# EFFECTS OF DIFFERENT EXPOSURE METHODS TO 1-METHYLCYCLOPROPENE ON QUALITY OF PARTIALLY RIPENED BANANAS

by

MANSI TRIVEDI

A Thesis submitted to the

Graduate School-New Brunswick

Rutgers, The State University of New Jersey

in partial fulfillment of the requirements

for the degree of

Master of Science

Graduate Program in Food Science

written under the direction of

Professor Kit L. Yam

and approved by

_____

_____

_____

_____

New Brunswick, New Jersey

January, 2012

## ABSTRACT OF THE THESIS

Effects of Different Exposure Methods to

1-Methylcyclopropene on Quality of Partially Ripened Bananas

By MANSI TRIVEDI

Thesis Director:

Professor Kit L. Yam

1-Methylcyclopropene (1-MCP) is being widely used as an ethylene antagonist to suppress ethylene induced ripening in fresh produce. It is commercially available in encapsulated form in cyclodextrin, which requires moisture triggers to release from encapsulation. There are several reports on efficiency of 1-MCP for inhibiting the action of ethylene during green life (mature – but unripe stage) of bananas, but the commercialization of 1-MCP application for bananas is still under the area of research due to inconsistence responses received by researchers on its effect. Also, there are limited studies showing its effects on yellow life (at and after partially ripened stage) of bananas, so the further investigation in this area was our subject of interest. In this study, different 1-MCP exposure methods were used to treat bananas to provide the scientific base for developing its commercial application. The overall objective of this study was to provide the better understanding for extending Controlled Release Packaging (CRP) system that can deliver the 1-MCP molecules from the package over longer period of time to increase the yellow life of bananas to

maintain greater quality at consumer market.

The study was divided into two parts: in the first part, the packaging system was used to treat partially ripened bananas with different 1-MCP exposure methods. The physiological responses of partially ripened bananas to these different 1-MCP exposure methods, controlled exposure (timed release - slow release for longer time) and one-time exposure, were studied. All experiments were conducted on Cavendish bananas (Dole) at partially ripened stage (ripening color stages 3 and 4). In the second part, the feasibility of 1-MCP to be incorporated in the CRP system was studied by controlling 1-MCP release through polyvinyl alcohol (PVA) film and studying its release from cyclodextrin through banana transpiration.

The physiological responses showed that both the 1-MCP exposure methods were effective to delay ripening of partially ripened banana by at least 5-6 days. But the one-time exposure method was more effective than controlled exposure after 6 days. The PVA was able to delay the release of 1-MCP: the release of 1-MCP through PVA was nearly 15 % in 6 hours, whereas the release of 1-MCP from cyclodextrin (control- without any film) was 100% in 6 hours. The bananas were able to provide sufficient moisture through transpiration to initiate 1-MCP release from cyclodextrin within the first two hours of the experiment.

## ACKNOWLEDGEMENT

My thesis was long journey with roller coaster ride at every steps of it, which I would have never finished without the support of some special people. I have learnt the different lessons from these special people, which not only helped me to construct my thesis, but it also helped me to construct my thinking ability to a different level. I would like to thank all these people without whom I would have not progressed in my life.

First a very special thanks to my advisor Dr. Kit L. Yam to tolerate me with my vacuumed mind. He has not only tolerated me but he has filled my mind with appropriate knowledge. His techniques of teaching were always different from the others. He has explained the science principals using philosophical analogies to make it easier. He does not believe in spoon feeding, instead he taught me the logical building skills, which enhanced my critical thinking ability. Apart from educational knowledge, he improved me in every aspects starting from communication skills to a complete professional presentation skills.

I am also very grateful to Dr. Nazir Mir, who served me as co-advisors even without any affiliated duties. He made himself available for my each questions even though he was always overloaded with his busy work schedules and heavy travels. The industrial visit with him had provided me a practical knowledge for conducting my experiments.

I owe my deepest gratitude to Dr. Dong Sun Lee, the visiting professor, who was

always available for any help, even without prior notice. He made all the calculations of fundamentals and engineering easier throughout my experiments.

I would like to thank my committee members Dr. Henryk Daun and Dr. Thomas Gianfagna for giving me the proper directions of writing and correcting my work wherever needed.

I have made a special friend, a guide, a sister during this journey. Thank you doesn't seem enough, but it is being said with great respects and love. Thank you so much Aishwarya for being with me always, you have made a very special space in my life.

I would also like to acknowledge the support of my student colleagues, who have supported me and encouraged me throughout my master's programme. They have always mirror backed my ideas, I could have never asked for the better friends cum family away from my family. I am indebted to Luni, Spurti, Saumi, Xi, Sai, Tulsi, Lakshmi, Peng, Saif, Yuthia, Simon, and Sunny for their support and encouragement.

I would also like to thanks my sister's friend Shana Hunter to help in rephrasing my writing.

I would also like to show my gratitude to my sister, brother and my in-laws for their continuous encouragement and loving support. Manank my husband without whom, I would have never finished this journey. He tolerated all my frustrations, granted all my wishes to make my life easier. He did all my shares of work to provide me most free time to work on my thesis. He started me to work all over again when I was losing the confident. I owe my loving thanks to my parents, who have been seeing me graduating since the day one. They have made sure that I never stay without any

resources required in my life till date. My dad has compromised his every joy to full fill all my wishes. My mom’s love is biggest courage of my life.

Last but not the least the almighty god for letting me through all the tests of life during this journey.

## TABLE OF CONTENTS

## LISTS OF TABLES

## LIST OF ILLUSTRATIONS

# 1. INTRODUCTION

## 1.1. Background

Fresh produce, especially fruits and vegetables, are considered an important part of our day to day diet because they are a major source of vitamins, minerals, organic acids, dietary fibers and also antioxidants. According to the food guide pyramid, a balanced diet should include at least 2-4 servings of fruit every day [1]. The consumption of fruits and vegetables has increased recently with greater consumer awareness about the health benefits of fresh produce over processed foods. Fruits and vegetables are highly perishable commodities, so proper post harvest handling is required to avoid unwanted losses and to retain the freshness and quality. During long distance transportation and distribution, risk of post harvest losses may increase, and therefore proper care and handling are being emphasized in recent years for post harvest commodities [2]. There are several causes of post harvest losses, including increased respiration rate, hormone production (i.e. ethylene), physiological disorders, general senescence, compositional and morphological changes. But the excess of ethylene (plant growth hormone) production mainly responsible for higher post harvest losses, especially for climacteric fruits. For this study, bananas have been chosen as the model for several reasons.

Bananas are considered as a staple fruit worldwide, and it is very common everyday fruit for most countries. Bananas are a typical climacteric fruit that exhibits a marked peak in ethylene production with a rise in respiration during the ripening process.

Ripening of climacteric fruit is either triggered by natural production of endogenous ethylene or by exogenous ethylene application [3-5]. The ripening is mainly triggered through the action of ethylene binding to receptor sites located on plant cells. The shelf life of a bananas is thereby reduced with an increase in available ethylene, and its binding with continuously forming plant receptor sites [6]. For this reason bananas are harvested at a mature-green stage for exporting, transporting, and distributing to avoid unwanted ethylene production [7]. When bananas are harvested at the green stage, they lose their ability to ripen naturally or they may show uneven ripening. Therefore, the exogenous ethylene treatments are used at storage facilities to initiate the ripening process and to confirm the even ripening, before distributing to consumer markets (Dr. Nazir Mir, personal communication). Commercially, once bananas are induced to ripen with exogenous ethylene their marketing life is only about 3-5 days, depending on ethylene treatment conditions and the holding temperature after the treatment [8].

In recent years ethylene antagonist agents for blocking the action of ethylene at the receptor level have provided promising results for controlling ripening and other ripening associated changes [9]. 1-Methylcyclopropene (1-MCP) is the well known ethylene antagonist which suppresses ethylene action by blocking ethylene receptor sites [6, 9-11]. The affinity of 1-MCP for the ethylene receptor is approximately 10 times greater than that of ethylene [10]. There are several reports on the efficiency of 1-MCP on inhibiting the action of ethylene on the green life of bananas and some combinations based on 1-MCP concentration, duration of treatment, and temperature

have been addressed [4, 7, 8, 12-15]. There is no commercially established technique found for treating bananas with 1-MCP. A common technique used to treat fresh produce (generally for all type of produce) with 1-MCP is by exposing fresh produce for several hours to a fixed 1-MCP concentration in a controlled room [16]. For bananas, generally the same procedure is being used by researchers to treat them at the green stage, before any exogenous ethylene application, which is found to be effective to extend the green life (mature – but unripe stage) of banana. But, there are limited research studies showing its effects on yellow life (at and after partially ripened stage) of banana. A method to slow down the ethylene induced ripening of bananas by cooling to 14 °C and using Modified Atmosphere Packaging (MAP) processes have shown promising results [8, 17, 18]. But bananas have to be repacked after the ethylene treatment into a polymeric film within which the appropriate modified atmosphere will be established. Due to the wide variation in respiration rates of fruits, and the different permeability of packaging, the MAP is not a feasible independent technique for commercial application [18]. Treatment with 1-MCP seems to be a more convenient method since repacking would not be required. Hence, there is the need for an alternative technique which can provide continuous exposure of 1-MCP to bananas to further delay ripening even after partially ripened stage. A novel technology known as Controlled Release Packaging (CRP) is being used for delivery of antioxidants and antimicrobials, which can be further extended for delivery of ethylene antagonist from the active packaging layer to delay the ripening of bananas. Before establishing the CRP system, the study of physiological

responses of partially ripe bananas to time release (controlled exposure) of 1-MCP and testing its effects on bananas in the packaging system is required.

### 1.2. Outcome for the industry

The overall aim of this work is to develop a different 1-MCP exposure technique for improving the quality and shelf life of fruits. This study will provide the scientific base for designing a Controlled Release Packaging (CRP) system for commercial application; and in part will investigate the role of 1-MCP on the quality of partially ripened bananas in the packaging system. This investigation will open the opportunities for extending a CRP system for delivery of 1-MCP to delay the ripening and maintaining the quality of fresh produce for longer period.

# 2. LITERATURE REVIEW

## 2.1. Banana

Bananas are the model for this study due to a combination of scientific and agricultural reasons. They have a typical climacteric pattern for ethylene production and respiration rate, and exhibits ripening by change in color, flavor, aroma, texture and other physiological characteristics [4]. So, it is very easy to observe the ripening and quality associated changes during the study.

Nutritionally, fresh bananas are a good source of carbohydrates, protein, fibers with ultimately a good amount of calories, and a low fat content. It contains approximately 35% carbohydrates, 6-7% fibers, 1-2% proteins, and also contains essential elements such as potassium, magnesium, phosphorus, calcium, iron, and vitamins A, B6, and C [7, 19].

Since 1981, banana production (worldwide) has increased from 40 million metric tons (MT) to 72.4 million MT, and the current report of the Food Agriculture Organization of the United Nations has estimated the banana production to be almost 83.4 million MT (FAO, 2008) [7]. In 2008, almost 75% of banana production came from six major banana producing countries out of 130 banana producing countries (FAO, 2008). There is no reported production of bananas in the United States (US). Therefore, there is a huge market for bananas imported into the US from Latin America, Mexico, and other banana producing countries.

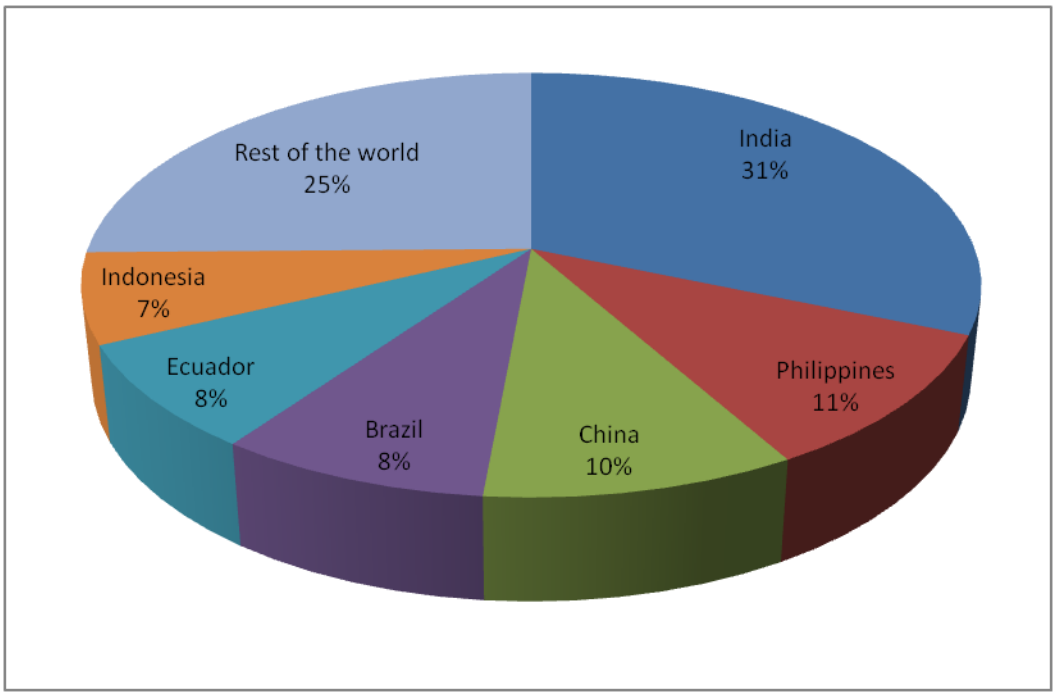



**Figure 1: Distribution of banana production in 2008 (FAO, 2008)**

### 2.1.1. Climacteric and non-climacteric fruit

Fruits are classified into two categories based on their respiratory pattern and capacity of producing ethylene after harvest. Fruits with capability of producing ethylene auto-catalytically after harvest, and which exhibit the peak of respiration rate during ripening are called climacteric fruits. In the case of climacteric fruits, both the respiration rate and ethylene production start increasing with the initiation of ripening. Fruits such as apples, bananas, tomatoes, and avocados fall into the category of climacteric fruits. After harvest, if exogenous ethylene is applied to these fruits, it accelerates the ripening process.

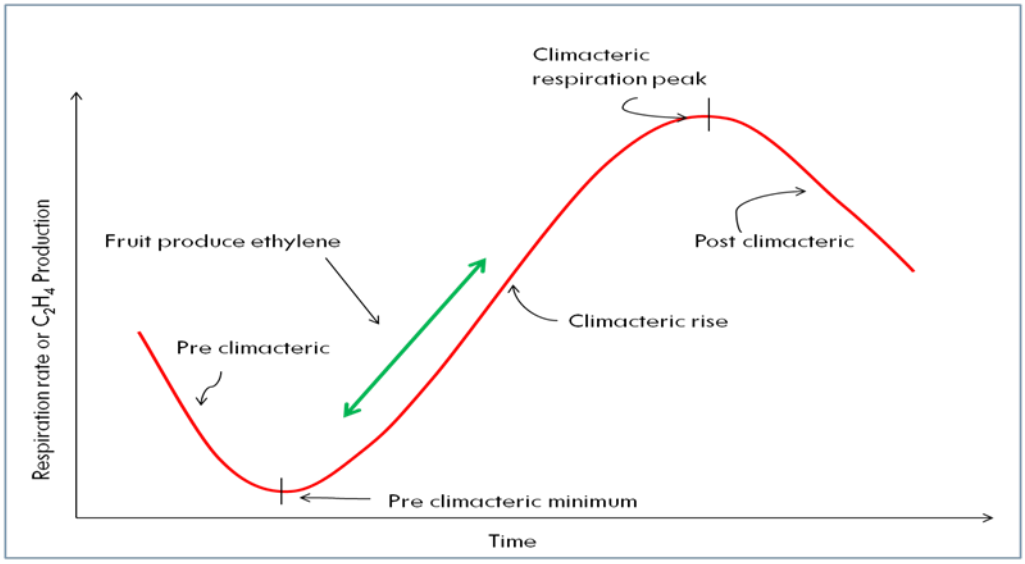



**Figure 2: Phases of the climacteric period showing trend of $CO_2$ and ethylene production during growth and development of climacteric fruits, [20]**

The figure above shows the different phases of the climacteric period with respect to respiration rate and ethylene production during the growth and development of the climacteric fruits. According to [20] the climacteric period can be defined as the development period of some plant parts during which the series of biochemical changes occurs, which are associated with respiration rate and autocatalytic ethylene production. During the peak climacteric phase, ethylene production and respiration rate both reach the maximum levels to triggers the ripening, which is accompanied by change in composition, color, texture and other sensory attributes of the fruits. This is then followed by senescence of fruits during post climacteric phase [20].

Fruits which do not show the sizable increase in the ethylene production, and respiration rate during ripening are called non-climacteric fruits [21-23]. For non-climacteric fruits, ethylene production and the respiration rate does not exhibit any sudden increases, so when their respiration rate is compared to climacteric fruits, it is very low. Also, they cannot produce ethylene auto-catalytically [24]. Fruits such as

strawberry, blueberry, cherry are categorized as non-climacteric fruits.

