increased to 0.21ml/kg/hr on day 20, 4 days later after the unpacked fruits in the cold room had reached the peak.

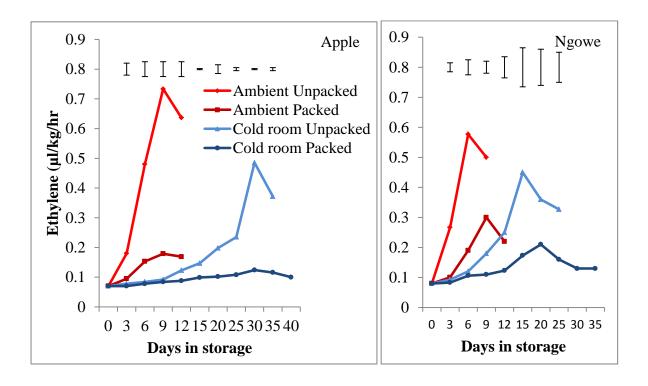


Figure 4.10. Rate of ethylene evolution in 'Apple' and 'Ngowe' mango varieties respectively stored in the cold room and at ambient room conditions either packed in active bag @ or left unpacked. Top bars represent LSD of means ($p \le 0.05$).

4.3.1.3. Percent cumulative weight loss

In all fruits, percent cumulative weight loss increased as they ripened. Fruits stored in the cold room retained a significantly (p≤0.05) higher percent of their initial weight as compared to those stored at ambient room conditions (Figure 4.11). A combination of cold storage and MAP significantly reduced rate of weight loss in the fruits subjected to this treatment as compared to cold storage without packaging. 'Apple' mango fruits stored at ambient room conditions lost 6.39% at the end of marketable shelf life (day 12) in comparison to 5.93% lost by fruits stored in the cold room 27 days later. Fruits stored in the cold room without packaging lost 5.93 % of the initial weight at the end of marketable shelf life (day 35) compared to 3.97% of packaged fruits in the cold room on the same day. In Ngowe, fruits stored at ambient room conditions had lost 5.52% of their initial weight by day 12 (end of their shelf life) while those in the cold room had lost 1.76%. Combination of cold storage and MAP caused significantly (p≤0.05) higher fruit weight retention. Fruits subjected to this treatment lost only 3.85% of the initial weight at the end of its storage life (day 35) compared to the 4.91% lost by cold stored fruits that were not packed in Active bags.

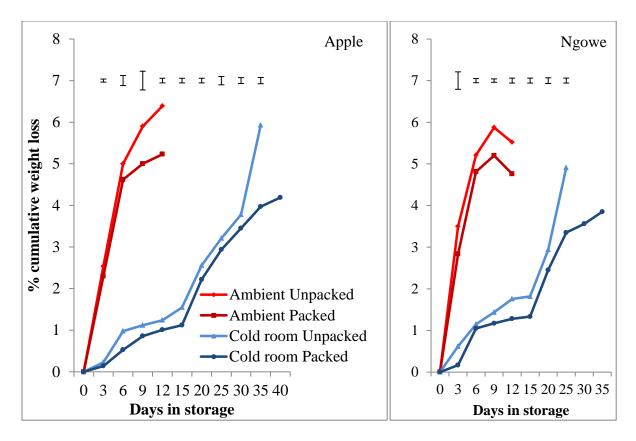


Figure 4.11. Percent cumulative weight loss of 'Apple' and 'Ngowe' mango varieties stored in cold room and at ambient room conditions either packed in Activebag® or left unpacked. Top bars represent LSD of means ($p \le 0.05$).

4.3.2. Changes in physical attributes of the fruits

4.3.2.1. Peel firmness

All fruits had reduced peel firmness as ripening progressed. Cold storage had a significant (p≤0.05) effect on peel firmness since cold stored fruits retained higher peel firmness during storage as compared to fruits at ambient room conditions whose peel softened at a faster rate (Figure 4.12). Fruits subjected to both cold storage and MAP treatment significantly (p≤0.05) remained firmer on all sampling days compared to fruits subjected to cold storage but left unpacked in Active bags®. In apple mango fruits stored at ambient room conditions, peel firmness reduced from 56.8N to 28.1N by the end of marketable shelf life (day 12) compared to cold-stored fruits whose firmness was 49.23N on the same sampling day. Peel firmness of cold-stored fruits that were not packed reduced to

37.43N (day 35) while those stored in the cold room and packed in Active bag® reduced to 40.57N on day 40. In 'Ngowe', peel firmness in fruits at ambient room conditions reduced gradually and faster from 37.53N to 8.9N by the end of their shelf life (day 9) compared to those stored in the cold room which reduced to 25.17N by the end of their shelf life on day 25 in storage. In the cold room, packaged fruits' peel softened at the slowest rate to 27.83N by day 35, 10 days later after unpackaged fruits had softened beyond marketable stage.

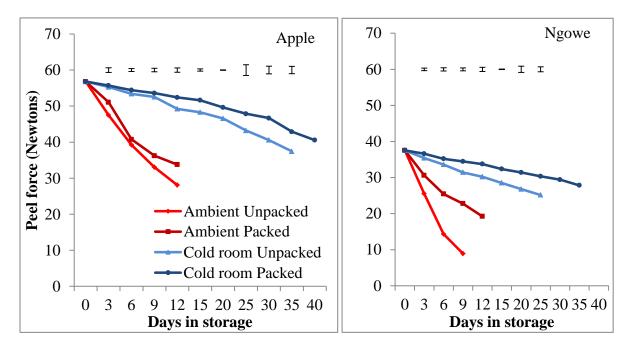


Figure 4.12. Peel firmness of 'Apple' and 'Ngowe' mango varieties stored in cold room and at ambient room conditions either packed in Active bag® or left unpacked. Top bars represent LSD of means ($p \le 0.05$)

