# Postharvest treatments to reduce chilling injury symptoms in stored mangoes.

By

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A thesis submitted to Graduate and Post-Doctoral Studies Office in partial fulfillment of the requirements for the degree of Master of Science

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### **ABSTRACT**

#### AZRA TASNEEM

#### M. Sc.(Bioresource Engineering)

# POSTHARVEST TREATMENTS TO REDUCE CHILLING INJURY SYMPTOMS IN STORED MANGOES.

The market life of many fruits and vegetables can be extended through storage at low temperatures. Chilling injury (CI) is a major postharvest storage problem for tropical commodities. Storing these products at temperatures below their critical temperature may result in severe physiological disorders known as CI symptoms. Mangoes (*Mangifera indica*. L) are susceptible to CI when stored below 12 °C. Visual CI symptoms include uneven ripening, surface pitting, discoloration, shriveling and scalding. Research has been conducted to overcome these serious problems using various postharvest treatments such as hot water, methyl jasmonate (MJ) or diphenylamine (DPA) with some reduction of the incidence of CI symptoms in fruits and vegetables.

Experiments were performed to assess and compare the potential of the abovementioned postharvest treatments to reduce the CI symptoms on mango cv. Kent. The obtained results indicated that MJ- and DPA-treatments gave significantly greater percentage of marketable fruits.

Experiments were also conducted with mangoes cv. Tommy Atkins treated with MJ and DPA before storing at low temperatures (1, 4, 7 and 10°C). The chemical treatments were successful at reducing CI symptoms of mangoes. Fruit decay was reduced during subsequent ripening. MJ-treated fruits had lower mass loss and higher total soluble solids (TSS) than the control treatment. The overall quality of MJ- and DPA-treated fruits was good with lower surface pitting and scalding compared with the control treatment. The best results were obtained at storage temperatures of 7 and 10°C. Both MJ and DPA postharvest treatments can reduce CI symptoms in mangoes cvs. Kent and Tommy Atkins when the mangoes are stored at below critical temperatures.

# RÉSUMÉ

# Traitements post-récolte visant à réduire les accidents de réfrigération lors de l'entreposage de mangues

La durée de conservation de nombreux fruits et légumes frais peut être prolongée par un entreposage réfrigéré. Dans le cas de produits tropicaux, des altérations causées par la réfrigération peuvent survenir et représentés un problème lors de la mise en marché. Ces produits végétaux conservés à l'état réfrigéré, à une température trop basse, peuvent développer des troubles physiologiques sévères. La mangue (*Mangifera indica*. L), est sujette aux accidents de réfrigération lorsqu'elle est entreposée à moins de 12 °C. Les symptômes communs de ces accidents de réfrigération sont une dysharmonie du métabolisme, le brunissement, et des dépressions de la peau qui limitent la valeur marchande des fruits endommagés. Ils existent quelques traitements post-récolte, ayant prouvé leur efficacité à réduire l'intensité des accidents de réfrigération de la mangue, tels l'eau chaude, le méthyle jasmonique, et la diphénylamine.

Des essais ont été effectués pour déterminer et comparer le potentiel de ces différents traitements à réduire l'intensité des accidents de réfrigération sur la mangue cv. Kent. Les résultats ont démontré que les traitements chimiques de méthyle jasmonique et de diphénylamine ont conservé un plus haut pourcentage de fruits de bonne qualité. Une autre série d'essais a été effectuée avec des mangues cv. Tommy Atkins, et les traitements chimiques avant entreposage de méthyle jasmonique et de diphénylamine ont tout deux réduit les accidents de réfrigération pour des températures d'entreposage de 1, 4, 7 et 10 °C. Le traitements de fruits au méthyle jasmonique a limité leur perte massique, et a donné un total de solides plus élevé que les fruits témoins. En général, la qualité des mangues traitées au méthyle jasmonique et au diphénylamine était supérieure à la qualité des fruits non-traités, avec les meilleurs résultats obtenus pour les températures d'entreposage de 7 et 10 °C.

Les résultats démontrent que les deux traitements chimiques, le méthyle jasmonique et la diphénylamine peuvent réduire les symptômes d'accidents de réfrigération pour les mangues cvs. Kent et Tommy Atkins.

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## FORMAT OF THESIS

This thesis is written in manuscript-based format, which is suitable for journal publication with slight amendments. The thesis format has been approved by the Faculty of Graduate Studies and Research, McGill University, as outlined in the "Thesis preparation and Submission Guidelines", Section I: Thesis preparation, Part C: Manuscript-based thesis:

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- (c) an introduction which clearly states the rational and objectives of the research;
- (d) a comprehensive review of the literature (in addition to that covered in the introduction to each paper);
- (e) a final conclusion and summary;
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## **CONTRIBUTION OF AUTHORS**

The reported work, which is presented here, was performed by the candidate and supervised by Dr. G. S. V. Raghavan of the Department of Bioresources Engineering, Macdonald Campus of McGill University, Montreal. Y. Gariépy, professional associate and Dr. V. Orsat, research associate, helped during the research project and in the writing of the thesis. Dr. Donald Smith, Department of Plant Science, Macdonald Campus of McGill University, Montreal contributed by providing information which helped during the research project. The entire research project was conducted in the Department of Bioresources Engineering, Macdonald Campus, McGill University. Montreal.

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# ABBREVIATIONS

a*	Chromacity coordinate (redness or greenness)	
ABA	Abscisic acid	
ACO	1-aminocyclopropane cycloxidase	
ACS	1-aminocyclopropane-1-carboxylic acid synthase	
b*	Chromacity coordinate (blueness or yellowness)	
C*	Chroma	
°C	degrees Celsius (unit for temperature measurement)	
$CaCl_2$	Calcium chloride	
CA	Controlled atmosphere	
CI	Chilling injury	
$C_2H_4$	Ethylene gas	
$CO_2$	Carbon dioxide gas	
CPLW	Cumulative physiological loss in weight	
CTs	Conjugated trienes	
DACP	Diazocyclopentadiene	
DPA	Diphenylamine	
FHAT	Forced hot-air treatment	
Fmax	Maximum force (N)	
g	Grams (unit of Mass)	
GA <sub>3</sub>	Gibberellic acid	
Gy	Gray (radioactivity unit)	
HSP	Heat shock protein	
hrs	Hours (unit of time)	
HW	Hot water	
HWT	Hot water immersion treatment	
JA	Jasmonic acid	
Krad	Kilorad (unit of absorbed radiation dose per unit mass)	
L*	Chromacity coefficient (lightness)	
MA	Modified atmosphere	

MCP	1-methylcyclopropene
Min	Minutes (unit of time)
MJ	Methyl Jasmonate
Ν	Newton (unit of force)
PVC	Polyvinyl chloride
RA	Regular atmosphere
RH	Relative humidity
RR	Respiration rate
SA	Salicylic acid
VHT	Vapor heat treatment
TSS	Total soluble solid

## I. GENERAL INTRODUCTION

#### **1.1 Introduction:**

According to FAO, United Nation report 2002, the world agricultural growth has slowed down from an average 2.2 per cent annually over the past 30 years to 1.5 per cent year until 2030. On the other hand, the world population growth will be growing at an average of 1.1 per cent a year up to 2030, compared to 1.7 per cent annually over the past 30 years. This will put an increased pressure on the land to produce more food to fulfill the requirement of the growing population all over the world. Fruit and vegetable production is lower than grain production, however, they contribute important nutrients to the diet, including vitamins A and C, folic acid, potassium, and dietary fiber (Mukherjee, 1997). In 1970, vegetables were considered as one of the leading sources for vitamin C and were contributing 50 % of the supply, while fruits contributed 39 %. By 1994, fruits contribution of vitamin C increased contributing 44 % of the international supply. Fruits contribute 12 % of the folic acid, a nutrient that is known to reduce the risk of heart disease and cancer. They also contribute 12 % of potassium, a mineral with beneficial effects on blood pressure (Putnam and Allshouse, 1997). Consumers tend to prefer fresh fruits and vegetables rather than processed or canned food and postharvest losses of fresh fruits and vegetables are one of the major problem of the food industry (Borrud et al., 1996). Surveys have revealed that a substantial portion of the harvest is wasted annually due to improper harvesting and postharvest practices, disease and lack of facilities and technology to extend storage life. Postharvest losses have been estimated in developed countries to range from 5-25 % while in developing countries it is 20-50 %, depending upon the commodity (Kader, 1992). This continues to cause heavy losses in revenue for the grower, wholesaler, retailers and exporters.

Prior to consumption it is extremely important for local and export markets to reduce the losses and retain high quality and freshness of harvested products. Furthermore, the export of fruits to distant markets needs special technology that ensures that the consumer receives a high quality product and value for their money. Mangoes (*Mangifera indica* L.) are considered as one of the choice subtropical fruit crops of the world due to their attractive color, delicious taste and nutritive value. The problem, which

many international traders are facing, is that the fruits are not evenly mature in a single consignment. Mango fruit have a short growing season and storage life, and the fruit prices after the seasonal peak are very high so it is not affordable for its consumers. Storage is essential for extending the consumption period of fruits, regulating their supply to the market and also for long distance transportation. Mangoes can be stored for 2-3 weeks in good condition at low temperatures. However, chilling injury (CI) is a problem associated with low temperature storage of mangoes. Some symptoms of CI, such as pitting, discoloration, internal breakdown, and decay, can result in large postharvest losses during storage. Therefore, it is important to establish techniques that can reduce CI of the fruits. We have found that several techniques are available to either reduce the development of CI symptoms or increase the resistance to CI. These techniques include low temperature conditioning, heat-treatment, intermittent warming, controlled atmosphere storage, treatments with calcium, chemicals, waxing, film packaging, genetic modification, and applications with ethylene, abscisic acid, methyl jasmonate (MJ), polyamines, or other natural compounds.

Studies have been done to minimize the incidence of CI symptoms in mango fruits. It is known that hot water (HW), MJ and diphenylamine (DPA) can reduce the CI symptoms in different fruits (Shellie and Mangan, 1994; Smock, 1961). Papers are available on MJ-treatment with mango (González-Aguilar et al., 2000; Buta and Moline, 1998) and on DPA-treatment on various fruits (Purvis, 2002; Whitaker, 2000). DPA is commercially used for preventing scald in pear and apples but not much work has been done with respect to DPA-treatments on mango fruit.

#### **1.2 Hypothesis**

This work is focused on the evaluation and the comparison of different postharvest treatments to alleviate the incidence of CI symptoms of mango fruits when stored at low temperature and to determine the minimum temperature at which mangoes can be safely stored. The hypothesis is that if the fruits were treated with the appropriate concentration of a growth regulator (MJ) or antioxidant (DPA) prior to storage at below the critical temperature, incidence of CI symptoms can be reduced and the shelf life of the fruits can be extended. Furthermore, it is also hypothesized that MJ-treatment will enhance fruit color development of the mango cv. Kent.

#### **1.3 Objectives**

The abovementioned hypotheses can be verified through the following objectives:

- 1. To evaluate the potential of postharvest heat-, MJ- and DPA-treatments on the alleviation of the CI symptoms in mangoes cvs. Kent and Tommy Atkins stored at temperatures below the recommended safe level.
- 2. To determine the temperature at which mangoes can be safely stored after heat-, MJ- and DPA-treatments with minimal CI symptoms.

#### 1.4 Scope

This study is based on mango fruits cvs. Kent and Tommy Atkins. Further research is required to find out the exact temperature at which CI will occur and what factors are responsible. Likely the applied method and postharvest CI-treatments can also be applicable to other cultivars and to other fruits. Further work with several mango cultivars is needed before such chemicals can be recommended for commercial use. More research is required to assess the various storage and ripening techniques.

# **II. LITERATURE REVIEW**

Mango (*Mangifera indica* L.) is an evergreen tree grown throughout subtropical and tropical regions. It is considered to be the oldest and best fruit in the world market (Singh, 1960). It belongs to the family Anacardiaceae which has 75 genera and 700 species (Lizada, 1993). The world production rate is roughly about 17 million tons over an area of two million ha in the tropics (Chadha and Pal. 1993). The origin of Mango was traced far back in the Indo-Burma region, and has been cultivated in India for over 4000 years and more than 1000 varieties are known there (Mukherjee, 1953). The countries with major production that export mangoes throughout the world are Philippines, Thailand, Mexico, and India. Tommy Atkins is one of the dominant cultivars in South America (Galan, 1993). The most appreciated cultivars in India are Dasheri, Chawsa, Alphanso and Totapari (Simon, 1970).

#### **2.1 Cultivation Characteristics**

Mango will grow in a slightly acidic (5.5-7.5) and well-drained soil, whether it is sandy, loam or clay (Young et al., 1965). It is somewhat tolerant to alkalinity (Kadman et al., 1976). Mango is also drought-tolerant, and can withstand occasional flooding (Singh, 1960). For best flowering and fruit set, good timely rainfall is necessary rather than the total rain fall. Temperature plays an important role in mango flowering and its influence varies with cultivars (Schaffer et al., 1994). Temperatures in the range of 24-30 °C are required for best flowering; however, during fruit development if sufficient water is provided the tree can withstand up to 48 °C. Flower deformation and loss of pollen viability can occur at low temperatures (Popenoe, 1957; Issarakraisila et al., 1992). Cold temperatures can also limit the growth of the plant and can cause damage or even kill young trees; while it has been reported that older trees can endure up to -4 °C for a few hours with limited damage (Crane and Campbell, 1991).

#### 2.1.1 Chemical composition, nutritive and medicinal value

Mango is usually called the king of tropical fruits. It contains high levels of vitamins, proteins, lipids, amino acids and minerals as shown in Table 2.1. Chinese traditional medicine believes that mango can produce saliva and eliminate fever; it is good for the stomach disorders; and can break kidney and gall stones. Mango juice improves the nervous system and can be used to treat mental illness. Mango is named as divine food (Kulkarni and Rameshwar, 1981; Doreyapa Gowda and Ramanjaneya, 1994).

Ingredients	Quantity
Water	83.9 g
Energy	234 kJ
Protein	0.3 g
Fat	0.1 g
Carbohydrate	15.0 g
Fiber	0.5 g
Ash	0.3 mg
Ca	8.0 mg
Fe	0.2 mg
Mg	12.0 mg
Р	10.0 mg
K	158.0 mg
Zn	0.1 mg
Vitamin C, ascorbic acid	15.1 mg
Riboflavin	0.1 mg
Niacin	0.3 mg
Pantothenic acid	0.2 mg
Vitamin B-6	0.1 mg
Vitamin A	3813 IU

**Table 2.1**: Nutritional value of mango cv. Haden per 100 gram of the edible part<br/>(Wenkam, 1990).

#### 2.1.2 Pests and diseases

Mango is prone to a number of diseases at any stage of its development. Hence, plants in the nursery and the fruits in storage or transit are susceptible to disease. In many cases these diseases are due to mismanagement leading to infections at the harvesting time. In such cases, prevention can be done by using chemical spray.

Bacteria, fungus, and flies are mainly responsible for the onset of diseases. Anthracnose disease caused by *Colletotrichum gloeosporioides* is one of the major common diseases for pre-and postharvested fruits and is associated with high rain fall and humidity (Fitzell and Peak, 1984; Jeffries et al., 1990). Alternaria rot, or black spot is another postharvest fruit disease, which infects fruits during ripening. Anthracnose (Figure 2.1) Stem end rot (Figure 2.2), black mould rot, bacterial black spot are common postharvest fruit diseases which can be prevented by using certain chemicals, or proper postharvest fruit treatment.

There are a number of fungi which attack mango fruit at their mature stage during storage and transit: *Pestalotiopsis Mangifera* (Figure 2.3), *Colletotrichum gloeosporioides* (Figure 2.4), *Ceratocystis fimbriata*, Gloeosporium species, *Dothiorella ribis*, *Penicillium*, and Cladiosporium are some fungi which commonly infect mango fruits. *Bactrocera dorsalis*, *Ceratitis capitata*, *Anastrepha suspense* and *Diaschamimorpha longicaudata* are common flies which can damage fruit.

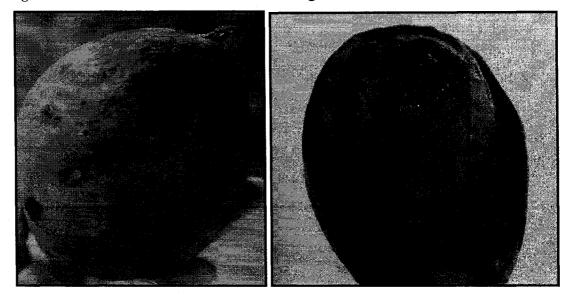


Figure 2.1: Tear shape pattern of anthracnose in mango (from APS digital image collection, 2001)

**Figure 2.2**: Stem end rot disease in mango. (from APS digital image collection, 2001)

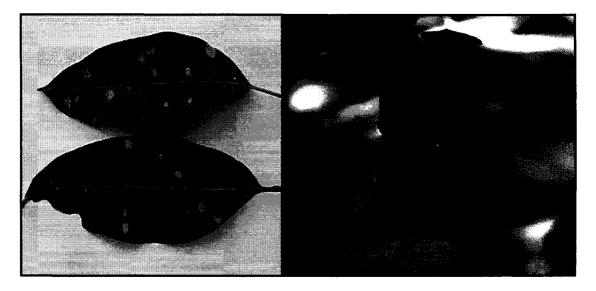


Figure 2.3: Whitish gray lesion on mango leaf caused by *Pestalotiopsis*. *mangifera* (Litz, 1997).

Figure 2.4: Typical anthracnose lesion on mango leaf caused by *Colletotrichum gloeo*sporioides (Litz, 1997).

For fresh mango fruit to be accepted by market, it has to be treated to ensure that it is free of fruit flies. Disinfestations can be done by chemical fumigation, or by nonchemical treatment, which consists of heating the fruits to specific temperature and maintaining this temperature for a defined period of time which could kill fly larvae and eggs. Vapour heat treatment (VHT), forced hot air treatment (FHAT), and hot water (HW) immersion are commonly used heat treatments for mango fruit (Esguerra and Lizada, 1990).

