AGROTECHNOLOGY & FOOD SCIENCES GROUP
WAGENINGEN UR

Freshly cut fruit salads

Final Report

March 2007

Daniela de Carvalho Ribeiro

Rapport no. 775

AGROTECHNOLOGY & FOOD INNOVATIONS
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Abstract

There is an increasing demand for various freshly cut fruit products. However, their short shelf-life has limited the market success of these products. This study aims to explore new combinations of packaging and pre-treatment technologies to achieve longer shelf lives (up to 2 weeks) without compromising on the product quality.

Quality changes (firmness, colour, odour, taste and visual appearance) of fresh-cut pineapple, melon, pear, kiwifruit, apple and mixes of these fruits were evaluated. Modified atmosphere packaging (MAP) with conventional and non-conventional gas mixtures in combination with chemical treatments to reduce browning, firmness loss and decay of fresh-cut fruits were evaluated. A modified atmosphere of 4% O_2, 10% CO_2, 86% N_2 in combination with 2% ascorbic acid dipping extended the shelf-life of fresh-cut pineapple was evaluated for more than 12 days. High O_2 atmospheres and air in combination with a dip solution of 2,5% calcium lactate or 2,5% $CaCl_2$ were found to be particularly effective in preserve the quality of fresh-cut melon for more than 12 days. Beneficial effects of 100% N_2 MAP combined with 2% ascorbic acid, 1% calcium lactate and 0.5% cysteine dip were found on enzymatic browning for fresh-cut pears during at least 12 days. During 7 days fresh-cut kiwifruits maintained the quality with 100% N_2O or 10% O_2, 90% N_2 in combination with a dip solution of 2% $CaCl_2$. N_2 atmospheres combined with a dip in ascorbic acid and calcium chloride inhibited most of the changes in colour and texture properties of fresh-cut apples for more than 12 days of storage.

During more than 12 days, mixtures of pineapple, apple, pear, melon and red grape packed in air with chemical treatment did not suffer quality changes. Colour, odour, taste and visual appearance remained unaltered. High O_2 atmospheres (80-90%) were very efficient in maintaining the fresh look appearance of pear, melon and red grape during 12 days. A mixture of pineapple, kiwifruit, melon and red grape package in N_2O atmosphere had 8 days of shelf-life. Nitrogen rich atmospheres for packaging only single cut fruits were very efficient, however for mixtures fruits proved to be unsuccessful yielding no more than four days of shelf-life.

Fresh-cut fruits packaged in modified atmosphere with air, high O_2 and N_2O prove to be beneficial in maintaining the quality and extending shelf-life in fruit salads. However, nitrogen rich atmospheres were not efficient in preserving fruit salads once the microbiologic activity was high.

1. Introduction

1.1. Fresh-cut fruit

During the 1990s, a new type of fruit products, fresh-cut fruits, was launched on the markets of western countries. Fresh-cut fruits appeared in response to the increasing demand for ready-to-eat convenient and healthy foods, which are of high quality and possess attributes similar to those of fresh non processed products.

Fresh-cut fruits are defined as those that have been lightly or minimally processed, thus undergo one or multiple operations such as washing, peeling, coring, cutting, hygienisation, dipping in stabiliser solutions or packaging under appropriate atmospheres. These produce have to be refrigerated during their entire shelf-life and, therefore, need to be distributed though reliable distribution channels in order to ensure safety and quality maintenance.

Fresh-cut fruits are a rapidly growing segment of the retail horticulture-based food service industry. Consumers in the United States and Europe are demanding more convenient but still healthy, ready-to-eat fruits due to their busy lifestyles and increasing purchase power. As a result, they willing to pay more for minimally processed fruit than the price paid for the whole fresh product. In addition, consumers pay for the convenience. This is because they also buy a series of services such a selection, washing, sanitation, cutting and packaging, which are added to the product and therefore increases product value.

The primary quality attributes of a fresh-cut product include colour, texture, appearance, flavour and aroma. Beyond these commercial quality attributes, consumers expect minimally processed fruits to be safe and have high nutritional values.

1.2. Respiration in fruits

Respiration in the cells of fruits is the metabolic process involving the breakdown of complex organic compounds such sugars, organic acids, amino acids and fatty acids into lower molecular weight molecules with the accompanying production of energy (ATP and heat) through oxidation-reduction.

Aerobic Respiration requires:

- Glucose
- Oxygen

Aerobic Respiration produces:

- Energy
- Carbon Dioxide
- Water

The summation of all the reaction for the aerobic respiratory breakdown of the most common substrate, glucose, is:

$$C_6H_{12}O_6 + 6O_2 \rightarrow 6CO_2 + 6H_2O + 2870 \text{ kJ}$$

In the absence of oxygen, anaerobic respiration (fermentation) occurs in fruits. Under anaerobic conditions, much lower amounts of energy and CO_2 are formed from 1 mole of glucose than that under aerobic conditions. Further the metabolic end-products are lactic acid, acetaldehyde and ethanol from the degradation of pyruvic acid. Off-flavours and off-odours sometimes are evident in fruits stored/package in atmospheres with very low levels of O_2.

Anaerobic Respiration requires:

- Glucose

Anaerobic Respiration produces:

- Energy (much less energy)
- Carbon Dioxide
- Alcohol

The summation of all the reaction for the anaerobic respiratory is:

$$C_6H_{12}O_6 \rightarrow 2CO_2 + 2C_2H_5OH + 92 \text{ kJ.}$$

1.2.1. Shelf-life and Respiration Rate

In general, there is an inverse relationship between respiration rates and postharvest-life of fresh commodities. The higher the respiration rate, the more perishable, shorter postharvest-life, the commodity usually is. Respiration plays a major role in the postharvest life of fresh commodities because it reflects the metabolic activity of the tissue that also includes the loss of substrate, the synthesis of new compounds, and the release of heat energy.

- Loss of substrate: Use of various substrates in respiration can result in loss of food reserves in the tissue and loss of taste quality (especially sweetness) and food value to the consumer.
- Synthesis of new compounds: Postharvest storage can be used to either prevent any reduction in quality, or to promote changes that increase quality. The quality of most non-climacteric fruit (e.g., strawberries, pineapple, and grape) is maximal at harvest and storage conditions are optimized to prevent quality loss. In contrast, many non-climacteric fruit (eg, lemons and oranges), and climacteric fruit (eg, bananas, apple, and pear) are harvested before they reach their best quality and storage conditions are optimized to permit the development of optimum quality. In the first case, the synthesis of new compounds is unnecessary because they lead to reduced quality. In the second case, synthesis of pigments and volatiles, loss of chlorophyll, and the conversion of starch to sugar are necessary for development of maximum quality. These synthetic reactions require energy and organic molecules derived from respiration.
- Release of Heat Energy: The heat produced by respiration (vital heat), which is about 673 kcal for each mole of sugar (180 g) utilized, can be a major factor in establishing the refrigeration requirements during transport and storage. Vital heat must be considered in selecting proper methods for cooling, package design, method of stacking packages, and refrigerated storage facilities. Some commodities have high respiration rates and require considerably more refrigeration than more slowly respiring produce to keep them at a specified temperature. For example, asparagus, broccoli, mushrooms and peas respire about 10-times faster than apples, cabbage, lemons, and tomatoes.

1.3. Quality aspects of fresh-cut fruits

Consumers chose fresh-cut fruit products in the shops based on their visual appearance. After opening the package they will experience the odour of the product, and then they will eat the product and notice the texture and taste. Hence, one quality aspect (visual appearance) is critical for the first purchase. For consumer satisfaction and repeat purchases also odour, texture and taste are important. Other quality aspects as nutritional quality, safety, sustainability usually play a minor or an indirect role in the consumer motivation to buy such a product in their supermarket.

1.3.1. Texture

Fruit texture is perceived by the consumer prior to taste. When biting into a piece of apple, crunchiness is perceived before juiciness. Softening or loss of tissue firmness is a quality defect that compromises the shelf-life of many fresh-cut fruits.

Texture is not a single, well-defined attribute. It is a collective term that encompasses the structural and mechanical properties of a food and their sensory perception in the hand or mouth. Although some definitions of texture restrict its use to only sensory attributes or to sensory attributes and the mechanical properties directly related to them, the term texture is sometimes extended to include some mechanical properties of commercial interest that may not be of direct interest to consumers, such as resistance to mechanical damage. A few of the many terms used to describe sensory texture of fruits or vegetables are hard, firm, soft, crisp, limp, mealy, tough, leathery, melting, gritty, woolly, stringy, dry, and juicy. There are no accepted instrumental methods for measuring each of these attributes.

Textural attributes of fruits and vegetables are related to the structural, physiological, and biochemical characteristics of the living cells; their changes over time; and their alteration by processes such as cooking or freezing. The continuous physiological changes in living cells plus the inherent variability among individual units of the commodity make the assessment of fruit or vegetable texture difficult. Because of their continuous change, textural measurements are often relevant only at the time of evaluation; that is, they usually cannot be used to predict condition much later in the storage period or marketing chain.

Textural changes in fresh-cut vegetables and fruits are minimized at low temperatures. Thus, temperature management is the first step to maintain the initial, fresh textural quality of these products. Several treatments have been proven to be effective in reducing fresh-cut firmness loss. A common treatment used to improve tissue firmness is to spray or dip fruit or vegetable pieces in aqueous calcium salts, as described for shredded carrots, zucchini slices, and fresh-cut pears, strawberries, kiwifruit, nectarines, peaches, and melons (Agar et al., 1999; Gorny et al., 1999; 2002; Izumi and Watada, 1994; 1995; Luna-Guzman et al., 1999; Luna-Guzman and Barrett, 2000; Main et al., 1986; Morris et al., 1985; Rosen and Kader, 1989). This is a result of the pivotal role played by calcium in maintaining the textural quality of produce due to its effect of rigidifying cell wall structure by cross-linking ester groups and also preserving the structural and functional integrity of membrane systems (Poovaiah, 1986).

However, there is a problem noticed when calcium chloride is used as a firming agent, which is that it may result in undesirable bitter flavour of the product. Fresh-cut cantaloupe cylinders dipped in calcium lactate solutions resulted in a textural improvement similar to calcium chloride-treated fruit cylinders. Sensory evaluation indicated that results were better since less bitterness and a more detectable melon flavour were perceived (Gorny et al., 2002; Luna-Guzman and Barrett, 2000).

1.3.2. Appearance

In order to trigger a purchase it is extremely important that fresh-cut products also look "fresh". "Fresh appearance" can be characterized by size, shape, colour, gloss, condition, and absence of defects. However, product colour contributes more than any other single factor. The factors that make a fresh-cut product look fresh can be very different for various products and usually have a strong cultural aspect. However, product colour contributes more than any other single factor. Defects can originate before harvest as a result of damage by insects, diseases, birds, and hail; chemical injuries; and various blemishes (such as scars, scabs, russeting, rind staining). Postharvest defects may be morphological, physical, physiological or pathological.

1.3.3. Flavour and aroma

Flavour quality of fresh-cut vegetables and fruits is critical to their acceptance and appreciation by consumers. Sensory attributes such as sweetness and characteristic aroma may be the most important indicators of shelf life from the consumer's point of view. The challenge in fresh-cut vegetable and fruit handling is to maintain the taste and aroma attributes of the original whole product. Due to the short shelf life of fresh-cut fruits and vegetables, starting with produce at its optimum maturity or ripeness stage and using the highest quality standards is extremely important in maintaining flavour shelf life (Beaulieu and Gorny, 2002).

Taste and aroma together make up flavour, which contributes to the recognizable nature of a food. Taste refers to detection of non-volatile compounds on the tongue while aroma is related to volatile compounds detected in the nose. These two aspects of flavour are inextricably linked as it has been shown that perception of aroma can be influenced by levels of taste components, and vice versa (Beaulieu and Baldwin, 2002).

The taste component in fruits and vegetables mainly depends on the sugar and organic acid levels and the relation between them. As a result of increased respiration due to wounding during preparation of fresh-cut produce, there is a depletion of sugar levels in the commodity and therefore organoleptic quality is reduced, especially in products like melons whose quality depends on high levels of sugars. This problem tends to be worse in fruits such as tomato or melon that have very limited capacity to replenish soluble sugars lost to accelerated respiration during storage or ripening, in comparison with fruits such as banana and apple that have a reserve of starch and can convert it to sugars during ripening. Organic acids are one of the major respiratory substrate and the increase in tissue pH in fresh-cut apples has been attributed to utilization of organic acids in respiration (Kim et al., 1993). Depletion of acids also can have negative organoleptic effects in fruits like apple, peach, and mango for which the balance of sweetness (sugars) and tartness (acids) is an important flavour attribute.

The second component of flavour, aroma, is related to synthesis of volatiles during the growth and development of fruits or vegetables, but the most dramatic production coincides with fruit ripening. These volatiles include alcohols, aldehydes, esters, ketones, lactones, and other compounds (Baldwin, 2002).

1.3.4. Water

Plant tissues are mainly composed of water and any small changes in water content may have a large impact on produce quality and could cause a variety of negative characteristics such as limpness, flaccidity, shrivel, wrinkle, and/or tissue desiccation.

On the other hand, crispness, an important characteristic of fresh produce that is related to water turgor pressure in the tissue, could be easily lost due to water loss. The loss in crispiness results in softening and flaccidity.

In case of fresh-cut products, the possibilities to undergo severe water loss are much higher than in the case of intact produce since during fresh-cut preparation the produce goes through peeling and cutting or shredding, slicing, etc. Losing the skin has a critical effect on many fresh-cut products because the skin is a protective waxy coating, highly resistant to water loss, and thus peeled, fresh-cut fruits and vegetables are more perishable and more susceptible to turgor loss and desiccation. On the other hand, the mechanical injury brought on by cutting, shredding and/or slicing, directly exposes the internal tissues to the atmosphere, promoting the evaporation of intercellular water and hence starting desiccation of the tissue. Furthermore, fresh-cut preparation increases the relative product surface area per unit mass or volume, which also increases the water loss.

The control of water loss in fresh-cut fruits and vegetables is possible through the use of appropriate handling techniques, including temperature and relative humidity control, which can greatly help minimize the rate of water loss. Reduction of water loss can be achieved basically by decreasing the capacity of the surrounding air to hold water, which can be achieved by lowering the temperature and/or increasing the relative humidity.

1.3.5. Nutritional aspects

Nowadays there is more awareness of the importance of fruits and vegetables as a great source of antioxidant compounds such as polyphenolics, vitamin C, vitamin E, β-carotene, and other carotenoids. It has been suggested that these phytonutrients have long-term health benefits and may reduce the risk of diseases such as cancer and heart disease.

Vegetables and fruits are the primary source of these antioxidant compounds in our daily diet and there is a lot of epidemiological work that shows strong correlations between the delay or suppression of certain diseases and consumption rates of fruits and vegetables. Retaining maximum bioactivity of these phytonutrients in fresh-cut fruits and vegetables is a very important goal and this could be achieved through better understanding of the effect of fresh-cut processing, packaging, and storage on bioactivity of these compounds. Many investigations with fresh-cut fruits and vegetables have demonstrated that concentrations of vitamins and other phytochemical compounds are reduced following fresh-cut operations as a result of wounding and are affected by conditions of handling, packaging, and storage. But, on the other hand, stress associated with processing may also initiate biosynthesis of many compounds that affect antioxidant content and product quality. The synthesis of wound ethylene after fresh-cut operations can stimulate a variety of physiological responses including loss of vitamin C and chlorophyll and induction of polyphenolic metabolism.

1.3.6. Microbial spoilage

The processing that fruit undergo turn them more vulnerable to microbiological risks, and the sensory quality is meaningless if the product is unsafe. Along the chains of steps involved in the production of fresh-cut products, from the growth of the raw material to the processing and distribution, there is potential for microbiological risk. Microbial contamination may (1) be of public health significance due to the presence of human pathogens, or (2) decrease the shelf life of the fresh-cut fruit due to spoilage microorganisms, and in addition render a major economic impact.

Microbial decay of fresh-cut fruit may occur much more rapidly than in vegetable products due to high levels of sugars found in most fruit. However, the acidity of fruit tissue usually helps suppress bacterial growth, but not growth of yeast and mould. There is no evidence to suggest that lower aerobic plate counts (APC) or total plate counts (TPC) immediately after processing correlate with increased shelf-life in fresh-cut vegetables. However, with fresh-cut fruit, very low APC, TPC and especially yeast and mould counts correlate with increased shelf-life.

1.3.7. Colour Retention – Enzymatic Browning

The organoleptic and biochemical characteristics of fruits may be profoundly modified by the appearance of brown pigments whose colour is superposed on the natural colours. Surface discoloration is probably the most quality defect of fresh-cut fruits and the factor most limiting shelf life.

Enzymatic browning occurs in fruits after bruising, cutting, or during storage, and its control during the processing of fruits is of great importance to fruit manufacturing. Enzymatic browning is a significant problem in apples, pears, bananas, peaches, and grapes, particularly.

Some browning may be non-enzymatic in origin and result from the Maillard reaction when mixtures of amino acids and carbohydrates are heated. However, most browning, and particularly the types that occur very rapidly, are caused by enzymatic oxidation of phenolic compounds under the effect of polyphenol oxidase.

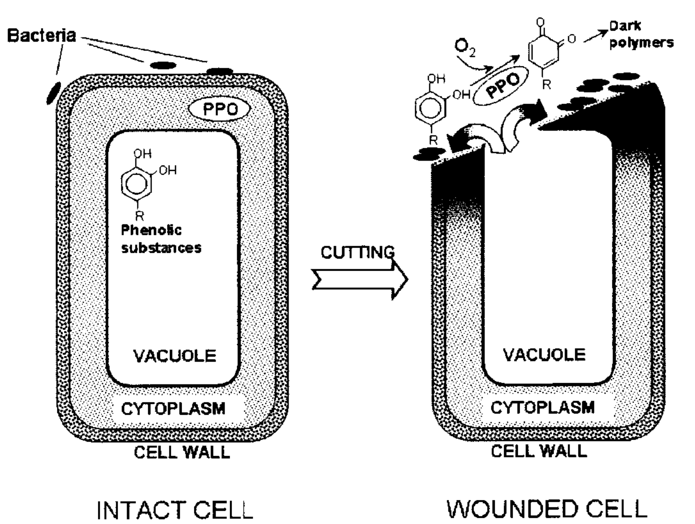


Figure 1 - Wounding effects stimulating oxidation of cellular components causing tissue browning, microbial development on the wound surface and tissue breakdown.

During the peeling and cutting operations, cells are broken (see figure 1), and their contents include previously compartmentalized enzymes that are suddenly liberated will now come into contact with their substrates and start the browning reactions

Polyphenol oxidase (PPP) is an example of an enzyme that can lower the quality of a food product by catalyzing the oxidation of phenolic compounds. The susceptibility to browning may depend on PPO activity and/or phenolic content (Coseteng and Lee, 1987).

Polyphenol oxidase catalyzes the initial step in the polymerization of phenolics to produce quinones, which undergo further polymerization to insoluble dark brown polymers known as melanins. These melanins form barriers and have antimicrobial properties, which prevent the spread of infection or bruising in plant tissues. The formation of yellow and brown pigments in fruit products during enzymatic browning reactions is controlled by the levels of phenols, the amount of PPO activity, and the presence of oxygen (Spanos and Wrolstad, 1992).

The enzymatic browning of fruit is always considered as a quality loss of both fresh and processed food products. Simple representation of enzymatic browning reactions is given in figure 2.