4.3.2.2. Flesh firmness

In all treatments, flesh firmness in fruits decreased gradually. In comparison to fruits at ambient room conditions, cold stored fruits retained significantly (p≤0.05) higher flesh firmness throughout the storage period (Figure 4.13). A combination of cold storage and MAP had a positive effect on flesh since the fruits maintained significantly higher flesh firmness compared to those stored in the cold room without packing. 'Apple' mango fruits in the control had the highest rate of flesh firmness reduction from 40.9N to 17.57N by day 12

compared to 33.67N for cold-stored fruits on the same day. Flesh firmness of unpacked fruits in the cold room reduced to 25.6N by the 35th day while those packed in Active bag®) softened at a significantly slow rate to 30.50N on day 40 (5 days later). Firmness in 'Ngowe' mango fruits stored at ambient room conditions reduced from 28.3N to 5.88Non day 9 compared to significantly slower rate of cold stored fruits to 13.53N, 16 days after fruits in the control had reached end of their marketable shelf life. Flesh firmness in cold stored packed fruits reduced to 17.45N on day 35, i.e. 5 days after cold stored unpacked fruits firmness had dropped to 13.53N by the 30th day.

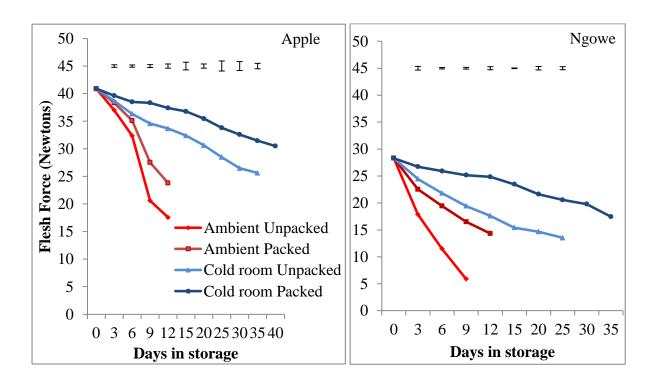


Figure 4.13. Flesh firmness of 'Apple' and 'Ngowe' mango varieties stored in cold room and at ambient room conditions either packed in Active bag® or left unpacked. Top bars represent LSD of means ($p \le 0.05$).

4.3.2.3. Peel colour

Peel colour, expressed as hue angle, decreased gradually in all fruits as they ripened. There was a significant effect of cold storage on color change from green to yellow. Complementing cold storage with MAP significantly ($p \le 0.05$) affected peel hue angle (Figure 4.14). 'Apple' mango fruits stored at ambient room conditions had significantly lower hue angles reducing from 103.5° to 62.13° at the end of the fruits' shelf life on day 12. Cold stored unpacked fruits retained significantly ($p \le 0.05$) higher hue angles that reduced to 63.6° on day 35 compared to control fruits though not as high as cold stored fruits that were packed in Active bags® whose peel hue angles' reduction was slow and delayed to 80.87° by the 40^{th} day (end of storage) with a high retention of original colour (green). In 'Ngowe' mangoes stored at ambient room conditions, peel hue angle reduced from 99.17° to 67.76° at the end of storage (day 9) compared to 63.37° in cold storage 16 days later. Interaction between cold storage and MAP retained significantly ($p \le 0.05$) high hue angle in fruits reducing to 77.77° by day 35.

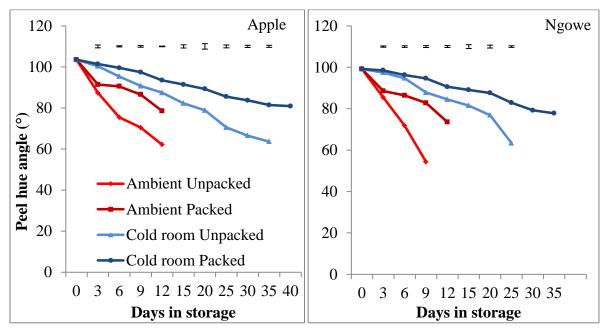


Figure 4.14. Change in peel hue angle of 'Apple' and 'Ngowe' mango varieties stored in cold room and at ambient room conditions either packed in Active bag® or left unpacked. Top bars represent LSD of means ($p \le 0.05$).

4.3.2.4. Flesh colour

Flesh colour (hue angle) reduced as ripening progressed in all fruits irrespective of the treatment. Cold-stored fruits retained significantly (p≤0.05) higher hue angle than fruits stored in the control. Interaction between cold storage and MAP was evidenced by high retention of flesh colour in fruits subjected to this treatment (Figure 4.15). In 'Apple' mango fruits stored at ambient room conditions hue angles reduced from 91.5° to 58.53° by the end of storage (day 12) compared to 79.57° in fruits stored in the cold room 23 days later. Packaging enhanced shelf life and reduced colour for fruits in the cold room whose hue angle was 72.98° at the end of storage, 5 days after unpacked fruits had reached end of their marketable shelf life. In 'Ngowe', flesh hue angle reduced from 87.97° to 58.43° at the end of storage (day 12) in the control room and to 60.67° for fruits in the cold room. Fruits in the cold room packed in Active bag® retained significantly (p≤0.05) more of the initial flesh colour compared to cold stored fruits that were not packed, reducing to 74.43° after 35 days in storage.

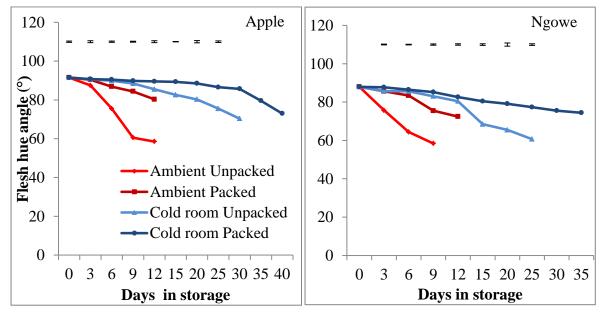


Figure 4.15. Change in flesh hue angle of 'Apple' and 'Ngowe' mango varieties stored in the CoolbotTM cold room and at ambient room conditions either packed in Active bag® or left unpacked. Top bars represent LSD of means ($p \le 0.05$).