#### 2.1.3 Harvesting

A good system of transport is necessary to develop an industry or market; it means a channel by which the consumer receives the commodity under best condition with minimum cost. Fruits are usually very perishable and mango is one of them. Their market value is affected if they receive any slight injury during their picking, packaging, and stocking in transit or on retail display. Mangoes are usually harvested green. Harvesting usually takes place after 15-16 weeks of fruit set when they are physiologically matured (Lakshminarayana et al., 1970). Later harvesting may result in uneven ripening, and can lower sugar to acid ratio. There is no particular parameter for judgment of fruit maturity; it depends on mango type, variety, production conditions, and location. Physical, chemical, and physiological parameters are used to define the maturity stage for harvesting of fruits. Useful chemical parameters are acidity, soluble solid content, phenolic constituents, and carbohydrate content. Physical parameters are size, shape, surface color, pit around the pedicel, lenticels and specific gravity (Popenoe and Long, 1957; Krishnamurthy and Subramanyam, 1970; Ketsa et al., 1991). Physiological maturity shows changes in the pulp color, breaking to yellow, hence it can be tested by slicing a fruit before harvesting. An ancient advice for mango harvesting says that, when first fruits begin to drop, the crop is ready for picking.

Usually harvesting is done by hand. Sometimes a long picking pole, ladder or hydraulic lifts are used to pick the fruits from tall trees (Singh, 1960). Picking of fruits begins from the lower side of the tree just to avoid sap oozing on the fruit below. At the time when mango is fully-grown and ready for picking, the stem will snap easily with a slight pull. If a strong pull is necessary, the fruit is still fairly immature and harvesting should be delayed (Ram Prakash, 1998). Some cultivars require multiple picking as not all the fruits mature at the same time. Usually fruits are picked with approximately 4-inch (10 cm) stem to avoid the spurt of milky/resinous sap; because in some cultivars it causes sap burn on skin of any fruit with which they comes into contact (Waskar et al., 1997). The fruits are then placed in field crates and after desapping, which can be done by breaking off the fruit stalk; they are placed stem-end-down to cease the flowing sap.



**Figure 2.5**: Picking of mango fruit by hand. (from PREDA, 2004. available at www.preda.org/fruitbiz.htm)

Before marketing, they are either allowed to ripen naturally, or with ethylene gas. After picking, the fruits are packed and graded according to their size, color and free of defect or injury. To prevent fruits from bruising they are packed in single or doublelayered cartons, which contain protective material.

#### **2.2 Storage of Fruit**

The basic concept of storage is to extend the shelf life of products by storing them in appropriate conditions to maintain their availability to consumers and processing industries in their usable form. They can either be stored naturally in the field, or in built storages (Raghavan and Gariépy, 1985 and Pantastico et al., 1975). In natural storage the product is left in the field and harvesting is delayed, while in artificial storage favorable conditions are provided which help to maintain product freshness and nutritional quality for a longer period. During storage, the mango physiology and its ripening involves many physiochemical activities, such as cumulative physiological loss in weight (CPLW) and volume, pulp and skin color change, acidity, loss in firmness, increase in total solids and sugar concentration.

According to FAO Year Book, 2000 the world import of fresh mango are projected to increase by 53 % to 459,000 tonnes by year 2005 and it can further be increased by improving storage conditions and controlling diseases and insect contamination. There are various techniques, which have been developed to improve the storage life and maintain the quality of fresh horticultural commodities.

#### **2.2.1.** Storage at low temperature:

International trade of fresh mango has been limited because of its highly perishable nature and susceptibility to postharvest disease and injury and hence mangoes are still considered as luxurious and expensive item in the markets of many industrialized countries. While storing the commodities there are many factors which influence product quality; temperature is one of them. For successful storage it is necessary to efficiently control the temperature throughout the storage period.

The principle behind cold storage is to delay the period of ripening of a product by slowing down its physiological activities. While storing fruits the first priority is to

maintain the quality of the product. Many commodities when stored at low temperature are subjected to damage caused by chilling which will promote fungus and diseases. Mangoes are tropical fruits and are therefore sensitive to chilling when stored below a critical minimum temperature (Chaplin et al., 1991; Lizada, 1991). If stored at low temperatures for prolonged time, storage could have an effect on ripening. It has been reported that cvs. Langra and Dasheri can safely be stored at 7-8 °C for up to 25 days (Mann and Singh, 1976). However, recommended temperatures are in the range of 10-15 °C and lead to storage life of 2 to 3 weeks. The best ripening temperature ranges from 21-24 °C. But at high temperature of 32 °C, the ripening process is retarded. Other essential factors are 98 % to 100 % relative humidity, and atmospheric pressure of 76 or 152 mm of Hg.

#### **2.2.2 Controlled and modified atmosphere storage:**

Respiration is the major physiological activity of concern in postharvest storage. It is a metabolic process that occurs continuously in all living cells. Respiration is the oxidative breakdown of complex material such as starch, sugar, and other organic compounds into simple molecules such as carbon dioxide, water and energy as represented by the following Equation (1) (Phan et al., 1975).

$$C_6 H_{12} O_6 + 6O_2 = 6CO_2 + 6H_2 O + Energy$$
(1)

Respiration rate (RR) is usually expressed in terms of  $O_2$  consumed and  $CO_2$  produced or heat released and it is expressed as mg.kg<sup>-1</sup>.h<sup>-1</sup>. RR can be lowered by lowering the temperature. During the postharvest life of the product, the respiration provides defence mechanism against spoilage. However higher RR, leads to faster deterioration of fruits and shortening of storage life.

By elevating carbon dioxide and lowering oxygen amount surrounding the product, there will be a decrease in respiration and a decrease in the rate of ethylene production. In closed storage, the respiration will simultaneously lead to build-up of carbon dioxide and depletion of oxygen. If  $O_2$  level goes too low it will create anaerobic conditions, which results in fermentation and unwanted by- products that can cause damage to certain products. If mango fruits are stored at 1 %  $O_2$  and 15 %  $CO_2$  level

condition, off-flavor and skin discoloration of fruits will occur (Hatton and Reeder, 1966).

The basic idea of Controlled Atmosphere Storage (CA) is to maintain the best product quality. This can be accomplished by keeping  $CO_2$ ,  $O_2$  and Ethylene gases at predetermined levels (gas levels differ depending on the type of fruit being stored). Usually decreased  $O_2$  and increased  $CO_2$  levels at a low temperature with high RH are suitable for stored commodities. CA can provide an effective storage environment for different fruits and vegetables (Raghavan et al., 2003, 1984; Bender et al., 2000; Gariépy and Raghavan, 1991, 1986, 1984; Kader 1986).

Modified atmosphere (MA) is referred to as a relationship between product respiration and gas exchange within any form of structural enclosure. MA storage technique can be used to maintain the postharvest quality of different fruits (Ding et al., 2002; Rodov et al., 2002; Meir et al., 1998; Illeperuma and Jayasuriya, 2002; Prabhanjan et al., 1992). MA packaging inhibited the mango ripening process (Sornsrivichai et al., 1989, 1992). In 1994, Yantarasri et al., found that film perforation delay softening and can reduce the weight loss of mango cv. Nam Doc Mai.

Diffusion channel method is a system where gas diffuses in a storage chamber, through a tube, that connects the ambient air with the chamber atmosphere. Several researchers (Ratti et al., 1998; Baugerod, 1980; Ramachandra, 1995) have successfully demonstrated that by using diffusion channels the gas exchange capability and the maintenance of equilibrium  $O_2$  level can be achieved over a long period of time. The advantage of these systems is that pressure and temperature fluctuations do not significantly affect the system. It should be noted that studies related to mango storage using diffusion channels is not available in the literature. There is scope to study this technique for extending shelf life of mangoes.

#### 2.2.3 Storage at low pressure

Low pressure storage, also called hypobaric storage, deals with the control of air pressure, temperature and humidity. The main principle of low pressure is that the pressure of the chamber is directly proportional to the  $O_2$  level. With a pressure decrease, the amount of  $O_2$  decreases reducing the respiration rate of the product. This system easily

maintains  $O_2$  and relative humidity. The advantage of this system is that it can easily remove the metabolites. But this system cannot be used commercially as it has certain construction disadvantages with airtight chambers being costly. Furthermore, this storage provides unsatisfactory ripening and poor aroma and flavor to the fruits (Ramachandra, 1995

Spalding and Reeder, 1997 noted that when cvs. Tommy Atkins and Kent were stored at 13 °C at a pressure of 76-152 mm Hg with a relative humidity of 98-100 % for up to 3 weeks, a greater percentage of market acceptable fruits were obtained; and when they were placed under normal pressure they took a longer time for ripening as compared with those stored at 760 mm Hg. The fruits were more greenish in color compared to those stored at normal pressure.

#### 2.2.4 Storage by use of coating

Waxes are commercially used to reduce the moisture loss from the fruits. Aqueous wax emulsion consisting of mineral petroleum like paraffin and vegetable waxes with or without emulsifier are commonly used to increase the storage life of mangoes (Dalal et al., 1971). However, coating of mango with refined mineral oil resulted in fruit injury (Mathur and Srivastava, 1955). Oil coating decreased the respiration more than wax coating and results in severe anaerobic condition that injured the fruit.

Fungicidal wax, wax emulsion containing hydrazide, maleic and polysaccharidebased coating also delays ripening process. Selective films like Polyvinyl chloride (PVC) films also prolong shelf life of mango fruit (Ketsa and Raksritong, 1992). The wax can be applied by roller brushes in a specifically designed wax applicator or by hand. Dipping in wax is to be avoided, and a uniform application of wax is necessary otherwise some fruits receive too much wax and others too little. Before application of wax fruits must be dried, otherwise foaming of water-emulsion waxes may occur.

#### 2.2.5 Storage by using ionizing radiation

For safe storage of fresh fruits and vegetables, the US Food and Drug Administration approved the use of irradiation at a dose of 100 Krad (United States Food and Drug Administration, 1986). Irradiation includes use of ionizing energy such as gamma rays, electrons, X-rays and microwaves. Spalding and von Windeguth (1988) reported that the percentage of decayed mango is minimized when they are exposed to 750 Gy or higher, but fruit peel shows scald-like symptoms with irradiation doses over 500 Gy.

Radiation also affected ripening, because of the specific biochemical processes. According to Spalding and Reeder (1986), combination of irradiation and HW (53 °C), or 0.2 % hot imazalil (53 °C) was more effective for storage purposes. Gamma irradiation (30 Krad) caused ripening delay of seven days in comparison to mango stored at room temperature.

#### 2.2.6 Storage by using chemicals

Treatment efficiency varies with infection level and storage regime. The length of shelf life depends on cultivar, injury, maturity at harvest, calcium spray, and exposure to ethylene (Anonymous, 1988, Coates et al, 1995). A dip in 4-6 % calcium chloride can increase the shelf life of some cultivars (Singh et al., 1993). Ethylene is used to reduce time for ripening initiation and it can also enhance skin color of the fruit (S. P. Burg and Burg, 1962). In South Africa, a benomyl dip for 5 min at 55 °C is recommended just after picking of fruits, which can control soft brown rot (Sepiah, 1986). Prochloraz also provides good protection from anthracnose and Alternaria rot in mango (Johnson and Coates, 1993). Prior to harvest Gibberellic acid (GA3) spray can retard mango ripening at ambient temperature for up to six days of storage (Khader, 1991). Calcium chloride treatment resulted in low ethylene production, low respiration, and helped to reduce the occurrence of storage decay (Eeden, 1992).

#### **2.3 Postharvest Heat Disinfestation Treatments**

For fresh mangoes postharvest heat disinfestation treatments have been well known over the past decades. Although heat has fungicidal and insecticidal action it often has detrimental effect on the crop. The fruit is heated for a defined period of time to ensure the energy gets transferred from the heating medium to the fruit. The heating medium used may be air or water. In 1989, Couey reported that heat-treatments are used to control postharvest diseases and insect pest. This method is cheap and environmentally friendly; so the interest in heat disinfestation has been revived. The temperature and treatment duration depend on commodity and cultivars and must be precise so that it only kills pests without affecting the commodity. Nowadays three heating methods are commonly used for mangoes.

- i. Vapor heat-treatment (VHT)
- ii. Forced hotair-treatment (FHAT)
- iii. Hot water-treatment (HWT)

#### **2.3.1 Vapor heat-treatment: (VHT)**

This is a conductive way of heat transfer and also called as high humidity air heating. In this process, saturated moist air is passed across the fruits when the temperature of fruit is at or below the dew point; condensation of the moisture appears on the fruit surface. The heat from the surface of fruit is then transferred towards the fruit center. It has been noted that the water droplet transfers heat more efficiently than air and allow faster fruit heating. But in this case risk of physical injury increases. Mangoes, imported into the USA and Japan must receive a VHT. Nowadays Japan requires mangoes from Thailand and Philippines to be treated with vapor heat at 46-47 °C for 10 min (Anonymus, 1987).

In ripe fruits, VHT can induce internal breakdown of the inner mesocarp, which is characterized by the presence of white, starchy, and tough lesions. In severe cases, this results in damage of the product because there is depletion of internal oxygen level, which gives fermented unwanted by-products (Esguerra et al., 1990).

#### **2.3.2 Forced hot air-treatment: (FHAT)**

It is also called non-condensing air heating. In this treatment, hot air is passed through a bed of fruits at a specified temperature and this leads to transfer of heat from hot air to cooler fruit via the skin and then goes to the center of fruits. In this treatment the fruit surface remains dry and the RH of the passing air is as low as 30 %. This method has been developed in USA for mango quarantine treatment (Mangan and Ingle, 1992). They reported that hot air-treated fruit whose pulp temperature is over 47 °C is able to kill all stages of West Indian fruit flies. Although this method is not recommended for avocado,

lychee, and nectarine; it has been approved for grapefruit, papaya, and mango. The disadvantage of this treatment is fruit weight loss and shrivelling which occurs due to low air humidity.

#### **2.3.3 Hot water-treatment (HWT)**

Hot water (HW) is an effective heat transfer medium and, within a short time a uniform temperature profile will be maintained (Couey, 1989). The additional benefit of HWT is that it can control postharvest diseases such as anthracnose and stem end rot (Couey 1989; McGuire 1991). This treatment is commonly used for disinfestation of mango from fruit flies (Sharp et al., 1988, 1989; Sharp 1986; Segarra-Carmona et al., 1990; Nascimento et al., 1992). This treatment is cheaper than any other heat-treatment and is also effective on commercial scale in the USA. Recommended temperature ranges are between 43-46 °C; above 46 °C the fruit experiences excessive damage. Usually a single dip procedure is used either in batch process or continuously for 65-90 min depending on fruit size and cultivars.

#### **2.3.4** Physiological responses to heat-treatment

Physiological response to heat treatment in the fruits has been summarized by Jacobi et al., (2001), according to that:

- i) Heat-treatment increases fruit heat tolerance which depends upon a number of factors including species, stage of fruit maturity, fruit size, exposure to different environmental factors, time, duration and type of application.
- When harvested fruits are transferred from ambient growth temperature to an elevated temperature, stress is induced and the impact depends upon length of exposure and temperature difference.
- iii) Heat-treatment also develops external or internal heat injuries in many cultivars.
- iv) Heat-treatment can affect fruits ripening either inhibiting, promoting or disrupting the ripening process.
- v) Heat-treatment accelerates the yellowing of the mango fruit skin and uniformity of skin color is also observed. However, in many cultivars fruit

becomes soft due to heat-treatment. Actual mechanism by which heattreatment accelerated mango fruit ripening is not yet known but it has been hypothesized that it is associated with increased synthesis of carotenoid, degradation of chlorophyll and synthesis of cell wall degrading enzymes.

- vi) Immature mangoes have lower heat tolerance compared to mature ones. When immature mangoes get treated with VHT at 46 °C for 10 min internal breakdown has been noted by Esguerra and Lizada (1990) in the form of spongy white starch tissue in fruit mesocarp; however, no external damaging was noted.
- vii) If fruits and vegetable treated with HW before storage at low temperatures, the treatment reduces the incidence of CI (González et al., 2000b; McCollum et al., 1993).

#### **2.4 Physiological and Physical Disorders**

During ripening, mango fruits are susceptible to several physical and physiological postharvest disorders, which affect fruit quality. Some disorders are inherent while some are induced. The inherent physiological disorders include spongy stem-end disorder, soft nose, and spongy or soft tissue. Best example of induced disorder is CI when commodity is exposed to low temperature.

#### 2.4.1 Sapburn

The stem of picked mango exudes large quantities of latex / sap which has low pH and high oil content and has tendency to burn the fruit skin. According to Joel (1978, 1980) latex repels fruit flies. Terpinolene is the main ingredient which is responsible for skin burn and high nitrogen levels in the fruit cause more severe sap-burn.

#### 2.4.2 Spongy tissue

In the pulp of ripened fruit, a desiccated sponge-like tissue is found which is called a spongy tissue (Amin, 1967). The fruit pulp remains unripe and due to physiological and biochemical disturbances a deposition of non-hydrolyzed starch occurs. Mechanical injury can also be responsible for spongy tissue like symptom in fruits. The

affected fruit has no external symptoms either at the time of picking or while ripening. The affected portion gets prominent when the fruit gets cut. The exact cause of spongy tissues is still unknown. It has been noted that the affected fruit pulp has higher acidity, low pH, low  $\beta$ -carotene content, sugar and ascorbic acid. The amylase and invertase activity is also reduced. In Alphanso mangoes the incidence of spongy tissue may be reduced if fruits are harvested earlier and ethylene is used for subsequent ripening (Lad et al., 1985).

#### 2.4.3 Black-tip

In black-tip disorder, the distal end of the fruit becomes yellow, and mesocarp and seed are unaffected in early stage, while later on the entire tip of the fruit turns brownish black in color (Ram, 1989). The affected portion gets hard and its growth is retarded. The fruit becomes unattractive and loses its quality. Sprays of sodium carbonate, sodium hydroxide and borax can prevent the incidence of black tip in mango.

#### 2.4.4 Soft-nose

In 1957, Young described soft-nose as breakdown of fruit flesh at the fruit apex. The mesocarp shows premature softening at distal end. This disorder may be related to calcium deficiency. Cultivars Kent and Ameeri from Canary Island were found more susceptible to this disease (Galan Sauco et al., 1984). The fruit, which has low calcium content, is the most affected by this disease.

#### 2.4.5 Chilling injury (CI)

To maintain the quality of harvested horticultural crops, temperature is one of the most predominant factor. Rate of metabolic processes of the commodity can be reduced by lowering the temperature. When tropical and subtropical crops are stored at low temperatures, certain physiological disorder have been observed such as CI. Due to this phenomenon, product storage life is limited, and leads to significant degradation of quality.

The primary cause of CI is thought to be the damage of cell membrane that initiates a cascade of secondary reaction. CI is a time and temperature problem. Mango fruits are subjected to CI when stored below 10 °C. The symptoms include grayish scald-

like discoloration of the skin, skin pitting, uneven ripening, and reduction in the level of carotenoids, aroma and flavor during ripening (Abou-Aziz et al., 1976; Hatton et al., 1965; Thomas and Oke, 1983; Wardlaw and Leonard, 1936).

