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**THE EFFECT OF SURFACE TREATMENTS  
AND MODIFIED ATMOSPHERE PACKAGING (MAP) ON  
THE QUALITY OF FRESH CUT SLICED ANJOU PEARS**

presented by

**Raafia Siddiq**

has been accepted towards fulfillment  
of the requirements for the

          M.S.           degree in           Packaging          



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**THE EFFECT OF SURFACE TREATMENTS  
AND MODIFIED ATMOSPHERE PACKAGING (MAP) ON  
THE QUALITY OF FRESH CUT SLICED ANJOU PEARS**

**By**

**Raafia Siddiq**

**A THESIS**

**Submitted to  
Michigan State University  
in partial fulfillment of the requirements  
for the degree of**

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## **ABSTRACT**

### **THE EFFECT OF SURFACE TREATMENTS AND MODIFIED ATMOSPHERE PACKAGING (MAP) ON THE QUALITY OF FRESH CUT SLICED ANJOU PEARS**

**By**

**Raafia Siddiq**

Shelf life and quality of fresh-cut Anjou pears was assessed using two surface treatments at 2% concentration levels (Nature Seal-NS and Sodium Acid Sulfate-SAS) with two package types (Modified Atmosphere Packaging–MAP and non MAP). The NS no MAP, NS + MAP, SAS no MAP, and SAS + MAP pears were studied for headspace gas, color, pH, TSS, TA, bacteria, yeast and mold counts over a storage period of 21 days at 4°C. Preliminary storage studies found that treatment type and MAP, showed significant differences between the pears in O<sub>2</sub> levels, color a\* value, pH, bacteria, yeast, and mold growth. Visual observation showed significant browning and tissue softening in the SAS pears. Thus, SAS was not included in further studies due to its effect on pear quality. NS + MAP pears had significantly better physical quality and shelf life.

Final storage studies with NS no MAP and NS + MAP found significant differences in CO<sub>2</sub> and O<sub>2</sub> levels, color, bacteria, yeast, and mold growth. No significant difference occurred in pH, TSS and TA. No significant difference in the aroma, flavor, texture and overall acceptability was found for NS + MAP pears versus freshly prepared control. Appearance was the only attribute to be significantly different. NS + MAP pears had a shelf life of up to 21 days at 4°C.



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## INTRODUCTION

Today's consumer has an increased interest in living a healthy lifestyle and eating right plays a big role in achieving that. When it comes to eating healthy foods, finding ways to incorporate fruits and vegetables into daily meals, not just for adults but also for children, is extremely important. The production and consumption of fresh cut fruits and vegetables is increasing substantially in the U.S. market. According to United Fresh's "Fresh Facts on Fresh Cut" report, fresh-cut produce continued to experience strong sales at retail with 2006 Q3 sales totaling more than \$1.5 billion. This number represents an 11.5% increase from 2005 (UFPA, 2006). However, the most recent statistics show that the retail sales of fresh cut fruits and vegetables lost both dollar sales and volume in the third quarter of 2009. Fresh-cut fruit increased its volume by 3 percent in 2009, but dollar sales declined 3 percent and the average retail price was down 6 percent to \$3.85. The fresh-cut fruit category experienced a 5% decline from Q3 2008, while the fresh-cut vegetable category also experienced similar declines (Christie, 2010). The decline could be due to many reasons, however most importantly, the recession probably played big role in these declines.

Fresh cut produce is defined as "Any fresh fruit or vegetable or any combination thereof that has been physically altered from its original form, but remains in a fresh state" (IFPA, 2004). Today's consumer has an increasingly busy lifestyle and has limited time to prepare meals. Thus, it is very important to continue to find ways to provide fresh fruits and vegetables that have extended shelf lives but are quick and convenient to access. Development of fresh-cut



produce systems, also called “minimally processed” should include two goals: (1) Keeping the produce fresh by extending its shelf-life without losing its nutritional and sensory quality or compromising product safety, and (2) ensuring a product shelf-life that is sufficient to make its distribution feasible within the region of its consumption (Vasconcellos, 2000; Laurila and Ahvenainen, 2002).

It is extremely important to monitor various physical, chemical and biological aspects of fresh cut products in order to determine whether any alterations to the product occur, and whether these increase or decrease the overall quality of the product. There are many methods available that can maintain the shelf-life and quality of fresh cut produce. Many of these methods can also be combined to increase the overall quality. These include but are not limited to: barrier materials technology, sanitizing methods, product anti-browning treatments, optimization of processing and storage temperatures and headspace control techniques such as modified atmosphere packaging (MAP).

MAP is a term applied to a range of food packaging technologies that rely on mixtures of the atmospheric gases; oxygen ( $O_2$ ), carbon dioxide ( $CO_2$ ), and nitrogen ( $N_2$ ), in concentrations different than those in air, to retard deterioration processes in foods (Brody and Marsh, 1997). MAP is one of the most beneficial ways to extend product shelf life from farm to fork. Although there are many various forms of MAP, in this study a passive MAP approach was used by taking medical grade (sanitary) air and injecting it into packages. Also, the choice of package type helps to regulate the exchange of gas going in and out of the package. The optimal range for keeping pears from ripening too much is to keep

the O<sub>2</sub> level below 1% or between 0-1% (Mitcham et al, 2009). In the passive MAP environment an atmosphere high in CO<sub>2</sub> and low in O<sub>2</sub> passively evolves within a sealed package over time as a result of the respiration of the product. The gas permeability's of the packaging films are such that the proper amount of O<sub>2</sub> can enter while excess CO<sub>2</sub> can exit, so that the product does not go into anaerobic respiration and spoil (Robertson, 2006).

D'Anjou (also known as Anjou) pears were chosen as the produce to test. This pear variety is available from October to June which allowed for both preliminary testing and storage study to be completed in close proximity. The two treatments used in this study were Sodium Acid Sulfate (SAS) and Nature Seal (NS). SAS is a natural food acid with the ability to lower the pH of a product without creating a sour taste. It also helps in maintaining microbial stability something that is important for all food products (Jones-Hamilton). NS is a patented blend of vitamins and minerals that extend the shelf life of fresh-cut produce for up to 21 days by inhibiting the respiration and oxidation process (Nature Seal, 2004). Not much research has been done on the use of SAS or NS on pears, thus, there was an interest to see the effect of both treatments on the quality of the pears. NS was also used as the control treatment, because it was known that pears without any treatment would not last longer than a few days. Therefore, NS due to its past research and literature was used as the control to compare to SAS. Pears are also not available today in snack packs the way apples are at food chains such as Subway and McDonalds. Therefore, there was interest to see if a packaging could be developed which would make

fresh sliced pears available to consumers on the go. Pears are important to the diet because they are low in fat, cholesterol and sodium free as well as a great source of fiber and vitamin C.

Pears were bought fresh from the same location and rinsed with 30 grams of SC Johnson Fruit/Vegetable wash in 2 gallons of water. They were then cut into slices using a corer/slicer machine and dipped into the corresponding treatments of NS or SAS for 60 seconds, which had already been dissolved in 1 gallon of distilled water. Following this they were manually shaken to release excess treatment solution and then packaged 4 pear slices per container. Each container was sealed off with a lid and put into cold storage immediately. The half of the samples that were MAP were quickly taken to the multi-vac machine which had been loaded with the roll stock film. Proper sanitation techniques were used throughout the study to limit the chance of contamination. On days 0, 3, 7, 10, 14, 17 and 21 two packages of each treatment were taken out and evaluated using a series of tests. For the second storage study the SAS treatment was removed due to lack of acceptable physical quality.

The objectives of this research were to: (1) assess the physical-chemical changes in fresh cut pears over time in both a modified atmosphere package and a control package, (2) To determine if MAP can be used to extend the shelf life of fresh-cut pears, and (3) To determine if the two product treatments, NS and SAS helped improve product quality.



## **LITERATURE REVIEW**

### **I. Fresh-Cut Fruits and Vegetables**

#### **a. History**

Fresh fruits and vegetables have been a staple of human consumption for many centuries. A key part of every human diet, fruits and vegetables provide many different types of vitamins and minerals that are essential to a healthy and functional body. The fresh-cut industry is a relatively new sector which has achieved prominence during the past decade or so, especially with the ever changing consumer lifestyle (Beaulieu, 2007). Traditionally, most produce is bought whole from grocery stores, brought home, washed, chopped, cut, or diced in whatever size desired. The leftovers would then need to be stored away for later use. Healthy living was not as much of a driving factor ten years ago, however, due to the increase in its importance now, the fresh-cut business is booming (Lamikanra, 2002). Some of the first fresh-cut products that appeared in the retail market were lettuce and cut salads, however, now there is a wide variety of different fresh-cuts available.

#### **b. Usage**

Fresh-cut produce is one of the fastest growing sectors of the food industry in the United States. Retail sales of fresh-cut produce grew from \$5 billion in 1994 to over \$10 billion in 2003 (Gertmenian, 2003). The consumption trends usually vary by age groups. For example, seniors eat fewer French fries and potatoes chips and more fresh and canned potatoes compared to younger

consumers (USDA). The USDA report also showed that toddlers ate more apples both fresh and processed, whereas adults ages 20-59 consumed the fewest apples. Another trend is that women over the age of 40 eat the most spinach, whereas teenage girls eat the least (USDA).

Another factor that plays in to the usage of fresh cuts is family income. High income families (USDA) ate more of many vegetables including fresh celery, garlic, cucumbers, bell peppers, mushrooms and tomatoes. High income families also drank more orange juice, whereas low income families drank more orange flavored drinks, which had less than 10% juice.

### **c. Nutritional and Health benefits**

A concern among many consumers is whether or not fresh-cuts are as nutritious as whole produce. A great deal of research has been done on this topic. In general, whatever process is used to preserve the freshness in a product is also used to preserve the nutrient content. There is no significant difference in the nutrient content of important vitamins like Vitamin A and Vitamin C when comparing fresh-cuts to whole products. Fresh-cuts tend to spoil before they are given a chance to experience significant nutrient loss. One of the main areas where nutrition loss may occur is in harvesting of the product (PBH, 2005). The nutritional quality of a fresh product is at its best immediately after harvesting. The longer the product is stored, the more it experiences a decrease in quality.

The Produce for Better Health Foundation (PBH, 2005) published a study on broccoli which showed that when a whole head of broccoli was stored, at the

same temperature and same period of time as florets there was no difference in the nutritional quality. Furthermore, both samples maintained adequate amounts of Vitamin C. When shredded carrots (PBH, 2005) were tested at a storage temperature of 38°F - 45°F for 10 days they showed no decrease in the amount of carotenoid content. Kiwi fruits were sliced and tested at 32°F for 6 days and showed no decrease in Vitamin C. In the same study by PBH, fresh mangos were cut into cubes and dipped into Vitamin C to prevent any surface browning. After being stored for 10 days at 41°C, they had the same amount of Vitamin C as the freshly cut fruit. Also Fresh-cut oranges were tested for 12 days at 39°C and showed no loss in Vitamin C. Fresh-cut strawberries that were held for 7 days at 41°C showed no loss in Vitamin C compared to whole strawberries (PBH, 2005).

A diet rich in fruits and vegetables has been shown to lower blood pressure, reduce the risk of heart disease, stroke, and some cancers and lower the risk of eye and digestive problems as reported by the Harvard School of Public Health (Vegetables and Fruits). For good health the USDA urges Americans to eat more fruits and vegetables, about 5-9 servings per day.

#### **d. Convenience and Functionality**

The increase in the popularity of fresh-cut produce is especially important for the busy life style of today. With a tough economy as well as both parents working there is less time available for families to prepare meals from scratch; fresh-cuts provide a healthy and nutritious way to consume fruits and vegetables directly or as accessories to meal preparation. It is very important to find a quick

and convenient way to provide consumers with their daily servings of fruits and vegetables. At the same time it is also important to provide an increased shelf life (~ 2 weeks) for these products, so that consumers have adequate time to use fresh-cuts in their diet. For the on the go lifestyle; ease of use, convenience and freshness are key factors that consumers look for when grocery shopping.

Fresh-cut fruit and vegetable trays with dips have become quite popular appetizers in both professional and social gatherings. Fresh-cuts have also become more important in fast food restaurants such as McDonalds and Subway where fresh-cut apples are available (Agriculture Research Service of the USDA, 2006). More and more companies and grocery stores are beginning to understand the importance of fresh-cuts and are beginning to provide more options for consumers. Fresh-cut vegetables include but are not limited to salad mixes, shredded cabbage, shredded lettuce, sliced and diced tomatoes, peeled baby carrots, chopped broccoli/cauliflower, sticks of celery and much more. The Agriculture Research Service also reported that fresh-cut fruits include melons, strawberries, pineapples, grapefruits, and many more (Bliss 2006). Many of these fresh-cut options are essential meal components and having them available can significantly cut down on meal preparation time.

The convenience of fresh-cuts in making the lives of consumers easier is important in today's lifestyles. Various uses of fresh-cuts in food delivery are important whether as snacks, meals, desserts, and healthy appetizers, for social or professional gatherings. Providing alternative ways for consumers to get their

daily servings of fruits and vegetables both quickly and conveniently is and will continue to be a very important part of the modern lifestyle and diet.

## **II. Processing and Preservation Techniques**

### **a. Washing and Sanitizing**

Washing and sanitizing is usually done as soon as possible after the product is harvested and often times a second and third wash will be used after they have been cut or peeled. Thomas Ohlsson and Nils Bengtsson (2002) reported in their book "Minimal Processing Technologies in the Food Industry" that washing and sanitizing products immediately after they have been cut greatly reduces the risk of microbial growth and oxidation since the tissue fluid and microbes are often removed. The enzymatic browning that occurs after a fruit or vegetable is cut can be inhibited by dipping or rinsing cut products in a specific solution such as Nature Seal or fruit and vegetable wash. The preferred methods for washing include flowing water, or air bubbling water. However, rinsing with water alone is not recommended since the microbes and dirt that come from the prior washes can contaminate the next batch of product. The temperature of the water used should be around 41°F (5°C) as reported by Ohlsson and Bengtsson (2002). Preservatives can also be used in the water to decrease the amount of microbes and to slow down enzymatic activity. This can help to increase the shelf life of products. Chlorine can also be used before or after the cutting and peeling step in combination with water to help control bacteria and viruses. It is very important to make sure the concentration of



chlorine used to rinse the product is reduced to the same level of drinking water in order to allow the product to be edible (Ohlsson and Bengtsson, 2002).

Sanitizers may include various other treatments, such as: chlorine dioxide, trisodium phosphate and hydrogen peroxide (Russell and Gould, 2003). Hydrogen peroxide has been shown to decrease the amount of microbes on fresh-cut bell peppers, zucchini and cucumber, resulting in an increase in the shelf life. In addition, no residues are left behind. In a study done on mushrooms; adding a pre-wash step using 0.5% to 1% H<sub>2</sub>O<sub>2</sub>, increasing the wash solution H<sub>2</sub>O<sub>2</sub> concentration from 3% to 5%, and substituting 4% sodium erythorbate + 0.1% NaCl for a more complex browning inhibitor formulation showed that mushrooms were free of adhering soil, less subject to brown blotch than conventionally washed mushrooms, and at least as resistant to enzymatic browning as unwashed mushrooms during storage at 4 degrees C (Sapers et al., 2001).

After rinsing, the water etc. can be removed by gently centrifuging or by using an air tunnel depending on the size and shape of the product. The amount of time used to dry products should be carefully selected so as to only remove the excess water and not damage any of the cells (Lamikanra, 2002). In the present study, a sanitizing/washing treatment was used in dissolving 30 grams of the powder (SC Johnson Fruit and Vegetable wash) in 2 gallons of distilled water, before the pears were cut. This was then used to remove any surface contaminants. Two main sanitizing and anti-browning treatments were used in this project. A sodium acid sulfate (SAS) treatment and a calcium ascorbate

treatment commonly known as Nature Seal were used. SAS is a “natural” food acid with the unique ability to lower the pH on the surface of products without generating a sour taste and has recently been used for fresh- cut produce. In a study by Fan et al (2009), granny smith apples were tested with various anti-browning agents, one of which was SAS. Results from the study showed that SAS was the most effective in inhibiting browning and microbial growth; however, it caused some skin discoloration and damage.

Nature seal has often been compared to SAS and is used extensively in the treatment of fresh-cut produce. It is a combination of vitamins and minerals which inhibit discoloration (browning), while maintaining the natural taste, texture and color of the product, thus extending its shelf-life.

**b. Anti-Browning (including MAP)**

One of the biggest problems with fresh-cuts is enzymatic browning. This causes a substantial decrease in the shelf life of many fresh produces. The shelf life of many fresh produce products is limited due to excessive tissue softening and cut surface browning. The cut surface browning in sliced pears is caused by the action of polyphenol oxidase (PPO) on phenolic compounds released during the cutting process (Amiot et al., 1995). Browning requires four different components: oxygen, an enzyme, copper and a substrate (Vamos-Vigyazo, 1981). Removing any one of these components will prevent browning from occurring. Browning not only causes a color change; but it also affects the taste/texture of the product and can cause a decrease in nutrients. PPO activity

varies among different fruits and vegetables as well as within cultivars of the same crop.

Washing with water alone does not inhibit the oxidation reactions, further steps need to be taken in order to control the problem. Numerous chemical and physical preservation strategies can be used to reduce enzymatic browning and fruit tissue softening after cutting. However, some of these treatments impart off flavors or the compounds used may not be generally recognized as safe (GRAS) by the Food and Drug Administration (McEvily and Iyengar, 1992). Some of the safer ways to prevent browning are; heating to inactivate the enzyme, removal of the oxygen and phenols, lowering the pH two or more units below the optimum browning level, or adding compounds that inhibit PPO.

Blanching and cooking processes decrease the potential for enzymatic browning. Since PPO has low thermal stability, it can be readily inactivated by cooking a product at high temperatures (Mayer, 1987). However, high heating can also cause loss of nutrients, texture and flavor. Thus, care needs to be taken when cooking a product to prevent browning, because there could be negative consequences.

Chemical inhibition methods are often used to prevent browning. One such method is using acidulants, which are chemicals that lower the pH of a product. Malic acid and citric acid are chemicals commonly used to do this. Reducing agents are also another type of anti-browning agent. These cause a chemical conversion of colorless *o*-quinones back to *o*-diphenols. One reducing agent commonly used is ascorbic acid, since it also lowers the pH and has a

direct effect on the PPO (Vamos-Vigyazo, 1981). Rosen and Kader (1989) indicated that a combination of 1% CaCl<sub>2</sub> dip with 0.5% O<sub>2</sub> and 12% CO<sub>2</sub> atmosphere maintained firmness and reduced the browning rates of Bartlett pear slices. Another study showed that when combining 1% calcium lactate and 2% ascorbic acid as a post cutting dip, it reduced the incidence of cut surface browning and firmness loss in fresh-cut pears (Gorny et al., 1998). Dong et al. (2000) reported that a 2 minute dip in 0.01% 4-hexylresorcinol + 0.5% ascorbic acid + 1% calcium lactate resulted in a shelf life of 15-30 days at 2-3° C for Anjou, Bartlett and Bosc pears. Sliced Anjou pears also had browning free color for 30 days by dipping them in 1% ascorbic acid and 1% calcium lactate; however the texture was softened due to the juice leakage (Dong et al., 2000). The peroxidase activity also decreased with storage and was inhibited by using an ascorbic acid treatment in fresh-cut cantaloupe melons (Lamikanra, 2001).

The thiol containing amino acid cysteine has been reported to effectively inhibit the PPO mediated enzymatic browning of fruits and vegetables (Josyln and Ponting, 1951) and has been reported to completely inhibit enzymatic browning in fresh-cut potatoes (Gunes and Lee, 1997) when a mixture of 2% citric acid and 0.5% L-cysteine was used. Cysteine is a naturally occurring amino acid that has GRAS status. When cysteine was used as an inhibitor of enzymatic browning on sliced apples (Walker and Reddish, 1964) and pears (Sapers and Miller, 1998), a pinkish-red colored compound was formed due to phenol regeneration with deep color formation (Richard-Forget et al., 1992).

Chelating agents can also be used to inhibit the PPO as well as complexing agents, which entrap or form complexes with PPO. An enzyme inhibitor such as 4-hexylresorcinol is very useful for products such as shrimp, however it still needs to be approved by the FDA for use on fresh-cuts. Sodium chloride is also known to inhibit PPO, as the pH decreases. Sometimes calcium is used to make tissue firmer, thus decreasing its browning. Generally, the chemicals used to inhibit browning are in a solution in combination with other chemicals and the fresh-cuts are dipped into (Lamikanra, 2002) the solution.

Physical methods used to help control enzymatic browning include lowering the storage temperature, using modified atmosphere packaging (MAP), adding edible coatings, or treating with gamma radiation or high pressure (Lamikanra, 2002). One of the most common methods is lowering the temperature during the handling, processing and storage steps. Another method is to decrease the amount of oxygen, using modified atmosphere packaging (MAP), and another is gamma radiation. Reducing the amount of available oxygen decreases the chances of a reaction taking place on the surface of the cut fruit or vegetable. You cannot, however, remove all the oxygen, because the living tissues still need a certain amount of oxygen. Atmospheres reduced in O<sub>2</sub> and elevated in CO<sub>2</sub> can also extend the postharvest life of many whole commodities (Kader, 1986). Low O<sub>2</sub> and/or elevated CO<sub>2</sub> environments as generated by modified atmosphere packaging of fresh-cut produce can extend produce shelf-life by: slowing the browning reaction at the cut surfaces, reducing the rates of product respiration and reducing C<sub>2</sub>H<sub>4</sub> biosynthesis and action

(Gorny, 1997). Fresh-cut iceberg lettuce products are routinely commercially packaged in plastic bags that have low O<sub>2</sub> (< .5%) and elevated CO<sub>2</sub> (>10%) atmosphere to reduce the cut surface browning.

By using MAP, a controlled environment for the product can be created in a package. This process creates an ideal gas composition in the package either through a commodity generated modified atmosphere, passive MAP or by active creation (Active MAP) of a modified atmosphere. Careful control of the appropriate gas composition needs to be made to cater to a specific product. MAP has become a widely used food preservation technique because it preserves the freshness of food and fits well with consumer preferences for fresh and additive-free foods (Day, 1994). A study done on shredded iceberg lettuce at 5°C in a polyethylene package showed browning inhibition for 10+ days. After the storage period the lettuce visual quality was still good, while the unsealed package of lettuce showed poor quality after the same time period (Peabody, 2005).

Coating and enrobing a product can also increase the shelf life of a product and prevent browning; these are thin protective layers, which are applied to a specific product. These replace the natural protective tissue (epidermis) or the peel, which are often times removed in processing. When the peel and tissue are removed it provides a way for microbial contamination to take place. Removing the peel also causes an increase in the respiration rate, which has an inverse effect on shelf life. Increasing the rate of respiration will decrease shelf life. An example of this is with carrots, in one study peeled carrots showed an

increase of 15% in their respiration rate compared to unpeeled carrots (Ohlsson and Bengtsson, 2002). Ohlsson and Bengtsson also found that with shredded lettuce there was an increase of 35-40% in the respiration rate compared to quartered lettuce. Many factors affect the shelf life of fresh-cut pear slices including the cultivar, stage of ripeness at cutting and storage regime before processing (Sapers and Miller, 1998; Gorny et al., 2000). Edible coatings help reduce the amount of respiration, decrease water loss and color change, improve the texture and mechanical integrity, improve the handling characteristics, help retain volatile flavor compounds and reduce microbial growth. Edible coatings can include one or more of the following compounds, proteins, resins, waxes and oils. Their characteristics can be improved by adding plasticizers, surfactants, and emulsifiers. Edible coatings need to be chosen carefully to make sure they adhere to the product as well as provide the proper barrier properties. When careful measures are taken and the appropriate coatings are applied they can provide superior browning control.

Low temperatures are also very helpful in controlling the browning mechanism. At low temperatures the enzyme activity and the metabolic rate decrease; combined, these effects give the product a longer shelf life. Fresh-cuts should be kept at temperatures slightly above freezing. The correct temperature needs to be chosen in order to prevent chilling injury. Often times, the fresh-cuts are immediately rinsed with cold water after being peeled or cut. Then they are usually dried off in a centrifuge or air tunnel to remove excess water (Cardwell, 2007).

Gamma Radiation can also be applied to fresh-cuts, especially to control insects and diseases, and to slow down ripening and sprouting. Gamma radiation can be used to increase the shelf life; however, it can also induce biochemical changes. In one case, apples and pears were irradiated, and after the treatment they were shown to have a decrease in their firmness (Ohlsson and Bengtsson, 2002). Many consumers are weary of eating a product that is radiated, because they fear they will somehow be affected by the radiation.

### **c. Firmness/Texture preservation**

Maintaining the product firmness and texture are very important when trying to sell products to consumers. Many consumers will not buy products that are bruised or oddly shaped. Over 90% of consumers take appearance into account when purchasing products. To improve this characteristic several different methods can be used. One method is to use calcium chloride or calcium lactate in combination with ascorbic acid and cysteine in a two-minute dip to slow down the browning and increase firmness retention. This process is used mainly in fresh-cut fruits. By combining it with a low temperature blanching the enzyme pectinesterase can be inactivated. This should be done before the actual dip to help preserve the texture. Gorny et al. (2002), used a post-cutting dip of 2% (w/v) ascorbic acid, 1% (w/v) calcium lactate and 0.5% (w/v) cysteine adjusted to pH 7.0 to significantly extend the shelf-life of Bartlett pear slices, and to reduce the loss of slice flesh firmness, while preventing cut surface browning. Heat treatments have also been shown to benefit the texture of fresh-cuts (Lamikanra, 2002).



In a study on potatoes, various multiple step processes were used to delay the enzymatic browning, once browning had begun, to limit the extent of that enzymatic browning. The process caused little or no hardening or separation of the potato surface tissue in the final cooked product and was found to be a better alternative than using sulfites for raw, peeled potatoes (Martin et al, 1999). In another study, peeled potatoes were treated with heated ascorbic/citric acid solutions to control the browning which helped firm and separate superficial tissues which affected the texture after being mashed and sliced. The superficial parenchyma cells were examined in cooked potatoes using scanning and transmission electron microscopy. After examination it was found that the cell wall rigidity and middle lamella retention were higher in samples treated with browning inhibitors compared to untreated controls. Mashed potato lumps, prepared from treated samples also behaved similarly (Sapers et al, 1997).

Appearance and textural changes are related to tissue deterioration, and are used as measures of freshness and quality decline in fresh-cuts (Cantwell and Suslow, 2002). Firmness retention is a very important quality characteristic for most fruits and vegetables. Pectin occurs in most plant materials and is a major part of the cell wall. Pectin provides firmness to the plant tissues by adhering to the juxtaposed cell walls in the plant. When pectic substances start to depolymerize, plant tissue starts to soften. Storage temperature influences pectic enzyme activity and fruit firmness. The firmness decreases more quickly at lower temperatures. When the cell structure is destroyed it can lead to

biochemical spoilage such as texture breakdown, off-flavor development, and browning (Ohlsson and Bengtsson, 2002).

Lamikanra (2002) stated that gamma irradiated apple slices showed a reduction in firmness that was dependant on the amount of the dose applied. Softening often occurs due to radiation. However, when a calcium chloride treatment is instituted prior to irradiation, it decreases the softening of the product. Controlled atmospheres and lower respiration rates also slow down tissue softening. Preventing water loss is important in maintaining firmness and texture. Once fresh-cuts are peeled or cut, they are no longer intact. The peel is a very important barrier to the loss of turgor or desiccation. Thus, removing the peel makes a product more perishable and susceptible to water loss (Lamikanra, 2002). Many fresh-cuts have waxy coatings that are very resistant to water loss.

The use of mild heat treatments has been found to have a positive impact on the physiological characteristics of fresh-cut fruits and vegetables. Using heat shock treatments of 45°C for 120 seconds, 50°C for 60 seconds, or 55°C for 30 seconds reduced the wound inducing phenols. When apples were put through this treatment before cutting they proved to have less browning as well as a firmer texture. Heat treatments inhibit ethylene synthesis, tissue responses to ethylene and degradation of the cell wall. Heat treatments of up to 60°C for one minute also improve the calcium chloride intake in products and improve their firmness (Lamikanra, 2002).

PPO has been shown to have maximum activity in the temperature ranges of 20-35°C. Since PPO is not a very heat stable enzyme, its inactivation can

occur at temperatures higher than 40°C (Lamikanra, 2002). Depending on the cultivar, pH and location where the product was grown this temperature can vary. Low temperature blanching is also effective in preventing and controlling enzyme activity in fresh-cut produce. For cubed pears blanching can be done for 3 minutes at 95°C, which can result in complete inhibition of enzymatic browning (Lamikanra, 2002).

#### **d. Shipping/storage conditions**

Proper storage and handling conditions need to be maintained for any perishable product, especially fresh fruits and vegetables. Different conditions and methods are used to assure preservation of products. One general practice is having a “forward only” movement on the processing line (Lamikanra, 2002). This ensures that there is no crossover between raw material and the clean products. There also needs to be adequate separation between the trimming room, the washing room and the packing room to prevent cross contamination. Temperature control is very important especially during processing. Many types of control devices can be put in place to make sure the temperature in each processing room is correct. The airflow also needs to be monitored to maintain room temperatures and to prevent both circulation of dust, and condensation from occurring (Lamikanra, 2002).

Waste removal is very important and needs occur as soon as possible to avoid any type of contamination. Waste cans used for edibles and non-edibles should be properly labeled and easy to wash and sanitize. All other equipment and materials as well as utensils should also be easy to wash. This can be done

via scrubbing, brushing, water jet spraying, and using chemicals such as acidic or alkali detergents. Sanitizing should be done through using either chemicals or steam, or both.

Training of employees properly, encouraging personal health and hygiene among all staff, keeping both neat and sanitary buildings and equipment, having sanitary operations in place, and making sure the amount of time from processing of produce to packaging and transport is kept at a minimum are very important. One of the most important elements of the packaging stage includes printing the “best before date” to ensure customers use the product before its “use by” date (Lamikanra, 2002).