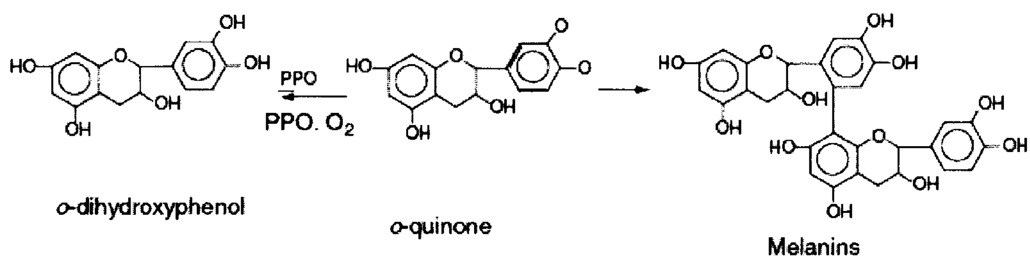


Figure 2 - Simplified mechanism for the transformation of a diphenol to dark colored melanins by PPO.

PPO activity, as for most of enzymes, may be minimized by reducing agents, heat inactivation, lowering the pH of the fruit product, and the presence of enzyme inhibitors, among other techniques. To effectively inhibit or control the enzymatic browning in fruit products, an accurate determination of the kinetics of these catalyzed-oxidative reactions is required. The kinetics of deterioration can be followed through colour measurements, which is a simple and effective way for studying the phenomenon.

The substrate specificity of polyphenol oxidase varies in accordance with the source of the enzyme. Phenolic compounds and polyphenol oxidase are in general directly responsible for enzymatic browning reactions in damaged fruits, during postharvest handling and processing. The relationship of the rate of browning to phenolic content and polyphenol oxidase activity has been reported for various fruits such as apples (Coseteng and Lee, 1987), grapes (Lee and Jaworski, 1988), and peaches (Lee et al., 1990). In addition to serving as polyphenol oxidase substrates, phenolic compounds act as inhibitors of polyphenol oxidases (Walker, 1995). Their inhibitory action decreased in the following order: cinnamic acid>p-coumaric acid>ferulic acid>benzoic acid.

Although relatively few of the phenolic compounds in fruits serve as substrates for polyphenol oxidase, as catechins, cinnamic acid esters, 3,4-dihydroxy phenylalanine (DOPA), and tyrosine (Table 1), the stoicheometry of complex reactions like enzymatic browning in fruits as substrate is practically unknown. Therefore, instead of determination of consumption of reactives (phenols), or formation of products (melanins), the kinetics of colour development is commonly used for studying the browning reactions.

Table 1 - Phenolic substrates of PPO in fruits.

Fruit	Phenolic substrates
Apple	Chlorogenic acid (flesh), catechol, catechin (peel), caffeic acid, 3,4-dihydroxyphenylalanine (DOPA), 3,4-dihydroxy benzoic acid, *p*-cresol, 4-methyl catechol, leucocyanidin, *p*-coumaric acid, flavonol glycosides
Apricot	Isochlorogenic acid, caffeic acid, 4-methyl catechol, chlorogenic acid, catechin, epicatechin, pyrogallol, catechol, flavonols, *p*-coumaric acid derivatives
Avocado	4-methyl catechol, dopamine, pyrogallol, catechol, chlorogenic acid, caffeic acid, DOPA
Banana	3,4-dihydroxyphenylethylamine (dopamine), leucodelphinidin, leucocyanidin
Eggplant	Chlorogenic acid, caffeic acid, coumaric acid, cinnamic acid derivatives
Grape	Catechin, chlorogenic acid, catechol, caffeic acid, DOPA, tannins, flavonols, protocatechuic acid, resorcinol, hydroquinone, phenol
Mango	Dopamine-HCl, 4-methyl catechol, caffeic acid, catechol, catechin, chlorogenic acid, tyrosine, DOPA, *p*-cresol
Peach	Chlorogenic acid, pyrogallol, 4-methyl catechol, catechol, caffeic acid, gallic acid, catechin, dopamine
Pear	Chlorogenic acid, catechol, catechin, caffeic acid, DOPA, 3,4-dihydroxy benzoic acid, *p*-cresol
Plum	Chlorogenic acid, catechin, caffeic acid, catechol, DOPA

Adapted from Marshall, Kim and Wei, 2000.

1.4. Addition of antibrowning compounds

Enzymatic browning may be controlled through the use of physical and chemical methods; often both are employed. Physical methods commonly used include reduction of temperature and oxygen, and use of modified atmosphere packaging or edible coatings. Chemical methods depend on either treatment with compounds that inhibit polyphenol oxidase, remove its substrates (oxygen and phenolics), or function as preferred substrates. Various antibrowning agents can be used in postcutting dip solutions.

1.4.1. L-Ascorbic acid

Ascorbic acid is a moderately strong reducing compound, which is acidic in nature, forms neutral salts with bases, and is highly water-soluble. L-ascorbic acid (vitamin C) and its various neutral salts and other derivatives have been the leading antioxidants for use on fruits and vegetables and in fruit juices, for the prevention of browning and other oxidative reactions.

Ascorbic acid also acts as an oxygen scavenger for the removal of molecular oxygen in polyphenol oxidase reactions. Polyphenol oxidase inhibition by ascorbic acid has been attributed to the reduction of enzymatically formed *o*-quinones to their precursor diphenols. Ascorbic acid is however irreversibly oxidized to dehydroascorbic acid during the reduction process, thus allowing browning to occur upon its depletion (figure 3). More stable forms of ascorbic acid derivatives, such as erythrobic acid, 2- and 3-phosphate derivatives of ascorbic acid, phosphinate esters of ascorbic acid, and ascorbyl-6-fatty acid esters of ascorbic acid, have however been developed to overcome these problems. Ascorbic acid esters release ascorbic acid upon hydrolysis by acid phosphatases. Their relative effectiveness as browning inhibitors varies in accordance with the food product. Compounds containing reactive amino or thiol groups can greatly affect the reactivity of *o*-quinones.

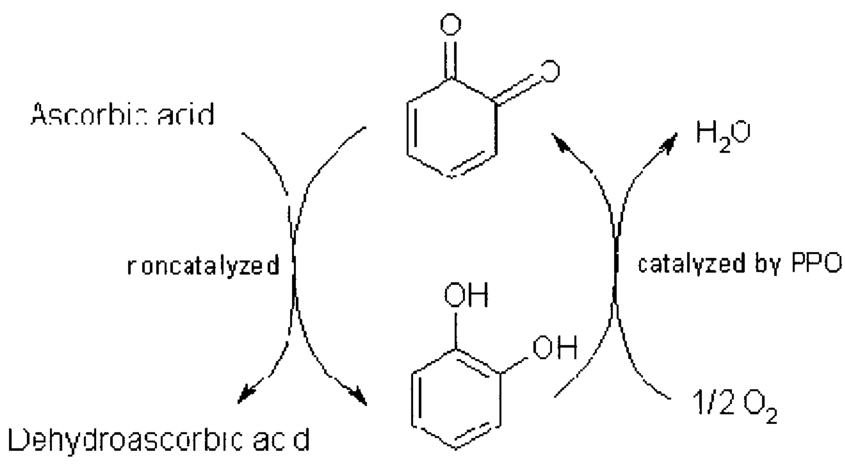


Figure 3 - Mechanism of prevention of colour formation by ascorbic acid.

1.4.2. Cysteine

Cysteine is an effective inhibitor of enzymatic browning. Concentrations of cysteine and other thiols required for the achievement of acceptable levels of browning inhibition have however been shown to have negative effects on taste. The inhibition of melanosis by cysteine is thought to be due to the formation of colourless thiol-conjugated *o*-quinones. Cysteine has also been shown to reduce *o*-quinones to their phenol precursors.

A mode of action for cysteine and cysteinyl addition in the control of browning proposed by Richard-Forget *et al.* (1992) is illustrated in figure 4. Cysteine-quinone adducts serve as competitive inhibitors of polyphenol oxidase

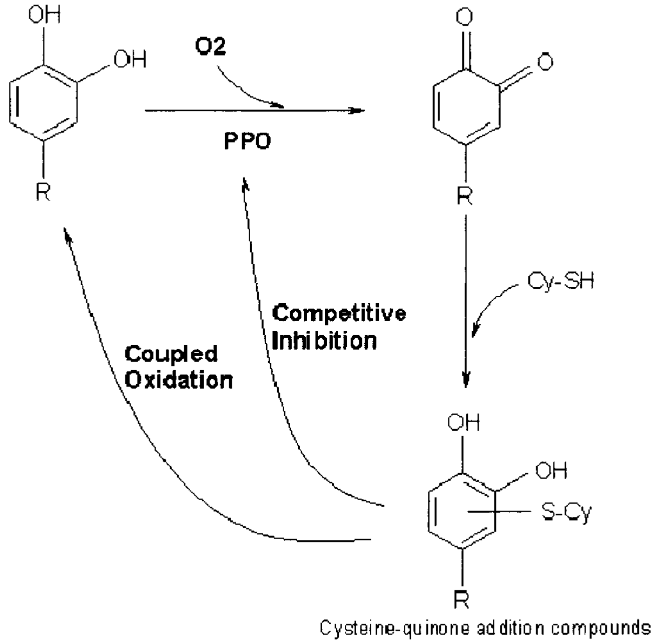


Figure 4 - Effect of cysteine and cyteinyl addition compounds with o-quinones on the enzymatic oxidation of o-diphenols

1.4.3. Citric acid

Citric acid is the one of the most widely used acidulants in the food industry. It is typically applied at levels ranging between 0.5 and 2 percent (*w/v*) for the prevention of browning in fruits and vegetables. In addition, it is often used in combination with other antibrowning agents such as ascorbic or erythorbic acids and their neutral salts, for the chelation of prooxidants and for the inactivation of polyphenol oxidase. Recommended usage levels for citric acid typically vary between 0.1 and 0.3 percent (*w/v*) with the appropriate antioxidant at levels ranging between 100 and 200 ppm. Citric acid exerts its inhibitory effect on polyphenol oxidase by lowering the pH as well as by chelating the copper at the active site of the enzyme.

1.4.4. 4-Hexylresorcinol

Substituted resorcinols, which are *m*-diphenolic compounds that are structurally related to phenolic substrates, have a competitive inhibitory effect on polyphenol oxidase activity. Hydrophobic substitution with hexyl, dodecyl, and cyclohexyl groups at the 4-position of the aromatic resorcinol ring increases the effectiveness of their competitive inhibitory effect on polyphenol oxidase.Studies conducted by McEvily *et al.* (1992) revealed cyclohexyl-substituted resorcinols to have the lowest I_{50}, i.e. the inhibitor concentration that resulted in 50 percent inhibition of polyphenoloxidase activity, at a concentration of 0.2 mM of the substituted resorcinol. Both the monophenolase and diphenolase activities of tyrosinase are inhibited by 4-hexylresorcinol (4-HR). Four-hexylresorcinol has a long history of use in pharmaceuticals and is considered to be safe and effective in use as an anti-browning agent.

Four-hexylresorcinol has several advantages over sulphites when applied in the control of browning in foods. These include its specific mode of inhibitory action, effectiveness at low concentrations, inability to bleach preformed pigments, and chemical stability. It has a synergistic effect with ascorbic acid in the prevention of browning. Ascorbic acid reduces quinones generated by polyphenoloxidase while 4-HR specifically interacts with polyphenol oxidase, and renders it incapable of catalysing the enzymatic reaction.

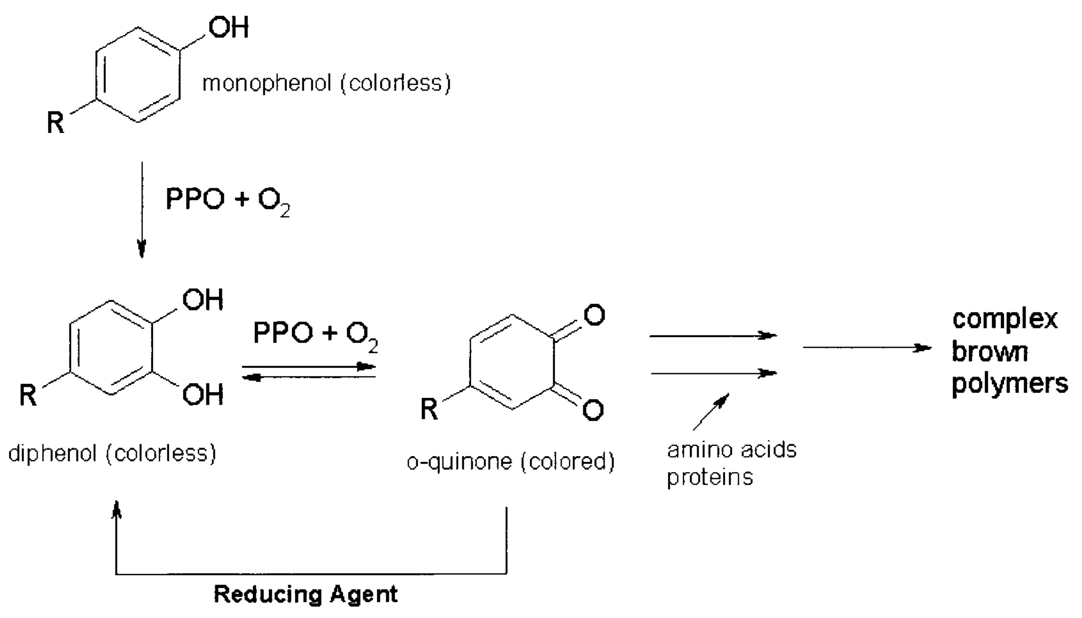


Figure 5 - The inhibitory effect of 4-hexylresorcinol on PPO.

1.5. Unit operations in fresh-cut fruits preparation

In order to maintain the highest possible quality of a fresh-cut fruit, it is important to observe adequate handling of the produce before preparation. It is necessary to keep produce under adequate storage conditions (temperature and relative humidity) and use gentle handling to minimize bruising and other physical injuries. Strict temperature control is the single most important factor in maintaining quality throughout the fresh-cut preparation and distribution. All unit operations should be carried out in cold processing rooms (≤5°C), and cleanliness is very important to ensure a high-quality and safe product. The overall steps involved in the preparation of fresh-cut fruit follow.

RAW MATERIAL GROWING
⇓
HARVEST
⇓
PRE-COOLING
⇓
RANSPORT
⇓
STORAGE
⇓
SELECTION
⇓
WASHING/BRUISING
⇓
PEELING
⇓
CUTTING
⇓
WASHING
⇓
PRE-TREATMENTS
(ANTIBROWNING / ANTISOFTENING)
⇓
DRAINING/DRYING
⇓
WEIGHING/PACKAGING
⇓
COLD STORAGE/COLD CHAIN
⇓
TRANSSPORT
⇓
DISPLAY/SALE

Figure 6 - Flow-sheet of fresh-cut horticultural products processing

1.5.1. Receiving, inspection and storage of raw material

Fruit has to be inspected and evaluated according to high standards of safety and quality. It is well known that no processing can increase the quality of already strongly decayed or misshapen ingredients. Therefore, it is very important that while awaiting processing, fruit should be kept under refrigeration (in general, 1 to 5°C) according to produce specifications. If a commodity is sensitive to chilling injury, it requires appropriate storage conditions, separate from no sensitive fruits. For example, symptoms of chilling injury are seen in pineapple stored for prolonged periods at temperatures below 12°C. Whereas some fruits, such as pears and apples, can be stored for extended periods of time while waiting to undergo processing, the effects of such storage on fresh-cut products may be undesirable.

1.5.2. Cleaning and disinfection

With many fruit that have a smooth surface, such as apples and pears, the use of cold clean water may be enough for a first wash. However, some fruits may harbour high microbial populations on their external surface. Fruits, like cantaloupes and pineapples should be washed, scrubbed, and dipped in solutions disinfectants. The most commonly used sanitizer is chlorinated water, but other possible chemicals for disinfection are hydrogen peroxide, surfactants, peroxyacetic acid among others.

1.5.3. Peeling, deseeding, trimming, coring and cutting

In contrast to vegetables, which in many instances can be peeled and cut mechanically (such as carrots), for most fruits whose texture is soft, the removal of skins and rinds is carried out by hand. It is very important to use clean and sharp knives during this operation. The use of blunt blades causes excessive physical injury (tissue crushing) to the fruit tissue adjacent to the cut, accelerating quality deterioration. Fresh-cut cantaloupe melon pieces prepared using blunt knives have increased ethanol levels, off-odours, and electrolyte leakage compared to sharp-cup pieces. The marketable visual quality of sharp-cut melon lasted longer; blunt-cut pieces developed a translucency, which is a common visual defect that indicates cell disruption in commercially prepared fresh-cut cantaloupe.

After this treatment, peeled fruit have to be rinsed by immersion in water for removal realeased enzymes and nutrients for microbial growth.

1.5.4. Washing and cooling

During the cutting steps there is a release of tissue fluids that should be removed to avoid undesirable microbiological or chemical reactions. It is imperative to rinse the cut surfaces of the fruit. At this stage the use of cold water (~0°C) accomplishes the washing of the cut surfaces as well as cooling of the fruit pieces. For safety reasons, chlorinated water (usually 50 to 100 ppm) is frequently used. All the equipments used in fruits the processing (cutting tools and boards) are a potential source of contamination and require adequate sanitation. In some cases it is necessary to use dipping solutions containing processing aids that will help prevent nonmicrobial quality deterioration such as surface and softening.

1.5.5. Dewatering

Excess moisture picked up during the washing operation should be removed prior to packaging. This step helps to prevent growth of microorganisms that remained after produce disinfection. Due to their delicate texture, fresh-cut fruit require passage through semi-fluidized beds with forced air to remove moisture or a simple gently centrifugation.

1.5.6. Packaging and distribution

In order to ensure the longest possible shelf-life for fresh-cut fruits, it is important to choose appropriate packaging materials and storage conditions.

Temperature is always a critical factor in the shelf life of fresh-cut fruits. The importance of temperature control is related to food safety and extension of fresh-cut life by slowing respiration rate and preventing quality deterioration. Throughout the unit operations involved in preparations and up to consumption, a cold chain should be maintained; ideally temperatures should not exceed 5°C, although preferably they should be closer to 1°C. During the distribution of fresh-cut products, it is also important to avoid rough handling. Very often products are subjected to shock and vibration stress, which cause injury, leading to more rapid quality loss.