In controlled atmosphere storage enhanced  $CO_2$  accumulation alleviated CI symptoms while low  $O_2$  has no significant effect (O' Hare and Prasad, 1993). Peroxidase, invertase and cellulase activities in the peel of the fruit increased while amylase activity decrease during the development of CI. Freezing injury is different from CI as ice crystals are formed in frozen tissues. When fruits are stored below their freezing point ice crystals are formed which causes cell damage. It has been thought; initially CI damaged the cell membrane, which initiated a cascade of secondary reactions which includes ethylene production, toxic compounds accumulation, reduction in photosynthesis, increased respiration and cellular structural alteration.

CI symptoms are not only due to storage of fruits at low temperatures but also depend on the maturity of harvested fruits and the duration of exposure of commodities to low temperatures and protective packaging of fruits while they are stored at low temperatures (Medlicott et al., 1990). Short heat-treatment at 38 °C is effective in reducing CI in various fruits, including tomato (Lurie and Klein, 1991) and avocado (Sanxter et, al., 1994; Woolf et al., 1995). Heat-treatments such as hot air, HW also reduced CI symptoms in mango although they failed to retard the ripening process (McCollum et al., 1993).

#### **2.5 Post harvest Treatment for Reducing CI Symptoms**

Several techniques have been developed to alleviate the onset of CI symptoms in different products. In 1990, Hatton described three broad categories of temperature management, which included:

- (i) Storage of product at threshold temperatures, so that it will not allow development of CI symptoms.
- (ii) Intermittent warming and cooling.
- (iii) Temperature conditioning.

Intermittent warming procedure can increase the shelf life of a number of products. In a number of crops it has been found useful in minimizing CI stored at lower

temperatures with one or more exposure to non-chilling (higher) temperatures. Rewarming allows metabolism of the toxic products which accumulates in the tissues at low temperature (Pentzer and Heinz, 1954) and also restores certain depleted essential metabolites. Rewarming also allows the revival of oxidative phosphorylation in tissues as it gets suppressed at low temperatures.

Temperature conditioning involves storage of products above the chilling threshold temperature before they are stored at a low temperature (Wade, 1979). This may be a single step operation or may be achieved in multi-steps by gradually decreasing the temperature. However, a single step conditioning is less effective than a multi-step conditioning.

Jacobi et al., (1995, 1996) reported that conditioning of mango fruit cv. Kensington at 40°C increased heat tolerance and reduced the effect of heat injury. According to these reports, those fruits which received HW conditioning treatment at 40 °C for 8-12 h had minimum heat injury and showed internal and external cavities and starchy layer beneath the skin. Heat-treatment effectively increases fruit heat tolerance and reduces CI in stored avocado (Woolf et al., 1995). There are different techniques which are used to minimize the incidence of CI.

#### 2.5.1 Use of heat-treatment

Different heat-treatments have been used to alleviate the incidence of CI symptoms in mango fruits. VHT-treated mango fruits showed increased level of putrescine and its accumulation helped the reduction of CI (Esguera and Lizada, 1990). As stated by González et al. (2000) peppers treated with HW at 53 °C for 4 min, had a reduced incidence of CI after 14 and 28 days storage at 8 °C. It has been hypothesized that HW-treated fruits show increased level of polyamine which reduces CI symptoms in fruits (González, 1997).

Ethylene-treated fruits, irrespective of the method of application, cause severe mesocarp discoloration in avocado fruits. Loss of green color has been observed in plant tissues while they are treated with Ethylene. The treatment also decreases fruit firmness, and CI during prolonged storage (Watada, 1986). The mechanism by which ethylene

regulates these attributes is not yet known. High temperature treatment inhibited ethylene production, which affected fruit ripening process.

Effect of postharvest heat-treatment on fruits are varied, since in some fruits it affects fruit firmness, color development and ethylene production. Chaplin et al., (1982), reported that pre-treatment of avocado fruits at 38 °C for 12 hrs before storage at 0 °C showed reduced symptoms of flesh injury. There are several possible explanation to support the concept that heat-treatments provide protection against CI.

- i) Heat-treatments induce heat shock protein, which provide protection against heat injury, chemical stress as well as from CI.
- ii) It is suggested that exposure of plant tissue to one stress (heat-treatment) provides protection of the plant from another stress.
- iii) Increase in ethylene synthesis means increased level of CI and heattreatments decrease the synthesis of ethylene.

In 1999, Fallik et al., established a HW-brushing treatment in which fruits, moving along with brush roller, received HW-treatment. This treatment was used for disinfestation of fruits and vegetables but it was noted that it induces tolerance to low temperatures in grapefruit cv. Star Ruby.

Modified atmosphere horticultural crop packaging leading to high  $CO_2$  and  $H_2O$  level and low  $O_2$  level protected many chilling sensitive crops against CI (Forney and Lipton, 1990). Further, HWT with controlled atmosphere storage also alleviated CI in 'Fuyu' persimmons.

#### 2.5.2 Use of Chemicals

#### (a) Jasmonic acid and Methyl Jasmonate:

Different chemicals are used before storage of fruits and vegetables at low temperatures to minimize the incidence of CI symptoms. Growth regulators and antioxidant compounds are among them. Abscisic acid (ABA), Salicylic acid (SA) and Jasmonic acid (JA), its methyl ester methyl Jasmonate (MJ) and methyl salicylic acid (MSA) are naturally found in a wide range of higher plants and are able to induce a mechanism that protects plants from CI (González et al., 2001).

SA is a natural signaling molecule, which mediates defense against many pathogens and also plays an essential role in thermogenesis. In mammals prior treatment with SA induces heat shock protein in response to heat stress (Jurivich et al., 1992). JA and MJ are called growth regulators and MJ is more volatile than JA. It is one of the main odor compounds of jasmine. MJ is also a flavor ingredient of semi-black and black tea. Plants that are attacked by insects or mechanically damaged produce higher levels of JA and MJ. Wounded parts release an 18 amino acid polypeptide, systemin, which, activates cell membrane lipase enzyme which ultimately releases linolenic acid, a main precursor of JA and MJ. As these compounds are volatile in nature as such they provide quick signals to neighboring cells and promote them to produce defensive chemicals before they are attacked. When strawberries and papaya treated with MJ (10<sup>-5</sup>M), and stored in MA packaging at 10 °C, loss of firmness, inhibition of fungal decay and reduction in CI has been noted (González-Aguilar et al, 2003). Wang and Buta (1994) reported delay of CI symptoms for 2-4 days in zucchini squash fruits treated with MJ prior to cold storage. MJ also effectively reduced CI in mangoes (González-Aguilar et al., 2000a), grapefruit, bell pepper and avocado (Meir et al., 1996). MJ either applied in gas or vapor form has similar effect on CI incidence.

## (b) Diphenylamine

Postharvest application of Diphenylamine (DPA) is commercially used to control scald production in fruits. During scald production, hypodermic cells (the layer of loose connective tissue immediately deep to the dermis of the skin) are affected at the depth of 40-150  $\mu$ m from the surface. It is an antioxidant for carotene and other unsaturated substances. Carotene synthesis by microorganisms is also decreased by DPA-treatments, which inhibit the dehydrogenation of saturated polyenes to carotenoid. It has variable effect on ester production in stored apples.

Apple and pear surfaces have  $\alpha$ -farnesene hydrocarbon along with its antioxidation products which are mainly responsible for black scalding of fruits after cold storage (Anet, 1972). Anet (1972) reported the oxidation of  $\alpha$ -farnesene and identified two conjugated trienes (CTs) from the skin of stored apples. These CTs are toxic to the fruit tissues as they initiate free-radical mediated chain reaction and decomposition to harmful volatiles (Whitaker, 2000). Treatments with DPA, in combination with low oxygen ( $\leq 1.5$  %) atmosphere are also used for limiting superficial scald in apple fruits.

## **2.6 Ripening of Mangoes**

Mangoes are harvested at the mature green stage and subsequently allowed to ripen. The recommended temperature for ripening of mango fruits ranges from 20-25 °C but it varies according to the variety and origin. Ripening at higher temperatures results in off flavor and spotting of the fruits. During ripening of the mango fruit different occurrences have been observed:

- During ripening of mango fruit a notable peak of ethylene has been noted. Ethylene plays an important role in ripening of fruit, breakdown of carotenoid in the peel and increased respiration is also observed in ripe mangoes along with increased amount of catalase and peroxidase (Mattoo and Modi, 1969; Krishnamurthy and Subramanyam, 1970).
- ii) Hydrolysis of starch and formation of sugar is associated with the ripening process. Main monosaccharides include glucose, fructose and sucrose. Sucrose being the predominant sugar it contributes 57 % of total sugar in ripe mangoes cv. Keitt while fructose and glucose are present at 28 and 15 % respectively (Medlicott and Thompson, 1985).
- iii) Concentration of organic acid decreases as the fruit ripens. Main organic acids include citric acid, glycolic, malic and ascorbic acid. In mango cv. Keitt the predominant acid was found as citric acid while oxalic, tartaric, ascorbic and  $\alpha$ -ketoglutaric acid were also present (Medlicott and Thompson, 1985).
- iv) Peel color of the fruit changes during ripening as chloroplast in the peel is converted into chromoplasts, which has red or yellow pigments, while some cultivars show reddish blush because of anthocyanins, while some remain green (Lizada, 1993). During ripening the carotenoid pigment level also varies among the cultivars. It has been reported that the level of carotenoid increases with a gradual decrease of anthocyanins in mangoes cv. Tommy Atkins (Medlicott et al., 1986).

- v) As the fruit ripens the flesh becomes softer. At the peak of ripening the fruit firmness decreases and softening of the fruit is associated with an increased solubility of cell wall pectins (Roe and Bruemmer, 1981).
- vi) Certain monoterpene hydrocarbons and lactones are responsible for mango flavor (Wilson et al., 1990).
- vii) At the early stage of growth polyphenolic content are high and decrease with the ripening and remain fairly steady (Lakshminarayana et al., 1970).
- viii) During ripening lipid content increases along with increases in glyceride and linolenic acid (Bandyopadhyay and Gholap, 1973)

#### 2.6.1 Ripening of mangoes by using chemicals

Mango fruits ripen unevenly on the tree and natural ripening can be very slow and unpredictable. Hence, to overcome these problems certain chemicals are used to ripen the fruits artificially. Fruits were briefly exposed to ethylene or similar gases like acetylene to initiate the ripening process. Ethylene is known to be a plant hormone that triggers fruit ripening. It has been reported that if ethylene is applied exogenously it helps fruit ripening (Medlicott et al., 1988). Ethylene-treatment is usually given at the packing house or at the point of distribution.

Ethephon is known as one of the most common ethylene-generating chemical and postharvest treatments. Ethephon accelerates ripening and improves the peel color of the mangoes (Lakshminarayana et al., 1975). Use of chemicals promotes the ripening process and improves color development of the fruits. Aside from this, ethylene is an explosive gas and is very expensive. Certain developing countries use calcium carbide as a fruit ripening agent. Acetylene gas is generated from calcium carbide, which initiates the ripening process. This practice is commercially used in Brazil and Senegal (Medlicott, 1986a). Acetylene is also an explosive gas and calcium carbide is more commonly used in welding applications. It is very toxic and has disadvantages compared to ethylene. Calcium carbide is considered as extremely hazardous sometime it contains traces of arsenic and phosphorus hydride, which is harmful for human consumption (Delpierre, 1974). MJ-treatment reduces the CI symptoms of mangoes cv. Kent and enhances the skin color development of the fruit (González-Aguilar et al., 2001).

## **2.7 Quality Evaluation**

Product quality can be evaluated by using different methods. According to Salunkhe et al., (1991) quality characteristics of the product can be divided into three types; sensory, hidden and quantitative. Sensory category includes color, size, shape, defects, gloss, taste and odor. Hidden deals with nutritive value, presence of contaminants and poisonous materials while quantitative deals with overall fruit quality.

### 2.7.1 Measurement of color

Color of fruits plays an important role in fruit consumption and is one of the most important quality attributes in the selection process. Sometime color influences flavor recognition and it affects consumer perception. Human eye posses three types of light judgment capacity and each correspond to a different band of wavelengths, which are red, green and blue. Spectrophotometer method of color description is also based on three dimensions. Nowadays colorimeter techniques are used for color determination. In 1931 CIE or Commission Internationale de I' Eclairage (International Commission on Illumination) standardized the color order systems which provides a qualitative as well as quantitative description of the color. It is based on the theory that the color is a combination of three primary colors, including red, green and blue. To locate a color in a color space the CIE color system utilizes three co-ordinates. The color spaces are:

- CIE XYZ
- CIE L\* a\* b\*
- CIE L\* C\* h°

Minolta chromameter can be used for determination of color attributes expressed in CIE L\* a\* b\* co-ordinates where L\* defines lightness, a\* represent red/green value and b\* denotes the blue/yellow color value as shown in Figure 2.6. From left to right a\* axis run and +a direction shows shift towards red while along the b\* axis +b movement shows shift toward yellow. The center L\* axis shows the degree of lightness (L=0 for black to 100 for white) on a vertical axis (McGuire, 1992).

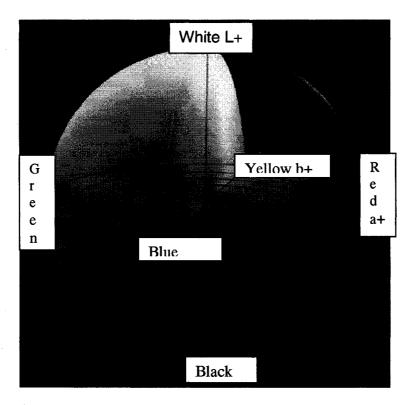


Figure 2.6: Representation of a color solid for L\* a\* b\* color space (Minolta, 1994).

The distance of a color from the center where the two dimensions cross each other indicates the chroma (C\*). We can get this angle by drawing a simple line from the center of the  $a^*$  and  $b^*$  plane through the location of the paint and this line define a hue angle (h\*) (Figure 2.7).

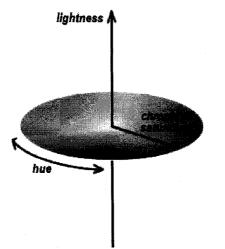


Figure 2.7: The geometrical arrangement of color attributes.

Ripe mango shows a wide range of colors from green to greenish yellow, red, violet and yellow. The main pigments in the mango fruit are chlorophyll, carotenes, xanthophylls and anthocyanin. It has been reported that during ripening of cv. Tommy Atkins chlorophyll is degraded while anthocyanins accumulate (Medlicott et al., 1986).

### 2.7.2 Textural properties

Firmness is defined, as specific force required to deforming a fruit. The fruit, which is more firmed, shows less deformation from a given applied force. The measurement of texture is an important criteria and it can be measured by instrumental or by sensory methods. Instrumental method is more sensitive and reproducible. Different instruments are designed to measure the textural properties of different food products, vegetable and fruits. There are basic five elements which are necessary for the instruments: the driving mechanism, a probe which will be in contact with the sample, a sensory system, a sensor for rate application and system for read out. Usually, for texture evaluation, two methods are very common. One involves the use of a precision penetrometer, which is used for compression tests as well as for puncture resistance with a blunt needle. The other method involves the use of an Instron Universal Testing machine. MJ-treatment of papaya cv. Sunrise combined with modified storage packaging improved the quality by preventing water and firmness loss and delaying yellowing of the fruit (González-Aguilar et al., 2003).

## **2.8.** Conclusions

Mango is the world's most popular fruit with a short season and a short storage life. One of the problems faced by mango traders are variations in physiological maturity leading to fruits to be marketed at different time intervals. When fruits are harvested they are mature and half matured fruits, and neglecting postharvest treatments results in uneven ripening, which decreases the market value. In the case of mango ripening, the process is quite rapid as the fruits ripen within six to seven days and after that they become over ripe and lose their market value.

Proper harvesting, postharvest treatments, appropriate handling and packaging all affect the final market value of the product. Different treatments have been established to

solve this problem but still this field requires more research and development activities leading to appropriate treatment through which shelf life of mango fruit can be increased.

Careful handling minimizes mechanical injuries and currently available technology like low temperature storage, CA and hypobaric storage can improve the shelf life for about 2 to 4 weeks. Postharvest heat-treatments provide protection from pests, insects and other diseases to some extent, but microbial control must be considered and treatment against pathogenic organisms need further research.

More research is required in the storage and ripening field as CI is also a major storage problem and it is still uncertain as to at what temperature CI will occur in different cultivars and what factors are involved? Further work with several mango cultivars by using different heat-treatments and chemicals at different concentrations need to be found for commercial use.

# III. POSTHARVEST TREATMENTS TO REDUCE CHILLING INJURY SYMPTOMS IN MANGOES cv. KENT

## 3.1 Abstract

This study aims at comparing the ability of postharvest treatments in reducing the incidence of chilling injuries on mangoes. Mangoes (*Mangifera indica* L.), like many tropical and subtropical fruits, are prone to chilling injuries when stored below 12 °C. The most frequent visual chilling injury (CI) symptoms of mango fruits are dark, scald-like discoloration and pitting on the skin. These symptoms affect the market value of the fruits and increase post harvest losses. Hot water (HW), methyl jasmonate (MJ) and diphenylamine (DPA), are three post harvest treatments known to reduce CI symptoms.

The mangoes used for the experiment were grown in Ecuador, South America, and shipped by containers to Montreal (QC, Canada). At their arrival, mangoes were divided into four lots that were either HW-treated ( $50 \pm 2 \,^{\circ}$ C) for 10 min, dipped in a  $10^{-4}$  M solution of MJ for 2 min, or dipped in 12 mM solution of DPA for 2-3 min, or left untreated (control). Then, each lot was subdivided into groups and stored at either: 1, 4, 7 or 10 °C for 21 days. Best results were obtained with the MJ and DPA CI- treatments with the percentage of marketable fruits being significantly greater than those under the control CI-treatment. With more than 50 % of marketable fruits, best storage conditions for mangoes cv. Kent was obtained with the following factorial combinations: MJ at 1 and 7 °C and with DPA at 4 and 7 °C. This experiment demonstrates that both growth hormones (MJ) and antioxidant (DPA) postharvest treatments have the potential of improving tolerance to cold of mango fruits cv. Kent thereby reducing the incidence of CI symptoms. This would eventually allow storage of mangoes at temperatures below 10 °C, which would increase the potential storage life of the fruits.