In 2006, the FDA issued draft guidelines for the safe production of fresh-cut fruits and vegetables. One of the key points mentioned was encouraging the implementation of safe practices by everyone involved in the supply chain. To enhance product shelf life means to start from the very beginning including the produce grower, and then the packers, distributors, transporters, importers, exporters, retailers, food service operators, and finally the consumers.

Distribution logistics present significant challenges as national or regional processors deliver most fresh-cut products. In the United States, most of the processors are located near the centers of crop production and processed products are shipped as far as 2500 miles by the national processors (Watada et al., 1996). For regional processors, the shipping distances may be significantly less than that of the national processors. Long-distance shipping and distribution operations can have a detrimental effect on the quality of fresh-cut

products. Fresh-cut products are also subjected to stress during the physical action of transportation, such as vibration. Other factors that may contribute negatively to product quality and safety during transit include temperature and relative humidity fluctuations, and damage to packaging.

Product quality especially needs to be monitored during the transporting and retailing steps. During transportation refrigeration should be used to maintain the temperature below 5°C. The time between transportation of products and loading times should also be managed so that temperature fluctuations, which occur, can be balanced out in such a way to not cause quality problems. When products are brought to the retail stores they are often left in storage for extended time and products may be left on the loading dock for long amounts of time, both of which can be very detrimental to the product. By careful control of processing and of the supply chain a product of optimal quality can be delivered (CFSAN, 2001).

Temperature is one of the key factors in maintaining product quality after harvest, since refrigeration is vital in controlling the respiration rate of fruits and vegetables. Respiration generates heat as sugars, fats, and proteins in the cells of the crop are oxidized. The loss of these nutrients through respiration means decreased food value, loss of flavor, and more rapid deterioration. The respiration rate of a product strongly determines its transit and postharvest life. The higher the storage temperature, the higher the respiration rate (Wilson, 1995). In order for refrigeration to substantially postpone deterioration, the temperature in cold storage rooms has to be kept as constant as possible. When

temperature fluctuations occur between warm and cold there may be a certain amount of moisture accumulation on the surface of packages and products which can then drip onto the fresh produce. The storage room should have proper insulation and adequate refrigeration capacity. There should be an air circulation mechanism in place to prevent temperature variation. Temperature controls and monitors such as: thermometers and thermostats should be checked periodically to ensure high quality and accuracy (Hardenburg, 1986). The presence of cooling facilities on the farm is very important to produce operation. A grower that is able to cool and store their produce immediately has a greater chance of being able to provide a product to the market at the same or nearly same quality as when it was harvested.

Pre cooling is a key step in managing the temperature of fresh produce. The heat which a product holds needs to be removed as quickly as possible before any shipping, processing or storage. This pre-cooling step is particularly important for produce with high respiration rates (Hardenburg, 1986). There are three different methods for pre cooling: room-cooling which is where produce is put into an insulated room with refrigeration units; fan-cooling where fans are used along with a cooling room to push cool air through the packages of produce (fans also have thermostats so they can automatically shut off when the desired product temperature is reached), and finally; hydro-cooling where the produce is dumped into cold water or cold water is flowed over produce. The hydro-cooling process helps both at cleaning and cooling the product, however, it can't be used with produce that is susceptible to wetting (Wilson, 1995).

Top or liquid icing is useful for dense products and palletized products that can't be easily cooled with forced air. In this process crushed ice is added to the top of produce by hand or machine. For liquid icing, slurry of water and ice is injected into distribution packages through vents or handholds without removing the package from the pallet (Anon, 1992). This icing method works well for high respiration produce such as sweet corn or broccoli (Howell, 1993). Vacuum cooling is another method where produce is put in a chamber and a vacuum is created which helps remove the heat from the tissues. This is a method more commonly used for leafy crops such as lettuce. Although this is a useful method, it is quite a bit more costly than other methods (Sasseville, 1988). Refrigerated trucks are made to maintain the cool temperature of the product, not to actually cool produce down since there are no mechanisms for air movement.

In storage or prior to processing many fresh produces require a mix of low temperature and high humidity, which is not easily attainable. Some produce require low temperatures and low humidity, and others require something in between. Root crops are best stored at 32°F and 90% humidity; these include products such as beets, carrots, turnips and leeks. Potatoes prefer temperatures of 40-60°F and 90% humidity. Garlic and onions prefer a temperature of 32°F but less humidity at about 65-75%. Winter squash prefer temperatures of 50-60°F but very little humidity (Byczynski, 1997). These methods can help to maintain the shelf life of fresh produce. It is important to make sure that as soon as a product is harvested it is immediately put in some form of pre-cooling so that when it is being transported between markets it can maintain a temperature that

is ideal for extending its shelf life. Improvements are always being made on how to provide optimal environmental conditions (relative humidity, temperature, gas concentrations) within transport vehicles. The use of controlled atmospheres (CA) and modified atmospheres (MA) in fresh fruits and vegetables continues to grow in transport vehicles since the ability to monitor the oxygen and carbon dioxide levels has increased. The CA process helps to maintain proper relative humidity and optimal temperature during the transport and storage of fresh fruits and vegetables. Both edible coatings and films with appropriate gas permeability's, and modified atmospheres can be of assistance during transportation and distribution. MAP is especially helpful during shipping and storage since it maintains the proper ratio of oxygen and carbon dioxide in a package (Kader, 2002).

### **III. Quality (Physical and Chemical Characteristics)**

The quality of fresh-cut produce depends upon the quality of the intact commodity from the farm, through preparation, and subsequent handling and transport. Quality factors include visual appearance (freshness, color, defects, and decay), texture (crispiness, turgidity, firmness, and tissue integrity), flavor (taste and smell), nutritive value (vitamins, minerals, and dietary fiber), and safety i.e., absence of chemical residues and microbial contamination (Kader and Mitcham 1996; Piagentini et al., 2002). Thus, it is a combination of attributes that determine their value as a food. From a consumers' perspective, the color or appearance is one of the most important attributes among those mentioned earlier. If the color of a fresh-cut fruit/vegetable is not attractive or of acceptable



quality, the consumer is less likely to purchase it regardless of its excellent texture, flavor, taste, or other quality attributes. Therefore, in addition to acceptable physico-chemical properties, a high level of sensory quality is a prerequisite for successful marketing of fresh-cut fruits and vegetables.

**a. Color and Texture**

Color or appearance and texture are two of the most important characteristics that help determine the acceptability of fresh-cut fruits and vegetables. Understanding the process by which changes occur in these characteristics can help to establish ways in which to control or reduce problems. This in turn can help to increase the shelf-life of products both in the store and at the consumer's home (Toivonen, 2008). Fresh fruits and vegetables are often sliced or cut in some method. This can induce much of the surface degradation. Finding ways to reduce the degradation, while maintaining shelf life, is an important task in the fresh-cut industry.

Color is one of the most important features that consumers use to judge a product before buying it. Browning causes discoloration and spots thus presenting the product as old. Any sort of wounded product also looks undesirable to the consumer. A loss of green pigmentation is also another quality that is observed when products are too ripe or not ripe enough. Freshness is associated with the vibrant green color, anything dull or turning yellow conveys a decaying product to the buyer (Lamikanra, 2002).

Textural quality factors include firmness, crispiness, juiciness, and toughness. These depend on the specific product and can vary from one product to another. The texture is affected during processing, and in shipping of the product. It is also affected by storage temperature, controlled/modified atmosphere and by ethylene accumulation. With soft fruits, long shipping times can cause great physical damage to the product. Tissue softening is also a concern since juices can leak from the cells of the fresh-cuts. Dong et al (2000) found that the firmness retention in “Bosc” and “Bartlett” fresh-cut pears was higher in under-ripe fruits. In another study it was found that there was no relationship between the oxygen concentration and firmness of fresh cut “Bartlett” pears (Gorny et al, 2002). Tissue wounding has been shown to induce a high respiration rate, which triggers faster texture deterioration compared to tissue that is intact (Rosen and Kader, 1989). Calcium in solution is commonly used to maintain the firmness of fresh-cuts. Calcium dips are often combined with chemicals such as ascorbate or cysteine to help prevent browning. The dipping treatment itself rinses enzymes and solutes from injured cells at the cut surface (Toivonen, 2008). Many studies have been done to look at the texture and firmness of fresh-cuts, in one study it was found that fruits treated with calcium are generally firmer than control fruits (Poovaiah, 1986).

Cut edge browning is a problem particularly in white flesh fruits such as apples and pears. This is due to the interaction of Polyphenol oxidase (PPO) and oxygen (Sapers, 1993). Research on the cellular structure of plant cells helps practitioners to understand the biochemical mechanisms involved in

browning of fresh cuts. The initial event in the oxidative browning process is the breakdown of membranes within cells of plant tissues. These findings have been found elsewhere as well (Toivonen, 2004). Once the physical stress or deteriorative process has begun, the compartmentalization of the cell begins to fail (Marangoni et al., 1996). Once this breakdown starts to occur, it results in the mixing of polyphenol substrates with PPO or phenol peroxidases, which causes the browning effect (Degl'Innocenti et al, 2005).

The yellowing or loss of green color that occurs in plants is due to chlorophyll degradation (Brown et al, 1991). However, this same process can also cause the color changes similar to browning in fresh-cuts which have been treated with some salad dressings. A low pH dressing has been shown to inactivate the chlorophyll degrading enzymes (Heaton and Marangoni, 1996). A related study showed that "white blush" in carrots could be controlled with treatments which altered the pH, which in turn controlled the enzyme activity (Bolin and Huxsoll, 1991; Bolin, 1992). "White blush" is a surface discoloration that minimally processed cut and peeled carrots undergo. When the carrots experience cutting or processing, the cells at the surface are damaged and exposed to the environment. When surface drying then occurs, it is called "white blush" (Tatsumi et al, 1991). Onions, garlic and leeks can develop a pink, red, green, blue-green, or blue discoloration from cell disruption (Josyln and Peterson, 1960). Depending on the severity of the cutting or handling operation, there can be severe tissue damage which induces such color changes (Howard et al, 1994).

More recently, secondary browning has been found to be a limiting factor in fresh-cut apples (Toivonen, 2006). It is different than cut-edge browning in that it is more localized unlike cut-edge which is diffuse in nature. In cut-edge browning, the process starts within a few hours of cutting, but secondary browning starts to appear in fresh-cut fruits between 1 to 3 weeks of storage. The exact time varies depending on the storage and handling of the product (Toivonen and Delaquis, 2006).

Texture can be categorized in various ways such as crispiness, hardness, mealiness, flouriness, grittiness, chewiness, stickiness, etc. The two most important factors that affect the mouth feel for consumers are firmness and juiciness. Produce with small cells have firmer tissue and thus are less juicy. The cells of ripening fruit flesh tend to be fairly weak in comparison to vegetables. The cell walls of fruits also go through a natural degradation process during fruit ripening which reduces the cell wall firmness; however, this is not generally seen in vegetables (Toivonen, 2008). Vegetables tend to have more cells with thicker secondary walls, and are therefore firmer in nature and not as susceptible to softening. However, in products such as spinach, wilting is an issue which causes loss in both the texture and visual appearance (Piagentini et al, 2002). Factors which affect the texture can change greatly during pre or post-harvest storage due to changes in the cell size, intercellular adhesion, starch/sugar conversion, water loss, cell wall composition and cell wall strength (Toivonen, 2008). Fresh-cut products are wounded tissues; therefore, they deteriorate more rapidly and physiologically are much different than whole fruits

and vegetables. Since preparation requires chopping, shredding, cutting, slicing, coring, and dicing through the cells, the cell contents are released at the site of wounding (Toivonen, 2008).

Other factors that affect the texture are water loss and osmotic changes. Water loss leads to a loss in both crispiness and turgor. It is most common in fresh-cut products since there is a lack of a cuticle and sub-epidermal layer, which causes exposure to internal tissues. However, appropriate packaging can be used to slow down the water loss (Toivonen, 2008). In fresh-cut pears it was found that firmness retention was much greater in an atmosphere of 100% nitrogen instead of oxygen. Oxidative damage can cause a reduction in the membrane integrity, resulting in cellular leakage and the flooding of intercellular spaces (Soliva-Fortuny et al, 2002). Other physical processes can cause a loss of product quality, in addition to chemical and microbiological mechanisms.

**b. pH, Total Soluble Solids, Titratable Acidity, Headspace Gas, and Microbiology**

Numerous tests can be used in order to look at the overall quality of fresh-cuts. Monitoring the pH over time can help to see whether there is a possible decrease in the quality of products especially fresh-cut fruits and vegetables. Lamikanra (2002) stated that lowering the pH of foods within the range of 3.0-5.0, can help in restricting the growth of many types of micro-organisms. Thus, it can help to reduce the risk of spoilage or growth of organisms. If the pH changes during the storage study, this may indicate that the product is becoming more or less susceptible to microbial growth. An acidic ingredient is used on the produce

to help lower it. In this study, SAS was one of the treatments used to help lower the pH. For some fruits the pH may already be low enough to reduce the risk of spoilage, however, in some cases the pH may increase during storage. King et al (1991) noted that the pH of lettuce increased during storage and, therefore, bacterial populations had also grown. A decrease in pH was found for shredded carrots, because lactic acid bacteria were present (Kakiomenou et al, 1996). Fresh-cuts can also have a dressing such as mayonnaise or sour cream added to them in order to lower the pH. By combining hurdles such as low temperature and low pH, bacterial growth can be controlled or reduced.

Total soluble solids are another test, which can be used to help determine the quality of fruit juices. Generally, the higher the total solids, the better the quality of the juice. The ratio of solids in relation to acidity is also an important factor that affects the flavor (El-Samahy et al, 2008). Soluble solids have been found to vary in amount depending on what area of the fruit they are in. For example, Archbold and Barter (1934) found that the soluble solids content was higher in the blushed versus unblushed side of apples. Also, the SS concentration was higher from the inside to the outside. Another study found that the soluble solids in watermelon increased from the rind to the core, with the greatest concentration near the seeds (Tucker, 1934). Generally, as the ripening of fruits occurs, an increase in soluble solids occurs.

Titrateable acidity or total acidity, as it is sometimes called, is used to find out how much acidity of a particular acid is present. These can include tartaric, malic or citric acid. Most often the process is used for producing optimal quality

wine (Titratable Acidity, 2008). In apples, malic acid is most predominantly found and its level decreases as the fruit ripens. The organic acids are what give fruits more of an acidic taste. It is important to monitor the level of acidity over time, as was done in this study to see if the acid component of the flavor of the product changes over time.

Headspace gas analysis is used to monitor the composition of accumulated gases in the headspace of specific package. The headspace gas is mostly made up of oxygen and carbon dioxide which results from the respiration of the product and diffusion through the package wall. However, it can also contain ethylene gas and other volatile components. Fresh produce has different respiration rates. For Anjou pears the respiration rate is 3-6 ml CO<sub>2</sub> /kg hr at 41°F. This respiration value is lower than for Bosc and Comice pear varieties, which have higher respiration rates (Mitcham, 2009). The optimum gas concentrations for pears are 1-2% oxygen and 0-1% carbon dioxide. If the carbon dioxide levels are too high, or the oxygen levels are too low the pears will not be able to respire properly which will cause browning, off flavors, and overall quality deterioration. It is very important to monitor the amount of oxygen and carbon dioxide in a package due to their effect on the fresh product. Overall, monitoring the headspace gas during storage can help to discern what the shelf life will be. It can also help to evaluate the overall quality of products while determining if the package system is durable and efficient in its design.

Microbiological testing can be used to determine the overall growth of bacteria, yeast and mold. In this study, the microbiological stability of fresh-cut

pears was determined by counting bacteria, yeast and molds. For this testing it is crucial to have as sanitary of an environment as possible in order to avoid contamination from outside environments. It is important to monitor the microbiological activity at frequent intervals to make sure the product is safe and edible throughout its shelf life. Soliva-Fortuny (2003), found that modified atmospheres inhibited the growth of yeast and molds, whereas bacteria grew rapidly on the fresh-cut Conference (English winter variety) pears. The availability of oxygen in the headspace of the bags, even in the smallest amount, was enough to start bacterial growth while the moderate level of carbon dioxide present did not prevent this growth. The same study concluded that low oxygen atmospheres could improve the microbial stability of fresh-cut pears which preserved them from nutritional losses. Ensuring a safe product free of microbes is very important in the fresh cut fruits and vegetable industry. The more the microbial presence is controlled the higher the possibility of extending the product shelf life.

**c. Sensory attributes (9 point Hedonic scale)**

Sensory testing can provide various types of information including whether or not a product might be successful in the market. It can also help to determine what consumer's perceptions are regarding a certain product. Lack of consistent quality is one of the factors that influence consumers from repeating purchases. Often the flavor may change before a visual change has occurred. This needs to be prevented in order to not mislead customers. For this reason the flavor and texture are two attributes that need to be studied carefully, in addition to being



monitored throughout the products shelf life. Even though the process can be expensive, it can help with quality assurance and quality control.

Lamikanra (2002) reported that flavors of each and every fruit and vegetable are very different and unique; for example, an apple and an orange both have quite different flavors. The different tastes that consumers can sense are sweet, sour, bitter, astringent, caramelized, honey, chemical, rotten, estery, fermented, floral, woody, musty, earthy, raw, fruity, citrus, artificial, and many more. Some of these are off flavors and are associated with poor quality of a product.

Generally, vegetables are not labeled as sweet, except for vegetables such as carrots and sweet potatoes, which have low intensity sweetness. Flavor intensity is the strength that a particular flavor has within a certain food. Different intensity scales are used to measure the response, such as universal scales, product specific scales or attribute specific scales. In addition to flavor, texture is also widely studied (Lamikanra, 2002).

Texture is the structure and orientation of the components in a food and the reaction of the food to any type of force applied to it. Each of the attributes can be defined and measured. Texture can be divided into four different categories: surface properties, first bite properties, chew down and after swallowing properties. Surface moisture (wetness) and roughness are surface properties. Springiness, cohesiveness, denseness, hardness, moisture release, juiciness, crispiness, and uniformity of bite are all based on the very first bite taken. The chewiness and cohesiveness of mass are determined during the

chew down phase, and mouth coating is evaluated after swallowing (Lamikanra, 2002).

The intensity scales are also unique for each food; the most important categories include crispiness, hardness, and juiciness. Crispiness is important because it indicates the degree of turgor pressure inside of the cells, or how much the product has wilted. Juiciness or moisture release can indicate the amount of dehydration that has occurred in a product. Hardness shows whether or not senescence has occurred which indicates if a product has grown old.

In sensory evaluation, there are various test categories which can be used depending on the type of information a researcher needs. The categories include: affective tests, descriptive tests, and difference/discrimination tests (Harte, 2009). Affective tests include: preference; where the subject is forced to say which product they prefer or an acceptance test, which measures the liking or eating behavior of a product. In a preference test, subjects compare one product against another and only one sample is selected in the end. Often, after an affective test, an additional question is asked about the preference of the tester for the samples. Overall, affective tests assess the personal response of consumers to a product, product idea, or product characteristic. Descriptive tests are usually the most sophisticated of sensory tests. These describe the perceived sensory characteristic in order of their occurrence. This test helps in determining actual differences found between samples, and provides results which help explain the data collected in other sensory tests (such as acceptance, preference or difference). Discrimination tests also known as difference tests.

These are used to determine whether a difference exists between samples. In this type of test a trained panel is usually used (Harte, 2009). Discrimination tests include: simple difference, triangle and duo-trio. These help test how attributes (texture, flavor, aroma, etc.) differ. Descriptive tests not only look at the difference, but also the intensity of it. In a typical test consumers are asked if the samples are the same or different.

The better the understanding of a consumers' perception of a product the more likely a product has of being successful. When consumers are asked to study product attributes, various scales can be used. These include: a 9-point hedonic scale, a 5 or 7 point scale, a "just right" scale or a "smiley scale". A hedonic scale means "having to do with pleasure." A hedonic scale is not used for trained panelists since they are familiar with different levels of good and bad within each attribute. Hedonic scales are usually used in a 75-100 consumer panel with untrained subjects (Harte, 2009). This is cost and time effective. With number scales the lower numbers usually correspond to "like extremely" whereas the higher numbers correspond to "dislike extremely." A 9-point scale is the most commonly used scale. In cases where there is a "just right" scale subjects are asked whether the strength or weakness of an attribute is too much, too little or just about right. The smiley scale is one that is used most often with children. The scale is set up with smiley faces ranging from a sad face to a neutral face to a happy or really happy face. Similarly, pictures or photos can correspond to a certain numerical value of like or dislike. Overall, the scales help to interpret the

consumer's perception, whether it is good or bad (Harte, 2009). Without a proper scale, results would be obscure and nonsensical.

#### **d. Shelf Life considerations**

Various factors play a role in the shelf life of products. These include processing methods and preservation techniques, packaging, and the environment. For consumers shelf life is one of the most important aspects since consumers want products to be the same over a significant time period. The industry is very devoted to researching new and improved methods for improving quality and shelf life (Lamikanra, 2002). Ultimately longer shelf life increases the consumers' chances of purchasing a product since it saves them trips to a grocery store.

Most salads and vegetables have a 12-14 day refrigerated shelf life. Fruits on the other hand have a shorter shelf life of around 8-10 days (Lamikanra, 2002). Efforts to preserve and prolong product shelf life are an area of importance to the fresh-cut industry, which is on-going.

#### **e. Packaging**

One of the most important ways to preserve food is through good packaging. Without it, the product, no matter how it was treated before hand, will not be able to withstand physical, chemical, or biological degradative forces. Therefore, it is vital to choose the appropriate material for a package in order to provide the highest level of protection to a product at the most reasonable cost. The role of packaging is to provide the following: containment of the product,

protection, convenience/utility (ease of use) and communication of how to use the product and keep it safe. These are the major aspects which should be used in designing any package (Lockhart, 1997).

If a consumer were to walk down any fresh-cut aisle in a store they would see one main feature among all the packages; it is the ability to see the product. The visual aspect of a product is the first attribute that interacts with consumers and the first attribute used in the purchase decision. A clear container or film provides consumers the ability to see a product and many base their decision as to or not buy from that initial glance.

There are hundreds of films that can be used for packaging, all of which have their own oxygen transmission rate, and each will affect how a sliced or cut product “breaths” throughout distribution and storage. If the film oxygen transmission rate is too high for a certain product, then it will age and brown. However, if the transmission rate is too low, then the product will go into anaerobic respiration and decay. Since fresh-cuts continue to respire in the display cabinet, the permeability of a film as well as the amount of oxygen that diffuses into a package is vital to know as it affects its shelf life. Modified atmosphere packaging (MAP) can be used to balance the oxygen and carbon dioxide in a package for a particular fresh produce. It allows products to respire slowly and stay fresher longer even in long storage conditions. .

MAP retards senescence, lowers the respiration rate, and slows down the rate of tissue softening. It has been shown that low oxygen and high carbon dioxide storage conditions are helpful in extending the shelf life of foods. MAP

alters the gasses around a product, creating an atmosphere that is different from air. This is, however, different from controlled atmosphere storage, where a fixed gas composition is created. MAP can be used to create a pre-determined composition of gas in a package, specific to a certain product. The gas levels are not only determined by the product and its physiological characteristics, but also by the environment and the barrier properties of the packaging material (Lamikanra, 2002). Out of the various methods for MAP, a passive MAP is commonly used for respiring products. In the case of this study, an atmosphere high in CO<sub>2</sub> and low in O<sub>2</sub> passively evolved within the sealed package over time due to the respiration of the product. In combination with the permeability of the film, the optimal headspace gas content is formed which helps to increase the shelf life of the pears (Robertson, 2006).

When selecting a film to use for MAP it is important to look at the gas permeability rates, the water vapor transmission rates, mechanical properties, sealing properties, and the design of the package. The Wiley Encyclopedia of Packaging Technology (1997) states that “most products are packed in laminated or co-extruded copolymers of LDPE (low density polyethylene), PS (polystyrene), and PP (polypropylene).” Other additives and comonomers are added to facilitate sealing, printing and to add antifog properties (Brody, 1997). Co-extruding technology has also increased the number of material variations available. Blends of different polymers like linear low density (LLD) and medium density (MD) polyethylene with ethylene vinyl acetate (EVA) are also quite useful for MAP packaging applications. Polyethylene (PE) has good shrink and is a

good moisture barrier. EVA on the other hand provides better sealability and has higher oxygen permeability than PE (Brody, 1997).

Fresh-cuts are produced in store and via product manufacturers. For fruits, one way is for retailers to assemble/cut the products in store, and then package them in trays, using a variety of closing techniques. Fruits can be sold in halves or quarters, and mixed with other varieties of fresh fruits. A popular method is to use a clamshell or plastic tub, and then fill with fruit chunks or diced product (Lamikanra, 2002). Recently, much research has been done with polylactic acid (PLA) which is biodegradable and useful for fresh-cut packaging in different container styles.

Cut carrots, sliced mushrooms, cut onions, cut celery, etc are often packaged similarly. They are packaged in trays individually or with mixtures of other vegetables. They can also be packed in clamshells and plastic tubs. Tubs of dips may be included to provide a value added package. The correct packaging of fresh cuts is critical to pro-long shelf-life and provide added value.

#### **IV. Food Safety Concerns related to Fresh-cuts**

##### **a. Outbreaks**

One of the biggest concerns with processing produce into fresh-cut fruits and vegetables is the increased risk of contamination of spoilage bacteria and pathogens. This increased risk can occur due to the destruction of the natural protective outer layer during the initial slicing, dicing, peeling, mashing, trimming, and coring of the particular fruit or vegetable. There are many variables involved

in the growth of pathogens in fresh-cuts. It is often difficult to run an actual experiment where all risk factors can be addressed for contamination (CFSAN, 2007). Numerous pathogens have been linked to outbreaks in fresh-cuts. Many of these cases and reports have been filed through the Center for Disease Control (CDC).

Over the past 30 years or so, the reporting of outbreaks has increased due to the better surveillance as well as the increase in consumer awareness. More and more people are eating fresh-cuts and so there is a greater risk that pathogenic outbreaks will occur. With the increase in popularity of salad bars there is also an increase in potential for illness. Summertime seems to be one of the most prevalent times due to inadequate temperature control, and the higher consumption (Lamikanra, 2002).

A great deal of research is being done in order to figure out better ways to prevent pathogenic contamination. Governmental agencies, such as the United States Department of Agriculture (USDA), and the Food and Drug Administration (FDA), as well as other public health agencies are helping consumers, by providing informative brochures and online material related to fresh-cut products and safety.

#### **b. HACCP**

HACCP stands for Hazard Analysis Critical Control Point and it is very important in ensuring the safeness of food products. The potential for risk can take place during cultivation, harvesting, and cutting and slicing operations.



Many different operations are involved in the supply chain necessary to produce fresh-cuts. Each can have multiple control points, making it difficult to fully monitor all sources of contamination. However, by implementing a good HAACP program as well as Good Agricultural Practices (GAPs), Standard Operating Procedures (SOPs), Good Manufacturing Practices (GMPs), and Sanitation Standard Operating Procedures (SSOPs) it is much easier and better to prevent any contamination and illnesses than to deal with the consequences of food poisoning.

HAACP consists of seven steps. Step one involves conducting a hazard analysis and creating a flow diagram of the steps in a process to determine where certain hazards can exist and what control measures should be used. Step two determines the critical control points that need to be established in order to control hazards. In step three the critical limits (CLs) are established, by developing the specifications (target values and/or tolerance) that have to be met in order to make sure that the critical control points (CCPs) are working. Step four makes sure that there are set procedures in place to monitor the CCPs. These can be used to adjust the process in order to make sure there is proper control of the CCPs. Step five establishes corrective actions when the monitoring system indicates any sort of deviation from the critical limit. Step six establishes a verification procedure for determining if the HACCP system is working properly. Finally, step seven specifies the establishment of an up to date and descriptive record of the procedures used in the HACCP system by documenting them.

In the fresh-cut industry HACCP plans incorporate many different preservative techniques to control growth of pathogens. HACCP can result in a cost effective system to maximize food safety by focusing on hazard control at the beginning. It establishes a systematic way to control potential risks in production and handling of foods, starting with raw materials to consumer preparation. It also helps consumers become confident in what they are buying.

### **c. Impact on Food Industry**

With sales of over \$10 billion in U.S. retail and food service the fresh-cut produce industry has a significant economic importance as stated by the International Fresh-Cut Produce Association (IFPA) in 2000. Each year the consumption of fresh produce grows more and more. With the added convenience offered by fresh-cuts it is all the more reason for consumers to desire it. The IFPA has been very helpful in providing food safety information to people around the world. They have published guidelines for food safety, designed different HACCP models for fresh-cut operations and promoted the industry (Herndon, 2006). The fresh-cut industry is very sensitive to pathogens and spoilage and its image can be ruined if proper care is not taken to produce a good, safe product. Safety is one of the biggest consumer concerns and therefore needs to be given adequate consideration.

If consumers are not confident that they are buying and eating a safe product they will be much less likely to purchase it. Therefore, products need to be as safe as possible. Implementing a proper HACCP plan for products and

using the hurdle concept can really decrease the chances of creating a dangerous product (Lamikanra, 2002). The food industry relies heavily on the good will consumer. If this good will is altered in any negative way then sales can drop and the industry could find itself fighting to survive. Therefore, it is really important to make sure consumers are being provided with a safe and healthy product.