### 2.1.2. Ripening process of banana

Ripening brings a series of biochemical changes, which are responsible for the change of color, pigment formation, starch breakdown, textural changes, volatile and aroma development, and finally abscission of fruit [21]. During ripening, peel color of banana changes from green to yellow and followed by browning. There are mainly two pigments responsible for peel color change of banana, chlorophyll and carotenoid [25]. The peel color of banana is one of the important visual quality indicators being observed by consumer to determine the eating quality. During ripening firmness of banana decrease, this can also be used as a quality indicator. The softening of banana mainly caused by the enzyme activities in cell wall which involves, polygalacturonase (PG), pectin methyl esterase (PME), pectate lyase (PL) and cellulose, and activities of these enzymes are mainly ethylene dependent [23]. The flavor development is also an important indictor to differentiate the ripe and unripe fruit. As banana ripens, the starch hydrolyzed to sugar to enhance the edibility and gives sweet flavor. The hydrolysis of starch is mainly catalyzed by activity of α-amylase and β-amylase [26].

### 2.1.3. Respiration behaviors of banana

Respiration plays a major role in maintaining the quality and shelf-life of fresh produce. When harvested produce respires, it takes oxygen in and gives off heat, moisture and carbon dioxide. The storage conditions play an important role in

controlling the respiration rate of bananas. The optimum temperature for banana storage is suggested as 13 °C to 14 °C with relative humidity of 85-90 % [27]. It is well understood from the studies that low oxygen and high carbon dioxide atmospheres are suitable for extending the storage life of climacteric fruits, for bananas these levels are suggested as 2-5% $O_2$ and 2-5% $CO_2$ [4, 18, 28-30]. $CO_2$ level higher than 7-8 % and $O_2$ levels below 1% may cause undesirable change in texture and flavor of bananas [27].

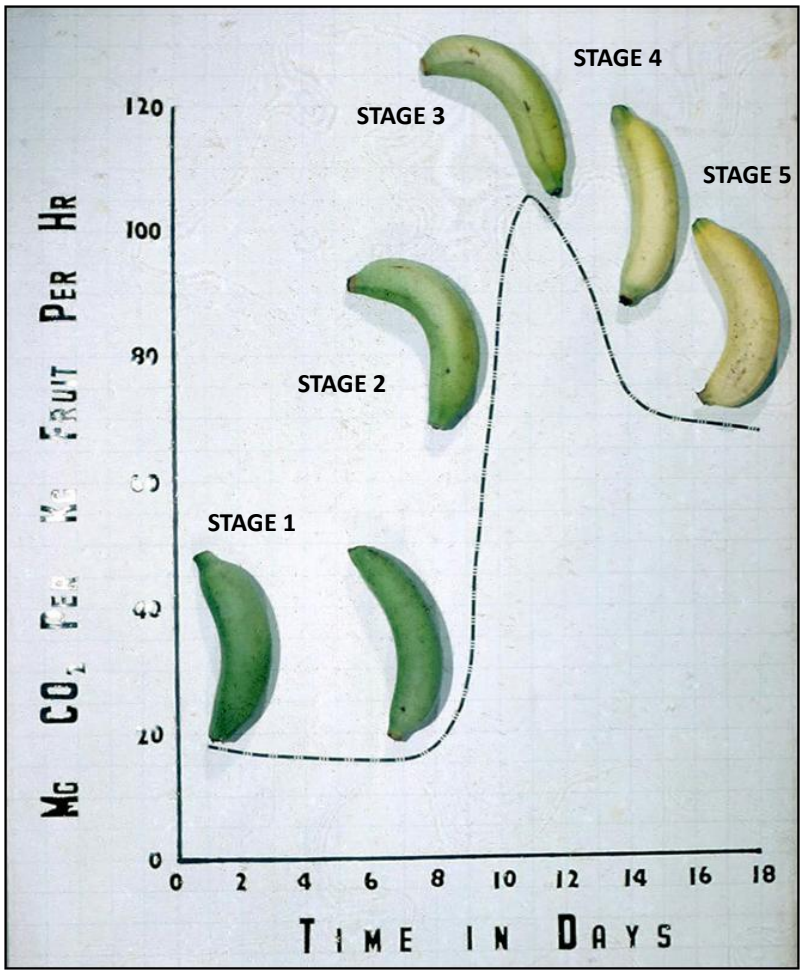



**Figure 3: $CO_2$ production by bananas during ripening process, (Adapted from: [31])**

As mentioned earlier bananas follows typical climacteric fruit; which shows a sharp

peak in ethylene production with increase respiration rate to onset the ripening process, which is followed by volatile production at post-climacteric phase. [3, 4, 28].The figure 3 above shows the typical carbon dioxide production rate of banana during different stage of ripening.

## 2.2. Ethylene

Ethylene is a naturally produced plant hormone, primarily responsible for the ripening of climacteric fruit and several processes associated with ripening. Ethylene is a simple gaseous hydrocarbon with molecular formula $C_2H_4$. It can easily diffuse in and out of the plant tissue from exogenous as well as endogenous sources [32]. The ethylene production induced during several growth stages of plant such as germination, ripening of fruits, abscission of leaves, and senescence of flowers. The action of ethylene causes dramatic changes in the color, flavor, and aroma of fruits during the ripening process [5, 33]. The quantity of ethylene production is highly dependent on the type of plant organ or tissue [34]. Plant cells contain ethylene binding receptors, an ethylene receptor is a protein that sits on the cell membrane and has a site for binding ethylene on the outside of the cell, which chemically reacts with ethylene and triggers ripening [35].

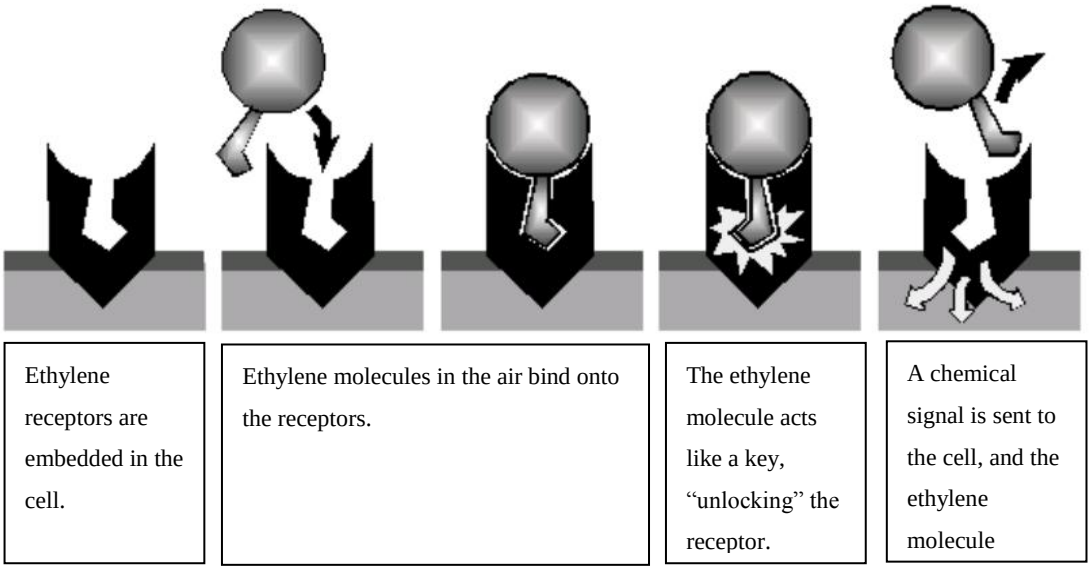
Ethylene can greatly affect the quality of harvested produce. It can be advantageous or deleterious depending on the produce, its ripening stage, and its desired use [32, 36]. Ethylene production is greatly affected by storage temperature of produce, the ethylene production generally reduced at low temperatures. However, a lower

temperature can cause chilling injury in chilling sensitive produce like banana and can enhance the ethylene production. Excess ethylene gas produced during stress like situations including senescent breakdown of fruit, chilling-related disorder, ethylene –induced disorders can cause superficial scald (e.g. in apples), browning (e.g. internal flesh browning of avocados, pineapple), undesirable chemical changes (e.g. isocoumarin in carrots, water-soaking of water melon) softening of tissue, and many other negative effects in produce [32, 37]**.** Fruits are highly perishable commodities, from the moment they are picked. They require proper ethylene management in post-harvest handling to maximize freshness, quality, and shelf life from the field to the table. To slow down the ripening process of fresh produce, we need to inhibit or slow down the action of ethylene gas. Thus, there will be slow ripening due to less available ethylene [38, 39].

### 2.2.1. Mechanism of ripening by ethylene

Ethylene receptors are embedded in the cells of fruits, the ethylene molecules in the air bind to the receptor sites and act like a “key” to unlock them. The receptor sites, then sends a chemical signal to the fruits’ cells to perform a series of chemical reactions[40, 41]. These chemical reactions result in the ripening of the fruits by changing the color, flavor, aroma, and composition of fruit (water content, starch content, sugar content

etc.).




"Binding of ethylene molecule with the receptor "unlocks" the receptor and leads to a chemical reaction in the plant tissue"

**Figure 4: Ethylene binding mechanism, (Adapted from: )**

### 2.2.2. Ethylene and ripening of banana

As mentioned earlier, bananas are harvested at a mature-green stage for transportation purposes to non-banana producing countries. Due to early harvesting at the green stage, bananas lose their ability to ripen naturally or they may shows uneven ripening. Commercial ripening with exogenous ethylene is common practice to treat bananas in storage rooms to ensure even ripening before distributing them to the consumer market. The ethylene treatment is given in a controlled room with fixed ethylene concentration, at ambient temperature and relative humidity required to ripen bananas from stage 1 to stage 2.5 - 3 [42]. After which the yellow life (at and after partially ripened stage) of banana is 3-5 days at the consumer market. The most identifiable difference in ripe and unripe banana is the color; it is the first indicator being used by a consumer to differentiate between unripe, and ripe, or over-ripe bananas. For this study, banana ripening color chart [43] was used as a

reference for ripening color stage identification. Most of the consumers prefer to eat bananas at stage 5-6, so maintaining this ripening stage at the consumer market for a more than the 3-5 days can be economically more beneficial.

2.2.2.1. The current system used to treat banana with ethylene at storage facilities

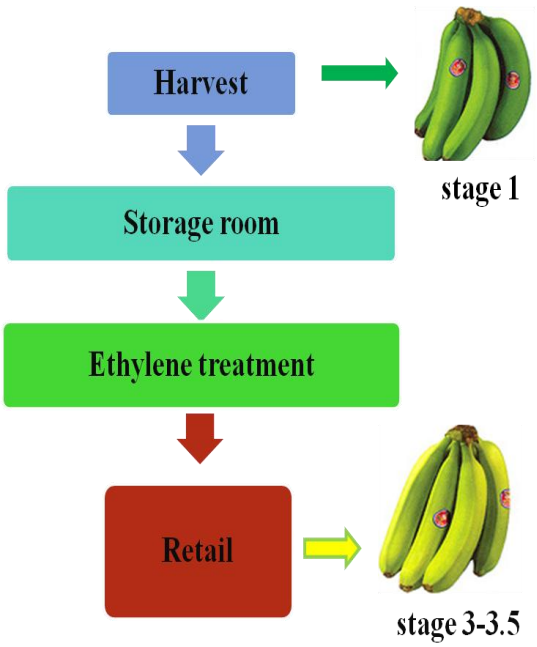



**Figure 5: Flow chart showing commercial treatment of bananas with exogenous ethylene at storage facilities, [36, 42]**

Figure 5 displays the commercial way of treating bananas with exogenous ethylene. This method is generally used to initiate the ripening of some of the climacteric fruits including bananas. Commonly 10-150 µL/L of ethylene is applied to fruits at 15-25º C in ripening rooms at storage facilities [36, 42]. For this exogenous ethylene application, ethylene is used in gas form from a cylinder of compressed ethylene gas. The gas required for the treatment in storage rooms is either being diluted in the atmospheric air using fans or being generated from the catalytic decomposition of

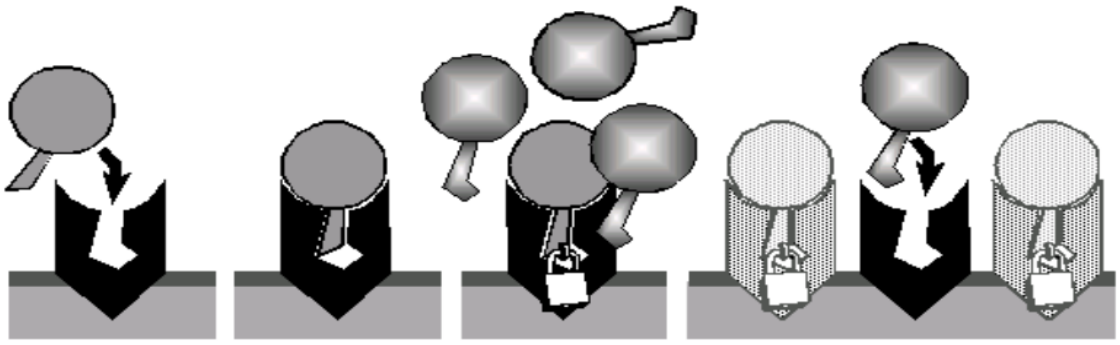
ethanol [36].

### 2.3. 1-Methylcyclopropene (1-MCP)

1-MCP is a novel compound that can prolong the shelf life of fresh produce by inhibiting the action of ethylene at receptor level [9-11, 32]. 1 MCP is a gaseous compound with the molecular weight of 54.09 at standard temperature and pressure. Its chemical formula is $C_4H_6$. Commercially, it is registered under trade name of SmartFresh™ or EthylBloc™ by Agrofresh Inc. / Rohm and Haas Company (Philadelphia, USA) [44]. 1-MCP is a highly volatile gas, thus commercially it is available in an encapsulated form in α-cyclodextrin (CD). The appropriate amount of water or KOH buffer are required as a trigger to release this encapsulated 1-MCP gas molecules [32, 45]. 1-MCP has been successfully used on flowers, some fruits, vegetables, and plotted plants, but its efficiency varies for various products. 1 -MCP (EthyleBloc™) use was approved for ornamental plants in 1999 by Environmental Protection Agency (EPA). The commercialization of 1-MCP for edible horticulture products was approved in 2002 by EPA. Apples were the first crop to receive registration for receiving 1-MCP treatment commercially [32, 36]. Then gradually pears, tomatoes, melons, and some other fruits have received an approval from EPA for using 1-MCP. But still 1-MCP use has not been commercialized for bananas, due to inconsistent responses received by researchers for its effect on banana. Therefore, more investigation needs to be done to identify the effect of 1-MCP on bananas for its commercial use.

### 2.3.1. Mechanism of blocking ethylene receptor sites

1-MCP reacts with the ethylene receptor and inhibits the action of ethylene. When 1-MCP molecules sit on ethylene receptor sites, it binds the receptors sites and does not allow the receptor to "unlock" like the ethylene molecule does. Therefore, no signal can be sent for a chemical reaction, which delays the further ripening. But as mentioned earlier, ethylene and receptor site formation are a continuous process, and 1-MCP does not bind the receptor site permanently. So, eventually new receptor sites can be formed and ethylene can regain its sensitivity for them, once the entire available 1-MCP molecule has been used up to block available receptor sites. See the figure below to understand the mechanism of 1-MCP blocking ethylene receptor sites[41].




"When 1-methylcyclopropene (1-MCP) binds to the ethylene receptor, it does not "unlock" the receptor and remains locked to the receptor preventing the binding of ethylene and the chemical reaction does not occur"

**Figure 6: Mechanism of 1-MCP blocking ethylene receptor sites, (Adapted from: [41])**

### 2.3.2. Effect of 1-MCP on delay ripening of bananas

The ability of 1-MCP to delay ripening of mature-green, pre-climacteric bananas have been shown widely in studies [9, 12]. The wide range of 1-MCP concentration ranging from 0.1 $nl \cdot L^{-1}$ to 1000 $nl \cdot L^{-1}$, as well as different application durations ranging from 1 to 72 hr. have been tested and reported [3, 4, 8, 11, 13]. Most of the literature have mentioned the effects of 1-MCP on green bananas, but studies showing treatment of 1-MCP on partially ripe bananas are limited and no studies have shown effect of 1-MCP treatment on specific stages of ripeness [8, 13].

### 2.3.3. Effect of 1-MCP on respiration rate of bananas

Ripening of climacteric fruit is accompanied by a peak in respiration and also by increasing ethylene production [46, 47]. Various researches have been done on 1-MCP and its effect on ethylene suppression, and thereby quality retention with longer shelf life of banana. But there are limited studies showing the relation of 1-MCP and respiration, when a 1-MCP treatment is given at a later ripening stage, such as stage 3 or 4. Some studies have also shown a respiration pattern of typical climacteric fruits during peak of ripening with increased ethylene and respiration rates, which has been temporarily suppressed by 1-MCP [3, 4, 22]. Particularly, there is no data available showing the effect of 1-MCP in controlling the respiration rates of partially ripened bananas. So, there is further research required to investigate the role of 1-MCP gas in altering respiration rates of bananas at partially ripened stages.