# **3.2 Introduction**

Mango (*M. indica* L.), is grown throughout the subtropical and tropical regions and it is one of the world's most important fruit crops. Most mango fruits are consumed fresh and they are good sources of vitamin A and C. Mangoes are climacteric fruits that ripen quickly once harvested. As with most fruits, it is possible to slowdown the ripening process and extends the storage life by reducing the temperature of the fruits. However, like many other tropical and subtropical fruits, mangoes are prone to chilling injuries when stored below critical minimum temperature (Chaplin et al., 1991; Lizada, 1991).

Recommended storage temperatures are generally within the range of 10 to 15 °C and lead to storage life after harvest of 2 to 3 weeks. Longer storage life could be achieved if the resistance of mangoes to low temperature could be improved. This would allow more fruits to be shipped to overseas' markets.

#### **3.2.1 Chilling injuries**

Mangoes exposed to temperatures below the critical minimum temperature develop severe physiological disorders referred to as chilling injury (CI) symptoms. The primary cause of CI is to damage the cell membrane that initiates a cascade of secondary reactions. CI is a time-temperature problem and some cultivars are more sensitive than others (Mann and Singh, 1976; Farooqui et al., 1985; Fornaris-Rullan et al., 1989; and Saucedo Veloz et al., 1977). CI symptoms include: grayish scald-like discoloration of the skin, skin pitting, uneven ripening, reduction in the level of carotenoids, and poor aroma and flavor (Abou-Aziz et al., 1976; Hatton et al., 1965; Thomas and Oke, 1983; Wardlaw and Leonard, 1936).

#### **3.2.2 Control of CI symptoms**

Postharvest heat-treatments, plant growth regulators and antioxidants have been successfully used to alleviate CI symptoms of tropical fruits. Moderate heat-treatments reduce the incidence of CI symptoms in many tropical crops (Shellie and Mangan, 1994). Heat-treatments induce heat shock protein (HSP), which protected products from both heat injuries and CI. HSP also called stress proteins, and are a group of proteins that present in all cells in all life forms. They are induced when a cell undergoes various types of environmental stresses like heat, cold and oxygen deprivation (Woolf et al., 1995; Klein and Lurie, 1991). The desired temperature and duration of the treatment is product-dependent and its efficacy varies from product to product. The treatment is best applied by immersion in a temperature controlled hot water (HW) bath. High temperatures affect

the firmness of fruits such as plums, tomatoes and avocados (Eaks, 1978; Tsuji et al., 1984; Biggs et al., 1988), their chemical composition (Klein and Lurie, 1990, 1992; Klein et al., 1990), color (Klein et al., 1990; Klein and Lurie, 1992), respiration (Kerbel et al., 1985; Lurie and Klein, 1991) and ethylene production (Biggs et al., 1988).

Jasmonic acid and its methyl ester, methyl jasmonate (MJ) are known as plant growth regulators. They are naturally produced by a wide range of plants and play an integral role in intracellular signal-transduction cascades, which induce plant defense mechanisms. It has been demonstrated that MJ, applied as vapor or as a solution before exposure to low temperatures, can significantly decrease the incidence of CI in zucchini squash (Wang and Buta, 1994), bell pepers, avocado (Meir et al., 1996) and mango (González-Aguilar et al., 2000).

Diphenylamine (DPA) is an antioxidant used commercially to control scald associated with CI in apples (Smock, 1961). The actual mechanism by which DPA reduces scalds is not fully understood but it has been hypothesised that DPA prevents oxidation of  $\alpha$  –farnesene to conjugated trienes (CTs). These compounds are toxic to the product tissues as they initiate free radical chain reactions (Whitaker, 2000).

## **3.3 Objectives**

The objective of this study was to evaluate and compare the benefits of using HW, MJ and DPA postharvest treatments to alleviate CI symptoms of mango fruits stored at temperatures below recommended safe levels at 1, 4, 7 and 10 °C for 21 days.

## **3.4 Materials and Methods**

The effect of the proposed treatments on CI of mangoes was studied using a laboratory-scale experiment. The experiment was laid down using a factorial design in which the treatments consisted of combinations of factors described in Table 3.1. Each factor combination was replicated three times for a total of 48 experimental units. An experimental unit consisted of a lot of 10 mangoes taken at random.

## 3.4.1 Procedure

The mangoes (*M. indica* cv. Kent) used in this experiment were obtained from a local distributor. They were grown in Ecuador, South America and shipped to Montreal

(QC, Canada) in refrigerated containers maintained at 10 °C. They were received for the experiment on January 20, 2003. Fruits were approximately 17 days from harvest at the start of the experiment. Fruits of uniform size, shape and free of defects were used for the experiment. The average mass of an individual fruit was 650 g. Three lots of 5 mangoes were used to assess the quality of the mangoes on their arrival in the laboratory.

Prior to the start of the experiment, the mangoes were randomly divided into four lots of 60 mangoes. One of the four CI-treatments was then applied to each lot. The first lot did not receive any CI-treatment and was used as control. The second lots of 60 mangoes were immersed in HW (50  $\pm$  2 °C) for 10 min. A vibrating water bath was used for this purpose. Mangoes from the third lot were exposed to MJ. A solution of 9:1 ratio of Ethanol and MJ was prepared and then diluted with water to get final concentration of  $10^{-4}$  M. Mangoes were dipped in this solution for 2 min and air dried. Mangoes from the fourth lot were dipped for 2.5-3.0 min in a solution made of 12 mM DPA in 5 % ethanol and 0.05 % Tween 20, and air dried.

After receiving their respective CI-treatment, each lot was then divided into four samples of 15 mangoes each and stored in paper bags for 21 days at either 1, 4, 7 or 10 °C. At the end of storage, the quality of mangoes was assessed and compared.

#### 3.4.2 Quality assessment

The parameters used to monitor the quality of the mangoes before and after storage were: skin and flesh color, pH, sugar content, texture, percentage of mass loss and percentage of good mangoes.

Skin and flesh colors were measured with a Minolta Chromameter Model CR-300X (Minolta camera Co. ltd., Japan) equipped with a 5 mm diameter measuring area. Measurements were reported in terms of CIE (Commission Internationale de I' Eclairage)  $L^*a^*b$  color space and expressed as  $L^*$  (whiteness/darkness, ranged from 0 to 100, while 100 being the lightest), a\* (redness for positive value and greenness for the negative one) and b\* (yellowness for positive and blueness for negative value) (McGuire, 1992). Calibration was performed against a standardized white calibration plate according to the manufacturer specifications.

pH of the flesh was measured using Fisherbrand Alkacid Test Ribbons (Fisher Scientific Ltd, Nepean, Ontario, Canada). The total soluble solids were determined with a hand-held Fisher brand refractometer model 0-90 % (Fisherbrand by Fisher Scientific Ltd, Nepean, Ontario, Canada). The fruits were pressed in order to obtain the juice needed for measurement. An Instron Universal Testing Machine Model 4502 (Canton, MA, USA) was used to measure the textural properties of mango fruits. The test method was defined in the Instron Series IX software which is the data acquisition, control and analysis software for material testing. The test consisted of measuring the force required to push at a constant speed of 25 mm·min<sup>-1</sup> a 6 mm cylindrical probe into the flesh of peeled mangoes. Results were expressed in terms of the maximum force recorded (Fmax) in N and modulus (initial slope of the loading curve) in N·m<sup>-1</sup>. Percentages of mass loss as well as percentages of good mangoes were also calculated at the end of storage for all factor combinations tested. The collected data were subjected to analysis of variance (ANOVA) using Statistical Package System Version 8.0 (SAS Institute Co., 1990). Duncan multiple-range tests were used to locate significant differences between treatment means with the level of significance set at 0.05.

# **3.5 Results**

#### **3.5.1 Initial quality**

The initial quality of the mangoes was measured and the results are presented in Table 3.2. The skin color of mangoes was mostly light green with some mangoes with reddish and/or yellowish patches. The flesh color ranged from pale to dark yellow. The pH of the juice extracted from the flesh varied from 4.0 to 6.0 with an average of 4.8. The total soluble solids (TSS) ranged from 12.0 to 17.0 % with an average of 14.1 %. The mangoes were fairly soft to touch and the average Fmax measured was 10.2 N with the Modulus at 7.2 N·m<sup>-1</sup>. Although not fully ripe, these values suggested that the mangoes were at an advanced state of maturity. This was not surprising since it took about 15 days for the mangoes had good appearance with less than 20.0 % of the fruits showing signs of superficial scalding, and less than 10.0 % of the fruits with minor mechanical injuries.

#### **3.5.2** Quality of mangoes after storage

The quality of the control and the CI-treated mangoes was assessed after 21 days of storage at the prescribed temperatures and the results are presented and discussed below.

## (a) Skin and flesh color

As shown in Tables 3.2 and 3.3, noticeable changes in the skin and flesh color occurred during storage. After 21 days of storage, mangoes skin was darker (lower L\* value), slightly more red (greater a\* value) and less yellow (lower b\* value). Comparison among the four CI-treatments tested (Table 3.4) indicated that the HWT yielded the lightest colored fruits with more red and yellow. MJ-treated fruits were slightly bright (greater L\* value) more yellow in skin color as compare to control fruits while the difference between MJ and DPA CI-treated fruits were not significant at 0.05 level. Aside from this observation, no specific trend could be derived from the data.

As shown in Table 3.5 the analysis performed to assess the effects of storage temperatures on mango skin color did indicate that fruits stored at 1 °C were brighter (greater L\* value), slightly more red (greater a\* value) and less yellow (lower b\* value) in color. Difference in L\*, a\* and b\* values among 7 and 10 °C were small and not significant at 0.05 level. No specific trends could be derived from the data.

Color measurements made on the flesh portion of the fruits indicated that after storage (Tables 3.2 and 3.3), the color was not as bright, less red and less yellow as that measured on fruits before storage. As shown in Table 3.4, it is interesting to note that the best color retention was observed on the control treatment with both the L\* and b\* values significantly greater than those of other CI-treatments. Differences in L\* and a\* among the three other CI-treatments were small and most were not significant at the 0.05 level. The flesh portion of the HW CI-treated fruits was significantly more yellow than control or DPA CI-treated fruits, but not significantly different from MJ CI-treated fruits. As shown in Table 3.5, best flesh color retention was observed on mangoes stored for 21 days at 1 °C with both L\* and a\* values significantly greater than those observed at the other temperatures. In most cases, differences in flesh color expressed as a\* and b\* for the mangoes stored for 21 days at 4, 7 or 10 °C were not significant at the 0.05 level.

#### (b) pH, total soluble solids and texture

As shown in Tables 3.2 and 3.3, the pH of mangoes after 21 days of storage averaged 4.5 down from 4.8 at the start of the experiment. Comparing among the four CI-treatments tested (Table 3.4) indicated that pH value for MJ-treated fruits was significantly lower than for control fruits. Differences among other three CI-treatments were not significant at 0.05 level. In contrast, storage temperatures did not appear to have affected normal changes in fruit pH (Tables 3.5). No specific trends could be derived from the data.

With the exception of mangoes stored at 1 °C, the TSS concentrations increased during storage. As shown in Table 3.3 the averaged TSS concentration of stored fruits was 15.2 % and no significant differences were observed among the four CI-treatments tested (Table 3.4). The averaged TSS concentrations of the fruits stored at 1 °C was 14.0 % which was very close to the percentage observed at the start of the experiment (Tables 3.2 and 3.5). This value was significantly lower than the TSS values measured at the three other temperatures tested.

As expected, all mangoes soften during storage, as this is part of the normal ripening process (Tables 3.2 and 3.3). After 21 days of storage, the overall averaged values of Fmax and Modulus were 5.8 N and 3.4 N·m<sup>-1</sup>. As shown in Table 3.4, differences observed in Fmax and Modulus between the four CI-treatments tested were not significant at the 0.05 level. Although no significant differences in Modulus was observed among the four temperatures tested, the mean values of Fmax recorded on mangoes stored at 10 °C was 7.4 N and it was significantly greater than those obtained at other. This result was unexpected and could be attributed to the higher moisture loss that occurred at that temperature or to the method used to measure fruit firmness.

## (c) Mass loss and marketable mangoes

Comparative percentages of mass loss and marketable fruits are presented in Tables 3.4, 3.5 and in graphical form in Figures 3.1 and 3.2. After 21 days of storage, the averaged mass loss recorded under the control treatment was 4.4 % and it was significantly lower than those recorded under the three other CI-treatments which ranged from 4.8 to 5.0 % (Table 3.4). Differences between the later ones were not significantly different at 0.05 level. On the counter part, as shown in Table 3.5 the storage

temperatures had predominant effects on mass losses. At 1 °C, the averaged mass loss was 2.9 % and it was significantly lower than values observed at 4, 7 and 10 °C. Mass losses recorded at 4 and 7 °C were not significantly different from each other but significantly lower than that at 10 °C. This demonstrates the potential benefit of storing mangoes at temperatures below 10 °C provided that a suitable treatment to prevent CI can be found.

As it can be seen in Figure 3.2, after 21 days of storage, the percentage of marketable fruits were affected by both factors studied. Best results were obtained with the MJ and DPA CI-treatments with the percentage of marketable fruits being significantly greater than that under the control CI-treatment. The lowest percentages of marketable fruits were observed at 10 °C (Table 3.5). The values obtained at 1, 4 and 7 °C were respectively 24.0, 33.9 and 37.0 % and were not significantly different from one another. With more than 50 % of marketable fruits, best storage conditions for mangoes cv. Kent was obtained with the following factor combinations: MJ at 1 and 7 °C and with DPA at 4 and 7 °C.

#### (d) Overall appearance and CI symptoms

In this experiment, untreated (control CI-treatment) mangoes had the lowest percentages of marketable fruits. More than 70 % of these fruits had prominent CI symptoms in the form of black scalding. In addition to these CI disorders, untreated fruits stored at 10 °C had skin discolorations, pitting and wrinkled skins.

More than 50 % of the HW-treated mangoes stored at 1, 4 and 7 °C had black scalding with surface pitting and skin discolorations. In addition to these symptoms, HW-treated mangoes stored at 10 °C had noticeable wrinkle marks on their skin.

With less than 30 %, MJ- or DPA-treated mangoes had the lowest percentage of fruits showing signs of CI symptoms. Here also, symptoms took the form of black scalding, surface pitting and skin discolorations. As with the other treatments, noticeable wrinkle marks were present on fruits stored at 10  $^{\circ}$ C.

# **3.6 Discussion**

In this study on mangoes cv. Kent, it was noted that the skin color of HW and MJ CI-treated fruits were brighter and more yellow than control fruits. These results substantiated the work of McGuire, (1991) and Coates et al., (1993). In their experiments, HWT as well as VHT improved skin color of the mango fruit. Prakash and Pandey, (2000) noted that HWT at 52 °C for 30 min did not effected fruit ripening. González et al., (2001) also mentioned that MJ-treatment improved the color development in mango cv. Kent. In this study, it was established that CI-treatments and storage temperatures significantly affected the mass losses of the stored fruits. González et al., (2000) observed reductions in mass losses for mangoes treated with MJ ( $10^{-4}$ M) and stored for 28 days at 7 °C.

In this study, it was demonstrated that HW CI-treatment had no significant affect on the incidence of CI symptoms and fruit firmness. In a study by Thompson (1987), HWT had little or no effect on the quality of marketable fruit. However, McIntyre (1993) reported that HWT increased shriveling and reduced fruit firmness during prolong storage. In our experiment with cv. Kent highest percentages of marketable fruits were obtained under MJ and DPA CI-treatments. González et al., (2000) reported similar results on mango fruit treated with MJ ( $10^{-4}$ M) before subsequent storage for 21 days at 7 °C.

It should be remembered that the mangoes cv. Kent utilized in this experiment were imported from Ecuador and it was 15 days in transit and that the normal storage period for mangoes is 2 to 3 weeks. At the end of the storage period in this study, the fruits were about 36-days-old following harvest. Therefore, it is very likely that better results (higher percentage of marketable fruits) would have been obtained if freshly harvested mangoes had been used.

This experiment demonstrated that both growth hormone and antioxidant postharvest treatments have the potential of improving tolerance to cold of mango fruits cv. Kent thereby reducing the incidence of CI symptoms. This would eventually allow storage of mangoes at temperatures below 10 °C which would increase the potential storage life of the fruits.

## 3.7 Conclusions

The present study was conducted to assess and compare the ability of HW, MJ and DPA postharvest treatments in reducing the incidence of CI on mangoes. Quality of treated fruits were assessed and compared to a control treatment after 21 days of storage at 1, 4, 7 or 10 °C. Both MJ- and DPA-treatments were successful in retarding the development of CI symptoms in mangoes. An increase in mass loss was observed when fruits were stored at 10 °C. Best results were obtained with the following factor combinations: MJ at 1 and 7 °C and DPA at 4 and 7 °C.

This study demonstrated that both MJ and DPA applied as postharvest treatments have the potential to improve cold tolerance of mango fruits cv. Kent. This could eventually allow storage of mangoes at temperatures below 10 °C which would increase the potential storage life of fruits.

# **3.7 Acknowledgement**

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Factors	Levels	Description
CI-Treatments	1	Untreated mangoes, control
	2	Hot water (HW)
	3	Methyl jasmonate (MJ)
	4	Diphenylamine (DPA)
Storage Temperatures (°C)	1	1
_ •	2	4
	3	7
	4	10

 Table 3.1. Description of the experimental design.

Quality attributes	Average values*	
Skin Color		
L	88.9	
a	-2.1	
b	30.4	
Flesh Color		
L	110.2	
a	-9.3	
b	68.4	
pH	4.8	
TSS, %	14.1	
Fmax, N	10.2	
Modulus, N·m <sup>-1</sup>	7.2	

**Table 3.2**. Quality of mangoes cv. Kent on their arrival in the laboratory.

\*Values presented in this table are averages based on 15 mangoes.

Quality attributes	Average values*
Skin Color	
$\mathbf{L}$ .	56.2
a	4.1
b	20.9
Flesh Color	
L	74.0
a	-3.5
b	43.7
pH	4.5
TSS, %	15.2
Fmax, N	5.8
Modulus, N·m <sup>-1</sup>	3.4

 Table 3.3. Quality of mangoes cv. Kent after 21 days of storage for all CI-treatments and temperatures combined.

\*Values presented in this table are averaged values based on 240 mangoes.

**Table 3.4**. Quality of mangoes cv. Kent after 21 days of storage for each CI-treatment tested and all temperatures combined.