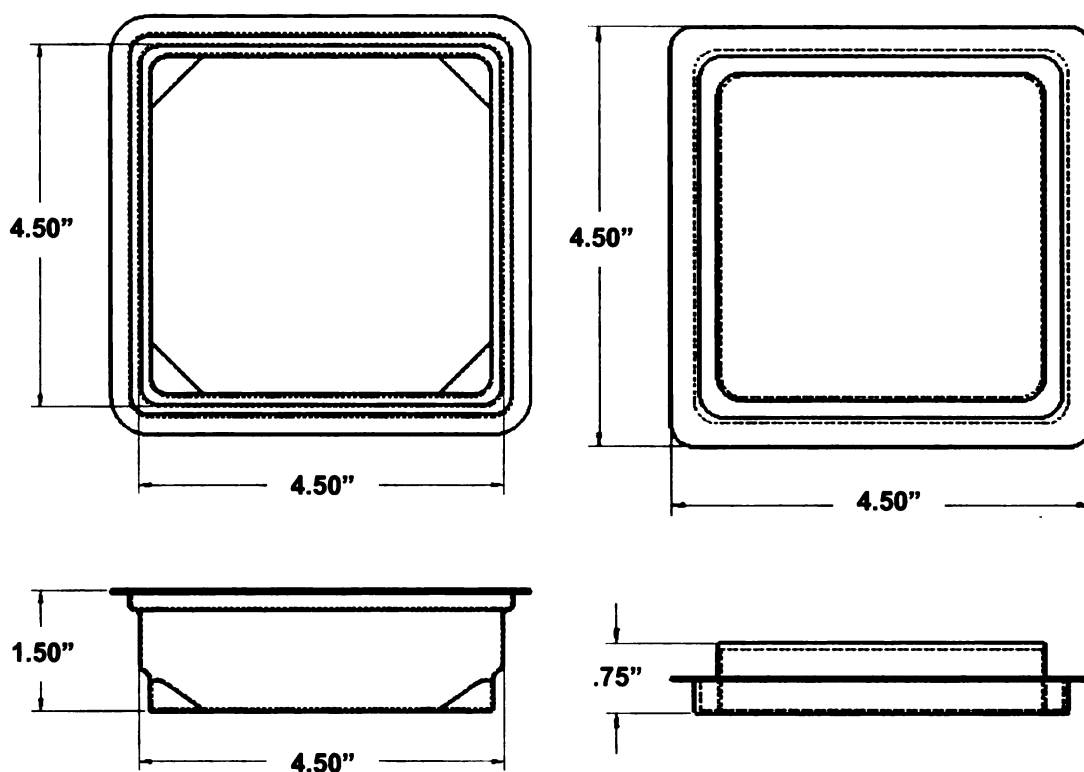
Factors that influence consumers to choose fresh-cuts include convenience and their exceptional health benefits. Their “on the go convenience” and ability to “travel” are very beneficial for people who are short on time and need something quick. Many people are concerned with becoming ill from fresh-cuts; however, with the increase in research being done to package, process and preserve fresh-cuts, it is becoming easier to keep consumers safe. By combining different methods of processing and preservation as well as discovering new and more productive ways to package fresh-cuts, the industry will continue to be prosperous. Learning how to maintain the optimal product quality throughout distribution, keeping it safe and marketing it as a healthy alternative will keep consumers happy and the industry growing.

## MATERIALS AND METHODS

### Materials

Commercial package containers were obtained for use in this study, and the Pears were purchased from a local source.

### I. Packaging/Product



**Figure 1: PET Package and Lid Diagram (Top and Side View)**

- **Packaging:** 8 oz. square deli flange containers were used for the entirety of the study. The containers were made of Polyethylene Terephthalate (PET) and its dimensions were 4.5" x 4.5" x 1.5" (Figure 1). The containers with the roll film seal had an O<sub>2</sub> permeability of .13 cc/[pkg-day], CO<sub>2</sub> permeability of .57 cc/[pkg-

day], and WVTR of .34 gm /[pkg-day]. Approximately 340 containers were used in the entire study. Snap lock lids were used on half of the containers, the other half were heat sealed with PET roll film by the Multi-Vac machine (discussed under MAP). Lid dimensions were 4.5" x 4.5" x .5". To seal the roll film the Multi-Vac machine was set at 95°C with a sealing time of 2 seconds to provide an optimal heat seal on the containers. Once packages were placed in the molds on the Multi-vac they were manually pushed into the machine cavity. The motorized film feed positioned the film on top of the container and using the proper amount of heat and pressure the film was sealed onto the container. The water cooled top section of the Multi-Vac helped to set the seal. Then, the integrated knife in the machine cut around the perimeter of the tray. Following this the tray evacuation system pushed the tray upward and the machine rewound film remnants automatically. Both lids and containers were ordered from Clear Lam Packaging, Inc. located in Elk Grove Village, Illinois. Item numbers were as follows for both: Lids- SDOFLID (thickness of ~13 microns for the top of the lid and ~18 microns for the side of the lid) and Containers- SD8F (thickness of ~17 microns for the bottom of the container and ~ 14 microns for the sides of the containers).

- **Product:** D'Anjou pears were purchased from Horrock's Farm Market located in Lansing, Michigan. Pears are usually grown in

Washington and shipped in refrigerated trucks to either the Detroit or Grand Rapids produce warehouse. Here they are stored in coolers set at 40-45°F at which point a produce buyer from Horrock's comes weekly to purchase a the pears. Pears are shipped daily to the store and not kept in storage (40°F) for longer than 1 week. Coolers in Horrock's are set at 42°F, however, from the point pears are picked to the point they are purchased by consumer and in between, pears can go through a temperature range of 34°F-54°F. There are various growers for the pears; however, when a case of pears is bought it is guaranteed to be from the same grower/producer. D'Anjou pears are available for approximately year round and so they were selected over other varieties of pears, also due to the limited amount of literature on this variety. The required number of pears was purchased by the case within 24 hours of each processing day maintain quality and consistency. The pears were picked up the night before processing, and packaging and were kept in refrigerated storage until used (40°F). A total of 4 cases (60 pears per case) were purchased to do the research. Pears were medium sized, approximately 166 grams each, and only pears with no brown spots or bruising were used for the study.

## II. Treatments

- a. **Sodium Acid Sulfate (SAS)** also known as pHase® was provided to the MSU Food Science and Human Nutrition department by Jones-Hamilton Co. (Walbridge, Ohio) for use on fresh-cut produce. SAS is a “natural” food acid with the unique ability to lower the pH on the surface of products without generating a sour taste. A 2% wt./wt. basis concentration (73.2g) was used to dissolve the SAS into a bucket containing 1 gallon of distilled water. This concentration was chosen as per the opinion of Dr. Muhammad Siddiq, Ph.D. Associate Professor in the Food Science and Human Nutrition department and expert in Fruit and Vegetable Processing. Half of the pears were dipped into this solution. SAS was used only during the preliminary studies. Due to its inability to physically preserve the sliced pears the SAS treatment was not continued during the remainder of the project. This will be explained further within the Results and Discussion section.
- b. **Nature Seal (NS)** was also used in this study as a treatment to extend product shelf life. It was provided by the Mantrose-Haeuser Co., Inc (Westport, Connecticut) and has been used extensively in treatment of fresh-cut produce. A 2% wt./wt. basis concentration (73.2g) was dissolved into a bucket with 1 gallon of distilled water. Again due to the expertise of Dr. Muhammad Siddiq. The other half of the samples was dipped into the treatment solution. NS is a

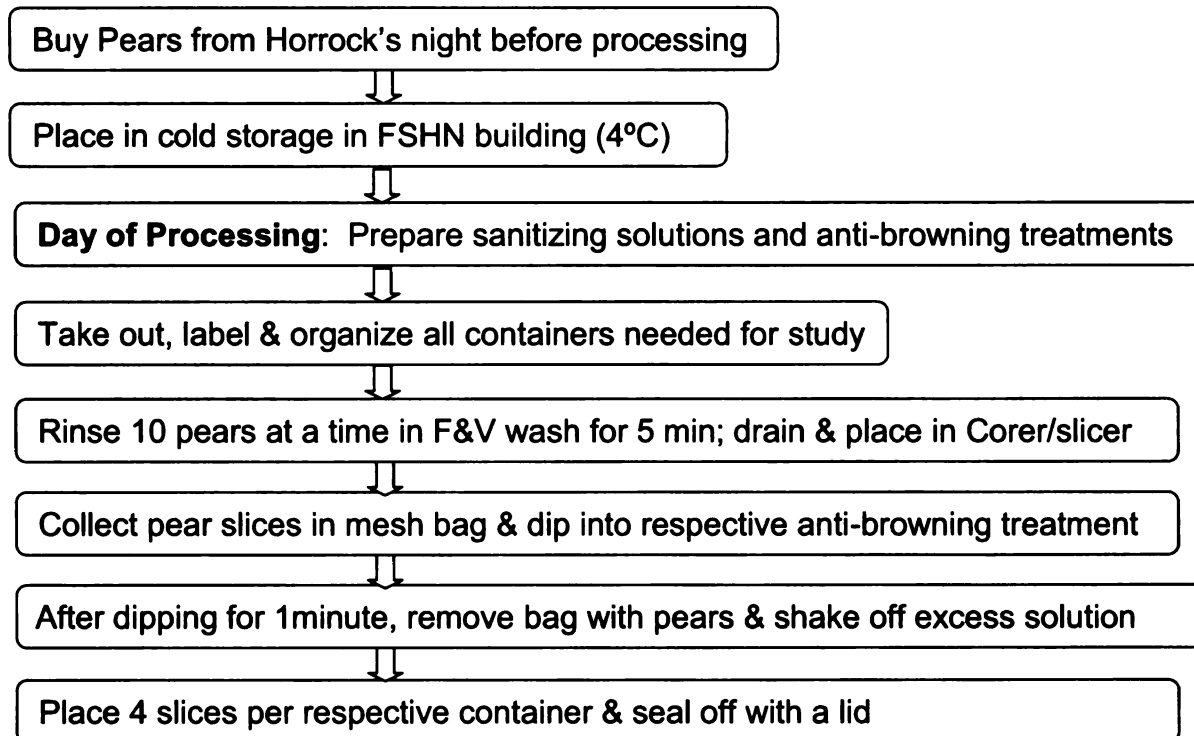
patented blend of GRAS (Generally Regarded As Safe) vitamins and minerals which inhibit discoloration (browning), while maintaining the natural taste, texture and color of the product. NS was used both in the preliminary study as well as the storage studies. NS is also often referred to as Calcium Ascorbate.

- c. **Fruit and Vegetable Wash-Cleaner and Sanitizer** (SC Johnson or Johnson Wax Professional/ JP Optimum CRS) was used to remove any soil, bacteria, yeasts or molds from the surface of the pears before processing. It consists of 4% Sodium Hypochlorite and 96% inert ingredients. To make the solution, 30 g of powder were mixed into 2 gallons of water to give a concentration of 150 ppm. The whole pears were dipped for a period of 5 minutes prior to any cutting and then excess solution was removed by lifting up the inner bucket of pears, which had holes drilled in the bottom designed for drainage. The wash was used during both the preliminary and storage study.

### **III. Processing**

For both the preliminary and storage studies, the processing of the pears was done in the Pilot Plant, located in the Food Science and Human Nutrition (FSHN) Building on the Michigan State Campus in East Lansing, MI. The following is a flow process of the procurement of the pears to the packaging in final containers.





**Figure 2: Flow Diagram for Pear Processing to Packaging**

The procedure used to process the pears during both the preliminary and actual study was quite similar. Prior to any processing gloves and lab coats were put on by each of the assistants. The amount of pears needed was purchased to ensure that enough samples were prepared for all necessary testing over the course of three weeks (21 days). Pears were purchased the night before from Horrock's Farm Market. At Horrock's they were kept in a cooler room set at 40-50°F (4-10°C), and then brought to the Food Science building where they were kept in a walk in cooler set at ~40°F (~4°C). They were purchased the night before to ensure maintenance of quality. Next, 73.2 grams of both the NS and SAS powder were weighed out on a balance and poured into zip lock bags. Each bag was then poured into separate 7.5 quart buckets which contained 1

gallon of distilled water. The buckets were labeled with the words “NS or SAS” to avoid putting samples in the wrong solution. The buckets were pre-rinsed with distilled water prior to the addition of either SAS or NS. Each bucket containing water and powder was stirred for a sufficient period of time using spatulas to fully assure dissolution of the powders. In addition, a 10 quart bucket was rinsed with distilled water and then filled with 2 gallons of distilled water. Thirty (30) grams of the Fruit and Vegetable wash powder was weighed, poured and mixed into the water in the bucket. A second 10 quart bucket with 10-1 cm holes drilled into the bottom was used for holding the pears. This bucket having a different profile than the other 10 qt buckets, allowed the pears to be dipped into the bucket containing the Fruit and Vegetable wash. The bucket with the holes allowed the wash to be drained from the pears w/out having to physically dip hands into the bucket, potentially causing contamination.

A total of ten (10) pears were rinsed with the Fruit and Vegetable wash at a time for a total of 5 minutes. Using a stainless steel knife that had been sanitized in 1 gallon of distilled water containing 1 cup of sanitizing solution (Clorox ® bleach), the top inch or so of each pear was cut to remove the stem and also to provide slices of even length. Of the ten pears, five were cut in the corer/slicer machine (Bock Engineered Products Inc., Toledo, OH.) at a time. Two mesh produce bags were combined (one inside of the other) to catch slices as they exited the machine. As soon as slices were caught in the mesh bags they were taken to the NS solution bucket and dipped for a period of 1 minute. Samples were agitated and stirred within the solution to provide proper coating of

all slices. After evenly coating slices with the NS solution, the mesh bag was taken out and physically shook in a repetitive manner for a period of 30 seconds to remove any excess solution. Samples were then taken and deposited into a clean aluminum tray. One assistant, stationed in a clean room was in charge of putting 4 slices of pears in a container and placing a lid on it. Containers were labeled as either: NS + MAP, NS no MAP, SAS + MAP, or SAS no MAP (the four treatments used in this study). The same amount of containers was set out for each treatment with a lid for each container. Once all NS no MAP containers were filled and packaged they were put into aluminum trays that were labeled with the treatment name and immediately taken to the walk in cooler. Once all NS + MAP containers were filled and packaged they were also put in properly labeled aluminum trays and then directly into the cooler.

Following the processing of the NS samples, the solutions were dumped and the buckets were rinsed and set aside for future use. For all SAS pear samples the same procedure was used (Figure 2): whole pears were each dipped for 5 minutes in the fruit and vegetable wash, excess moisture was removed, pears were then sliced 5 at a time and collected in mesh bags, and then dipped into the SAS solution for 1 minute, and then the remaining moisture was removed and samples were placed in a tray to be packaged. Once all SAS samples were packaged and put into the appropriately labeled trays they were also taken to the cooler. At this point, the half of the samples that required MAP packaging were immediately taken across the street to the Packaging Building. The time the MAP samples spent outside the cooler was kept as minimal as

possible. To prevent oxidation and browning, temporary lids (the same lids used for non-MAP samples) were placed on the MAP samples, while they were collected and ready to be packaged using the Multi-Vac. They were also kept in the cooler to slow down any chance of enzymatic browning.

#### **IV. Modified Atmosphere Packaging (MAP)**

The following materials and method were used in this study: Multi-Vac Traysealer T200, Medical Grade Air Tank, Clear Lam Lidding Film–PET with EVA coating ( $O_2$  permeability of  $59.30 \text{ cc}/[\text{m}^2\text{-day}]$  and WVTR of  $9.14 \text{ gm}/[\text{m}^2\text{-day}]$  ) and Clear Lam Packages. Before the start of the actual pear processing the Multi-Vac T200 was turned on and allowed to warm up to the desired sealing temperature of  $95^\circ\text{C}$ . In order to use the Multi-Vac the following steps needed to be completed:

Step 1: Pull the MAP tray holder out. This is where containers were placed for packaging.

Step 2: Turn red switch clockwise to start up machine

Step 3: Turn green knob counter-clockwise as well as valve marked with x to allow for water to go into the machine.

Step 4: Turn blue knob counter-clockwise and turn gas tank knob counter-clockwise as well to allow for gas and pressure to be flowing.

Step 5: Turn on machine settings by pressing power on the panel

Step 6: Press P6 or desired parameter which has sealing time and temperature already programmed into it.

At the completion of these steps the machine was given the proper amount of time required to heat up to the sealing temperature, in order to ensure strong seals. Containers were packaged four at a time and it took approximately 30 seconds to complete each set. During the sealing process, air is first removed out, and then purified (Medical grade) air is flushed into the package from a tank attached to the machine. Finally, the packages are heat sealed to the lidding film. A roll of polymeric film (PET coated with a thin layer of Ethylene Vinyl Acetate (EVA)) was loaded into the film feeding device. Adding the EVA coating to the PET film allowed for better heat seals with less wrinkling of the film. This ensured a tight seal between the package and film, preventing an exchange of gases through the sealed area. After sealing all MAP samples, the temporary lids were discarded or saved if in good condition. All MAP samples were then quickly taken back to the cooler where the non-MAP samples were being held. All of the samples remained in cold storage until the test day, at which point a specific number of packages of each treatment variable were taken from storage and evaluated using a series of test procedures. On days 0, 7, 14, and 21, two packages of each treatment were evaluated, using the following tests: headspace gas content, color, pH, Total Soluble Solids (TSS), and Titratable Acidity (TA), in that order. The microbiological tests were conducted on days 0, 7, 14 and 21, three packages of each treatment were taken from storage. For days 3, 10, and 17 only the headspace and color were evaluated using 2 packages from each treatment. At approximately three day intervals, photos were also taken of all samples to document any physical/color changes.

During the afternoon of the day the pears were processed, 2 packages of each treatment were removed from storage to obtain the initial day 0 data. A paper copy of the data was maintained along with Excel files.

The same procedure was used to process the pears for the second study. The main difference between the preliminary and secondary storage study was that the Sodium Acid Sulfate solution was eliminated since it did not yield pears having positive physical/color characteristics. The same tests were also conducted on samples at the same time intervals using the same number of packages.

## **V. Analytical tests used to evaluate the pears**

### **a. Headspace Gas**

The gas headspace analysis of the packaged pears was conducted using the following materials and method: an Illinois Instrument 6600 Oxygen/Carbon Dioxide Headspace Analyzer, Moisture Particle Filter (Part PFL-305143), Sampling needles (Part PSM-201000) and Septa (Part PPL-193456).

The percent oxygen and carbon dioxide in the headspace of each container was determined using the Illinois Instrument 6600 O<sub>2</sub>/CO<sub>2</sub> Headspace Analyzer (Johnsburg, Illinois). Prior to using the instrument, it was to be calibrated for both O<sub>2</sub> and CO<sub>2</sub> using the instructions provided by the manufacturer. The carbon dioxide level was calibrated to 0% and the oxygen level was calibrated to 20.9%. Sampling needles (to penetrate the package) and septum were purchased from the manufacturer. A septum was attached to each

package to help keep the package (where the needle was inserted) intact to only allow the package atmosphere to be drawn out through the needle. This also prevented any internal atmosphere to escape through holes in the film that may have occurred if the needle had been directly inserted through the film. Without the septum the headspace gas readings would likely have been inaccurate. A particle filter was also attached to the needle to prevent any large particles or samples from plugging the needle. The same particle filter was used repeatedly, unless it became blocked. The same needle was also used on each testing day, unless the needle touched the pear sample. If so, it was discarded and a new one was used.

The machine was allowed to warm up for a period of one hour before evaluation of the packages. Once ready, the needle was pushed through the septa and through the package 2 cm into the package headspace, making sure not to touch the actual pear sample. The analysis was begun and testing was conducted for a period of 25 seconds. During this time, atmosphere was sucked out of the package and the amount of oxygen and carbon dioxide in the headspace was determined. The measurements were taken on all four treatments as well as all four replicate samples. Testing was conducted on Day 0, 3, 7, 10, 14, 17 and 21. To prevent the package from imploding the time of testing was adjusted. This time was found by testing multiple empty but sealed containers.

**b. Photos**

Numerous photos were taken during both the preliminary and storage study using a Canon Powershot SD 750. Pictures were taken on Days 0, 3, 7, 10, 14, 17, and 21 and were taken under bright lights to properly display the color differences. Images were taken during the preliminary and final storage study showing each treatment individually as well as comparing the different samples over time.

**c. Color ( $L^*$ ,  $a^*$ , and  $b^*$ )**

For color tests the following materials and method were used: A LabScan XE made by Hunter Lab, Easy Match QC program on Computer, 17 mm diameter measurement port, glass cup for holding sample, Kimtech Wipes, Distilled Water and a 3" diameter black sample holder cover.

Color measurements were taken every three days using the LabScanXE colorimeter (Day 0, 3, 7, 10, 14, 17, and 21). Prior to taking measurements, the machine was calibrated using both a black and white tile. The same packages that were used for the Headspace Gas analysis were used in measuring the color of the pears. The EasyMatchQC program was used to pre-calibrate and to read the samples. After calibrating the machine, a pear slice was taken from the first treatment and cut in half in order to fit it into the sample glass cup. The cup was placed on the 17mm measurement port and the sample was placed into the glass cup. A black cup larger than the glass cup was used as a lid to avoid interference in the readings from outside light or objects. A reading was taken



from both sides of the pear and then the sample was discarded. Additional readings, one from each side, were taken from a second slice from the same container. This resulted in four measurements per container. The same process was used on the second package, thus giving a total of eight readings per treatment. The same process was followed for the remaining three treatments as well as for all four replicate treatments. The glass cup was rinsed between each reading with distilled water and wiped dry with KimTech Wipes. The color “L”, “a”, and “b” values were taken for each sample and exported to an excel file.

#### **d. Juice Extraction and pH**

The following materials were used for the juice extraction and pH testing: Black and Decker JE 2100 Fruit and Vegetable Juice Extractor, 20ml plastic vials for sample collection, distilled water, Omega pHB-212 Microprocessor pH Meter (pH/mV/Temperature bench top), pH buffer solutions of pH 4, 7 and 10, and Kimtech wipes.

The remaining pear samples (from the headspace and color test packages) were blended in a Black and Decker juicer. Approximately 20ml of juice was extracted from the samples and poured into plastic vials which were labeled according to treatment. The blender was rinsed with distilled water between treatments to avoid any contamination. The pH meter was pre-calibrated using pH buffer solutions of 4, 7 and 10 (Stamford, Connecticut). Following the calibration, each treatment as well as the replicates were tested a total of 4 times. After each reading, the testing probe was rinsed with distilled

water and wiped off carefully with a KimTech Wipe. The pH was determined on Days 0, 7, 14 and 21.

**e. Total Soluble Solids (TSS)**

The following materials and procedure was used for the TSS: A Pal 1-Atago Pocket Refractometer, Distilled water, 15.5 cm Disposable plastic pipettes and Kimtech wipes.

The percent soluble solids were calculated using the Pal 1-Atago Pocket Refractometer. Results were recorded in units of °Brix for all treatments as well as replicates. The same juice that was extracted from samples and used for measurement of pH was used to determine TSS. The refractometer was first calibrated using distilled water to make sure it was zeroed. Then using a 15 mm disposable pipette, two to three drops of each treatment were put into the refractometer and a reading was taken. Measurements were taken twice for each treatment and all replicates. Between each treatment the refractometer was cleaned with distilled water and wiped dry with a KimTech wipe. All readings were recorded along with the pH values. The TSS was determined on Days 0, 7, 14 and 21.

**f. Titratable Acidity**

For the titratable acidity the pH meter used earlier in the project was used again with the following materials: 0.1N NaOH solution and 0.1 N HCl solution (prepared by the researcher), a Phenolphthalein indicator, 10ml juice samples (stored in 20 mL plastic vials), 2 gallons of distilled water, a 250 mL beaker, 10

mL cotton plugged glass pipette, 50mL pipette filter (rubber bulb), a 50 mL burette with a stand, 6 plastic cups for waste, 500 gram bottle of Sodium Hydroxide (NaOH) pellets, 500 ml bottle of Hydrochloric Acid (HCl) solution, Reynolds Aluminum Foil, 1000 ml Beaker, 1000ml Erlenmeyer flask, and 9" Stainless steel spatula.

The following method was used to test the TA. Three solutions were made: A 0.1% Normal solution of NaOH using 4 g NaOH pellets and 1000 mL distilled water; A 0.1% Normal HCl solution using 8.26mL HCl and 1000mL of distilled water (both solutions set aside for 30 minutes prior to use); and 1% phenolphthalein indicator using 1g of phenolphthalein and 100mL of ethanol. The NaOH solution was made weekly and the HCl was made fresh every 2 weeks. After preparing the solutions the next step was to standardize in order to determine the normality of the NaOH and HCl solution (further explained in Appendix II).

Using the juice samples from the pH and TSS tests the following procedure was used to conclude how much NaOH it took to neutralize the pH of the juice samples to a pH of 8: (1) 10 mL of juice sample was combined with 100 mL of distilled water in a 200 mL beaker. (2) Calibrate pH meter using pH buffer 4, 7, and 10. Once calibrated take pH wand and dip into the beaker and mix, making sure not to touch sides of beaker. While stirring the mixture, carefully release the NaOH drop by drop into the beaker, making sure not to stop stirring. (3) As each drop is added, carefully monitor the pH and stop adding NaOH once the beaker solution has been neutralized to pH 8. Note the difference in volume

from the beginning of the NaOH addition to the end of its addition; this was done for all treatments to find the amount of NaOH used to neutralize each sample. These numbers were then used to calculate the total acidity for each sample. This test was conducted on Days 0, 7, 14 and 21.

**g. Microbiological**

For the microbiological study the following materials were used: petri dishes, whirl pack bags, 5.5" square weigh boat containers, balance, TSA-YE- Trypticase Soy Agar, Difco™ PDA- Potato Dextrose Agar, PBS- Phosphate Buffer Solution, 70 ppm Ampicillin, yeast extract, distilled water, 4 - 1000ml Erlenmeyer Flasks, a 100ml Graduated Cylinder, aluminum Foil, autoclave, incubator, water bath, walk in cooler, Bunsen Burner and lighter, test tube rack, pipette 100 µl, pipette 1000 µl, pipette tips for both sized pipettes, hazardous waste disposal container, L-Spreaders (plastic), vortex test tube agitator and a plate Spinner.

Microbiological testing was done to determine total count of bacteria, yeast and mold on the pears. TSA-YE (Trypticase Soy Agar) and PDA (Potato Dextrose Agar) media were prepared and poured into petri-dishes prior to each testing day (Day 0, 7, 14, and 21). An in depth procedure is provided in Appendix III. After preparing plates the next step was to plate the samples. Three packages of each treatment were taken out of storage at each of the testing points. Next, 25 grams of sample were taken and added to 100 mL of Phosphate buffer solution (PBS) in a whirl-pack bag, labeled properly and

agitated by hand for 60 seconds. This was done for all samples. All materials/devices were made sure to be properly sanitized beforehand between each sample.

The next part was to take petri dishes out and properly label 2 plates for each dilution (0, -1, -2, and -3) for both the PDA and TSA-YE plates. A total of 48 plates were used for each treatment on each test day. To plate samples, 1000  $\mu\text{L}$  of the solution from each whirl pack bag was added to 9000  $\mu\text{L}$  of PBS in a test tube. This test tube was mixed and labeled as dilution -1. Autoclaved pipette tips were used and discarded between dilutions. Next, 1000  $\mu\text{L}$  of dilution -1 were pipetted out and put into the second test tube which became dilution -2. This was repeated until dilution -3 was reached. Each solution was mixed by a Vortex test tube agitator before being diluted further. The original whirl pack bags with the sample and buffer solution were labeled as Dilution 0. After all dilution test tubes were prepared, they were ready to be plated. A 100  $\mu\text{L}$  pipette was taken and 100  $\mu\text{L}$  of each dilution was placed on to its corresponding plate. Using an L-spreader the solution was spread on to the agar by simultaneously spinning the plate spinner and moving the L-spreader back and forth. Pipette tips were discarded in between each dilution. Dilutions were plated two plates at a time, from 0, -1, -2, and -3 to avoid contamination. Each whirl pack bag was discarded at the end.

After plating, petri dishes were allowed to sit and dry for 15 minutes after which all TSA-YE plates were put on a tray and into the incubator set at 29°C. The plates were left for 24 hours and examined the next day. All PDA plates

were set on a tray at room temperature conditions and checked 2-3 days later. The number of colonies formed was counted on each plate for each dilution and the numbers were recorded. Then, plates were discarded. On each testing day, 0, 7, 14 and 21 the same process was followed for plating, labeling, and counting colonies.

#### **h. Sensory**

For the sensory analysis the following materials were used: SIMS Questionnaire, consent forms, flyers (advertisement), incentives (ice cream cups), 100 dixie cups, 2 Gallons of Distilled Water, 200 Paper Plates, 100 napkins, 100 spit cups, and 100 white 8.5" x 11" trays with printer paper on each.

A consumer panel was used to evaluate the quality and acceptability of the pear samples. A total of 100 participants were recruited and a questionnaire was developed using the SIMS program. The demographic for the study included males and females between the ages of 18-60. Since the study was conducted on campus, a majority of consumers were either students or faculty of Michigan State University. Consumers were asked to observe and eat some of each sample and then fill out the questionnaire. The following attributes were evaluated by the sensory panel: appearance, aroma, flavor, texture and overall acceptability using a 9 point hedonic scale ranging from 1, Dislike Extremely, to 9, Like Extremely. Follow up questions were also asked about both the packages presented and product (Questionnaire in Appendix IV). Samples were prepared 10 days prior to the testing. Control samples were prepared 12 hours

before the test in order to present fresh samples. The control was packaged in the same containers but with pop off lids instead of the MAP peel OFF lids. Thus, these samples were nearly as fresh, similar to what a consumer would prepare for themselves. Seven sets of the following packages were also prepared: A snack sized zip lock bag with 4 pear slices, a MAP package with 4 pear slices, and a PET container with the pop OFF lid and 4 pear slices (NS without MAP sample). These 3 packages were placed on a tray and set at each booth for consumers to look at and touch. Consumers were asked to not open any of the packages to prevent rapid browning.

At the completion of the test all results were recorded in the SIMS computer system and then extracted and exported to excel. The Consent form and advertisement can be found in Appendix IV.