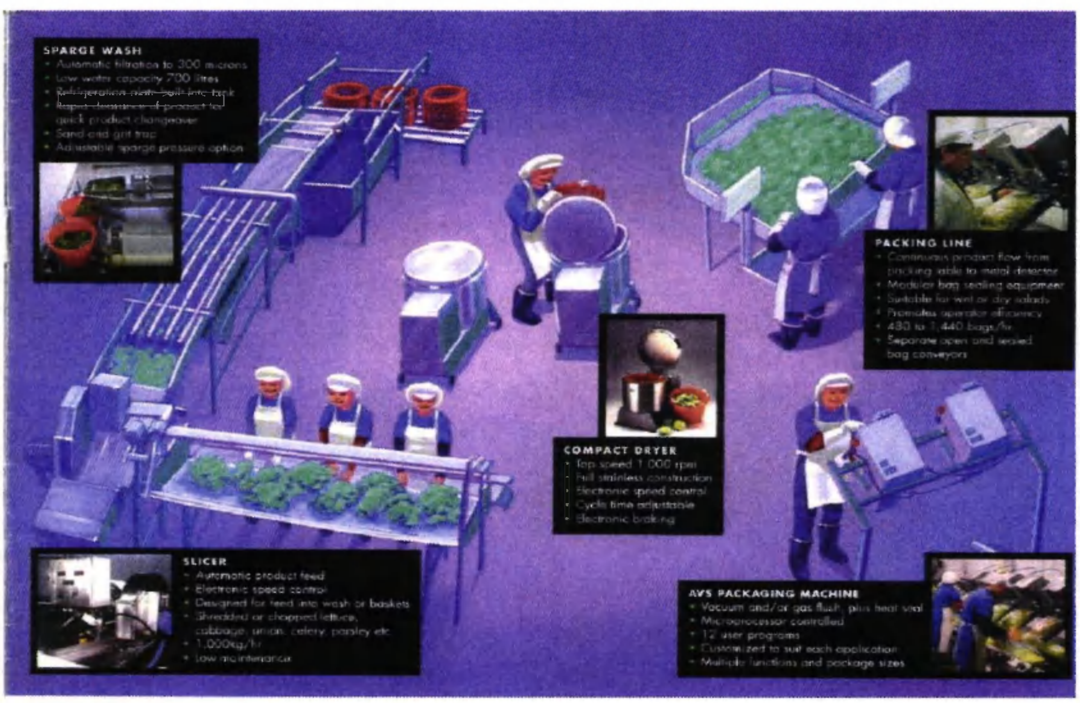


Figure 7 - Modern technology process for fresh-cut

1.6. Modified atmosphere packaging

During the last few decades there has been a trend towards an increased demand for chilled food products with prolonged shelf-lives. Consumers prefer pre-processed products that are fresh or fresh like, convenient and easy to prepare and without additives. Pre-packed products offer today's busy consumers a considerable saving in time and money. This has promoted the development of alternative technologies for foodstuff packaging, distribution and storage, such as modified atmosphere packaging (MAP), resulting in products with an increased shelf-life and higher quality. MAP has become a commercial and economic reality in markets that have a well-established and controlled cold chain and which can sustain a high-priced quality product.

1.6.1. MAP principles

The principle of MAP is the replacement of air in the package with a fixed gas mixture. Once the gas mixture is introduced, no further control of the gas composition is exercised, and the composition will inevitably change due to respiration, diffusion, etc. MAP is also known as "gas packaging" or "gas exchange packaging", but these are not recommended on packaging labels since consumers often perceive the word gas negatively. Lately MAP has also been referred to as *protective atmosphere packaging* or, when used in labeling, "packaging in a protective atmosphere".

The most apparent advantage of MAP is the achieved increased shelf-life, but MAP also has several other advantages and also disadvantages (table 2)

The effectiveness of MAP on extending shelf-lives, however, is dependent on several factors: type of food, initial quality of the raw material, gas mixture, storage temperatures, hygiene during handling and packaging, gas/product volume ratio and the barriers properties of the packaging material.

Table 2 – Advantages and disadvantages with modified atmosphere packaging

Advantages	Disadvantages
Shelf-life increase by possible 50-400%	Added costs
Reduced economic losses due to longer shelf-life	Temperature control necessary
Decreased distribution cost, longer distribution distances and lower delivery frequencies	Different gas formulations for each product type
Provides a high quality product	Special equipment and training required
Easier separation of sliced products	Product safety to be established
Centralised packaging and portions control	Increased pack volume – adversely affects transport costs and retail display space
Improved presentation – clear view of products and all-around visibility	Loss of benefits once the pack is opened or leaks
Little or no need for chemical preservatives	Dissolved CO2 into the food could lead to pack collapse and increased drip
Sealed packages, barriers against product re-contamination and drip from package	
Odourless and convenient packages	

1.6.2. Components of gas mixtures in MAP

Oxygen, CO_2, and N_2, are most often used in MAP. Other gases such as nitrous and nitric oxides, sulphur dioxide, ethylene, chlorine, as well as ozone and propylene oxide have been suggested and investigated experimentally. However, due to safety, regulatory and cost considerations, they have not been applied commercially already.

These gases are combined in three ways for use in modified atmospheres: inert blanketing using N_2, semi-reactive blanketing using CO_2/N_2 or $O_2/CO_2/N_2$ or fully reactive blanketing using CO_2 or CO_2/O_2.

1.6.3. Carbon dioxide (CO_2)

Carbon dioxide is the major anti-microbial factor of MAP. Generally the inhibitory effect is seen as increases in the lag phase and generation time during the logarithmic phase of growth of the organisms. The effectiveness of carbon dioxide is influenced by the original and final concentrations of the gas, the temperature of storage and the original population of organisms. Microbial growth is reduced at high concentrations of carbon dioxide in a variety of products and this effect increases as storage temperature decreases.

Geniogeorgis (1985) suggested that the antimicrobial activity of carbon dioxide was a result of the gas being absorbed onto the surface of the food forming carbonic acid, subsequent ionization of the carbonic acid and a reduction in pH. However, this minimal decrease in pH probably wound not cause any significant bacteriostatic activity. The main theories suggested have been summarised by Farber (1991):

- Alteration of cell membrane function including effects on nutrient uptake and absorption;
- Direct inhibition of enzyme systems or decreases in rate of enzyme reactions;
- Penetration of membranes resulting in changes to intracellular pH;
- Direct changes to physico-chemical properties of proteins.

The effectiveness of carbon dioxide as an anti-microbial agent is not universal and depends on the microbial flora present and the product characteristics. Yeasts which produce carbon dioxide during growth are stimulated by high levels of carbon dioxide and thus for some products where they are potentially a major cause of spoilage, MAP may not be an advisable option. Also the food-associated pathogens *Clostridium perfringens* and *Clostridium botulinum* are not affected by the presence of carbon dioxide and their growth is encouraged by anaerobic conditions. In general carbon dioxide is most effective in foods where the normal spoilage organisms consist of aerobic, gram negative psychrotropic bacteria.

1.6.4. Oxygen (O_2)

Food deteriorates due to physical, chemical and microbiological factors. Oxygen is probably the most important gas in this context being used metabolically by both aerobic spoilage micro-organisms and plant tissues and taking part in some enzymatic reactions in food including the compounds such as vitamins and flavours. For these reasons, in modified atmosphere packaging, oxygen is either excluded or the levels set as low as possible. In MAP, oxygen levels are normally set as low as possible to reduce oxidative deterioration of foods. Oxygen will generally stimulate the growth of aerobic bacteria and can inhibit the growth of strictly anaerobic bacteria, although there is a very wide variation in the sensitivity of anaerobes to oxygen.

1.6.5. Nitrogen (N_2)

Nitrogen is an inert tasteless gas, which displays no antimicrobial activity on its own. Because of its low solubility in water and fat, the presence of N_2 in a MAP food can prevent pack collapse that can occur when high concentrations of CO_2 are used. In addition, N_2, by displacing O_2 in the pack, can delay oxidative rancidity and also inhibit the growth of aerobic micro organisms. Nitrogen can also indirectly influence the micro organisms in perishable foods by retarding the growth of aerobic spoilage organisms (Farber, 1991; Phillips, 1996). The second role of nitrogen in MAP is to act as a filler gas and keeps flexible packages from developing a vacuum.

The most optimal combination of gases for a food product depends on many factors, such as the type of the product, packaging materials and storage temperature. The packaging system selected must have sufficient headspace to provide enough gas to interact with the entire product. The headspace must contain a reservoir of CO_2 to compensate for gas absorbed by the product and lost across the packaging material (Parry, 1993). The longer the required shelf-life then the larger the headspace should be.

1.6.6. Non-conventional modified atmosphere - High O_2

A number of packers of fresh prepared green vegetables in the United Kingdom have been experimenting with O_2 mixtures between 70 and 100% (Day 1996). The treatment, referred to as "oxygen shock" or "gas shock," has been found to be very effective in inhibiting enzymatic discoloration, preventing anaerobic fermentation reactions, and inhibiting aerobic and anaerobic microbial growth. High levels of O_2 can inhibit the growth of both anaerobic and aerobic microorganisms since the optimal O_2 level for growth (21% for aerobes, 0-2% for anaerobes) is surpassed. However, there have also been reports of high O_2 (that is, 80-90%) stimulating the growth of foodborne pathogens such as *Escherichia coli* and *Listeria monocytogenes* (Amanatidou and others 1999). Recent studies by Kader and Ben-Yehoshua (2000) and Wszelaki and Mitcham (2000) examining the use of superatmospheric O_2 levels to control microorganisms on produce, have found that only O_2 atmospheres close to 100 kPa or lower pressures (40 kPa) in combination with CO_2 (15 kPa), are truly effective. These requirements may be difficult to achieve in industry since working with such high O_2 levels can be hazardous due to flammability issues. As with most MAP gases, superatmospheric O_2 has varied effects depending on the commodity, and further research is required in this area to elucidate the utility of this technique in the fresh-cut produce industry.

1.6.7. Non-conventional modified atmosphere - Nitrous Oxide (N_2O)

N_2O is a naturally occurring gas produced primarily by aerobic denitrifying bacteria in the soil. The linear N_2O molecule is isosteric with CO_2 (same number of valence electrons arranged in a similar manner). N_2O is an antagonist of ethylene production and action which may be related in part to its biophysical similarity to CO_2. (Innovation in controlled atmosphere technology)

It seems that N_2O binds lipids and also proteins, such as cytochrome c oxidase (Gouble et al., 1995; Day, 1996). Sowa and Towill (1991)[1] reported the action of N_2O on partial and reversible inhibition of respiration and cytochrome c activity in mitochondria isolated from seeds, leaves or cellular suspensions.

Moreover continuous nitrous oxide gas treatment has shown a significant ripening inhibition effect by extending the lag phase, which precedes the ethylene rise and delayed colour change in pre-climacteric fruits of tomatoes and avocados. Studies in nitrous oxide prove is capacity to inhibit the proliferation of some typical micro-organisms present in modified atmosphere packaging.

2. Results and Discussion – 1st Part

2.1. Pineapple

Pineapple product is not canned in Europe, but it is used as main ingredient in fruit salad. Pineapple (*Ananas comosus L.* Merrill) is a non-climacteric tropical product that shows moderate to low rates of respiration and ethylene production. Quality of fresh-cut pineapple consists in a combination of visual appearance, texture and flavour characteristic for their type.

M. Martínez-Ferrerre *et al.* (2002) reported that a MAP system of 4% O_2, 10% CO_2, 86% N_2 maintained good colour, texture, odour and taste after 25 days of storage at 5°C. This treatment did not show detrimental effects on sensory quality compared with fresh fruits, and significantly inhibited the growth of spoilage microorganisms, particularly moulds and yeasts. MAP in combination with blanching, ascorbic acid and refrigeration had beneficial effects on the inhibition of enzymatic browning.

The aim of this study was to examine in which gas mixture the quality of freshly cut pineapple can be maintained the best. Several gas mixtures were evaluated including the optimal gas mixture reported for M. Martínez-Ferrerre et al. (2002).

In order to evaluate which is the kind of package that allows preserving minimal processed pineapple four different gas compositions, in combination with 2% ascorbic acid treatment, were analyzed for a period of 12 days at 7 °C: Air; 100% O_2; 100% N_2O; 4% O_2, 10% CO_2, 86% N_2.

2.1.1. Experimental procedures

Fresh-cut pineapples were washed, hand-peeled, dicing, dipped in ascorbic acid solution (2%) for 10 minutes and packed in different atmospheres. The number of packages for each treatment was 9, each one having approximately 200g of fruit. Pineapple cubes were placed in a CPET tray covered by APET-PA top film. All the trays were stored in a cold room at 7 °C. Three trays (of 3 sample replicates) of each treatment were randomly selected for analysis.

The O_2 and CO_2 concentrations inside the packages were measured with a gas chromatograph (GC Dansensor Checkmate II).

The quality evaluation of the fresh-cut pineapple was measured for 4 times during the experiment at day 1, 3, 7 and 12. The following aspects were evaluated: odour, sweetness, bitterness, translucency, firmness, juice leakage, taste and visual scale.

Product quality was evaluated by an untrained panel composed per two persons.

Visual quality scale was scored based on the following hedonic scale: 9 = excellent (essentially no symptoms of deterioration); 7= good (minor symptoms of deterioration, not objectionable); 5 = fair (deterioration evident, but not serious, limit of saleability); 3 = poor (serious deterioration, limit of saleability) and 1 = *extremely poor (*not usable, off-odours, fungal decay).

Texture was evaluated by chewing process and was scored as: 1 = pineapple texture, 0 = some texture and -1 = no pineapple texture. Juice leakage was scored as: 1= no juice leakage, 0 = some juice leakage, -1 = juice leakage. Sweetness was scored as: 1 = sweet, 0 = some sweetness and -1 = no sweetness. Bitterness was scored as: 1 = no bitter taste, 0 = some bitter taste -1 = bitter taste. Translucency was scored as: 1 = no translucency, 0 = some translucency -1 = translucency. Taste was scored in good, fair or bad.

The evaluation of odours in the headspace was determined immediately after opening the package and after some minutes to check if it persisted. Odours were scored as 1= pineapple odour, 0 = some pineapple odour and -1 = no pineapple odour.

2.1.2. Results and discussion

All the product quality evaluation results are showed in Appendix 1. The treatment that better preserved the pineapple characteristics was: 4% O_2, 10% CO_2, 86% N_2. This treatment kept the pineapple initial colour, odour and flavour and maintained a better fresh-like texture. The pineapple with this treatment after 12 days kept the bright yellow colour and strong sweet-bitter flavour and aroma. Pineapple under this treatment showed a more desirable colour and visual appearance than the other treatments.

Pineapple of other treatments, at the 7^{th} day of storage, lose texture and begun to look translucency. Texture of fresh-fruits becomes soggy as the cell walls break down and the cells lose water.

Juice leakage was observed in all the four treatments.

Changes in CO_2 and O_2 concentrations inside the package of fresh-cut pineapple are shown in figure 8.

A relatively high concentration of CO_2 (10%) inhibits microbial growth and with a low concentration of O_2 the fruits continues is respiration process and inhibit the switch from aerobic to anaerobic environment. High oxygen package had a higher concentration of oxygen that the other three packages and also resulted in a higher concentration of CO_2.

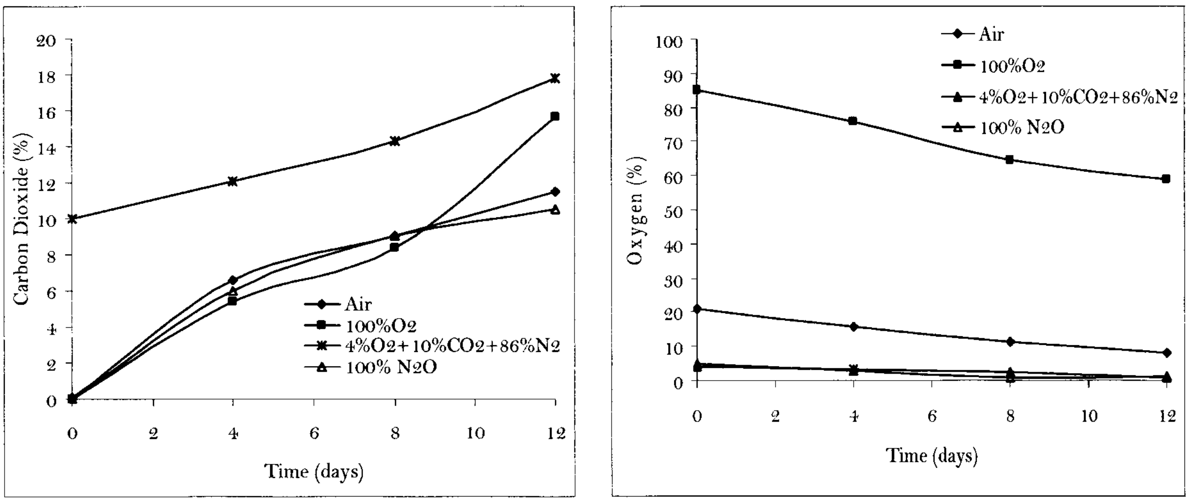


Figure 8 - Carbon dioxide and oxygen concentration (%) in different packages of fresh-cut pineapple during 12 days of storage at 7°C.

With the other tested treatments the main problem observed was the appearance of off-odours. With these same treatments at the 7th day of the experiment and thereafter pineapple fruits lost colour and became translucent. Flesh translucency, also called porosity, is associated with greater fruit sensitivity to mechanical injury, indicated by leakage of cellular fluids.

The major indications of quality loss for all the treatments tested and the respective shelf-life are shown in table 3.

Table 3 - Shelf-life of fresh-cut pineapple packaged in different gas compositions stored at 7°C

Gas composition	**Shelf-life (days)**	**Limiting factor**
Air (20%O_2, 80% N_2)	7	Loss of firmness and translucency
100% O_2	7	Loss of firmness, translucency and off-odours
100% N_2O	7	Loss of firmness, translucency and off-odours
4% O_2, 10% CO_2, 86% N_2	>12	None

2.2. Melon

Muskmelons (*Cucumis melo* L. var. reticulatus) are climacteric fruits and the main factors affecting their eating quality are texture, colour, flavour and sweetness.

Irene Luna-Guzma'n et al. (2000) found that fresh-cut melon cylinders dipped for 1 min in 2.5% solutions of either calcium chloride ($CaCl_2$) at ≈25 °C or calcium lactate at ≈25 °C and 60 °C helps to maintain firmness throughout cold storage.

The aim of this study was to examine the melon reaction to different mixture of gases and to some dipping solutions. Some analyses were based on literature and others were based in free choice of modified atmospheres with the goal of achiving better results.

In order to evaluate which is the kind of package and/or dipping solution that allows preserving minimal processed melon, different gas compositions and two dipping solutions were analyzed for a period of 12 days at 7 °C: Air; 100% O_2; 80% O_2, 20 % N_2; 95% N_2, 5% O_2; 90% N_2, 5% O_2, 5% CO_2; 100% N_2O, calcium chloride and calcium lactate solutions.

2.2.1. Experimental procedures

Fresh-cut melon were washed, hand-peeled, dicing, and some samples dipped into: 2.5% (w/v) $CaCl_2$ or 2.5% (w/v) Calcium lactate for 1 minute and then packaging in different atmospheres. The number of packages for each treatment was 9, each one having approximately 200g of fruit. Melon cubes were placed in a CTEP tray covered by APET-PA top film. All the trays were stored in a cold room at 7 °C. Three trays (triplicate samples) of each treatment were randomly selected for analysis.

Product quality was evaluated by an untrained panel composed per two persons.

The quality evaluation of the fresh-cut melon was measured for 12 days. The following aspects were evaluated: odour, sweetness, bitterness, crunchiness/hardness, juice leakage and taste.

Product quality was evaluated by an untrained panel composed per two persons.

Texture (crunchiness/hardness) was evaluated by chewing process and was scored as: 1 = melon texture, 0 = some texture and -1 = no melon texture. Juice leakage was scored as: 1= no juice leakage, 0 = some juice leakage, -1 = juice leakage. Sweetness was scored as: 1 = sweet, 0 = some sweetness and -1 = no sweetness. Bitterness was scored as: 1 = no bitter taste, 0 = some bitter taste -1 = bitter taste. The evaluation of odours in the headspace was determined immediately after opening the package and after some minutes to check if it persisted. Odours were scored as: 1= melon odour, 0 = some melon odour and -1 = no melon odour.

2.2.2. Results and discussion

All the product quality evaluation results are showed in Appendix 2. Of all the analyzed packaging atmospheres, the one that better preserved the melon's essential properties (texture, flavour and sweetness) was 100% of O_2.

This atmosphere holds the typical melon flavour as well as it keeps the natural melon sweetness and its characteristic foreign bitter taste. For the period of 12 days, this gas composition maintained the analyzed parameters unchanged at least at sensorial level. Juice leakages and strong bitterness were not observed. The use of high oxygen levels in treatment was effective on inhibition of enzymatic discoloration and on the preventing of anaerobic fermentation reactions. In the package water condensation was observed due to the fruit respiration.