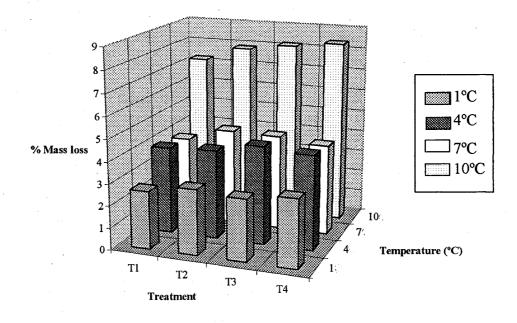
	Treatments			
Quality attributes	No CI- Treatment <i>(Control)</i>	Hot water (HW)	Methyl Jasmonate <i>(MJ</i> )	Diphenylamine <i>(DPA)</i>
Skin Color				
L	54.2c*	58.2a	56.9a,b	55.4b,c
a	5.7a	6.7a	3.4a,b	0.5b
b	18.5b	22.4a	22.0a	20.8b,a
Flesh Color				
L	76.0a	72.6b	73.5b <u>,</u> a	73.7b,a
a	- 3.4a	· - 3.2a	- 3.5a	- 3.8a
b	44.2b	47.2a	42.4b,c	40.8c
pH TSS, %	4.6a 15.4a	4.5a,b 15.5a	4.4b 15.0a	4.6a,b 15.1a
Fmax, N	6.0a	6.2a	5.1a	6.0a
Modulus, N·m <sup>-1</sup>	3.2a	3.4a	3.2a	3.7a
Mass loss, % Marketable	4.4b	4.8a	5.0a	5.0a
mangoes, %	9.9b	21.4b,a	<u>34.9a</u>	<u>31.8a</u>

\*Means in the same row with the same letters are not significantly different at the 0.05 level.

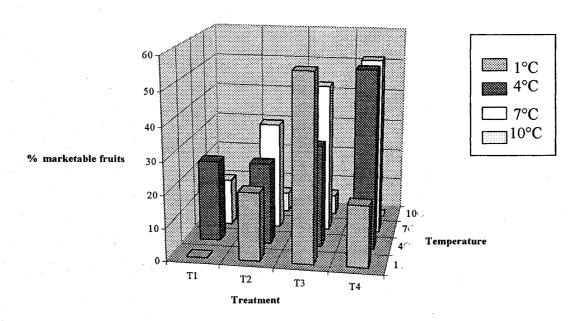
Temperatures (°C)Quality14710				
attributes	<b>ل</b> 	•		
Skin Color		• • •		
L	59.1a	54.1c	56.8a,b	54.7c,b
a	6.7a	1.6b	2.9b,a	5.3a,b
b	19.4b	20.4b,a	22.5a	21.5b,a
Flesh Color		ан на селото на селот На селото на		
L	77.0a	71.6b	73.4b	73.9b
a	-3.9b	-3.5b,a	-3.5b,a	-3.1a
b	50.2a	41.3c	39.0d	44.1b
pH	4.5a	4.6a	4.5a	4.5a
TSS, %	14.0c	16.2a	15.2b	15.7b,a
Fmax, N	4.5c	6.0b	5 1h a	7.4a
Modulus, N·m <sup>-1</sup>	4.30 3.3a		5.4b,c	7.4a 3.4a
Mass loss, %		3.2a	3.7a	
Marketable	2.9c	4.2b	4.2b	7.9a
mangoes, %	24.0a	33.9a	37.0a	3.1b

**Table 3.5**. Quality of mangoes cv. Kent after 21 days of storage for each temperature tested and all CI- treatments combined.

\*Means in the same row with the same letters are not significantly different at the 0.05 level.



**Figure. 3.1:** Percentage mass loss for all CI-treatment / temperature combinations tested after storage for 21 days (where T1 shows control, T2 shows HW-treated, T3 shows MJ-treated and T4 shows DPA-treated fruits).



**Figure.3. 2:** Percentage of marketable fruits all CI-treatment / temperature combinations tested after storage for 21 days (where T1 shows control, T2 shows HW-treated, T3 shows MJ-treated and T4 shows DPA-treated fruits).

# **CONNECTING TEXT**

Experiments were conducted using mangoes cv. Kent and the effect of different postharvest treatments to reduce chilling injury symptoms were noted before storing the fruits at lower temperatures. A lot of 120 fruits were stored at 1, 4, 7 or 10 °C for 21 days and were assessed in terms of quality as presented in Chapter III. Another lot of 120 fruits was transferred after 21 days storage for ripening at 20 °C for 5 days and these results are evaluated in Chapter IV. Further, it should be noted that the experiments discussed in Chapters III and IV were conducted at the same time using the same cultivar.

# IV. POSTHARVEST TREATMENTS TO REDUCE CHILLING INJURY SYMPTOMS IN RIPENED MANGOES cv. KENT

# 4.1 Abstract

Mangoes (*Mangifera indica* L.), like many tropical and subtropical fruits, are subjected to chilling injuries when stored below 12 °C. The symptoms included discoloration, uneven ripening, poor color and pitting on the skin. The intensity of chilling injury (CI) symptoms varies among the cultivars. These symptoms affect the market value of the fruits. Among the various techniques developed Hot water (HW), methyl jasmonate (MJ) and diphenylamine (DPA), are three postharvest treatments known to reduce CI symptoms. This study aims to compare the ability of these postharvest treatments in reducing the incidence of CI symptoms, and to increase the storage life of mango fruits.

The mangoes used for the experiment were grown in Ecuador, South America. At the beginning the fruits were divided into four lots and were either received treatment of HW for 10 min, dipped in a 10<sup>-4</sup> M solution of MJ for 2 min, or dipped in 12 mM solution of DPA for 2-3 min, or left untreated (control). Each lot was subdivided into samples and stored at either: 1, 4, 7 or 10 °C for 21 days. The fruits were allowed for subsequent ripening for 5 days at 20 °C. Fruit quality was assessed and compared after ripening of the fruits. The results indicated that DPA-treated fruits had brighter flesh color with more red and yellow than the other fruits. Higher mass loss was recorded in fruits stored at 10 °C while highest percentage of marketable fruits was obtained at 7 °C. The study demonstrated that mangoes could be successfully stored at temperature below 10 °C, which resulted in increased storage life.

# **4.2** Introduction

Mango (*M. indica* L.), is considered as one of the choice subtropical fruit crops. It has a short growing season and storage life. It can generally be stored for 2-3 weeks by using proper storage conditions and low temperatures. However, chilling injury (CI) is a problem associated with low temperature storage for some crops, mangoes being one of them. The severity of CI symptoms depends upon the storage temperature and duration of

exposure. CI symptoms include scalding, surface pitting, poor aroma, poor color development and increased susceptibility to diseases (Abou-Aziz et al., 1976; Wardlaw and Leonard., 1936).

Many studies have been conducted and various techniques established to alleviate the incidence of CI symptoms in mangoes. It has been demonstrated that heat-treatment reduces the CI symptoms in many tropical crops (Shellie and Mangan., 1994). It is also known that plant growth regulator Methyl Jasmonate (MJ) and antioxidant Diphenylamine (DPA) have potential to either reduce the development of CI symptoms or to increase the resistance to CI (Wang and Buta., 1994; Gonzàlez-Aguilar et al., 2000, Smock., 1961).

# 4.3 Objectives

The objective of this study was to evaluate and compare the potential of HW, MJ and DPA postharvest treatments to alleviate CI symptoms in mango cv. Kent when the fruits were stored at temperatures below the recommended safe levels and ripened at 20 °C for 5 days.

# 4.4 Materials and Methods

The effect of different postharvest treatments including HWT, MJ and DPA was studied by conducting a laboratory-scale experiment. In this experiment, the storage temperatures utilized were 1, 4, 7 and 10 °C with a storage duration of 21 days, followed by an additional 5 days at 20 °C for ripening. The experiment was arranged according to a factorial design in which the two factors were CI-treatments and storage temperatures. In this study, a treatment consisted of a combination of two factors. Each combination was replicated three times for a total of 48 experimental units. An experimental unit consisted of a lot of 10 mangoes taken at random.

#### 4.4.1 Procedure

The mangoes cv. Kent used in this experiment were obtained from a local distributor and had been grown in Ecuador and shipped to Montreal (QC, Canada) in refrigerated containers (10 °C). Fruits were approximately 17 days after harvest at the

start of the experiment. Three lots of 5 mangoes were used for initial quality assessment. It should be noted that the mangoes used for conducting this experiment and the experiment as discussed in Chapter III belonged to the same lot.

The mangoes were randomly divided into four lots of 60 mangoes each. The same procedure as mentioned in Chapter III was applied and the mangoes were stored in paper bags for 21 days at 1, 4, 7 or 10 °C. The fruits were then kept for an additional 5 days at 20 °C for ripening. At the end of ripening, the quality of mangoes was assessed and compared. The parameters used to evaluate the quality of the stored mangoes were: skin and flesh color by using Minolta Chromameter Model CR-300X (Minolta camera Co. ltd., Japan), pH by using Fisherbrand Alkacid Test Ribbons (Fisher Scientific Ltd, Nepean, Ontario, Canada), sugar contents with a hand-held Fisher brand refractometer model 0-90 % (Fisherbrand by Fisher Scientific Ltd, Nepean, Ontario, Canada), texture using Instron Universal Testing Machine Model 4502 (Canton, MA, USA), percentage of mass loss and percentage of good mangoes. The data was subjected to analysis of variance (ANOVA) using SAS Institute Co Statistical Package System Version 8.0 (SAS Institute Co., 1990). Duncan multiple-range tests were used to locate significant differences between treatment means at a 0.05 level.

## 4.5 Results

The mangoes used for this experiment and for experiment as discussed in Chapter III belonged to the same lot. After storage, these fruits were allowed to ripen for 5 days at 20 °C and their quality was subsequently assessed and the results are discussed below:

#### 4.5.1 Skin and flesh color

After ripening, DPA-treated fruits were significantly darker in color (lower L\* value) than control- or MJ CI-treated fruits but not significantly different to HWT fruits (Table 4.1). Differences among other three CI-treatments were not significant at 0.05 level. No significant differences in skin color expressed in terms of a\* and b\* were observed between the four CI-treatments tested (Table 4.1). As for the flesh color, only DPA-treated fruits were slightly brighter (greater L\* value) with more red (greater a\*

value) and yellow (greater  $b^*$  value). Differences among other three CI-treatments in terms of L<sup>\*</sup>, a<sup>\*</sup> and b<sup>\*</sup> value were not significant at 0.05 level.

The analysis performed to assess the effect of storage temperature (Table 4.2) after ripening on mango skin color indicated that the fruits stored at 7 °C were brighter with slightly more red color than fruits stored at either 1, 4 or 10 °C. Also, the flesh color of the fruits stored at 10 °C had significant lower a\* values. With a b\* value of 45.3, fruits stored at 1 °C had the yellowiest color of the four temperatures tested. Although this value was significantly lower than the mean value measured at 7 °C, it was not significantly different from that of fruits stored at either 4 or 10 °C. Differences in L\* and a\* values among the fruits stored at either 4, 7 or 10 °C were not significant at 0.05 level. No specific trend could be derived from the analyzed data.

#### 4.5.2 pH, total solids and texture

After 21 days of storage at 1, 4, 7 or 10 °C and 5 days ripening at 20 °C, pH, TSS, Fmax and modulus of the fruits were measured and the results are presented in Tables 4.1 and 4.2. The analysis of the data indicated that CI-treatments and storage temperatures studied did not interfere with normal changes in fruit pH and TSS. As shown in Table 4.1, DPA CI-treated fruits had lower Fmax value than other three CI-treatments. HW CItreated fruits had significantly lower modulus than MJ or DPA CI-treated fruits, but not significantly different from the control fruits. No specific trends could be derived from the collected data.

### 4.5.3 Mass loss and marketable mangoes

Comparative percentage of mass loss and marketable fruits are presented in Tables 4.1, 4.2 and in Figures 4.1 and 4.2. The results indicated that DPA CI-treated fruits had significantly greater mass loss than control and the HWT fruits, but the difference with MJ-treated fruits was not significant at 0.05 level (Table 4.1).

On the counter part, storage temperatures had prominent affect on the percentage of mass loss and the amount of marketable fruits (Table 4.2). Highest mass loss was observed for fruits stored at 10 °C. After 21 days storage and subsequent ripening the highest percentage of marketable fruits were those stored at 7 °C. This demonstrates the

potential benefit of storing mangoes at temperatures below 10 °C, which would increase the potential storage life of the fruits.

## 4.6 Discussion

The quality attributes of mangoes cv. Kent after storage at 1, 4, 7 and 10 °C for 21 days and after subsequent ripening at 20 °C for 5 days were compared (Tables 3.4, 4.1 and 3.5, 4.2). The results indicated that the skin color of the fruits were more red and yellow after ripening. Fruit pH values remained relatively unchanged. As expected during the ripening process, TSS concentrations increased and flesh firmness decreased. More mass loss and less marketable fruits were recorded after ripening.

It should be noted that the mangoes cv. Kent are highly perishable and that their normal useful life after harvest is 14 to 21 days when kept at 10 to 12 °C. The mangoes used in this experiment, were imported from Ecuador and they were about 17 days old at the start of the experiment. Adding the 21 days of storage and the five days of ripening leads to an overall physiological age of roughly 43 days. This is well in excess of the recommended storage life. This could explain why the statistical analysis performed on the data collected after ripening was less discriminating than the analysis performed after storage.

## 4.7 Conclusions

The present study was conducted to assess and compare the ability of postharvest treatments of HW, MJ and DPA in reducing the incidence of CI on mangoes. Quality of treated fruits were assessed and compared to a control treatment after 21 days of storage at 1, 4, 7 or 10 °C and ripening at 20 °C for 5 days.

Among all the quality parameters studied, it was demonstrated that greater mass losses were recorded for fruits stored at 10 °C and that the highest percentage of marketable fruits were observed for fruits stored at 7 °C. The reason brought forward to explain high losses observed was the advanced physiological age of the fruits. Nonetheless, this study demonstrated that with appropriate postharvest treatments, mangoes could be successfully stored at temperatures below the recommended values of 10-12 °C, which would increase the potential storage life of the fruits.

# 4.8 Acknowledgement

The authors are grateful to Aliments Imex Foods Inc. For providing the mangoes necessary for the study. The authors greatly appreciate the grant obtained from Natural Sciences and Engineering Research Council of Canada (NSERC).

# 4. 9 References:

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Wang, C.Y. and J. G. Buta. 1994. Methyl jasmonate reduces chilling injury in Cucurbita pepo through its regulation of abscisic acid and polyamines levels. *Environ. Exp.Bot.* 34: 427-432.

Wardlaw, W.W. and E.R. Leonard. 1936. The storage of west Indian mangoes. Low Temperature Research Station, Imperical College of Tropical Agriculture, Trinidad: pp.1-47.

Quality attributes	No CI- Treatment <i>(Control)</i>	<u>Treatments</u> Hot water <i>(HW)</i>	Methyl Jasmonate <i>(MJ)</i>	Diphenylamine (DPA)
Skin Color				· · · · · · · · · · · · · · · · · · ·
L	56.7a	55.9b,a	57.13a	53.2b
а	7.4a	7.8a	7.0 <b>a</b>	7.3a
b	28.6a	28.9a	29.9a	29.2a
Flesh Color L a b	67.3b - 2.0a 40.8b	68.4b - 2.0a 41.5b	69.4b -1.8a 41.7b	82.3a - 2.7b 50.1a
pH	4.6a	4.6a	4.5a	4.4a
TSS, %	16.3a	16.2a	16.2a	16.3a
Fmax, N Modulus, N·m <sup>-1</sup> Mass loss, % Marketable	4.2a 2.3a,b 11.8b	4.1a 2.0b 12.3b	3.5a 2.4a 12.5b,a	3.1b 2.5a 13.6a
mangoes, %	15.0a	20.0a	15.0a	20.0a

Table 4.1: Quality of the mangoes cv. Kent after 21 days of storage and subsequent 5days of ripening at 20 °C for each CI-treatment tested and all temperaturescombined.

\* Means in the same row with the same letters are not significantly different at the 0.05 level.

<u>Temperatures (</u> <sup>o</sup> C)					
Quality attributes	1	4	7	10	
Skin Color					
L	54.9b,c	56.0b	59.4a	52.6c	
a	4.0 <b>c</b>	5.9c,b	10.4a	9.2b,a	
b	29.1b,a	30.1a	30.5a	26.8b	
Flesh Color					
L	72.1a	72.8a	71.5a	71.0a	
а	-2.5b	-2.8b	-2.1b	-1.0a	
b	45.3a	43.3b,a	42.4b	43.1b,a	
pH	4.5a	4.6a	4.5a	4.6a	
TSS, %	16.1a	16.4a	16.1a	16.3a	
Fmax, N	3.8a	3.5a	3.8a	3.7a	
Modulus, N·m <sup>-1</sup>	2.1a	2.2a	2.4a	2.4a	
Mass loss, %	11.3c	13.2b	10.6c	15.2a	
Marketable					
mangoes, %	6.7b	16. <b>7</b> b	36.7a	10.0b	

 Table 4.2: Quality of cv. Kent mangoes after 21 days of storage and subsequent 5 days of ripening at 20 °C for each temperature tested and all CI-treatments combined.

\* Means in the same row with the same letters are not significantly different at the 0.05 level.

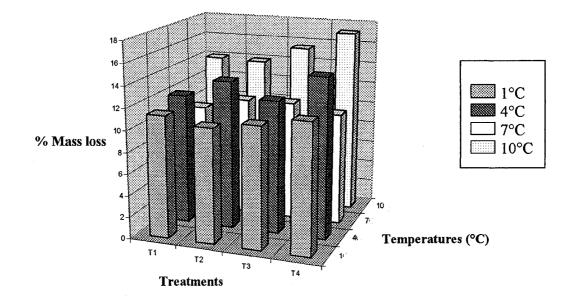


Figure 4.1: Percentage mass loss for all CI-treatment/ temperature combinations tested after storage for 21 days and subsequent 5 days ripening at 20 °C (where T1 shows control, T2 shows HW-treated, T3 shows MJ-treated and T4 shows DPA-treated fruits).

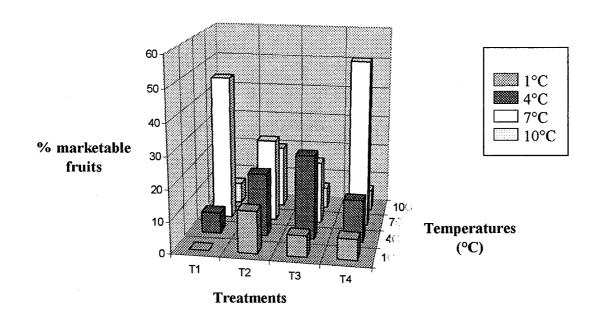


Figure 4.2: Percentage of marketable fruits for all CI-treatment/ temperature combinations tested after storage for 21 days and subsequent 5 days ripening at 20 °C (where T1 shows control, T2 shows HW-treated, T3 shows MJ-treated and T4 shows DPA-treated fruits).

