The Effect of Controlled Atmospheres on the Composition and Quality of Dill (*Anethum graveolens* L. cv. Ducat) Cultivated in Spring and Stored at Two Temperatures

D. Tsamaidi, I.C. Karapanos and H.C. Passam
Laboratory of Vegetable Production, Agricultural University of Athens
Iera Odos 75, 11855 Athens, Greece

Keywords: chlorophyll, vitamin C, total phenolics, weight loss, oxygen, carbon dioxide, shelf life

Abstract

This study was performed to evaluate four atmospheres varying in CO_2 and N_2 concentrations on the composition and quality of dill (*Anethum graveolens* L. cv. Ducat) during storage. Seeds were sown directly in the soil on 16/01/2010 and the plants harvested 93 days after sowing. The leaves (lamina and petiole) were weighed at harvest and immediately transferred to airtight plastic bags within which the atmospheres were modified by injection of O_2, CO_2 and N_2. The treatments that were applied (O_2-CO_2-N_2) were: (1): 20-0-80, (2): 20-5-75, (3): 20-10-70, (4): 20-15-65. All samples were stored for 14 days at 2 or 5°C and the gas composition within the bags maintained at the desired levels. From the results, it was found that weight loss during storage varied between 7 and 12% at 2ºC and 13 and 17% at 5ºC irrespective of the gas composition. Vitamin C concentration decreased during storage, but at both storage temperatures the decrease was less at the highest CO_2 level. The concentration of total phenolics decreased during storage, irrespective of the composition of the storage atmosphere. There was a loss of chlorophyll during storage, which tended to be greater at 5ºC than at 2ºC, but was prevented at both temperatures by the highest CO_2 concentration. In consequence, leaves stored under a gas composition of 20-15-65 (O_2-CO_2-N_2) were greener at the end of storage. This, in conjunction with the relatively higher vitamin C concentration in this treatment, indicates that 15% CO_2 was the most suitable storage atmosphere for quality retention in dill, while the preferred temperature was 2ºC due to lower weight loss.

INTRODUCTION

Dill (*Anethum graveolens* L.), an annual aromatic herb of the Apiaceae (Umbelliferae) family, is grown widely throughout Europe, America and Asia for its aromatic foliage, which is used either fresh or dried to flavor foods. It is best, however, when used fresh because flavor and aroma decrease during drying (Grieve, 2011).

The shelf-life of fresh vegetable produce is limited by water loss (O'Beirne, 1990), which leads to weight loss and shriveling, and physiological ageing (senescence), which occurs when the starch/sugar stored within the product is consumed during respiration; therefore shelf-life is inversely proportional to respiration rate (Day 1990). Fresh green herbs, like dill, are highly perishable due to rapid senescence after harvest. Wounding, temperature and water loss are the major factors affecting the rate of senescence, and deterioration during storage is manifested by reduced nutritional value, chlorophyll loss, lamina abscission and the onset of decay (Aharoni et al., 1993).

Controlled atmosphere (CA) storage is a method in which the concentrations of gases within the storage environment are continually monitored and adjusted to maintain optimum levels. Elevated CO_2 concentrations may reduce the respiration and ethylene production rates of fruit, thereby delaying ripening and extending storage life. Moreover, CO_2 concentrations above 10% can reduce the incidence of some physiological disorders induced by C_2H_4, and inhibit the growth of pathogens (Zagory and Kader, 1988). Chlorophyll loss is delayed both by low O_2 (<5%) and by elevated CO_2 (>5%) (Lipton and Mackey, 1987). However, temperature must be carefully controlled during CA storage to avoid fluctuations in respiration, water condensation and even anaerobiosis, while low temperatures are normally applied to reduce respiration and prevent spoilage (Stiles, 1991).

Recent years have seen an increasing demand for both fresh and dried herbs for local markets and export, thus encouraging their cultivation by small-scale farmers (Matthews and Jack, 2011). For quality retention, temperatures during transit and marketing should be low. Additionally, modified atmosphere (MA) packaging of herbs, including dill (Tsamaidi et al., 2011) may delay leaf senescence due to the elevation of CO_2 concentration (Aharoni et al., 1988).