**i. Statistical Analysis**

Statistical Analysis was done using two-way or three-way factorial analysis of variance (ANOVA) model (Proc Mixed, SAS 9.2). The models of treatments, storage time, and package type, as well as interactions of all 3 were used for all analytical and microbiological tests. The data was treated for multiple comparisons by analysis of variance with least significant difference (LSD) between means determined at the 5% level. For the two way ANOVA model, the following equation was used:  $y_{ij} = \mu + MAP_i + Time_j + MAP_i * Time_j + e_{ij}$ ,  $i = 1, 2, j = 1, 2, \dots$  where  $y_{ij}$  is the response variable,  $\mu$  is the overall mean,  $MAP_i$  is the fixed effect of package type,  $Time_j$  is the fixed effect of storage time,  $MAP_i * Time_j$

is the interaction effect between package type and storage time, and  $e_{ij}$  is the residual term. For the three way ANOVA model, the following equation was used:  $y_{ijk} = \mu + Trt_i + MAP_j + Time_k + Trt_i * MAP_j + Trt_i * Time_k + MAP_j * Time_k + Trt_i * MAP_j * Time_k + e_{ijk}$ ,  $i = 1, 2, j = 1, 2, k = 1, 2, \dots$  where  $y_{ijk}$  is the response variable,  $\mu$  is the overall mean,  $Trt_i$  is the fixed effect of treatments,  $MAP_j$  is the fixed effect of package type,  $Time_k$  is the fixed effect of storage time,  $Trt_i * MAP_j$ ,  $MAP_j * Time_k$ ,  $Trt_i * Time_k$  and  $Trt_i * MAP_j * Time_k$  are the two-way and three-way interaction effects between these three factors, and  $e_{ijk}$  is the residual term.

#### **j. Permeability (O<sub>2</sub>, CO<sub>2</sub>, and WVTR)**

In this study the PET containers oxygen, carbon dioxide and water vapor permeability were tested in addition to the PET (with EVA coating) films oxygen and water vapor permeability. For both the film and container each test was run until a steady state was reached. The average Oxygen Transmission Rate (OTR), Water Vapor Transmission Rate (WVTR), and Carbon Dioxide Transmission Rate (CO<sub>2</sub>TR) was then calculated for the data obtained for each sample using replicate cells. The averages of Cell A and Cell B were then calculated to give the final value. In depth results for all samples are shown in Appendix V.

#### **The following procedure was used for the film:**

The OTR was measured using an Ox-Tran Model 2/21 Oxygen Permeability (Mocon, Inc.) in accordance with ASTM 3985-05 (standard method



for oxygen gas transmission rate through plastic film and sheeting using a coulometric sensor). A mixture of nitrogen and hydrogen was used as a carrier gas at a flow rate of 10cc/min and test conditions were set at 23°C, 0% RH, and 100% oxygen. A flow rate of 20 cc/min was used for the test gas. Samples were conditioned in the instrument for 2 hours at 23°C, 0% RH, and 100% oxygen before data collection began.

The WVTR was measured using a Permatran-W Model 3/31 (Mocon, Inc.) in accordance with ASTM F 1249-06 (Water Vapor Transmission Rate Through Plastic Film and Sheeting Using a Modulated Infrared Sensor). Nitrogen was used as a carrier gas at a flow rate of 100 cc/min, and test conditions were 23°C and 100% RH. A test time of 30 minutes was used for both cell A and cell B. Samples were conditioned in the instrument for 1 hour at 23°C and 100% RH before data collection began. Sponges soaked in water were used to generate the relative humidity for the test.

**For the sealed container (lidding film attached) the following methods were used to determine the OTR, WVTR, and CO<sub>2</sub>TR:**

The OTR was measured using an Ox-Tran Model 2/21 Permeability (Mocon, Inc.), tested in accordance with ASTM 3985-05 (same as film). A mixture of nitrogen and hydrogen were used as carrier gases through copper tubes connected from the machine to the containers at a flow rate of 10 cc/min, and test conditions were 23°C, 0% RH, and 100% oxygen. Plastic tubes were run from the machine to the inside of the Ziploc bag providing the oxygen gas. A flow rate of 20 cc/min was used for the test gas. Samples were conditioned in a

Ziploc bag for 4 hours at 23°C, 0% RH, and 100% oxygen before data collection began.

The CO<sub>2</sub>TR was measured using a Permatran-C Model 4/41 (Mocon, Inc.), in accordance with ASTM F 2476 - 05, Standard test Method for the Determination of Carbon Dioxide Gas Transmission Rate (CO<sub>2</sub>TR) Through Barrier Materials Using an Infrared Detector. Nitrogen was used as a carrier gas through copper tubes connected from the machine to the containers at a flow rate of 100 cc/min, and test conditions were 23°C and 0% RH. Plastic tubing was run from the machine into the Ziploc bags providing the carbon dioxide gas. A test time of 45 minutes was used for both cell A and cell B. Samples were conditioned in a Ziploc bag for at least 4 hours at 23°C and 0% RH before data collection began.

The WVTR was measured using a Permatran-W Model 3/31 (Mocon, Inc.), in accordance with ASTM F 1249-06, Water Vapor Transmission Rate Through Plastic Film and Sheeting Using a Modulated Infrared Sensor. Nitrogen was used as a carrier gas through copper tubes connected from the machine to the containers at a flow rate of 100 cc/min, and test conditions were 23°C and 100% RH. A test time of 45 minutes was used for both cell A and cell B. Samples were conditioned in a Ziploc bag for 4 hour at 23°C and 100% RH before data collection began. Sponges soaked in water were put in the Ziploc to generate the relative humidity in the bags.

## **RESULTS AND DISCUSSION**

This section presents the experimental results from the various tests used to evaluate the fresh-cut pears. The work includes two studies, a preliminary storage study and a final storage study with results divided accordingly into these two sections. The four treatments used in the preliminary study were: T1-NS no MAP, T2- NS + MAP, T3- SAS no MAP, and T4-SAS + MAP. Two sets of each study were completed for all samples. The data points from the replicate study were combined with the first set of results. The anti-browning treatments NS and SAS refer to Nature Seal and Sodium Acid Sulfate, respectively. The average values for each test can be found in tables in Appendix VI.

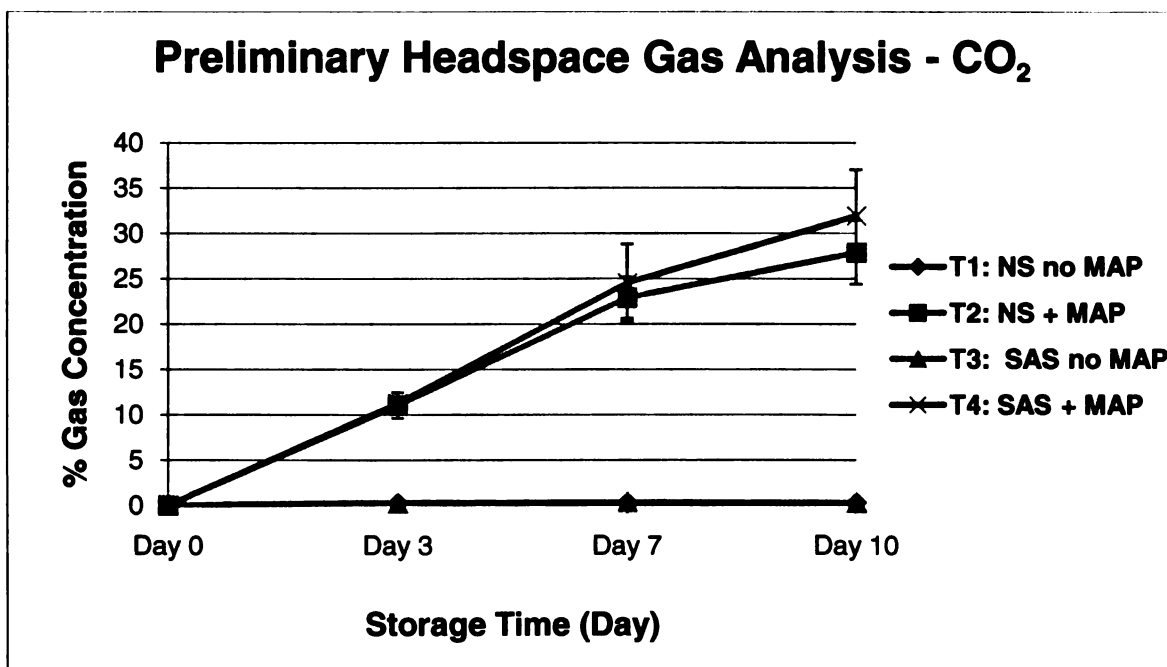
### **I. Preliminary Results**

#### **a. Headspace Gas Analysis**

The concentrations of oxygen and carbon dioxide in the headspace of the packages were studied over time. The initial CO<sub>2</sub> concentration was assumed to be 0% and the initial concentration for O<sub>2</sub> was assumed to be 20% on Day 0. In order to increase the shelf life of fresh fruit it is important to increase the carbon dioxide levels and reduce the oxygen levels. This helps to slow down the natural respiration rate of the fruit, thereby increasing its shelf life. Since fruits continue to respire after harvesting, it is very important to monitor the oxygen level. If there is no oxygen, the pears will go into anaerobic respiration, which will increase senescence and decay since pears cannot aerobically respire without oxygen. Kader and Saltveit (2003) stated that to preserve fresh-cut fruits,

oxygen levels should be <5 %, with elevated carbon dioxide levels above 3%. Using packaging, the levels of oxygen and carbon dioxide can be controlled.

Two measurements were taken from two separate containers for each treatment on each testing day, after which the samples were discarded. Figure 3 shows the levels of CO<sub>2</sub> over time for each of the four treatments including the replicate (second set) study.



**Figure 3: Headspace CO<sub>2</sub> Concentration of Pears over Time at 4°C**

In Figure 3, it can be seen that samples T2 and T4, both MAP, had carbon dioxide levels that steadily increased over time. Samples T1 and T3, however, did not show an increase in the carbon dioxide levels. This may have been due to the fact that there was not a tight fitting seal on these packages; therefore gases were able to flow back and forth between the package headspace and

external environment, thus limiting the amount of gas accumulation inside the package.

The following table shows the p-values for the carbon dioxide levels over the 10-day storage time based on the different factors.

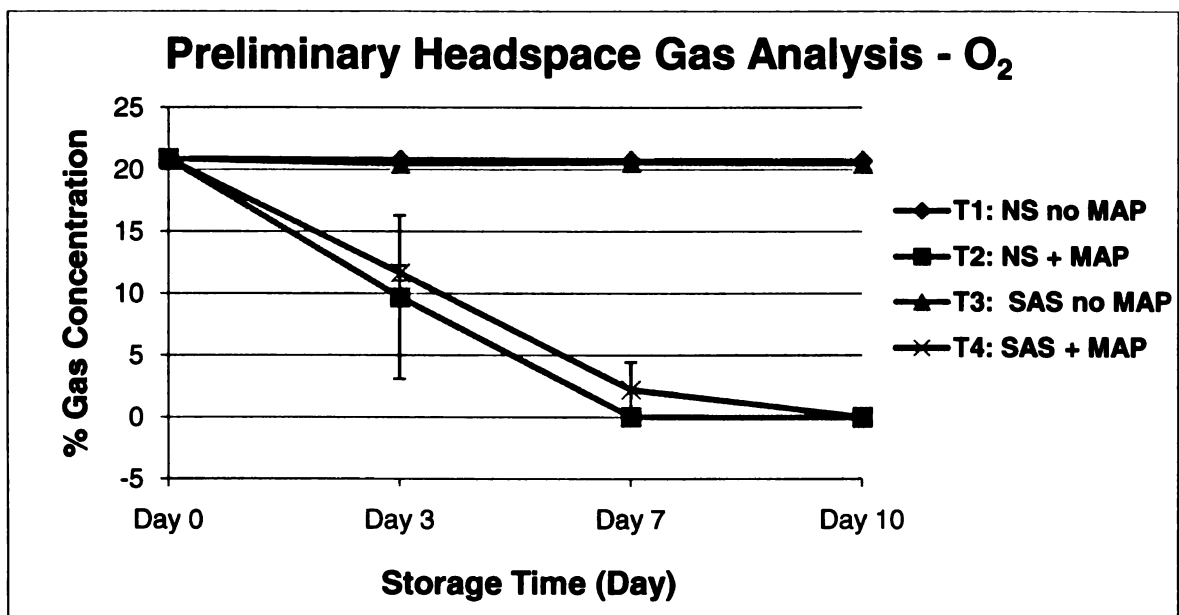
**Table 1: p-values for CO<sub>2</sub> level as affected by the various factors**

<b>Effect</b>	<b>P-Value</b>
Treatment	.5381
MAP	<.001
Treatment * MAP	.7651
Time	<.0001
Treatment * Time	.7262
MAP * Time	.0011
Treatment * MAP * Time	.8333

*p<.05 is significantly different*

Table 1 shows that there was no significant difference in the carbon dioxide level in the packages over the 10-day period between the individual treatments of NS and SAS (p-value .5381). This was also true for the interaction between Treatment \* MAP (p-value .7651), the interaction between Treatment \* Time (p-value .7262) and the overall interaction between treatment \* MAP \* Time (p-value .8333). There WAS a significant difference in the carbon dioxide levels when a package did and did not contain a modified atmosphere (p-value <.0001). The effect of time (p-value < .0001), as well as the interaction of the effects of MAP\*Time (p-value .0011) did result in significant differences. A significant difference was also observed between days 3 and 7 as well as day 3 and 10 (p-value <.0001 for both). However, between days 7 and 10 there was no significant change or difference in the level of carbon dioxide (p-value .5546).

The oxygen levels for all four treatments were also monitored over the 10 day period. Two readings were taken from two separate containers. As a container was tested and results recorded, it was discarded. In Figure 4, the oxygen concentration is shown over the 10 day period for all four treatments.



**Figure 4: Headspace O<sub>2</sub> Concentration of Pears over Time at 4°C**

In Figure 4, it can be seen that for samples T1 and T3 no change occurred in oxygen levels over time. This may have been due to the lack of a tight seal between the package and the lid (NO MAP), thus allowing a continuous transfer of oxygen. Samples T2 and T4, however, showed a decrease in their level of oxygen. As the level of oxygen decreases, respiration will slow, leading to an increased shelf life. However, as the oxygen concentration nears zero, the pears will go into anaerobic respiration, which will then cause the pears to rot. Sample T2 showed a decline in oxygen level between Days 0 and 7, after which point the concentration remained constant. Sample T4 also declined; however, it

continued to decline throughout the storage time, arriving at the same concentration as T2 by day 10. Both MAP pear samples had oxygen values which declined due to the pears continuing to respire. This means that the pears were taking in large amounts of oxygen and using it to aerobically respire while releasing carbon dioxide. This is why we see higher levels of CO<sub>2</sub> and lower levels of O<sub>2</sub>.

The following table (Table 2) shows the p-values for oxygen level over the 10-day storage time based on the different factors that were tested. A statistical evaluation of the four treatments was done for individual treatments and for the interaction of the various factors. This helped to identify the specific factors which had a greater effect on the samples and which did not make a significant difference in quality.

Each treatment in Table 2, individually and collectively, caused a significant difference in the oxygen levels over the 10 day period. All p-values were below the .05 level.

**Table 2: p-values for O<sub>2</sub> level as affected by the various factors**

<b>Effect</b>	<b>P-Value</b>
Treatment	.0036
MAP	<.0001
Treatment * MAP	.0032
Time	<.0001
Treatment * Time	.0116
MAP * Time	<.0001
Treatment * MAP * Time	.0119

*p<.05 is significantly different*

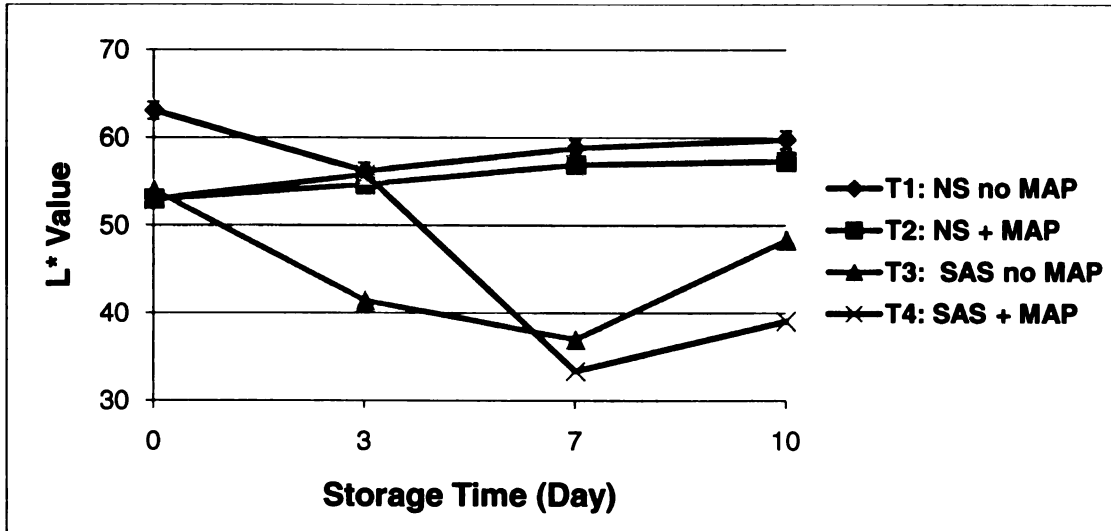
Overall, both MAP and Time, caused the levels of oxygen and carbon dioxide to change significantly in the sample packages. The interaction of MAP and Time also resulted in a significant difference in the concentration levels of both oxygen and carbon dioxide. Thus, MAP was beneficial in achieving the packaging environment necessary to extend the shelf life. The treatments of NS and SAS did not cause a significant difference in the level of carbon dioxide; however, they did cause a significant difference in oxygen concentration.

**b. Color L\*, a\*, and b\* Analysis**

The color coordinates, L, a, and b were determined on the pear samples using a LabScan XE made by Hunter Labs, Inc. Using the Easy Match QC program, the values of L, a, and b were read for each pear sample (using the color scale) for each treatment and recorded in the computer program. The coordinates L\*, a\* and b\* represent the following according to the Hunter Lab scale published by Hunter Labs, Inc.; “the L\* axis represents lightness and darkness and runs from top to bottom, the maximum value for L\* is 100 pertaining to white or a very light colored sample, whereas the minimum value for L\* is 0 pertaining to a black or very dark sample. Both a\* and b\* scales do not have specific numerical limits, +a\* values refer to red, -a\* values refer to green, +b\* values refer to yellow and -b\* values refer to blue.” The pear samples were tested at Day 0, 3, 7 and 10. A total of four readings from each sample were taken for each of the four treatments (T1, T2, T3 and T4). A replicate set was also evaluated following the same procedure, with the same number of readings taken for samples T1, T2, T3 and T4.



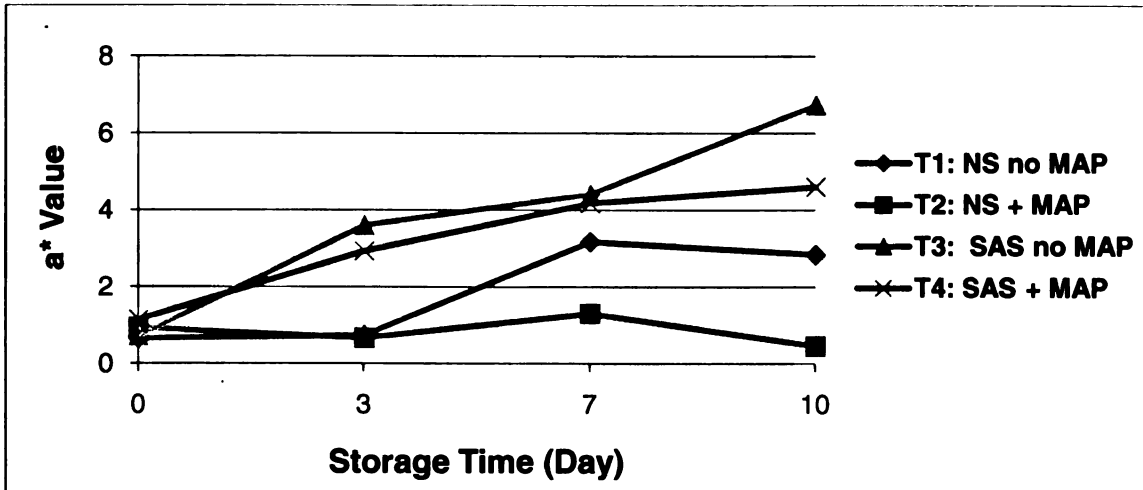
By analyzing the average values of the L coordinate for both sets (set 1 and 2), the change in color was found using the color scale. Figure 5 shows the changes in the average L\* values over time for all samples.



**Figure 5: L\* color of Pears over Time for the treated samples**

For the L\* values from Figure 5 the following can be stated; Sample T1: NS non MAP became slightly darker in color over the 10 day period since L\* values decreased (despite some variation), sample T2: NS + MAP became slightly lighter in color, and samples T3: SAS non MAP and T4: SAS + MAP both became darker in color, however, there was some variation between Day 0 and Day 3. Even though there was an overall change in the pears from the initial values to the final values, some variation occurred over time for each sample. Overall, the results showed that the NS + MAP was the only sample that consistently increased in lightness. This did not occur in any of the other samples.

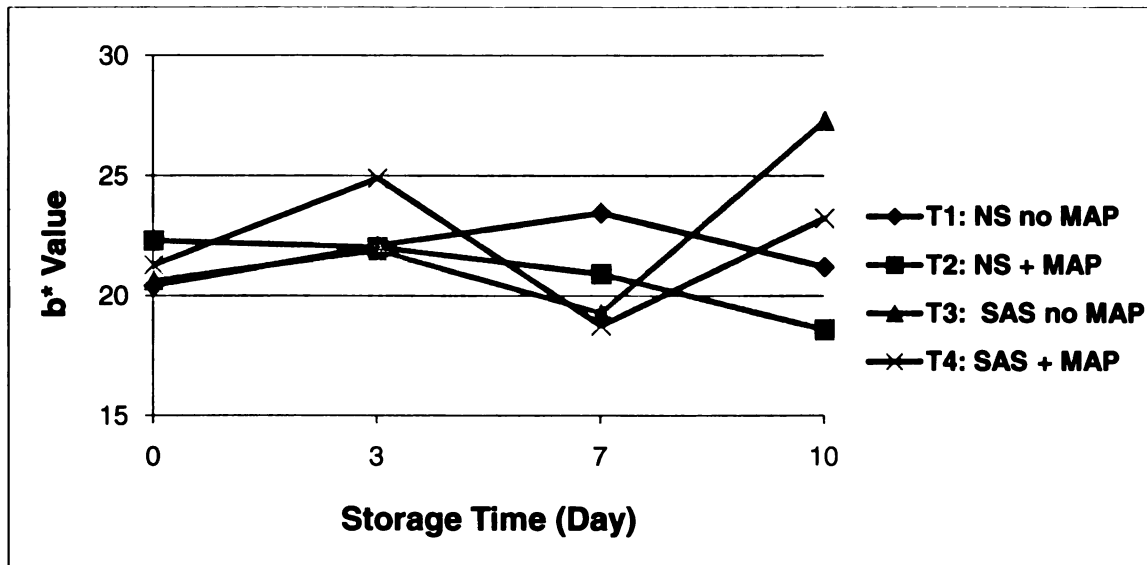
The second color component was the  $a^*$  value, and the results are depicted in Figure 6.



**Figure 6:  $a^*$  color of Pears over Time for the treated samples**

Samples T1 showed an overall increase from the initial value to the final value (Figure 6), meaning that the pears tended towards a more red color which indicates a higher level of browning. However, there was some variation between Days 0, 3 and 7. Sample T2 (NS + MAP) had an overall decrease in the initial value to the final value, although it was small. Samples T3 and T4 (SAS samples), show a steady increase in  $a^*$  values meaning they were trending towards more to a reddish color (increased browning). Overall, it was found that sample T2: NS + MAP maintained the most consistent  $a^*$  value, and closer to a more green color. The SAS samples were a more reddish color and more discoloration occurred in these samples.

The results for the average  $b^*$  color values are shown in Figure 7:



**Figure 7: b\* color of Pears over Time for the treated samples**

From Figure 7, it can be seen that for sample T1 (NS no MAP) the pears had an overall slight increase in the b\* value which means that they were moving towards a more yellow color, however, there was some variation between the values from Day 3, 7 and 10. In sample T2 (NS + MAP), the values decreased slightly (more blue) throughout the study. In sample T3 (SAS no MAP), there was an overall increase from the initial value to the final value which indicates that the sample was tending more to a yellow color, however, there were some significant variations between Days 3 and 7. Sample T4 (SAS + MAP) showed a slight overall increase in value which meant it was tending towards a more yellow color even though there were some big variations between Day 3 and 7. Overall, the NS non MAP sample was the most consistent from the initial to final test date. Table 3 shows which factors caused a significant difference in the L, a, and b values.

**Table 3: p-values for L, a, and b values as impacted by the various factors**

Effect	p-values		
	L*	a*	b*
Treatment	.2703	<.0001	.5052
MAP	.5624	.0053	.6603
Treatment * MAP	.2210	.5192	.7951
Time	.8648	<.0001	.4944
Treatment * Time	.8691	.0002	.0876
MAP * Time	.4206	.0189	.4079
Treatment * MAP * Time	.5551	.6189	.8741

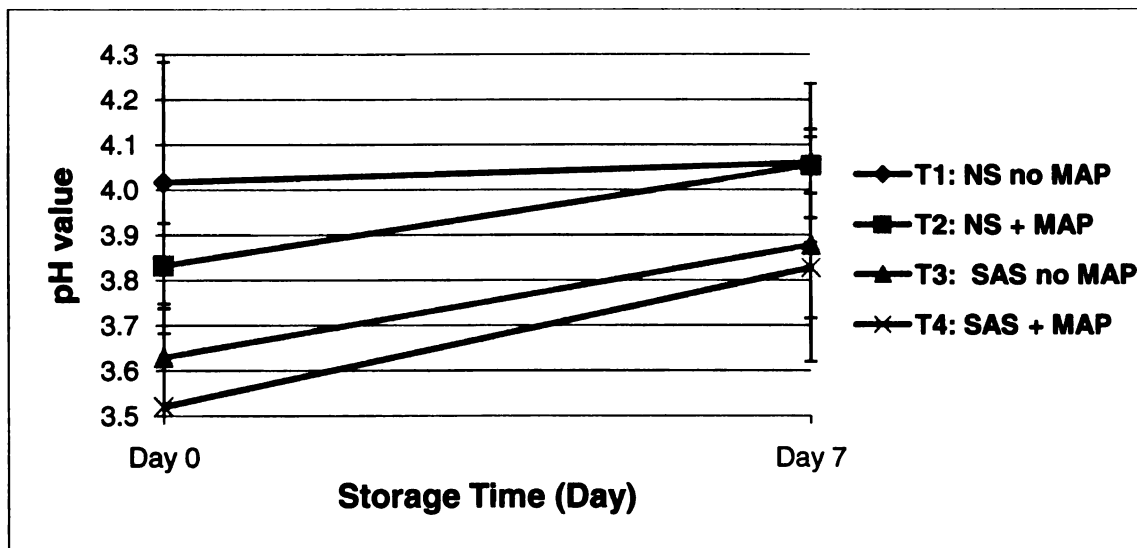
*p<.05 is significantly different*

Table 3 shows that none of the treatment factors caused a significant difference in the L\* value. For the a\* values the treatment type, MAP, Time, interaction of Treatment \* Time, and interaction of MAP \* Time caused significant differences. The interaction of Treatment and MAP, as well as the interaction between Treatment \* MAP \* Time did not cause a significant difference in the a\* values of the pears. For the b\* values none of the factors caused a significant difference in the b scale.

Overall; the use of NS or SAS did not significantly affect the L\* or b\* values, using MAP compared to non MAP did not significantly affect the L\* or b\* values, and Time did not cause significant difference in either values. The color a\* value showed the most significant differences due to the treatment factors studied.

### c. pH Analysis

The pH measurements were taken at weekly intervals; however, due to the visual quality of the SAS pear samples, the analysis was not continued after the first week. The pH measurements help to indicate a change in acidity of the pears. A change in pH can also be an indicator of product decay. Understanding the change in acidity can help to determine how different the taste of the pear sample will be. Since D'Anjou pears have a slightly acidic flavor to begin with, it is important to monitor how much that acidity changes, thus indicating how much the flavor will also be affected. In order to do pH testing, pear samples were juiced using a juicer into 20 ml plastic vials. Two readings were taken from each sample on each testing day. Figure 8 shows the change in the pH over time.



**Figure 8: pH values of Pears over Time for the treated samples**

In Figure 8 it can be seen that none of the samples had a significant change in pH over the 7 day period. All pear samples steadily increased in value, while T1 remained the most constant over time. Each sample, regardless of treatment or MAP, started out acidic and did not become significantly more acidic or basic over time. The SAS samples did have overall lower pH values throughout the study. When pH values increase it increases the susceptibility of the fresh-cut pears to microbes and pathogens. Many pathogens and microbes do not thrive in low pH environments, therefore, when the pH is low, there is greater chance of avoiding microbial spoilage. Table 4 presents the p-values for pH based on the different treatment factors. Treatment and Time were the only factors that made a significant difference in the pH level of the pears (p-value of .0006 and .0025, respectively). MAP or non MAP did not make a significant difference in pH of the pears over time.

