Effect of Modified Atmosphere Packaging under Ice Cooling on the Postharvest Storage Life and Quality of Spinach (Spinacea oleracea L)

Leaves

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ABSTRACT

The effect of the use of modified atmosphere packaging (MAP) in combination with ice cooling on postharvest storage life (6 days) and market quality attributes of spinach (Spinacea oleracea L.) leaves were investigated. Crates containing the leaves were stored at ambient conditions (temperature 23.7 \pm 0.5°C and relative humidity 75 \pm 4%) to simulate market conditions. Leaves were evaluated during storage for changes in weight loss, moisture content, quality and wilting condition (based on sensory characteristics), chlorophyll, soluble protein and ascorbic acid contents. Modified atmosphere packaging plus ice cooling reduced moisture loss (0.6% compared to 5% in MAP under ambient conditions), wilting and prolonged market quality of the leaves. In addition, this treatment reduced loss of ascorbic acid and soluble protein (10% compared to 30% under ambient conditions). Modified atmosphere packaging plus ice cooling did not have any added advantage in terms of weight loss and chlorophyll degradation compared to control samples held under MAP at ambient temperature and the treatments were not significantly different (p<0.05). These results indicate that a combination of MAP and ice cooling can considerably extend the postharvest storage life of spinach leaves as well as retaining their market quality attributes. We recommend that farmers and/or traders who transport or handle bulk leafy vegetables to adopt such a method since it has the potential of extending and maintaining product quality.

1.0. Introduction

Spinach (Spinacea oleracea L.) is one of the highly perishable leafy vegetables which is often packed in crates, bags or placed directly on top of vans during transportation to markets without any cooling. A lot in food value and/or quality is compromised or lost

during the transportation period as a direct result of the high temperatures, poor harvesting and poor postharvest handling among others (Abe et al., 1996; Kays, 1991). In developed countries, means of removing field heat of fresh produce prior to and during storage are often employed to maintain the perishables in a fresh state during the marketing chain. However, the primary factors in maintaining quality and extending the postharvest life of fresh fruits and vegetables are harvesting at optimum maturity, minimizing mechanical injuries and providing the optimum temperature and relative humidity during transportation and/or marketing (Kader et al., 1989). Once these primary requirements have been met, further maintenance of quality at all stages can be achieved through modification of the atmosphere surrounding the produce and modified atmosphere packaging (MAP) provides such a means (Zagory and Kader, 1988).

Modified atmosphere packaging has been used as a supplement to appropriate temperature and relative humidity conditions during transportation and/or storage of some fruits and vegetables (Barth et al., 1993b). Modified atmosphere packaging utilizes polymeric films of differential permeabilities to oxygen, carbon dioxide, ethylene and water vapour to extend the shelf life of various fruits and vegetables. Atmospheric modification within the package develops as a result of the respiration of the plant tissue and gas diffusion characteristics of the film (Kader et al., 1989; Barth et al., 1993b). Semi-permeable films are, however, the most popular barriers to create modified atmospheres (Talasila et al., 1995). Modified atmosphere packaging has the advantage of not requiring expensive equipment that is used in controlled atmosphere storage for regulating the gas composition. In addition, MAP in combination with low temperature storage has been recognized as a promising and inexpensive way to improve shelf life of fresh fruits and vegetables while minimizing-quality impairment (Zagory and Kader, 1988; Kader et al., 1989). Loss in quality and limited shelf life are major problems faced in the transportation and marketing of spinach leaves in developing countries (Kays, 1991). The fact that most of the spinach is grown in the rural areas, far away from the urban centres where bulk of it is consumed, brought into consideration the application of MAP in combination with ice cooling for preservation of commercial quality, in terms of texture, appearance, flavour and nutritive value. Icing has the advantage of lowering the product temperature, particularly during transportation (a step in postharvest handling where other cooling methods may not be

appropriate) to market places, as well as being relatively cheaper compared to other methods of cooling such as hydrocooling, vacuum cooling and forced-air cooling (Mitchell, 1992). Maintenance of low temperature by other albeit, expensive means has been shown to extend postharvest life of several fruits and vegetables (Lazan et al., 1987; Watada et al., 1987; Kader et al., 1989, Amanatidou et al., 1999; Ahmadi et al., 1999). Maintaining proper temperatures without compromising the quality of perishables is a difficult problem for most developing countries, particularly in the tropics and subtropics. Spinach is one vegetable that can provide the necessary nutrients in the diets for many disadvantaged communities if its postharvest handling is well addressed. Spinach is rich in ascorbic acid (vitamin C), an important nutrient and being a green leafy vegetable, its senescence is related to wilting and loss of greenness with degradation of chlorophyll. In Kenya, for example, such loss could be as high as 60% depending on locality (Abe et al., 1996).