# **CONNECTING TEXT**

In Chapters III and IV the results indicated that postharvest hot water-treatment (HWT) had no significant effect on the reduction of chilling injury (CI) symptoms of mangoes cv. Kent when the fruits were stored at low temperatures or when they were kept at higher temperature for ripening.

In the next chapter a different mango cultivar cv. Tommy Atkins was used and the effect of postharvest Methyl Jasmonate (MJ) and Diphenylamine (DPA)-treatments were investigated. The objective of this study was to evaluate and compare the potential of these treatments to alleviate the CI symptoms in mango cv. Tommy Atkins.

# V. REDUCING CHILLING INJURY IN REFREGRATED STORAGE OF MANGOES cv. TOMMY ATKINS

## 5.1 Abstract

The market life of many fruits and vegetable can be extended through storage at low temperatures. Chilling injury (CI) is one of the major postharvest storage problems for tropical commodities. Storing these products at temperatures below their critical temperature may result in severe physiological disorders referred as CI symptoms. Mangoes (*Mangifera indica* L.) are susceptible to CI when stored below 12 °C. Visual CI symptoms include uneven ripening, surface pitting, discoloration, shriveling, and scald. The appearance of these symptoms decreases the market value. Many researchers have conducted studies to overcome these serious problems. Among the various techniques developed, postharvest treatment with methyl jasmonate (MJ) or diphenylamine (DPA) has been efficient in reducing the incidence of CI symptoms in fruits and vegetables.

This study aims at assessing and comparing the potential of MJ and DPA to reduce CI symptoms in mangoes (cv. Tommy Atkins) stored at temperatures between 1 and 10 °C. Laboratory scale experiments were carried out on mangoes that were either dipped in MJ solution (10<sup>-4</sup> M), or in 12 mM DPA solution, or left untreated (control). The mangoes were stored at 1, 4, 7 and 10 °C for 18 days. Then, the fruits were allowed to ripen at 20 °C for 4 days. Fruit quality was assessed and compared after storage and after ripening. Both MJ- and DPA-treatment had the ability to reduce the CI symptoms in mango fruits stored at low temperatures. Decay was reduced during subsequent ripening. MJ-treated fruits had lower mass loss, brighter colored skin, and higher total soluble solids (TSS) than the control treatment. The overall quality of MJ- and DPA-treated fruits was good with less surface pitting and scalding compared with the control treatment. The best results were obtained at storage temperatures of 7 and 10 °C. Both MJ and DPA postharvest treatment have the ability to reduce CI symptoms in mangoes cv. Tommy Atkins.

## **5.2 Introduction**

Mango (*M. indica* L.), a member of the Anacardiaceae family, is one of the most popular tropical fruits. There are hundreds of mango cultivars, among which cv. Tommy Atkins is one of the most popular cultivars in North America. It is oval to oblong in shape and the skin is orange yellow in color with dark red blushes and numerous white dots. Mangoes are harvested at physiologically mature stage and are allowed to ripen under optimum conditions. Their shelf life is usually 2 to 3 weeks.

The shelf life can be extended by precooling, chemical treatments and storage at low temperatures. Chilling injury (CI) in mangoes is one of the major postharvest storage disorders. It occurs when the fruits are stored at temperatures below their minimum critical temperature and result in the development of severe physiological disorders (Chaplin et al., 1991; Lizada; 1991). These symptoms include scalding, pitting on fruit skin, poor aroma, failure to ripen, poor color development and increased susceptibility to different diseases (Abou-Aziz et al., 1976; Wardlaw and Leonard, 1936). The severity of CI depends upon temperature and duration of exposure. CI symptoms become more prominent, when the fruits are transferred from low to higher temperatures. Many studies have been conducted to overcome the problem and various techniques were applied to alleviate the incidence of CI symptoms on mangoes. These methods included heattreatments, controlled and modified atmosphere storage, application of different chemicals or biological agents, including plant growth regulators and antioxidants (McCollum et al., 1993; Yahia, 1998; Gonzàlez-Aguilar et al., 2000).

Jasmonic acid (JA) and methyl jasmonate (MJ) are known as natural plant growth regulators and they are usually found in higher plants (Koda 1992; Meyer et al., 1984). It plays an important role in the plant defense mechanism. It has been reported that MJ can affect other physiological processes including the inhibition of seed germination, callus growth and pollen development, and the stimulation of root formation, synthesis of ethylene and senescence (Koda, 1992; Staswick, 1992). JA induces the synthesis of stress proteins in affected tissues which could lead to increases in tolerance to CI. It has been reported that the postharvest applications of MJ before storage at a low temperature can reduce CI symptoms in grapefruit, avocado, bell pepper (Meir et al., 1996), zucchini squash (Wang and Buta, 1994), papaya and mango (González-Aguilar et al., 2003, 2000).

In addition, postharvest treatment of fresh cut celery and peppers with MJ increased shelf life and reduced microbial contamination (Buta and Moline, 1998).

DPA is commercially used to control superficial scald in apples (Smock, 1961). Although the actual working mechanism is not yet fully understood, it is believed that DPA has ability to block the oxidation of  $\alpha$ -farnesene to conjugated trienes (CTs). These compounds play an important role in the development of superficial scalding as they initiate free radical mediated chain reaction, which are subsequently decomposed in to harmful volatiles (Huelin and Coggiola, 1970, 1968).

## 5.3 Objectives

The main objective of this study is to evaluate the potential of MJ and DPA postharvest treatments to reduce CI symptoms in mangoes cv. Tommy Atkins stored at different temperatures below the recommended safe level.

## **5.4 Materials and Methods**

Effect of MJ- and DPA-treatments on CI of mangoes has been studied by conducting a laboratory experiment. The experiment was laid down using a factorial design in which the treatments consisted of combinations of factors described in Table 5.1. Each treatment was replicated three times for a total of 36 experimental units. Each unit consisted of a lot of 5 mangoes taken at random.

#### 5.4.1 Procedure

Mangoes (*M. indica* L. cv. Tommy Atkins) used in this experiment were obtained from a local distributor. They were grown in Mexico and shipped to Montreal (Quebec, Canada) in refrigerated containers maintained at 10 °C. They were received on June 11, 2003, approximately 5 days after harvest. Before the start of the experiment, the fruits were conditioned at 10 °C for 16-18 hrs. Fruits of uniform size, shape, maturity and free of defect were used. Three lots of 5 mangoes were used for initial quality assessment.

### 5.4.2 Treatments and storage conditions

Prior to the start of the experiment, the fruits were randomly divided into three lots of 60 fruits each. The first lot was used as a control and did not receive any CI treatment (CI-1). The second lot (CI-2) of 60 mangoes was dipped in the MJ solution for 2 min and then air-dried. The solution was prepared by mixing 1 part of MJ into 9 parts of ethanol and adding distilled water to get a final concentration of 10<sup>-4</sup>M. The third lot (CI-3) received the DPA-treatment. A solution of 12 mM DPA in 5 % ethanol with 0.05 % Tween-20, was made and the fruits were dipped in the solution for 2 to 2.5 min and then air-dried. After receiving their respective CI-treatment, each lot was randomly divided into 12 experimental units of five mangoes each. They were placed in paper bags and stored for 18 days at either: 1, 4, 7 or 10 °C. After storage, the quality of the fruits was evaluated. Then, the fruits were kept for an additional 4 days at 20 °C for ripening. At the end of ripening, the quality of mangoes was again assessed and compared.

### 5.4.3 Quality evaluation

The parameters utilized to evaluate the quality of the fruits after storage were as follows: percent mass loss, visual appearance, incidence of CI symptoms, and skin color. In addition to these parameters, changes in flesh color, total soluble solids (TSS), and texture of the fruits were also measured at the end of the ripening process. The visual appearance was rated using a scale ranging from 1 to 5 (Table 5.2). The incidence of CI symptoms was evaluated using the amount of pitted surface area, scalding, discoloration and shriveling. The scales utilized for this part of the evaluation are presented in Table 5.2. Skin and flesh color was measured with a Minolta Chromameter (Model CR-300X, Minolta camera Co. ltd., Japan) and expressed in 3-dimentional L\*, a\*, b\* color coordinates (McGuire, 1992). Hand-held Fisherbarnd refractometer (Fisherbrand by Fisher Scientific Ltd, Nepean, Ontario, Canada) was used for determining the TSS content. An Instron Universal Testing Machine Model 4502 (Series IX Automated Materials Testing System) was used to measure the textural properties of fruits. The test consisted of measuring the force required to push at a speed of 25 mm/min, an 8 mm cylindrical probe into the flesh of the peeled mangoes. Parameters recorded included the maximum Force (Fmax) in N and the modulus in  $N \cdot m^{-1}$ .

#### **5.4.4 Data analysis**

The experiments were conducted in three replicates. The data collected was subjected to analysis of variance (ANOVA) using the Statistical Package System Version

8.0 (SAS Institute Co., 1990). Duncan's multiple range tests were used to determine if treatment means were significantly different at the 0.05 level.

## 5.5 Results

#### 5.5.1 Initial quality of the mango fruits

The initial quality of the mangoes was assessed at the start of the experiment and the results are presented in Table 5.3. The overall quality of fruits was good with less than 10 % showing signs of mechanical injuries. The predominant skin color was dark green with a large dark purple-red area and some green-yellow patches. This color pattern is characteristic of the cv. Tommy Atkins. Flesh color varied between fruits from light to dark yellow. The average mass of an individual fruit was 355.7 g. Total soluble solids (TSS) ranged from 9.0 to 19.0 % with an average value of 12.8 %. Data on fruit firmness was inconsistent and only the modulus values are reported.

#### 5.5.2 Quality of mangoes after storage and ripening

The quality of the mangoes was assessed after 18 days storage and after 4 days of subsequent ripening. The results were compared among the basis of CI-treatments and temperatures.

### 5.5.3 Effects of CI-treatments on mango quality

## (a) Treatment CI-1 (Control)

#### Mass loss

Mass losses recorded in CI-1 (control) fruits after 18 days of storage ranged from 0.9 % (4 °C) to 4.1 % (1 °C). All differences among the four temperatures tested were significant at the 0.05 level (Table 5.4). At the end of the subsequent ripening period, the highest mass losses (6.1 %) were recorded for mangoes stored at 1 °C and the lowest (3.3 %) for those stored at 4 °C.

## Visual appearance

After 18 days of storage, the appearance of the fruits stored at 4 °C and above was good and significantly better than that of fruits stored at 1 °C (Table 5.4). The best score was given to mangoes stored at 10 °C, and it was not significantly different from the score given to fruits stored at 7 °C. The evolution of the visual quality over the 4 days ripening

period was characterized by further reductions in appearance. The mean visual appearance value of mangoes stored at 10 °C was fair to good and it was significantly greater than the values recorded at other storage temperatures. Mangoes stored at 1 °C had the lowest value and it was significantly lower than the values recorded at other temperatures. The difference in visual appearance observed for mangoes stored at 4 and 7 °C was not significant at 0.05 level.

## Incidence of CI symptoms

The incidence of CI symptoms was evaluated on the basis of surface pitting, discoloration, shrivelling and scald formation of fruits and the results are presented in Table 5.4. Measurements made after storage and after ripening demonstrated the sensitivity of mangoes to CI. Fruits stored at 10 °C showed the lowest incidence of surface pitting and discoloration. After ripening, surface pitting and discoloration became more and more apparent on fruits stored at temperatures below 10 °C. Fruits stored at either 1 or 4 °C had the highest incidences and the difference between these two temperatures was not significant at the level of 0.05. At the end of the storage as well as after ripening, shriveling in all fruits stored at either 1, 4, 7 or 10 °C were not significant. After storage, fruits stored at 1 °C had the most scald formation but it was not significantly different from that of fruits stored at 4 °C. Scald formation progressed during ripening and at the end; levels were found to be inversely related to the storage temperatures. The levels recorded at 1 °C was significantly greater than those recorded at other temperatures tested (Table 5.4).

#### Skin and Flesh Color

Storage temperature had marginal effects on mangoes' skin color. After 18 days storage and subsequent ripening, differences among the four temperatures studied in  $L^*$  and b\* values were not significant at 0.05 level (Table 5.4). Mangoes stored at 10 °C had significantly greater a\* values suggesting that they were slightly redder than the others, but not significantly different from that of fruits stored at 4 °C.

Color measurements made on the flesh portion of the CI-1 (control) fruits indicated that fruits stored at 7 and 10 °C were significantly bright in color (greater L\* value) than those stored at 1 and 4 °C. Differences in a\* values were not significant

among the four temperatures tested. Fruits stored at 10 °C were more yellow in color than those stored at the three other temperatures.

#### TSS and Fruit Firmness

As expected, the TSS concentrations increased during ripening (Tables 5.3 and 5.4). Fruits stored at 1 °C had the highest TSS content. However, the value was not significantly different from the values recorded for the fruits stored at either 4 or 10 °C.

As expected, a reduction in flesh firmness was observed during storage and ripening. This was associated with the sharp decrease in modulus, which was felt from about 8.0 to below  $3.1 \text{ N} \cdot \text{m}^{-1}$  (Tables 5.3 and 5.4). As shown in Table 5.4, highest value of modulus was recorded on fruits stored at 7 °C. Although greater, this value was not significantly different from the value recorded at 4 °C. Differences in modulus recorded between 1, 4 or 10 °C, were not significantly different from one another. Differences in Fmax were not significant among the four temperatures tested.

### (b)Treatment CI-2 (MJ -treatment)

#### Mass Loss

After 18 days of storage, mass losses recorded under CI-2 treated fruits ranged from 0.7 % to 4.2 % with the highest losses being observed on fruits stored at 1 °C (Table 5.5). After ripening, mangoes stored at 1 °C had lost more than 6.6 % of their mass and this was significantly greater than the values observed at 4 and 7 °C, but not significantly different from the mass loss at 10 °C. Difference between 4 and 7 °C was small and not significant at the 0.05 level. Magnitudes and trends observed under treatment CI-2 were similar to those under CI-1.

#### Visual Appearance

As shown in Tables 5.4 and 5.5, for all four temperatures studied, the visual appearance of MJ-treated (CI-2) was better after storage and after ripening than that of control fruits (CI-1). At the end of storage and after ripening, fruits stored at either 4, 7 or 10 °C (Table 5.5) had the highest score and differences were not significant at the 0.05 level. Fruits stored at 1 °C had the lowest score.

## Incidence of CI symptoms

After 18 day storage, CI-2 fruits stored at 1 and 4 °C showed significantly greater incidence of surface pitting and discoloration than those stored at 7 and 10 °C (Table 5.5). At the end of the ripening period, fruits stored at 1 and 4 °C had significantly greater amount of surface pitting while differences in discoloration were no longer significant. The amount of shriveling did not appear to be affected by storage temperatures as the differences observed were not significant at the 0.05 level. The amount of scalding seemed to be inversely related to storage temperature. Highest level was recorded on fruits stored at 1 °C and it was significantly greater than those observed on fruits stored at either 7 or 10 °C, but not significantly different from that fruits stored at 4 °C (Table 5.5). When compared to untreated fruits (CI-1), fruits that underwent the postharvest MJtreatment (CI-2) had less surface pitting, discoloration and scald indicating that MJ was effective in reducing the incidence of CI symptoms in mangoes stored at lower temperatures (Tables 5.4 and 5.5).

#### Skin and Flesh Color

The effect of temperature on the skin and flesh color of CI-2 mangoes was minimal. Measurements made after storage and after ripening indicated that most differences in skin color or in flesh color, expressed in terms of L<sup>\*</sup>, and b<sup>\*</sup>, were not significant at the 0.05 level (Table 5.5). Flesh color of the fruits stored at 7 °C had significantly greater a<sup>\*</sup> value than those stored at either 1, 4 or 10 °C.

## TSS and Fruit Firmness

As shown in Table 5.5, at the end of the ripening period, the measurements of the TSS concentrations made on CI-2 treated fruits indicated that the storage temperature studied had no impact of fruit TSS content. The TSS concentrations ranged from 18.1 to 19.0 %.

Firmness measurements made on the fruits at the end of ripening indicated that fruits kept at 1 and 7 °C had significantly lower Fmax values. Also, fruits stored at 7 °C had significantly lower modulus than those stored at either 4 or 10 °C, but not significantly different from those stored at 1 °C.

## (c) Treatment CI-3 (DPA-treatment)

#### Mass Loss

Under CI-3, mass losses recorded after 18 days storage ranged from 0.7 % (4 °C) to 4.6 % (1 °C). All differences between the four temperatures tested were significant at the 0.05 level. After ripening, mangoes stored at 1 °C had lost more than 7.1 % of their mass and this was significantly greater than the values observed at the other temperatures (Table 5.6). After 18 days storage and subsequent ripening lower mass loss was observed in fruits stored at 4 °C. Under treatment CI-3, magnitudes and trends in mass loss were similar to those observed under CI-2 and CI-1 (Tables 5.4, 5.5 and 5.6).

## Visual Appearance

With a score of 4.4 over 5, fruits stored for 18 days at 7 °C had the best visual appearance among the DPA (CI-3) treated fruits (Table 5.6). This score was significantly greater than that of fruits kept at either 1 or 10 °C. After 4 days of ripening, the highest scores were observed at 4 and 7 °C, and the difference was not significant at 0.05 level. This trend was similar to what was observed with CI-2 treated fruits (Figure 5.1).

### Incidence of CI Symptoms

The incidence of CI symptoms was evaluated after 18 days storage and after the following 4 day ripening period. In both evaluations, fruits stored at 1 °C showed the highest incidence of surface pitting (Table 5.6). Differences observed in surface pitting among the three other temperatures tested were not significant at the 0.05 level. At the end of ripening, discoloration of fruits stored at 4 °C was significantly greater than the value recorded at 7 °C. As for the other CI symptoms monitored, differences between temperatures were small and, in most cases, not significantly different. When compared to control fruits (CI-1), both CI-2 and CI-3 treated fruits had lower incidence of CI symptoms. This is illustrated in Figure 5.2 which shows the effects of CI-treatments and temperature on the incidence of superficial scald on mangoes.