**Table 4: p-values for the pH values of the pear samples as affected by the various factors**

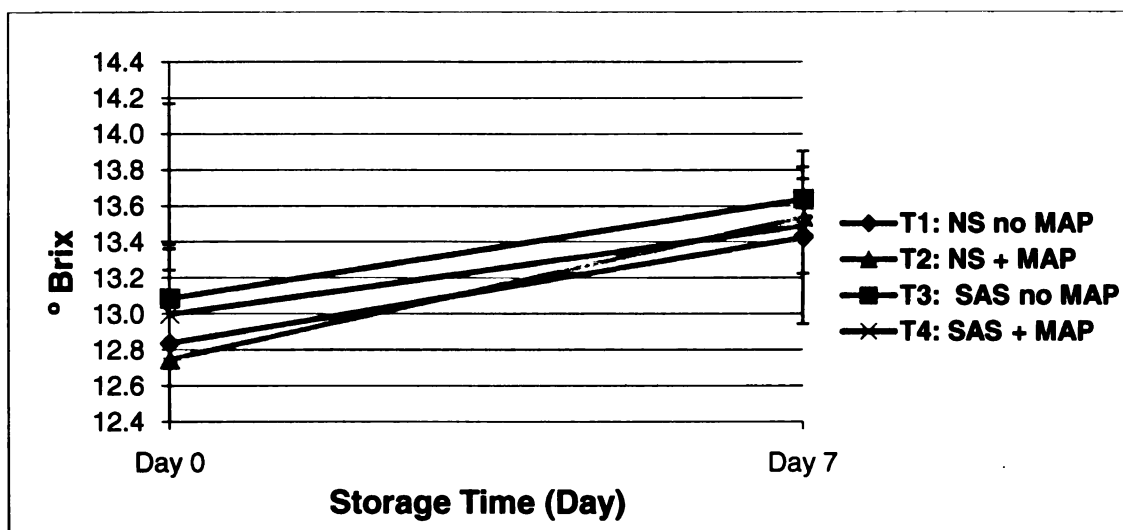
<b>Effect</b>	<b>P-Value</b>
Treatment	.0006
MAP	.2009
Treatment * MAP	.7104
Time	.0025
Treatment * Time	.3569
MAP * Time	.3569
Treatment * MAP * Time	.7104

*p < .05 is significantly different*

#### **d. TSS (Total Soluble Solids) Analysis**

The percent soluble solids were calculated using the Pal 1-Atago Pocket Refractometer. Results are presented in °Brix units, which represent the sugar content in an aqueous solution. One degree Brix corresponds to 1 gram of sucrose in 100 grams of solution, the higher the number the more concentrated the solution. The TSS does not always indicate the amount of sucrose since some fruits have only a small sucrose content; in these cases the total sugar content is measured. With fresh fruits, respiration converts the sugars that are stored in the cells into energy. The stored sugar molecules are combined with oxygen to release carbon dioxide, water and energy. This energy will then allow the pears to carry out any necessary metabolic functions. Using a MAP environment can slow down the respiration rate, thus the sugar content can be maintained for a longer period of time. The same juice that was extracted from the pear samples and used to measure pH was used to determine the TSS. Samples were analyzed over a period of one week.

Figure 9 shows the change in TSS values of the pears during the preliminary storage evaluation.



**Figure 9: TSS values of Pears over Time for the treated samples**

Pear sample T1: NS no MAP showed a slight increase (Figure 9) in the soluble solids over the 7 day period. Sample T2: NS + MAP showed a slightly larger increase in soluble solids, while T3: SAS no MAP and T4: SAS + MAP both showed a fairly constant and steady increase in soluble solids content. However, none of the treatments actually caused any significant difference in the soluble solids content of the pears. The p-values for each treatment factor studied are reported in the following table (Table 5).

**Table 5: p-values for TSS content of the pear samples based on various factors**

Effect	P-Value
Treatment	.3710
MAP	.8351
Treatment * MAP	.7289
Time	.0022
Treatment * Time	.6280
MAP * Time	.8351
Treatment * MAP * Time	.7289

*p < .05 is significantly different*



The only factor that caused a significant difference in the TSS values of the pears was time (Table 5). Neither NS nor SAS treatments caused a significant difference in the TSS values. This was also true for the packaging treatment factors, as neither MAP nor non MAP, had a significant effect on the pears. Time (one week), however, was enough to cause significant changes in the TSS values of the pears.

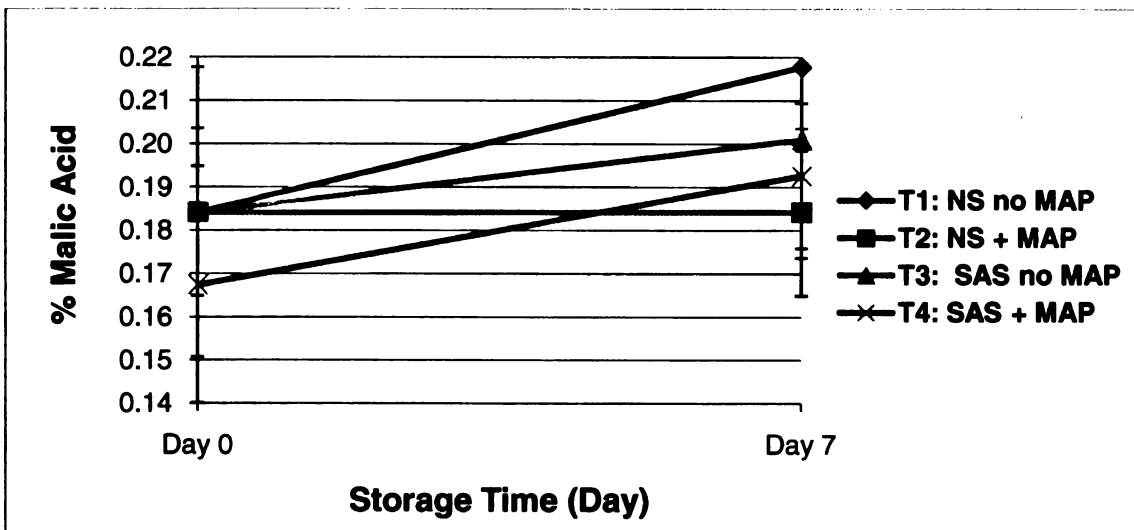
Overall, the soluble solids content did not significantly change due to the surface treatment and package type. The only factor to have a significant effect on the pears was “time”. This means that the longer the pears were in storage the more significant their difference in TSS levels. The ripening process is what causes a fruit to become softer, sweeter and less green. It was evident from the physical quality of the pears that many were softening and undergoing browning. Over time, as the pears started to ripen there was an increase in the sugar content which caused the pears to be softer, thus causing a slight increase in the soluble solids content.

**e. TA (Titratable Acidity or Total Acidity) Analysis**

Titratable acidity or total acidity is used to determine how much of a particular acid is present in a product. The balance of sugars to acid content is quite important in fruit and fruit juices because they are the main factors which determine the level of taste appeal. If a sample is too acidic it will taste sour which is considered an off flavor for pears. The TA values in this study are reported in % malic acid using the formula: % malic acid = mL NaOH x N NaOH

x 0.067 meq x 100/wt. of sample. The same juice sample, which was used for testing the pH and TSS, was used to determine the TA.

Figure 10 represents the change in TA over time for all samples.



**Figure 10: TA values of Pears over Time for the treated samples**

The only sample that did not experience any increase or decrease in % malic acid was the NS + MAP sample (Figure 10). This was probably because the pears in this sample underwent the least amount of physical breakdown (physical observation and color measurements). During the respiration process, the pear tissue is chemically broken down and there is a loss in sugars and acids which affects the TA, TSS and pH. In the case of the NS + MAP pears, the respiration rate was at a level which was optimal for a passive atmosphere, thus allowing the pears to properly respire and maintain their color and chemical structure (World Food Science, 2009). All other samples had a slight increase in TA over the 7 day period due to both the browning reaction and headspace gas

levels in the package. Table 6 gives the p-values for TA based on the various treatment factors studied.

**Table 6: p-values for TA content of pear samples based on various factors**

<b>Effect</b>	<b>P-Value</b>
Treatment	.4876
MAP	.1130
Treatment * MAP	.9302
Time	.0325
Treatment * Time	.8162
MAP * Time	.4876
Treatment * MAP * Time	.1955

*p < .05 is significantly different*

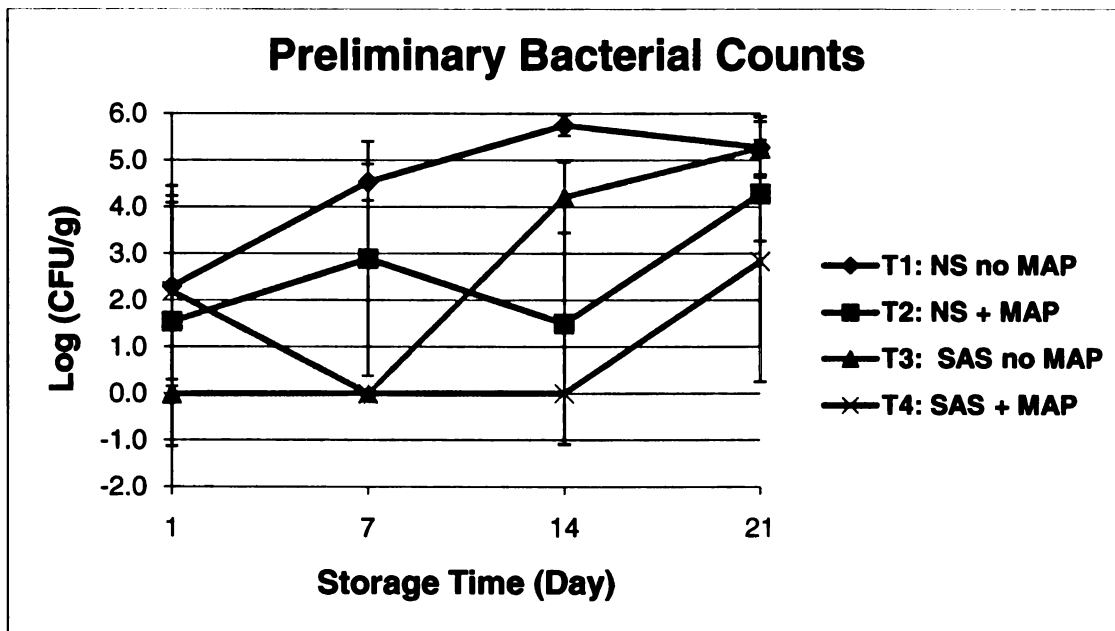
Results from Table 6 show that Time was the only factor which caused a significant difference in the TA in the pear samples. Over the 7 day time period there was a significant difference in the TA of the pears which was not due to the treatment or package type. Furthermore, it was found that the interaction of Treatment with Time and the interaction of MAP and time did not cause any significant differences in the amount of malic acid in the pears, which isolates time as the only factor causing the change. Had there been a more extended time period of study there may have been more significant differences caused by the treatments or package type.

**f. Microbiological Analysis**

The microbiological stability of fresh-cut pears was evaluated over a period of 21 days to determine the total count of bacteria, yeast and mold populations. Microbial numbers were determined for 21 days since there was an

interest in seeing how the SAS affected the microbial numbers. Due to the poor physical quality of SAS pear samples, other analytical tests were not studied past 7 or 10 days. Trypticase Soy Agar (TSA) was used for determining bacterial count and Potato Dextrose Agar (PDA) was used to determine yeasts and mold counts.

Figures 11 and 12 depict the change in microbial numbers during the 21 day period.



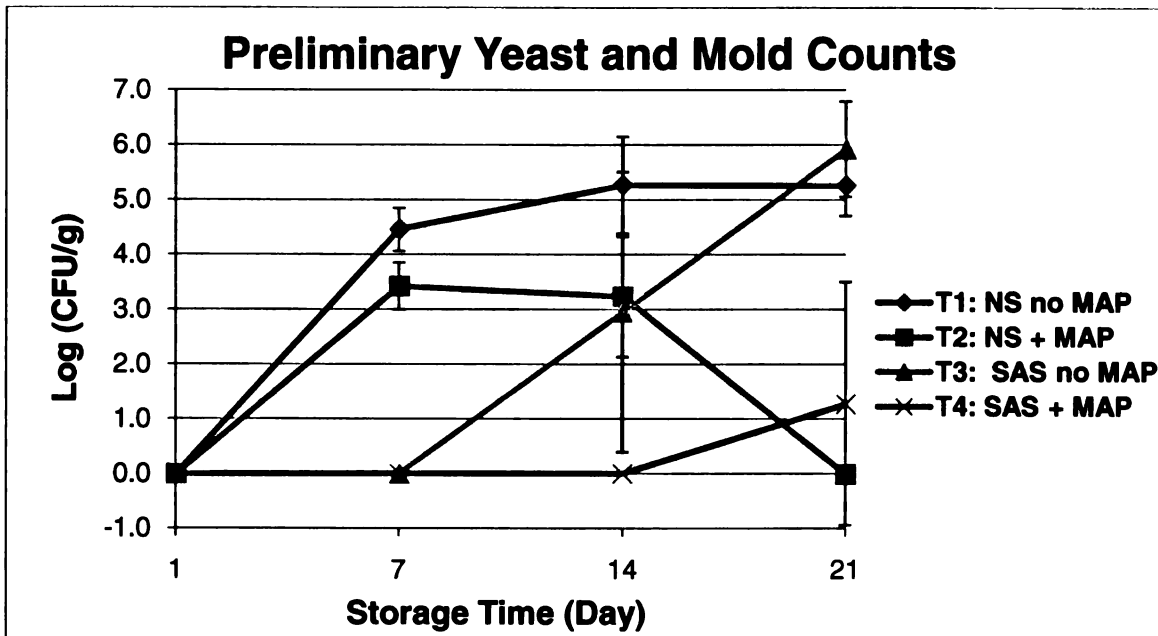
**Figure 11: Bacterial counts of Pears over 21 day period for the treated samples**

The initial bacteria counts (Figure 11) in pear sample T1: NS non MAP were approximately 2.3 log (CFU g<sup>-1</sup>) which then increased to 5.3 log (CFU g<sup>-1</sup>) by the end of the 21 day period. Sample T2: NS + MAP had an initial bacterial count of 1.5 log (CFU g<sup>-1</sup>) which then increased to 4.3 log (CFU g<sup>-1</sup>) over the

course of the study. The T3: SAS non MAP sample had an initial bacterial count of 0.0 log (CFU g<sup>-1</sup>) which did not change until day 14 at which point it was 4.2 log (CFU g<sup>-1</sup>) and at the end of the study, it was at 5.3 log (CFU g<sup>-1</sup>). Sample T4: SAS + MAP had an initial count of 2.2 log (CFU g<sup>-1</sup>) which went down to 0.0 at day 7 and by day 21 it had increased only slightly to 2.8 log (CFU g<sup>-1</sup>). No microbial growth was found on several of the SAS pear samples, whereas the NS samples always had some amount of growth each test day. In general, the SAS acted as an anti-microbial treatment since it inhibited bacterial growth on several days of testing, this was because the chemical properties of SAS work to maintain microbial activity in whichever product it is used in.

Overall, SAS + MAP pears had the lowest overall bacterial growth. The MAP helped to regulate the gas exchange within the package which helped decrease the amount of oxygen related spoilage. The MAP also helped to decrease the growth of contaminants (pathogens and spoilage microbes) from infecting the pears by providing a tight barrier to the outside environment.

In Figure 12 (below) the yeast and mold counts over time are depicted for each sample.



**Figure 12: Yeast and mold counts of Pears over 21 day period for the treated samples**

Sample T1 had a change in yeast and mold counts (Figure 12) from 0 to 5.3 log (CFU g<sup>-1</sup>) over the 21 days. Sample T2 also had an initial count of 0 log (CFU g<sup>-1</sup>) which increased to 3.4 after one week and then decreased to 3.2 after the second week. However, at day 21 there was no yeast or mold found, which may have been due to the MAP atmosphere regulating the headspace and changing the environment that was found in the previous 2 weeks (high carbon dioxide levels will inhibit yeast and mold). For sample T3, the initial count went from 0 to 5.9 log (CFU g<sup>-1</sup>) over the 21 day period. Finally, T4 increased only a slight amount from 0 to 1.3 log (CFU g<sup>-1</sup>), however, not until Day 14. Almost no yeast or mold growth was present on the SAS + MAP pear samples. SAS samples in general showed a greater inhibition of bacteria, yeast and mold in

comparison to the NS samples. Again, the low growth may have been due to the MAP environment, which created high levels of CO<sub>2</sub>, low levels of O<sub>2</sub> and prevented microbes from entering the package.

Table 7 shows the p-values based on the various treatment factors that may have affected the growth of bacteria, yeasts and molds.

**Table 7: p-values for Microbial counts in pear samples due to the various factors**

Effect	P-Value	
	TSA-YE	PDA
Treatment	.0006	<.0001
MAP	.0018	<.0001
Treatment * MAP	.3725	.7416
Time	.0002	<.0001
Treatment * Time	.0813	<.0001
MAP * Time	.0038	<.0001
Treatment * MAP * Time	.3449	.6329

*p<.05 is significantly different*

The Treatment type, package type (MAP), Time, and interaction of MAP \* Time all caused a significant difference in the bacteria level over time (Table 7). For the pear yeast and mold counts, the interaction of Treatment \* MAP and interaction of Treatment \* MAP \* Time were the two factors that did not cause a significant difference. Treatment type, package type (MAP) and time caused significant differences to occur in the bacteria, yeast and mold counts.

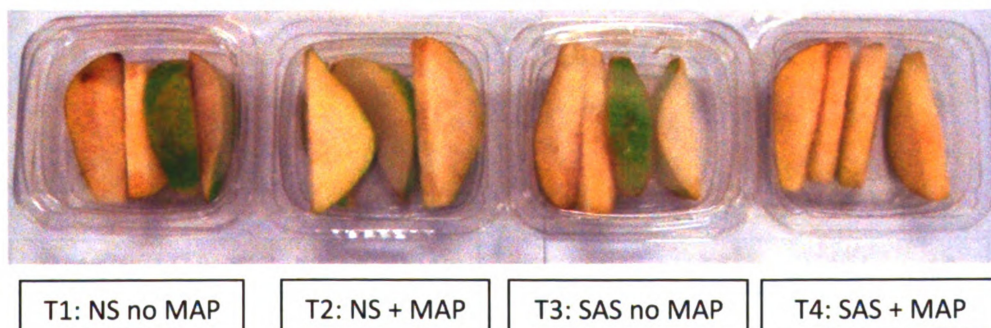
Treatment and package type had a significant effect on the growth of bacteria, yeast and mold. The lower counts in the MAP may have been due to the effectiveness of the oxygen concentration in the package headspace and the

hermetic nature of the package seal. The MAP proved to inhibit many bacteria, yeasts and molds. Although it did not completely prevent all types of growth, the MAP samples had lower overall counts as compared to the non MAP pear samples. The SAS samples also had lower growth levels compared to the NS pear samples. Since the SAS treatment has an acid component in it, this was one of the reasons for the total inhibition of microbes in some cases and the smaller numbers in others (Jones-Hamilton).

**g. Visual Observation of Pear Quality**

There was also a substantial visual and physical difference in the SAS and NS pears which is shown in the following photographs. The following photographs represent the evolution of the physical quality of the pear samples over a 21 day period. Although the chemical and biological tests had concluded much earlier, there was an interest to see how the SAS treatment affected the pears over a longer time period. The second preliminary round of testing resulted in pear samples that were the same, or very similar, to the first set. Therefore, set one of the photos is only shown (Figure 13).

**Day 0**



**Figure 13: Preliminary photos of pear treatments over 21 days**



**Day 3**



T1: NS no MAP

T2: NS + MAP

T3: SAS no MAP

T4: SAS + MAP

**Day 7**



T1: NS no MAP

T2: NS + MAP

T3: SAS no MAP

T4: SAS + MAP

**Day 10**



T1: NS no MAP

T2: NS + MAP

T3: SAS no MAP

T4: SAS + MAP

**Day 14**



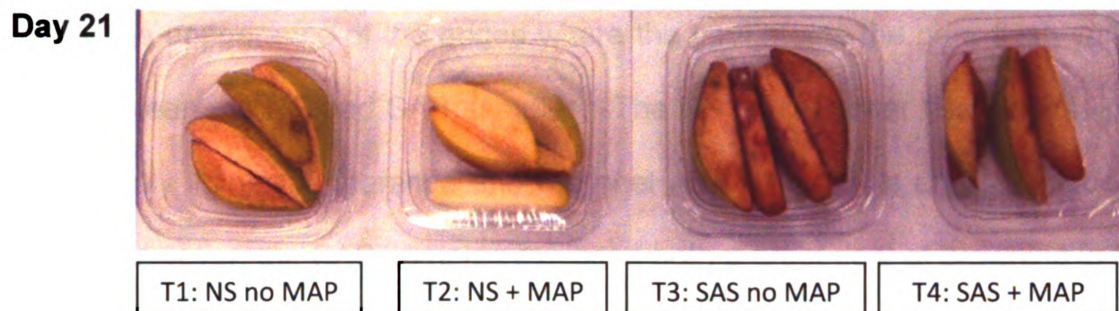
T1: NS no MAP

T2: NS + MAP

T3: SAS no MAP

T4: SAS + MAP

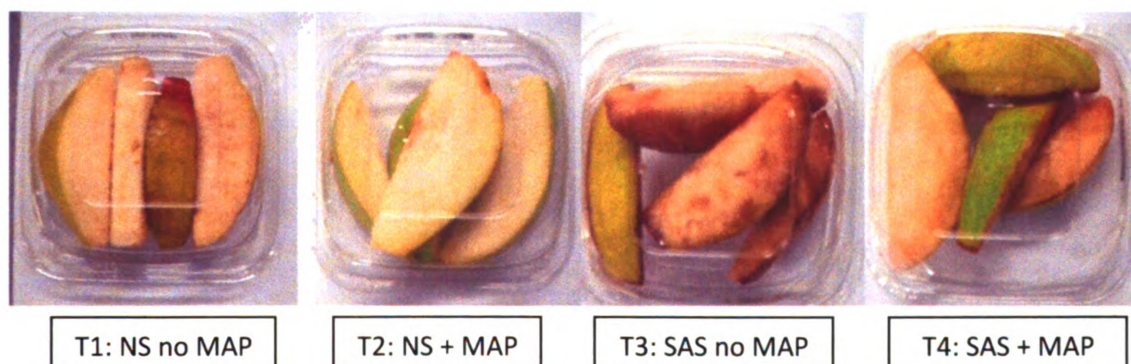
**Figure 13: Preliminary photos of pear treatments over 21 days (continued)**



**Figure 13: Preliminary photos of pear treatments over 21 days (continued)**

From the photos it is fairly evident that the NS samples did not undergo as severe browning as did the SAS samples. The SAS samples exhibited a higher amount of surface deterioration and softening of the pears. The edges of the SAS treated pears (area where peel meets flesh), showed significant amounts of color leaching from the peel to the surface of the pears (see below for additional photos). The NS + MAP sample maintained the whitest and freshest looking appearance throughout the study, whereas the NS non MAP pears showed slightly more change over time.

**Closer look at peel and flesh browning on Day 21:**



**Figure 14: Preliminary photos of pear peel and flesh**

The appearance of the edges where the peel meets the pear flesh varied a lot between the NS and SAS samples. The NS + MAP pear flesh maintained its relatively lighter appearance and the peel also retained its original shade of green. However, the SAS samples exhibited both a great deal of browning in the flesh as well as in the peel. The SAS treatment may have resulted in a faster aging process in the pears thus causing the cut surface to experience more decay, softening and browning. The SAS treatment was used at a 2% concentration that may have also caused the more severe browning in pears. The SAS treatment may also have negatively affected polyphenol oxidase activity, thus inducing enzymatic browning (Jones-Hamilton).

Overall, the extreme softening and browning of the SAS pears resulted in their elimination from the final storage study. Since visual characteristics are one of the first measures of quality that consumers have access to, the pears must look fresh.

## **II. Final Results**

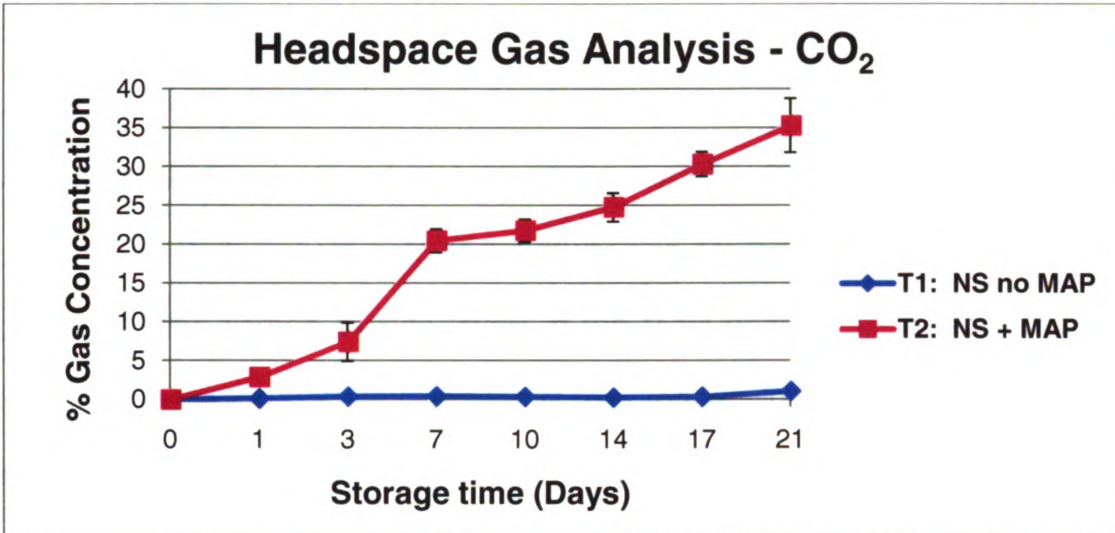
The following results and discussion are based on the final storage study. At the completion of the preliminary study it was decided that SAS would not be further pursued. The samples in the final study were NS no MAP and NS + MAP. The average results tables for each test evaluation can be found in Appendix VII.

### **a. Headspace Gas Analysis**

The headspace gas levels of oxygen and carbon dioxide were studied over a period of 21 days. Initial levels of O<sub>2</sub> and CO<sub>2</sub> on Day 0 were assumed to be 20% and 0%, respectively (atmospheric environment). In order to help increase the shelf life of products, it is important to increase the CO<sub>2</sub> levels and reduce the O<sub>2</sub> levels. This helps to maintain a slower respiration rate in the pears. Without the proper amount of oxygen, the pears will go into anaerobic respiration and spoil since they cannot respire without oxygen. Kader and Saltveit (2003) stated that to preserve fresh-cut fruits, oxygen levels should be <5% and elevated carbon dioxide levels above 3%.

Two measurements were taken from two separate containers on each testing day (0, 1, 3, 7, 10, 14, 17, and 21). The containers were discarded after the headspace, color, and photo analysis were completed. The following figure (Figure 15) shows the levels of CO<sub>2</sub> in the package headspace over time for both package types, which includes the second set (replicate) study.





**Figure 15: Headspace CO<sub>2</sub> Concentration of Pears over Time at 4°C**

In Figure 15, it can be seen that the carbon dioxide level in the pears with MAP steadily increased, despite the variation around day 7. For the non MAP pears there was no change in the gas concentration over time. The non MAP samples did not have a tight fitting seal; therefore, gases were continuously migrating in and out of the package in connection to the outside environment.

Table 8 shows the p-value (or significant difference) in the carbon dioxide levels of the pears in both package types over the 21 day period.

**Table 8: p-values for CO<sub>2</sub> level as affected by the package type**

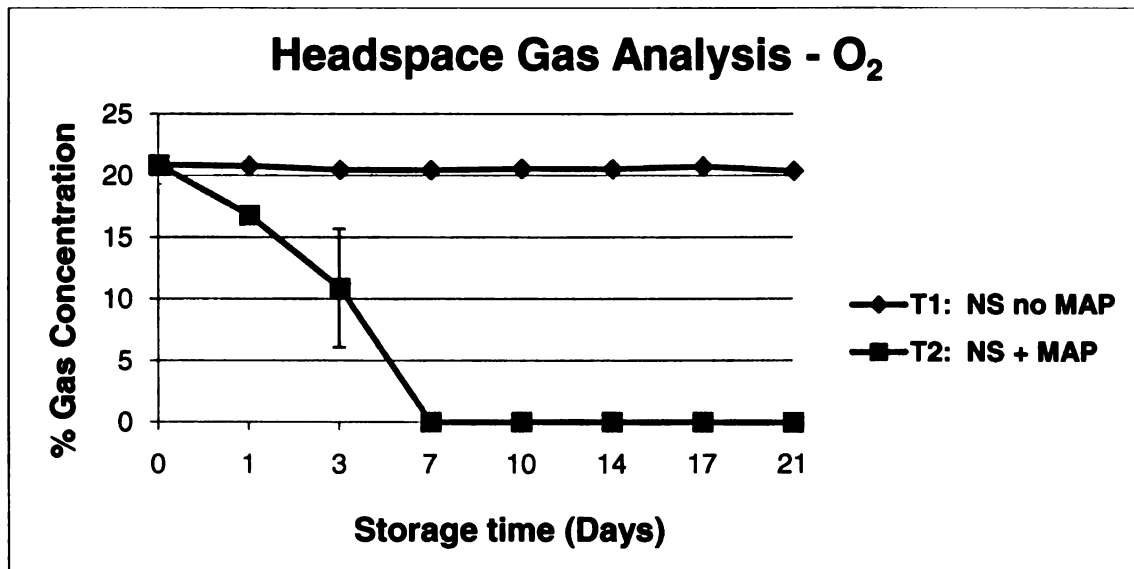
Effect	P-Value
MAP	<.0001
Time	<.0001
MAP * Time	<.0001

*p<.05 is significantly different*

From Table 8, it can be seen that all three factors made a significant difference in the carbon dioxide levels of the pears. The presence of MAP versus no MAP was found to have a p-value of  $<.0001$ . The MAP package behaved similarly in both sets of the study and proved to be the more effective package. The carbon dioxide levels in the MAP pear samples increased over time, which meant that their aerobic respiration would be slowed. "Time" also significantly affected the carbon dioxide levels in the pears as is evident in Figure 15. Thus, as time increased the concentration levels significantly differed between pear samples. A fairly steady increase is seen in the concentration between each of the testing days for the MAP samples. A positive correlation is seen between time and concentration for the MAP samples; so as the time period increases, the concentration does as well. This trend was not evident for the non MAP pear sample, which exhibited a constant concentration as time increased throughout the study. The interaction of MAP and time also had a significant difference in carbon dioxide concentration; thus the effect on pears between time with MAP and time without MAP significantly differed. Pears use oxygen to aerobically respire which releases carbon dioxide, thus slowing down their respiration and increasing the likelihood of lower microbiological growth. The lower the rate of respiration (as long as it does not go into anaerobic respiration) the better the chance of prolonging the pears shelf life and quality.