The objective of this study was to investigate the possibility of extending the postharvest life of spinach through a combination of MAP and ice cooling with particular reference to nutritional and market quality attributes. The results obtained from this study will be useful to small and medium scale establishments involved in transportation, distribution and/or marketing of green leafy vegetables.

2.0. MATERIALS AND METHODS

Sample preparation

Freshly harvested spinach (*Spinacea oleracea* L.) leaves were obtained from a local supplier in Thika, Kenya. The leaves were sorted for freedom from defects and uniformity in terms of colour, size and shape. The leaves were then transported in cool boxes to the Laboratory of Postharvest Technology, Department of Food Science and Technology at the Jomo Kenyatta University of Agriculture and Technology, where the experiments were conducted. Upon arrival at the laboratory, the leaves were washed in running tap water to remove soil and other contaminants and then dipped in 50µg/l NaOCl (chlorine) solution for 1 minute. The leaves were separated into two sublots of two replicates each for treatment at ambient temperature and under ice cooling. For each treatment, one set was sealed in polyethylene (PE) bags (gauge 150), the other in perforated PE bags (sixteen 6 mm diameter holes) and the other set was unpackaged. The ice was packed in PE bags with relatively

uniform volumes (2000 ml) and placed in crates where the prepared leaves were stored. The coolant comprised of 60% fine crushed ice, 40% water and 1% sodium chloride. The sodium chloride was used to lower the temperature. Replacement of the ice was done every morning during each of the six days of storage. The crates were placed in a laboratory bench to simulate marketing and/or transportation conditions. The mean room temperature was $23.7 \pm 0.5^{\circ}$ C and the relative humidity was 75 ± 4 %.

Analyses

Determination of moisture content

Moisture content of the control and treated samples was determined by the oven dry method of AOAC (1984). Duplicate samples (10g each) per treatment were dried in an oven at 105 - 110^o C for 3 hours. Crucibles containing the dried samples were cooled in a desiccator for 30 min and weighed to the nearest 0.001g and percent moisture loss was computed.

Determination of chlorophyll content

Total chlorophyll content of the control and treated samples was determined by the spectrophotometric method of Arnon (1949). Five grams of vegetable tissue was ground in a mortar and pestle in 20 ml of cold 80% (v/v) acetone and some acid-washed sand. The extract was filtered and the residue was washed with 80% acetone until it was colourless. The final volume was then brought to 100 ml using 80% cold acetone. Aliquots of samples were transferred to 5ml quartz cuvettes (1 cm light path) prior to spectrophotometric reading at 645 and 663 nm using a Shimadzu double beam spectrophotometer (Model UV-120, Shimadzu Corp., Kyoto, Japan). The determinations were replicated three times per treatment. Total chlorophyll content expressed as μg total chlorophyll per gram of vegetable sample on fresh weight basis was calculated using the Arnon formula: Total chlorophyll content ($\mu g/g$) = 20.2A₆₄₅ + 8.02A₆₆₃. Percent total chlorophyll was calculated to indicate percentage loss during storage.

Determination of soluble protein content

Aliquots of the control and treated spinach samples (5g each) were ground in a mortar and pestle with five volumes of 0.2 M potassium phosphate buffer (pH 7.2) in the presence of acid-washed sand. The extract was centrifuged at 27,000 x g for 20 minutes. The determinations were replicated three times per treatment. Protein content in the supernatant was then determined by the protein dye-binding method of Bradford (1976) using bovine scrum albumin (Sigma Chemical Co., St. Louis, MO) as a standard. The protein content was expressed as a percentage of total weight.

Determination of ascorbic acid content

Ascorbic acid content of the control and treated samples was determined according to AOAC (1984) methods. Five grams of sample were ground in a mortar and pestle with acid-washed sand and 10% trichloroacetic acid. Ascorbic acid content in the extract was then determined by visual titration with 2,6-dichlorophenolindophenol. Ascorbic acid content was expressed as mg/100 g of fresh weight and then percent loss during storage was computed.