So, it could be concluded that the 100% O_2 atmosphere is efficient and allows preserving the quality of minimal processed melons for at least 12 days at 7°C.

The efficiency of the Calcium lactate and $CaCl_2$ to preserve firmness was also evaluated. The results shown both types of dipping had equal efficiency and in combination with air (20% O_2, 80% N_2) they could hold the melon quality parameters for at least during 12 days at 7 °C.

The main problems observed with the other packaging gases (with or without dipping solutions) were: translucency of melon cubes, appearance of off-flavours (ex: yogurt flavour) and fungal growth.

The major indications of quality loss for all the treatments tested and the respective shelf-life are shown in table 4.

Table 4 - Shelf-life of fresh-cut melon packaged in different gas compositions stored at 7°C

Gas composition	Shelf-life (days)	Limiting factor
Air (20% O_2, 80%N_2)	8	Off-flavours
100% O_2	> 12	None
80% O_2, 20%N_2	8	Off-flavours
95% N_2, 5% O_2	8	Yogurt flavour
90% N_2, 5% O_2, 5% CO_2	8	Yogurt flavour and loss of firmness
100% N_2O	8	Loss of firmness and translucency
100% N_2O (plus $CaCL_2$)	8	Off-flavours and fungus
Air (plus Calcium Lactate)	> 12	None
Air (plus $CaCL_2$)	> 12	None

2.3. Pears

Fresh-cut pear might be a suitable component for fruit-salad or other food service or consumer products, but are subject to rapid enzymatic browning and tissue breakdown.

X.Dong et al. (2000) reported that a combination treatment of 0,01% 4-hexylresorcinol (4-HR), 0,5% ascorbic acid and 1,0% calcium lactate can provide 15 to 30 days shelf-life for Anjou, Bartlett and Bosc pears when the pears are sliced at the average ripeness of 43, 49 and 38 N respectively, with a 2 minutes dipping, and partial vacuum packaging. Adel A. Kader et al. (2001) found that a post-cutting dip of 2% ascorbic acid, 1% calcium lactate and 0.5% Cysteine adjusted to pH 7.0 did significantly extend shelf-life of 'Bartlett' pear slices, by inhibiting loss of slice flesh firmness and preventing cut surface browning. They also report that low O_2 (0.25 or 0.5 kPa) elevated CO_2 (air enriched with 5, 10 or 20 kPa CO_2), or superatmospheric O_2 (40, 60, or 80 kPa) atmospheres alone did not effectively prevent cut surface browning or softening of fresh-cut pear slices.

The aim of this study was to make various experiments to examine the response of the freshly cut pear to different mixture of gases and to some dipping solutions. Some analyses were based on literature and others were based on own experience.

Quality of fresh-cut pear consists in a combination of good colour (no enzymatic browning), texture and flavour characteristic for their type.

In order to evaluate which is the kind of package that allows preserving fresh-cut pear four different gas compositions and two dipping solutions were analyzed for a period of 12 days at 7 °C: Air (with and without dipping solution); 100% O_2 (with and without dipping solution); 100% N_2 (with and without dipping solution); 80% O_2, 20% N_2 (with and without dipping solution).

2.3.1. Experimental procedures

Fresh-cut apples were washed, hand-peeled, coring, dicing, and some of them dipped in two different solutions: (1). 0,01% (w/v) 4-hexylresolcinol (4-HR), 0,5% (w/v) ascorbic acid, 1,0% (w/v) calcium lactate; (2). 2% (w/v) ascorbic acid, 1% (w/v) calcium lactate and 0.5% (w/v) Cysteine (adjust to pH 7.0) for 2 and 5 minutes respectively and then package in different atmospheres. The number of packages for each treatment was 9, each one having approximately 200g of fruit. Pear slices were placed in a CPET tray covered by APET_PA top film. All the trays were stored in a cold room at 7 °C. Three trays (triplicate samples) of each treatment were randomly selected for analysis.

The O_2 and CO_2 concentrations inside the packages were measured with a gas chromatograph (GC Dansensor Checkmate II). The quality of the freshly cut pears was evaluated during 12 days (d1, 4, 8, 12). The following aspects were evaluated: browning, firmness, crispness, initial juiciness, crunchiness, mealiness, flouriness, odour, taste and visual scale. Product quality was evaluated by an untrained panel composed per two persons. Percentage of pear browning was evaluated using the subjective test shown in table 5.

Table 5 - Subjective test of browning area (%) for colour evaluation of fresh-cut pears.

Scale	Browning area (%)
0	white
1	< 5% surface
2	< 20%
3	< 33%
4	< 50%
5	> 50%

Firmness was evaluated by breaking manually pieces of pear and was score in: 1 = firmness, 0 = some firmness and -1 = no firmness. Crispness (1 = crispness, 0 = some crispness and -1 = no crispness), initial juiciness (1 = initial juiciness, 0 = some initial juiciness and -1 = no initial juiciness), crunchiness (1 = crunchiness, 0 = some crunchiness and -1 = no crunchiness) , mealiness (1 = no mealiness , 0 = some mealiness and -1 = mealiness), flouriness (1 = flouriness, 0 = some flouriness and -1 = no flouriness) and taste (good, fair and bad) were evaluated by chew process.

Visual quality scale was scored based on the following hedonic scale: 9 = excellent (essentially no symptoms of deterioration); 7= good (minor symptoms of deterioration, not objectionable); 5 = fair (deterioration evident, but not serious, limit of saleability); 3 = poor (serious deterioration, limit of saleability) and 1 = extremely poor (not usable, off-odours, fungal decay).

The evaluation of odours in the headspace was determined immediately after opening the package and after some minutes to check if it persisted. Odours were score on 1= pear odour; 0 = some pear odour and -1 = no pear odour.

2.3.2. Results and discussion

All product quality evaluation results are showed in Appendix 3. Of all the analyzed combinations of pre-treatment and initial gas atmospheres the one that the pears the best in time, were: 100% O_2 (2% ascorbic acid, 1% calcium lactate, 0.5% Cysteine); 100% N_2 (0,01% 4-hexylresolcinol (4-HR), 0,5% ascorbic acid, 1,0% calcium lactate; and 100% N_2 (2% ascorbic acid, 1% calcium lactate, 0.5% Cysteine)

The development of the gas composition of the headspace of the pear packages is shown in Figure 9.

Pear slices kept in air and 85% O_2 atmosphere showed similar variance in the CO_2 and O_2 concentrations during storage time. However, pear slices with 85% O_2 atmosphere (4-HR and Cysteine solutions) exhibited much less tissue necrosis and cut surface browning. Oxygen concentration showed no significant changes throughout the storage period in package filled with 100% N_2. However, a strong increase in respiration was monitored in all cases as a consequence of the wounding produced by cutting. Hence CO_2 concentrations rose to values of 25% after 12 days of storage.

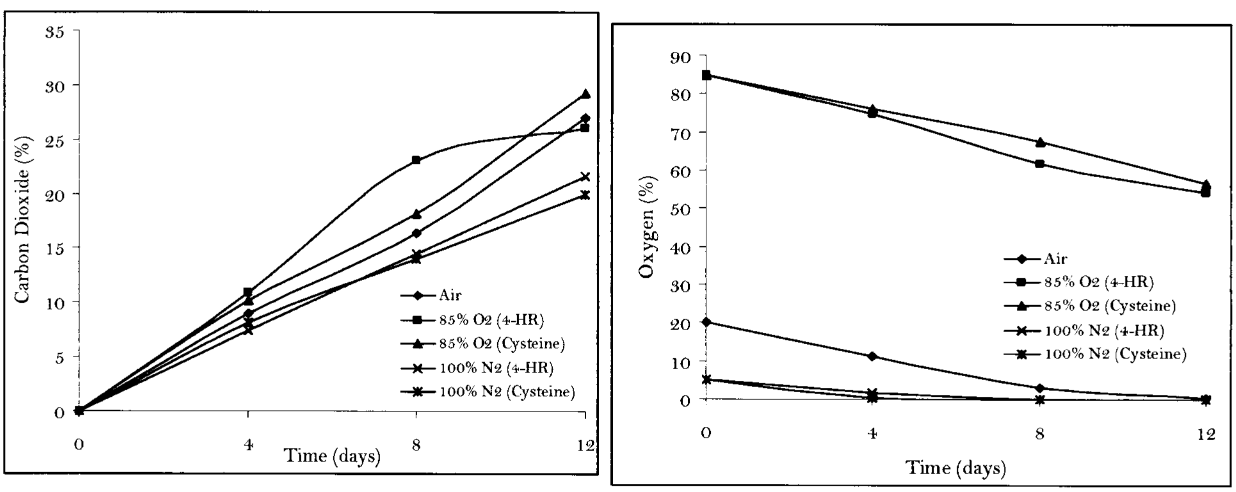


Figure 8 - Carbon dioxide and oxygen concentration (%) in different packages of fresh-cut pears during 12 days of storage at 7°C.

Browning of minimally processed pears occurred less when they are dipped in cysteine solutions (100% O_2 and 100% N_2). High CO_2 injury occurred in a dose responsive manner with damage occurring earlier and more severe in air and samples with 4-HR dipping solution

(100% O_2 and 100% N_2) (figure 9). It must be considered that the carbon dioxide levels were always in higher concentrations since the beginning of storage and when the levels of CO_2 are above 10% symptoms like tissue browning, necrosis and softening become visible since pears are very susceptible to CO2 injury. Tissue necrosis and severe surface browning first occurred in the flesh tissue close to the core and spread radially outward over time. Loss of gloss at the cut surface as well as development of an abrasive surface texture were noticed in 100% N_2 atmosphere, because of mass loss in water content.

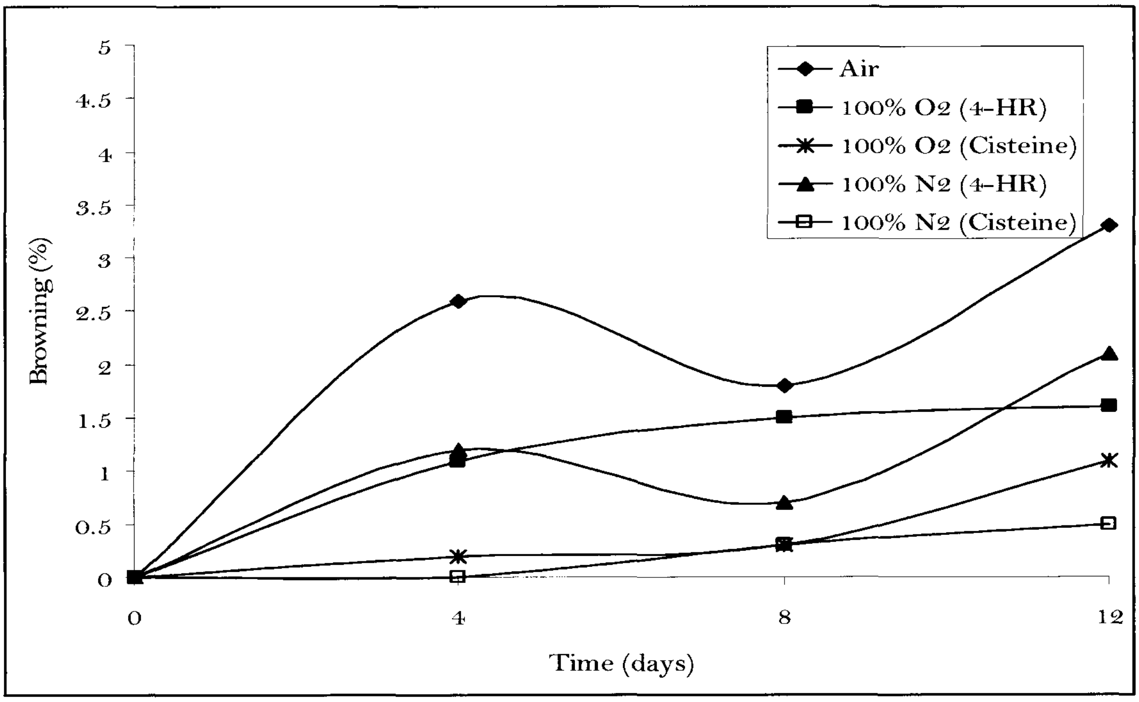


Figure 9 - Enzymatic browning values of the modified atmospheres pears samples during refrigerated storage during 12 days.

Between the 1st and the 4th day of storage, firmness and texture (firmness, crispness, initial juiciness, crunchiness, mealiness and flouriness) declines for samples package in air and 100% O_2 (4-HR). In all the other three treatments these two parameters had remained unalterable.

The most efficient treatment to avoid off-odours was cysteine in combination with 100% O_2 or 100% N_2, just on the 12 day of storage was noticed a light odour of alcohol. All the other treatments evidenced alcohol odour since the 4 day of analyse.

Throughout the 12 days of storage just 100% N_2 (with cysteine) atmosphere keep the pears taste unalterable, all the other treatments lost pear taste during this time.

- Enzymatic browning values of the modified atmospheres samples during refrigerated storage during 12 days

The low quality of pears during the storage time results of changes in colour by enzymatic browning, flavour loses and off-odours. The major indications of quality loss for all the treatments tested and the respective shelf-life are shown in table 6.

Table 6 - Shelf-life of fresh-cut pears packaged in different gas compositions stored at 7°C

Gas composition	**Shelf-life (days)**	**Limiting factor**
Air (20%O_2, 80% N_2)	< 4	Colour (enzymatic browning), fair taste and off-odours
100% O_2 (with 4-HR)	8	Colour (enzymatic browning) and fair taste
100% O_2 (with cysteine)	8	Fair taste and off-odours
100% N_2 (with 4-HR)	8	Fair taste and off-odours
100% N_2 (with cysteine)	>12	None

2.4. Kiwifruit

Consumer preference for kiwifruit (*Actinidia deliciosa*) is determined primarily by the sugar-acid balance with fruit firmness and fruit volatile content. Loss of cellular compartmentalisation, due to peeling and cutting, causes mixing of previously sequestered metabolites of the ethylene generating system stimulating ethylene production. Enzymatic activities, softening and ripening of kiwifruit are promoted by ethylene. Therefore, it is difficult to maintain the quality of kiwifruit slices once they have been cut. Agar et al. (1999) reported that fresh-cut kiwifruit slices had a shelf-life of 9-12 days if treated with 1% calcium chloride or 2% calcium lactate, and stored at 0-2°C and 90% RH in an C_2H_4-free atmosphere of 2kPa O2 and/or 10 kPa CO_2 calcium treatments and modified atmosphere were used. Pietro Rocculi et al. (2004) found that a modified atmosphere with 90% N_2O was the best mixture of tested gases in order to maintain the quality of kiwifruit slices.

The objective of this study was to examine the kiwifruit reaction to different mixture of gases. Several gas mixtures were evaluated including the optimal gas mixture reported for Pietro Rocculi et al (2004) and Agar et al. (1999).

In order to evaluate which is the kind of package that allows preserving minimal processed kiwifruit, three different gas compositions and a calcium chlorite dip were tested for a period of 12 days at 7 °C: Air; ; 100% N_2O; 10% O_2, 90% N_2.

2.4.1. Experimental procedures

Fresh-cut kiwifruits were washed, hand-peeled, dicing, and some samples dipped (in: 2% (w/v) $CaCl_2$) for 5 minutes and then packed in different atmospheres. The number of packages for each treatment was 9, each one containing, approximately, 200g of fruit. Kiwifruit packages were placed in a CPET tray covered by APET-PA top film. All the trays were stored in a cold room at 7 °C.

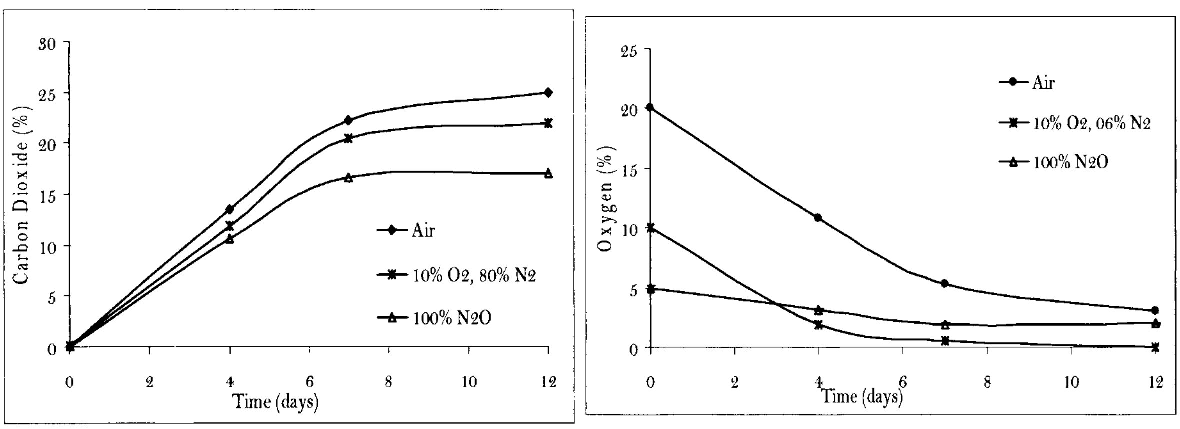
The O_2 and CO_2 concentrations inside the packages were measured with a gas chromatograph (GC Dansensor Checkmate II). The quality evaluation of the fresh-cut kiwifruit was measured for 12 days (d1, 3, 7, 12). The following aspects were evaluated: odour, sweetness, acidity, translucency, colour, texture, juiciness, juice leakage, taste and visual scale.

Product quality was evaluated by an untrained panel composed per two persons.

2.4.2. Results and discussion

All product quality evaluation results are showed in Appendix 4. In general, 100% N_2O and 10% O_2, 90% N_2 modified atmospheres showed analogous results in kiwifruit slices during storage. These atmospheres had been good in preserving odour, sweetness, texture, juiciness and prevent juice leakage of samples for only 7 days. However they were not effective in inhibiting the phenomena of flesh and pericarp translucency responsible of the low quality of kiwifruit as well as the very characteristic acidity. The control (air) and 100% O_2 samples were very affected by browning after only 4 days, in both pericarp and core surfaces.

The development of the gas composition of the packaging headspaces of fresh-cut kiwifruits is shown in figure 10. Packages with 100% N_2O had the lowest CO_2 production during storage and the biggest value of CO_2 production was reached for packages with air as expected. The O_2 levels in packages with air and 100% N_2O was almost depleted and totally consumed in packages with 10% O_2, 90% N_2. The observed O_2 and CO_2 results in packages with 100% N2O confirm the inhibition effect on vegetables tissue respiration reported to Sowa and Towill (1991).



ure 10 - Carbon dioxide and oxygen concentration (%) in different packages of fresh-cut kiwifruit during lays of storage at 7°C.

The main problems observed in the packages were the colour changes of kiwifruit (flesh and core translucency) and acidity reduction. The major indications of quality loss for all the treatments tested and the respective shelf-life are shown in table 7.