Oxygen concentration was also monitored over the 21 day period. Two readings were taken from two separate containers, and a second set of data was also collected for a replicate study. The oxygen and carbon dioxide

concentrations were measured simultaneously for each container. Figure 16 depicts the oxygen evolution over the 21 day time period for both sample sets.



**Figure 16: Headspace O<sub>2</sub> Concentration of Pears over Time at 4°C**

From Figure 16 it can be seen that the oxygen concentration remained fairly constant for the pear sample without MAP. So as storage time increased the oxygen concentration remained unchanged for T1, which was again due to the lack of tight seal between the package and the lid. For the MAP pear sample, there was an overall decrease in the concentration. The MAP pears showed a significant drop in concentration between days 0 and 7 after which the oxygen concentration reached a steady state for the remainder of the study. Between days 7 and 21 the MAP pear sample had a very low and steady level of oxygen, which meant that a steady respiration rate was achieved. Evidently, enough oxygen was still present to prevent the pears from going into anaerobic respiration. By having oxygen levels at or near 0% the pears were able to obtain

an extended shelf life. In this case the shelf life was able to be extended for 21 days.

Table 9 expresses the p-values which determine what factors played a significant difference in the oxygen concentration over the 21 day period. The values were calculated using both sets of data.

**Table 9: p-values for O<sub>2</sub> level as affected by the package type**

<b>Effect</b>	<b>P-Value</b>
MAP	<.0001
Time	<.0001
MAP * Time	<.0001

*p<.05 is significantly different*

From Table 9 we see that each of the factors studied had a significant effect on the oxygen concentration of the pears. There was a significant difference between the MAP and non MAP samples over the 21 day period. As time increased a significant difference was also seen between the concentration level of the MAP pears and the non MAP pears. This meant that the longer the pears spent in storage the longer they had time to reach a steady state of respiration within the MAP sample. Since pears have a lower respiration rate similar to apples it explains why they do not immediately show a lower oxygen concentration. The MAP helps to control the amount of oxygen that comes in contact with the pear so that the pears do not respire too quickly and spoil. The interaction between MAP and time also resulted in a significant difference which meant that the effects of MAP and time were different from the effects of non MAP and time on the oxygen concentration level. The package system with the

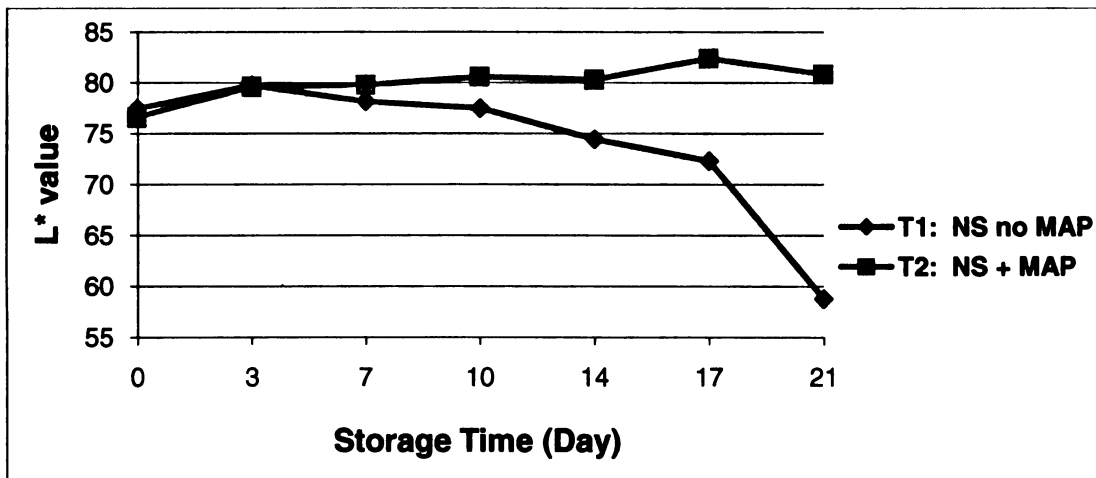


hermetic seal in combination with the respiration rate of the pears had a positive influence on extending the shelf life of the pears to 21 days by regulating the amount of oxygen entering the package, and allowing the optimal amount to be used for respiration.

**b. Color L\*, a\*, and b\* Analysis**

The color coordinates, L, a, and b were measured on the pear samples using the same method as in the preliminary study. The values of L, a, and b on the color scale were read from each pear sample and recorded. The pear samples were tested at Day 0, 1, 3, 7, 10, 14, 17 and 21. A total of four readings were taken from each of the two treatments (T1 and T2). A second set was also evaluated following the same procedure, and the same number of readings was taken from samples T1 and T2.

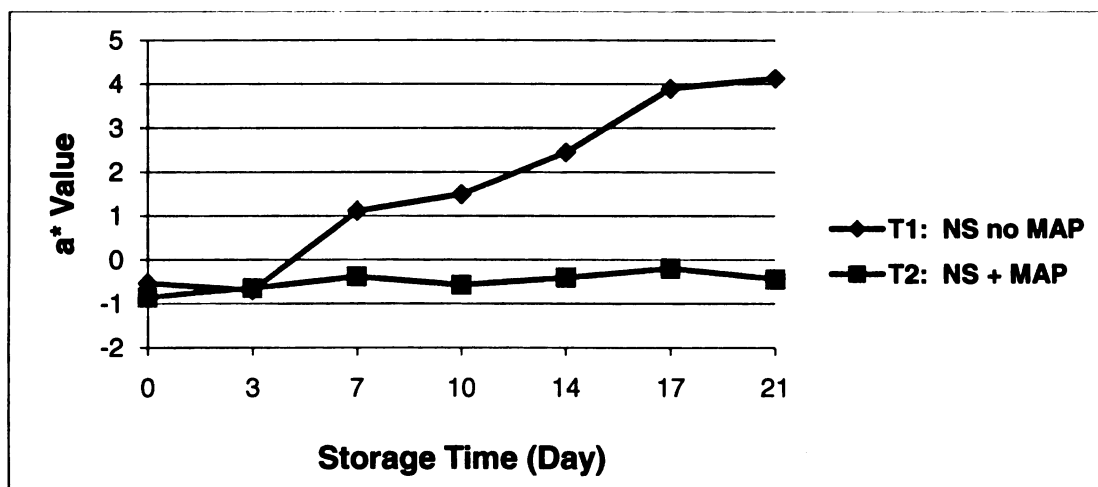
The following graph shows the evolution of the L\* coordinate of color, over the 21 day time period, for both the MAP and non MAP pear samples.



**Figure 17: L\* color of Pears over Time**

From Figure 17 it can be seen that the pear sample without MAP had a fairly significant decrease in the L\* value over the 21 period. The sample did not show a dramatic change during the first 14 days; however, after that point the color showed a significant amount of darkening which is highly undesirable in fresh-cut pears. This means that a storage period of 14 days was optimal for the non MAP sample, and that after that time it would no longer have acceptable physical quality. Although there was a slight increase in the lightness for the MAP sample it was no real significant change over time. Overall, this means that the MAP provided better control of oxidative browning.

The a\* value was the second color component which was measured for the pears. Again, two sets of data were collected over a period of 21 days and graphed below.



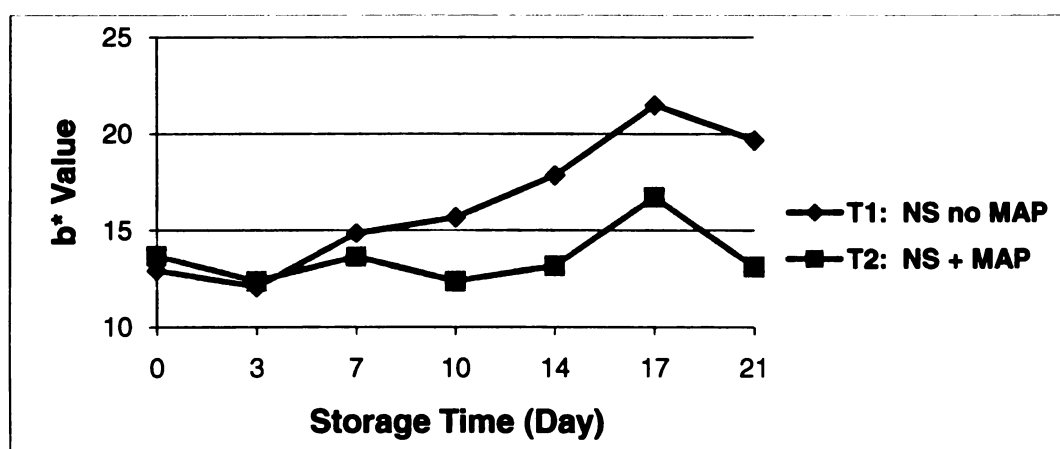
**Figure 18: a\* color of Pears over Time**

For the a\* color component it can be seen that the NS pears without MAP showed a significant difference compared to the NS pears with MAP (Figure 18). The pears without MAP had a significant increase in their a\* value over the 21

day period which means that the pears were trending towards a more red color and becoming more brown. The pears with MAP showed values that consistently remained below 0 over the 21 day period, which meant that they maintained their original color and stayed fairly green in color. Thus, T2 proved to be the more optimal packaging choice to preserve the green color of the pears. The MAP helped to regulate both the amount of oxygen and carbon dioxide in the package, which decreased the amount of oxidation as well as helped regulate the respiration rate in the pears. This is why MAP samples showed a more consistent color throughout the study and did not undergo excessive browning.

In both the preliminary and final study, NS with MAP was found to have the greenest looking pears. The MAP helped control the air flow of gases in the headspace of the samples, which caused the pears to have a much lower respiration rate, helping to prolong the shelf life.

The results for the average  $b^*$  values are shown in Figures19:



**Figure 19:  $b^*$  color of Pears over Time**

The b\* values for both the MAP and non MAP pears were shown to be significantly different from each other (Figure 19). The b\* values for NS + MAP stayed fairly consistent with time and did not show any significant changes in color. The non MAP samples had an increase in b\* values over the 21 day period, though they were not statistically significant. This means that the non MAP samples were starting to take on a more yellow color over time, indicating signs of decay.

The following table shows which factors had a significant difference in the color L\*, a\*, and b\* values of the pears (Table 10).

**Table 10: p-values for L, a, and b values as impacted by the package type**

Effect	P-Value		
	L*	a*	b*
MAP	<.0001	<.0001	<.0001
Time	<.0001	<.0001	<.0001
MAP * Time	<.0001	<.0001	<.0001

*p<.05 is significantly different*

When studying the effects of MAP, Time and the interaction of MAP\*Time it is evident that each factor significantly affected all aspects of the color value (Table 10). In conjunction with the graphs presented earlier, it can be seen that MAP caused significant differences in the L\*, a\* and b\* values. Since the MAP helped to regulate the respiration rate, the effect on the overall color of the pears was positive and there was color preservation. Time also had a significant effect on the color values so as the storage time increased the MAP and non MAP samples had significantly different overall values. As evident in Figures 17-19 as

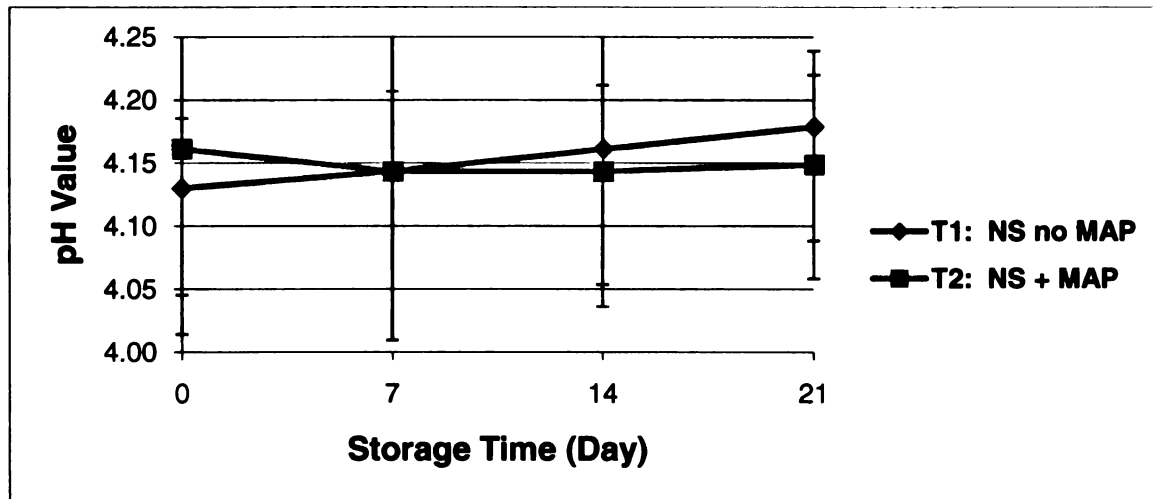
time increased the MAP samples retained their original color and did not undergo any significant browning, which was statistically different from the non MAP pears which showed significant browning over time. The interaction of MAP and Time was also found to be statistically different which means that the effects of MAP with Time compared to the effects of non MAP and time were statistically different in relation to the various aspects of pear color.

Overall, MAP had a significant positive influence on retaining the initial color of the pears over the 21 day period. The ability of the package system to allow enough oxygen to maintain the respiration process, and deter the pears from going into anaerobic respiration, helped the pears to avoid rotting and browning. MAP also limited the amount of oxygen available to the enzyme polyphenol oxidase which helped to decrease the oxidative browning and maintain the fresh color in the pears. Since the physical aspect of a product is one of the first that is seen by consumers, it is important to maintain fresh color both on the shelves as well as in the home after an extended period of time. Ultimately, the MAP proved to extend the physical shelf life of the pears over time.

### **c. pH Analysis**

The pH measurements were taken at weekly intervals (Day 0, 7, 14 and 21) for both samples T1 and T2. The pH measurements help to indicate a change in acidity of the pears. Understanding the change in acidity can help to determine how different the taste of the pear sample will be. As mentioned in the preliminary results, D'Anjou pears are slightly acidic to begin with, therefore, it is

important to monitor the change in pH to see how it may potentially affect the flavor, physical quality and chemical quality of the pears. Pear samples from the color and headspace test were juiced to test the pH, TSS and TA. Figure 20 represents the change in pH over time.



**Figure 20: pH values of Pears over Time**

From the results shown, no significant differences occurred in the pH values between the MAP and non MAP samples over the 21 day period. The NS non MAP sample had a slight increase in the pH while the NS + MAP had a slight decrease in pH. However, the changes in pH were not significant for either sample (Figure 20). The non MAP pears showed a slight increase in pH which could make them more susceptible to microbial growth and spoilage. The MAP samples had a slightly lower pH which could help to increase their shelf-life. The p-values for the pH as affected by the treatment factors are presented below.

**Table 11: p-values for pH as impacted by the package type**

<b>Effect</b>	<b>P-Value</b>
MAP	.8389
Time	.9255
MAP * Time	.4899

*p < .05 is significantly different*

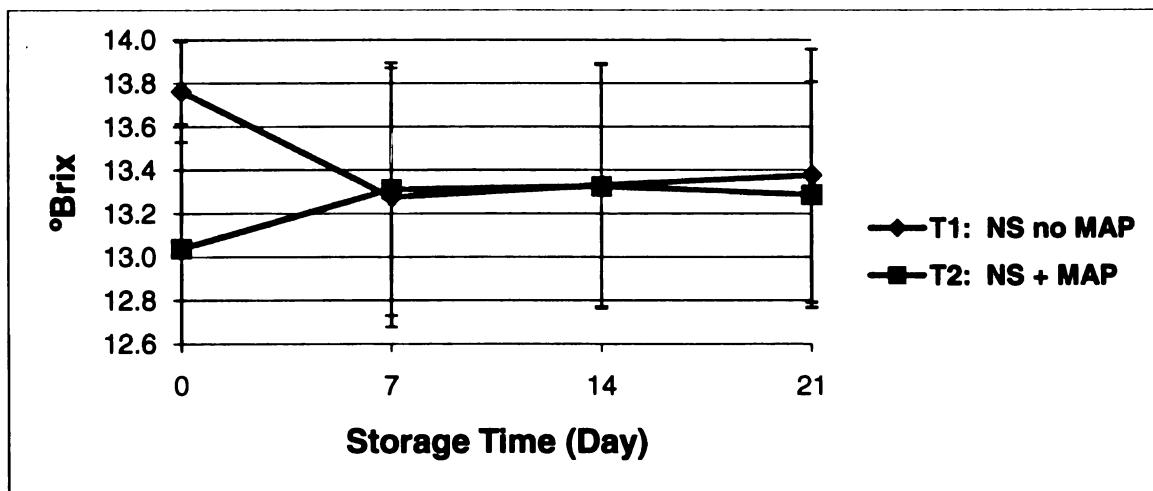
When assessing the affects of MAP, Time and the interaction of MAP and Time, it was found that none of the factors caused a significant difference in the pear samples. The use of MAP did not statistically resulted in a difference in the pH between the pears. The time period of 21 days also did not have a significant effect on the pH value of the pears either. MAP and non MAP samples had pH values that did not vary drastically, even though there was some variation. Finally, the interaction of MAP and Time was also not significantly different, despite some small variations in the data.

Overall, there were no significant differences in the pH level of the pears, some minimal differences occurred. It is still important to note the differences between the MAP pears and the non MAP pears. It is also important to note that the pH values were decreasing in the MAP pears, which means that it was providing the NS a way to effectively be used in lowering the pH and in inhibiting browning.

**d. TSS (Total Soluble Solids) Analysis**

The percent soluble solids was also calculated and analyzed using the same pear juice sample as used for pH testing. Results are presented in °Brix

units, which represent the sugar content in an aqueous solution. One degree Brix corresponds to 1 gram of sucrose in 100 grams of solution, the higher the number the more concentrated the solution. The TSS does not always give the amount of sucrose since some fruits have small sucrose contents and large amounts of non-sucrose rings; in these cases the total sugar content is measured. When fresh fruits respire it converts the sugars into energy. The stored sugar molecules are combined with oxygen to release carbon dioxide, water and energy. This energy will then allow the pears to carry out any necessary metabolic functions. Using a MAP environment can slow down the respiration rate, thus the sugar content can be maintained for a longer period of time. The change in the TSS content over the 21 day period is shown in Figure 21.



**Figure 21: TSS values of Pears over Time**

For the NS non MAP pears the degree of soluble solids decreased slightly over the 21 day period (Figure 21). For the NS + MAP pears there was a slight increase in the TSS, however, neither sample had significantly different results.



MAP was able to help keep the TSS level of the pears constant after the first week of the study. For the non MAP sample the TSS level dropped within the first week, however, it remained at a fairly constant value the remainder of the study. The MAP sample showed an increase within the first week which also somewhat steadied out over time. There were no dramatic changes in their initial and final TSS values, even though some variations occurred. In the ripening process the soluble solids content tends to increase thus causing a product to become less green and sweeter. When the respiration rate is higher, as was in the case of NS non MAP pears (compared to NS + MAP pears), there can be more moisture loss within the pears and more tissue softening, thus decreasing the soluble solids content, as seen from Figure 21. With loss in moisture there can then also be a decrease in the acidity of the pears. This could be due to the loss of organic acids because of the higher respiration rate. This decrease in acidity was also apparent from the pH levels of the NS non MAP pears (Figure 20).

When the respiration rate is lower, there tends to be more moisture retention in products, as in the case with the MAP pears. This moisture retention can result in a higher level of solids (seen in Figure 21) and can also decrease the amount of tissue softening that occurs. With the headspace gas composition that developed due to passive MAP, the respiration rate would have fallen, resulting in extended shelf-life. The statistical significance of the treatments on the TSS was determined and is shown in Table 12.

**Table 12: p-values for TSS as impacted by the package type**

<b>Effect</b>	<b>P-Value</b>
MAP	.3422
Time	.9669
MAP * Time	.4679

*p < .05 is significantly different*

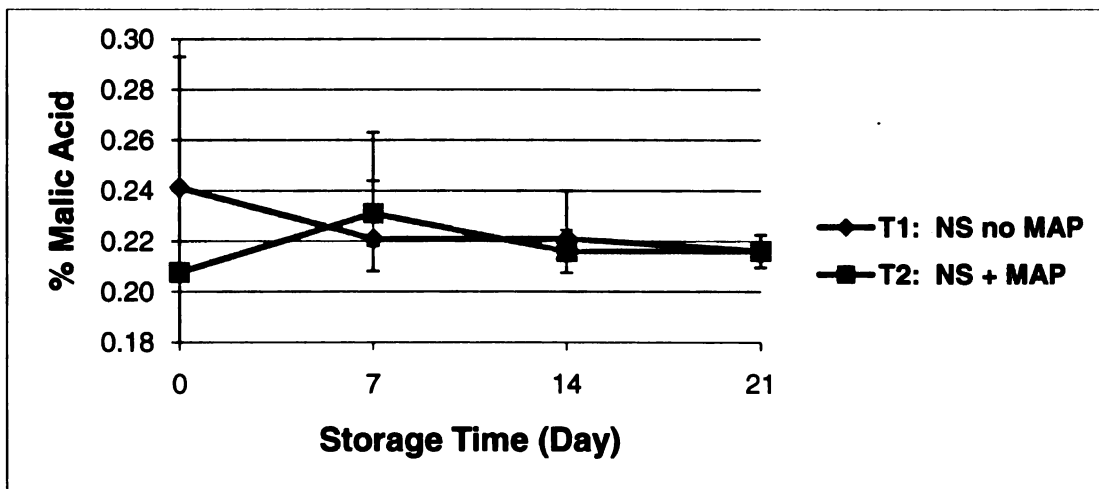
None of the factors had a significant effect on the TSS levels of the pears. Even though variations occurred due to the different metabolic processes, they were too minimal to have any significant effect on the pears. It is still important to understand and recognize what is happening with the pears, to determine even the slightest increases and/or decreases in the soluble solids level. Although there were slight changes in the TSS, MAP did not have a significant effect on the level of soluble solids in the pears.

**e. TA (Titratable Acidity or Total Acidity) Analysis**

Titratable acidity or total acidity is used to determine how much of a particular acid is present in a product. The balance of sugars to acid content is important in fruit and fruit juices because they are the main factors which determine the level of appeal in taste. If a sample is too acidic it will taste sour which is not a preferred taste for pears. Higher acid levels in fruits tend to mean a lower pH, which can play a major role in the taste of a product, in the color and in its microbial stability.

The change in the TA for the different pear samples is presented in Figure 22. The MAP sample showed a slight increase in the % malic acid over time,

although it was not a significant increase. The non MAP sample showed a decrease in the % malic acid over time, but again it was not significant. Although there were variations in the TA, none of the treatments caused a significant difference in the TA of the pears. It is still important to note that with the slight decrease in pH that occurred in the MAP samples, the TA would be expected to rise, which is what is shown in Figure 22 (and vice versa for non MAP pears), even though the variations were small over the storage period. Since the pH did not vary much during the study, it corroborates well with the changes in TA. Also, since there was little physical ripening of the pears, the TA would not be expected to change substantially.



**Figure 22: TA values of Pears over Time**

The p-values for the different factors studied are presented in Table 13.

**Table 13: p-values for TA as impacted by the package type**

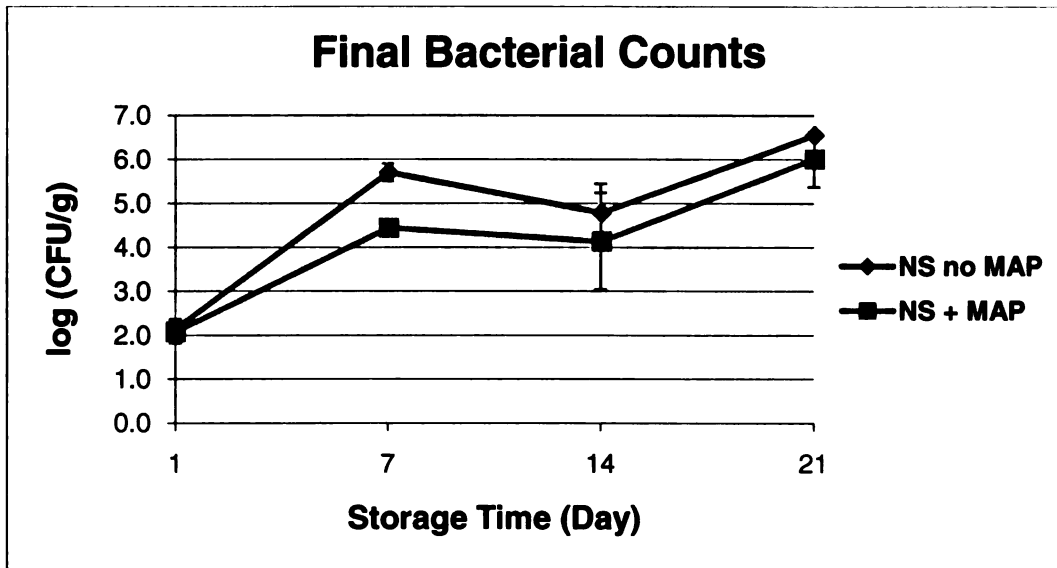
<b>Effect</b>	<b>P-Value</b>
MAP	.6151
Time	.8347
MAP * Time	.5118

*p < .05 is significantly different*

None of the factors had a significant effect on the TA levels of the pears. MAP did not significantly influence the TA levels in a positive or negative way. The length of the storage period did not affect the TA levels in the pear samples. Since the pears did not ripen or brown significantly, it is likely that the respiration rates did not change substantially over time.

**f. Microbiological Analysis**

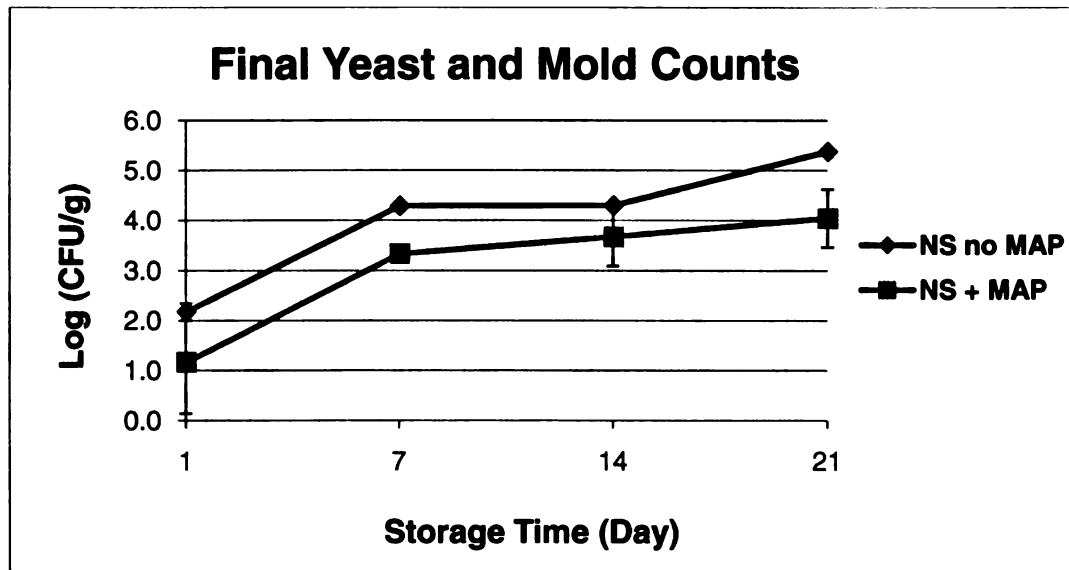
The microbiological stability of the fresh-cut pears was evaluated over a period of 21 days to determine the total bacteria, yeast and mold populations. Trypticase Soy Agar (TSA) was used in determining the bacterial count and Potato Dextrose Agar (PDA) was used to determine the yeasts and mold counts.



**Figure 23: Bacterial counts of Pears over 21 day period**

The initial bacterial populations of the non MAP samples was found to be 2.15 log (CFU g<sup>-1</sup>) which increased to 6.55 log (CFU g<sup>-1</sup>) over the 21 day period. The increase may have been due to the lack of a tight seal, which allows no oxygen to enter the package. The MAP pears had an initial bacterial count of 2.08 log (CFU g<sup>-1</sup>) which increased to 6.02 log (CFU g<sup>-1</sup>) by the end of the storage period (Figure 23). The MAP pears had slightly lower bacterial counts at each test day. Since a general decrease occurred in the pH of the MAP pears, this could have resulted in lower overall bacterial populations than the non MAP pears. MAP also helped to regulate the amount and type of gas in the package which decreased the amount of oxygen available to organisms that thrive in oxygen environments. The tight barrier provided by the MAP also eliminated any physical pathway through which microbes could enter. The yeast and mold

population was also studied for both pear samples over a period of 21 days (Figure 24).



**Figure 24: Yeast and Mold counts of Pears over 21 day period**

The yeast and mold results are similar for both the MAP and non MAP pears. The non MAP pears had higher numbers on each test day as compared to the MAP pears. The initial yeast and mold count for the non MAP samples was 2.2 log (CFU g<sup>-1</sup>) which increased to 5.4 log (CFU g<sup>-1</sup>). For the MAP pears the initial count was 1.2 log (CFU g<sup>-1</sup>) which increased to 4.0 log (CFU g<sup>-1</sup>) over the storage period (Figure 24). The MAP pears consistently had lower counts on each test day as compared to the non MAP pears, which indicates that MAP was a somewhat better in controlling the growth of yeasts and molds. The tight seal provided by the MAP also eliminated the likelihood that yeasts and molds would have been able to enter the package. The statistical significance of each factor

on the growth of bacteria, yeast and mold in the pear samples is shown in Table 14.