Gaseous monitoring

The composition of carbon dioxide and ethylene gases within each of the sealed PE bags were monitored during storage using gas chromatograph. Samples of internal gas atmosphere were withdrawn using a 1ml gas tight syringe through a self-sealing septum affixed on the surface of the bags. Carbon dioxide concentration (%) was analyzed by injecting 1 ml of the headspace gas into a Shimadzu gas chromatograph (Model GC 8A, Shimadzu Corp., Kyoto, Japan) equipped with a thermal conductivity detector and a Porapak Q column. Ethylene concentration (ppm) was determined by injecting 1 ml of the headspace gas into a Shimadzu gas chromatograph (Model GC 9A, Shimadzu Corp., Kyoto, Japan) equipped with a flame ionization detector and activated alumina column. Due to some technical problems, we could not analyze the concentration of oxygen in the PE bags.

Assessment of visual appearance and determination of weight loss

Wilting condition of the spinach leaves was expressed as an index using a five point hedonic scale: 1 = not wilted; 2 = slightly wilted; 3 = moderately wilted; 4 = very wilted; 5 = extremely wilted. Marketability and edibility of the spinach leaves was rated on a five point hedonic scale as follows: 1= all leaves are highly marketable; 2 = majority of leaves are very marketable; 3 = majority of leaves are marketable; 4 = majority of leaves are edible, but not marketable; 5 = majority of leaves are neither marketable nor edible. Visual appearance involved relating wilting, weight loss and moisture loss to other quality parameters such as colour and texture. Weight loss of two replicates per treatment was followed on a daily basis using a weighing balance to the nearest 0.001 g and the weight loss was expressed as a percent. The data presented are means of duplicate determinations.

3.0. RESULTS AND DISCUSSION

Modified atmosphere packaging reduced moisture loss in spinach leaves held at both ambient temperature and under ice cooling (Table 1).

Table 1. Effect of modified atmosphere packaging on percent moisture loss in spinach leaves during storage at ambient temperature or under ice cooling

	Moisture loss (%) Storage period (days)				
Ambient temperature	С	0	3.9 ^a	39.0°	ND
	P	0	2.4 ^b	9.7°	22.3ª
	UP	0	0.6°	4.7°	5.7 ^b
Ice Cooling	C	0	0.9^{c}	3.1°	8.6°
	P	0	0.9^{c}	1.7°	1.9 ^d
	UP	0	0.0^{c}	0.6^{c}	0.7^{d}

C = control; P = perforated bags; UP = unperforated bags ND = Not Determined. Values are means of two determinations. Means in a column followed by the same letter are not significantly different at p< 0.05.

The change in moisture content under ice cooling was minimal but was significantly different (p < 0.05) compared to that of control. Perforating the packages also reduced moisture loss (8.5% moisture loss after 4 days) and this effect was more pronounced under ice cooling. Such effects of MAP on reducing moisture loss have been reported for other perishable commodities, including broccoli florets (Zhuang et al., 1994), broccoli spears (Forney et al., 1989; Barth et al., 1993a, b), carrots (Amanatidou et al., 2000) and some tropical leafy vegetables (Lazan et al., 1987) in which samples retained turgidity. Zhuang et al., (1994) observed that in broccoli florets, moisture content was related to change in soluble proteins.

Weight loss during storage increased with increase in time in both treatments (Fig. 1A and B) and was highest in unpackaged samples. However, ice cooling significantly (p < 0.05) reduced weight loss in unpackaged samples compared to unpackaged samples held at ambient temperature. Modified atmosphere packaging with perforation reduced weight loss under both treatments with samples held under ice cooling losing the least. The pattern of weight loss was linear under MAP (without perforation) for both treatments and weight loss appeared not to be related to the treatment, that is, under ambient or ice cooling conditions. The reduced weight loss in samples under MAP was probably due to high relative humidity maintained within the packages and this could have counteracted the effect of the storage temperature. In tomato fruit, weight loss was greatly reduced by high relative humidity (Bhowmik and Pan, 1992) and MAP (Nakhasi et al., 1991). Similarly, unpackaged samples held at ambient temperature wilted considerably compared to those held under ice cooling (Fig. 2A and B). Wilting was directly related to the mode of packaging and storage conditions. Watada et al. (1987) observed that in spinach, the wilting condition was greater at higher temperature and in unpackaged samples, a change that was similar to that of weight loss. Wilting and weight loss was extensive in unpackaged samples, and because of the large changes, the correlation between wilting condition and weight loss was readily apparent. Similar observations were made by Watada et al. (1987). The extensive wilting condition and weight loss in unpackaged samples reduced the marketability and edibility of the spinach leaves by about 60%, a situation that was alleviated by MAP and storage under ice cooling (Fig. 3A and B).