Because the effect of CA storage on dill has not been previously reported, we examined the effect of maintaining increased concentrations of CO_2 within the storage atmosphere on the quality characteristics of this herb at two temperatures.

MATERIALS AND METHODS

Plant material and cultivation

The experiment was carried out at the Laboratory of Vegetable Production of the Agricultural University of Athens. Seeds of dill (*Anethum graveolens* L. cv. Ducat) were sown on 16 January 2010 in soil enriched with 150g potassium monophosphate, 40g potassium sulphate, 20g magnesium sulphate, 10g trace elements (Nutrileaf) and 300g marble dust per m^3 in an unheated, plastic-covered greenhouse. During cultivation, plants were fertilized with 300 ppm N in the irrigation water two or three times a week, beginning from the stage of 3-4 leaves. Plants were harvested on 19 May 2010 (123 days after sowing) just prior to flowering.

Plants were harvested 1cm above the soil and, after the removal of any old, senescent leaves, transferred to the laboratory where plant height, leaf number and foliage fresh weight were recorded.

Treatments

Randomly selected leaves were placed in sealed, airtight high density polethylene bags (18 x 41 cm, volume 600 ml) which were first aspirated and then injected with O_2, CO_2 and N_2 through a gas mixer (PBI Dansensor, Map Mix 9000 ME, Denmark) to achieve the desired atmosphere. Each bag contained 3 leaves (together with their petioles), weighing 15g. The CA treatments that were applied (O_2-CO_2-N_2) were: (A1): 20-0-80, (A2): 20-5-75, (A3): 20-10-70, (A4): 20-15-65. All samples were stored for 14 days at 2°C or 5°C and the gas composition within the bags was maintained constant $\pm$ 1-1.5% by air renewal every two days using a gas sensor (PBI Dansensor Checkmate II) and gas mixer. Each treatment consisted of 5 bags replicated 5 times.

Determination of quality characteristics

At the end of storage, leaves were weighed to determine weight loss and then dried at 72°C for 3 days for the determination of dry matter. Quality characteristics were monitored before and after storage. Chlorophyll a, chlorophyll b and total chlorophyll concentrations were measured using the method of Arnon (1949), vitamin C (L–ascorbic acid) was measured according to Bajaj and Kaur (1981), while total phenolics were measured using the method of Lisiewska et al. (2006).

Statistical analysis

The results were subjected to analysis of variance (ANOVA) and means compared by the application of Duncan's multiple range test using the statistical package SPSS 15.0.

RESULTS

Weight loss at the end of 14 days' storage varied between 7 and 12% at 2ºC and 13 and 17% at 5ºC irrespective of the gas composition of the storage atmosphere, and was significantly higher at 5ºC than at 2ºC, except in treatment A3 (Table 1).

Vitamin C concentration within the leaves decreased during storage in all treatments, but at both storage temperatures the decrease was less at the highest CO_2 level (treatment A4). The decrease in vitamin C concentration was similar at 2ºC and 5ºC except in treatment A2, where the decrease in vitamin C concentration was higher at 5ºC (Table 2).

The concentration of total phenolics decreased during storage at both temperatures. At 5ºC, the decrease in phenolics was not influenced by the gas composition, whereas at 2ºC the decrease was greater in treatment A4 than in treatment A3. Significant differences between the two temperatures were not observed in any of the treatments (Table 3).

The total chlorophyll content of the leaves decreased during storage in atmospheres containing 0-5% CO_2 (treatments A1-A2) at 2ºC and in those containing 0-10% CO_2 (treatments A1-A3) at 5ºC. At both temperatures, there was no loss of total chlorophyll in 15% CO_2 (treatment A4), but in the other treatments the loss was greater at 5ºC than at 2ºC, although not always to a statistically significant level. The pattern of decrease in chlorophyll a and b was similar to that of the total chlorophyll (Table 4).

Dry matter (Table 5) increased during storage at both temperatures and was not significantly affected by the gas composition or the storage temperature.