## Skin and Flesh Color

As for CI-1 and CI-2 treated fruits, the effect of temperature on the skin and flesh color of CI-3 mangoes was minimal (Table 5.6). After storage as well as subsequent ripening, the fruits stored at 1 °C were slightly greener in color. Differences among the four temperatures studied in L\* and b\* values were not significant at 0.05 level. Color

measurements made on the flesh portion of the fruits indicated that fruits stored at 1 °C had lower L\* value than the fruits stored at either 4 or 7 °C. Differences in a\* and b\* values among the four temperatures studied were not significant at 0.05 level. Temperatures in the range of 4 to 10 °C did not interfere with flesh color development of mangoes cv. Tommy Atkins. A separate analysis performed on skin and flesh color data indicated that the postharvest MJ- and DPA-treatments did not appear to affect noticeably fruit color when compared to untreated fruits.

### **TSS and Fruit Firmness**

As mentioned in Table 5.6, after ripening, the TSS content of the CI-3 treated fruits ranged from 17.6 to 18.6 % and none of the differences observed among the four temperatures tested were significant. Comparing between the treatments, CI-2 and CI-3 treated fruits gave significantly greater TSS value. At 7 °C, both CI-2 and CI-3 treated fruits had significantly greater TSS than the control treatment (Tables 5.4, 5.5 and 5.6). For the four temperatures studied, firmness measurements made on the CI-3 fruits at the end of the ripening period indicated that the differences observed in Fmax and modulus were not significant at the 0.05 level (Table 5.6).

#### 5.6 Discussion

In this study on mangoes cv. Tommy Atkins, it was demonstrated that storage temperatures had significantly affected mass losses while, at any given temperature, no significant differences were observed among the three CI-treatments tested. In the previous experiment on cv. Kent (Chapter III and IV) it was also observed that storage temperature had significantly affected mass losses. However, MJ- and DPA-treated fruits had significantly greater mass losses. On the other hand, González et al., (2000) reported reductions in mass losses for mangoes treated with MJ (10<sup>-4</sup>M) and stored for 28 days at 7 °C. No other report could be found on either DPA-treated mangoes or on mangoes stored at temperature below 7 °C.

In this study, it was clearly demonstrated the both MJ (CI-2) and DPA (CI-3) were beneficial in reducing the incidence of CI symptoms on mangoes when the fruits were stored at temperatures below the critical value. Similar results were obtained with the mangoes cv. Kent and the results were discussed in Chapter III and Chapter IV. These results corroborate the work performed by González et al. on cv. Tommy Atkins (González et al., 2000) and cv. Kent (González et al., 2001) mangoes. They also mentioned that the MJ-treatment delayed color development in mangoes cv. Tommy Atkins while improving color development in mangoes cv. Kent. However, there was no effect on the fruit quality. In our study, MJ-treatments did not noticeably affect the color development when compared to the untreated fruits. A similar trend was also observed with the DPA-treated mangoes. Finally, González et al., 2000 reported that cv. Tommy Atkins mangoes treated with MJ and stored for 28 days at 7 °C and kept for 5 days at 20 °C, had greater TSS content than untreated fruits stored under the same conditions. In our study, mangoes treated with MJ (CI-2) or DPA (CI-3), stored for 18 days at 7 °C and kept for 4 days at 20 °C, had significantly greater TSS than the control fruits (CI-1). In our study on mangoes cv. Kent (Chapter III) no significant differences in TSS value were obtained among the four CI-treatments tested. In light of the above, four factors that appeared to have affected fruit response to MJ-treatments were fruit cultivar, maturity, storage temperature and storage duration.

## **5.7 Conclusions**

Like many tropical fruits, mangoes are prone to CI when they are stored at temperatures below the recommended value. A laboratory experiment was conducted on mangoes cv. Tommy Atkins to assess and compare the ability of postharvest MJ- and DPA-treatments to alleviate CI symptoms in mangoes stored at 1, 4, 7 and 10 °C.

Parameters used to compare the effectiveness of the treatments indicated that both MJ and DPA have the potential to reduce the incidence of CI symptoms in mango fruits cv. Tommy Atkins. In all CI-1, CI-2 and CI-3 treated fruits, excessive mass losses were observed in fruits stored for 18 days at 1 °C. Measurements made on skin and flesh color indicated that storage temperature as well as the MJ- and DPA-treatments did not interfere with the normal color development of mangoes.

## 5.8 Acknowledgement

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Factors	Levels	Description
<b>CI-Treatments</b>	1	Untreated mangoes, control
	2	Methyl jasmonate (MJ)
	3	Diphenylamine (DPA)
	1	1
Storage Temperatures (°C)	2	4
	3	7
	4	10

 Table 5.1. Description of the experimental design.

 Table 5.2 Parameters utilized to evaluate the quality of the stored mangoes.

Quality	Score	Description
Visual appearance <sup>1</sup>	1	Very poor
	2	Poor
	3	Fair
	4	Good
	5	Very good
Surface Pitting and Scalding <sup>2</sup>	1	No injury
0	2	10-20 % of total surface
	3	11-25 % of total surface
	4	26-50 % of total surface
	5	more than 50 % of total surface
Discoloration		
	1	No discoloration
	2	Slight discoloration
	3	Moderate discoloration
	4	Severe Discoloration
Shrivelling	1	No shriveling
-	2	0 to 30 % of total surface
	3	31 to 50 % of total surface
	4	More than 51 % of total surface

1 Gonzalez-Alguilar, 1997 2 Purvis, 2002

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Quality attributes	Average values	
Mass (g)	355.7	
Skin Color	39.8	
L	34.1	
a	16.9	
b		
Flesh Color	75.5	
L	-0.9	
a	54.0	
b	12.8	
TSS, %	8.0	
Modulus, N·m <sup>-1</sup>	0.0	

 Table 5.3. Quality of mangoes cv. Tommy Atkins on their arrival in the laboratory.

<u> </u>		Temperatures °C (after storage)				8 days of storage and after 4 days ripening at 20 °C. Temperatures °C (after ripening)			
Quality attributes	1	4	7	10	1	4	7	10	
Mass loss, %	4.1a	0.9d	1.4c	2.1b	6.1a	3.3c	4.0b,c	4.6b	
Visual appearance	1.9c	2.9b	3.1a,b	3.8a	1.5c	2.3b	2.1b	3.5a	
CI assessment									
	3.0a	2.6a,b	1.5b,c	1.1c	3.5a	3.3a	2.6a,b	1.5b	
Surface pitting	3.0a	2.9a	2.0b	1.3c	3.5a	3.4a,b	2.7b	1.7c	
Discoloration	1.5a	1.0a	1.0 <b>a</b>	1.0a	1.7a	1.2a	1.5a	1.3a	
Shrivelling	3.8a	3.0a,b	2.3b	1.9b	4.1a	3.4b	3.1b,c	2.6c	
Scald formation									
Skin Color	39.5a	41.6a	40.3a	42.8a	38.9a	42.0a	42.9a	41.6a	
L	25.2b	31.0a,b	26.5b	34.2a	25.1b	28.4a,b	26.5b,c	32.2a	
a	14.0a	19.4a	19.3a	19.1a	13.6a	17.2a	18.6a	16.9a	
b									
Flesh Color					60.3b	60.1b	65.3a	67.8a	
L					0.8a	0.2a	-0.8a	0.4a	
a					49.4b	48.9b	51.2b	56.6a	
b									
					18.1a	17. <b>7</b> a,b	17.2b	17.7a,t	
TSS, %					5.9a	6.8a	8.6a	5.7a	
Fmax, N Modulus, N·m <sup>-1</sup>					1.8b	2.4a,b	3.1a	1.6b	

Table 5.4. Quality of the mangoes cv. Tommy Atkins, CI-1 (control) afte	er 18 days of storage and after 4 days ripening at 20 °C.
Temperatures °C (after storage)	Temperatures °C (after ripening)

\* Means in the same row either after storage or after ripening, with the same letters are not significantly different at the 0.05 level.

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	Temperatures °C (After storage)				Temperatures °C (After ripening)			
Quality attributes	1	4	7	10	1	4	7	10
Mass loss, %	4.2a	0.7c	1.3b,c	2.4b	6.6a	3.0c	4.3b,c	5.0a,t
visual appearance	2.6b	3.5a,b	4.4a	4.3a	2.0b	3.4a	3.8a	3.9a
CI assessment								
	2.1a	1.9a	1.0b	1.1b	2.3a	2.0a	1.3b	1.2b
Surface pitting	2.8a	2.2a,b	1.3b	1.3b	2.9a	2.4a	1.6a	1.7a
Discoloration	1.1a	1.0a	1.0a	1.0 <b>a</b>	1.2a	1.0a	1.0a	1.0a
Shrivelling	3.3a	2.4a,b	1.5b	1.4b	3.4a	2.7a,b	2.0b	1.7b
Scald formation								
Skin Color	40.0a	40.7a	42.8a	40.9a	39.3a	40.4a	43.8a	39.8a
$\cdot$ L	27.3a	31.5a	28.6a	32.6a	26.6a	30.1a	26.4a	31.1a
a	13.4a	16.7a	21.2a	15.5a	12.9a	15.2a	19.6 <b>a</b>	14.0a
b								
Flesh Color					59.2a	61.7a	62.8a	66.1a
L					1.0b	0.5b	2.4a	1.1b
a					53.1a	51.2a	55.4a	53.0a
b								
					18.8a	18.1a	19.0a	18.7a
ГSS, %					5.3b	8.1a	5.1b	7.4a
Fmax, N Modulus, N∙m⁻¹					2.0a,b	2.6a	1.6b	2.8a

Table 5.5. Quality of the CI-2 (MJ-treated) mangoes after 18 days of storage	and after 4 days ripening at 20 °C.
Temperatures °C (After storage)	Temperatures °C (After ripening)

\* Means in the same row either after storage or after ripening, with the same letters are not significantly different at the 0.05 level.

Table 5.6. Quality o		nperatures °C	· · · · · · · · · · · · · · · · · · ·		Temperatures °C (After ripening)			
Quality attributes	1	4	7	10	1	4	7	10
Mass loss, %	4.6a	0.7d	1.2c	2.2b	7.1a	3.2c	3.7b,c	4.7b
Visual appearance	2.8b	3.5a,b	4.4a	3.4b	2.5c	3.7a	3.5a,b	2.7b,c
CI assessment								
	2.2a	1.2b	1.0b	1.3b	2.7a	1.0b	1.0b	1.0b
Surface pitting	2.3a	2.0a	1.3a	1.8a	2.4a,b	2.9a	1.6b	2.2a,b
Discoloration	1.0a	1.0a	1.0 <b>a</b>	1.0a	1.1a	1.1a	1.0a	1.0a
Shriveling	3.1a	2.3a	1.7a	2.3a	3.3a	2.6a	2.5a	2.7a
Scald formation								
Skin Color	44.3a	42.6a	38.6a	41.9a	45.1a	42.6a	40.0 <b>a</b>	41.0a
L	19.4b	27.0a	33.2a	31.1a	23.1b	29.4a	28.8a	29.4a
a	19.5a	15.9a	20.0a	17.5a	21.4a	17.4a	15.6a	16.6a
b								
Flesh Color					61.0b	65.2a	65.8a	62.5a,b
L					0.8a	2.6a	0.4a	1.4a
a					51.4a	53.3a	53.5a	52.9a
b								
					17.6a	18.0a	18.1a	18.6a
TSS, %					5.1a	7.3a	6.6a	6.2a
Fmax, N Modulus, N·m <sup>-1</sup>					1.9a	2.9a	2.4a	2.2a

Table 5.6 Quality of the CL 2 (DDA treated) managers after 18 days of storage and after 4 days ringping at 20 °C

\* Means in the same row either after storage or after ripening, with the same letters are not significantly different at the 0.05 level.

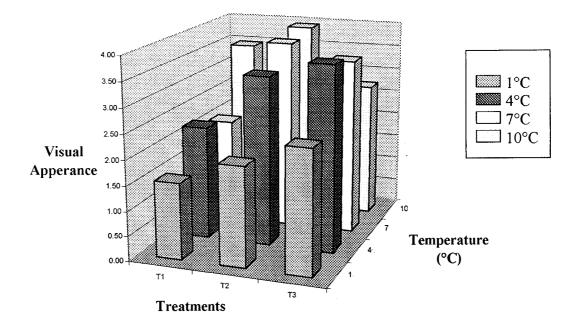


Figure 5.1: Visual appearance of mango fruits cv Tommy Atkins after 18 days of storage and 4 days subsequent ripening (where shows control, T2 shows MJ-treated and T3 shows DPA-treated fruits).

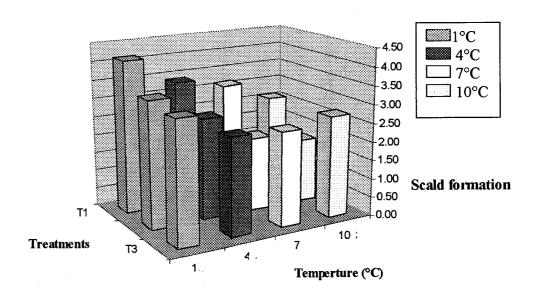


Figure 5.2: Scalding of mango fruits cv Tommy Atkins after 18 days of storage and 4 days subsequent ripening (where T1 shows control, T2 shows MJ-treated and

# **CONNECTING TEXT**

In the previous chapters use of different postharvest treatments to reduce the incidence of chilling injury (CI) in stored mangoes has been discussed. After determining the best treatments to reduce CI symptoms the next step is to focus our idea on fruit color development. Color development of the fruits plays an important role in the fruit marketing and in the following chapter, the use of different chemicals and gasses for mango color enhancement was reviewed has been discussed. Their advantages and disadvantages were considered. Use of calcium carbide for mango color development is evaluated critically as many developing countries continue to use it for fruit color development. Other alternatives are suggested.

# VI. ARTIFICAL METHODS USED FOR FRUIT COLOR DEVELOPMENT.

## 6.1 Abstract

For export or for local market display, ripening of mango fruit is required since color and texture of the fruit play an important role in attracting consumers. To increase the shelf life of fruits, the ripening process can be delayed through different treatments. Mangoes do not ripen at once on the tree, therefore mangoes are harvested mature but in their un-ripped or green form. Unscrupulous traders, farmers and retailers are using certain chemicals to force the ripening of fruits. Harvesting at green stage facilitates handling of fruits in boxes and crates. There is a saying that "an apple a day keeps the doctor away" that may be transformed to " a mango a day will rush you to the doctor". As most of us do not know that, certain chemicals used for artificially ripening fruits are severely harmful for the health. Calcium carbide is one of them, which is commonly used by many developing countries as a fruit ripening agent especially for banana and for mangoes. Commercial calcium carbide sample has many impurities and some of them are very dangerous to human health like arsenic and phosphorus hydride, which are contained in calcium carbide as impurities. Calcium carbide is basically used for welding purposes and is explosive in air.

Certain other chemicals like ethephon, methyl Jasmonate (MJ) and ethylene are also used for mango ripening purposes and to enhance the color development of the fruits. These chemicals are environmental friendly but are quite expensive. The aim of this study is to compare the available techniques used to enhance the fruit color development and their advantages and disadvantages are also being considered.

## 6.2 Objectives

The objective of this study is to discuss different techniques, which are available for artificial fruits color development; and also to focus on their advantages and disadvantages.

### **6.3 Introduction**

Ripening of fruits is a perfectly natural phenomena leading to increased sweetness

flavor development and softening of the fruits. The visual appearance of fruits especially their bright color and texture is one of the major factors, which directly affects the quality of the fruit as well as fresh fruit consumption. Color is not a physical phenomena but it is a sensation experienced by an individual. Usually climacteric fruits are harvested in a mature but unripe condition and then they are allowed to ripen either naturally or by using certain chemicals. Mango fruits ripen unevenly on the tree and fruits are picked by hand at an average maturity. Tree ripe fruits show bright skin color with uniformly softened flesh and developed flavor, but those fruits have a very short shelf life. For distant markets or for export, half ripe or unripe mangoes are used depending on the market distance, but ripe fruits are preferred for local marketing. To extend the shelf life of the product, certain treatments are used to delay the ripening process of the mangoes.

## 6.4 Treatments Affecting Postharvest Ripening

- i) It has been reported that certain preharvest treatments may affect the postharvest ripening of mango fruits. According to Khader (1991) if gibberellic acid (GA<sub>3</sub>) in the concentration of 100-300 mgl<sup>-1</sup> in the form of foliar spray applied prior to harvesting, it can retard the ripening of mango fruits up to 6 days.
- Ethylene is involved in many aspects of fruit ripening and its production is inhibited by high temperatures (Field, 1984). Inhibition of ethylene production was found when mango fruits cv. Nam Dokmai were kept at 38 °C for 3 days and then transferred to 20 °C. Heat-treatment inhibits the activities of 1-aminocyclopropane-1-carboxylic acid synthase (ACS) and 1-aminocyclo propane-carboxylic oxidase (ACO) in the heated fruits. ACS and ACO are the key regulatory enzymes for ethylene biosynthesis. Further, it has been noted that the activity of ACO recovers fully after heat-treatment while ACS activity recovers partially, but enough to allow the heated fruit to reach an ethylene peak (Ketsa et al., 1999). Certain inhibitors of ethylene such as diazocyclopentadiene (DACP) and 1-methylcyclopropene (MCP) also have ability to delay the ripening process (Sisler et al., 1996).

- iii) It has been reported that if calcium chloride or calcium nitrate (0.6-2.0 %) spray are used before harvesting, they can delay the ripening process of fruits (Singh et al., 1993). Pressure or vacuum infiltration with 2-8 % calcium chloride (CaCl<sub>2</sub>) solution can also delay the ripening of mangoes upto 12 and 8 days respectively (Yuen et al., 1993).
- iv) Using inhibitors of respiration and/or ethylene production can retard fruit ripening. A variety of inhibitors have been reported including maleic acid, auxins, vitamin K and salicylic acid (Vendrell., 1970).
- v) Certain fungicidal wax application can also delay the ripening process of mangoes such as wax containing o-phenylphenol, microcrystalline petroleum wax, terpene resin, oleic acid and triethanolamine (Mathur and Subramanyam, 1956).

## 6.5 Artificial Methods Used for Fruits Ripening and Color Development

The natural ripening process can be very slow and unpredictable. To overcome this problem, fruits can be ripened artificially by exposing the fruits to certain chemicals, which initiate the ripening process. Ripe mangoes show a wide range of colors from green to greenish yellow, red, violet and yellow. As external color of the fruit is an important criterion in consumer preference, exogenously application of different chemicals can be used for ripening and color development of fruits.