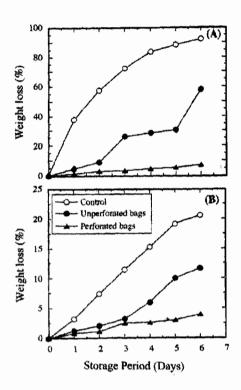


Figure 1. Effect of modified atmosphere packaging on weight loss in spinach leaves kept at ambient temperature (A) and under ice cooling (B)

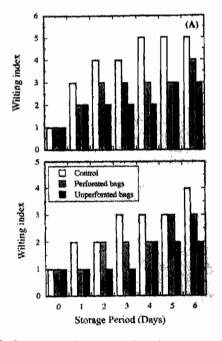


Figure 2. Effect of modified atmosphere packaging on wilting condition in spinach leaves kept at ambient temperature (A) and under ice cooling (B)

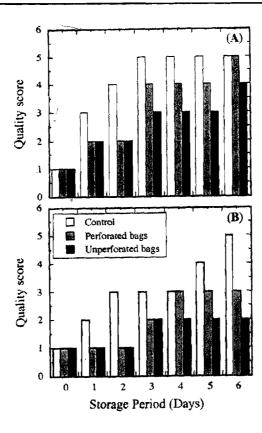


Figure 3. Effect of modified atmosphere packaging on quality attributes of spinach leaves kept at ambient temperature (A) and under ice cooling (B)

Indeed, a combination of MAP and ice cooling extended the marketing period up to at least 6 days while MAP under ambient temperature extended the same to 5 days. The experiment was stopped after 6 days when samples in most of the treatments had lost more than 3% of water (Table 1) beyond which spinach becomes unsalable (Kays, 1991). Watada *et al.* (1987) reported that spinach leaves held in PE bags maintained saleable quality for 9 days. There are many factors that may be attributed to this difference including growing conditions, the film thickness, variety and postharvest handling prior to the treatment. However, based on the observation made in this study, there is need to carry out further research to establish the maximum storage time under these conditions as well as variety responses. Green leafy vegetables wilt rapidly in unfavourable environments due to their extensive and rather permeable surfaces (Kailasapathy and Koneshan, 1986). Humidity conditions are of greater importance in the preservation of ascorbic acid in vegetables subject to rapid loss of moisture than those resistant to wilting (Ezell and Wilcox, 1959).

Therefore, in addition to serving as an index to visual quality, wilting can be used as an index of nutritional quality. Modified atmosphere packaging also extended the postharvest quality of rutabaga (Zhu et al., 2001), yellow passion fruit (Arjona et al., 1994) and pepper (Lownds et al., 1994) by reducing shriveling mainly through restriction of moisture loss.

One of the symptoms of senescence in harvested green leafy vegetables is loss of greenness with degradation of chlorophyll (Yamauchi and Watada, 1991) while ascorbic acid is very labile and its retention is often used when evaluating postharvest storage effects on nutritional quality in fruits and vegetables (Klein and Perry, 1982; Barth *et al.*, 1993a). Unpackaged samples under both treatments lost chlorophyll (from 640.8 µg/g to 384.5 µg/g) (Fig. 4A and B) and ascorbic acid (from 38.5 mg/100g to 23.1 mg/100g) (Fig. 5A and B).

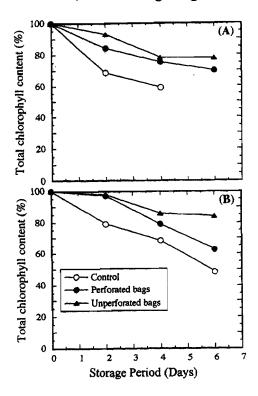


Figure 4. Effect of modified atmosphere packaging on total chlorophyll content in spinach leaves kept at ambient temperature (A) and under ice cooling (B). Initial chlorophyll content was 640.8 µg/g fresh weight.

Before deterioration at day 4, unpackaged samples held at ambient temperature had lost 40% chlorophyll while those held under ice cooling had lost 35% of chlorophyll by day 4.

Yamauchi and Watada (1991) observed that chlorophyllase played a minor role in the degradation of chlorophyll in spinach leaves held at 25°C and that most of the chlorophyll appeared to be degraded by the peroxidase pathway where the porphyrin ring is opened and the resulting compound is colourless. Similarly, unpackaged samples held at ambient temperature had lost 60% of ascorbic acid before deterioration while those under ice cooling had lost 50% by day 4.