DISCUSSION

Controlled atmospheres recommended for the storage of fresh herbs (chervil, chives, coriander, dill, sorrel and watercress) are 5-10% O_2 + 4-6% CO_2 at temperatures of 0-5ºC (Saltveit, 1997; Saltveit, 2003). Aharoni et al. (1988) found that 5% CO_2 reduced chlorophyll loss in dill and chives, but a combination of 5% CO_2 and 5% O_2 was more effective than either gas alone (Lipton and Mackey, 1987). At 0.5ºC and 95-100% R.H., postharvest life ranges from 1 week for chervil to 2 weeks for coriander and dill, and up to 3 weeks for savory, whereas basil can be stored satisfactorily at 12°C and 95-100% R.H. (Gorini, 1981; Aharoni et al., 1993) for up to 2 weeks (Lange and Cameron, 1994).

High quality dill is characterized by its fresh appearance and dark green leaves. The prevention of water loss and chlorophyll degradation during storage is therefore of the greatest importance. In the present experiment, weight (water) loss was less at 2ºC than at 5ºC, indicating that the lower storage temperature is preferable. Moreover, it is likely that quality during storage could be further improved by rapid cooling to just above 0°C immediately after harvest (Aharoni et al., 1988). Although vacuum-cooling of leafy produce may be preferable to hydro-cooling (Aharoni et al., 1988), dill is a relatively minor crop and the cost of vacuum application is likely to be prohibitive.

Aharoni et al. (1989) reported that senescence (yellowing and decay) of chives packed in polyethylene-lined boxes was delayed more by the accumulation of CO_2 (5.7%) than by the decrease in O_2 (12%). In dill, high CO_2 concentrations (5-11%) reduced chlorophyll loss, so that visual quality after storage in polyethylene for 5 days at 6ºC + 2 days at 12ºC was between 'fair' and 'good' (Aharoni et al., 1993). Similarly, chlorophyll retention was achieved by MA storage of dill in permeable low-density polyethylene for 7 days at 2ºC or 7ºC (Tsamaidi et al., 2011). Under MA storage, the CO_2 concentration within the package increased with time, and O_2 decreased. Under CA conditions, however, the concentrations of these two gases were virtually constant. Therefore, it is clear from the data of Table 4 that for total inhibition of chlorophyll degradation during the storage of dill, CO_2 should be maintained at a concentration of 15%.

Positive effects of increased CO_2 concentration on the maintenance of vitamin C is known in leafy vegetables (Souzan and Abd el-Aal, 2007), while the reverse has been observed for phenolics (Oboh, 2005). During CA storage of dill, vitamin C loss was less at the highest CO_2 concentration (15%), whereas total phenolics were reduced more in 15% CO_2 at 2ºC (but not at 5ºC) (Table 3). Similarly during MA storage of dill for 7 days, vitamin C decreased less at high CO_2 levels at 7ºC (but not at 2ºC), whereas no effect of CO_2 concentration on the phenolics concentration was observed (Tsamaidi et al., 2011).

It is well known that increases in CO_2 concentration lead to a reduction in the respiration rate of stored produce, e.g. chives (Ishi and Okubo, 1984). The reduction of respiratory activity by high CO_2 levels correlates with improved retention of chlorophyll and vitamin C (Kader, 1986)and is apparently greater under CA storage with a high CO_2 concentration (15%) than under MA storage, where the CO_2 concentration fluctuates (Tsamaidi et al., 2011).

CONCLUSION

For quality retention of fresh dill leaves during CA storage, a constantly-maintained atmosphere of 20-15-65 (O_2-CO_2-N_2) and a temperature of 2ºC are recommended. At the end of storage (14 days) leaves stored under these conditions were fresher and greener (no chlorophyll loss) and had a higher vitamin C content than leaves stored under lower levels of CO_2 or at a higher temperature (5ºC).

ACKNOWLEDGEMENTS

This research has been co-financed by the European Union (European Social Fund – ESF) and Greek national funds through the Operational Program "Education and Lifelong Learning" of the National Strategic Reference Framework (NSRF) - Research Funding Program: Heracleitus II. Investing in knowledge society through the European Social Fund.

Literature Cited

Aharoni, N., Dvir, O., Chalupowis, D., Zhalupowisz, Z. and Aharon, Z. 1993. Coping with postharvest physiology of fresh culinary herbs. Acta Hort. 344: 69-78.