**Table 14: p-values for Microbial counts in pear samples due to various factors**

Effect	P-Value	
	TSA-YE	PDA
MAP	.0109	.0001
Time	<.0001	<.0001
MAP * Time	.3209	.6371

*p<.05 is significantly different*

MAP pears had a significant lower number of bacteria, yeast and mold counts compared to non MAP pears. From Figures 23 and 24, it is seen that MAP helped to keep the total counts lower over the storage period compared to the non MAP pears. The MAP pears had a slightly lower pH which may have reduced the growth of micro-organisms. The MAP pears also had a package system that was a better barrier to contaminants compared to the non MAP system. Most importantly, the oxygen level in the MAP pears was lower, and the CO<sub>2</sub> level was higher, which would reduce the growth of mold and certain spoilage bacteria.

Time was also found to be a factor which induced significant differences in the bacteria, yeast and mold counts of both pear samples. From Figure 23 and 24 it is shown that as time increased the bacteria, yeast and mold counts also increased significantly. The difference in the initial and final growth numbers in both pear samples was found to be significantly different over time. The longer

the pears were in storage, the more time was available for microbes to infect their host and start to grow. Since NS is not an anti-microbial treatment it was expected that some growth would occur. However, due to the MAP it was expected that the microbial growth would be substantially less for those pears as compared to the non MAP pears. Even though the microbial counts were less, the expectation was that the difference would be more.

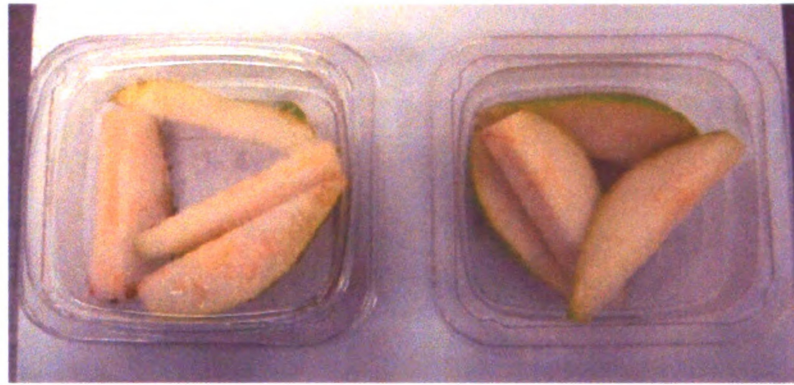
The interaction of MAP and Time did not have a significant effect on the microbial growth of pears. This means that as the time increased having MAP or non MAP did not cause a significant difference in the microbial growth of pears (also seen in Figures 23 and 24). Individually, MAP and Time had caused significant differences; however, collectively they did not. Overall, MAP did have a positive influence on the level of bacteria, yeast and mold counts as compared to the non MAP samples.

**g. Visual Observation of Pear Quality**

The following photos represent the change in the physical quality of the pears in both a non MAP and MAP environment over the 21 day storage period. Since the second set of pears looked very similar to, or the same as the first set, only the first set of pears is presented (Figure 25).



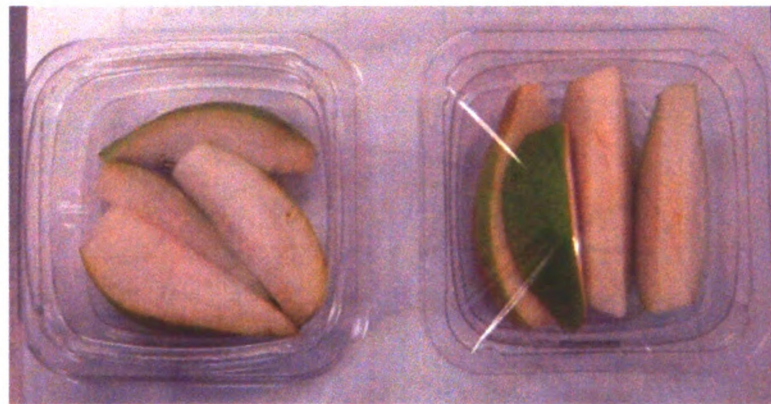
**Day 0**



T1: NS no MAP

T2: NS + MAP

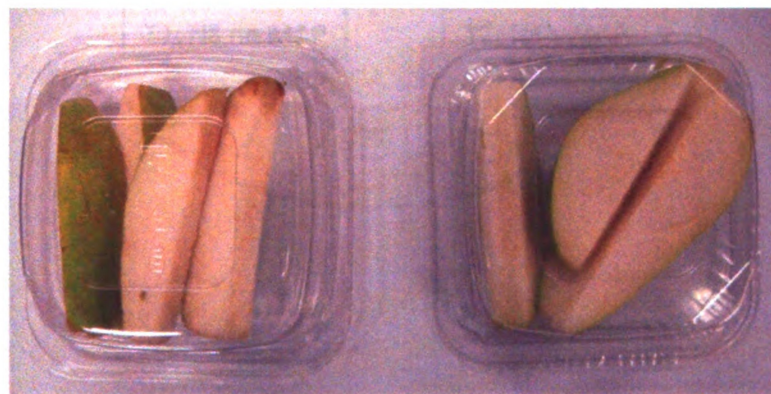
**Day 3**



T1: NS no MAP

T2: NS + MAP

**Day 7**



T1: NS no MAP

T2: NS + MAP

**Figure 25: Final photos of pear treatments over 21 days**

**Day 10**



T1: NS no MAP

T2: NS + MAP

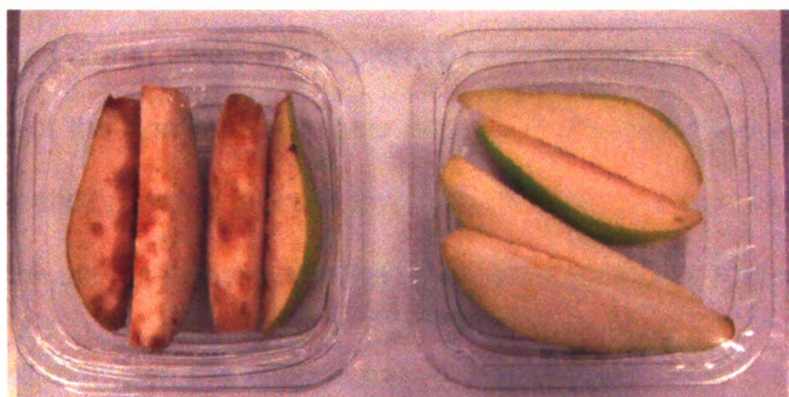
**Day 14**



T1: NS no MAP

T2: NS + MAP

**Day 17**



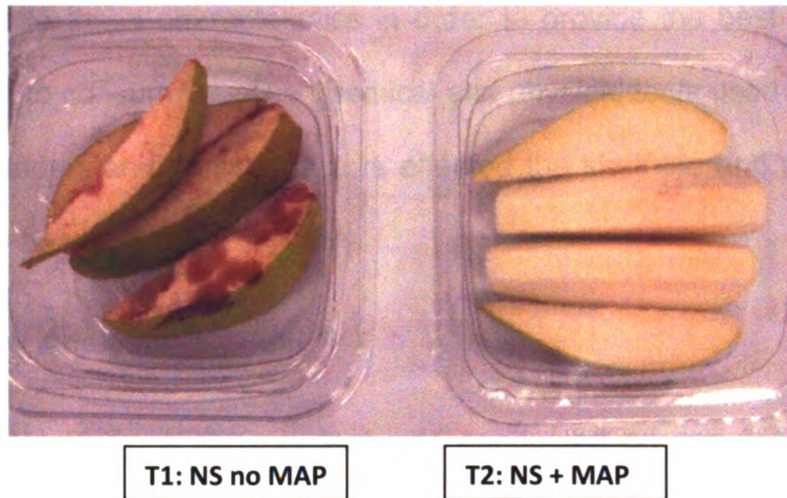
T1: NS no MAP

T2: NS + MAP

**Figure 25: Final photos of pear treatments over 21 days (continued)**



Day 21



**Figure 25: Final photos of pear treatments over 21 days (continued)**

From the photos it is very evident that the MAP pears had the best physical quality. The overall quality over the 21 day period remained more consistent in the MAP pears compared to the non MAP pears. The MAP pears looked the freshest and had the whitest looking flesh, whereas the non MAP pears had a significant amount of browning from day 3-7. The appearance of browning is consistent with the hunter color results which showed that the non MAP pears had higher levels of discoloration and browning. A major factor in the discoloration of the non MAP samples was the higher level of oxygen available for the enzymatic browning process involving PPO. Oxygen acts as a catalyst for the PPO which ultimately produces the brown color on the surface of the pears.

Overall, between the chemical tests and physical photos, the MAP pears proved to be the freshest looking product with the most optimal characteristics. As is the case of any product on the retail shelf, the color and physical appearance of a product are key factors in product purchasing. High priority

must be given to these characteristics in order to provide the best looking and tasting product to consumers. The chemical and physical tests used in this study were vital to understanding what factors affected the shelf-life and provided the optimal quality pears.

#### **h. Sensory Analysis**

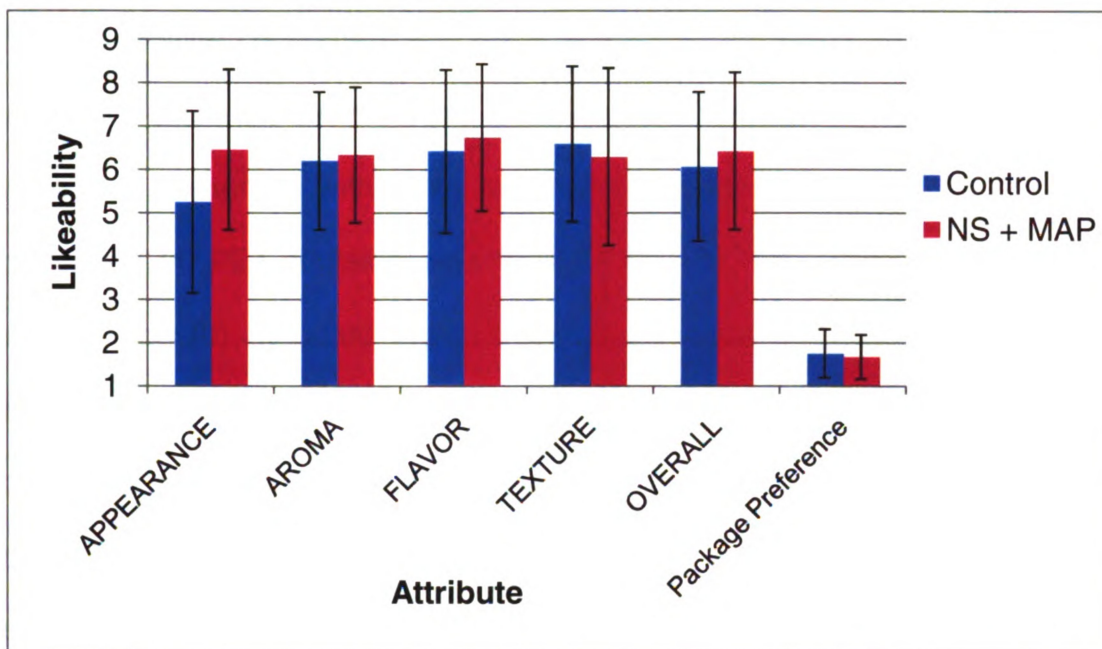
As a final study, a sensory test was conducted on non MAP and MAP pears after a storage time of 10 days. This storage time was selected because of the loss of the physical quality of non MAP pears after day 10. One hundred consumers participated in the study by answering a series of questions which were followed by several supplemental questions relating to packaging preference. The questions were all based on the products they tasted and packages they visually studied. Since MAP was found to have the best pears these were compared to a set of freshly cut pears (<12 hours). The following table (Table 15) presents the average values, the p-values, and significant differences between the control pears (fresh) and MAP pears (10 day storage), from the consumer study.

**Table 15: Summary of Mean scores and p-values for Control vs. NS + MAP pears based on various attributes at 10 days of storage**

<b>Attribute</b>	<b>Control</b>	<b>NS + MAP</b>	<b>P-Value</b>	<b>Sig</b>
APPEARANCE	5.25	6.46	0.0001	***
AROMA	6.2	6.34	0.5309	NS
FLAVOR	6.42	6.74	0.2067	NS
TEXTURE	6.59	6.3	0.2866	NS
OVERALL	6.07	6.43	0.1509	NS
Package Preference	1.76	1.68	.456	NS

*p < .05 is significantly different*

From the sensory test it can be seen that in terms of the aroma, flavor, texture and overall acceptability there was no significant difference between the two pear samples. From the scale used, a value of 9 referred to “Like Extremely” and a value of 1 referred to “Dislike Extremely.” The appearance parameter was the only characteristic to show a significant difference, in that the consumer panelists preferred the NS + MAP pears over the control pears, since it had a higher mean score (Table 15 and Figure 26). The results were very positive in that the consumers, between the ages of 18-60 (all Michigan State University students or faculty) overall liked both samples fairly equally. This means that the MAP pears (which were in storage for 10 days) tasted very similar to the freshly cut pears and there wasn’t a preference of one over the other. Figure 26 is a visual comparison of sensory attributes between the control pears and NS + MAP pears.



**Figure 26: Attribute comparison between Control and NS + MAP pears**

The goal of any researcher is to develop a product that tastes and looks as similar to the freshly cut product, which for this study was found to be true for the taste characteristic. The appearance was the only attribute which was found to be significantly different between the pears; however, this should be expected since it is difficult to keep a product looking as fresh as the moment it was cut. The mean score for the MAP pears was 6.46, which is between “like slightly” and “like moderately.” Thus, consumers did not dislike the product appearance totally, and might not reject the product based only on appearance.

One of the supplemental questions asked was: “In what form do you typically eat pears?” The options listed were: Fresh, Frozen, and Canned, of which >75% consumers, between the ages of 18-60, preferred to eat pears fresh

(Appendix IV). It is helpful to know this information, because it shows that finding ways to provide consumers with fresh pears (with an extended shelf-life) is very important, since it is the most preferred way in which they are eaten. Consumers were also presented with three package types and asked to pick which they preferred: (1) PET container with Snap on Lid, (2) PET packaging with PET film sealed, and (3) Standard Ziploc bag. The overall preference was split fairly evenly between options 1 and 2. Although consumers really liked the ability to peel off a seal and throw it away, they also wanted to be able to eat half of the pears in one sitting and consume the rest later, thus the snap on lid feature was preferred (Table 15).

Overall, it was found that the MAP pears did not significantly differ from the fresh control in any of the attributes except for appearance. The fresh control did not have as good of an appearance as compared to the MAP pears. Since the appearance is the first attribute available to consumers, it is very important to provide the freshest looking pears. Overall, the taste was found to be similar, which meant that as far as MAP is concerned, it helped to preserve the flavor of the product, which means the consumer panel liked the MAP pears for both their appearance and flavor.

## CONCLUSION

The preliminary results showed that the process treatments (NS and SAS) resulted in significant differences between the pear samples in the areas of O<sub>2</sub> levels, color a\* value, pH, bacterial growth, and yeast/mold growth (p-values of .0036, <.0001, and .0006, <.0001, and .0006, respectively). In regards to MAP, significant differences were found between the MAP and non MAP pears samples in the areas of headspace gases, color a\* value, bacterial growth, and yeast/mold growth (p-values of <.0001, <.0001, .0053, <.0001, and .0018, respectively). The MAP pears passively created an optimal headspace gas concentration which resulted in less microbial growth as compared to non MAP pears (World Food Science, 2009).

Preliminary testing showed there was a significant amount of browning and discoloration (colorimeter results and physical observation) in the SAS samples, regardless of MAP. The SAS treatment caused the pigments to leach from the peel to the flesh of the pears, and a great deal of tissue softening, neither of which occurred in the NS pears. Even though there was some microbial growth on the SAS pears, the SAS treatment did act as an inhibitor to the growth of microbes after several days of storage. By the completion of the initial storage study, the SAS treated pears were in very poor physical conditions and thus, SAS was removed from the study due to its effect on the physical quality of the pears.



NS pear samples (preliminary tests) showed a very slight amount of browning, yet overall they maintained the freshest and greenest color. The pears had very similar headspace gas levels to the SAS pears, both in the MAP and the non MAP. The pH was slightly higher in NS pears, while the TSS was very similar to SAS pears. The TA was higher in the NS no MAP sample; however, there was no change at all in the NS+MAP sample. Despite having the freshest visual characteristics, the NS no MAP pears had the highest number of bacteria, yeast and mold, while the NS + MAP pears in comparison had much lower microbial counts over the 21 day period.

Final test results found that MAP versus non MAP pear samples had results that were significantly different in the areas of CO<sub>2</sub> and O<sub>2</sub> levels, color L\*, a\* and b\* values, bacterial growth, and yeast/mold growth (p-values <.0001 for headspace and color, .0109, and .0001, respectively). No significant differences were seen in the samples in pH, TSS and TA even though there were slight variations. Passive MAP created an optimal headspace gas composition (World Food Science, 2009) which allowed for the extension of shelf-life, by maintaining the physical quality of the pear samples. Sensory test results from 100 consumers concluded that MAP pears had no significant difference in aroma, flavor, texture and overall acceptability compared to the fresh control which had been prepared within 12 hours of testing. Thus, MAP pears stored over a 10 day period tasted the same or very similar to control pears prepared fresh. Appearance was the only attribute to significantly differ between the MAP and Control samples, where NS + MAP pears were found to appear fresher than the

control. The pH, TSS and TA tend to vary and change as metabolic reactions occur. This means that since there were no significant differences between the pH, TSS and TA, it is likely that there was no significant differences in the metabolic reactions that were occurring between the NS no MAP and NS + MAP pears.

Overall, MAP in combination with NS proved to extend the shelf life of the fresh-cut pears for up to 21 days, while maintaining good color quality, which is vital to consumers. Microbial growth was also found to be significantly less in MAP pears, thus reducing microbial decay and potential safety concerns associated with micro-organisms. Fresh-cut MAP pears were found to be comparable to freshly prepared samples after 10 days of storage. Thus, fresh cut pears, treated with Nature Seal had a shelf life of ~10 days in a passive modified atmosphere package. This would allow consumers the convenience of purchasing a ready to eat, sliced pear to satisfy their lifestyle needs.

A recommendation for future research would be to use a smaller concentration of SAS to see if the effects on the physical quality are less detrimental. It would also be helpful to see how 21 day MAP pears compared to freshly prepared pears using a consume panel. Combining NS and SAS as a treatment could also provide interesting results depending on the compatibility of the two treatments individually. Since both treatments provide benefits separately it would be interesting to see if they could be combined to provide an even better quality for the pears.

## **APPENDIX I – Materials for Processing**

- Lab coats, and gloves for each participant helping (4-5 students)
- Pears, SAS (only in the preliminary study), NS, Fruit and Vegetable Wash
- PET Packages
- PET Lids
- Multiple Gallons of Distilled water (bought from University General Store)
- Stainless Steel Knife (sanitized in 1 gallon Distilled water with 1 cup Clorox® bleach)
- 2- 17" Spatula for stirring treatment mixtures
- 2 Paper towel rolls
- Timer/Watch
- Aluminum Foil Trays ( 20.5" x 13")
- 5 Buckets (7.5 quart square bucket for NS treatment, 7.5 quart square bucket for SAS treatment, 2- 10 quart buckets for Fruit and Vegetable Wash treatment: one with holes drilled in bottom and one without, and one 10 quart bucket for pear cores)
- Fresh Produce Net Bags (mesh)
- Maestro Cutting System (Wedger/Corer) from North Star Engineered Products
- Walk in Cooler set at 40°F (Room 111B in FSHN)
- Two shelf cart with wheels for transporting between buildings

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## **APPENDIX II- Procedure for Titratable Acidity**

For titratable acidity there were multiple steps that needed to be completed before testing any of the samples for TA level. The first step was to prepare all necessary solutions. To make 0.1% Normality NaOH (Sodium Hydroxide) solution the following steps were followed: (1) Weigh out 4 grams of NaOH pellets and dissolve into 900 mL of distilled water in a 1000 mL beaker. (2) Mix thoroughly and top off with more distilled water until the beaker has reached 1000 mL and mix again. (3) Cover solution with aluminum foil and set aside for 30 minutes before any use. To make the 0.1% Normality HCl (Hydrochloric Acid) solution the following steps were used: (1) Take 8.26 mL of HCl and dissolve in 500 mL of distilled water in a 1000 mL Erlenmeyer flask. (2) After mixing thoroughly top off flask with an additional 500 mL of distilled water giving 1000 mL of solution and mix thoroughly, making sure to cover with aluminum foil once prepared. The final solution to be made was the 1% phenolphthalein indicator. (1) Dissolve 1g of phenolphthalein in 100 mL of ethanol. (2) After dissolving thoroughly put solution in a stoppered bottle, using an eye dropper when needed. The 0.1% N NaOH solution was made weekly and the 0.1% N HCl was made every 2 weeks.

After preparing the solutions the next step was to standardize in order to determine the normality of the NaOH and HCl solution. For the standardization 3 plastic cups were filled with 10 mL of HCl. A 10 mL cotton plugged glass pipette fitted with a rubber bulb was used to carefully measure out 10 mL of HCl in each cup. Prior to filling each cup the pipette was rinsed with HCl solution twice into a

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waste cup. After preparing the 3 sample HCl cups, 5 drops of the phenolphthalein were carefully added to each cup and set aside. Next the 50 ml burette was closed at the bottom and 50 mL of NaOH was added to the burette, once filled, the solution was released into a different waste cup. This process was repeated in order to rinse the burette. After rinsing the burette it was again closed at the bottom and filled 50 mL of NaOH solution ready to be used in the standardization process. Next, the first sample cup with HCl and phenolphthalein solution was set underneath the burette of NaOH solution. Drop by drop the NaOH was titrated into the cup until the entirety of the solution turned a pink/fuchsia color and remained for at least 30 seconds. The amount of NaOH used to neutralize the solution in the cup was recorded and then plugged into the normality equation to ensure that the normality of NaOH remained .1N.

This process was repeated with the remaining two cups of HCl with phenolphthalein in order to give a triple check. After the preparation of all 3 solutions and the standardization, the actual test was ready to be conducted. Using the same juice samples from the pH and TSS tests the following process was used to conclude how much NaOH it took to neutralize the pH of the juice samples to a pH of 8: (1) Take 10 mL of juice sample and combine with 100 mL of distilled water in a 200 mL beaker. (2) Keep beaker over burette with NaOH, turn on pH meter and calibrate using pH buffer 4, 7, and 10. Once calibrated take pH wand and dip into the beaker and mix around making sure not to touch sides of beaker. While stirring the mixture, carefully release the NaOH drop by drop into the beaker, making sure not stop stirring. (3) As each drop is added,

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carefully monitor the pH meter and stop adding NaOH once the beaker solution has been neutralized to pH 8. At this point note the difference in volume from the beginning of the NaOH addition to the end of its addition. This same process was used for all treatments and replicates and the difference in volume was noted for each. After attaining the amount of NaOH used to neutralize each sample the total acids for each sample was calculated. This method was followed on each of the testing periods of Day 0, 7, 14 and 21.

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### **APPENDIX III- Microbiological Method**

Microbiological testing was done to determine total count amount of bacteria, yeast and mold on the pears. The process consisted of several steps that required the very sanitary environment. The process for microbiology can be split into 2 major parts: Preparing media/pouring plates and Plating Samples. The preparation of the media was usually done 1 week prior to testing and once petri dishes were filled with agar they were put into cold storage. Two types of media were prepared; TSA-YE (Trypticase Soy Agar) and PDA (Potato Dextrose Agar). TSA-YE was used to identify the amount of bacteria growing, while the PDA was used to show the amount of yeast and mold present. Prior to any preparation of any sort, workspace and most utensils were sprayed with 70% Ethanol wash and dried off. The microbiology portion of the study was done in triplicates to provide accurate amounts of samples to look at.

To make the TSA-YE agar the following steps were followed: (1) Take 1 L of distilled water and a magnetic stir bar in a 1000mL Erlenmeyer flask and dissolve 40 grams of TSA-YE powder as well as 6 grams of Yeast Extract. Place the flask on the hot plate and turn both the heat and stir power to maximum levels and allow the solution to come to a boil. Cover the top of the flask with foil paper labeled with the letters TSA-YE on autoclave tape. Carefully monitoring the solution, as soon as bubbles start to form, remove the flask from the heat. Set TSA-YE solution aside in a tray. At the same time that the TSA-YE media was being prepared the PDA was also prepared. The steps were identical except for the powder going into the second flask was 39 grams of PDA powder.

As both solutions came to a boil and were taken off the heat, they were set aside in a tray, awaiting the autoclave. During this time another liter of each media was made to give a total of 2 liters of both TSA-YE and PDA. Once all agar was prepared it was placed into the autoclave and left for a period of one hour to allow for proper sterilization of agar.

Following the autoclave process, agar was carefully removed with heavy duty heat resistant mitts and placed into water baths set at 55°-57°C for a period of 30 minutes to allow it to be cooled down from the autoclave temperature. After the 30 minute period the PDA flasks were taken out and set onto hot plates, turning on ONLY the stir power. At this point 70 ppm of Ampicillin (which was prepared by lab personnel) was carefully added to each of the flasks, making sure to use a pipette and slowly adding it into the side of the flask not splashing or adding the solution too quickly, the agar was allowed to be stirred for a couple minutes to allow accurate dispersion of the Ampicillin. At this point, 200 empty petri dishes were taken out of bags and stacked 10 high. A bunsen burner was lit in order to flame the top of the flasks before pouring agar into plates. Plates were quickly and carefully poured to fill two thirds of each petri dish. The flask was also flamed on top after ever 10 or so dishes were filled. Due to the gelatinous nature of the media in solidified form, plates had to be poured as quickly as possible, therefore, while PDA plates were filled, the TSA-YE remained in the water bath.

Once all PDA plates were filled they were allowed to sit and harden overnight. Next, 200 empty petri dishes were taken out for the pouring of TSA-

YE plates. The bunsen burner was left on in order to also flame the tops of the TSA-YE flasks. The TSA-YE flasks were then taken out of the water bath one at a time and again plates were quickly and carefully poured and set aside. Once the entire agar had been poured out into petri dishes it was set out to dry overnight. There was no need to individually label each plate because once hardened, the TSA-YE plates turned into a dark yellow color and the PDA plates turned into a white opaque color.

The next step in the microbiology process consisted of actually plating samples. For this part, 3 packages of each treatment were taken out of storage at each of the testing points; Day 0, 7, 14, and 21. Once again the entire workspace was cleaned and sanitized including all materials such as the pipettes, weight boats, balance, and knife. Before starting the required numbers of plates needed were taken out of storage and allowed to warm up to room temperature. They were also set upside down to prevent condensation from dripping onto the agar. The following steps were followed for actually plating samples: Light bunsen burner and keep on for entire time of plating. Take the balance and place one weight boat container and tare the weight. Take the sterilized knife and cut anywhere between 2 and 3 slices into cubes in order to give 25 grams of sample. Measure out 100 mL of Phosphate buffer solution (PBS) in a graduated cylinder, making sure to flame the top of the PBS jar prior to each use. Take a whirl-pack bag and label with treatment 1, 2, 3 or 4 and package number 1, 2, or 3. Then add the PBS as well as the 25 grams of pear sample and seal the bag by folding the top of the bag over and folding the tabs

into place. Once sealed, agitate the bag by hand for a period of 60 seconds. Once this step is completed, repeat the same steps for all other sample treatments and packages making sure to have a total of 12 packages properly agitated and labeled. It is very important to properly sanitize the knife and each new weigh boat between each sample to prevent any cross contamination. Weigh boats can be discarded after each 25 gram sample is weighed out. After all steps are completed, take all the bags and set them aside as the plates are labeled. For this next part, each petri dish was labeled with which package and which treatment it pertained to. For example: Treatment 1, Package 1, 2 or 3. Then petri dishes were set aside, two for each dilution, and two per media type. The dilutions selected for this study ranged from 0, -1, -2, and -3. This gave 8 petri dishes of PDA and 8 of TSA-YE, all together giving 16 plates for each package of each treatment. A total of 48 plates were then used for each treatment. With the four treatments this amounted to 192 plates and with four testing days that gave a grand total of 768 plates for the entirety of the microbiology tests.

The next step was to take a test tube filled with 9000  $\mu$ L of PBS and pipette 1000  $\mu$ L of the solution from the treatment 1, package 1 whirlpack bag. This first test tube would be dilution -1. This process was done using autoclaved pipette tips which were discarded when going between dilutions. Next, 1000  $\mu$ L of dilution -1 were pipetted out and put into the second test tube which became dilution -2. Again the tip was discarded here and a new tip was applied which was used to pipette 1000  $\mu$ L out of the second test tube and added to a third test

tube which became dilution -3. Each time any solution was added to a test tube from another it was properly mixed by being placed on a Vortex test tube agitator before being diluted further. The original whirl pack bag with the sample and buffer solution was considered as Dilution 0. After all dilutions were prepared, they were ready to be plated accordingly. A 100  $\mu$ L pipette was taken and 100  $\mu$ L of each dilution were plated onto its corresponding plate. After 100  $\mu$ L of each dilution was added to a petri dish it was placed on a plate spinner. An L-spreader was then used to evenly distribute the 100  $\mu$ L solution onto the agar by simultaneously spinning the plate spinner and moving the L-spreader back and forth. As each dilution was plated the pipette tips were again discarded in between. Dilutions were plated two plates at a time, from 0, -1, -2, and -3 to avoid outside any environmental contamination. Each whirl pack bag with the sample was discarded last, in case any dilutions had to be redone.

Once plated, petri dishes were allowed to sit and dry for 15 minutes. After this time, all plates were flipped upside down in order to have the lids on the bottom. All TSA-YE plates were put on a try and put into the incubator which was set at 29°C. The plates were then left for 24 hours and examined the next day. All PDA plates were set out on a tray as well, but were allowed to sit in room temperature conditions and were checked on 2-3 days later, once checked and counted plates were discarded. Each week the same process was followed for plating and labeling petri dishes. Since enough plates had been poured with agar initially to last two weeks, the next batch of media was made at the third week and plates were again poured and set aside for the final two testing days.