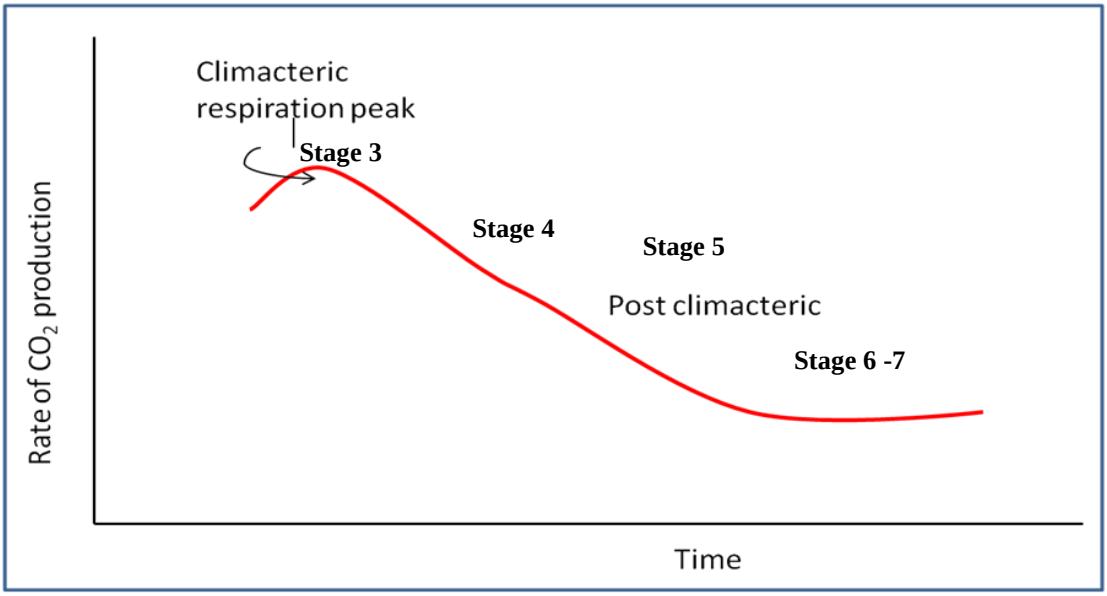



**Figure 7: $CO_2$ production trend at and after partially ripened stage of bananas, [20]**

Figure 7 has focused on the climacteric and post climacteric phase of typical climacteric fruits, which has been used in our study to understand the trend of $CO_2$ production at and after ripening stage 3 of bananas.

## 2.4. Summary

### 2.4.1. Current system for 1-MCP application

The current way of treating fresh produce with 1-MCP is by exposing it to 1-MCP at the desired concentration for several hours (1- 72 hr) depending on the requirements; or by immersing the produce in an aqueous solution of 1-MCP for the required time at storage room facilities. There is no commercially established method found for treating bananas with 1-MCP, therefore a similar 1-MCP treatment method as for treating other produce is generally used to treat bananas by researchers at laboratory level. This treatment is followed by an exogenous ethylene treatment for resuming

the ripening process under a controlled environment before sending the bananas to consumer markets. Generally ethylene treatments can be given before or after the 1-MCP treatment. At research laboratories 1-MCP treatments are given by immersing produce in an 1-MCP aqueous solution, or by injecting gas inside an enclose system like glass chamber or tent for a fixed time (6-12 hrs generally) with the desired concentration, followed by storing in an incubation chamber maintained with desired RH and temperature, or by leaving open to room temperature or required temperature for the produce [40, 48-50].

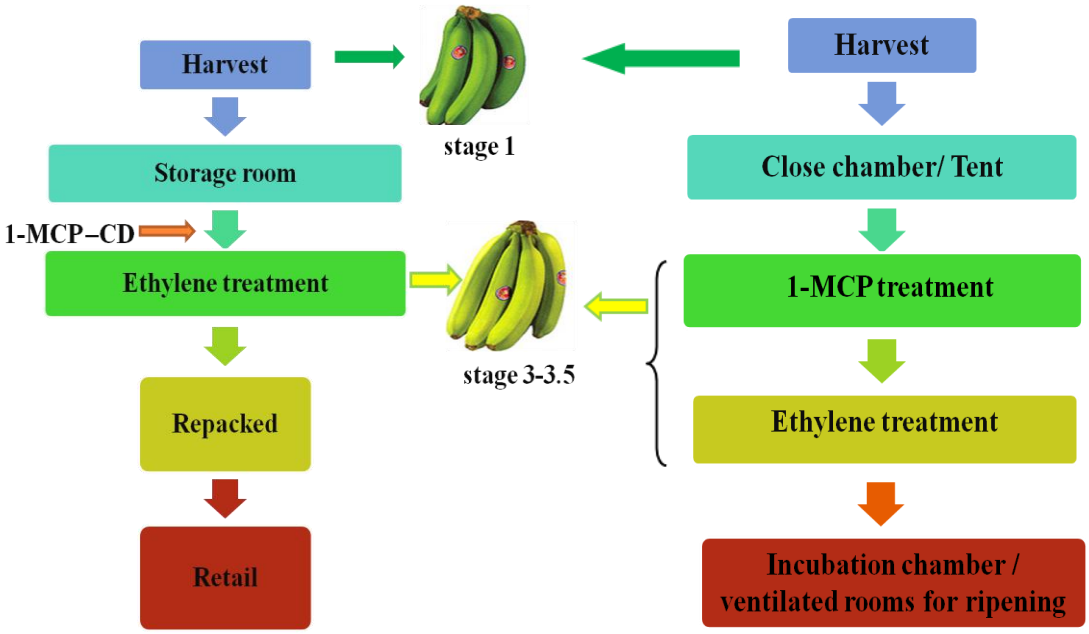



**Figure 8: The current system used to treat fruits with 1-MCP at storage facilities and laboratory (using banana as model), [4, 13]**

Implications of 1-MCP treatments level listed in published literature have shown effects for only the short period of time, until continuous dosages are applied [45]. There is no data found on the effect of 1-MCP with continuous dosage application, especially for climacteric fruits like bananas, which immediately start ripening after

ethylene treatment. The continuous dosage application would be a great alternative technique for fresh produce which continuously forms the receptor sites during the entire ripening process. Thus, there is a need for an alternative system which can provide the continuous replenishment of 1-MCP to extend the green life, as well as the yellow life, of bananas without long tedious procedures throughout the ripening process. Such systems are available for delivery of antioxidants or antimicrobials from packages, generally known as active packaging or Controlled Release Packaging (CRP). The researcher group [51] have reported to release 1-MCP from package sachets and group [16] have reported to release 1-MCP from different heat pressed polymer films. But no significant data has been found on the effect of the controlled (Continuous) release of 1-MCP from packages on fruits directly, and also no data has been found on what are the effects of controlled exposure of 1-MCP on fresh produce except [45], who tested the effect of continuous exposure of 1-MCP on the ripening quality of tomatoes. Therefore, first there is a need to study the effects of controlled exposure of 1-MCP on fresh produce to extend the CRP technology for continuous replenishment of 1-MCP from the package itself.

Also, in all of the published literature mentioned above, are showing the effects of 1-MCP on green life of bananas only. There is a lack of evidence showing the effects of 1-MCP on yellow life of bananas. As mentioned earlier for exporting purposes, bananas are harvested at the green stage and transported under controlled conditions to storage facilities, where they are treated with exogenous ethylene to resume the ripening process. This eventually shortens their shelf life to 3-5 days at the consumer

markets. In case of non-banana producing countries that require long distance transportation, the 1-MCP treatment after ethylene treatment would be advantageous. It would be more beneficial to have another dose of 1-MCP after bananas reach the ripening stage 2.5-3 at storage facilities to increase yellow life of banana at distribution centers and consumer markets. A technique which can help to extend the yellow life of bananas would be a better alternative to get the desired quality of produce at consumer market. Also, the storage room process is tedious and requires the constant atmosphere for several hours for the 1-MCP treatment that also increases the cost of treatment. Packaging bananas with 1-MCP releasing film would be an alternative to storage room application of 1-MCP, which can be executed by means of the CRP system. This system can be applicable at both the mature-green stage and the partially ripened stages of fruits.

### 2.4.2. Concept of controlled release packaging system

The Controlled Release Packaging (CRP) concept is designed to achieve a slow release (timed delivery) of active compounds over longer periods of time. CRP uses the packaging as a delivery system to release an active compound from packaging material to food in a controlled manner [52, 53]. Until now this technology has been successfully used for delivery of antioxidants and antimicrobials, it can be further extended to other active compounds such as ethylene antagonist (1-MCP) to serve fresh produce. For CRP of banana, bananas can be packed in 1-MCP containing active films or package containing 1-MCP sachets immediately after harvest. From which release of 1-MCP can be controlled in such a way that it releases 1-MCP gas

slowly during the entire banana transportation process, starting from the green life until the yellow life at the consumer market. The release of 1-MCP from the package can be varied by several factors including internal environment of the package, polymer type used to develop the package, quantity of bananas per package, etc. The CRP system can be designed by using the appropriate polymer for slow release of 1-MCP to ensure the availability of 1-MCP throughout the distribution process. This can continuously release 1-MCP to extend the shelf life of bananas during entire transportation and distribution process. It can also be designed to release 1-MCP during storage and local distribution to extend the yellow life at the consumer market.

Before establishing the CRP system, there is an obvious need to evaluate the effects of controlled release of 1-MCP on the quality of bananas treated in the packaging system instead of storage rooms. The figure below shows the process flow for the treatment of produce with 1-MCP within the package.

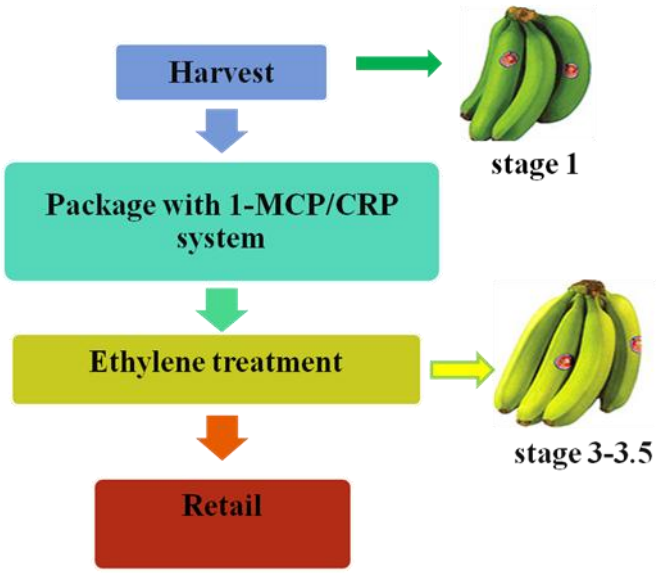



**Figure 9: Flow chart showing process of the 1-MCP application method in the package system**

# 3. OBJECTIVES

## 3.1. Long term objective

Our long term objective is to develop the controlled release packaging (CRP) system for delivery of 1-MCP to fresh produce to further delay its ripening. As part of the long term objective, this thesis has focused on evaluating the effects of different 1-MCP exposure methods given in the package to fresh produce. This study will provide an alternative for the current methods of treating fruits in storage rooms or controlled environment chambers to further extend the shelf life of fresh produce with better quality.

## 3.2. Objective for this study

The overall goal of this work is develop a different 1-MCP application system that will allow improving the current 1-MCP application to further improve the quality and shelf life of banana. This study will focus on developing the scientific base for designing a controlled release packaging (CRP) system and to investigate the role of 1-MCP for delaying further ripening of partially ripened bananas.

Objectives of this work are as follows:

1) To test the effects controlled exposure (continuous replenishment) and one-time exposure of 1-MCP given in the packaging system on quality of partially ripened bananas (yellow life).
2) To test the feasibility of 1-MCP to be incorporated in a controlled release packaging system by studying the followings:

- Release of 1-MCP from cyclodextrin through polyvinyl alcohol (PVA) film
- Release of 1-MCP from cyclodextrin through banana transpiration

The following figure shows the conceptual illustration of CRP system drawn for fresh produce.

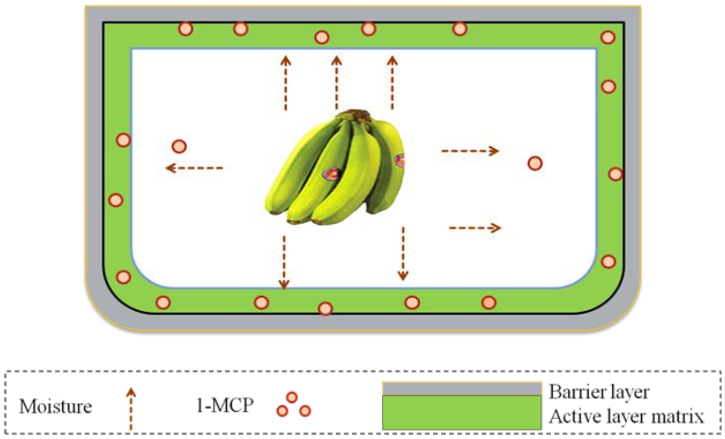



**Figure 10: Conceptual illustration of controlled release packaging for fresh produce, [53]**

### 3.3. Different exposure methods of 1-MCP

In this study two different 1–MCP exposure methods, one-time exposure and controlled exposure, were tested on partially ripened stages of bananas and responses were measured by quality analysis.

#### 3.3.1. One-time exposure

We have defined the term “one-time exposure” as “the injection of the total amount of active compound (1-MCP) needed for treatment are given at time zero, followed by no additional exposure”. 1-MCP was injected inside the permeable package containing fruits and the package was kept intact for entire duration of the

experiment. The idea of the one-time exposure application was obtained from "instant addition of the active compound –Active Packaging" with the assumption that the 1-MCP gas dosages directly applied in the package or released from the package may be able to be maintained through the transportation, and will be more beneficial than the current method of treating fruits in storage room after exogenous ethylene application. We have tried to mimic the active packaging system in terms of one time exposure.

### 3.3.2. Controlled exposure (timed delivery)

The controlled exposure is defined here as the "total amount of active compound needed is given in several doses at fixed timed intervals to provide the continuous replenishment of the active compound to mimic the controlled release from the package." There are several methods to give the controlled exposure treatment of 1-MCP to fresh produce. For example, 1-MCP gas can be delivered through syringe to the package or closed system in a timely manner; or by modifying the release of 1-MCP from a sachet containing 1-MCP or active film incorporated with 1-MCP, depending on the gas and moisture barrier property of polymer used for the sachet or active film. Here we have focused on the first method by injecting 1-MCP gas through a syringe over a period of time. Here the assumption was made that continuous replenishment of 1-MCP may inhibit the action of continuously forming ethylene receptors and can further delay the ripening. This assumption was supported by the hypothesis of [45], the "implication of the 1-MCP dosage applied in published literature are only effective for initial moments of the study, until

continuous dosages are applied". In this system we have mimicked the controlled release packaging system, to see if the technology is transferable and effective to gaseous compounds like 1-MCP and fresh produce systems.

### 3.3.3. Concept of one-time exposure and controlled exposure application

The purpose of one-time exposure and controlled exposure applications is to provide an alternative for existing methods of 1-MCP application. This application involves the exposure of fruits to several doses of 1-MCP or a single dose of 1-MCP using the injection method inside the package to maintain the 1-MCP environment. The concept developed for this thesis is based on a controlled release packaging system. Figure 11 below illustrates the permeable packaging system used for this study to give 1-MCP exposure using syringe injection.