Aharoni, N,. Dvir, O., Reuveni, A., Aharon, Z., Gur, G., Noi, M., Tene, A., Kuris, U. and Efrat, U. 1988. Film packaging and vacuum precooling of green herbs for export. Hassadeh, 68: 767-771.

Aharoni, N., Reuveni, A. and Dvir, O. 1989. Modified atmospheres in film packages delay senescence and decay of fresh herbs. Acta Hort. 88: 255-262.

Arnon, D.I. 1949. Copper enzyme in isolated chloroplast polyphenoloxidase in *Beta vulgaris*. Plant Physiol. 24: 1-15.

Bajaj, K.L. and Kaur, G. 1981. Spectrophotometric determination of L-ascorbic acid in vegetables and fruits. The Analyst, 106 (1) 117–120.

Day, B. 1990. Modified atmosphere packaging of selected prepared fruit and vegetables. In: Processing and Quality of Foods. Vol 3: Chilled Foods: The Revolution in Freshness., ed Zeuthen, P., Cheftel, J.C., Eriksson, C., Gormley, T.R.P., Linko, P. and Paulus, K. Elsevier, London, UK, pp 230-233.

Gorini, F. 1981. Vegetable schedules. 2. Leafy vegetables. Chervil. Inform. di Ortoflorofrutticolt. 22:3-4.

Grieve, M. 2011. Dill. A Modern Herbal. www.Botanical.com

Ishi, K. and Okubo, M. 1984. The keeping quality of Chinese chive (*Allium tuberosum* Rottler) by low temperature and seal-packaging with polyethylene bag. J. Jap. Soc. Hort. Sci. 53 (1): 87-95.

Kader, A.A. 1986. Biochemical and physiological basis for effects of controlled and modified atmospheres on fruit and vegetables. Food Tech. 40: 99-100.

Lange, D.D. and A.C. Cameron. 1994. Postharvest shelf-life of sweet basil (*Ocimum basilicum*). HortSci. 29:102-103.

Lisiewska, Z., Kmiecik, W. and Korus, A. 2006. Content of vitamin C, carotenoids, chlorophylls and polyphenols in green parts of dill (*Anethum graveolens* L.) depending on plant height. J. Food Comp.Anal. 19: 134–140.

Lipton, W.J. and Mackey, B.E. 1987. Physiological and quality responses of Brussels sprouts to storage in controlled atmospheres. J. Amer. Soc. Hort. Sci. 112: 491-496.

Matthews, M. and Jack, M. 2011. Spices and Herbs for Home and Market. Diversification Booklet No. 20, Food and Agriculture Organization, Rome, 70 pp.

O'Beirne, D. 1990. Modified atmosphere packaging of fruits and vegetables. In: Chilled Foods: The State of the Art, ed Gormley, T. Elsevier Applied Science, Barking, UK, pp 183-199.

Oboh, G., 2005. Effect of blanching on the antioxidant property of some tropical green leafy vegetables. Food Sci. Tech. 38: 513-517

Saltveit, M.E. 1997. A summary of CA and MA requirements and recommendations for harvested vegetables. In: Seventh International Controlled Atmosphere Research Conference. Volume 4: Vegetables and Ornamentals. Univ. Calif., Davis, Postharvest Horticulture Series 18:98-117.

Saltveit, M.E. 2003. A summary of CA requirements and recommendations for vegetables. Acta Hort. 600: 723-727.

Souzan, S.L. and Abd El-Aal, H.A. 2007. Minerals profile- shelf life extension and nutritive value of fresh green leafy vegetables consumed in Egypt. In: Afr. Crop Sci. Conf. Proc. 8: 1817-1826.

Stiles, M.E. 1991. Scientific principles of controlled/modified atmosphere packaging. In: Modified Atmosphere packaging of food, ed. Ooraikul, B. and Stiles, M.E., Ellis Horwood Ltd., Chichester, U.K., pp. 18-25.

Tsamaidi, D., Stavrinou, E., Petrou, F. and Passam, H.C. 2011. The effect of modified atmospheres during storage on the quality characteristics of dill (*Anethum graveolens* L.). Proc. of the Greek Society for Horticultural Science (Limassol) 15A, 268-271.