After each testing day all materials were properly rinsed or set aside for waste so they could be autoclaved. PDA and TSA-YE trays were examined each testing day. The number of colonies formed was counted on each plate of each dilution and the numbers were recorded.



## **APPENDIX IV- Sensory Procedure**

Before consumers looked at the packages the following steps were completed to start the test: Trays lined with paper were set up with a Dixie cup of distilled water, a spit cup, a napkin, and two plates labeled with a 3 digit code pertaining to each of the two treatments (Control and NS + MAP). The codes were in place to keep treatments anonymous. The SIMS questionnaire was also activated the morning of the test to allow each consumer to electronically evaluate all samples. Each tray with materials was set out by the booth and when a consumer had filled out a consent form and was ready to start testing, they would be pointed to a booth where they had to slide a card saying READY underneath the booth window. Then immediately two slices of each treatment was put on the proper plates and samples were sent through the window. When a consumer was done answering questions regarding the samples as well as packages on their side of the booth, a card with the word FINISH was slid underneath the booth window. At this point the window was pushed up and everything on the tray was taken back and discarded and the tray was reused for the next set of consumers.

Treatment samples were kept on ice in order to keep the temperatures cool and not allow them to warm up to room temperature. This also helped decreased the amount of variability of the sample temperature. Multiple students helped in the sensory test in setting up sample trays and placing samples on plates when ready to be tested. At the completion of a test, a student standing outside the room provided each consumer with their choice of flavor for the ice

cream cup incentive. The same testing procedure was followed for all consumers and student assistants had to make sure that a consent form was signed by each panelist, the right samples were on the properly coded plate, the used samples were discarded and an incentive was given out.

## Pear Questionnaire

You will be provided with two pear samples. Please follow the directions on the screen and answer the associated questions as you try the samples one at a time. Upon receiving the samples, please continue with this test.

How do you like the **APPEARANCE** of the sample?

REQUIRED: Please comment on the color and apparent freshness of the sample based on appearance.

How do you like the **AROMA** of the sample?

How do you like the **TEXTURE** of the sample?

REQUIRED: Please comment on the texture of the sample.

How do you like the **FLAVOR** of the sample?

How do you like the **OVERALL ACCEPTABILITY** of the sample?

How likely would you be to purchase this product as an on-the-go snack?

Convenience is an important factor in choosing my snacks.

How often do you eat pears?

In what form do you typically eat pears? **Fresh, Frozen or Canned?**

Now, please look at the three packages that you have been presented with, but

**DO NOT** open the packages.

Which of the three packages do you prefer?

REQUIRED: Why?

Do you know what modified atmosphere packaging is?

Modified atmosphere packaging is a technique used to prolong the shelf life period of fresh products by slowing down the natural deterioration of the product by adjusting the package material, dipping product in a safe combination of vitamins and minerals or adjusting the storage temperature.

**Sample 492** is a modified atmosphere package. Does this change your preference?

If so, why?

Are you **Male** or **Female**?

Please indicate into which of the following age categories you fall: (Age group options)

You have completed the evaluation. Thank you for your time. Please remember to pick up your reward as you leave.

Most Questions required a rating of 1 “**Like Extremely**” to 9 “**Dislike Extremely**”

Some questions required YES/NO answers.

**Consumer Consent Form**

**Fresh Pear Slices-Value Added Venture: Varieties, Processing Models &**

**Consumer Acceptance”**

**Department of Food Science and Human Nutrition and School of Packaging  
Michigan State University**

**Sample: Pear Slices**

Before you decide to sign this consent form and participate in our study, please read carefully and thoroughly the reverse side of this form for the sample ingredients and preparation information, purpose and procedure of this study, potential risks and benefits from your participation, our assurance of your privacy, your rights as a human subject in our study, etc.

Your responses are collected anonymously. We have no way to connect you, as an individual, to the completed survey form. You are free to not answer any question you choose, but please try to answer every question. Incentives will be provided for a good faith effort to participate and complete the questionnaire.

If you have any question during your reading this consent form, or during or after your participation, please do not hesitate to contact the on-site sensory evaluation leader and/or the principle investigator. Feel free to contact Dr. Janice Harte, the principle investigator of this study, via phone at 517-355-8474, ext. 105 (114 Trout Food Science and Human Nutrition Building, Michigan State University, East Lansing, MI 48823). You also can reach her via email at

harteja@msu.edu for any inquiry you might have due to your participation in our study.

If you have questions or concerns about your role and rights as a research participant, would like to offer information or offer input, or would like to register a complaint about this study, you may contact, anonymously if you wish, the Michigan State University's Human Research Protection Program at 517-355-2180, Fax 517-432-4503, or e-mail [irb@msu.edu](mailto:irb@msu.edu) or regular mail at 202 Olds Hall, MSU, East Lansing, MI 48823.

PARTICIPATION IN THIS STUDY IS VOLUNTARY. PLEASE NOTE UPON YOUR SIGNING THIS CONSENT FORM, YOU VOLUNTARILY AGREE TO PARTICIPATE IN OUR STUDY. YOUR SIGNATURE INDICATES YOU HAVE READ THE INFORMATION PROVIDED ABOVE AND THAT YOU HAVE HAD AN ADEQUATE OPPORTUNITY TO DISCUSS THIS STUDY WITH THE PRINCIPLE INVESTIGATOR AND HAVE HAD ALL YOUR QUESTIONS ANSWERED TO YOUR SATISFACTION. YOU WILL BE GIVEN A COPY OF THIS CONSENT FORM WITH YOUR SIGNATURE FOR YOUR RECORDS UPON YOUR REQUEST.

SIGNED \_\_\_\_\_

DATE \_\_\_\_\_

## Consent Form

### Fresh Pear Slices- Value Added Venture: Varieties, Processing Models &

### Consumer Acceptance

**Department of Food Science and Human Nutrition and School of Packaging  
Michigan State University**

#### INVITATION TO PARTICIPATE

You are invited to participate in this study that assesses the quality attributes of sliced pears.

#### PURPOSE OF THIS STUDY

This study is intended to study the consumer acceptability of pear slices dipped in an anti-browning ingredient. Texture, appearance, aroma and flavor characteristics of pear slices will be evaluated.

#### PROCEDURE OF THIS STUDY

Each participant will be presented with pear slices and will be asked to **evaluate the pears visually and after tasting**, score the attributes as presented on the score sheet for each sample. Samples will be presented using three digit random codes. Consumer marketing questions will also be asked.

#### SAMPLE INGREDIENTS AND SAMPLE PREPARATION

All the ingredients used in our samples are food-grade and FDA approved for foods. **The ingredients are: pear slices dipped in a solution of a FDA approved ingredient for color preservation called NatureSeal (calcium ascorbate).**

### **POTENTIAL RISKS**

Because all ingredients we use in our study are food grade and FDA approved for food applications, these samples pose no adverse health risk, provided the subject has not been identified as being susceptible to an allergic reaction to the previously listed sample ingredients. If you believe there is a potential of an **allergic reaction upon sniffing and tasting**, notify the on-site sensory evaluation coordinator and/or principle investigator immediately. You will be released from participating in this study. Please note if you are injured as a result of your participation in this study, Michigan State University will provide emergency medical care, if necessary, but this and any other medical expense must be paid from your own health insurance program.

### **POTENTIAL BENEFITS**

There are no benefits gained directly from your participation in this study. However, your participation and response will provide us valuable data, which can be used to identify optimum sliced pear storage conditions and help bring a new product to market.

### **ASSURANCE OF CONFIDENTIALTY**

Any information obtained in connection with this study that could be identified with you will be kept confidential by ensuring that all consent forms are securely stored. All data collected and analyzed will be reported in an aggregate format that will not permit associating subjects with specific responses or findings. **Your privacy will be protected to the maximum extent allowable by law.**



### **WITHDRAWAL FROM THIS STUDY**

Participation in this study is voluntary. Your decision to refuse participation or discontinue participation during this study will be honored promptly and unconditionally.

**ADVERTISEMENT**

Do you eat pears? Participate in an evaluation of a fresh sliced pear product!



An ice cream treat will be given for your participation!

Where? Room 101 G.Malcolm Trout Building  
(corner of Wilson and Farm Lane)

When? **Thursday, February 18th, 2010**  
**10 am-1pm**  
(or until 100 survey's are completed)



For any questions contact Janice Harte in the Department of Food Science and Human Nutrition Building, [harteja@msu.edu](mailto:harteja@msu.edu) 355-8474, ext. 105

## APPENDIX V- Permeability Results (O<sub>2</sub>, CO<sub>2</sub>, and WVTR)

Tables 16-20 represent the results from the OTR, WVTR and CO<sub>2</sub>TR for both the container and film.

**Table 16: OTR of PET Lidding Film**

<b>OTR of Lidding Film</b>	<b>Units (cc/ [m<sup>2</sup>-day])</b>
Average for Cell A	59.60285
Average for Cell B	59.00276
Average of A and B	<b>59.30281</b>
S.D	0.424328

**Table 17: WVTR of PET Lidding Film**

<b>WVTR of Lidding Film</b>	<b>Units (gm/ [m<sup>2</sup>-day])</b>
Average for Cell A	9.148077
Average for Cell B	9.125281
Average of A and B	<b>9.136679</b>
S.D	0.016119

**Table 18: OTR of PET container with Lidding Film**

<b>OTR of PKG and Film</b>	<b>Units (cc/ [pkg-day])</b>
Average for Cell A	0.119946
Average for Cell B	0.141826
Average of A and B	<b>0.130886</b>
S.D	0.015471

**Table 19: CO<sub>2</sub>TR of PET container with Lidding Film**

<b>CO<sub>2</sub>TR of PKG and Film</b>	<b>Units (cc/ [pkg-day])</b>
Average for Cell A	0.251611
Average for Cell B	0.893424
Average of A and B	<b>0.572517</b>
S.D	0.45383

**Table 20: WVTR of PET container with Lidding Film**

<b>WVTR of PKG and Film</b>	<b>Units (gm/ [pkg-day])</b>
Average for Cell A	0.297679
Average for Cell B	0.375247
Average of A and B	<b>0.336463</b>
S.D	0.054849

## APPENDIX VI- Preliminary Results In Depth

The following Appendix includes all data tables from which graphs were created for the results/discussion section of the thesis. Data tables have the average values listed, which were compiled from the raw data.

**Table 21: Average values for Headspace CO<sub>2</sub>-Set 1**

	CO <sub>2</sub> (%)			
	Day 0	Day 3	Day 7	Day 10
T1	0.0	0.3	0.2	0.2
T2	0.0	10.9	22.2	26.2
T3	0.0	0.2	0.3	0.3
T4	0.0	11.0	21.1	27.7

**Table 22: Average values for Headspace CO<sub>2</sub>-Set 2**

	CO <sub>2</sub> (%)			
	Day 0	Day 3	Day 7	Day 10
T1	0.0	0.3	0.5	0.4
T2	0.0	11.2	23.7	29.6
T3	0.0	0.3	0.5	0.4
T4	0.0	11.5	28.1	36.3

**Table 23: Average values for Headspace O<sub>2</sub>-Set 1**

	O <sub>2</sub> (%)			
	Day 0	Day 3	Day 7	Day 10
T1	20.9	20.9	20.8	20.9
T2	20.9	11.8	0.0	0.0
T3	20.9	20.5	20.7	20.5
T4	20.9	11.8	4.1	0.0

**Table 24: Average values for Headspace O<sub>2</sub>-Set 2**

	O <sub>2</sub> (%)			
	Day 0	Day 3	Day 7	Day 10
T1	20.9	20.7	20.7	20.6
T2	20.9	7.6	0.0	0.0
T3	20.9	20.5	20.5	20.5
T4	20.9	11.6	0.3	0.0

**Table 25: Average values for Headspace Gas Concentrations (Both Sets)**

Day	0	3	7	10		0	3	7	10
	CO <sub>2</sub> (%)					O <sub>2</sub> (%)			
<b>T1: NS no MAP</b>	0.0	0.3	0.3	0.3		20.9	20.8	20.7	20.7
<b>T2: NS + MAP</b>	0.0	11.0	23.0	27.9		20.9	9.7	0.0	0.0
<b>T3: SAS no MAP</b>	0.0	0.2	0.4	0.3		20.9	20.5	20.6	20.5
<b>T4: SAS + MAP</b>	0.0	11.2	24.6	32.0		20.9	11.7	2.2	0.0

**Table 26: Standard Deviation for Headspace Gas Concentrations (Both Sets)**

Day	0	3	7	10		0	3	7	10
	CO <sub>2</sub> (%)					O <sub>2</sub> (%)			
<b>T1: NS no MAP</b>	0.0	0.1	0.2	0.1		0.0	0.2	0.2	0.2
<b>T2: NS + MAP</b>	0.0	1.4	2.3	3.5		0.0	6.6	0.0	0.0
<b>T3: SAS no MAP</b>	0.0	0.0	0.2	0.1		0.0	0.1	0.3	0.0
<b>T4: SAS + MAP</b>	0.0	0.7	4.2	5.1		0.0	0.6	2.3	0.0

**Table 27: Average values for Hunter Color over 10 day period**

<b>L* Comparison</b>									
	<b>Round 1</b>					<b>Round 2</b>			
Day	T1	T2	T3	T4		T1	T2	T3	T4
0	69.31	49.37	57.08	62.26		56.87	56.64	50.89	43.68
3	67.13	62.05	41.23	52.65		45.25	47.28	41.60	58.98
7	57.96	64.85	31.07	26.70		59.61	49.03	42.82	39.98
10	55.40	65.02	44.90	32.90		64.17	49.68	51.72	45.26
<b>a* Comparison</b>									
	<b>Round 1</b>					<b>Round 2</b>			
Day	T1	T2	T3	T4		T1	T2	T3	T4
0	0.73	0.54	0.28	0.67		0.59	1.36	1.16	1.63
3	0.62	-0.27	3.37	2.34		0.87	1.41	3.84	3.54
7	2.07	0.85	4.03	3.18		4.27	1.76	4.78	5.17
10	1.57	1.25	7.01	3.31		4.11	-0.32	6.46	5.90
<b>b* Comparison</b>									
	<b>Round 1</b>					<b>Round 2</b>			
Day	T1	T2	T3	T4		T1	T2	T3	T4
0	21.25	20.27	21.94	24.18		19.61	24.35	19.34	18.41
3	23.84	21.60	22.25	23.93		20.35	22.47	21.53	25.89
7	22.36	21.82	16.95	13.59		24.53	20.01	21.62	18.44
10	17.53	21.25	26.62	19.88		24.89	15.97	27.36	26.50

**Table 28: Average values for Hunter Color over 10 day period (Data combined)**

<b>L Comparison</b>				
Average of Both Sets				
Day	T1: NS no MAP	T2: NS + MAP	T3: SAS no MAP	T4: SAS + MAP
0	63.09	53.01	53.98	52.97
3	56.19	54.67	41.42	55.82
7	58.78	56.94	36.95	33.34
10	59.78	57.35	48.31	39.08
<b>a Comparison</b>				
Average of Both Sets				
Day	T1: NS no MAP	T2: NS + MAP	T3: SAS no MAP	T4: SAS + MAP
0	0.66	0.95	0.72	1.15
3	0.75	0.68	3.61	2.94
7	3.17	1.30	4.40	4.18
10	2.84	0.46	6.74	4.61
<b>b Comparison</b>				
Average of Both Sets				
Day	T1: NS no MAP	T2: NS + MAP	T3: SAS no MAP	T4: SAS + MAP
0	20.43	22.31	20.64	21.30
3	22.10	22.04	21.89	24.91
7	23.45	20.92	19.28	18.75
10	21.21	18.61	27.30	23.24

**Table 29: Average values for pH over 1 week period**

	<b>pH Set 1</b>			<b>pH Set 2</b>	
	Day 0	Day 7		Day 0	Day 7
<b>T1</b>	4.2	4.2	<b>T1</b>	3.8	3.9
<b>T2</b>	3.9	4.1	<b>T2</b>	3.7	4.0
<b>T3</b>	3.8	3.8	<b>T3</b>	3.4	4.0
<b>T4</b>	3.7	3.9	<b>T4</b>	3.4	3.8



**Table 30: Average values and Standard Deviation for pH over 1 week period (Data combined)**

	Average			Standard Deviation	
	Day 0	Day 7		Day 0	Day 7
T1: NS no MAP	4.0	4.1	T1: NS no MAP	0.3	0.2
T2: NS + MAP	3.8	4.1	T2: NS + MAP	0.1	0.1
T3: SAS no MAP	3.6	3.9	T3: SAS no MAP	0.2	0.3
T4: SAS + MAP	3.5	3.8	T4: SAS + MAP	0.2	0.1

**Table 31: Average values for TSS over 1 week period**

	TSS Set 1			TSS Set 2	
	Day 0	Day 7		Day 0	Day 7
T1	13.6	13.5	T1	12.1	13.3
T2	12.6	13.5	T2	12.9	13.6
T3	13.3	13.7	T3	12.9	13.6
T4	12.9	13.3	T4	13.1	13.7

**Table 32: Average values and Standard Deviation for TSS over 1 week period (Data combined)**

	Average			Standard Deviation	
	Day 0	Day 7		Day 0	Day 7
T1: NS no MAP	12.8	13.4	T1: NS no MAP	1.3	0.5
T2: NS + MAP	12.8	13.5	T2: NS + MAP	0.6	0.1
T3: SAS no MAP	13.1	13.6	T3: SAS no MAP	0.3	0.2
T4: SAS + MAP	13.0	13.5	T4: SAS + MAP	0.2	0.3

**Table 33: Average values for TA over 1 week period**

	TA Set 1			TA Set 2	
	Day 0	Day 7		Day 0	Day 7
T1	0.20	0.22	T1	0.17	0.22
T2	0.18	0.18	T2	0.18	0.18
T3	0.18	0.22	T3	0.18	0.18
T4	0.15	0.20	T4	0.18	0.18

**Table 34: Average values and Standard Deviation for TA over 1 week period (Data combined)**

	Average			Standard Deviation	
	Day 0	Day 7		Day 0	Day 7
T1: NS no MAP	0.18	0.22	T1: NS no MAP	0.03	0.02
T2: NS + MAP	0.18	0.18	T2: NS + MAP	0.02	0.02
T3: SAS no MAP	0.18	0.20	T3: SAS no MAP	0.02	0.03
T4: SAS + MAP	0.17	0.19	T4: SAS + MAP	0.03	0.02

**Table 35: Average and Standard Deviation for bacterial count in pears over 21 days**

	Average Values			
	1	7	14	21
<b>NS no MAP</b>	2.3	4.5	5.7	5.3
<b>NS + MAP</b>	1.5	2.9	1.5	4.3
<b>SAS no MAP</b>	0.0	0.0	4.2	5.3
<b>SAS + MAP</b>	2.2	0.0	0.0	2.8
	Standard Deviation			
<b>NS no MAP</b>	2.1	0.4	0.2	0.7
<b>NS + MAP</b>	2.7	2.5	2.6	1.0
<b>SAS no MAP</b>	0.0	0.0	0.8	0.6
<b>SAS + MAP</b>	1.9	0.0	0.0	2.6

*Units are in log (CFU g<sup>-1</sup>)*

**Table 36: Average and Standard Deviation for yeast & mold count in pears over 21 days**

	Average Values			
	1	7	14	21
<b>NS no MAP</b>	0.0	4.5	5.3	5.3
<b>NS + MAP</b>	0.0	3.4	3.2	0.0
<b>SAS no MAP</b>	0.0	0.0	2.9	5.9
<b>SAS + MAP</b>	0.0	0.0	0.0	1.3
	Standard Deviation			
<b>NS no MAP</b>	0.0	0.4	0.9	0.6
<b>NS + MAP</b>	0.0	0.4	1.1	0.0
<b>SAS no MAP</b>	0.0	0.0	2.6	0.9
<b>SAS + MAP</b>	0.0	0.0	0.0	2.2

*Units are in log (CFU g<sup>-1</sup>)*

## APPENDIX VII- Final Results in Depth

The following Appendix includes all data tables from which graphs were created for the results/discussion section of the thesis. Data tables have the average values listed, which were compiled from the raw data.

**Table 37: Average values for Headspace CO<sub>2</sub>- Set 1**

	CO <sub>2</sub> (%)							
	0	1	3	7	10	14	17	21
T1: NS no MAP	0.0	0.1	0.3	0.4	0.3	0.2	0.4	1.2
T2: NS + MAP	0.0	2.9	6.4	21.3	21.5	23.5	29.6	38.1

**Table 38: Average values for Headspace CO<sub>2</sub>- Set 2**

	CO <sub>2</sub> (%)							
	0	1	3	7	10	14	17	21
T1: NS no MAP	0.0	0.1	0.4	0.4	0.3	0.2	0.3	1
T2: NS + MAP	0.0	3.0	8.5	19.6	22.0	26.1	31.1	32.6

**Table 39: Average values for Headspace O<sub>2</sub>-Set 1**

	O <sub>2</sub> (%)							
	0	1	3	7	10	14	17	21
T1: NS no MAP	20.9	20.9	20.5	20.5	20.5	20.5	20.8	20.4
T2: NS + MAP	20.9	16.8	12.5	0.0	0.0	0.0	0.0	0.0

**Table 40: Average values for Headspace O<sub>2</sub>- Set 2**

	O <sub>2</sub> (%)							
	0	1	3	7	10	14	17	21
T1: NS no MAP	20.9	20.8	20.5	20.4	20.6	20.6	20.75	20.5
T2: NS + MAP	20.9	16.9	9.3	0.0	0.0	0.0	0	0.0

**Table 41: Average and Standard Deviation for Headspace Gas Concentrations (Both Sets)**

	<b>0</b>	<b>1</b>	<b>3</b>	<b>7</b>	<b>10</b>	<b>14</b>	<b>17</b>	<b>21</b>
	<b>Average CO<sub>2</sub> (%)</b>							
<b>T1: NS no MAP</b>	0.0	0.1	0.3	0.4	0.3	0.2	0.3	1.1
<b>T2: NS + MAP</b>	0.0	2.9	7.4	20.5	21.7	24.8	30.3	35.3
	<b>Standard Deviation CO<sub>2</sub> (%)</b>							
<b>T1: NS no MAP</b>	0.0	0.0	0.1	0.1	0.0	0.0	0.1	0.1
<b>T2: NS + MAP</b>	0.0	0.2	2.5	1.5	1.5	1.8	1.6	3.5
	<b>Average O<sub>2</sub> (%)</b>							
<b>T1: NS no MAP</b>	20.9	20.8	20.5	20.5	20.6	20.5	20.8	20.4
<b>T2: NS + MAP</b>	20.9	16.8	10.9	0.0	0.0	0.0	0.0	0.0
	<b>Standard Deviation O<sub>2</sub> (%)</b>							
<b>T1: NS no MAP</b>	0.0	0.1	0.1	0.1	0.1	0.1	0.2	0.1
<b>T2: NS + MAP</b>	0.0	0.4	4.8	0.0	0.0	0.0	0.0	0.0

**Table 42: Average values for Hunter Color over 21 day period**

<b>L Comparison</b>				
<b>Round 1</b>		<b>Round 2</b>		
<b>Day</b>	<b>T1: NS no MAP</b>	<b>T2: NS + MAP</b>	<b>T1: NS no MAP</b>	<b>T2: NS + MAP</b>
0	76.82	76.77875	78.07	76.4025
3	79.25875	79.7925	80.24375	79.45375
7	77.39	80.24125	78.97	79.36625
10	75.82125	80.525	79.17875	80.735
14	73.58625	80.31875	75.3075	80.3375
17	71.71875	83.36625	72.88125	81.4425
21	56.95875	81.08625	60.595	80.61125
<b>a Comparison</b>				
<b>Round 1</b>		<b>Round 2</b>		
<b>Day</b>	<b>T1: NS no MAP</b>	<b>T2: NS + MAP</b>	<b>T1: NS no MAP</b>	<b>T2: NS + MAP</b>
0	-0.3425	-0.87125	-0.72125	-0.83625
3	-0.82	-0.90375	-0.56875	-0.365
7	1.39875	-0.575	0.835	-0.15625
10	2.1175	-0.4275	0.87375	-0.68875
14	2.30625	-0.44125	2.5875	-0.35125
17	2.7725	-0.3025	5.02125	-0.085
21	4.4825	-0.53375	3.78	-0.33625
<b>b Comparison</b>				
<b>Round 1</b>		<b>Round 2</b>		
<b>Day</b>	<b>T1: NS no MAP</b>	<b>T2: NS + MAP</b>	<b>T1: NS no MAP</b>	<b>T2: NS + MAP</b>
0	13.33125	13.37375	12.5225	13.965
3	11.78875	12.06375	12.33125	12.72625
7	15.61125	13.26	14.0825	14.07375
10	17.21	12.5775	14.1375	12.2225
14	18.24	13.3825	17.45625	12.995
17	21.0775	16.235	21.8775	17.20875
21	21.01625	12.5275	18.2925	13.71125

**Table 43: Average values for Hunter Color over 21 day period (Data combined)**

<b>L COMPARISON</b>		
<b>Day</b>	<b>T1: NS no MAP</b>	<b>T2: NS + MAP</b>
<b>0</b>	77.445	76.590625
<b>3</b>	79.75125	79.623125
<b>7</b>	78.18	79.80375
<b>10</b>	77.5	80.63
<b>14</b>	74.446875	80.328125
<b>17</b>	72.3	82.404375
<b>21</b>	58.776875	80.84875
<b>a COMPARISON</b>		
	<b>T1: NS no MAP</b>	<b>T2: NS + MAP</b>
<b>0</b>	-0.531875	-0.85375
<b>3</b>	-0.694375	-0.634375
<b>7</b>	1.116875	-0.365625
<b>10</b>	1.495625	-0.558125
<b>14</b>	2.446875	-0.39625
<b>17</b>	3.896875	-0.19375
<b>21</b>	4.13125	-0.435
<b>b COMPARISON</b>		
	<b>T1: NS no MAP</b>	<b>T2: NS + MAP</b>
<b>0</b>	12.926875	13.669375
<b>3</b>	12.06	12.395
<b>7</b>	14.846875	13.666875
<b>10</b>	15.67375	12.4
<b>14</b>	17.848125	13.18875
<b>17</b>	21.4775	16.721875
<b>21</b>	19.654375	13.119375

**Table 44: Average Values for pH over 21 day period**

	<b>Average Values</b>				<b>Average Values</b>			
	<b>Set 1</b>				<b>Set 2</b>			
	<b>0</b>	<b>7</b>	<b>14</b>	<b>21</b>	<b>0</b>	<b>7</b>	<b>14</b>	<b>21</b>
<b>T1: NS no MAP</b>	4.1	4.2	4.2	4.2	4.1	4.1	4.1	4.2
<b>T2: NS + MAP</b>	4.2	4.2	4.2	4.2	4.1	4.1	4.1	4.1

**Table 45: Average and Standard Deviation for pH over 21 day period (Data combined)**

	Average Values				Standard Deviation			
	0	7	14	21	0	7	14	21
<b>T1: NS no MAP</b>	4.1	4.1	4.2	4.2	0.1	0.1	0.1	0.0
<b>T2: NS + MAP</b>	4.2	4.1	4.1	4.1	0.1	0.1	0.1	0.1

**Table 46: Average values for TSS over 21 day period**

	Average Values				Average Values			
	Set 1				Set 2			
	0	7	14	21	0	7	14	21
<b>T1: NS no MAP</b>	13.9	13.4	13.4	13.5	13.6	13.2	13.3	13.2
<b>T2: NS + MAP</b>	13.1	12.9	12.9	12.9	13.0	13.8	13.8	13.7

**Table 47: Average and Standard deviation for TSS over 21 day period (Data combined)**

	Average Values				Standard Deviation			
	0	7	14	21	0	7	14	21
<b>T1: NS no MAP</b>	13.8	13.3	13.3	13.4	0.2	0.6	0.6	0.6
<b>T2: NS + MAP</b>	13.0	13.3	13.3	13.3	0.6	0.6	0.6	0.5

**Table 48: Average values for TA over 21 day period**

	Average Values				Average Values			
	Set 1				Set 2			
	0	7	14	21	0	7	14	21
<b>T1: NS no MAP</b>	0.27	0.20	0.21	0.22	0.21	0.24	0.23	0.21
<b>T2: NS + MAP</b>	0.20	0.23	0.22	0.22	0.22	0.23	0.21	0.21

**Table 49: Average and Standard Deviation for TA over 21 day period (Data combined)**

	Average Values				Standard Deviation			
	0	7	14	21	0	7	14	21
<b>T1: NS no MAP</b>	0.24	0.22	0.22	0.22	0.05	0.04	0.02	0.00
<b>T2: NS + MAP</b>	0.21	0.23	0.22	0.22	0.03	0.01	0.01	0.01

**Table 50: Average and Standard deviation for bacterial count in pears over 21 days**

Day	1	7	14	21
	11/9/2009	11/16/2009	11/23/2009	11/30/2009
	<b>Average</b>			
<b>NS no MAP</b>	2.15	5.71	4.79	6.55
<b>NS + MAP</b>	2.08	4.46	4.14	6.02
	<b>Standard Deviation</b>			
<b>NS no MAP</b>	0.22	0.20	0.66	0.08
<b>NS + MAP</b>	0.26	0.15	1.10	0.64

*Units are in log (CFU g<sup>-1</sup>)*

**Table 51: Average and Standard Deviation for yeast & mold count in pears over 21 days**

Day	1	7	14	21
	11/10/2009	11/16/2009	11/23/2009	11/30/2009
	<b>Average</b>			
<b>NS no MAP</b>	2.2	4.3	4.3	5.4
<b>NS + MAP</b>	1.2	3.3	3.7	4.0
	<b>Standard Deviation</b>			
<b>NS no MAP</b>	0.17	0.09	0.00	0.07
<b>NS + MAP</b>	1.02	0.16	0.59	0.58

*Units are in log (CFU g<sup>-1</sup>)*



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