4.3.3. Changes in biochemical quality attributes

4.3.3.1. Total soluble solids (TSS)

There was a gradual increase in total soluble solids in all fruits during ripening. Coldstored fruits had a slower rate of increase in TSS compared to fruits stored at ambient room conditions. Interaction of cold storage and MAP significantly affected TSS content of fruits (Figure 4.16). Fruits subjected to a combination of cold storage and MAP had significantly lower levels of TSS compared to those subjected to cold treatment without packaging. In 'Apple' mango fruits, TSS content increased from 4.2° to 12.7° at the end of storage in fruits stored at ambient room conditions compared to 13.3° on day 35 in cold room. TSS content for packed fruits in the cold room increased to 10.3° at the end of storage (day 40). In 'Ngowe' mango fruits, TSS increased from 3.43° to 12.7° at the end of storage for fruits stored at ambient room conditions compared to 13.6° for fruits stored in the cold room. TSS content in fruits packed in Active bag® increased to 11.23° by 35th day in storage which was significantly lower than unpacked fruits in the cold room (13.6° at day 25).

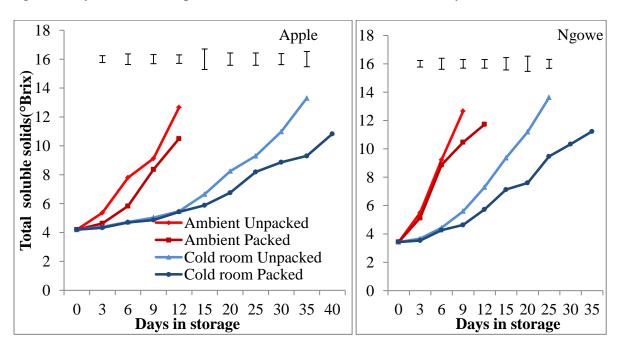


Figure 4.16. Change in Brix of 'Apple' and 'Ngowe' mango varieties stored in cold room and at ambient room conditions either packed or left unpacked. Top bars represent LSD of means ($p \le 0.05$).

4.3.3.2. Total Titratable Acidity (TTA)

TTA content decreased in all fruits as ripening progressed and was significantly affected by cold storage. Fruits stored in the cold room had significantly (p≤0.05) higher TTA content throughout the storage period. Interaction between cold storage and MAP significantly affected TTA content (Figure 4.17). All packed fruits stored in the cold room retained significantly (P \le 0.05) higher amount of TTA content irrespective of variety compared to unpacked fruits. In 'Apple' mangoes, TTA reduced from 0.75% citric acid equivalent to 0.03% citric acid equivalent at end of storage (day 12) in fruits stored at ambient room conditions compared to 0.05% in cold stored fruits at day 35. Cold-stored packed fruits' TTA reduced to 0.08% citric acid equivalent compared to the significantly (P≤0.05) lower TTA content (0.05% citric acid equivalent) in cold-stored unpacked fruits. In Ngowe mangoes, fruits in the control reduced TTA content from 0.87% citric acid equivalent to 0.08 % (day 9) citric acid equivalent compared to the delayed (16 days later) 0.08% citric acid equivalent observed on day 25 in cold-stored fruits at the end of their storage on day. Cold-stored fruits that were packed retained significantly (p≤0.05) high levels of TTA content (0.22% citric acid equivalent compared to 0.08% of unpacked fruits on day25). TTA content reduced to 0.09% citric acid equivalent on day 35.

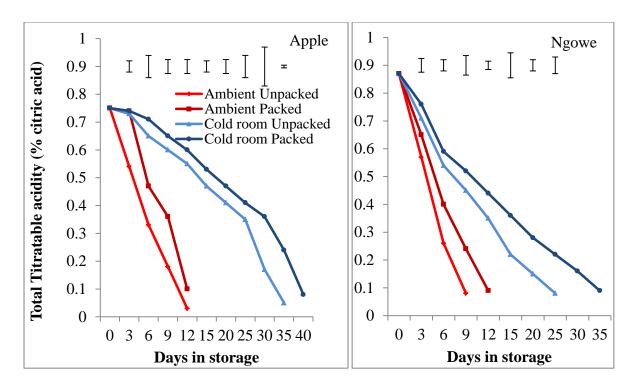


Figure 4.17: Change in Total titratable acidity (% citric acid) of 'Apple' and 'Ngowe' mango varieties stored in cold room and at ambient room conditions, either packed in Active bag® or left unpacked. Top bars represent LSD of means ($p \le 0.05$).

4.3.3.3. Beta carotene

There was a gradual increase in beta carotene content with ripening in all fruits irrespective of the variety. Cold storage significantly (p≤0.05) slowed down the rate of increase of beta carotene content (Figure 4.18). An interaction between cold storage and MAP had a significant effect on the level of beta carotene content on each sampling day. Cold stored packed fruits had significantly (p≤0.05) lower levels of beta carotene content. In 'Apple' mango, fruits at ambient room conditions increased beta carotene content from 0.43 mg/100ml to 9.23 mg/100ml at the end of storage on day 12 compared to 7.85 mg/100ml in cold stored fruits 23 days later. Cold-stored fruits packed in active bags stayed 5 days longer than unpacked fruits with significantly low beta carotene level of 1.93mg/100ml. In 'Ngowe', fruits at ambient room conditions increased beta carotene content from 0.5 mg/100ml to 8.46 mg/100ml compared to 7.41 mg/100ml for cold stored fruits 16 days later. Packed cold stored

fruits had slowest rate in increase of beta carotene content to 1.95mg/100ml, 0.26% of the observed beta carotene content in unpacked fruits stored in the cold room (7.41mg/100ml).