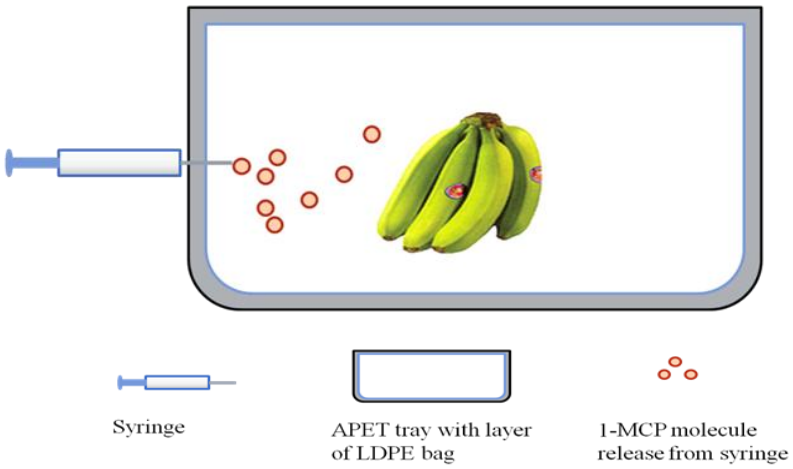



**Figure 11: Conceptual illustration for permeable package system used for different 1-MCP exposure application, [53]**

## 3.4. Experimental design

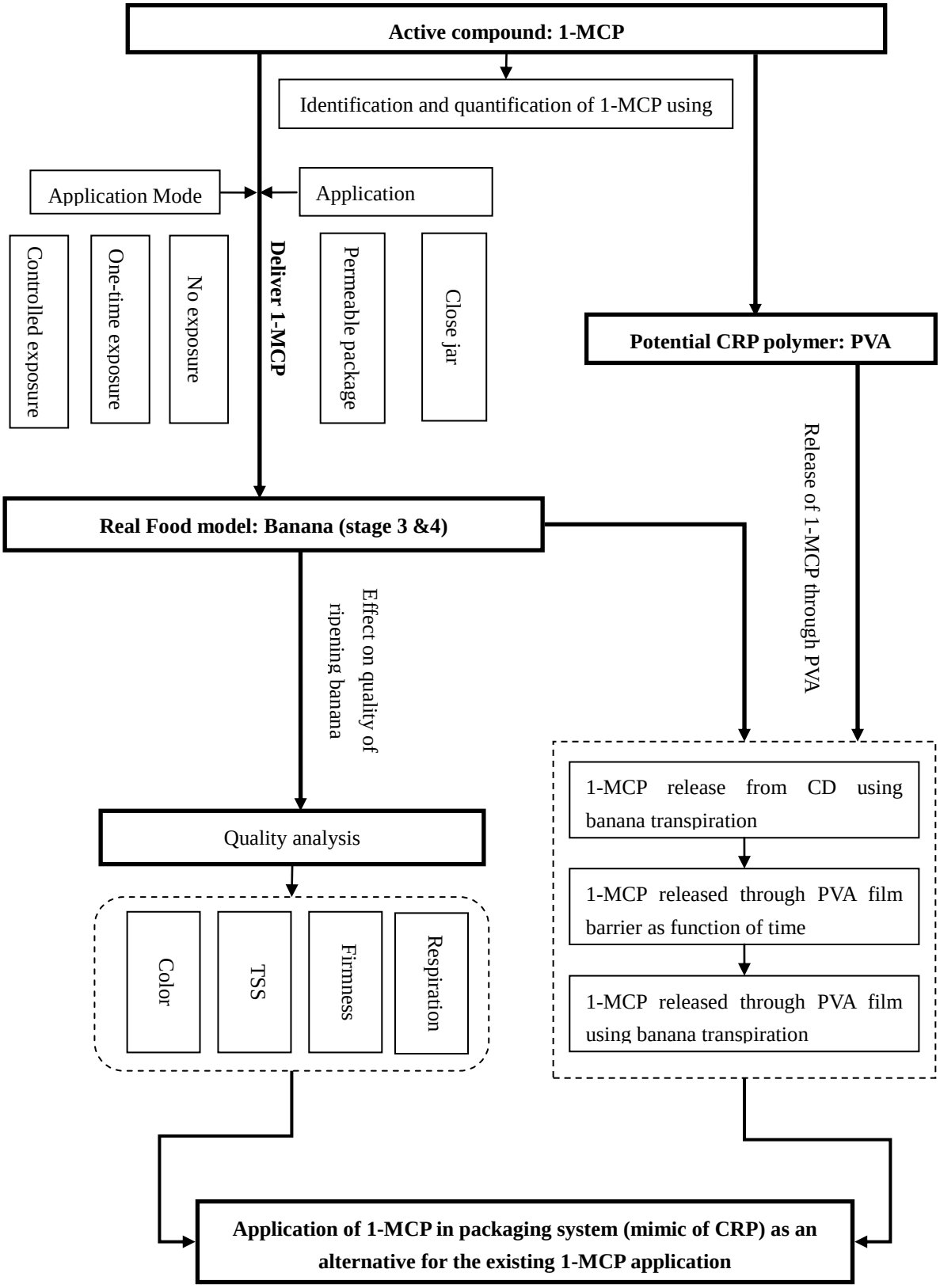



**Figure 12: Experimental design for different exposure methods to 1-MCP**

The experimental design for developing an alternative technique to treat fresh produce with 1-MCP is described in Figure 12. Ethylene antagonist 1-MCP was used as an active compound. 1-MCP was delivered to model food (Bananas) at different ripening stages (3 & 4) using different application modes including one-time exposure, controlled exposure, and no exposure of 1-MCP in different application systems that is a closed system and permeable system. The 1-MCP was delivered one-time and multiple times (controlled exposure) to study the effect of 1-MCP on inhibiting the action of continuously forming ethylene receptors. The effectiveness of different exposure methods in different application systems were observed by a change in color, texture, respiration rate, and TSS content of model food (bananas). To study the permeability of active compound -1-MCP from film, cast PVA film was used. The moisture activated release of 1-MCP from encapsulation of cyclodextrin (CD) using moisture released from bananas was also considered.

# 4. MATERIAL AND METHODS

## 4.1. Materials

### 4.1.1. Active compound - 1-methylcyclopropene (1-MCP)

1-MCP gas was obtained from AgroFresh Inc. (a division of Rohm and Hass Co., Philadelphia, PA, U.S.A) encapsulated in α-cyclodextrin (CD) in powder form containing 0.14% active ingredient by weight.

#### 4.1.1.1. 1-MCP stock gas preparation:

1-MCP stock gas was generated in a stock glass vial by dissolving weighed quantities of 1-methylcyclopropene - cyclodextrin complex (1-MCP-CD) (a.i. 0.14% (w/w) 1-MCP) powder in 1 ml of de-ionized water at 25°C with continuous stirring. When the encapsulated form of 1-MCP-CD powder was dissolved in water, 1-MCP released was initialized instantaneously into the headspace of the vial and was subsequently emitted over 2 hours of period [54]. The concentration of 1-MCP was calculated based on the volume of free space in the glass vial. After 2 hours the appropriate quantity was withdrawn from the stock of 1-MCP atmosphere and then injected into treatment containers containing bananas, consequential in the final 1-MCP concentration required.

#### 4.1.1.2. 1-MCP gas analysis

The concentration of 1-MCP was analyzed using a gas chromatography (GC) (HP Series 5890A, Hewlett Packard) fitted with 80/100 mesh porapak N column and a flame ionization detector. The temperatures were 120°C, 150° C, and 150° C for

column, injection, and detector respectively. Helium at 40 ml/min was used as the carrier gas. The GC-Mass spectrometry was used to detect the 1-MCP peak.

### 4.1.2. Application system

#### 4.1.2.1. Close Jar System

For treatment of the bananas in closed system (non-permeable) a 1.9L glass jar was used with metal leads. The metal lids fitted with two rubber septa, one for the sample intake and gas injection, and other for the air passage punctured with needle to avoid the suffocation of the fruits.

**Figure 13: Closed jar system used for 1-MCP treatment**

#### 4.1.2.2. Permeable package

An APET rectangular clear tray with dimensions of 9 X 9 X 2.6 IN. and Hefty bag (Type: Hefty baggies storage bag) with the dimensions of 11.5 X 12.18 IN. (1 gallon) was used to make permeable package for all experiments. There were a total of eight (8) holes made in the tray to avoid gas accumulation. One hole was sealed using silicon to make the injection port for injecting and withdrawing gas. The tray was kept inside the Hefty bag and was sealed using a hand sealer (Uline sealing

equipment, model # KF 200H) from edge to edge to make the bag fit to the size of tray.

**Figure 14: Permeable package system used for 1-MCP treatment**

4.1.2.3. Volume of permeable package

At time zero 5 ml of $CO_2$ gas was injected using gas tight syringe in to the permeable package through the injection port and then the package was kept on a stirrer for 10 minutes, to mix the gases well. The sample was then withdrawn using a 10 uL gas tight syringe and injected in the GC. The amount of $CO_2$ was calculated using a calibration curve of $CO_2$ to calculate the volume of the package.

4.1.2.4. Gas permeability of permeable package

To measure the permeability of the package, the permeable package was prepared in triplicate. The packages were flushed with $CO_2$ gas for 3-4 minutes, followed by

sealing it immediately using hand sealer. 10 uL samples were withdrawn every hour and analyzed using GC. Gas permeability was calculated from the slope obtained, the average of replicates was used for calculation.

4.1.2.5. Optimization of $CO_2$ level in permeable package

According to post harvest handling guide (UC Davis, post harvest handling guide), the banana packages should maintain the $CO_2$ level below 7-8% to avoid any stress to fruit. Therefore, the experiments were conducted by keeping 1-3 fingers of banana in the package and $CO_2$ levels were measured after 24 hours to calculate the amount of $CO_2$ gas accumulation in the permeable package with respect to amount of bananas kept in the permeable package.

4.1.3. Food model – bananas

Hands of mature, but not ripe, bananas (Cavendish variety - Dole fruits) were purchased from a local supermarket (the same brand of banana from the same vendor was obtained throughout the experiment to minimize the variation.), i.e. color change from green to yellow (stages 3-4) for preliminary experiments. The fruit fingers were cut from banana hands, selected on the base of uniform size , mass, and maturity index by measuring volume of the banana through water displacement techniques, taking a weight of each finger, and visual observation using the color chart [43] for ripening color stage identification respectively. Any bruised or diseased fruit fingers were discarded.

## 4.2. Preliminary experiment

### 4.2.1. Effect of 1-MCP on partially ripe bananas

This experiment was conducted to confirm the activity of available 1-MCP to delay further ripening of partially ripened bananas.

Partially ripe bananas that are stage 3 were treated with 1-MCP to confirm the activity of available 1-MCP and to test the effect of 1-MCP on the quality of partially ripened stages of bananas. Fruits for this experiment were treated with 0.52 PPM 1-MCP in a closed jar for 12 hours as mentioned earlier in reviewed literatures, and then left open to the air at room temperature for further observations.

### 4.2.2. Determination of effective range of 1-MCP

The 1-MCP stock gas was prepared as mentioned earlier in section 4.1.1. To determine the effective range (minimum and maximum concentration) of 1-MCP required to delay further ripening, partially ripened bananas (stages 3 & 4) were exposed to three different concentrations. The following 1-MCP concentrations 0.08 PPM, 0.15PPM, and 0.63 PPM were given to fruits for 6 hours at room temperature in a closed jar. For treatment the bananas were individually placed in a 1.9 L glass jar and sealed. 1-MCP gas was then injected in jar from the stock gas using gas tight syringe through rubber septum located on the lid of the jar. The control was not treated with 1-MCP and three replicates for each treatment were used for the experiment. After the treatment, the fruits were left open to the room air to ripen naturally.

### 4.3. Experiment 1: Effects of two different types of 1-MCP treatment on partially ripened bananas in a closed jar system

This experiment was conducted to study the effect of two different types of 1-MCP treatments: 1) one-time exposure 2) controlled exposure in delaying further ripening of partially ripened bananas. For treatment (1) “one time exposure” , banana ( stage 3 ) was weighed and placed individually into 1.9 L air tight glass jar and treated with 0.63 PPM 1-MCP gas for 6 hours at room temperature on day 0. For treatment (2), 0.08 PPM 1-MCP gas was exposed to the bananas for 6 hours every day for 12 days, to provide the “controlled exposure” of 1-MCP. All samples were prepared in triplicates and control for both treatments was kept without any 1-MCP treatment. 1-MCP treatment container was left unsealed after 6 hours of 1-MCP exposure in a closed jar to maintain a normal atmosphere. The rate of ripening was assessed by a change in peel color. Also the presence of CO2 gas was measured by head space analysis of treatment jar to study the change in the respiration pattern during the treatment.

Prior to daily headspace sampling to measure the $CO_2$, lids were sealed on each of the containers before 1 hour. Then four times 10 uL of head space gas from the jars were withdrawn over 6 hours to measure the respiration rate of bananas. The respiration rate was calculated from the slope obtained by four sampling points.

## 4.4. Experiment 2: Effects of two different types of 1-MCP treatment given to bananas in permeable package (to mimic the CRP environment) at ripening color stages 3 and 4

The permeable package was created by sealing PET trays in LDPE bags, as mentioned earlier. Three fingers of banana from ripeness stage 3 and stage 4 were sealed in permeable packages separately and similar 1-MCP treatments were given to both stages (3 & 4) of fruits as given in case of closed jar system experiment. For this experiment, 1-MCP gas was injected from the 1-MCP stock gas to create the desire 1-MCP concentrations in the package. For "one - time exposure" 0.63 PPM 1-MCP gas was injected inside the permeable package on day 0, for "controlled exposure" 0.08 PPM 1-MCP gas /day was injected throughout the duration of the experiment.

No 1-MCP was injected in the package serving as controls. For this experiment 12 replicates were used for each type of treatment including controls. The respiration rate, peel color, firmness, and TSS were used as quality indicators of the bananas.

### 4.4.1. Methods to access quality of bananas

#### 4.4.1.1. Respiration rate measurement

Three packages from each treatment, including controls, were kept intact throughout the experiment for assessment of respiration rate, which was calculated from $CO_2$ gas amount (%) in the headspace of the permeable package. 10uL samples were withdrawn daily for gas analysis from the headspace of those intact packages. The

level of $CO_2$ gas in headspace of package was calculated from $CO_2$ standard curve and presented in % value.

4.4.1.2. $CO_2$ gas analysis

The concentration of carbon dioxide was measured by sampling 10 uL through gas tight syringe. The withdrawn samples were analyzed by gas chromatography (HP Series 5890A, Hewlett Packard) fitted with CTR I column and a thermal conductivity detector. The temperature condition for oven, injection port and detector was 50°C for 6 min, 90° C and 110° C respectively. Hydrogen at 60 ml/min was used as the carrier gas. The calibration of carbon dioxide was done by injecting pure carbon dioxide in GC-TCD and a calibration curve was used to calculate concentration for all experiments.

4.4.1.3. Initial quality analysis

For initial analysis (day 0) six fingers of bananas at ripening stage 3 were analyzed for each physiological quality indicator peel color, firmness, TSS content, and overall visual quality. The mean value was used as initial values for all quality indicators. After that, three replicates from each type of treatment were destructed at the $6^{th}$ and $10^{th}$ / $12^{th}$ day (last day) for quality assessment.

4.4.1.4. Total soluble solids (%), (TSS)

To determine total soluble solids 5 g pulp of sample fruit without peel (5 g from 3 fruits of each sample) was homogenized with 5 ml distilled water and centrifuged at 13000 g for 10 minutes, then supernatant was collected for total soluble solids

analysis. TSS was measured using digital refractometer (model 10480, Mark Abbe II) and was reported in %.

4.4.1.5. Firmness (N)

A TA-XT2 Texture Analyzer (Stable Micro Systems) was used to evaluate the texture of the bananas. Penetrometry tests were conducted using a 5 mm stainless steel probe, penetrating 10 mm into sample. The test was performed with test speed of 1 mm/s with trigger force of 10.01 N. The test was performed on whole peeled bananas at two points, 3 fingers of banana per replicates were used to perform all tests. The maximum force (N) generated during probe travel were used for data analysis.

4.4.1.6. Peel color

Three fingers per replicates were used to evaluate the external skin color visually by the standard banana color chart every alternate day by two judges. The rate of ripening from green to yellow was measured using standard banana color chart [43]. A numerical value from (1 to 7) was given to each sample based on the color, and average was calculated for results. The color scale was determined as followings. 1 – green, 2 – more green than yellow, 3 – 50-50% green-yellow, 4 – more yellow than green, 5 – yellow with green tips, 6- completely yellow, 7 – yellow with brown spotting.

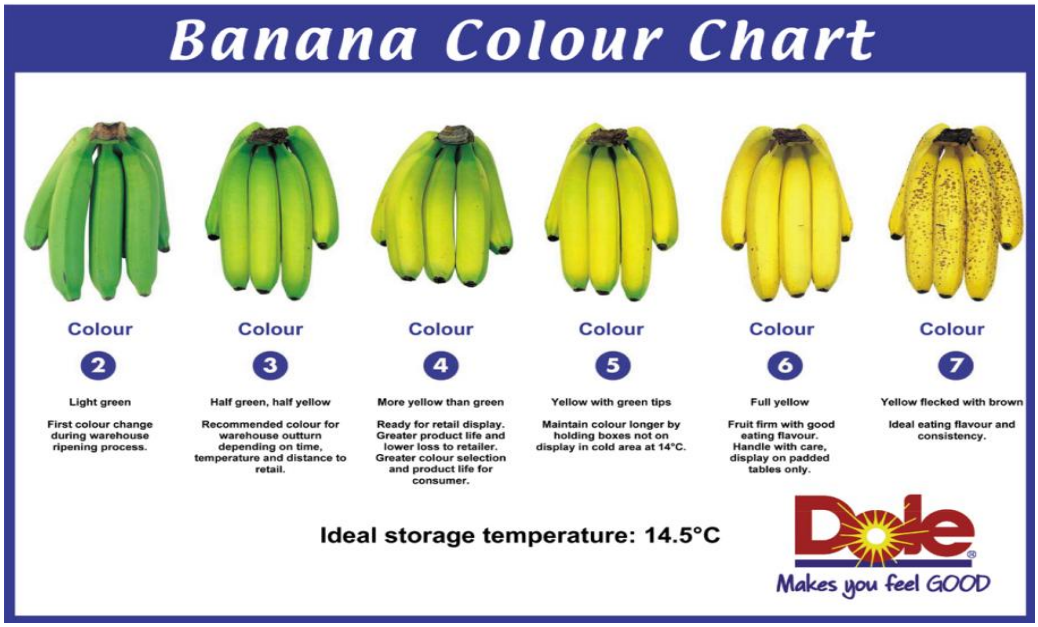



**Figure 15: Banana ripening color stage chart, ( Adapted from: [43])**

The above figure of banana ripening color chart was used for ripening color stage identification throughout the study.

4.4.1.7. Sugar spotting

Fingers of banana were chosen randomly from each replicates for sugar spotting on the banana peel using scale of 1-4, where 1 = yellow with no development of sugar spotting, 2 = little darker surface with some brownish spot, 3= spot all over the surface, with more intense browning and size and 4 = darker spot, even black and overlapping with bigger size on surface.