Table 7 - Shelf-life of fresh-cut kiwifruit packaged in different gas compositions stored at 7°C

Gas composition	**Shelf-life (days)**	**Limiting factor**
Air (20%O_2, 80% N_2)	< 4	Translucency, lose of texture and acidity
100% N_2O ($CaCl_2$)	7	Translucency, lose of texture and acidity
10% O_2, 90% N2($CaCl_2$)	7	Translucency, lose of texture and acidity

2.5. Apple

Sensory quality of apple fruits is dependent on a number of factors including appearance, texture, taste (sweet, sour, salt and bitter sensations) and aroma (volatile compounds). Among these attributes, combination of visual appearance, texture and flavour appear to be the most important for the consumer. Enzymatic browning is one of the principal quality deterioration factors of minimally processed apples. Browning reactions in apple become evident when, for instance, fruit is subjected to processing or mechanical injury. As for other fruits, methods to reduce senescence and browning processes in cut apples are being investigated. Adel A. Kader et al. (1998) report that apple slices treated with 2% ascorbic acid and held in an atmosphere of 0 kPa O_2 (100% N_2) at 10 °C had no significant browning or loss of visual quality for up to 15 days. Pietro Rocculi et al. (2004) showed that a non- conventional atmosphere (with Ar and N_2O) packaging, combined with a 0,5% of ascorbic acid, 0,5% of citric acid, and 0,5% of calcium chloride dipping treatment could permit the fresh quality maintenance of minimal processed apples for 12 days. C. Y. Lee (2000) report that 4-hexyl resorcinol was very effective in preventing apple browning and X. Dond showed that a combination of 2 minutes treatment of 0,01% 4-hexylresorcinol (4-HR), 0,5% ascorbic acid and 1,0% calcium lactate can provide 15 to 30 days of shelf life in some species of pears.

The objective of this study was to examine the apple reaction to different mixture of gases and different chemical treatments. In order to evaluate which is the kind of package that allows preserving fresh-cut apple five different gas compositions and three dipping solutions were analyzed for a period of 12 days at 7 °C: Air (0.5% ascorbic acid, 0.5% citric acid, 1% calcium chloride); 80% O_2, 20% N_2 (0.5% ascorbic acid, 0.5% citric acid, 1% calcium chloride); Air (2% ascorbic acid); 100% N_2(2% ascorbic acid); Air (0.01% 4-HR, 0.5% ascorbic acid, 1.0% calcium lactate); 100% N2O (0.01% 4-HR, 0.5% ascorbic acid, 1.0% calcium lactate); 100% O_2 (0.01% 4-HR, 0.5% ascorbic acid, 1.0% calcium lactate).

2.5.1. Experimental procedures

Fresh-cut apples were washed, hand-peeled, dicing, and dipped in: 0.5% ascorbic acid, 0.5% citric acid, 0.5% calcium chloride (10 minutes dipping), 2% ascorbic acid (5 minutes dipping and pH = 2,5), 0.01% 4-HR, 0.5% ascorbic acid, 1.0% calcium lactate (2 minutes dipping) and then packaging in different atmospheres. The number of packages for each treatment was 9, each one containing approximately 200g of fruit. Apples packages were placed in a plastic tray covered by plastic film. All the trays were stored in a cold room at 7 °C.

The O_2 and CO_2 concentrations inside the packages were measured with a gas chromatograph (GC Dansensor Checkmate II).

The quality evaluation of the fresh-cut apple was measured for 12 days (d1, 5, 8, 12). The following aspects were evaluated: browning, firmness, crispness, initial juiciness, crunchiness, mealiness, flouriness, odour, taste and visual scale.

Product quality was evaluated by an untrained panel composed per two persons.

Percentage of pear browning was evaluated using the following subjective test:

Table 8. Subjective test of browning area (%) for colour evaluation of fresh-cut apples.

Scale	**Browning area (%)**
0	white
1	< 5% surface
2	< 20%
3	< 33%
4	< 50%
5	> 50%

Firmness was evaluated by breaking manually pieces of apple and was score in: 1 = firmness, 0 = some firmness and -1 = no firmness. Crispness (1 = crispness, 0 = some crispness and -1 = no crispness), initial juiciness (1 = initial juiciness, 0 = some initial juiciness and -1 = no initial juiciness), crunchiness (1 = crunchiness, 0 = some crunchiness and -1 = no crunchiness) , mealiness (1 = no mealiness , 0 = some mealiness and -1 = mealiness), flouriness (1 = flouriness , 0 = some flouriness and -1 = no flouriness) and taste (score: good, fair and bad) were evaluated by chew process.

Visual quality scale was scored based on the following hedonic scale: 9 = excellent (essentially no symptoms of deterioration); 7= good (minor symptoms of deterioration, not objectionable); 5 = fair (deterioration evident, but not serious, limit of saleability); 3 = poor (serious deterioration, limit of saleability) and 1 = extremely poor (not usable, off-odours, fungal decay).

The evaluation of odours in the headspace was determined immediately after opening the package and after some minutes to check if it persisted. Odours were score on 1= apple odour; 0 = some apple odour and -1 = no apple odour.

2.5.2. Results and discussion

All product quality evaluation results are showed in Appendix 5. Of all the analyzed conditions, the one that better preserved the most important apple properties (visual appearance, texture and flavour) was the one holding 100% of N_2 with a chemical treatment of 2% ascorbic acid.

The apples packed in 100% of N_2 (2% of ascorbic acid) showed, without any doubt, the best colour during the storage time. Apples package with ascorbic acid treatments (in air or 100% of N_2 atmosphere) had great results in all analyzed parameters. The percentage of browning area for packages in 100% N_2 (2% ascorbic acid) was only 0,3 on the end of 12 days while apples packed in 100% of N_2O (0,01 4HR, 0,5% ascorbic acid, 1,0% calcium lactate) had 4,2. 4-HR proved not to be an efficient browning inhibitor in all tested packages (figure 11).

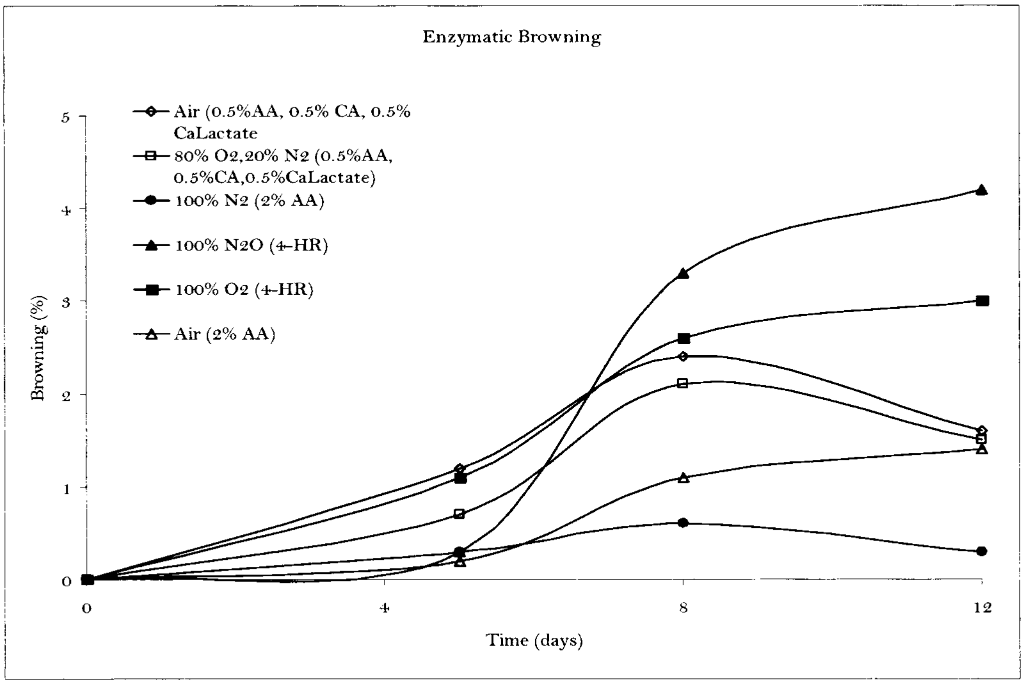


Figure 11 - Enzymatic browning values of the modified atmospheres samples during refrigerated storage during 12 days.

Changes in CO_2 and O_2 concentrations inside the package of fresh-cut apples are shown in figure 12. The amount of oxygen consumed was less in 100% N_2 and 100% N_2O packages. The packages with high oxygen atmospheres showed largest oxygen consumption rate. Relatively to the production of CO_2 all the packages stayed bellow the considered detrimental level (> 20%), however, the biggest value was observed on the best package (100% N_2 with 2% ascorbic acid dipping).

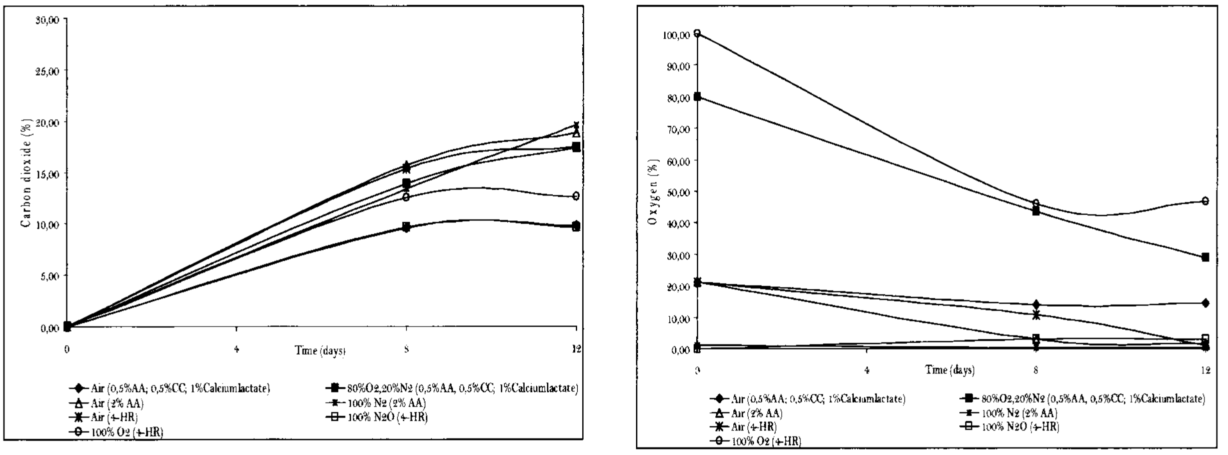


Figure 12 Carbon dioxide and oxygen concentration (%) in different packages of apples during 12 days of storage at 7°C

Any of chemical treatments were good in maintained the texture of apples, this parameter did not suffered any changes. Apple slices remained with excellent firmness, crispness, initial juiciness, crunchiness, flouriness and without mealiness.

In respect to odour, results the atmosphere with 100% N_2 was also the only one that did not had any off-odour. Apple slices had been with perfect fresh fruit odour during the 12 days of storage. The other atmospheres since the eight day of storage developed alcohol odour derived to the respiration of fruits.

About the apples taste the worse results had been the ones that had been treated with 4-HR, they developed a strong bitter taste, probably derived to 4-HR flavour. The other packages were in perfect conditions of taste.

In the end of 12 days the visual appearance was bad in treatments with 4-HR, good in treatments with 0.5% ascorbic acid, 0.5% citric acid, 0.5% calcium chloride and excellent in apple slices treated with 2% ascorbic acid.

Low quality of apple during the storage time results of changes in colour by enzymatic browning loses, flavour loses and off-odours. The major indications of quality loss for all the treatments tested and the respective shelf-life are shown in table 8.

Table 9 - Shelf-life of fresh-cut apples packaged in different gas compositions stored at 7°C

Gas composition/Chemical treatments	**Shelf-life (days)**	**Limiting factor**
Air (0.5% ascorbic acid, 0.5% citric acid, 0.5% calcium chloride)	8	Off-odours
80%O_2, 20% N_2 (0.5% ascorbic acid, 0.5% citric acid, 0.5% calcium chloride)	8	Off-odours
Air (2% ascorbic acid)	12	Off-odours
100% N_2 (2% ascorbic acid)	>12	None
Air (4-HR)	5-8	Colour (enzymatic browning), fair taste and off-odours
100% N_2O (4-HR)	5-8	Colour (enzymatic browning), fair taste and off-odours
100% O_2 (4-HR)	5-8	Colour (enzymatic browning), fair taste and off-odours

2.6. Conclusions – 1st Part

The main conclusions in the end of this study were:

- Pineapple – The best gas mixture to preserve the quality in fresh-cut pineapple was: 4% O_2, 10% CO_2, 86% N_2 in combination with a dip solution of 2% ascorbic acid provide more than 12 days of shelf-life;
- Melon – The best gases mixtures to preserve the quality in fresh-cut melon were: 100% O_2 or air (20% O_2, 80% N_2) in combination with a dip solution of 2,5% calcium lactate or 2,5% $CaCl_2$ provide more than 12 days of shelf-life;
- Pears – The best gas mixture to preserve the quality in fresh-cut pears was: 100% N_2 in combination with a dip solution of 2% ascorbic acid, 1% calcium lactate and 0.5% Cysteine provide more than 12 days of shelf-life;
- Kiwifruit – The best gases mixtures to preserve the quality in fresh-cut kiwifruit were: 100% N_2O or 10% O_2, 90% N_2 in combination with a dip solution of 2% $CaCl_2$ provide 7 days of shelf-life;
- Apple – The best gases mixtures to preserve the quality in fresh-cut apple were: air (20% O_2, 80% N_2) or 100% N_2 in combination with a dip solution of 2% ascorbic acid provide 12 and more than 12 days respectively of shelf-life.

MAP in combinations with chemical treatments and refrigeration had beneficial effects on the inhibition of enzymatic browning, firmness loss and decay of fresh-cut fruits. However, the pre-treatments alone (so combined with packaging under air) also proved to be efficient in extending the shelf-life in almost analysed fruits.

Maturity stage is an important parameter deserving consideration when producing minimally processed fruits. MAP success depends on different factors such as: good quality of initial product, good manufacturing practices, correct gas mixture and refrigeration of the product. Appearance of off-flavours and off-odours, changes in colour were the main limiting factors for fruits shelf life.

Further studies are necessary to determine the microbiological stability throughout the potential shelf-life obtained when combining dipping treatment with conventional or non-conventional atmosphere in fruits.

3. Results and discussion – 2nd Part

3.1. Fruit salad

Fruit salad is a product prepared from a mixture of basic cut fruits like pineapple, kiwifruit, papaya, mango, melon, apple, pear, banana and other fruits.

Fresh-cut products have increased their popularity, due to their convenience and fresh characteristics. Consumers demand for various fresh-cut fruits, particularly tropical fruits, is increasing rapidly in the world wide.

As well known, fruit is essential on human's beings feeding, so there is a look for in the market of non-processed fruit. We can already find fruits in market although not fresh; they contain addition of sugar and conserving additives.

So, the main objective of this experiences and study is to produce a mixed fruit salad of fresh-cut fruits, which consumers have the confidence to buy and consume the way they want.

Fresh-cut fruit salad quality consists in a combination of some parameters: Visual characteristics – fruit salad shall have a colour characteristic of the mixed minimally processed fruit; Flavour – Fruit salad shall have a normal flavour and odour characteristic for the blend of fruit; Texture – the texture of fruits shall be appropriate for the respective fruit.

The intention of this study was to determine the quality and shelf-life of various mixtures of fruits using for that the optimal conditions that we found for each type of fruit (in the first part of the work). With these optimal conditions the best combinations of pre-treatments and packaging gas mixtures are meant.

In order to evaluate what of package & pre-treatments solutions preserves fresh-cut fruit salad best, the following gas initial compositions were tested for a period of 12 days at 7 °C: Air (20% O_2, 80% N_2) , high oxygen atmosphere (87% O_2, 13% N_2), nitrogen atmosphere (96% N_2 , 4% O_2) and 100% N_2O atmosphere.

3.1.1. Experimental procedures

Fresh-cut fruit were washed, hand-peeled, dicing and dipped in diverse chemical solutions during different times (table 10).

Table 10 – Chemical treatments and respective dipping time used in fruits.

Fruit	Chemical treatment	t (dipping) minutes
Pineapple	2% Ascorbic Acid	10
Kiwifruit	1% $CaCl_2$	5
Pear	2% Ascorbic Acid + 1% Calcium lactate + 0.5% Cysteine	5
Apple	2% Ascorbic Acid	5
Melon	2.5% Ca lactate	1
Grapes	50% ethanol	0,05

After dipped the fruits were immediately package in the following atmospheres:

- Fruit Salad 1 - Air (20% O_2, 80% N_2) – pineapple, apple, pear and melon;
- Fruit Salad 2 - Air (20% O_2, 80% N_2) – pineapple, apple, pear, melon and red grape;
- Fruit Salad 3 - High oxygen atmosphere (87% O_2, 13% N_2) – pear and melon;
- Fruit Salad 4 - High oxygen atmosphere (87% O_2, 13% N_2) – pear, melon and grape;
- Fruit Salad 5 - Nitrogen atmosphere (96% N_2 , 4% O_2) – apple, kiwifruit and pear;
- Fruit Salad 6 - Nitrogen atmosphere (96% N_2 , 4% O_2) – apple, kiwifruit, pear and grape;
- Fruit Salad 7 - 100% N_2O atmosphere – pineapple, kiwifruit and melon;
- Fruit Salad 8 - 100% N_2O atmosphere – pineapple, kiwifruit, melon and grape.

The number of packages for each fruit salad was 9, each of about 300g of fruit. Minimal processed fruits were placed in a CPET tray covered by APET-PA top film. All the trays were stored in a cold room at 7 °C. Three trays (triplicate samples) of each treatment were randomly selected for analysis.

The O_2 and CO_2 concentrations inside the packages were measured with a gas chromatograph (GC Dansensor Checkmate II). The quality evaluation of the fresh-cut fruits was measured for 12 days (d1, 4, 8, 12). Three trays (triplicate samples) of each treatment were randomly selected for analysis. The following aspects were evaluated for each type of fruit constitutes the fruit salad: odour, colour, texture, taste, visual scale and juice leakage.

The product quality was evaluated by an untrained panel composed per two persons.

The evaluation of odours in the headspace was determined immediately after opening the package and after some minutes to check if it persisted. Odours were score on 1= fruit odour; 0 = some fruit odour and -1 = disturbing off-odour.

Texture was evaluated by breaking manually pieces of fruit or by chewing fruit pieces (depending on the fruit) and was score in: 1 = good texture, 0 = some texture and -1 = no texture. The colour was evaluated by analyse of individually peaces of each fruit that constitutes the salad fruit: 1 = characteristic colour, 0 = some characteristic colour and -1 = no characteristic colour. Taste was scored in good, fair or bad. Juice leakage was score in 1 = no juice leakage, 0 = some juice leakage and -1 = juice leakage.

Visual quality scale was scored based on the following hedonic scale: 9 = excellent (essentially no symptoms of deterioration); 7 = good (minor symptoms of deterioration, not objectionable); 5 = fair (deterioration evident, but not serious, limit of saleability); 3 = poor (serious deterioration, limit of saleability) and 1 = *extremely poor* (not usable, off-odours, fungal decay).

3.1.2. Fruit Salad 1 and 2

All product quality evaluation results are showed in Appendix 6. Fruit salad 1 and 2 were both composed by pineapple, apple, pear and melon, the difference was that red grape was incorporated in fruit salad 2.

Oxygen consumption and carbon dioxide production, in fruit salads 1 and 2 in the end of storage time, were very similar; however in fruit salad 2 the respiration of fruits was faster. Oxygen values approached zero; almost all oxygen was consumed in both fruit salads. In relation to carbon dioxide, after 12 days of storage, air atmosphere showed CO_2 content (> 20%) which potentially harmful for fruit quality. The CO_2 values were approached 30% after 12 days, although was not verified loss of quality in any fruit was detected with exception of apple.