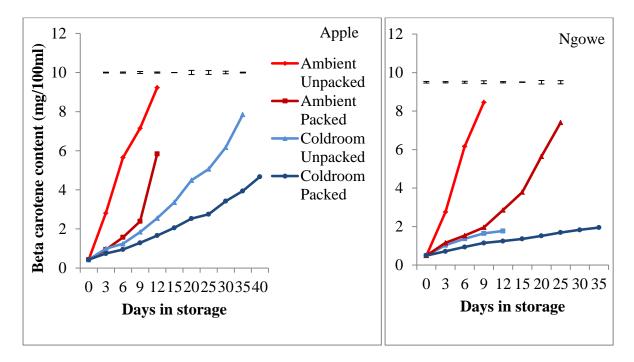


Figure 4.18. Change in beta carotene content (mg/100ml) of 'Apple' and 'Ngowe' mango fruits stored in the Coolbot™ cold room and ambient room conditions, either packed in Active bag® or left unpacked. Top bars represent LSD of means (p≤0.05)

4.3.3.4. Vitamin C

Vitamin C decreased gradually in all fruits as ripening progressed irrespective of variety. Cold storage affected retention of Vitamin C content in fruits. Interaction between cold storage and MAP had a significant (p≤0.05) effect on the rate of decrease of Vitamin C content (Figure 4.19). In 'Apple' mangoes at ambient room conditions it reduced from 110.70mg/100ml to 51.53mg/100ml at the end of storage (day 12) compared to 51.8mg/100ml in cold storage (day 35). Cold-stored packed fruits retained significantly higher content of Vitamin C (59.77mg/100ml) at the end of storage (day 40). In Ngowe mangoes stored at ambient room conditions, the content reduced from the initial 106.23mg/100ml to 45.53mg/100ml on the 9th day compared to the 49.1 mg/100ml in cold

stored fruits on day 25. Cold-stored packed fruits had highest retention of Vitamin C (58.17mg/100ml) at the end of storage on day 40.

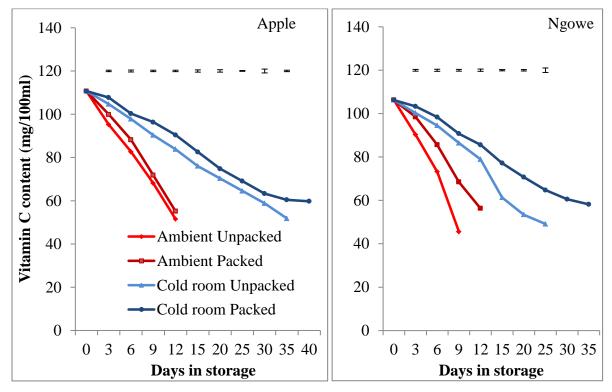


Figure 4.19. Change in Vitamin C content (mg/100ml) of 'Apple' and 'Ngowe' mango fruits stored in the Coolbot[™] cold room and ambient room conditions, either packed in Active bag® or left unpacked. Top bars represent LSD of means (p≤0.05).

4.3.4. Changes in major sugars

4.3.4.1. Fructose

Fructose content increased gradually in all fruits as they ripened. Increase in the fructose content was significantly (p<0.05) delayed in cold stored fruits. Combination of packaging and cold storage significantly (p<0.05) affected rate of increase in fructose content since cold stored that were packed in active bag® had less rapid increase compared to cold stored fruits not packed (Table 4.1 and 4.2). Fructose content in Apple mangoes stored at ambient room conditions increased from the initial 1.8mg/100ml to 7.9 mg/100ml at the end of storage on day 12 compared to 8.9 mg/100ml in cold stored fruits 23 days later. Cold stored packed fruits had significantly lower fructose levels (8.7 mg/100ml) at the end of storage period5days after unpacked fruits had reached end of marketable shelf life. In Ngowe

mangoes stored at ambient room conditions, fructose content increased from the initial 1.7 mg/100ml to 7.0 mg/100ml on day 9 compared to 8.1 mg/100ml in the cold room (day 25). Fructose levels in cold stored packed fruits increased to 8.4 mg/100ml 10 days after unpacked fruits had reached end of marketable shelf life.

Table 4.1. Change in fructose content (mg/100ml) in 'Apple' mango fruits stored in the CoolbotTM cold room and at ambient room conditions, either packed in Active bag® or left unpacked.

		DAYS	IN STO	ORAGI	Ε							
Storage	Packing	0	3	6	9	12	15	20	25	30	35	40
Condition	option											
Ambient	Unpacked	1.8a	2.5a	4.5a	6.3a	7.9a						
Ambient	Packed	1.8a	2.0b	4.1b	5.9b	7.6b						
Cold room	Unpacked	1.8a	1.9b	2.5c	3.8c	4.8c	5.8a	6.7a	7.1a	7.9a	8.9a	
Cold room	Packed	1.8a	1.9b	2.1d	2.9d	3.7d	4.8b	5.5b	6.0b	7.1b	7.8b	8.7
	Mean	1.8	2.1	3.3	4.7	5.9	5.3	6.1	6.6	7.5	8.3	8.7
	LSD		0.2	0.2	0.2	0.3	0.4	0.3	0.3	0.4	0.6	

Means within each column followed by different letters differ significantly at (p<0.05)

Table 4.2. Change in fructose content (mg/100ml) in 'Ngowe' mango variety stored in the CoolbotTM cold room and at ambient room conditions, either packed in Active bag® or left unpacked.

		DAYS IN STORAGE											
Storage	Packing	0	3	6	9		15	20	25	30	35		
Condition	option				12								
Ambient	Unpacked	1.7a	2.3a	6.1a	7.0a								
Ambient	Packed	1.7a	2.2a	4.2b	6.1b	7.3a							
Cold room	Unpacked	1.7a	1.9b	3.5c	4.2c	5.3b	6.3a	7.4a	8.1a				
Cold room	Packed	1.7a	1.9b	2.7d	3.6d	4.1c	5.0b	5.6b	6.3b	7.7	8.4		
	Mean	1.7	2.1	4.1	5.2	5.6	5.7	6.5	7.2	7.7	8.4		
	LSD		0.1	0.2	0.2	0.1	0.3	0.8	0.2				

Means within each column followed by different letters differ significantly at (p<0.05)