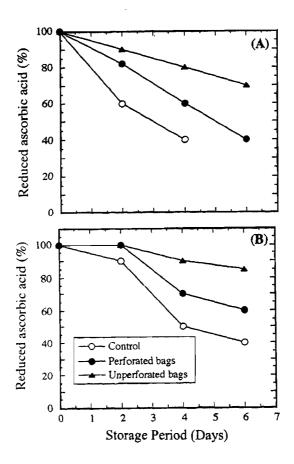


Figure 5. Effect of modified atmosphere packaging on ascorbic acid content in spinach leaves kept at ambient temperature (A) and under ice cooling (B). Initial ascorbic acid content was 38.5 mg/100g fresh weight.

Modified atmosphere packaging increased both chlorophyll and ascorbic acid retention. Perforated films have been used to package produce in an attempt to reduce moisture loss and avoid anaerobic conditions and carbon dioxide injury during MAP (Zhuang et al., 1994). In the present study perforated films reduced both chlorophyll and ascorbic acid

degradation but not as much as in unperforated films which maintained these parameters at about 15% higher. This is in agreement with previous observations which have indicated that vented films reduced chlorophyll and ascorbic acid losses in broccoli (Barth et al., 1993a, b; Zhuang et al., 1994) and green beans and spinach (Watada et al., 1987). Chlorophyll retention was more related to the mode of packaging than to the storage temperature, while ascorbic acid retention was related to both the mode of packaging and storage temperature. This is in agreement with data reported by Lazan et al. (1987) who observed that in some tropical leafy vegetables, storage temperature had little effect on chlorophyll degradation. Watada et al. (1987) indicated that packaging had no effect on chlorophyll content in spinach held at 10°C. Observations made in this study confirm previous reports which have indicated that loss of chlorophyll in vegetables is retarded by elevated carbon dioxide conditions in the microclimate created within the PE bags (Lipton and Harris, 1974) which tend to antagonise the effects of ethylene in degradation of chlorophyll (Kays, 1991). However, the level of gases in the polyethylene bags needs to be carefully maintained since it can affect colour negatively (Clark and Burmeister, 1999).

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Chlorophyll loss has mainly been used as a measure of green plant tissue senescence. From the present study it was observed that there was a positive correlation between chlorophyll retention and ascorbic acid retention. For spinach under ambient conditions the respective regression equations and correlation between chlorophyll retention and ascorbic acid retention were: control: v = 1.35x - 34.10, $R^2 = 0.9881$; perforated bags: v = 1.84x -80.14, $R^2 = 0.9420$; unperforated bags, y = 1.01x - 2.33, $R^2 = 0.8482$ while the corresponding values for the spinach under ice cooling were: control: y = 1.24x - 23.29, R^2 = 0.8795; perforated bags: v = 1.23x - 22.75, $R^2 = 0.9651$; unperforated bags, v = 0.96x + 1.23x - 1.23x5.20, $R^2 = 0.9575$. Thus, chlorophyll content can be used as an indirect measure of the nutritional value of green leafy vegetables. The high ascorbic acid retention in samples under MAP could be attributed to the gas composition inside the packages which has been shown to inhibit many biochemical and physiological processes including degradation chlorophyll and ascorbic acid (Kader et al., 1989). There was a good correlation between wilting index and ascorbic acid retention. For spinach under ambient conditions the respective regression equations and correlation between wilting index and ascorbic acid retention were: control: y = 0.73x + 27.69, $R^2 = 0.9918$; perforated bags: y = 0.92x + 12.32,

 $R^2 = 0.9435$; unperforated bags, y = 0.78x + 27.75, $R^2 = 0.9288$ while the corresponding values for the spinach under ice cooling were: control: y = 0.77x + 20.5, $R^2 = 0.8792$; perforated bags: y = 0.95 + 10.91, $R^2 = 0.8861$; unperforated bags, y = 0.6x + 40, $R^2 =$ 0.9474. It has been indicated that conditions that are favourable for wilting result in a more rapid loss of ascorbic acid (Ezell and Wilcox, 1959; Kailasapathy and Koneshan, 1986). The loss of ascorbic acid in wilted (unpackaged) leaves may be due to increased oxidation because of the reported increase in activity of ascorbic acid oxidase and other relevant oxidizing enzymes in wilted leaves (Lazan et al., 1987; Liao and Seib, 1987; Barth et al., 1993b). The atmospheric composition inside the package possibly served best in preserving ascorbic acid (Ezell and Wilcox, 1959; Barth et al., 1993a, b). This could also be due to reduced activities of peroxidase and ascorbic acid oxidase as a result of oxygen depletion inside the packages (Barth et al., 1993b). In the preservation of chlorophyll during MAP, Kader (1986) stated that the high carbon dioxide concentration in the package may result in chlorophyll protection due to direct action of carbon dioxide on the membrane or by slowing senescence. In addition, ascorbic acid has been found to be an anti-oxidant complex in chloroplasts and which maintains chlorophyll in the packaged spinach leaves by reducing oxidation of chloroplast components (Thompson et al., 1987).