#### 6.5.1 Straw-bed or rice straw method

In this method, green mangoes are picked and are spread for two days on a thick layer of mango leaves and then transferred to a straw-bed. In India fruits can be spread in multi-layers on thick rice straw, in ventilated storage rooms over a period of 1 week, where each layer is separated from other via straw and this has been found very good for ripening and it also gives attractive color to the fruit (Patwardhan., 1927).

#### 6.5.2 Use of different chemicals and gasses

#### a) Treatment with Ethephon/ethrel

Ethephon, a plant growth regulator is an ethylene-releasing chemical, which can be used to improve fruit color development. But it also stimulates ripening process of the fruit. It is also called chorethiphon, Ethrel, Florel, Cerone, Cepha. It is a gray ceraceous solid with melting point 74-75 °C and is soluble in water and alcohol. It cannot coexist with alkali, salts and metal. Its molecular structure is:

# O II CICH<sub>2</sub>CH<sub>2</sub>P(OH)<sub>2</sub>

# (Ethephon)

Ethephon has been successfully used to improve and enhance red color development and red pigment intensity in pepper fruits (Knavel and Kemp., 1973; Batal and Granberry., 1982). It has been reported that treatment with ethephon improves the peel color and accelerates the mango fruit ripening (Lakshminarayana et al., 1975).

Pal (1998a) reported that when unripe mangoes cv. Dashehri were treated with 200-400 ppm of ethrel and with subsequent storage at 25-28 °C with 85-90 % RH it resulted in a better ripening of the fruits; while low dose of ethrel maintained better fruit firmness on ripening compared to calcium carbide-treated fruits. When ethrel is applied to plants, it is absorbed and broken into ethylene, which is a natural substance that helps in ripening. Usually it is diluted with water and the containers are placed in a storage room. Sodium hydroxide is added to the mixture, ethylene gas is released and forces ripening of the fruits in three to five days. Ethylene is a natural ripening agent found in the fruits and it yields uniform ripening and flavor retention. The rooms must have sufficient air circulation to make uniform distribution of gas, while building of carbon dioxide is to be avoided as it reduces the effect of ethylene. The air should be changed every 4 hrs and ethylene should be reapplied. Ethephon can promote several benefits such as fruit thinning (apples, cherries), color development (apples), degreening (citrus), flower induction (pineapples) and it can stimulate lateral branching in potted plants (azaleas and geraniums).

#### b) Treatment with methyl jasmonate (MJ)

MJ is a known growth regulator produced by a wide range of higher plants. It is a fatty acid derivate and it is volatile in nature. It appears to induce the production of proteinase inhibitors and ethylene. It has been noted that MJ applications effectively

reduce the CI symptoms in different fruits when they are stored at below critical temperature (Wang and Buta., 1994). It has been reported that MJ vapor treatment at the concentration of  $10^{-4}$  M, reduces the CI symptoms in mangoes cv. Tommy Atkins when they were stored at 7 °C without altering the ripening process (González-Aguilar et al., 2000a). In an experiment with mangoes cv. Kent, the result showed that a MJ vapor treatment ( $10^{-5}$ M) for 20 hrs at 20 °C reduced the CI symptoms and enhanced the skin color development (González-Aguilar et al., 2001). Fruits show greater L\*, a\* and b\* values than untreated fruits and those which were treated with MJ ( $10^{-4}$ M).

Use of MJ is safe and it is an environmental friendly compound but it is an expensive substance and it is not commercially used for fruit color development.

### c) Treatment with ethylene

Ethylene is a plant hormone, which is known to be a trigger of the ripening of fruits and promotion of plant senescence. Ethylene is a colorless, sweet odor flammable gas soluble in water. Its molecular structure is:

# $CH_2 = CH_2$

# (Ethylene)

Mango is a climacteric fruit and ethylene plays an important role in ripening, breakdown of carotenoid in the peel, increased respiration, loss of firmness and softening with an observed increase in catalase and peroxidase in ripe mangoes (Mattoo and Modi, 1969; Krishnamurthy and Subramanyam, 1970). Exact mode of action of ethylene is not yet known but in literature a number of theories are available which offer possible explanation. In a harvested fruit small amount of ethylene is already present which is sufficient to initiate the ripening process. Ethylene production decreases as the fruits get matured and then unnoticeable for a time and reappear at the time of ripening (Akamine and Goo, 1973). In pineapple, at vegetative maturity, the ethylene gas is generated which activates the flowering and fruit cycle. In terms of ethylene production, two classes of fresh products exist. One is a climacteric product that produces a burst of ethylene as they ripen, as well as an increase in respiration. The second class is the non-climacteric

products that do not increase ethylene production when they ripen. Ethylene can affect both types of products (Table-6.1).

Climacteric (Ethylene producing)	Non-climacteric (Non ethylene producing)		
Apples, pears, quince	cherry, blackberry, strawberry		
Apricot, nectarine, peach	eggplant, cucumber, pepper		
Mango, avocado, banana	lemon, orange, mandarin		
Tomato, sapodilla	water melon, honey dew melon		
Rock melon, passion fruit	grape, lychee, loquat		

 Table 6.1: Examples of climacteric and non-climacteric products.

\*From Kader, (1992).

The concentration of ethylene required for ripening varies with the product, usually in the range of 1 to 100 ppm concentrations (Table. 6.2). The time and temperature of the treatment also affects the ripening process.

Fruits	Temperature (°C)	Ethylene (ppm)	Treatments (hrs)
Avocado	18 – 21	10	24-72
Banana	15 – 21	10	24
Mango	15 – 21	10	12-24
Kiwifruit	18 – 21	10	24
Persimmon	18 – 21	10	24
Tomato	13 – 22	10	Continuous

 Table 6.2: Ripening conditions for some fruit using ethylene.

\*From Wills *et al.*, (1998).

Depending on the degree of maturity ethylene-treatment causes green mangos to develop full color in 7 to 10 days, whereas untreated fruits require 10 to 15 days. The advantages are that there can be fewer pickings of the fruits and the color development after treatment is more uniform. If the fruits are picked at the proper maturity stage 24 hrs of exposure is usually sufficient. Therefore, ethylene-treatment is a common practice in Israel and Florida for ripening fruits for the local market. As ethylene-treated fruits soften

faster than non-treated one, it is a common practice to give ethylene-treatment at the packinghouse or at the retailer's point before distribution. Use of ethylene for ripening of different fruits has been studied well and no evidence of health related issues, in terms of nutritive value, have been reported (Chace., 1934; Clendennen., 1997).

### **Disadvantages and Environmental concerns**

Presence of ethylene has some disadvantages in terms of postharvest shelf life and can be harmful for the product quality.

- Ethylene is an explosive gas in air and it is colorless hence its presence can be overlooked. Safety measures in terms of electric wiring, piping used in the ripening rooms, and process of application must be considered. Operators must be well trained and prepared. The gas is explosive in air at concentrations ranging from 3.1 % to 32 %.
- Ethylene is very expensive and as such it is not used by many developing countries. For example in People's Democratic Republic of Yemen, the cost of ethylene production estimated is about 50 times greater than locally available calcium carbide used for ripening fruits (Medlicott et al., 1987).
- iii) Ethylene may come from other ripening fruits in the market or storage room or from exhaust gases of vehicle. This exposure may affect the product quality and a continuous exposure to a low concentration of ethylene throughout marketing can cause significant harm. About 10-30 % loss in shelf life of the fresh product has been reported (Wills et al., 2000).
- iv) Ethylene in atmosphere is degraded by UV light (present in the sun) and it can reduce ozone related pollution.
- v) Ethylene is phytotoxic in nature.

#### d) Treatment with calcium carbide

Use of ethylene for ripening of the fruit is a common practice in different countries but due to high cost and scarcity in terms of its availability, many developing countries uses low-cost calcium carbide to ripen fruit, a material more commonly used for welding purposes. Usually calcium carbide is imported from China, Taiwan and South Africa. Calcium carbide-treatments are extremely dangerous as commercial calcium carbide contains impurities of arsenic and phosphorous hydride, which are toxic to human health. Acetylene is generated from calcium carbide by the addition of water or by contact with moisture in air and act on fruits causing them to ripen in a similar manner to ethylene. Use of calcium carbide for fruit ripening has been known for many years but not enough literature is available for acetylene production from calcium carbide. Fruits ripened with calcium carbide are soft and have good peel color development but poor in flavor. A number of countries use calcium carbide to ripen a wide range of fruits as shown in Table-6.3.

 Table 6.3: The fruits and countries where acetylene liberated from calcium carbide have been used to ripen fruits.

Fruit species	Countries
Banana	Australia, Egypt, India, Philippines, South Africa
	Sudan, Taiwan, U.S.A, Yemen
Mango	Brazil, Costa Rica, India, Malaysia, Philippines,
0	Senegal, South Africa
Citrus	Australia, Philippines, South Africa
Tomatoes	Australia, Morocco, Philippines, U.S.A
Plums	South Africa
Peaches	South Africa

\*Sy and Wainwright, (1990).

It has been reported that compared to ethylene, high concentrations of acetylene are required to initiate the ripening process as  $0.01 \text{ ml.}1^{-1}$  of ethylene while  $1.0 \text{ ml.}1^{-1}$  of acetylene are required to initiate the ripening process of mango fruits. Acetylene in the concentration of  $0.1 \text{ ml.}1^{-1}$  delayed the ripening process compared to ethylene (Smith and Thompson, 1987).

Calcium carbide not only changes the skin color of the fruits but it also initiates the enzymatic action that breaks down the glucose resulting in a quick ripening of the fruits. Use of calcium carbide sometimes gives ripening color to a raw fruit. It also increases the shelf life and maintains the ripened color. Mango cv. Rataul were treated with different concentrations of ethrel (500, 1000, 1500 ppm) and calcium carbide. The results showed that fruits treated with ethrel 1000 ppm dip have better ripening attributes while calcium carbide-treated fruits showed greater incidence of rot and had a drastic decrease in ascorbic acid content (Pal., 1998b). The recommended way of application is to use a large vessel of water and added sufficient quantity of carbide and placing them in a well-ventilated chamber. Acetylene, which is then generated, is responsible for the fruit ripening as shown by the following equation (6.1) ( Sy and Wainwright, 1990).

$$CaC_2 + 2H_2O = Ca(OH)_2 + C_2H_2$$
 (6.1)

India is one of the major mango producing country and it also uses calcium carbide as a fruit ripening agent. Mann (1974) studied different doses of calcium carbide on mature hard green mango cv. Dashehri. The fruits were packed with calcium carbide and were moistened by a drop of water before being tightly covered with newspaper to prevent the leakage of acetylene. The fruits ripened within 8 days and the result showed that those fruits (4-5 kg), which were ripened with 2 g of calcium carbide, developed most desirable taste and flavor. Nagaraj et al., (1984) used ventilated wooden boxes with 2 g of calcium carbide kg<sup>-1</sup> of mango fruits, which were covered with straw and craft paper. The calcium carbide was removed after 96 hrs after which the fruit was kept in a well -ventilated room. The results showed that calcium carbide-treatments gave uniform yellow color and acceptable texture to the fruits. The treatment significantly reduced the number of days required for fruit ripening.

In Senegal, calcium carbide is commercially used to ripen banana and mangoes and fruits are harvested green. Two methods for calcium carbide application have been used. In one method fruits are placed in a basket made up of palm leaves and calcium carbide wrapped in a cloth or newspaper placed at the bottom of the basket. In order to increase the basket temperature and maintain the humidity level, the basket is covered with strong craft paper. The basket is placed in a closed room for 3-4 days and when the fruits developed a yellow skin color they are selected for sale. In another practice, a large heap of fruits (1 or 2 tons) is placed at the corner of a room with calcium carbide spread in several places and then the heap is covered with craft paper. The fruits ripened in both of these ways have good skin color with high acidity and low sugar content but poorly developed flavor (Sy and Wainwright, 1990).

In Malaysia, mangoes are picked slightly unripe and then calcium carbide is applied for artificial color development. In this method soapboxes or basket lined with banana leaves are used on which calcium carbide is sprinkled and then this box is filled to the top with the fruits and then the fruits are covered with more leaves on which calcium carbide is scattered. The fruits develop uniform yellow color within 2 or 3 days with a poor flavor (Berwick, 1940).

## Problems associated with using calcium carbide as a fruit-ripening agent

- i) Calcium carbide is produced by combining calcium oxide and carbon and when it combines with water it releases acetylene. Calcium carbide is generally used for gas welding because extreme heat is generated from this chemical. Acetylene is flammable and explosive even in a low concentration as compared to ethylene (Geesner, 1977).
- Acetylene gas is an analogue of ethylene and quickens the ripening process.
   Sometimes only the skin color is changed while the fruit remains unripe and raw. When a high dose of carbide is used on a raw fruit for ripening purposes it results in poor flavor of the fruit and possibly toxic.
- iii) Calcium carbide is considered as extremely hazardous as it may contain traces of arsenic and phosphorus Hydride (Delpierre, 1974). Early symptoms of arsenic and phosphorus poisoning include vomiting, diarrhea with or without blood, burning sensation of the chest and abdomen, thirst, weakness and difficulty in swallowing and speech. Other effects include numbness in the legs and hands, cold and damp skin and low blood pressure and in cases it can become fatal if not treated in time.
- iv) Acetylene gas had an unpleasant odor and produced a noticeable flavor in the treated fruits (Harvey, 1928).
- v) Acetylene is not only toxic to the fruits but it may be harmful to those who handle it.

vi) It has been noted that same weight of calcium carbide not always releases the same amount of acetylene. Stevenson, (1954) reported that in Australia 1 g of calcium carbide releases 312 ml of acetylene while according to Seymour (1984) in Sudan 240 ml acetylene librated from the same amount of calcium carbide while in Senegal, Medlicott, (1986b) calculated 150 ml acetylene generated per gram of calcium carbide. The liberation of acetylene depends upon the composition and size of the calcium carbide pieces.

The use of calcium carbide as a fruit ripening agent is harmful for human health as such nontoxic compounds like ethylene and MJ can be tested for fruit color development. It is important to establish their appropriate concentration and better techniques of application.

## 6.6 Conclusions

Use of ethylene and MJ for fruit ripening purposes are not harmful for human consumption but these compounds are quite expensive hence developing countries use low cost calcium carbide compound for such purposes.

Use of calcium carbide is harmful and has many disadvantages compared to ethylene. Acetylene is liberated from calcium carbide and for safety measure there is a need to investigate exact mode of action of acetylene for ripening of the fruits and optimum range of concentration and exposure time to the fruits. It is essential to control the delivery system of acetylene from calcium carbide, which must be safe and applicable to the wide range of users. It is important to develop new and better technique of application, which prevents direct contact of the substance with the fruits.

New compound, which are environmentally safe and not harmful for human health, must be discovered and tested.

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# VII. GENERAL SUMMARY AND CONCLUSIONS

The delicious and juicy mango is one of the world's most popular fruit. There are numerous varieties, including the cvs. Tommy Atkins, Kent, Keitt, and Haden which are grown in the US. Beyond being delicious, it is rich in vitamins, minerals and antioxidants. It has a short season and storage life. The fruits are generally not mature in a single consignment and proper storage of the fruits is essential for extending the consumption period. The fruits can be stored for 2-3 weeks in good condition. The shelf life of the fruit can be extended by storing them at low temperature. Chilling injury (CI) is a problem associated with low temperature storage of mangoes. CI symptoms include pitting, discoloration, internal breakdown, and scalding, failure to ripening and increased susceptibility to decay. It is important to improve and develop different postharvest techniques to reduce CI symptoms of the mango fruits. Therefore, experiments were performed by using different postharvest treatments on different mango cultivars in order to determine if postharvest treatments have the potential to reduce CI symptoms of the fruits when the fruits were stored below the critical temperature as well as when they were transferred to high temperature for further ripening.

Three different postharvest CI-treatments including hot water (HW) dip ( $50 \pm 2$  °C for 10 min), methyl jasmonate (MJ) dip ( $10^{-4}$  M for 2 min) and diphenylamine (DPA) dip (12 mM for 2.5-3 min) were used to assess and compare their potential to reduce the CI on mangoes cv. Kent when they were stored for 21 days at 1, 4, 7 or 10 °C. The parameters used to monitor the quality of mangoes were based on color values, pH, sugar content, texture characteristics, percentage of mass loss and percentage of good mangoes. The results were compared among the treatments and with the control fruits, which received no CI-treatment. The HWT showed no significant effect on the fruits and was not used in later experiments. The result indicated that MJ- and DPA-treatments were successful in retarding the incidence of CI symptoms in cv. Kent mangoes. Significant increase of mass loss was obtained when the fruits were stored at 10 °C. In case of cv. Kent mangoes, the best results were observed with MJ-treatment at 1 and 7 °C. DPA-treated fruits give best result when they were stored at 4 and 7 °C. So it is concluded that both MJ and DPA, when applied exogenously as a postharvest treatment, can reduce the

CI symptoms of mangoes cv. Kent and can increase the shelf life even when the fruits are stored below 10 °C.

In another set of experiment, mango fruits cv. Tommy Atkins were used and received the same CI-treatment as mentioned above, except for HWT. The fruits were stored at 1, 4, 7 or 10 °C for 18 days and then allowed to ripen at 20 °C for 4 days. The fruits were assessed for quality after storage as well as after ripening. The quality assessment parameters used after storage were color value, percent of mass loss, visual appearance and special emphasis was given to the presence of CI symptoms. Beside these parameters, total soluble solids and textural properties of the fruits were also determined at the end of the ripening process. The results indicated that fruits stored at 1°C showed excessive mass loss, irregardless if they received MJ- or DPA-treatment or were left untreated (control). Both MJ- and DPA-treatments efficiently reduced the incidence of CI symptoms in mango fruits cv. Tommy Atkins. Furthermore, these treatments did not interfere with the normal color development of fruits.

In the final chapter, use of different chemical compounds (ethephon, MJ, ethylene and calcium carbide) for ripening of mango fruit to enhance the fruit color development is discussed. Disadvantages of using calcium carbide for fruit color development were mainly focused, since it is toxic in nature, as such it is not commercially recommended.

Overall, this work demonstrates that postharvest application of MJ and DPA have the potential to reduce the incidence of CI symptoms in mangoes cvs. Kent and Tommy Atkins even when they are stored below recommended temperatures.

Further research in this area is needed to find out the most appropriate concentration of these chemicals and their exposure time which efficiently reduces the CI symptoms in mango fruits and extend their shelf life. More research is required to assess the various storage and ripening techniques on other popular mango cultivars.

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