4.3.4.2. Sucrose

A gradual increase in sucrose content was observed in all fruits as they ripened. Sucrose content increase was significantly (p<0.05) affected by cold storage. A combination of cold storage and MAP significantly delayed rate of increase of sucrose content compared to cold stored fruits that were not packed in Active bag® (Table 4.3 and 4.4). In 'Apple' mangoes stored at ambient room conditions, sucrose content increased from the initial 1.8mg/100ml to 7.8 mg/100ml at the end of storage on day 12 compared to 8.4 mg/100ml in cold stored fruits 23 days later. Cold stored packed fruits had significantly lower fructose levels (8.5 mg/100ml) at the end of storage period 5 days after unpacked fruits had reached end of marketable shelf life. In 'Ngowe' mangoes stored at ambient room conditions, sucrose content increased from the initial 1.3 mg/100ml to 6.8 mg/100ml on day 9 compared to 6.7 mg/100ml in the cold room (day 25). Cold stored packed fruits increased sucrose levels to 6.9 mg/100ml 10 days after unpacked fruits had reached end of marketable shelf life.

Table 4.3. Change in sucrose content (mg/100ml) in 'Apple' mango fruits stored in the CoolbotTM cold room and at ambient room conditions, either packed in Active bag® or left unpacked.

		DAY	S IN S	STORA	GE							
Storage condition	Packing option	0 :	3	6	9	12	15	20	25	30	35	40
Ambient	Unpacked	1.8a	3.5a	5.2a	6.6a	7.8a						
Ambient	Packed	1.8a	2.5b	4.4b	6.6a	7.7a						
Cold room	Unpacked	1.8a	2.1c	3.6c	4.1b	4.6b	5.3a	6.1a	6.9a	7.6a	8.3a	
Cold room	Packed	1.8a	1.9d	2.2d	2.7c	3.3c	4.0b	5.1b	5.7b	6.6b	7.5b	8.5
	Mean	1.8	2.5	3.9	5.0	5.8	4.7	5.6	6.3	7.1	7.9	8.5
	LSD		0.1	0.1	0.3	0.2	0.2	0.2	0.4	0.1	0.6	

Means within each column followed by different letters differ significantly at (p<0.05)

Table 4.4. Change in sucrose content (mg/100ml) in 'Ngowe' mango fruits stored in the CoolbotTM cold room and at ambient room conditions, either packed in Active bag® or left unpacked.

		DAY	S IN S	TORA							
Storage	Packing	0	3	6	9	12	15	20	25	30	35
condition	option										
Ambient	Unpacked	1.3a	3.1a	5.1a	6.8						
Ambient	Packed	1.3a	2.4b	3.8b	5.2b	6.4a					
Cold room	Unpacked	1.3a	1.8c	2.8c	3.7c	4.7b	5.2a	6.1a	6.7a		
Cold room	Packed	1.3a	1.7c	2.1d	2.1d	3.6c	4.1b	4.9b	5.3b	5.9	6.9
	Mean	1.3	2.3	3.5	4.6	5.5	4.7	5.5	6.0	5.9	6.9
	LSD		0.1	0.1	0.7	0.4	0.3	0.3	0.3		

Means within each column followed by different letters differ significantly at (p<0.05)

4.3.4.3. Glucose

Glucose content increased gradually as ripening progressed. The rate of increase was significantly (p<0.05) affected by cold storage (Table 4.5 and 4.6). Interaction between cold storage and MAP significantly (p<0.05) delayed rate of increase in glucose content compared to cold stored fruits not packed in Active bag®. In 'Apple' mangoes stored at ambient room conditions, glucose content increased from the initial 1.7 mg/100ml to 4.8 mg/100ml at the end of storage on day 12 compared to 4.9 mg/100ml in cold stored fruits 23 days later. Glucose content in cold stored packed fruits was 4.8 mg/100ml, not significantly different from that of unpacked fruits though it was delayed (day 40). In 'Ngowe' mangoes stored at ambient room conditions, glucose content increased from the initial 1.4mg/100ml to 4.2 mg/100ml on day 9 compared to 4.1mg/100ml in the cold room (day 25). Glucose levels in cold stored packed fruits increased to 4.4mg/100ml 10 days after unpacked fruits had reached end of marketable shelf life.

Table 4.5. Change in glucose content (mg/100ml) in 'Apple' mango fruits stored in the CoolbotTM cold room and at ambient room conditions, either packed in Active bag® or left unpacked.

		DAY	S IN S	TORA	GE.							
Storage	Packing	0	3	6	9	12	15	20	25	30	35	40
condition	option											
Ambient	Unpacked	1.7a	2.5a	3.1a	4.1a	4.8a						
Ambient	Packed	1.7a	2.4b	3.0a	4.1a	4.8a						
Cold room	Unpacked	1.7a	2.1c	2.5b	2.8b	3.2b	3.5a	3.7a	3.9a	4.3a	4.9a	
	Packed	1.7a	1.9d	2.3c	2.7b	3.1b	3.3b	3.5b	3.8a	4.1b	4.8b	4.8
Cold room	Mean	1.7	2.2	2.8	3.4	3.9	3.4	3.6	3.9	4.2	4.8	4.8
	LSD		0.1	0.1	0.1	0.1	0.1	0.1	0.3	0.2	0.5	

Means within each column followed by different letters differ significantly at (p<0.05)

Table 4.6. Change in glucose content (mg/100ml) of 'Ngowe' mango fruits stored in the CoolbotTM cold room and at ambient room conditions, either packed in Active bag® or left unpacked.