Net loss in protein has been recognized as a dominant feature of senescence of green plant tissues (Zhuang et al., 1994). Soluble protein content under both treatments generally decreased during storage but MAP significantly (p< 0.05) reduced loss of soluble proteins irrespective of the storage temperature (Fig. 6A and B). However, soluble protein levels were significantly (p < 0.05) higher in spinach leaves held under ice cooling. Zhuang et al. (1994) demonstrated that MAP maintained high soluble protein levels in broccoli held at 5°C. Modified atmosphere packaging in conjunction with low temperature storage also assisted in the preservation of soluble protein content in some tropical leafy vegetables (Lazan et al., 1987). At ambient temperature MAP retained 60% of soluble protein while MAP under ice cooling maintained 71% during the six days of storage period. We observed that in unpackaged leaves soluble protein content was significantly (p < 0.05) lower compared to other treatments, probably due to water stress caused by moisture loss from the spinach.

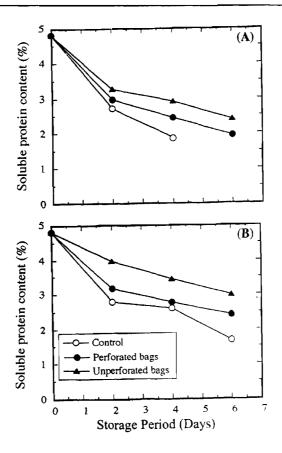


Figure 6. Effect of modified atmosphere packaging on soluble protein content in spinach leaves kept at ambient temperature (A) and under ice cooling (B).

Water stress may promote protein degradation as well as inhibit synthesis of proteins (Hsiao, 1973), and this could explain the decrease in soluble protein content in unpackaged leaves. Lazan et al. (1987) made similar observations using Brassica juncea and Amaranthus caudatus. Baxter and Walters (1990) observed that okra pods stored in controlled atmosphere retained more proteins than those held in air. The high loss of soluble proteins in commodities held in air could be due to respiratory activity to provide carbons for Kreb's cycle reactions. The physical state of the vegetable is also important in influencing its biochemical processes including protein degradation (Zhu et al., 2001; Amanatidou et al., 2000).

Carbon dioxide concentration in PE bags held under ambient temperature increased to 2.8% (Fig. 7A) within 24 hrs and thereafter remained constant. Ethylene concentration increased to 2.5 ppm (Fig. 7B) within the six days of storage.

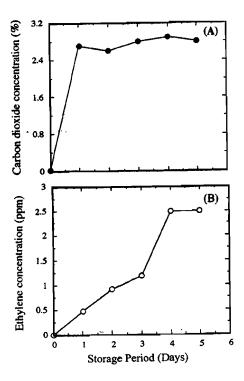


Figure 7. Changes in carbon dioxide concentration (A) and ethylene concentration (B) in packaged spinach leaves kept at ambient temperature

The carbon dioxide concentration reached the so called 'steady state' within 24 hrs. This is in agreement with previous studies which have indicated that carbon dioxide in PE bags reaches the 'steady state' within 24 hrs (Forney et al., 1989; Nakhasi et al., 1991; Shamaila et al., 1992; Barth et al., 1993a; Mathooko et al., 1993; Mathooko, 1995; Talasila et al., 1995) and that ethylene tends to accumulate in the PE bags (Nakhasi et al., 1991; Mathooko et al., 1993). If the time to reach 'steady state' is long relative to the storage life, the packaged product may not fully benefit from the MAP. The accumulation of ethylene in the PE bags did not in any way negate the effects of carbon dioxide on the regulation of various physiological and biochemical processes since carbon dioxide is a well-known inhibitor of ethylene action (Mathooko, 1995; 1996). Although the concentration of oxygen in the PE bags was not determined, it is expected that under such circumstances oxygen concentration would decrease. Therefore, the superior quality observed in spinach leaves held under MAP is due to decreased oxygen and increased carbon dioxide concentrations and probably decreased sensitivity to ethylene besides high relative humidity. The elevated carbon dioxide inside the packages may have prevented accumulation of higher levels of ethylene, and also

inhibited the biological activity of ethylene in enhancing senescence and chlorophyll degradation.