4.4.1.8. Overall appearance

The visual quality of the bananas were determined based on hedonic scale of 1 = inedible, 2 = limit of usability, 3 = limit of marketability and 4= very good and 5 = excellent. The scores of 3 banana fingers per replicates were given by two judges

based on color, visual structural integrity, and general visual appeal [30].

## 4.5. Experiment 3: Release of 1-MCP gas through PVA film barrier

### 4.5.1. Film formation

Cast film was obtained by mixing 4 g of PVA polymer powder in 96 ml of distilled water at 60°C. The prepared solvent was poured on a glass frame, followed by drying for 48 hours. After drying, the film was peeled off the frame and stored in a bag until use. PVA polymer was selected to make the film due to its good gas barrier properties and its ability of high moisture permeability, which can allow the moisture migration in the film to release 1-MCP from 1-MCP- cyclodextrin complex incorporated in films or sealed in films. It is also a completely bio-degradable material. If release of 1-MCP can be controlled through PVA film barrier, it will have the potential to be used as an active layer in the CRP system for incorporating 1-MCP.

### 4.5.2. Release of 1-MCP through PVA film at 100% RH

1-MCP was weighed in an aluminum cup and immediately sealed by PVA films with a silicon rubber sealant. Release of 1-MCP from PVA film was determined by placing the entire cup in a 500 ml glass jar containing distilled water to provide the desire RH. The jar was sealed with metal lids, fitted with rubber septa, and further secured with para films which served as a sampling port. The amount of 1-MCP released from 1-MCP-CD through PVA film barrier and 1-MCP-CD control were determined periodically for 1-MCP concentration in the jar headspace. Headspace (1

ml) was withdrawn by gas tight syringe and analyzed by gas chromatography equipped with flame ionization detector. GC-FID conditions for HP 5890 GC were the same as mentioned earlier for 1-MCP gas analysis.

### 4.6. Experiment 4: Release of 1-MCP from cyclodextrin using moisture released from bananas

This experiment was conducted to confirm the release of 1-MCP from cyclodextrin complex using the bananas' moisture as the trigger. For this experiment the banana was placed inside the 1.9 L glass jar with weighed quantity of 1-MCP –CD powder to generate 20 PPM concentration of 1-MCP gas. The jar was sealed with metal lids fitted with rubber septa, which served as a sampling port and further secured with para films. The amount of 1-MCP released from CD was determined periodically for 1-MCP concentration in the jar headspace. Headspace (1 ml) was withdrawn by gas tight syringe and analyzed by gas chromatography. In the closed jar system no water or RH salt was used to provide extra moisture to release 1-MCP, instead the bananas' moisture was used to release 1-MCP from CD. The amount of gas present in headspace jar was calculated using 1-MCP gas standard curve and presented in % value.

# 5. RESULTS

## 5.1. Results of preliminary experiments

Following picture was taken to show the effect of 1-MCP exposure on the color of ripening bananas.

### 5.1.1. Effect of 1-MCP on bananas (visual observation)

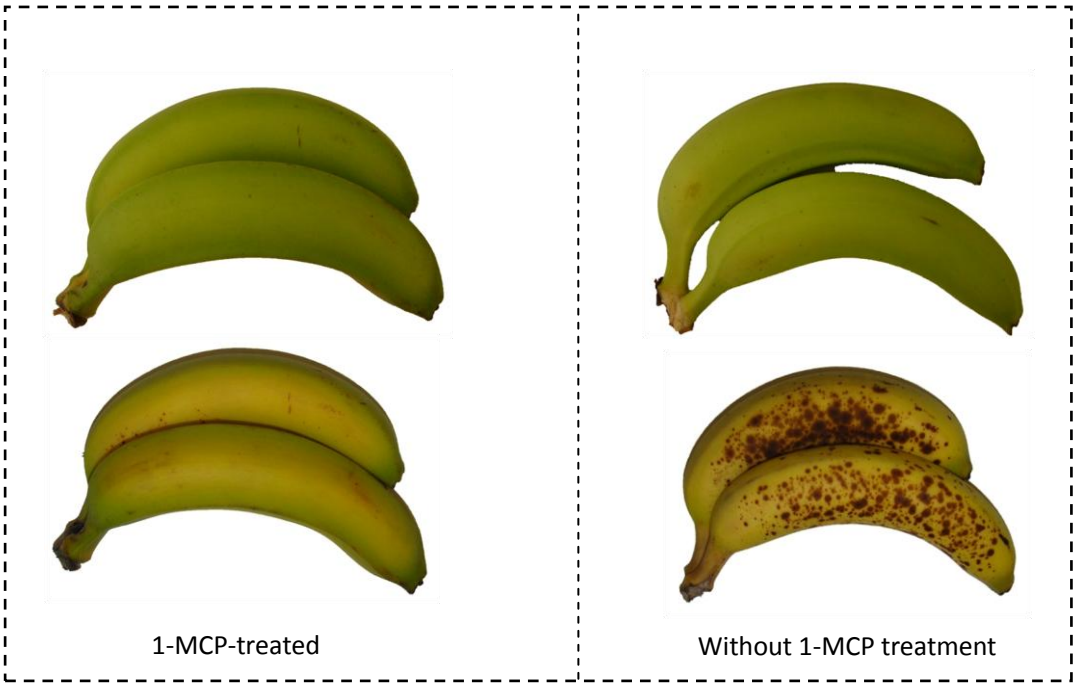



**Figure 16: Effect of 1-MCP treatment on visual quality of bananas**

Figure 17 shows the effects of 1-MCP on delaying the ripening color stage of banana using graphical illustration.

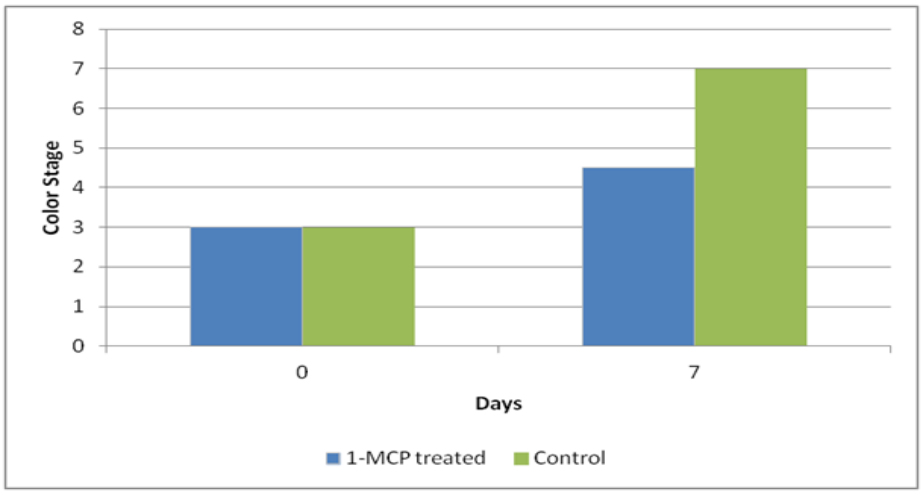



**Figure 17: The effect of 1-MCP treatment on color changes of ripening bananas**

Preliminary experiments showed the clear effect of 1-MCP on partially ripened bananas as indicated by a change in color. The 1-MCP treated sample has a better appearance with much less browning and sugar spotting. The treated bananas had developed less yellow color even after 7 days of the treatment, whereas the control bananas without any treatment had developed brown spot with fully grown yellow color.

5.1.2. Effective range of 1-MCP concentration to treat bananas at ripening stages 3 and 4

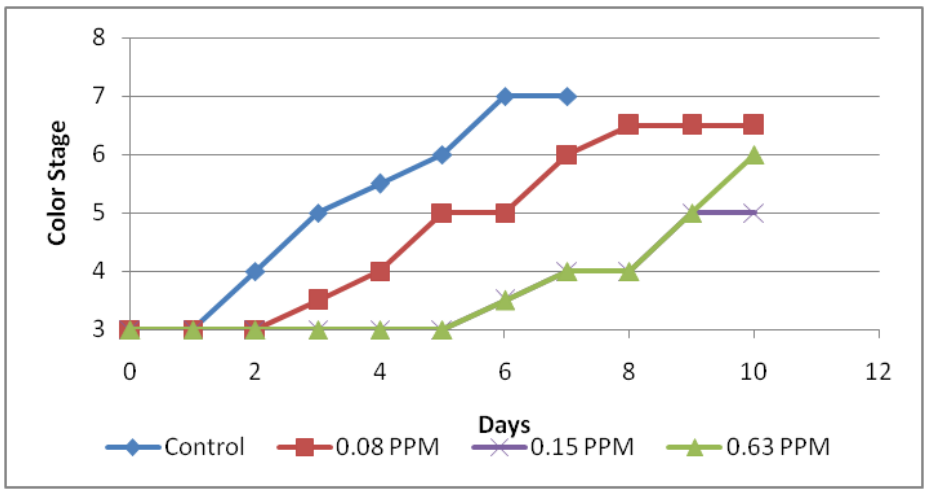


**Figure 18: Effective 1-MCP concentration range for stage 3 bananas**

The effective 1-MCP concentration range was observed by a change in the ripening color stage of the bananas. Bananas with ripening color stage 3 were exposed to a different concentration for 6 hours in 1.9 L close jar and then left to ripen at room temperature. Each datum represents the mean of three replicates. Treatment with different 1-MCP concentrations starting from 0.08 to 0.63 PPM showed delays in further ripening of bananas compared to controls. The delay in ripening was judged by change in color of bananas from green to yellow using the standard banana ripening color chart[43]. Observation for fruits treated at stage 3: The control banana reached stage 7 on $6^{th}$ day of treatment, whereas 1-MCP treated fruit with a concentration higher than 0.08 PPM reached only ripening color stage 3.5 during the same time period. Even the minimum concentration that is 0.08 showed the effect by holding the bananas at ripening color stage 5 until the sixth day of treatment. There was no difference observed between the 0.15 PPM and 0.63 PPM treated fruits until the 9th day of treatment. Only on the 10th day a difference was observed between fruits treated with 0.15 PPM and 0.63 PPM, fruits treated with 0.15 PPM had maintained the same ripening color stage 5, unlike the fruits treated with 0.63 PPM which reached ripening stage 6. The fruit with minimum concentration seemed to be the least effected amongst all three. There was no sugar spotting observed in 1-MCP treated sample at all until the last day of treatment.

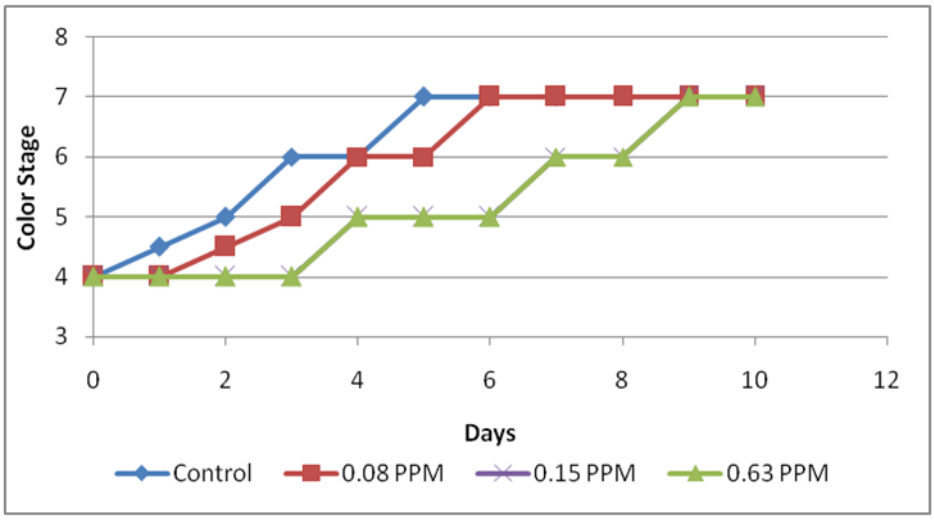



**Figure 19: Effective 1-MCP concentration range for stage 4 bananas**

Figure 19 shows the effect of 1-MCP on the peel color of the bananas treated at ripening stage 4. Effective 1-MCP concentration range for stage 4 bananas was observed by a change in color of the bananas. Bananas with ripening color stage 4 were exposed to different concentrations for 6 hours in 1.9 L closed jars and then left to ripen at room temperature. Data represents the means of three replicates. A similar observation was made for the 1-MCP treatment given to stage 4 bananas as stage 3. However, a 1-MCP treatment seems less effective here compared to treatments given at stage 3. Fruits started ripening on day 4 even with 1-MCP treatment. The concentration at 0.15 PPM and 0.63 PPM seems effective for 7-8 days, whereas not much difference was observed between control fruits and fruits treated with 0.08 PPM.

## 5.2. Different type of 1-MCP treatments given to partially ripened bananas in close jar

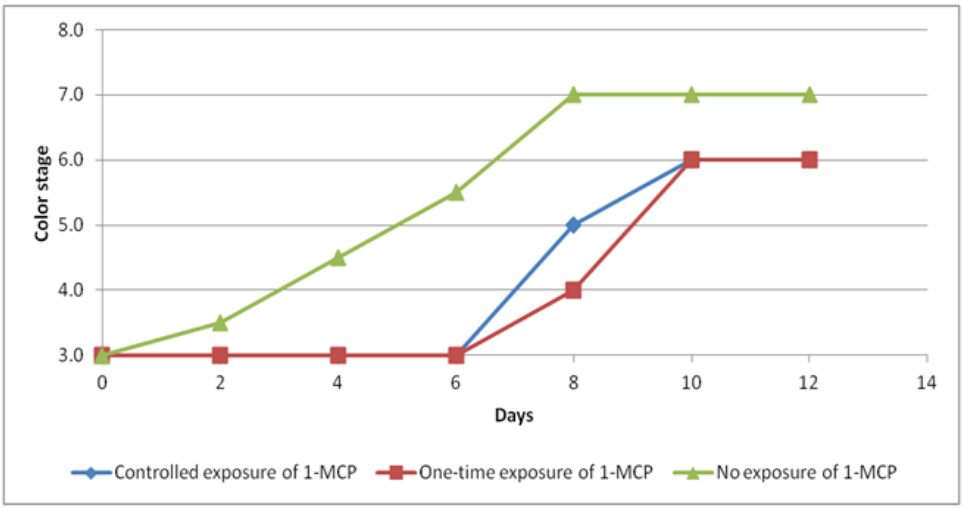



**Figure 20: Peel color development of bananas treated with 1-MCP in close jar**

Figure 20 shows the effect of 1-MCP on the peel color of the bananas. Fruits were exposed to 0.63 PPM for 6 hours on (one –time exposure), 0.08 PPM/day for 6 hours (controlled exposure), and no exposure (control). The initial peel color of the control fruits remained unchanged only for first two days, after which it increased rapidly with yellowing and brown spots forming in over the next 4 days. 1-MCP treated fruits with "controlled exposure" were able to maintain the same ripening color stage for six days, and then they gradually started ripening and very few brown spot formations were observed, even up to the last day of the experiment. The fruits treated with “One-time exposure” seem to be the most effected in terms of delaying the yellowing of the fruits, until the 8$^{th}$ day of treatment the fruits seemed to be holding nearly ripening color stage 4. After the 8th day the peel color of the fruits treated with both types of 1-MCP treatments showed a similar change in the peel

color.

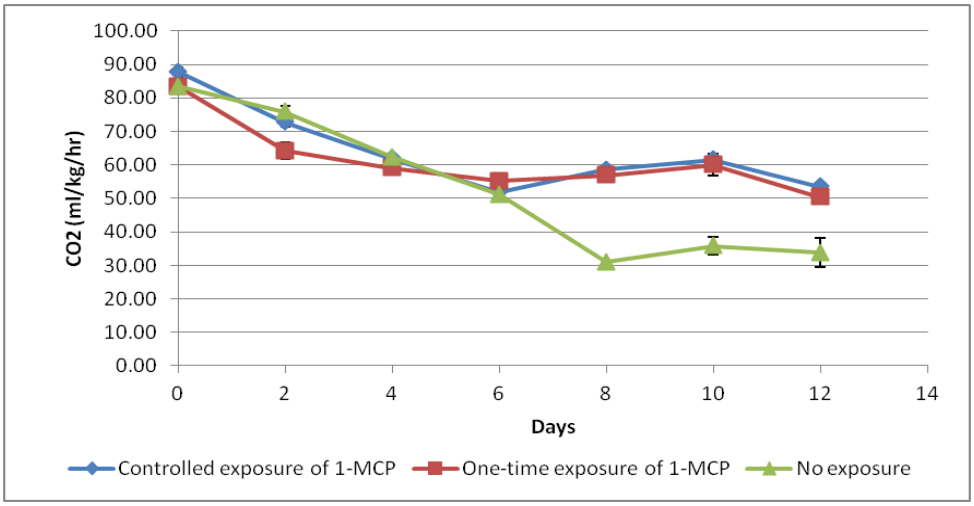



**Figure 21: Respiration rate (CO2 ml/kg/hr) of bananas treated with 1-MCP in close jar system**

Fruits were exposed to 0.63 PPM for 6 hours on (one-time exposure), 0.08 PPM/day for 6 hours (controlled exposure), and no exposure (control). Figure 21 shows the respiration rate of bananas treated with 1-MCP. For the initial 2 day sample treated with one time exposure of 1-MCP has shown a lower rate of respiration compared to both controlled exposures of 1-MCP sample and control sample. On day 4 all three samples including controls had decrease in reparation rate with no significant difference among all three samples including the controls. After the 4$^{th}$ day the respiration rate was constant for samples treated with one-time exposure of 1-MCP and controlled exposure of 1-MCP. Where, in the case of the control samples, the respiration rate continued to drop. The respiration rate of 1-MCP treated fruits ranged from 50.5 to 89.9 $CO_2$/kg/hr, whereas for the control it ranged from 33.8 to 83.4 ml $CO_2$/kg/ hr.