		DAYS IN STORAGE											
Storage	Packing	0	3	6	9	12	15	20	25	30	35		
condition	option												
Ambient	Unpacked	1.4a	2.5a	3.8a	4.2a								
Ambient	Packed	1.4a	2.3b	2.9b	3.6b	4.4a							
Cold room	Unpacked	1.4a	1.7c	2.1c	2.2c	4.2b	2.9a	3.8a	4.1a				
Cold room	Packed	1.4a	1.7c	1.8d	2.1d	2.6c	2.8b	3.2b	3.5b	4.1	4.4		
	Mean	1.4	2.0	2.7	3.0	3.4	2.9	3.5	3.8	4.1	4.4		
	LSD		0.1	0.1	0.1	0.1	0.1	0.1	0.2				

Means within each column followed by different letters differ significantly at (p<0.05)

CHAPTER 5

5.0. DISCUSSION AND RECOMMENDATIONS

5.1. DISCUSSION

Temperature management is a critical component to maintaining perishable horticultural produce in good quality for a longer period of time (Bachmann and Earles, 2000). Temperature affects shelf life since it is estimated that deteriorative processes in perishable commodities increase 2-3 fold for every 10°C increase in temperature (KMUTT, 2007). Poor temperature management is a major contributor to the high post harvest losses (ranging between 45-50%) experienced along horticultural commodities supply chain (Atanda *et al.*, 2011) hence the need for cold chain maintenance. Most cold chain facilities are integrated to meet specific need especially in Kenya where most investment has been on cold storage for flowers. However, expensive conventional cold rooms are out of reach for small holder farmers (Huyskens-Keil *et a.*, 2015).

The critical need for cold storage has necessitated research in low-cost cold storage technologies as alternative to conventional cold rooms. The CoolbotTM is one such technology which has been used effectively in other countries and on various commodities. Installing a CoolbotTM cold room of 1 tonne capacity costs about USD 3,500. The CoolbotTM cooling system is comprised of a standard air conditioner costing about USD 1000 and the CoolbotTM gadget of about USD 300 (John, 2012) plus the insulated room whose cost can vary 1,000-3,000 USD depending on the level of sophistication. In contrast, installing a conventional cold room of the same capacity requires about USD 10,000 which is more than twice the cost of installing a CoolbotTM cold room. This is due to sophisticated refrigeration system used in the conventional cold room making it expensive beyond the reach of small holder famers (Alexiades *et al.*, 2014).

Other than cold storage, there are several other technologies that have been used to reduce post harvest losses which include Modified Atmosphere Packaging (MAP), Controlled Atmosphere Storage (CAS) and use of edible coatings among others (Kader, 2002). These technologies can be used to complement cold storage for enhanced shelf life and quality preservation of horticultural commodities. In previous studies, cold storage has been coupled with waxing and MAP to extend shelf life of avocado (Mendeta *et al.*, 2016) and mango fruit (Kelany *et al.*, 2010) respectively. In the present study, the efficacy of a new low-cost cold storage technology, CoolbotTM, in reducing post harvest losses among small holder mango farmers was investigated. Poor cold chain management can lead to very high post harvest losses and cold storage is key.

In the first experiment (season 1), the efficacy of CoolbotTM in lowering and maintaining cold temperatures during storage of mango fruits was established. CoolbotTM cold room conditions were compared with the ambient room conditions, where most farmers store their produce after harvest awaiting transport to the market. Results showed that the CoolbotTM was effective in lowering air temperature in the cold room to the set (10±1°C) during the first 6 hours and maintaining it throughout the storage period compared to the fluctuating temperature in the ambient room. Internal temperature of the mango fruits stored in the CoolbotTM cold room was lowered and maintained at 10±1°C compared to that of fruits stored at ambient room conditions which fluctuated between 25°C to 28°C. In conventional cold rooms, cooling produce from 20°C to 10°C takes 6-8 hours (Jobling, 2000). The findings are similar to a study on chili, brinjals and okra (Neeru, 2014) where the pulp of the products stored in the CoolbotTM cold room had significantly lower temperature (between 10°C and 13°C) compared to the fluctuating temperatures (between 20°C and 29°C) at ambient room conditions. Products in the CoolbotTM cold room stayed fresh and marketable longer (up to 21 days) compared to those stored at ambient room conditions. Generally, CoolbotTM cold

storage was found to be effective in lengthening shelf life as evidenced by significantly (p<0.05) slower or delayed ripening related changes including respiration rate, peel/flesh colour and firmness.

Respiration rate of mango fruits in both storage conditions increased gradually as ripening progressed. Respiration rate of fruits in the CoolbotTM cold room was lower and the respiratory peak was delayed by 16 days. Low temperatures slowed down the rate of O₂ consumption and CO₂ evolution. In a study on cold storage of mangoes, a 3 fold increase in respiration was observed with increase in temperature from 5°C -20°C (Mohammed and Brecht, 2002; Devanesan *et al.*, 2012).

Respiratory activity is known to affect other ripening related changes in most climacteric fruits. As ripening progressed, peel and flesh firmness decreased in all fruits irrespective of storage conditions. The decrease has been attributed to enzymes that degrade the cell wall and the insoluble protopectins to more simple soluble pectins (Hoffman et al., 1994). However, cold storage delayed rate of softening of peel and flesh. The slower rate of peel and flesh softening has been attributed to reduced enzymatic reactions of the enzymes (polygalacturonase (PG), pectin methylesterase (PME), pectatelyase and endo-β-1, 4glucanase (EGase) activities) at low temperature (Jarimopas and Kitthawee, 2007). The faster rate of softening in fruits stored at ambient conditions has been attributed to higher rates of starch hydrolysis to soluble sugars since starch has a structural function in the cell (Thompson, 1996) and accelerated rate of ripening associated with high temperature (Kelany et al., 2010). Similar findings were reported in a study conducted on apple fruits (Jalal et al., 2010), bananas (Ahmad et al., 2001) and mangoes (Thanaa and Rehab, 2011). Peel colour, expressed as hue angle, decreased gradually in all fruits as ripening progressed though it was significantly (p<0.05) slower in cold stored mango fruits. Colour change in mango fruits could be attributed to destruction of chlorophyll (green colouring matter) as new pigments

(carotenoids) are synthesized which are responsible for colour (Ueda, 1999). Delayed colour change in cold stored mango fruits was attributed to the suppressed activity of enzymes involved in synthesis of carotenoids due to low temperatures. A study by Suslow and Cantwell (1997) on tomatoes and on bananas (Ahmad *et al.*, 2001) on the effect of cold storage on peel and flesh colour showed similar findings.