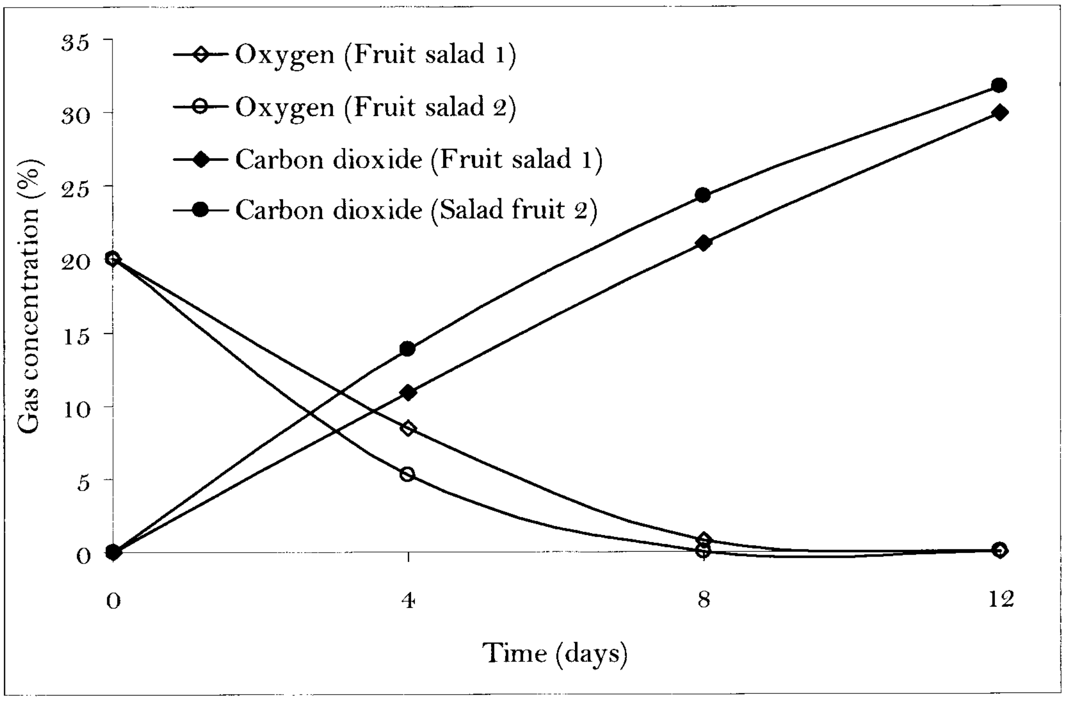


Figure 13 - Carbon dioxide and oxygen concentration (%) of fruit salad 1 and fruit salad 2 packed in air (20% O_2, 80% N_2) during 12 days of storage at 7°C.

The limiting factor of fruit salad 1 was apple slices appearance, in the 4th day of storage they already presented different colour than the original. This rapid deterioration of apple slices happened due to the fast consumption of oxygen, this led to the oxidation of phenolic compounds by the enzyme polyphenoloxidase (PPO). This resulted in browning reactions and loss of water that yielded in a dry appearance. However in fruit salad 2 that phenomenon did not occurred.

Odour, texture and taste had remained unalterable during the storage time for both fruit salads.

3.1.3. Fruit Salad 3 and 4

All product quality evaluation results are showed in Appendix 7. Fruit salad 3 and 4 were both composed by pear and melon, the difference was that red grape was incorporated in fruit salad 4.

Oxygen consumption and carbon dioxide production in both fruit salad was huge (≈ 40%).

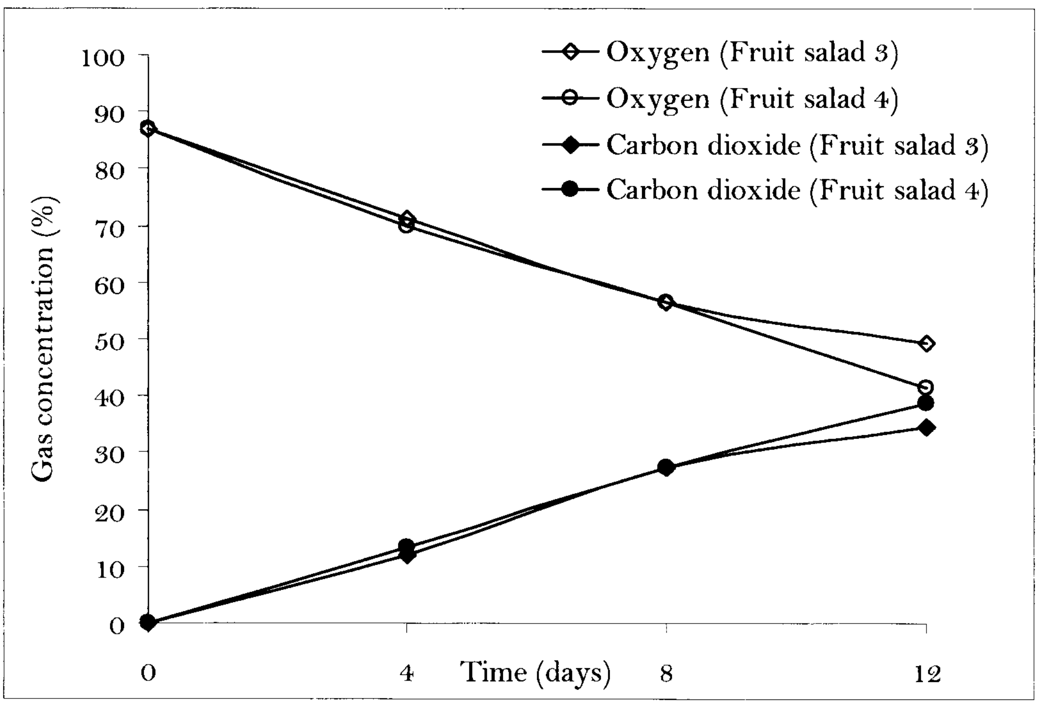


Figure 14 - Carbon dioxide and oxygen concentration (%) of fruit salad 3 and fruit salad 4 packed in air (20% O_2, 80% N_2) during 12 days of storage at 7°C.

On the 8th day of storage pears start to look dry on fruit salad 3 because of water loss and in the 12th day was verified colour changes in pears caused by enzymatic browning that was promoted by the elevated respiration rate occurred inside the packages. Although this more accentuated verified browning it wasn't a limiting factor to the shelf life of fruit salad 3.

The limiting factor of fruit salad 3 was the strong acid taste. Between the 8th and 12th day the quantity of CO_2 was so large that the fruits obtained an acid taste from carbonic acid, this carbonic acid is formed by the dissolution of carbon dioxide in the water content in fruits. In fruit salad 4 the acidity taste was not verified. The difference observed in this two salads (same packaging atmosphere and dipping solutions) can be due to the fact of fruit salad 4 had one more kind of fruit in is constitution (red grapes), in this way the amount of CO_2 in the package could be divided by the three fruits and therefore small amounts of carbonic acid are formed in each fruit resulting in no acidity taste (at least these acidity taste was not detectable). Microbiological activity in the red grapes can be also a hypothesis in the verified differences of the results. This is only a supposition with no literature supporting this hypothesis.

In fruit salad 4 the odour, texture and taste remained the same of respective fruit during the storage time. No juice leakage was observed in both fruit salads packages.

3.1.4. Fruit salad 5 and 6

All product quality evaluation results are showed in Appendix 8. Fruit salad 5 and 6 were both composed by apple, kiwifruit and pear, the difference was that red grape was incorporated in fruit salad 6.

In fruit salad 5 and 6 all the oxygen available was consumed until the 4th day of storage and the production of carbon dioxide in the end of storage reached levels in order of 33% for fruit salad 5 and 26% for fruit salad 6. During the storage time a sharp was increase in the CO_2 concentration was observed, while O_2 concentrations remained at $\approx$ 0%, suggesting a shift from aerobic to anaerobic respiration within the packs. Overpressure (well evident in 4th day of storage and thereafter) and no available oxygen suggest production of fermentative gases inside the packages. In fact the pressure inside the packages was so big that between the 8th and the 12th day the collapse was verified in fruit salad 6. However the sensorial analyse were performed until the 12th day of storage. In fruit salad number 5 no differences in colour, texture and taste of pears and apples slices were detected in time. Kiwifruit lost his green colour and texture between the 4th and 8th day of storage. A dry appearance due to the lost of water was noted in the 4th day of storage.

Fruit salad 6 lost the taste between the 8th and 12th and in some kiwifruit slices was observed fungal growth.

The limiting factor for fruit salads, 5 and 6, in shelf-life was microbiologic activity.

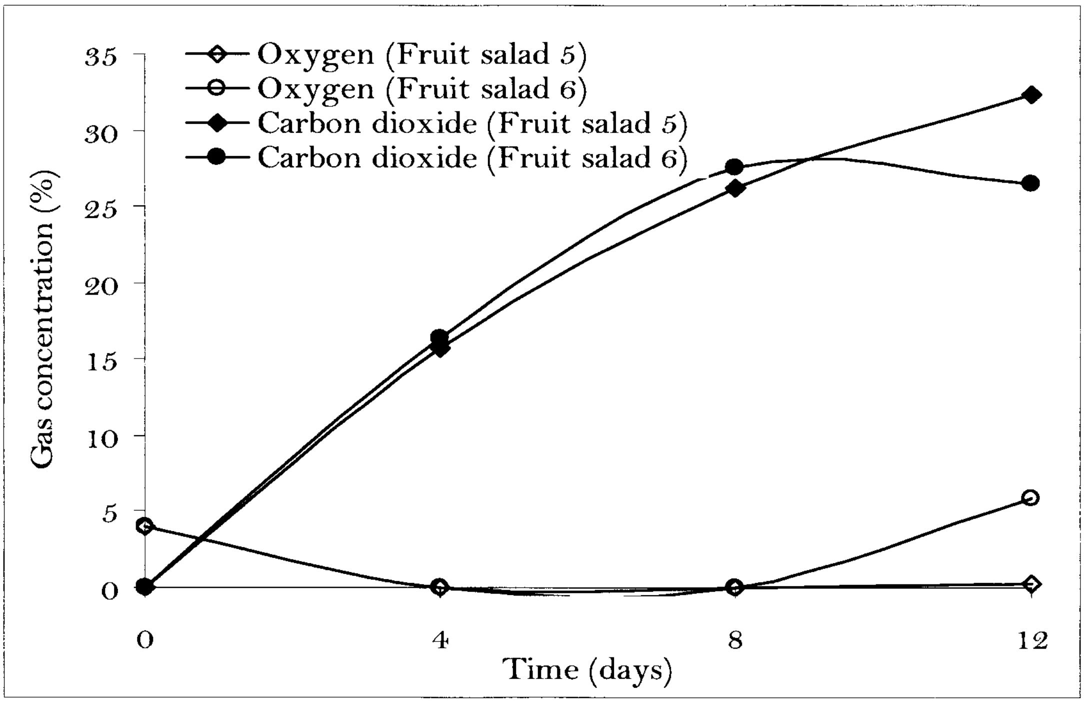


Figure 15 - Carbon dioxide and oxygen concentration (%) of fruit salad 5 and fruit salad 6 packed in air (20% O_2, 80% N_2) during 12 days of storage at 7°C.

3.1.5. Fruit salad 7 and 8

All product quality evaluation results are showed in Appendix 9. Fruit salad 7 and 8 were both composed by pineapple, kiwifruit and melon, the difference was that red grape was incorporated in fruit salad 8.

Minimal oxygen consumption in fruit salad 7 and 8 was observed. In the beginning was present 2% of oxygen was present inside the packages and in the end of storage time the oxygen concentration was about 1.4%. The levels of carbon dioxide in the end of storage for fruit salad 7 was 29% and for fruit salad 8 35%. Even though these values are very high and in theory detrimental for the fruits, it seems that the fruits were not affected.

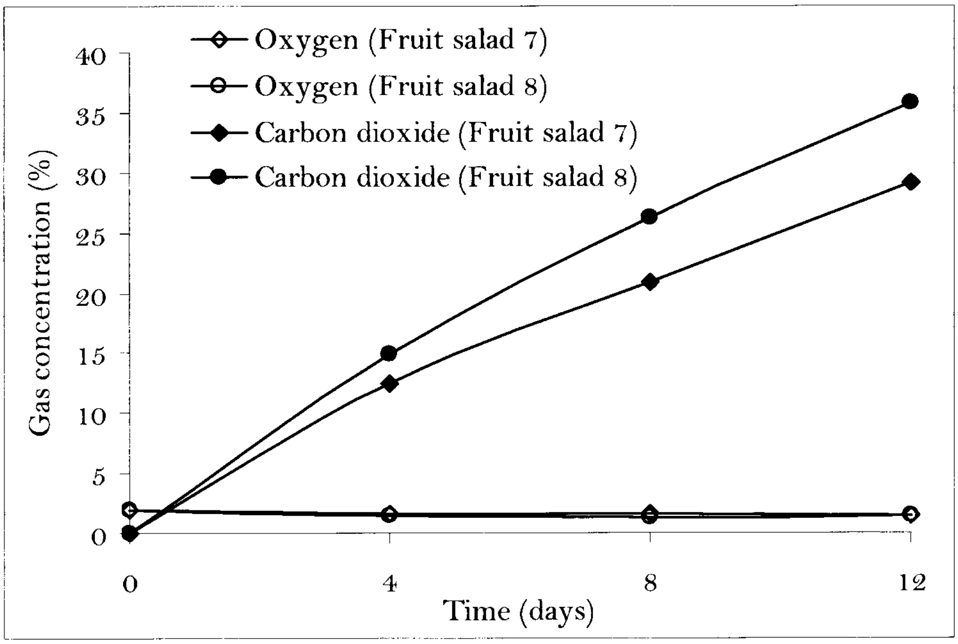


Figure 16 - Carbon dioxide and oxygen concentration (%) of fruit salad 5 and fruit salad 6 packed in air (20% O_2, 80% N_2) during 12 days of storage at 7°C.

The main problem of this both fruit salads was the kiwifruit appearance. Kiwifruit slices lose colour and texture between the 8th and 12th day, however taste was good during the 12 days of storage. If kiwifruit slices were not included in this two fruits salads the shelf-life of both will be extended at least in 4 days.

Odour, colour, texture and taste remained perfect for the rest of fruits in both fruit salads.

4. Conclusions – 2nd Part

During more than 12 days, mixtures of pineapple, apple, pear, melon and red grape packaged in air with chemical treatment did not suffer quality changes. Colour, odour, taste and visual appearance remained unchanged. High O_2 atmospheres (80-90%) were very efficient in maintain the fresh look appearance of pear, melon and red grape during 12 days. A mixture of pineapple, kiwifruit, melon and red grape package in N_2O atmosphere had a shorter shelf-life because of kiwifruit. If kiwifruit was remove of this fruit salad at least more four days of shelf-life was reached.

Nitrogen rich atmospheres for packaging single fruits was very efficient (demonstrated in the 1st part of this researched) however for mixtures of apple, kiwifruit and pear prove to be very unsuccessful yielding no more than four days of shelf-life to this mixture.

Fresh-cut fruits packaged in modified atmospheres with air, high O_2 and N_2O proved to be beneficial in maintaining the quality and extending shelf-life in fruit salads, however, nitrogen rich atmospheres were not efficient in preserving fruit salads once microbial growth becomes important.

When packed alone the shelf-life of fruits are not the same than when are packed in groups, they exhibit different behaviour.

Modified atmosphere packaging (MAP) with conventional and non-conventional gas mixtures in combination with chemical treatments can be very efficient in protecting freshly cut fruits and mixed fruit salads by reducing browning, firmness loss and decay of fruit salads.

5. References

- Belloso, O.M., and Fortuny, R.S., Effect of modified atmosphere packaging on the quality of fresh-cut fruits. Stewart postharvest review,2006
- Willey RC. Introduction to minimally processed refrigerated fruits and vegetables. In: Minimally processed refrigerated fruits and vegetables. Edited by RC Wikey. New York: Chapman & Hall pp 1-14
- Ahvenainen R. New approaches in improving the shelf-life of minimally processed fruit and vegetable. Trends in food science and technology 1996:7:179-187
- Barrett, D. M., Somogyi, L and Ramaswamy, H. 2005. Processing fruits – science and technology, 2nd ed. CRC Press, Boca Raton, pp.26-30, 63-64
- Mir N and Beaudry RM. Modified atmosphere packaging. In: The commercial storage of fruits, vegetables, and florist and nursery stocks. Edited by Gross KC, Wang CY, Saltveit M. Agriculture Handbook Number 66. Washington DC: USDA, ARS; 2004:10.
- Salunkhe, D.K., Bolin, H.R., Reddy, N.R. 1991Storage, processing and quality of fruits and vegetables. 2nd ed. Vol.2 CRC Press, Boca Raton, pp 29-39
- Ohlsson, T., Bengtsson N. Minimal Processing Technologies in the Food Industry 2002, CRC Press, Boca Raton
- Robertson, G. 2005 Food Packaging: prinpiples and practice 2nd ed. CRR Press, pp 318-320
- Sowa, S., Towill, L.E., 1991. Effects of nitrous oxide on mitochondrial and cell respiration and growth in Distichilis spicata suspension cultures. Plant Cell. Tiss. Org
- Alzamora, S.M., Tapia, M.S. and Malo, A.L. (2000) Minimally processed fruits and vegetables – fundamental aspects and applications. Gaithersburg, Maryland. pp – 319-339
- Ferrer, M. M., Harper, C., Muñoz, F.P. and Chaparro, M. 2002 Modified atmosphere packaging of fruits minimally processed mango and pineapple fruits. Journal of food science
- Guzmán, I. L., and Barret, D.M., Comparison of calcium cholide and calcium lactate effectiveness in maintaining shelf stability and quality of fresh-cut cantaloupes. (2000) Posthasvest biology and technology

- Gorny J.R., Hess-Pierce B., Cifuentes R.A., Kader A.A. Quality changes in fresh-cut pear slices as affected by controlled atmospheres and chemical preservatives (2002) Postharvest Biology and Technology
- Jaeger, S.R., Rossiter, K.L., Wismer, W.V., Harker, F.R., 2003. Consumer-driven product development in the kiwifruit industry. Food Qual. Pref. 14, 187–198.
- Watada, A.E., Abe, K., and Yamuchi, N. 1990. Physiological activities of partially processed fruits and vegetables. Food Technol. 44(5): 116-122.
- Arpaia, M.L., Mitchell, F.G., and Kader, A.A. 1986. Ethylene and temperature effects on softening and white core inclusions of kiwifruit in air or controlled atmospheres. J. Amer. Soc. Hort. Sci. 111: 149-153
- Ooraikul, O and Stiles, E., Modified atmosphere packaging of food. 1991. Ellis Horwood. New York. Pp. 169-228
- Arthey, D. and Ashurst, P.R., (2001). Fruit processing – nutrition, products, and quality management. 2nd ed. Aspen publishers, inc. Gaithersburg, Maryland. Pp. 1-19
- Agar I.T., Massantini R., Hess-Pierce B., Kader A.A., Postharvest CO2 and ethylene production and quality maintenance of fresh-cut kiwifruit slices. (1999) Journal of Food Science
- Kader, A.A. (1986) Biochemical and physiological basis for effects of controlled and modified atmospheres on fruits and vegetables. Food techonol.
- Rocculi P., Romani S., Rosa M.D. Effect of MAP with argon and nitrous oxide on quality maintenance of minimally processed kiwifruit. (2005) Postharvest Biology and Technology
- Son S.M., Moon K.D., Lee C.Y. Inhibitory effects of various antibrowning agents on apple slices (2001) Food Chemistry, 73 (1).
- Dong et al., 2000 X. Dong, R.E. Wrolstad and D. Sugar, Extending shelf life of fresh cut pears, Journal of Food Science 65 (2000).
- Marshall, M.R., Kim, J. and Wei, C-I. (2000) Enzymatic browning in fruits, vegetables and seafoods. Nutrition and food science department. Alabama.
- Wszelaki A.L., Mitcham E.J. Effects of superatmospheric oxygen on strawberry fruit quality and decay (2000) Postharvest Biology and Technology, 20 (2), pp. 125-133.