4.0. CONCLUSION

This work was conducted to evaluate the use of MAP in combination with ice cooling in extending postharvest quality and shelf-life of green leafy vegetables using spinach as a model. The degree of benefit derived from this technique differed with the quality attribute. Modified atmosphere packaging and ice cooling of spinach leaves resulted in greater ascorbic acid retention thereby providing higher ascorbic acid to the consumer, greater chlorophyll retention thereby contributing to greener appearance, reduced weight and moisture losses, reduced wilting, increased marketability as measured subjectively using hedonic quality scale and maintained higher soluble protein levels. It is, therefore, concluded that this technique has a great potential in the extension of postharvest storage life of leafy vegetables during transportation, distribution and/or marketing especially in tropical countries where the temperatures may rise up to 40°C. However, if this method is to be applied for bulk transportation, there is need to evaluate the residue effect of MAP on the quality of the various commodities or devise methods of ascertaining the quality of packaged produce without violating the integrity of the package. For MAP to be a viable alternative to the conventional controlled atmosphere, more research is needed on film manufacture, package technique, film permeability for individual commodities as well as market trials.

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REFERENCES

Abe K., Mathooko F.M., Kiiyukia C., Koaze H., Muigai C.W. and Mwaniki M.W. (1996). Surveillance of postharvest handling of perishable food crops in Kenya. Technical Report, Department of Food Science and Postharvest Technology, Jomo Kenyatta University of Agriculture and Technology, Nairobi, Kenya. Pp. 36. Ahmadi H., Biasi W.V. and Mitcham E.J. (1999). Control of brown rot decay of

- nectarines with 15% carbon dioxide atmospheres. Journal of the American Society for Horticultural Science, 124, 708-712.
- 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. *Journal of Applied Microbiology*, 86, 429-438.
- Amanatidou A., Slump R.A. Gorris L.G.M. and Smid E.J. (2000). High oxygen and carbon dioxide modified atmospheres for shelf life extension of minimally processed carrots. *Journal of Food Science*, 65, 61-66.
- AOAC. (1984). Official Methods of Analysis. (14th ed). Association of Official Analytical Chemists. Washington D.C.
- Arjona H. E., Matta F. B. and Garner Jr. J. O. (1994). Wrapping in polyvinyl chloride film slows quality loss of yellow passion fruit. Hortscience, 29, 295-296.
- Arnon, D. (1949). Copper enzymes in isolated chloroplasts: polyphenoloxidase in *Beta vulgaris*. Plant Physiology, 24, 1-15.
- Barth M. M., Kerbel E. L., Perry A. K. and Schmidt S. J. (1993a). Modified atmosphere packaging affects ascorbic acid, enzyme activity and market quality of broccoli. *Journal of Food Science*, 58, 140-143.
- Barth M. M., Kerbel E. L., Broussard S. and Schmidt S. J. (1993b). Modified atmosphere packaging protects market quality in broccoli spears under ambient temperature storage. *Journal of Food Science*, 58, 1070-1072.
- Baxter L. and Walters L. (1990). Chemical changes in okra stored in air and controlled atmosphere. *Journal of the American Society for Horticultural Science*, 115: 452-454.
- Bhowmik S. R. and Pan J. C. (1992). Shelf life of mature green tomatoes stored in controlled atmosphere and high humidity. *Journal of Food Science*, 57, 948-953.
- Bradford M. M. (1976). A rapid and sensitive method for the quantification of microgram quantities of protein utilizing the principle of protein-dye binding. *Analytical Biochemistry*, 72, 248-254.
- Clark C.J. and Burmeister D.M. (199). Magnetic resonance imaging of browning development in "Braeburn" apple during controlled atmosphere storage under high CO₂. *HortScience*, 34, 915-919.
- Ezell B. D. and Wilcox M. S. (1959). Loss of vitamin c in fresh vegetables as related to