In the second experiment (season 2), the synergistic effect of the Coolbot™ cold storage and modified atmosphere packaging in preserving quality and extending shelf life of mango fruits was observed. Ripening progression of cold-stored mango fruits packed in Activebag® was compared with the unpacked cold-stored mango fruits. Cold storage extended shelf lifeof stored mango fruits.. Combining cold storage with modified atmosphere packaging further extended shelf life of "Apple" and "Ngowe" mango fruits.. Packaging with Activebag® alters the gaseous composition around the packed produce and this has been found effective in delaying ripening processes in fruits even without cold storage (Batu and Thompson, 1998 and Karacay *et al.*, 2009). The delayed ripening in cold-stored fruits was evidenced by lower rates of ripening related changes including respiration, ethylene evolution, softening, colour and weight loss. Cold storage and MAP also preserved quality attributes of the stored fruits as evidenced by high levels of vitamin C. Other quality related changes such as sugars, titratable acidity and total soluble solids progressed less rapidly in cold-stored fruits.

Respiration rate increased gradually with storage time and as ripening progressed regardless of the storage conditions. Climacteric peak in cold-stored 'Apple' mango fruits was significantly (p<0.05) delayed compared to the fruits at ambient room conditions. Increase in temperature by 10°C has been shown to increase respiration rate three times in mango fruits (Mohammed and Brecht, 2002; Devanesan *et al.*, 2012). In the cold-stored fruits, MAP further reduced respiration rate compared to unpacked fruits. The lower

respiration rate in packed fruits is attributed to low O₂ and high CO₂ levels within the bag. Not only was the peak delayed but the peak level was significantly lower compared to unpacked fruits'. Similar findings of delayed and lower climacteric peaks were also observed in previous studies on MAP in mangoes (Ouma, 2015). Low respiration rate in packed fruits stored in cold room was attributed to reduced permeability of the Active bag®) film causing accumulation of CO₂. In high temperatures, film permeability is altered leading to reduced accumulation of CO₂ hence high respiration rate (Joles *et al.*, 1994 and Beaudry *et al.*, 2000). The findings of increased respiration rate as temperature is increased in MAP storage are similar to those of a study on strawberries' (Silva *et al.*, 1999) and melon's (Gomes *et al.*, 2012).

Similarly, ethylene evolution in cold-stored packed fruits was slowest in comparison to other treatments. This has been attributed to low temperature that hinders the activity of Aminocyclopropane-1-carboxylic acid (ACC) synthase, hence interfering with ethylene biosynthesis (Amoros *et al*, 2008). The atmosphere in the package, low O₂ and high CO₂ is also known to reduce product's sensitivity to ethylene (Kader and Rolle, 2004). The findings correspond to a similar study on 'Kent' mangoes with low ethylene evolution at low low O₂ and high CO₂ and vice versa (Kelany *et al.*, 2010).

As ripening progressed, physiological weight lost by the fruits increased irrespective of storage conditions or packaging. At ambient room conditions physiological weight loss was significantly higher (6.39%) than in cold-stored fruits. Weight loss is due to water loss from the produce through transpiration and substrate breakdown during respiration (Rathore *et al.*, 2007). Cold-stored packed fruits had significantly lower weight loss on each sampling day. In cold-stored 'Apple' mango, the low physiological weight loss (5.93% on day 35) was further reduced under MAP where 4.19% loss was reported on day 40. Reduced rate of physiological weight loss in packed cold stored fruits is attributed to reduced water loss via

transpiration due to the low water permeability of the polymeric films used to make the Activebag® (Batu and Thompson, 1998). Modified atmosphere packaging (MAP) generates high relative humidity and water vapor pressure around the produce hence reducing water vapor pressure deficit (Serrano *et al.*, 2006). However, at high temperatures the film becomes more water permeable hence allowing more water to be lost via transpiration thus increased weight loss (Irtwange, 2006). With low respiration rate, there is slow substrate breakdown hence slower physiological weight loss Lower respiratory activity under MAP has been previously reported in mango (Githiga *et al.*, 2015; Ouma, 2015), passion fruit (Yumbya, *et al.*, 2014) and grapes (Martinez *et al.*, 2006).

A gradual decrease in mango peel and flesh firmness was observed irrespective of storage condition or packaging. Cold stored fruits packed in Activebag® however remained firmer than fruits in all other treatments. Decrease in firmness during ripening is attributed to depolimerization of cell wall pectins to soluble ones. Enzymes involved in firmness reduction include polygalacturonase (PG), pectin methylesterase (PME), xylanase and cellulase (Ahmad *et al.*, 2013). The slower reduction in firmness in cold stored packed fruits is attributed to the combined effect of low temperature and oxygen levels which reduced the activity of enzymes involved in fruit softening especially polygalacturonase (Lazan *et al.*, 1990). The slower softening can also be linked to the lower rate of moisture loss in packed fruits due to the reduced transpiration rate. This might explain the retained firmness in packaged fruit as the cells possibly remained more turgid resulting in firmer fruits than those which were unpacked (Lazan *et al.*, 1993; Irtwange, 2006). The findings are similar to those observed in a study on avocado (Hershkovitz *et al.*, 2005), banana (Isaac *et al.*, 2007) and mango (Githiga *et al.*, 2015).

Peel and flesh colour (hue angle) decreased as ripening progressed irrespective of storage conditions. Fruits stored at ambient conditions had the highest rate of colour change

from green to yellow compared to fruits stored in the cold room. Packaging delayed and retarded colour change in both storage conditions. The findings are similar to those reported on a study in papaya (Rohani *et al.*, 1997), mango (Pesis *et al.*, 2002; Githiga *et al.*, 2015) and loquat (Amoros *et al.*, 2008). Colour change in fruits has been attributed to degradation of chlorophyll (green colouring matter) by enzyme chlorophyllase and synthesis of carotenoids (Blackenship *et al.*, 2003). Poor color development in packed fruits is attributed to a reduced rate of metabolic activities in the fruit related to chlorophyll degradation and biosynthesis of carotenoids, the color pigments (Mathooko, 2003). The findings are similar to those of Ding *et al.*, (2002) where poor colour development was observed in loquats subjected to MAP.