- Amanatidou, A., Smid, E.J. and Gorris, L.G.M., 1999. Effect of elevated oxygen and carbon dioxide on the surface growth of vegetable associated microorganisms. J. Appl. Microbiol. 86, pp. 429–438.
- Ke, D., Rodriguez-Sinobas, L. and Kader, A.A., 1991. Physiological responses and quality attributes of peaches in low oxygen atmospheres. Sci. Hort. 47, pp. 295–303.
- Kader, A.A., 1995. Regulation of fruit physiology by controlled/modified atmospheres. Acta Hort. 398, pp. 59–70.
- Day, B.P.F., 1996. High oxygen modified atmosphere packaging for fresh prepared produce. Postharvest News Inform. 7, pp. 31N–34N.
- http://www.codexalimentarius.net/download/standards/262/CXP_002e.pdf
- Church, N. (1994). Developments in modified-atmosphere packaging and related technologies. Trends in Food Science and Technology, 5, 345–352.
- Day, B. (2001). Fresh Prepared Produce: GMP for High Oxygen MAP and Non-sulphite Dipping. Guideline No. 31. 76 Pp. : Campden & Chorleywood Food Research Association Group, Chipping Campden.
- Hoogerwerf, S.W., Moezelaar, R. & Kets, E. (2000). Effect of novel high oxygen atmospheres on growth of food related associated moulds. In: Second International Symposium on Food Packaging-Ensuring the Safety and Quality of Foods, Vienna. P. 107.
- Jacxsens, L., Devlieghere, F., Van der Steen, C. & Debevere, J. (2001a). Effect of high oxygen atmosphere packaging on microbial growth and sensorial qualities of fresh-cut produce. International Journal of Food Microbiology, 71, 197–210.
- Phillips, C.A. (1996). Review: Modified atmosphere packaging and its effects on the microbiological quality and safety of produce. International Journal of Food Science and Technology, 31, 463–479.
- Van der Steen, C., Devlieghere, F. & Debevere, J. (2003). High oxygen concentrations in combination with elevated carbon dioxide to affect growth of fresh-cut produce micro-organisms. Acta Horticulturae, 599, 141–148.
- Narenda S.S. and Nirankar, N., (2006) Effect of Calcium Lactate, 4-Hexyl Resorcinol and Vacuum Packing on Physico-chemical, Sensory and Microbiological Qualities of Minimally Processed Litchi (*Litchi chinensis* Sonn.) International Journal of Food Science & Technology 41 (9), 1073–1081.

6. Appendix

Appendix 1 - Fresh-cut Pineapples: Product quality evaluation

Samples	Time (d)	Physico-chemical determinations									
		Odour	Sweetness	Bitterness	Translucency	Firmness	Juice leakage	Total	Taste	Visual Scale	Observations
Air	0	1	1	1	1	1	1	6	1	9	
	3	1	1	1	1	1	1	6	1	9	Cond.
	7	1	1	1	0	0	0	3	1	7	Cond.
	12	0	1	1	-1	-1	0	0	0	7	Cond. and off-odour
100% O_2	0	1	1	1	1	1	1	6	1	9	
	3	1	1	1	0	1	0	4	1	7	Cond.
	7	0	1	1	-1	0	0	1	1	7	Cond. and off-odour
	12	0	1	1	-1	-1	0	0	0	7	Cond. and off-odour
100% N_2O	0	1	1	1	1	1	1	6	1	9	
	3	1	1	1	1	1	1	6	1	9	Cond.
	7	1	1	1	0	0	0	3	1	7	Cond.
	12	0	1	1	0	0	0	2	0	7	Cond. and off-odour
4% O_2 + 10% CO_2 + 86% N_2	0	1	1	1	1	1	1	6	1	9	
	3	1	1	1	1	1	1	6	1	9	Cond.
	7	1	1	1	1	1	0	5	1	9	Cond.
	12	1	1	1	1	1	0	5	1	9	Cond.

Figure 17 – Pineapples package with the following atmospheres: 4% O_2, 10% CO_2, 86% N_2 (left image), 100 % O_2 (middle image) and 100% N_2O (right image) with 12 days of storage at 7°C.

Appendix 2 - Fresh-cut Melons: Product quality evaluation

	Time (d)	Physico-chemical determinations						
Samples		Melon flavour	Sweetness	Bitterness	Crunchiness/Hardness	Juice Leakage	Total	Observations
Air	12	-1	1	0	-1	-1	-2	cond./off-flavours
100% O_2	12	1	1	1	1	1	5	cond./very good flavour
80% O_2, 20%N_2	12	-1	1	0	1	1	2	cond./off-flavours
95% N_2, 5% O_2	12	-1	1	0	1	-1	0	cond./yoghurt flavour
90% N_2, 5% O_2, 5% CO_2	12	-1	1	0	0	-1	-1	cond./yoghurt flavour
100% N_2O	12	0	1	1	0	1	3	cond./transparent
100% N_2O, $CaCl_2$	12	-1	1	1	1	1	3	cond./off-flavours/fungus
Air + Ca lactate	12	0	1	0	1	1	3	cond.
Air + $CaCl_2$	12	0	1	0	1	1	3	cond.

Appendix 3 - Fresh-cut Pears: Product quality evaluation

Samples	Time (d)	Physico-chemical determinations							
		Browning class average	Firmness	Crispness	Initial Juiciness	Crunchiness	Mealiness	Flouriness	Total (Texture)
Air	0	0	1	1	1	1	1	1	6
	4	2.6	0	0	1	0	1	0	2
	8	1.8	0	0	1	0	1	0	2
	12	3.3	0	0	1	0	0	0	1
100% O_2	0	0	1	1	1	1	1	1	6
4-HR	4	1.1	0	0	1	0	1	0	2
	8	1.5	0	0	1	0	1	0	2
	12	1.6	1	0	1	0	1	0	3
100% O_2	0	0	1	1	1	1	1	1	6
Cysteine	4	0.2	1	1	1	1	1	1	6
	8	0.3	1	1	1	1	1	1	6
	12	1.1	1	1	1	1	1	1	6
100% N_2	0	0	1	1	1	1	1	1	6
4-HR	4	1.2	1	1	1	1	1	1	6
	8	0.7	1	1	1	1	1	1	6
	12	2.1	1	1	1	1	1	1	6
100% N_2	0	0	1	1	1	1	1	1	6
Cysteine	4	0	1	1	1	1	1	1	6
	8	0.3	1	1	1	1	1	1	6
	12	0.5	1	1	1	1	1	1	6

Samples	Time (d)	Physico-chemical determinations						
		Pear odour	Alcohol	Fermentation	Total (odour)	Visual Scale	Taste	Observations
Air	0	1	1	1	3	9	Good	
	4	1	0	1	2	5	Fair	Cond./ Green odour (hexanol)
	8	1	0	1	2	4	Fair	Green odour (hexanol)
	12	1	0	1	2	4	Fair	Green odour (hexanol)
100% O_2	0	1	1	1	3	9	Good	
4-HR	4	1	0	1	2	7	Good	Cond.
	8	1	0	1	2	5	Fair	
	12	1	0	1	2	5	Fair	Green odour (hexanol)
100% O_2	0	1	1	1	3	9	Good	
Cisteine	4	1	1	1	3	9	Good	
	8	1	1	1	3	9	Good	
	12	1	0	1	2	7	Fair	Green odour (hexanol)
100% N_2	0	1	1	1	3	9	Good	
4-HR	4	1	0	1	2	8	Good	
	8	1	0	1	2	7	Good	
	12	1	0	1	2	7	Fair	Green odour (hexanol) / Slices surface with a dry aspect
100% N_2	0	1	1	1	3	9	Good	
Cisteine	4	1	1	1	3	9	Good	
	8	1	1	1	3	9	Good	
	12	1	0	1	2	8	Good	Slices surface with a dry aspect

Figure 18 – Pears packed in several atmospheres in combination with chemical treatments at 4th and 12th day of storage at 7°C.

Appendix 4 - Fresh-cut Kiwifruit: Product quality evaluation

Samples	Time (d)	Physico-chemical determinations											
		Odour	Sweetness	Acidity	Translucency	Colour	Texture	Juiciness	Juice leakage	Total	Taste	Visual Scale	Observations
Air	0	1	1	1	1	Green	1	1	1	7	Good	9	
2% $CaCl_2$	4	1	1	0	0	Green/Yellow	0	1	1	4	Good	6	
	7	1	1	0	0	Green/Yellow	0	1	1	4	Good	6	
	12	1	1	-1	-1	Green/Yellow	-1	1	1	1	Fair	5	
100% N_2O	0	1	1	1	1	Green	1	1	1	7	Good	9	
2% $CaCl_2$	4	1	1	0	0	Green	1	1	1	5	Good	8	Cond.
	7	1	1	0	0	Green	1	1	1	5	Good	7	Cond.
	12	1	1	-1	-1	Green	-1	1	1	1	Good	5	
10% O_2, 90% N_2	0	1	1	1	1	Green	1	1	1	7	Good	9	
2% $CaCl_2$	4	1	1	0	0	Green/Yellow	1	1	1	5	Good	7	Cond.
	7	1	1	0	0	Green	1	1	1	5	Good	7	Cond.
	12	1	1	-1	-1	Green	-1	1	1	1	Good	5	

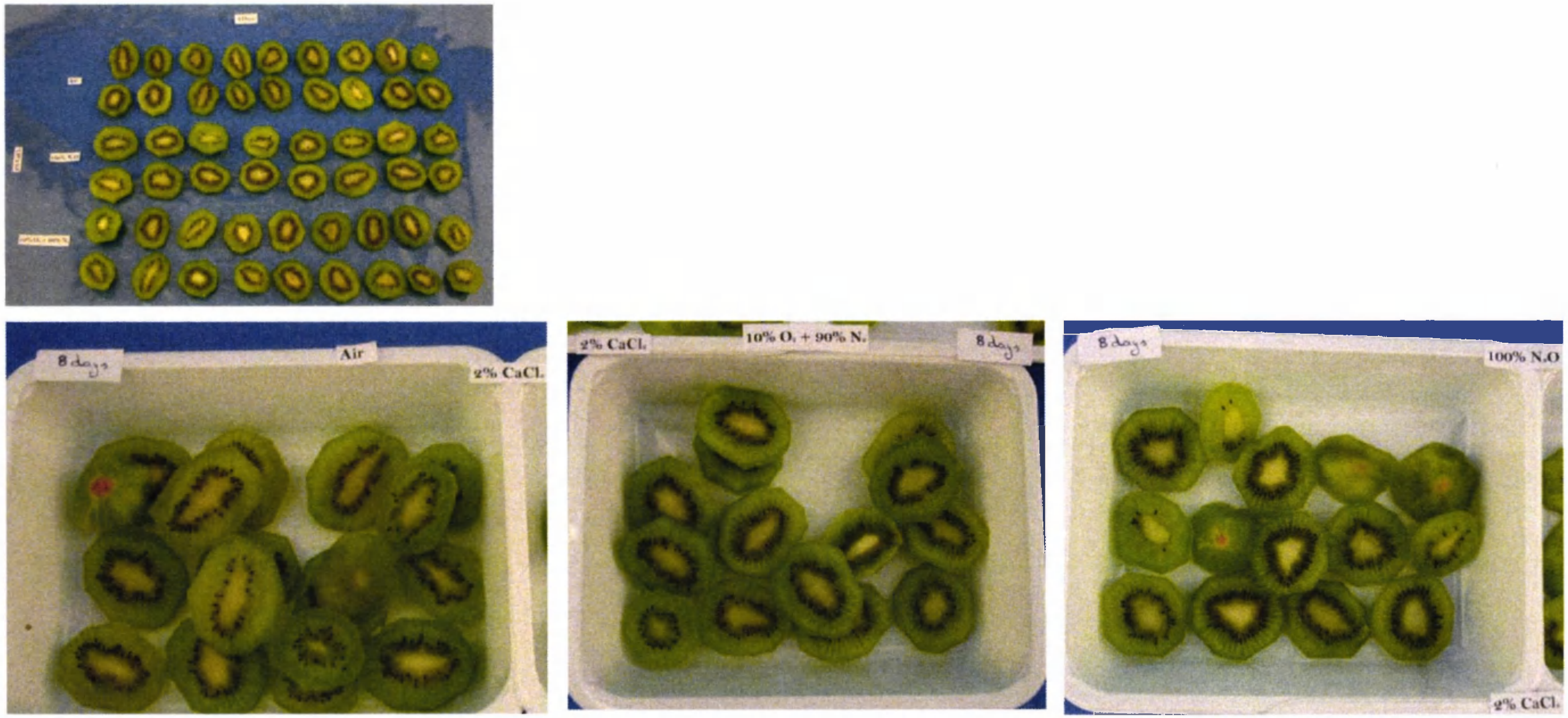


Figure 19 - Kiwifruit packed in several atmospheres in combination with chemical treatments at 4[th] (above image) and 8[th] (down image) day of storage at 7°C.

Appendix 5 - Fresh-cut Apple: Product quality evaluation

		Physico-chemical determinations							
Samples	Time (d)	Browning class average	Firmness	Crispness	Initial Juiciness	Crunchiness	Mealiness	Flouriness	Total (Texture)
Air	0	0	1	1	1	1	1	1	5
0,5% ascorbic acid	5	1.2	1	1	1	1	1	1	5
0,5% citric acid	8	2.4	1	1	1	1	1	1	5
0,5% calcium chloride	12	1.6	1	1	1	1	1	1	5
80% O_2, 20% N_2	0	0	1	1	1	1	1	1	5
0,5% ascorbic acid	5	0.7	1	1	1	1	1	1	5
0,5% citric acid	8	2.1	1	1	1	1	1	1	5
0,5% calcium chloride	12	1.5	1	1	1	1	1	1	5
Air	0	0	1	1	1	1	1	1	5
(2% ascorbic acid)	5	0.2	1	1	1	1	1	1	5
	8	1.1	1	1	1	1	1	1	5
	12	1.4	1	1	1	1	1	1	5
100% N_2	0	0	1	1	1	1	1	1	5
(2% ascorbic acid)	5	0.3	1	1	1	1	1	1	5
	8	0.6	1	1	1	1	1	1	5
	12	0.3	1	1	1	1	1	1	5
Air	0	0	1	1	1	1	1	1	5
0,01% 4HR	5	2.4	1	1	1	1	1	1	5
0,5% ascorbic acid	8	3.2	1	1	1	1	1	1	5
1,0% calcium lactate	12	1.8	1	1	1	1	1	1	5
100% N_2O	0	0	1	1	1	1	1	1	5
0,01% 4HR	5	0.3	1	1	1	1	1	1	5
0,5% ascorbic acid	8	3.3	1	1	1	1	1	1	5

1,0% calcium lactate	12	4.2	1	1	1	1	1	1	5
100% O_2	0	0	1	1	1	1	1	1	5
0,01% 4HR	5	1.1	1	1	1	1	1	1	5
0,5% ascorbic acid	8	2.6	1	1	1	1	1	1	5
1,0% calcium lactate	12	3	1	1	1	1	1	1	5

		Physico-chemical determinations							
Samples	Time (d)	Apple odour	Alcohol	Fermentation	Total (odour)	Observations	Taste	Visual Scale	
Air	0	1	1	1	3			9	
0,5% ascorbic acid	5	1	1	1	3	Cond.	Good	7	
0,5% citric acid	8	1	0	1	2	Cond.	Good	7	
0,5% calcium chloride	12	1	0	1	2		Good	7	
80% O_2, 20% N_2	0	1	1	1	3		Good	9	
0,5% ascorbic acid	5	1	1	1	3	Cond.	Good	7	
0,5% citric acid	8	1	0	1	2	Cond.	Good	7	
0,5% calcium chloride	12	1	0	1	2		Good	7	
Air	0	1	1	1	3		Good	9	
(2% ascorbic acid)	5	1	1	1	3	Cond.	Good	9	
	8	1	0	1	2	Cond.	Good	7	
	12	1	0	1	2		Good	7	
100% N_2	0	1	1	1	3		Good	9	
(2% ascorbic acid)	5	1	1	1	3	Cond.	Good	9	
	8	1	1	1	3	Cond.	Good	9	
	12	1	1	1	3		Good	9	
Air	0	1	1	1	3		Good	9	

0,01% 4HR	5	1	1	1	3	Cond.	Good	5	
0,5% ascorbic acid	8	0	0	1	1	Cond.	Fair	5	
1,0% calcium lactate	12	0	0	0	0	Cond.	Bad	5	
100% N_2O	0	1	1	1	3		Good	9	
0,01% 4HR	5	1	1	1	3	Cond.	Good	9	
0,5% ascorbic acid	8	0	0	0	0	Cond.	Fair	5	
1,0% calcium lactate	12	-1	0	-1	-2	Cond.	Bad	3	
100% O_2	0	1	1	1	3		Good	9	
0,01% 4HR	5	1	1	1	3	Cond.	Good	7	
0,5% ascorbic acid	8	1	0	1	2	Cond.	Fair	7	
1,0% calcium lactate	12	0	0	-1	-1	Cond.	Bad	5	

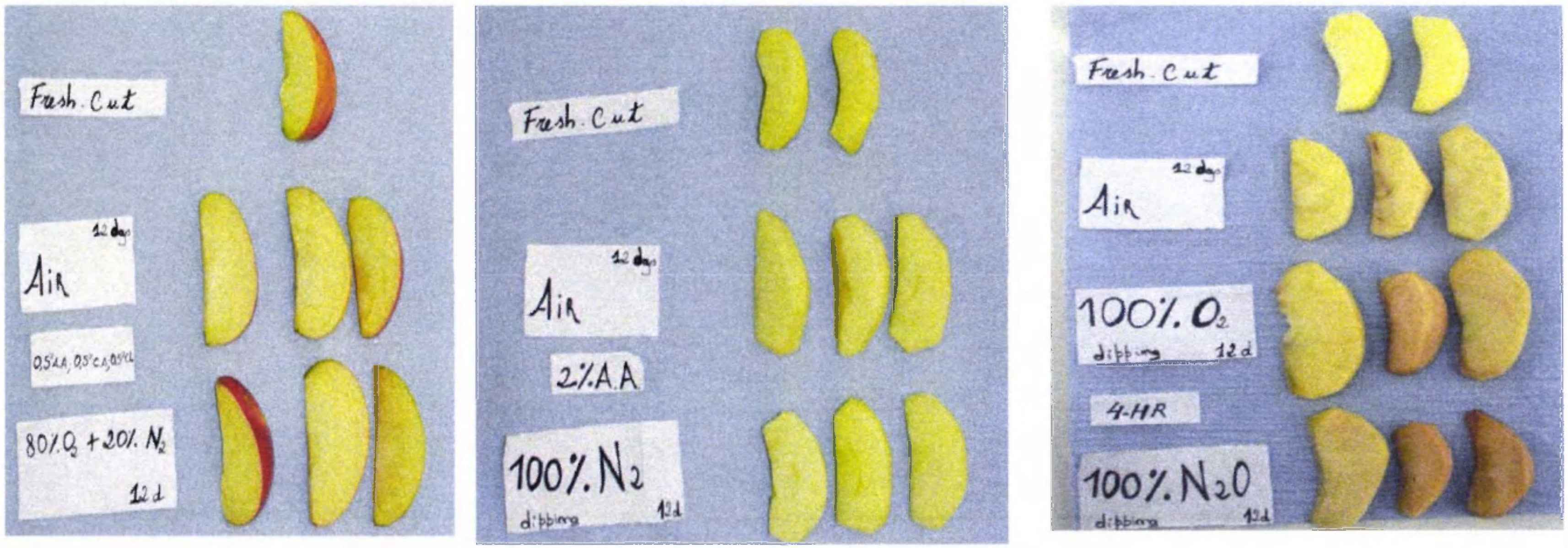


Figure 20 - Apples packed in several atmospheres in combination with chemical treatments at 12^{th} day of storage at 7°C.