## 5.3. Effect of two different types of 1-MCP treatment given to bananas in a permeable package (to mimic the CRP environment) at ripening stage 3 and 4

### 5.3.1. Effect of 1-MCP on respiration rate of bananas treated at ripening stages 3 and 4

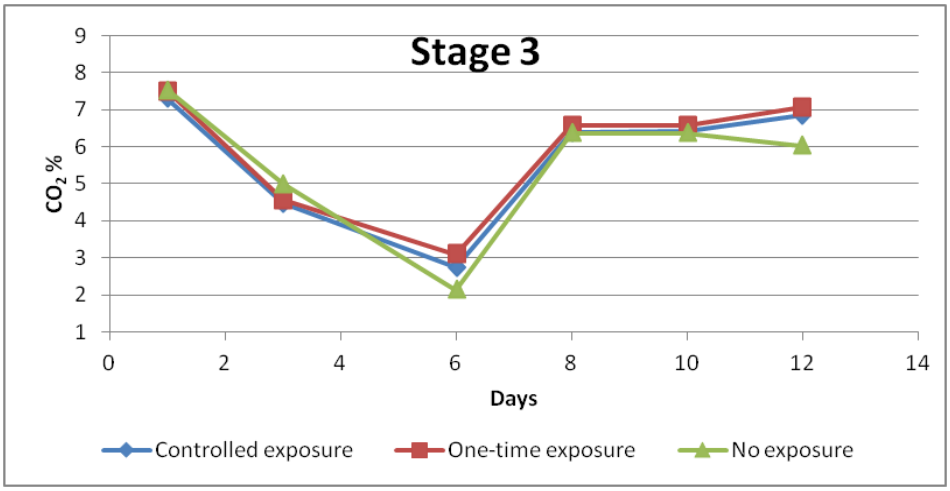



**Figure 22: Effect of 1-MCP on respiration rate of bananas treated at ripening color stage 3 in permeable package**

Figure 22 shows the effect of 1-MCP on the respiration rate of bananas. Fruits treated at ripening stage 3 with 1-MCP had the highest respiration rate on the first day of the treatment and similar responses were observed for the controls. For the first 6 days of the treatment no significance difference was found in respiration rate of fruits treated with 1-MCP and fruits without treatment. The respiration rate for all three samples were declining continuously until the $6^{th}$ day, than it rose a little bit on the $8^{th}$ day and maintained a constant rate until the $12^{th}$ day. Towards the end of the experiment after the $10^{th}$ day, the respiration rate of the control sample seemed to be

dropping a little faster compared to both 1-MCP treated samples. In conclusion, there was no significant difference found in the respiration rate of bananas for all three types of samples. The highest $CO_2$ production in the permeable package was observed at nearly 7.5 % and the lowest was nearly 3 %. None of the packages exceeded the $CO_2$ production above 8 % or dropped below 1-2 %, so we can assure that the package had no impact on quality deterioration of banana due excess of $CO_2$ accumulation or less $CO_2$ gas in the package.

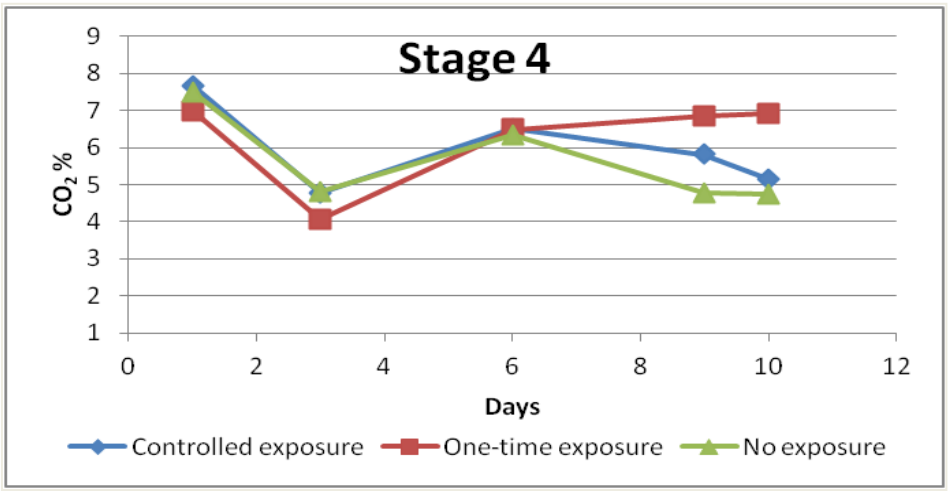



**Figure 23: Effect of 1-MCP on respiration rate of bananas treated at ripening color stage 4 in permeable package**

A similar trend was observed for the respiration rate of fruits treated at stage 4 in permeable packages at stage 3. No significant difference was found in the respiration rate of bananas treated with 1-MCP and without 1-MCP. The highest $CO_2$ production in the permeable package was observed at nearly 7.5 % and the lowest was nearly 4 %. Also fruits treated at ripening stage 4 did not exceed the $CO_2$ production of 8% in the package. From figure 21 it can be observed that the $CO_2$ production follows the typical respiration trend of climacteric fruits and showed a drop in the respiration

rate due to the post climacteric phase.

5.3.2. Effect of 1-MCP treatment on total soluble solids (TSS) content of bananas treated at ripening stages 3 and 4

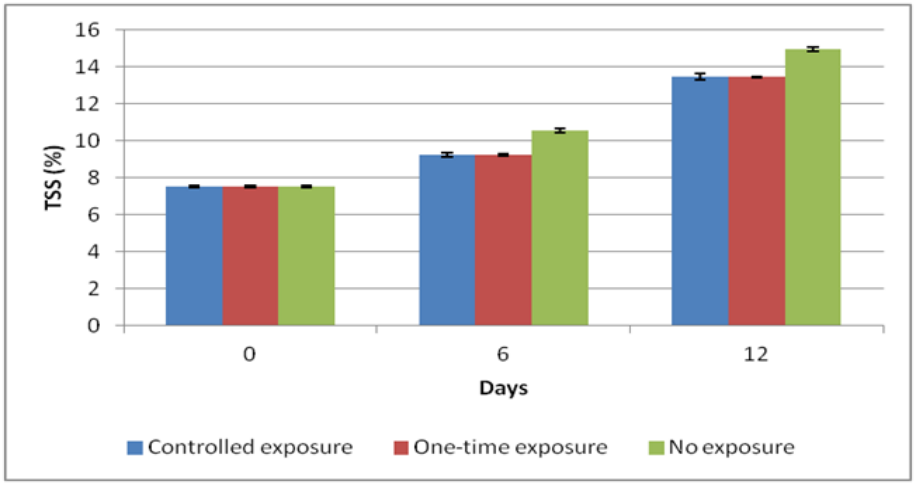



**Figure 24: Effect of 1-MCP on TSS content of bananas treated at ripening color stage 3 in permeable package**

As the bananas start ripening, the TSS content of the bananas increases due to breakdown of starch. In this experiment a significance difference was observed in TSS content of fruits treated with 1-MCP and without any 1-MCP treatment. At the $6^{th}$ day the TSS content for fruits treated with 1-MCP ranged between 9 to 9.4 %, whereas for control fruits TSS content was found to be nearly 10.7%. This shows that the control samples were already at an advanced stage of ripening compared to 1-MCP treated samples. There was no significant difference found between samples treated with controlled exposures and one time-exposure at the 6th day or even on the $12^{th}$ day of the experiment. On the $12^{th}$ day the TSS content of 1-MCP treated samples were nearly 13%, whereas for controls it was measured at 15 %. This

indicates that the controlled exposure treatment, even with half of the dose than that of one-time exposure, has the same capacity to delay the ripening associated processes.

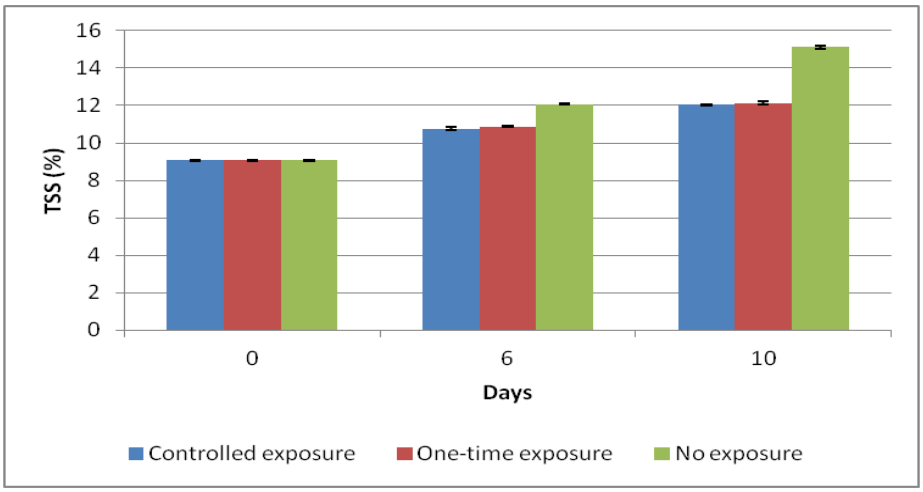



**Figure 25: Effect of 1-MCP on TSS content of bananas treated at ripening color stage 4 in permeable package**

A similar observation was made with respect to the TSS content of fruits treated at ripening stage 4 as for fruits treated at ripening stage 3. Fruits treated with both types of 1-MCP treatments showed a significant difference up to the end of the experiment. Final TSS content of fruits treated with 1-MCP ranged between 12 to 12.3 %, whereas for control fruits the TSS content was found between 15.1 – 15.3 %. No significant difference was found between samples treated with controlled exposure and one time-exposure. These results implied that 1-MCP treatments were effective to delay breakdown of starch content, which is one of the ripening indicator for bananas.

5.3.3. Effect of 1-MCP treatment on firmness of bananas treated at ripening stages 3 and 4

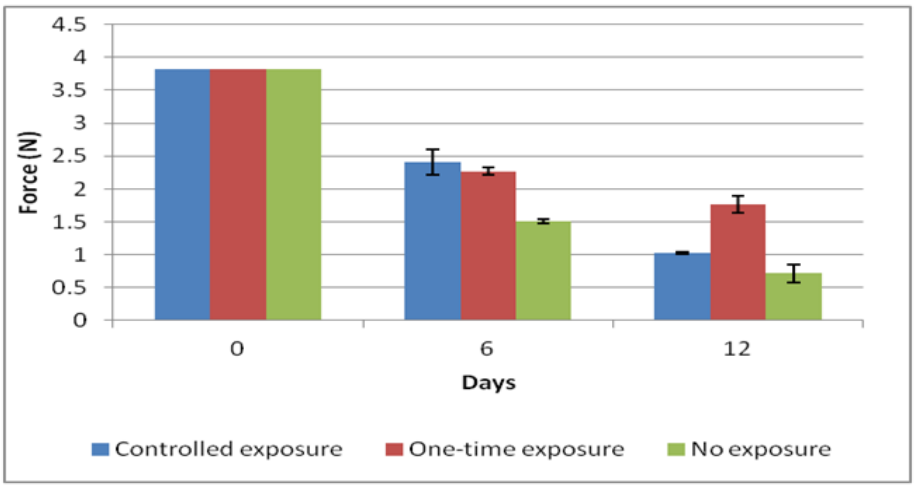



**Figure 26: Effect of 1-MCP on firmness of bananas treated at ripening color stage 3 in permeable package**

Figure 26 shows the effect of different 1-MCP treatments on maintaining the firmness in bananas. As the bananas ripen they tend to soften more, especially during climacteric phase to post climacteric phase. As the fruits start getting softer, it they required less penetration force. From Figure 26 it can be clearly observed that the force required to puncture the banana dropped as the fruits started ripening. An initial average force of 3.8 N was noted for penetrating the fruits on day 0, which dropped to 2.7 – 2.2 N for 1-MCP treated samples, and 1.8 to 1.5 for control after 6 days. After 12 days it dropped down further to nearly 1.6-1.1 for 1-MCP treated and nearly 0.7 for without 1-MCP treated fruits. No significant difference observed in one-time exposure and controlled exposure treatment until the $6^{th}$ day, both showed good effects of 1-MCP on maintaining the firmness of fruit compared to fruit

without any treatment. After that the controlled exposure treated sample softened faster than that of the one-time exposure treated sample.

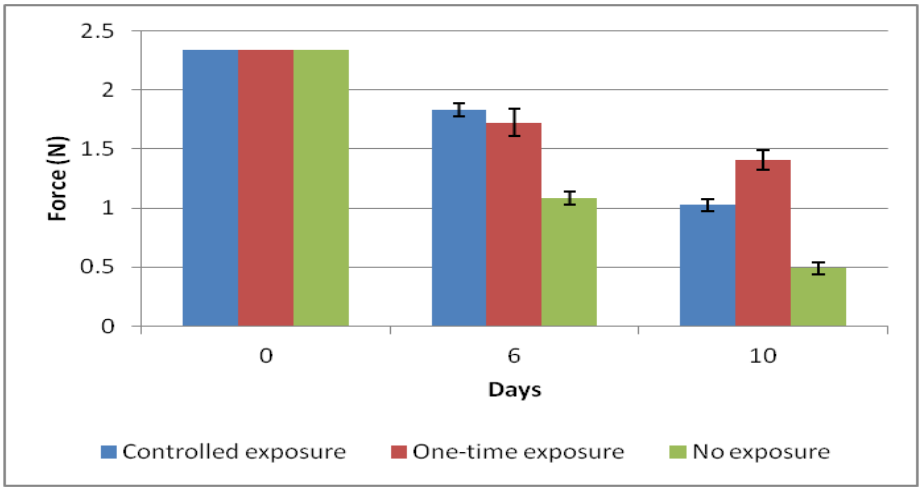



**Figure 27: Effect of 1-MCP on firmness of bananas treated at ripening color stage 4 in permeable package**

A similar observation was made for the firmness of fruits treated at stage 4, as fruits treated at ripening stage 3. The maximum force required to puncture the banana dropped from an initial average force of 2.4 to 1.8– 1.7 for 1-MCP treated samples and 1.1 to 0.9 for control after 6 days. No significant difference was observed in the one-time exposure and controlled exposure treatment up to the $6^{th}$ day. The one-time exposure treated samples for both stages; treatments given at ripening stage 3 and stage 4 were able to maintain the highest firmness amongst all three samples. Controlled exposure treated samples also had better firmness retention than the controls, but was less firm than the one-time exposure treated samples. Overall, samples treated with either of the 1-MCP treatments had maintained definitely more firmness for at least 6 days than fruits without any 1-MCP treatment.

5.3.4. Effect of 1-MCP treatment on peel color of bananas treated at ripening stages 3 and 4

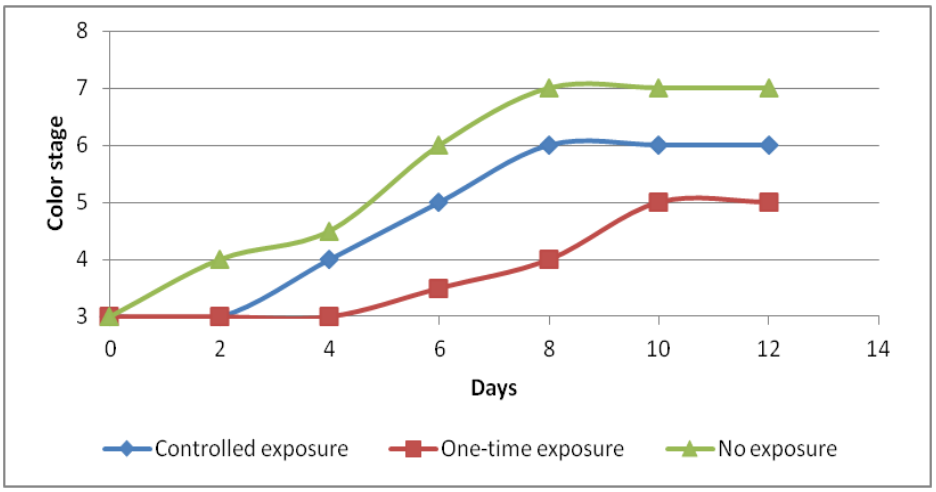



**Figure 28: Effect of 1-MCP on peel color of bananas treated at ripening color stage 3 in permeable package**

Figure 28 shows the peel color of the fruits treated with 1-MCP and without 1-MCP, using the banana ripening color chart [43]. Peel color index using the banana color chart indicated that there was a significant difference in the color of the 1-MCP treated sample and the sample without any treatment. It is clear from Figure 28 that there is a definite effect of 1-MCP on peel color of bananas, even at a partially ripened stage. For 1-MCP treatment given at ripening color stage 3, controlled exposure has maintained ripening color stage 3 for the first two days, and then it started turning more yellow after that. It reached stage 6 on the $8^{th}$ day and maintained the same ripening color stage until the end of the experiment. Whereas the one-time exposure sample had maintained stage 3 color until almost the $6^{th}$ day and were at ripening color stage 5 at the end of the treatment. In contrast with both

the 1-MCP treated samples, the control started turning yellow from the next day of treatment and it reached to stage 7 on the 8th day of the experiment. Not much sugar spotting was found on the controlled exposure samples, but some discoloration was observed after it turned to stage 6.