- wilting and temperature. Journal of Agricultural and Food Chemistry, 7, 507-509.
- Forney C. F., Rij R. E. and Ross S. R. (1989). Measurement of broccoli respiration rate in film-wrapped packages. *Hortscience*, 24, 111-113.
- Hsiao T. C. (1973). Plant responses to water stress. Annual Reviews of Plant Physiology, 24, 519-535.
- Kader A. A. (1986). Biochemical and physiological basis for effects of controlled and modified atmospheres on fruits and vegetables. *Food Technology*, 40(5), 99-104.
- Kader A. A., Zagory D. and Kerbel E. L. (1989). Modified atmosphere packaging of fruits and vegetables. *CRC Critical Reviews in Food Science and Nutrition*, 28, 1-30.
- Kailasapathy K. and Koneshan T. (1986). Effect of wilting on the ascorbate content of selected fresh green leafy vegetables consumed in Sri Lanka. *Journal of Agricultural and Food Chemistry*, 34, 259-260.
- Kays S.J. (1991). Postharvest Physiology of Perishable Plant Products. Pp. 356, 451. Van Nostrand Reinhold, New York.
- Klein B. P. and Perry A. K. (1982). Ascorbic acid and vitamin a activity in selected vegetables from different geographical areas of the united states. *Journal of Food Science*, 47, 941-948.
- Lazan H., Ali Z. M, Mohd A. and Nahar F. (1987). Water stress and quality decline during storage of tropical leafy vegetables. *Journal of Food Science*, 52, 1286-1288.
- Liao M-L. and Seib P. A. (1987). Selected reaction of l-ascorbic acid related to foods. Food Technology, 41(11), 104-107
- Lipton W. J. and Harris C. M. (1974). Controlled atmosphere effects on the market quality of stored broccoli. *Journal of the American Society for Horticultural Science*, 99, 200-205.
- Lownds N. K., Banaras M. and Bosland P. W. (1994). Postharvest water loss and storage quality of nine pepper (*capsicum*) cultivars. *Hortscience*, 29, 191-193.
- Mathooko F. M. (1995). Studies on the biochemical and physiological responses of fruits and vegetables to treatment with elevated levels of carbon dioxide. Ph.D. Thesis, Okayama University, Okayama, Japan.
- Mathooko F. M. (1996). Regulation of ethylene biosynthesis in higher plants by carbon dioxide. *Postharvest Biology and Technology*, 7, 1-26.

- Mathooko F. M., Sotokawa T., Kubo Y., Inaba A. and Nakamura R. (1993). Retention of freshness in fig fruit by CO₂-enriched atmosphere treatment or modified atmosphere packaging under ambient temperature. *Journal of the Japanese Society for Horticultural Science*, 62, 661-667.
- Mitchell F.G. (1992). Cooloing horticultural commodities. In: Postharvest Technology of Horticultural Crops. A.A. Kader (Editor). Pp. 53 63. University of California, USA.
- Nakhasi S., Schlimme D. and Solomos T. (1991). Storage potential of tomatoes harvested at the breaker stage using modified atmosphere packaging. *Journal of Food Science*, 56, 55-59.
- Shamaila M., Powrie W. and Skura B. J. (1992). Sensory evaluation of strawberry fruit stored under modified atmosphere packaging (map) by quantitative analysis. *Journal of Food Science*, 57, 1168-1174.
- Talasila R. C., Chau K. V. and Brecht J. K. (1995). Design of rigid modified atmosphere packages for fresh fruits and vegetables. *Journal of Food Science*, 60, 758-761.
- Thompson J. E., Legge R. E. and Barber R. F. (1987). The role of free radicals in senescence and wounding. *New Phytology*, 105, 317-321.
- Watada A. E., Kim S. D., Kim K. S. and Harris T. C. (1987). Quality of green beans, Bell peppers and spinach stored in polyethylene bags. *Journal of Food Science*, 52, 1637-1641.
- Yamauchi N. and Watada A. E. (1991). Regulated chlorophyll degradation in spinach leaves during storage. *Journal of the American Society for Horticultural Science*, 116, 58-62.
- Zagory D. and Kader A. A. (1988). Modified atmosphere packaging of fresh produce. *Food Technology*, 42(9), 70-77.
- Zhu, M., Chu, C.L., Wang, S.L. and Lencki R.W. (2001). Influence of oxygen, carbon dioxide and degree of cutting on the respiration rate of rutabaga. *Journal of Food Science*, 66, 30-37.
- Zhuang H., Barth M. M. and Hildebrand D. F. (1994). Packaging influenced total chlorophyll, soluble protein, fatty acid composition and lipoxygenase activity in broccoli florets. *Journal of Food Science*, 59, 1171-1174.