Appendix 6 – Fruit salad 1: Product quality evaluation

Fruit Salad 1	**Time (d)**	**Physico-chemical determinations**								
		Odour	Colour	Texture	Taste	Visual Scale	Juice Leakage	Observations	Limiting Factor	Shelf-life (days)
Pineapple	0									
Apple	0		1	1	1	9				
Pear	0		1	1	1	9				
Melon	0		1	1	1	9				
Mixture	0	1	1	1	1	9	1			
Pineapple	4		1	1	1	9				
Apple	4		0	1	1	7			Apple colour	
Pear	4		1	1	1	9				
Melon	4		1	1	1	9				
Mixture	4	1					1	Condensation		<4
Pineapple	8		1	1	1	9				
Apple	8		0	1	1	7			Apple colour	
Pear	8		1	1	1	9				
Melon	8		1	1	1	9		Condensation		
Mixture	8	1		1	1		1			
Pineapple	12		1	1	1	9				
Apple	12		0	1	1	7			Dry aspect/Apple colour	
Pear	12		1	1	1	9				
Melon	12		1	1	1	9				
Mixture	12	1		1	1		1	Condensa		

tion

Figure 21 – Salad fruit 1(pear, melon, pineapple and apple) during the 12 days of storage in air at 7°C.

Appendix 7 – Fruit salad 2: Product quality evaluation

Fruit Salad 2	**Time (d)**	**Physico-chemical determinations**								
		Odour	Colour	Texture	Taste	Visual Scale	Juice Leakage	Observations	Limiting Factor	Shelf-life (days)
Pineapple	0		1	1	1	9				
Apple	0		1	1	1	9				
Pear	0		1	1	1	9				
Melon	0		1	1	1	9				
Grape	0		1	1	1	9				
Mixture	0	1	1	1	1	9	1			
Pineapple	4		1	1	1	9				
Apple	4		1	1	1	9				
Pear	4		1	1	1	9				
Melon	4		1	1	1	9				
Grape	4		1	1	1	9				
Mixture	4	1	1	1	1	9	1	Condensation		
Pineapple	8		1	1	1	9				
Apple	8		1	1	1	8				
Pear	8		1	1	1	9				
Melon	8		1	1	1	9				
Grape	8		1	1	1	9				
Mixture	8	1	1	1	1	9	1	Condensation		
Pineapple	12		1	1	1	9				
Apple	12		1	1	1	8				
Pear	12		1	1	1	9				
Melon	12		1	1	1	9				
Grape	12		1	1	1	9				
Mixture	12	1	1	1	1	9	1	Condensation		> 12

Figure 22 - Salad fruit 2 (pear, melon, pineapple, apple and red grape) during the 12 days of storage in air at 7°C.

Appendix 8 – Fruit salad 3: Product quality evaluation

Fruit Salad 3	Time (d)	Physico-chemical determinations								
		Odour	Colour	Texture	Taste	Visual Scale	Juice Leakage	Observations	Limiting Factor	Shelf-life (days)
Pear	0		1	1	1	9				
Melon	0		1	1	1	9				
Mixture	0	1	1	1	1	9	1			
Pear	4		1	1	1	9				
Melon	4		1	1	1	9				
Mixture	4	1	1	1	1	9	1	Condensation		
Pear	8		1	1	1	8		Dry aspect		
Melon	8		1	1	1	9				
Mixture	8	1	1	1	1	9	1			<8
Pear	12		0	1	0	7				
Melon	12		1	1	0	9				
Mixture	12	1		1	0	9	1		Taste	

Figure 23 - Salad fruit 3 (pear and melon) during the 12 days of storage in air at 7°C

Appendix 9 – Fruit salad 4: Product quality evaluation

Fruit Salad 4	Time (d)	Physico-chemical determinations								
		Odour	Colour	Texture	Taste	Visual Scale	Juice Leakage	Observations	Limiting Factor	Shelf-life (days)
Pear	0		1	1	1	9				
Melon	0		1	1	1	9				
Grape	0		1	1	1	9				
Mixture	0	1	1	1	1	9	1			
Pear	4		1	1	1	9				
Melon	4		1	1	1	9				
Grape	4		1	1	1	9				
Mixture	4	1	1	1	1	9	1	Condensation		
Pear	8		0	1	1	8				
Melon	8		1	1	1	9				
Grape	8		1	1	1	9				
Mixture	8	1		1	1		1			
Pear	12		0	1	1	8				
Melon	12		1	1	1	9				
Grape	12		1	1	1	9				
Mixture	12	1		1	1		1			> 12

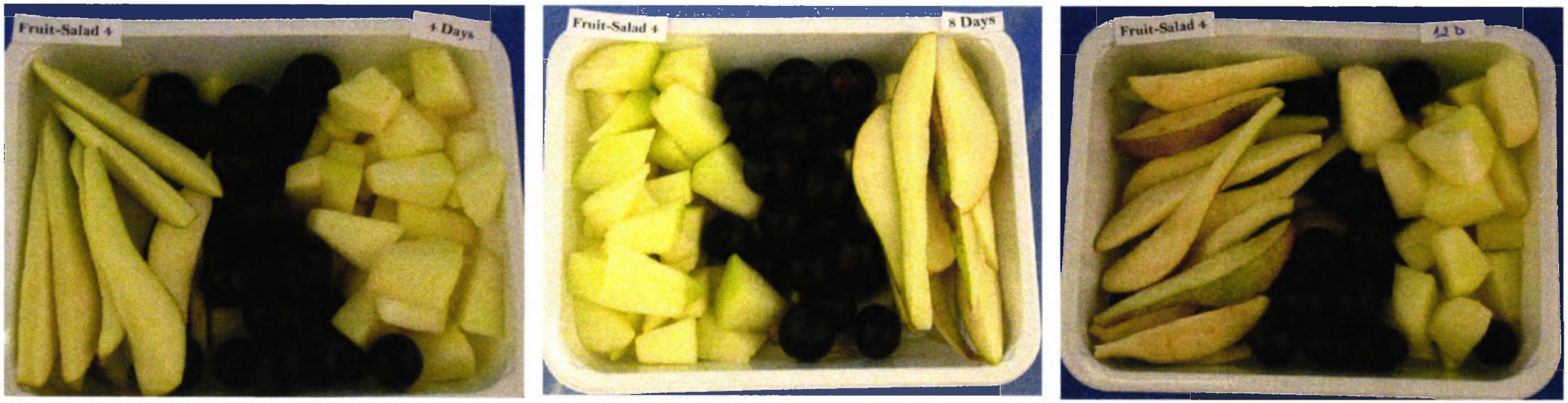


Figure 24 - Salad fruit 4 (pear, melon and red grape) during the 12 days of storage in air at 7°C

Appendix 10 – Fruit salad 5: Product quality evaluation

Fruit Salad 5	Time (d)	Physico-chemical determinations								
		Odour	Colour	Texture	Taste	Visual Scale	Juice Leakage	Observations	Limiting Factor	Shelf-life (days)
Apple	0		1	1	1	9				
Kiwifruit	0		1	1	1	9				
Pear	0		1	1	1	9				
Mixture	0	1	1	1	1	9	1			
Apple	4		1	1	1	9				
Kiwifruit	4		1	1	1	8		Dry aspect		
Pear	4		1	1	1	9				
Mixture	4	1	1	1	1		1	Condensation and Overpressure	Microbiology activity	<4
Apple	8		1	1	1	9				
Kiwifruit	8		0	0	1	7		Dry aspect		
Pear	8		1	1	1	9				
Mixture	8	1			1		1	Condensation and Overpressure		
Apple	12		1	1	1	9				
Kiwifruit	12		0	0	1	6		Dry aspect		
Pear	12		1	1	1	9				
Mixture	12	1			1		1	Condensation and Overpressure		

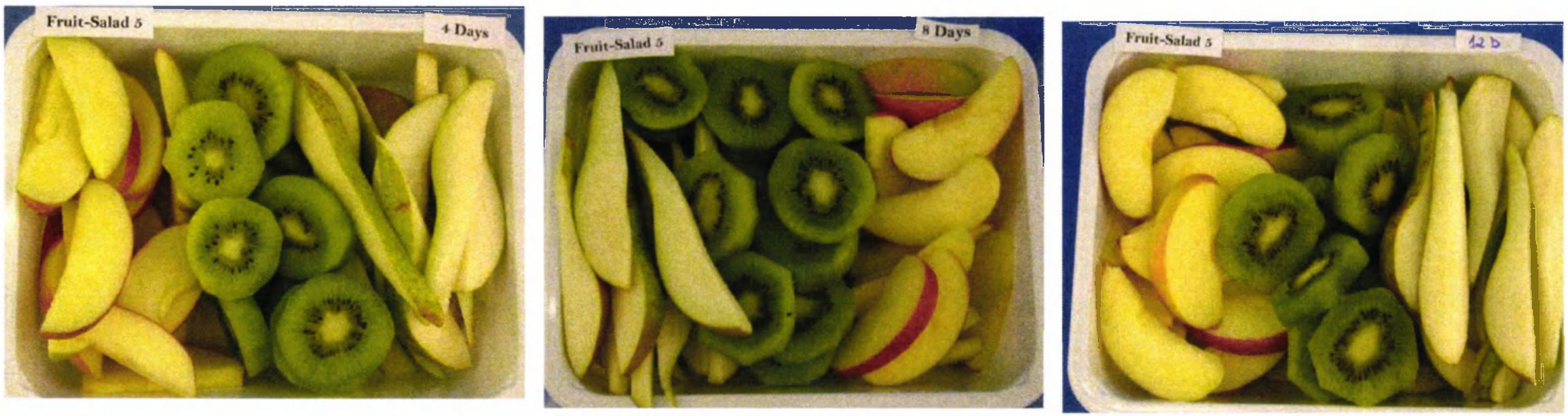
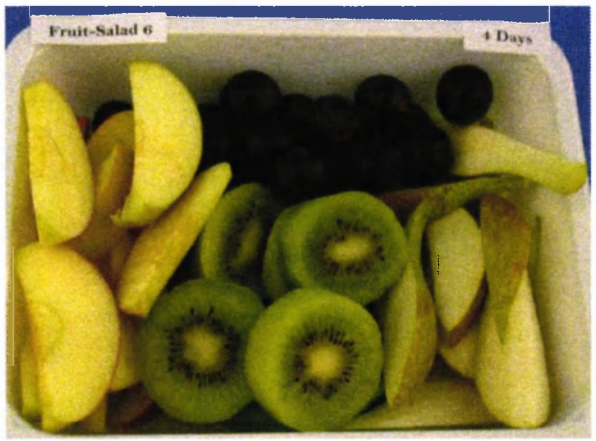


Figure 25 - Salad fruit 5 (pear, apple and kiwifruit) during the 12 days of storage in air at 7°C

Appendix 11 – Fruit salad 6: Product quality evaluation

Fruit Salad 6	**Time (d)**	**Physico-chemical determinations**								
		Odour	Colour	Texture	Taste	Visual Scale	Juice Leakage	Observations	Limiting Factor	Shelf-life (days)
Apple	0		1	1	1	9				
Kiwifruit	0		1	1	1	9				
Pear	0		1	1	1	9				
Grape	0		1	1	1	9				
Mixture	0	1	1	1	1	9	1			
Apple	4		1	1	1	8				
Kiwifruit	4		1	0	0	7				
Pear	4		1	1	1	9				
Grape	4		1	1	1	9				
Mixture	4	1	1				1	Condensation and Overpressure	Microbiology activity	<4
Apple	8		0	1	1	8				
Kiwifruit	8		0	0	1	7		Dry aspect		
Pear	8		1	1	1	9				
Grape	8		1	1	1	9				
Mixture	8	1					1			
Apple	12		0	1	0	8				
Kiwifruit	12		0	0	0	7		Dry aspect and fungus		
Pear	12		1	1	0	9				
Grape	12		1	1	0	9				
Mixture	12	1			0		1			



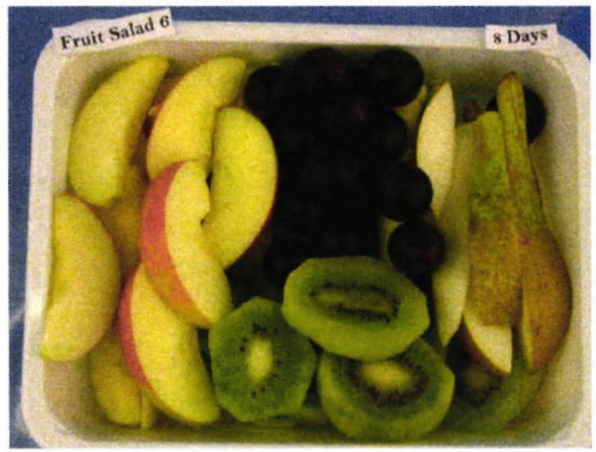


Figure 26 - Salad fruit 6 (pear, apple, kiwifruit and red grape) during the 12 days of storage in air at 7°C

Appendix 12 – Fruit salad 7: Product quality evaluation

Fruit Salad 7	Time (d)	Physico-chemical determinations								
		Odour	Colour	Texture	Taste	Visual Scale	Juice Leakage	Observations	Limiting Factor	Shelf-life (days)
Pineapple	0		1	1	1	9				
Kiwifruit	0		1	1	1	9				
Melon	0		1	1	1	9				
Mixture	0	1	1	1	1	9	1			
Pineapple	4		1	1	1	9				
Kiwifruit	4		1	1	1	9				
Melon	4		1	1	1	9				
Mixture	4	1	1	1	1	9	1	Condensation		
Pineapple	8		1	1	1	9				
Kiwifruit	8		1	1	1	8		Dry aspect		
Melon	8		1	1	1	9				
Mixture	8	1	1	1	1	9	1			8
Pineapple	12		1	1	1	9				
Kiwifruit	12		0	0	1	7		Dry aspect and translucency	Appearance	
Melon	12		1	1	1	9				
Mixture	12	1			1		1			

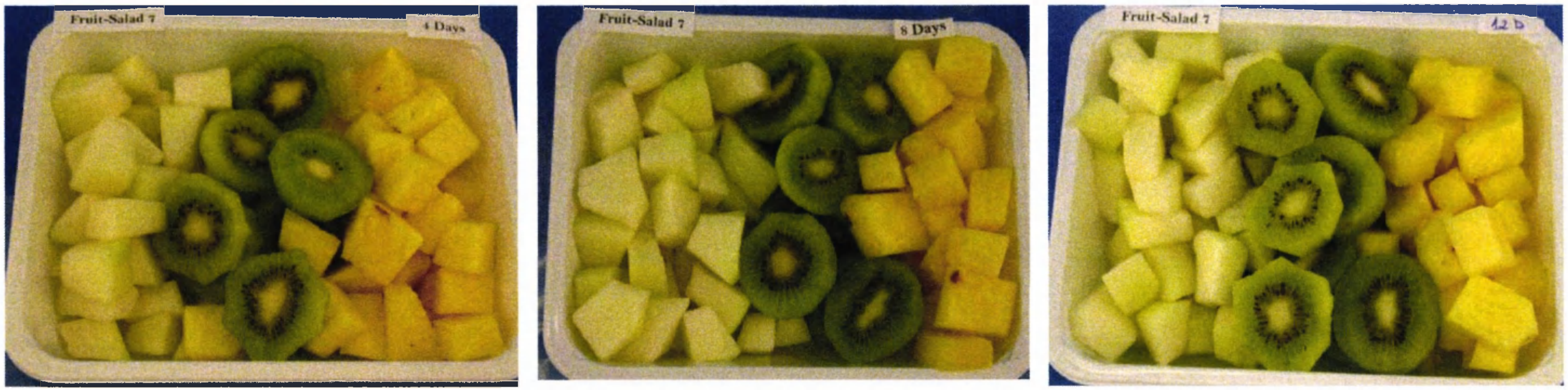


Figure 27 - Salad fruit 7 (melon, pineapple and kiwifruit) during the 12 days of storage in air at 7°C

Appendix 13 – Fruit salad 8: Product quality evaluation

Fruit Salad 8	Time (d)	Physico-chemical determinations								
		Odour	Colour	Texture	Taste	Visual Scale	Juice Leakage	Observations	Limiting Factor	Shelf-life (days)
Pineapple	0		1	1	1	9				
Kiwifruit	0		1	1	1	9				
Melon	0		1	1	1	9				
Grape	0		1	1	1	9				
Mixture	0	1	1	1	1	9	1			
Pineapple	4		1	1	1	9				
Kiwifruit	4		1	1	1	8		Dry aspect		
Melon	4		1	1	1	9				
Grape	4		1	1	1	9				
Mixture	4	1	1	1	1		1	Condensation		
Pineapple	8		1	1	1	9				
Kiwifruit	8		1	1	1	8				
Melon	8		1	1	1	9				
Grape	8		1	1	1	9				
Mixture	8	1	1	1	1		1			8
Pineapple	12		1	1	1	9				
Kiwifruit	12		0	0	1	7		Dry aspect and translucency	Appearance of kiwifruit	
Melon	12		1	1	1	9				
Grape	12		1	1	1	9				

Mixture	12	1			1		1			

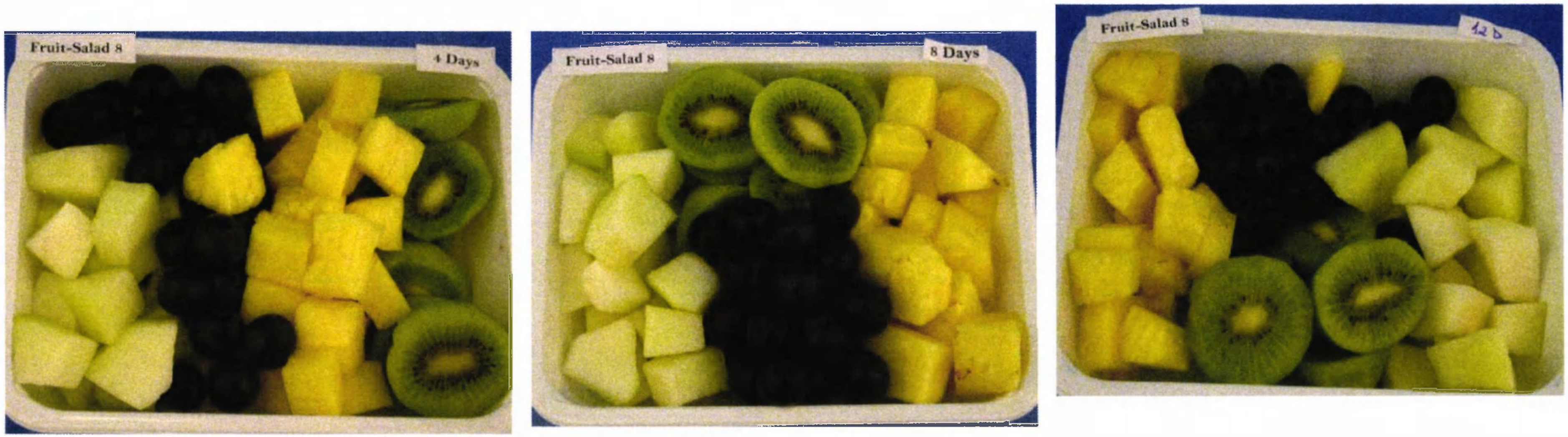


Figure 28 - Salad fruit 8 (melon, pineapple, kiwifruit and red grape) during the 12 days of storage in air at 7°C