Zagory, D. and Kader, A.A., 1988. Modified atmosphere packaging of fresh produce. Food Technology 42 (9): 70-77.

Tables

Table 1. Effect of controlled atmospheres and storage temperature on the weight loss of dill leaves after 14 days' storage.

	Weight loss of leaves (%)	
Treatment	**2ºC**	**5ºC**
A1 (20/0/80)	7.07 (b)	13.50 (a)
A2(20/5/75)	8.04 (b)	17.70 (a)
A3 (20/10/70)	12.76	13.73
A4(20/15/65)	8.71 (b)	17.57 (a)

Means within each column did not differ significantly (p=0.05)
Means within rows followed by a different letter in parenthesis are significantly different (p=0.05)

Table 2. Effect of controlled atmospheres and storage temperature on the concentration of vitamin C in dill leaves after 14 days' storage.

	Vitamin C (Ascorbic acid) (mg/100g f.m.)	
Treatment	**2ºC**	**5ºC**
A0 (Fresh)	511.77 a	511.77 a
A1 (20/0/80)	190.29 d (a)	217.06 c (a)
A2 (20/5/75)	290.88 bc (a)	210.59 c (b)
A3 (20/10/70)	236.18 cd (a)	218.53 c (a)
A4 (20/15/65)	320.00 b (a)	273.82 b (a)

Means within each column followed by a different letter are significantly different (p=0.05)
Means within rows followed by a different letter in parenthesis are significantly different (p=0.05)

Table 3. Effect of controlled atmospheres and storage temperature on the concentration of total phenolics in dill leaves after 14 days' storage.

	Total phenolics (mg GAE/100g f.m.)	
Treatment	**2ºC**	**5ºC**
A0 (Fresh)	273.90 a	273.90 a
A1 (20/0/80)	192.93 bc	179.83 b
A2 (20/5/75)	201.97 bc	200.04 b
A3 (20/10/70)	220.10 b	211.55 b
A4 (20/15/65)	165.90 c	205.97 b

Means within each column followed by a different letter are significantly different (p=0.05)
Means within rows are not significantly different (p=0.05)

Table 4. Effect of controlled atmospheres and storage temperature on the concentrations of chlorophyll a, chlorophyll b and total chlorophyll in dill leaves after 14 days' storage.

	Chlorophyll a (mg/100g f.m.)		Chlorophyll b (mg/100g f.m.)		Total chlorophyll (mg/100g f.m.)	
Treatment	**2ºC**	**5ºC**	**2ºC**	**5ºC**	**2ºC**	**5ºC**
A0 (Fresh)	94.83 a	94.83 a	38.74 ab	38.74 a	133.57 ab	133.57 a
A1 (20/0/80)	68.84 b (a)	15.56 c (b)	30.58 b (a)	7.52 c (b)	99.42 cd (a)	23.08 c (b)
A2 (20/5/75)	48.76 b	32.32 c	29.78 b (a)	14.80 bc (b)	78.53 d (a)	47.12 c (b)
A3 (20/10/70)	67.04 b	68.22 b	41.27 ab (a)	22.68 b (b)	108.31 bc	90.89 b
A4 (20/15/65)	96.80 a	89.13 ab	45.52 a	32.28 a	142.31 a	121.42 a

Means within each column followed by a different letter are significantly different (p=0.05)
Means for each parameter within each row followed by a different letter in parenthesis are significantly different (p=0.05)

Table 5. Effect of controlled atmospheres and storage temperature on the dry matter of foliage biomass after 14 days' storage of dill leaves.

	Dry matter (%)	
Treatment	**2ºC**	**5ºC**
A0 (Fresh)	1.89 b	1.89 b
A1 (20/0/80)	13.08 a	13.73 a
A2 (20/5/75)	13.83 a	13.47 a
A3 (20/10/70)	14.00 a	13.30 a
A4 (20/15/65)	13.69 a	14.28 a

Means within each column followed by a different letter are significantly different (p=0.05)
Means within rows are not significantly different (p=0.05)