Total soluble solids (TSS) increased gradually in all fruits during ripening irrespective of storage conditions or packaging. This is attributed to an increase in breakdown of starch into simple sugars as ripening progressed (Siddiqui *et al.*, 2010). Packaging cold-stored fruits in Activebag® further reduced rate of increase of TSS. This can be attributed to low enzymatic activity (due to low temperatures) and retarded metabolic activities due to low O₂ and high CO₂ concentration (Mathooko *et al.*, 2003). High rate of TSS increase in unpacked fruits is attributed to high respiration rate hence the need for respiratory substrates to provide energy for metabolic activities (Saranwong *et al.*, 2003). The faster increase in TSS for fruits stored at ambient room conditions are similar to those obtained in a study on papaya (Azene *et al.*, 2014) and passion (Yumbya *et al.*, 2014). In contrast, total titratable acidity (TTA) in all fruits decreased gradually as ripening progressed irrespective of the storage conditions or packaging. However, cold stored fruits that were packed in Activebag® had delayed rate of reduction of TTA. Reduction of TTA with ripening is attributed to use of organic acid (dominant acid in mangoes is citric acid) as a substrate in respiration (Girdardi *et al.*, 2005). High temperatures increase enzymatic processes leading to rapid biochemical breakdown of

fruits (Yoshida *et al.*, 1984). The retarded TTA reduction in cold stored packed fruits is attributed to low respiration rate due to the modified atmosphere around the fruit in the package (low O₂ and high CO₂ concentrations) and low temperatures which enhance decreased metabolism activities hence reduced loss of respiratory substrates, citric acid being one of them (Girardi *et al.*, 2005). These findings (retarded TTA reduction in cold-stored fruits) concur with past studies in papaya (Rohani *et al.*, 1997; Singh and Rao., 2005) plums (Diaz, 2011) and purple passion fruits (Yumbya *et al.*, 2014).

Beta carotene content increased gradually in all fruits as ripening progressed. The increase in beta-carotene content was slower under cold storage and MAP. Low levels of beta carotene content in cold stored fruits under MAP has been attributed to interference of carotenoids biosynthesis due to the low O₂ and high CO₂ conditions (Artes et al., 2006) and reduced enzymatic processes by low temperature (Jarimopas and Kitthawee, 2007). Ascorbic acid (Vitamin C) decreased gradually in all fruits as ripening progressed irrespective of storage conditions or packaging. This could be attributed to its enzymatic oxidation which is aggravated by poor storage conditions (Appiah et al., 2011). A combination of cold storage and MAP synergistically reduced the decrease in Vitamin C content. This is attributed to low enzymatic activity due to low temperatures and reduced enzymatic oxidation due to altered gaseous composition in the package (low O2 and high CO2 concentration). High vitamin C retention observed in cold stored fruits subjected to MAP is also attributed to its water soluble nature. Under MAP, there was reduced transpiration water loss which may have translated to retention of high ascorbic acid levels. Valero and Serrano (2010) showed a positive correlation between transpiration water loss and rate of reduction of ascorbic acid. A study by Aina (1990) showed susceptibility of ascorbic acid to oxidation at ambient storage conditions and to transpiration loss. Previous studies have shown higher Vitamin C retention

in cold-stored fruits subjected to MAP in papaya (Singh and Rao, 2005) and in mango (Thanaa et al., 2011).

Major sugars (fructose, sucrose and glucose) followed a similar trend as that of total soluble solids. A gradual increase was observed as ripening progressed in all fruits irrespective of storage conditions or packaging. However, the sugars remained relatively low in cold stored packed fruits compared to unpacked fruits. An increase in sugar levels as ripening progressed is attributed to breakdown of carbohydrates into simple sugars during respiration (Siddiqui and Dhua, 2010). The significantly low sugar level in cold stored packed fruits is attributed to the low rate of respiration which is due to low storage temperatures (Kundan *et al.*, 2010) and Modified Atmosphere Packaging (Saranwong *et al.*, 2003). The results are similar to findings of Marero and Kader (2006) in pineapples, in loquats (Amoros *et al.*, 2008), mangoes (Githiga *et al.*, 2015) and passion (Yumbya *et al.*, 2014).

In conclusion, the findings showed that the Coolbot gadget was effective in lowering and maintaining low temperatures in an insulated room fitted with air conditioner during the storage period. Due to low temperatures in the Coolbot cold room, fruits' shelf life was extended and quality preserved as evidenced in instrumental analysis. Coolbot™ cold storage and MAP had a synergistic effect in extending shelf life and maintaining post harvest quality of Ngowe and Apple mango fruits by slowing down ripening and other deteriorative processes. Cold-stored packed mango fruits had longest shelf life of 35 and 40 days compared to the 25 and 35 days for unpacked 'Ngowe' and 'Apple' mangoes respectively. Fruits stored at ambient room conditions had the lowest shelf life of 9 and 12 days for 'Ngowe' and 'Apple' mangoes respectively. The technology can therefore be promoted as a low-cost alternative cold storage option for adoption by smallholder farmers.

5.2. RECOMMENDATIONS

- The Coolbot[™] cold storage technology has been found to be effective in extending shelf life and preserving quality of mango fruits.
 - O It is recommended for adoption by small holder farmers so as to reduce the high post harvest losses they incur. Farmers will also be able to store their produce longer after harvest when there is market glut and sell when the prices go up hence increasing their profit margin.
 - Research should be done on other perishable commodities and their response to CoolbotTM cold storage documented for wider use of the technology.
 - Other technologies (such as waxing) used to extend shelf life of perishable commodities should be tested in combination with CoolbotTM cold storage to evaluate their synergistic effect in extending shelf life and quality preservation.
- 2. Since temperature is critical in reducing the high post harvest losses in most perishable commodities, capacity building should be enhanced among small holder farmers on best possible ways to reduce temperature abuse on harvested produce.

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