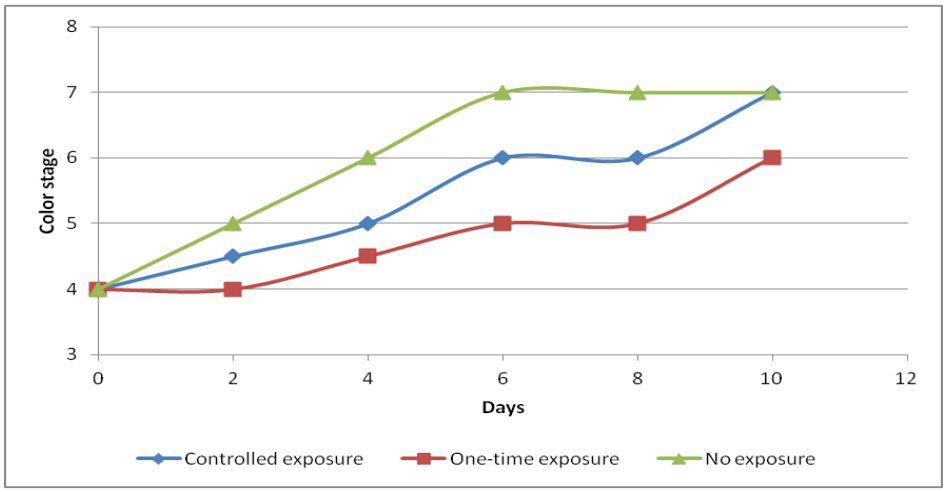



**Figure 29: Effect of 1-MCP on peel color of bananas treated at ripening color stage 4 in permeable package**

The same trend was observed in bananas treated at stage 4 with controlled exposure of 1-MCP. Controlled exposure maintained ripening color stage 4 for the first 2 days and then reached stage 6 on the 6th day. Whereas the onetime exposure sample maintained ripening color stage 3 until almost the 4th day, and was at ripening color stage 6 on the 10th day. Controlled exposure did not have a great effect when treated at ripening color stage 4 compared to one-time exposure.

5.3.5. Effect of 1-MCP on delaying sugar spotting in bananas

| Days | Treatment stage 3 | | | Treatment stage 4 | | |
|---|---|---|---|---|---|---|
| | Control | Controlled exposure of 1-MCP | One time exposure of 1-MCP | Control | Controlled exposure of 1-MCP | One time exposure of 1-MCP |
| 0 | 0 | 0 | 0 | 1 | 1 | 1 |
| 2 | 0-1 | 0 | 0 | 2 | 1 | 1 |
| 4 | 1 | 1 | 0-1 | 2-3 | 1-2 | 1 |
| 6 | 2-3 | 1 | 0-1 | 3 | 1-2 | 1 |
| 8 | 3 | 1 | 0-1 | 3 | 2-3 | 2 |
| 10 | 4 | 1 | 0-1 | 4 | 2-3 | 2-3 |
| 12 | 4 | 2 | 0-1 | | | |

**Table 1: Effect of 1-MCP on controlling sugar spotting on bananas treated at ripening color stage 3 &4**

Overall bananas treated with 1-MCP did not exhibit much sugar spotting throughout the experiment when treated at stage 3. Any of the 1-MCP treated bananas did not show the spotting score 4 during the entire experiment. 1-MCP treated samples at stage 3 did not reach spotting score 2 until the $12^{th}$ day. Whereas banana treated at stage 4 did exhibit the spotting score 2 after 8 days. For all control samples without any 1-MCP treatment spotting score was found to be 4 after 8 days.

| Days | Treatment stage 3 | | | Treatment stage 4 | | |
|---|---|---|---|---|---|---|
| | Control | Controlled exposure of 1-MCP | One time exposure of 1-MCP | Control | Controlled exposure of 1-MCP | One time exposure of 1-MCP |
| 0 | 5 | 5 | 5 | 5 | 5 | 5 |
| 2 | 4 | 5 | 5 | 3 | 4 | 4 |
| 4 | 3 | 4 | 4 | 3 | 3 | 4 |
| 6 | 3 | 4 | 4 | 2 | 3 | 3 |
| 8 | 2 | 3 | 4 | 2 | 3 | 3 |
| 10 | 2 | 3 | 4 | 2 | 2 | 3 |
| 12 | 2 | 3 | 3 | | | |

**Table 2: Effect of 1-MCP on maintaining overall appearance of bananas treated at ripening color stage 3 &4**

### 5.3.6. Effect of 1-MCP on visual quality of bananas

None of the bananas treated with 1-MCP or without 1-MCP reach visual score 1-inedible in 10-12 days experiments. But the untreated banana did displayed the visual score 2 – limit of usability after 6-8 days. Bananas treated with 1-MCP did not show the limit of usability at all when treated at stage 3. But it showed some limitation for bananas use after 8 days when treated at stage 4. Overall banana treated with 1-MCP showed very good visual appearance even up to 10 days compared to controls. Hence it can be concluded that 1-MCP can maintain a better appearance and visual quality for at least 5-6 days more than fruits without any treatment.

## 5.4. Release of 1-MCP gas through PVA film barrier

**Figure 30: PVA film obtained using casting method**

The figure above shows the cast film obtained by processing PVA polymers. The film obtained was transparent and had an even thickness as measured by a film thickness meter at several points.

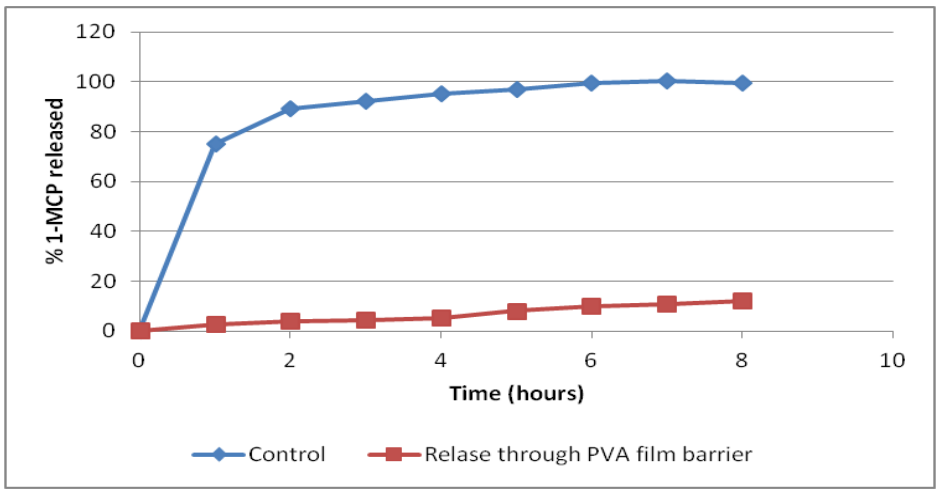



**Figure 31: Release of 1-MCP through PVA films barrier from 1-MCP-CD at 25° and 100 % RH**

The figure above shows the release of 1-MCP gas from CD complex in the headspace of a sealed glass jar over 8 hours period. This experiment was conducted to study the release of 1-MCP controlled by PVA polymer into the headspace of a package. Here a glass jar was used to mimic the package and the desired RH was

provided to release 1-MCP from CD. The PVA film was used as an active layer to slowdown the release of 1-MCP in the package. The control sample without any active film layer showed almost 80 % of 1-MCP in headspace released from CD within 1 hour, whereas samples with PVA film barrier had less than 5% 1-MCP gas in headspace in 1st hour of experiment released from CD. The control sample without any barrier showed 100 % of 1-MCP gas in headspace released from cyclodextrin in nearly 5 hours, in the case of the sample with the barrier layer, there was only about 12 % headspace concentration for 1-MCP released from CD in 8 hours of the period.

## 5.5. Release of 1-MCP from CD using moisture released from bananas

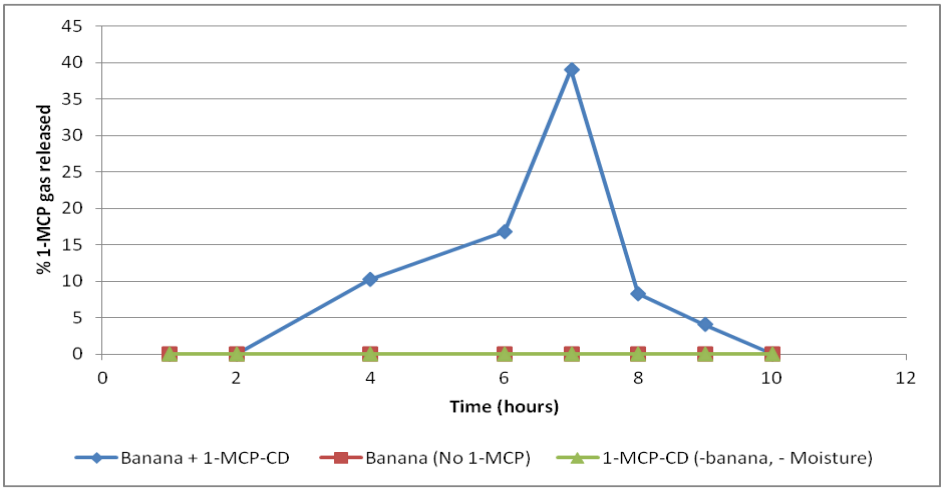



**Figure 32: Release of 1-MCP from CD complex via moisture released from bananas**

The above figure is showing the release of 1-MCP achieved by moisture released from bananas.

The Figure 32 shows that under normal room temperature and desired RH, 100% release of 1-MCP can be achieved within 5 hours. Here the 1-MCP released with the

bananas moisture, initiated after 2 hours and reached maximum level at around 7 hours, followed by a declined in headspace concentration of 1-MCP. A maximum of 40 % 1-MCP gas release was observed in headspace at the $7^{th}$ hour. At the $10^{th}$ hour no 1-MCP was observed in the headspace of the jar with bananas and 1-MCP. For the control sample of 1-MCP without bananas, no release was observed at all in 10 hours and in the case of only bananas, no other gas was detected which interfere with the 1-MCP gas analysis by GC.

# 6. DISCUSSION

## 6.1. Introduction

In recent years, new trends and systems have been developed in response to consumers' preference for fresh produce, moving towards mildly preserved and convenient with a prolonged shelf-life.

1-Methylecycloprpene (1-MCP), an ethylene antagonist compound is in keen interest of post harvest biologists for past few years. But, commercialization of 1-MCP is still limited to apples, pears, tomatoes, melons and flowers [32]. So, researchers are attempting to provide more data on potential application of 1-MCP for other plant commodities. 1-MCP application for delaying the ripening of bananas has been also studied widely by researches, but inconsistent responses received by researchers for its effects is limiting the commercialization of 1-MCP application for bananas. Hence, the further research, to study 1-MCP effects on bananas using different exposure techniques would be useful for establishing its commercial application. Bananas are perishable, climacteric fruit which can continuously ripen even after harvesting; handling, transporting, and distributing of such perishable commodities require more attention. For long distance transportation bananas have to be harvested at the green stage where they are treated with exogenous ethylene in a storage room before being distributed to supermarkets. Thereafter the yellow life (at and after partially ripen stage) of bananas is reduced to only about 3-5 days at supermarkets. The current 1-MCP application found in citations has focused on extending the green life (mature- but unripe banana) before ethylene treatment, which has shown

inconsistent response for its commercial use[32]. Also the effect of 1-MCP using current application was found to be effective for only short period of time unless continuous exposure is applied [11, 45]. So, the general aim of this work was to enhance the yellow life of partially ripened bananas using different 1-MCP exposure application that can provide the scientific base for establishing commercial 1-MCP application for bananas. An innovative technology, known as controlled release packaging (CRP), is being used widely for delivery of active compounds such as antimicrobials and antioxidants, which can be further extended for delivery of 1-MCP to commercialize 1-MCP application. By providing an active layer of film to fresh produce packages, we can change the internal environment of packaging to extend shelf life and enhance the food safety and sensory quality. [38]. To extend the yellow life of bananas for transporting them to non-banana producing countries, the CRP system would be a great tool, where the packaging itself serves as a delivery system. 1-MCP can serve as an active compound to delay ethylene induced ripening of the fresh produce and especially for climacteric fresh produce. Two different 1-MCP exposure methods, controlled exposure (to mimic continuous replenishment) and one-time exposure were used in this study to mimic the CRP system.

## 6.2. Summary of main conclusions

The results of two different exposure methods of 1-MCP on partially ripened bananas, the release suppression of 1-MCP from cyclodextrin complex through PVA film barrier and the release of 1-MCP from cyclodextrin using moisture released through banana transpiration has served a scientific base for designing a CRP system

for fresh produce using 1-MCP as an active compound.

### 6.2.1. Effect of controlled exposure and one time exposure on quality parameter of partially ripened bananas

The published literature on the effect of 1-MCP has either reported a suppressed rate of respiration for 1-MCP treated fruit, or no significant effect. Also, some of the citations mentioned that there were delays in the peak of the respiration rate, but no significant difference in the height of the respiration peak of bananas was observed for samples treated with 1-MCP versus untreated[4] . Here we have seen very little difference in the respiration rate for 1-MCP treated samples and untreated, and that is also with close jar system. This observation might be due to the stage of treatment. As mentioned earlier that banana is typical climacteric fruit which follows the same trend of respiration as shown in Figure 2. The work which has been published in literature where respiration is suppressed by 1-MCP, the treatments were given in stage 1 (mature-green – stages 1-2) during pre-climacteric phase. In this study, treatments were given at partially ripened stages where bananas were already at their peak of respiration and were moving towards the post climacteric phase, where the respiration rate actually starts decreasing. So, if bananas follow the typical climacteric fruit respiration trend, then their respiration rate should drop gradually after the peak of respiration. The less $CO_2$ production here is an indicator of more aging in fruit in an advanced stage of ripening during the post climacteric phase. Overall there was no significant difference found in the respiration rate of bananas when treated at a partially ripened stage with 1-MCP when compared with bananas

without 1-MCP treatment.

The controlled exposure treatment was effective to maintain the quality of banana for longer time compared to the no exposure, but was less effective than one time exposure; the results showed controlled exposure was effective for the initial 5-6 days. This can be due to the very lower dosage of 1-MCP used to treat bananas in the case of controlled exposure treatment, whereas the one-time exposure with higher initial dosages showed very good effects on the quality parameters of bananas. Effects of 1-MCP get diminished over time, so there is also a possibility that the 1-MCP amount given for the controlled exposure method was not sufficient to saturate the package environment throughout the treatment. In addition, the 1-MCP gas can also escape from the permeable package, which can deplete the 1-MCP environment inside the package. Therefore the 1-MCP was not able to inhibit the action of ethylene and delay further ripening after 6 days. These results indicate that for controlled exposure to be effective, an initial higher dosage is required to saturate the environment within the package. Also, not all quality parameters were affected in the same magnitudes by 1–MCP. The strongest effect of 1-MCP was found on firmness, the weakest was found on the respiration rate, and the in between was on TSS and color change of treated samples. In this study the rapid change in color was observed in the cases of untreated fruits. Exposure with 1-MCP was able to delay color change from partially green to fully yellow which was found consistent with other studies that were mentioned earlier. Also, there was much  less sugar spotting developed in bananas treated with 1-MCP than bananas without any 1-MCP treatment, in fact

there was no sugar spotting observed in bananas treated with the one-time exposure method. In addition, the color changes also accompanied by softening of the bananas, which occurred faster in untreated bananas than 1-MCP treated bananas. The softening of bananas was significantly delayed with 1-MCP. The 1-MCP treated bananas took longer to turn soft and were more firm than untreated bananas, even after 12 days. The starch to sugar conversion which was measured in terms of TSS content found less in 1-MCP treated fruits compared to untreated fruits, which was consistent for all the experiments. But there was no significant difference observed in the TSS content of bananas for samples treated with controlled exposure treatment or one-time exposure treatment; this indicates that TSS content of bananas may not be influenced greatly by concentration or timing of 1-MCP treatment. All of the quality indicating results found was consistent with the other studies done on bananas. Though there was no particular study found on bananas with different exposure methods of 1-MCP, the results were compared for accuracy with other similar studies.

### 6.2.2. Release of 1-MCP from CD

1-MCP is available in encapsulated form in cyclodextrin due to its highly volatile nature, which requires a moisture trigger to release from cyclodextrin complex. In a CRP system the 1-MCP will be incorporated in an active layer. To be effective on fresh produce 1-MCP must release from the encapsulated form under the atmosphere created in the fresh produce package. Thus the release of 1-MCP is dependent on the humidity of the package headspace which is again dependent on the transpiration of

fresh produce. Therefore the moisture in the package must first penetrate the active layer of film in order to trigger the release of 1-MCP from the cyclodextrin. In this study the experiment was conducted to see if bananas can provide enough moisture to permeate through the film layer and reach the 1-MCP-CD to release the 1-MCP gas. The results showed that moisture released from bananas was sufficient enough to permeate through the film layer to trigger the release of 1-MCP form CD complex. Before designing the CRP system with 1-MCP incorporation for any produce, the moisture activation study is recommended, as different produce have different respiration patterns and the moisture released can be difference for each of them. The moisture release may vary with the type of produce and their respiratory pattern, which can ultimately affect the release of 1-MCP from CD. This study can also help to quantify the amount of produce required per package, in terms of moisture production, to activate the 1-MCP release.

The selection of active film material is mainly dependant on water vapor transmission and 1-MCP gas transmission as they impact the release of 1-MCP molecules from and through the film, which plays a major role in the design of entire packaging system. The water vapor permeability of PVA films is almost 10 times higher than LDPE [51], which is commonly being used as fresh produce packing material. PVA film was used as an example of an active film to study the 1-MCP gas permeability through the film. The result shows that cast PVA film was able to permeate the moisture through the film to facilitate the release of 1-MCP gas from cyclodextrin. The PVA film was also able to greatly slow down the release of 1-

MCP. The casted PVA film slow down the permeation of 1-MCP gas released from cyclodextrin, the1-MCP gas levels for sample with PVA film barrier in headspace of jars were nearly 80 % less even after 6 hours than that of control samples, which shows that it has a good 1-MCP gas barrier. For establishing a CRP system, film such as PVA with moderate to higher moisture permeability and lower 1-MCP permeability would be a best fit.

# 7. CONCLUSIONS

Ethylene antagonist 1-MCP was able to delay the further ripening of partially ripened bananas, which was evaluated by studying the physiological responses of banana to different 1-MCP exposures. Overall, 1-MCP treated bananas were able to maintain better eating quality for 5 to 6 days more than the untreated bananas.

Both the 1-MCP treatments, controlled exposure and one-time exposure were proven to be effective on the quality of partially ripened bananas in the packaging system. But not all quality parameters were affected in the same magnitudes by 1–MCP. The strongest effect of 1-MCP was found on firmness, the weakest was found on the respiration rate, and the in between was on peel color and TSS of treated samples. The effect of 1-MCP on respiration is still not clear. In fruits treated with 1-MCP had a delayed progression of the climacteric phase and maintained the higher respiration rate at the post climacteric phase compared to untreated fruits in the closed jar system. But the result was not completely reproducible when the 1-MCP treatment was given in the permeable package. In the permeable package there was no significant difference found in $CO_2$ gas levels amongst all three samples in headspace of the packages. From this study, it can be concluded that the effect of 1-MCP on the respiration rate of partially ripened bananas is not significant. In the future, research needs to be done to investigate the role of 1-MCP in controlling the respiration rate of partially ripe bananas, especially in a packaging system to further confirm these results for commercial application. The consideration of testing the permeability of packages for 1-MCP and respiration gases would help to provide a

more accurate calculation for respiration rate of bananas in the packaging system.

Both the one time exposure and controlled exposure of 1-MCP treatments showed similar effects on delaying ripening of bananas at least for the initial 6 days. The effects of controlled exposure seem to be less effective after 6-7 days in permeable packages; which may be due to the loss of 1-MCP gas from the packages. The assumption was made for the lessened effect of controlled exposure is that there may be permeation of some 1-MCP gas molecules outside the permeable package. The controlled exposure of 1-MCP was a very small dose, even though with continuous replenishment it may not be sufficient to be effective after some loss of 1-MCP gas leaked from the package. One-time exposure of 1-MCP was almost 10 times larger dose, the small loss of 1-MCP occurred may not affect the package system, assuming that it will have more than a sufficient amount to saturate the environment of the package throughout the treatment cycle. Therefore, while designing an actual controlled release packaging system, the barrier layer has been considered as one of the important factor to avoid the loss of active compound from the package. Also, these results indicate that for controlled exposure to be more effective, it requires initial higher dosages to saturate the package environment. Both types of 1-MCP treatment have potential to be incorporated into the actual packaging system and bring it to a CRP system of 1-MCP to delay further ripening of fresh produce.

The PVA film was used here as an example of film material that can serve as an active layer to control the release of 1-MCP in a CRP system for fresh produce. From the results of experiments it can be concluded that a moderate moisture barrier

film like PVA has potential to be used as an active layer with a barrier layer on the outside of the package to achieve the slow or controlled release of 1-MCP to fresh produce. From the results it can also be concluded that fresh produce can provide enough moisture to activate the release of 1-MCP from CD complex. This experiment was required to confirm the release of 1-MCP from CD through the bananas' moisture for the availability of released 1-MCP gas molecules to fresh produce.

The practical implication of the controlled exposure and one-time exposure of 1-MCP treatment at partially ripened stages is that it can be applied to the fruits in the package itself when exporting the fruits to long distance. While being transported long distances the fruit can have continuous doses of 1-MCP from the package until it reaches the consumer market. Also, initial higher doses of 1-MCP before packaging can be considered for highly perishable fruits to hold the unripe or partially ripened stage throughout the distribution cycle.

Combining everything, the results obtained in this study fulfilled our objective of developing the different 1-MCP exposure methods to treat partially ripened bananas to further delay the ripening. Also, it has provided preliminary information for developing scientific base for designing the CRP system for 1-MCP application to fresh produce.

# 8. FUTURE WORK

The followings are the suggestions for conducting future work based on the work in this thesis.

- Higher doses of 1-MCP for the controlled exposure with different exposing patterns need to be analyzed with different fresh produce.
- More ranges of polymers need to be analyzed to achieve a wider range of controlled release of 1-MCP.
- A multilayered structure in which the outer layer is a high barrier to 1-MCP can be use to prevent loss of 1-MCP gas molecules to the general environment.
- The future work will also need attention set towards preventing the loss of 1-MCP during 1-MCP incorporated through film/sachet storage, which can be prevented by storing films or sachets in low-humidity environments or high gas barrier packages until use.
- Also we know that respiration plays an important role in shelf life of climacteric fruits. But, there is not clear evidence found for a relation of 1-MCP and interchanging respiration gases during this study. However, there are studies showing the effects of 1-MCP suppressing the respiration rate or delaying the respiration peak when applied at the green life of bananas. If these results are consistent, than 1-MCP will be an important factor to consider while designing a CRP system for fresh produce. As this may affect the internal environment of the package directly or indirectly, and can change

the levels of respiration gases in the package. Also, the gas permeability of polymer affect the internal environment of the package, which can affects the respiration rate directly or indirectly, thus the selection of the polymer and calculation for designing the CRP needs to be revised before combining CRP and 1-MCP. If the respiration rate goes higher, the excess heat production can cause faster ripening. And if the respiration rates go really low it may cause fermentation of fruits due to the anaerobic conditions formed in the package. So it requires balancing of the respiration rate in the package based on the effects of 1-MCP.

# 9. REFERENCES


1. USDA, *The food guide pyramid*, in *Home and garden bulletin*. 1992, Center for nutrition policy and promotion: Washington, DC. p. 29.
2. Wu, C.T., *An overview of post harvest biology and technology of fruits and vegetables*, in *Techology on reducing post harvest losses and maintaining quality of frutis and vegetables* 2010, AARDO. p. 2-11.
3. Golding, J.B., et al., *Relationships between respiration, ethylene, and aroma production in ripening banana.* Journal of Agricultural and Food Chemistry, 1999. **47**(4): p. 1646-1651.
4. Jiang, W., et al., *Regulation of 1-MCP-treated banana fruit quality by exogenous ethylene and temperature.* Food Science and Technology International, 2004. **10**(1): p. 15-20.
5. Payasi, A. and G. Sanwal, *Ripening of climacteric fruits and their control.* Journal of food Biochemistry, 2010. **34**: p. 679-710.
6. Serek, M., E.C. Sisler, and M.S. Reid, *Novel gaseous ethylene binding inhibitor prevents ethylene effects in potted flowering plants.* J. Amer. Soc. Hort. Sci., 1994. **119**(6): p. 1230-1233.
7. Farid, M., *Effect of 1-Methylecyclopropene (1-MCP) on shelflife and quality of Cavendish bananas*, in *School of Agriculture, Food and Wine*. 2006, The University of Adelaide. p. 220.
8. Pelayo, C., et al., *Variability in responses of partially ripe bananas to 1-methylcyclopropene.* Postharvest Biology and Technology, 2003. **28**(1): p. 75-85.
9. Sisler, E.C. and M. Serek, *Inhibitors of ethylene responses in plants at the receptor level: Recent developments.* Physiologia Plantarum, 1997. **100**(3): p. 577-582.
10. Blankenship, S.M. and J.M. Dole, *1-Methylcyclopropene: a review.* Postharvest Biology and Technology, 2003. **28**(1): p. 1-25.
11. Nanthachai, N., et al., *Absorption of 1-MCP by fresh produce.* Postharvest Biology and Technology, 2007. **43**(3): p. 291-297.
12. Golding, J.B., et al., *Application of 1-MCP and propylene to identify ethylene-dependent ripening processes in mature banana fruit.* Postharvest Biology and Technology, 1998. **14**(1): p. 87-98.
13. Jiang, Y., D.C. Joyce, and A.J. Macnish, *Responses of banana fruit to treatment with 1-methylcyclopropene.* Plant Growth Regulation, 1999. **28**(2): p. 77-82.
14. Macnish, A.J., et al., *1-Methylcyclopropene treatment efficacy in preventing ethylene perception in banana fruit and grevillea and waxflower flowers*. Vol. 40. 2000, Collingwood, AUSTRALIE: CSIRO.
15. Harris, D.R., et al., *Effect of fruit maturity on efficiency of 1-methylcyclopropene to delay the ripening of bananas.* Postharvest Biology and Technology, 2000. **20**(3): p. 303-308.
16. Hotchkiss, J.H., C.B. Watkins, and D.G. Sanchez, *Release of 1-*

*methylcyclopropene from heat-pressed polymer films.* Journal of Food Science, 2007. **72**(5): p. E330-E334.

17. Ahmad, S., et al., *Effects of storage of bananas in controlled atmosphere before ethylene treatments on its ripening and quality.* Journal of Agricultural Research, 2006. **44**(3): p. 219-228.
18. Noomhorn, A. and S.D. Potey, *Modified atmosphere packaging of banana.* Journal for Hawaiian and Pacific Agriculture, 1993. **4**: p. 69-78.
19. *Banana facts*, in *IITA Research to nourish Africa*. 2008.
20. Watada, A.E., et al. (1984) *Terminology for the description of developmental stages of horticultural crops*. Hortscience **19**, 2.
21. Knee, M., *Fruit quality and its biological basis*. 2002: Sheffield Academic Press. 279.
22. Lippert, F. and M.M. Blanke, *Effect of mechanical harvest and timing of 1-MCP application on respiration and fruit quality of European plums Prunus domestica L.* Postharvest Biology and Technology, 2004. **34**(3): p. 305-311.
23. Lohani, S., P.K. Trivedi, and P. Nath, *Changes in activities of cell wall hydrolases during ethylene-induced ripening in banana: effect of 1-MCP, ABA and IAA.* Postharvest Biology and Technology, 2004. **31**(2): p. 119-126.
24. Sisler, E.C. and S.F. Yang, *Ethylene, the gaseous plant hormone*, in *BioScience*. 1984, American Institute of Biological Sciences. p. 234.
25. Subagio, A. and N. Morita, *Changes in carotenoids and their fatty acid esters in banana peel during ripening.* Food Science and Technology International, Tokyo, 1997. **3**(3): p. 264-268.
26. Cordenunsi, B.R. and F.M. Lajolo, *Starch breakdown during banana ripening: sucrose synthase and sucrose phosphate synthase.* Journal of Agricultural and Food Chemistry, 1995. **43**(2): p. 347-351.
27. Kader, A.A., *Banana, recommendations for maintaining postharvest quality*. 2005, Postharvest Technology Research Information Center , UC DAVIS: Devis, CA.
28. Jiang, Y. and D.C. Joyce, *Softening response of 1-methylcyclopropene-treated banana fruit to high oxygen atmospheres.* Plant Growth Regulation, 2003. **41**(3): p. 225-229.
29. Yahia, E., *Modified and controlled atmospheres for the storage, transportaion, and packaging of horticultural commodites*, ed. E. Yahia. 2009, Boca Raton, FL: CRC Press. 589.
30. Vilas-Boas, E.V.d.B. and A.A. Kader, *Effect of atmospheric modification, 1-MCP and chemicals on quality of fresh-cut banana.* Postharvest Biology and Technology, 2006. **39**(2): p. 155-162.
31. Kader, A.A., *Banana respiration*, in *Banana, Recommendations for Maintaining Postharvest Quality*. 2005, Postharvest Technology Research Information Center , UCDAVIS: Devis, CA.
32. Watkins, C.B., *The use of 1-methylcyclopropene (1-MCP) on fruits and vegetables.* Biotechnology Advances, 2006. **24**(4): p. 389-409.
33. Bleecker, A.B., *The ethylene receptor family from Arabidopsis: structure and*

*function.* Philosophical Transactions of the Royal Society of London. Series B: Biological Sciences, 1998. **353**(1374): p. 1405-1412.

34. Corbineau, F., F. Engelmann, and D. Came, *Ethylene production as an indicator of chilling injury in oil palm (Elaeis guineensis Jacq.) somatic embryos.* Plant Science, 1990. **71**(1): p. 29-34.
35. Ayoub, J.A., Driver, M. G., Taub. I. A. , *Use of ethylene absorbers in extending shelf life*, U.S.A.N.R. Food engineering directorate, Development, and Engineering Center, Editor. 1976, U.S. Army Natick
36. Saltveit, M.E., *Effect of ethylene on quality of fresh fruits and vegetables.* Postharvest Biology and Technology, 1999. **15**(3): p. 279-292.
37. Devlieghere, F., et al., *Modeling the relation between ethylene production rate, respiration rate and their influence on climacteric and non-climacteric fruits.* Acta Horticulture, 2003. **600**: p. 647-651.
38. Ponce, P., Guilherme, L. R., Carbonari, Lugao. A. B., *Active packaging using ethylene absorber to extend shelf-life*, in *International Nuclear Atlantic Conference* 2009, ABEN Rio de Janeiro,RJ, Brazil
39. Watkins, C.B., J.F. Nock, and B.D. Whitaker, *Responses of early, mid and late season apple cultivars to postharvest application of 1-methylcyclopropene (1-MCP) under air and controlled atmosphere storage conditions.* Postharvest Biology and Technology, 2000. **19**(1): p. 17-32.
40. Choi, S.T. and D.J. Huber, *Influence of aqueous 1-methylcyclopropene concentration, immersion duration, and solution longevity on the postharvest ripening of breaker-turning tomato (Solanum lycopersicum L.) fruit.* Postharvest Biology and Technology, 2008. **49**(1): p. 147-154.
41. Blankenship, S., *Ethylene effects and the benefits of 1-MCP.* Perishables Handling Quarterly, 2001(108).
42. Mir, N., *Personal communication*, M. Trivedi, Editor. 2009.
43. Dole, *Banana colour chart*, in *Quality Chain*, Dole Packaged Foods Europe SAS.
44. USEPA, *1- Methylcyclopropene, Registration Action Document*, O.o.P.P. U.S. Environmental Protection Agency, Editor. 2006, Biopesticides and Pollution Prevention Division.
45. Mir, N., et al., *Inhibiting tomato ripening with 1-methylcyclopropene.* J. Amer. Soc. Hort. Sci., 2004. **129**(1): p. 112-120.
46. Tassoni, A., C.B. Watkins, and P.J. Davies, *Inhibition of the ethylene response by 1-MCP in tomato suggests that polyamines are not involved in delaying ripening, but may moderate the rate of ripening or over-ripening.* J. Exp. Bot., 2006. **57**(12): p. 3313-3325.
47. Giovannoni, J.J., *Genetic regulation of fruit development and ripening.* Plant Cell, 2004. **16**(suppl_1): p. S170-180.
48. Gutierrez, M.a.S., et al., *Different responses of goldenberry fruit treated at four maturity stages with the ethylene antagonist 1-methylcyclopropene.* Postharvest Biology and Technology, 2008. **48**(2): p. 199-205.
49. Bellincontro, A., et al., *Postharvest ethylene and 1-MCP treatments both*

*affect phenols, anthocyanins, and aromatic quality of Aleatico grapes and wine*. Australian Journal of Grape and Wine Research, 2006. **12**(2): p. 141-149.

50. Rizzolo, A., et al., *Effect of 1-methylcyclopropene on aroma compounds in "big top" nectarines after shelf life.* Journal of Food Quality, 2006. **29**(2): p. 184-202.
51. Lee, Y.S., et al., *Development of a 1-Methylcyclopropene (1-MCP) sachet release system.* Journal of Food Science, 2006. **71**(1): p. C1-C6.
52. Yam, K.L., Schaich, K. M., *Development of controlled release packaging, USDA proposal*. 2005, Rutgers University.
53. Zhu, X., *Development of target release rate concept for controlled release packaging*, in *Graduate Program in Food Science*. 2008, Rutgers, The State University of New Jersey: New Brunswick. p. 206.
54. Macnish, A.J., et al., *A simple sustained release device for the ethylene binding inhibitor 1-methylcyclopropene.* Postharvest Biology and Technology, 2004. **32**(3): p. 321-338.