Responses of Parthenocarpic Cucumbers to Low-oxygen Storage

Angelos K. Kanellis[1], Leonard L. Morris, and Mikal E. Saltveit, Jr.[2]
Department of Vegetable Crops, Mann Laboratory, University of California, Davis, CA 95616

Additional index words. *Cucumis sativus*, controlled atmosphere, CA, respiration, respiratory quotient, RQ, storage, postharvest

Abstract. **Parthenocarpic cucumber fruit (*Cucumis sativus* L. cv. Deliva) of marketable maturity (10 to 14 days after anthesis) were held at 12.5° or 20°C in reduced O_2 levels for 5 or 18 days before transfer to air. Carbon dioxide production at reduced O_2 levels was generally less than in air; however, at O_2 levels < 0.5%, anaerobic respiration resulted in increased rates of CO_2 production. Upon transfer to air after 18 days, all samples from reduced O_2 showed increased CO_2 production rates that equalled or exceeded that of the air controls. Except at 0.0% and 0.25% O_2 levels, ethylene production was increased in reduced O_2. After transfer to air, ethylene production increased and the increase was inversely related to the previous O_2 level. Ethanol and acetaldehyde production were measureable for fruit held in 1% O_2 after 18 days at 12.5° and showed dramatic increases at lower O_2 levels. Low-O_2 injury (pitting) developed on most fruit held at 0.0% O_2 and on many fruit held at 0.25% O_2. Only minimal commercial benefits are likely to be realized from storage of 1 to 3 weeks in 0.5% to 2.0% O_2 at 12.5°.**

Previous CA studies with cucumber fruit have focused on determining optimum O_2 and CO_2 levels for retention of visual quality (2, 3, 17, 18). When only O_2 levels were modified, either short exposure (4) or one concentration (13) was tested. Little information existed on the physiological responses of cucumbers to reduced O_2 levels, and no studies have included parthenocarpic cucumbers. Eaks (4) suggested 1% O_2 as a critical level for aerobic respiration of cucumbers, based on his observation that CO_2 production was minimal at this level.

The main objective of this research was to determine the critical O_2 level for aerobic respiration of parthenocarpic cucumbers. Production of CO_2, C_2H_4, ethanol, and acetaldehyde, and consumption of O_2 also were studied. The effect of low-O_2 storage on retention of visual quality was also observed.

Materials and Methods

Parthenocarpic 'Deliva' cucumber fruit were obtained from commercial greenhouses near Lodi, Calif. Fruit of marketable maturity (≈450 to 500 g) were harvested 10 to 14 days after anthesis and transported directly to the Mann Laboratory, Davis, Calif. Cucumbers for Expt. 1 were spring-grown and from older plants than those for Expt. 2, which were summer-grown and from a different grower. Fruit were washed in a 0.05% sodium hypochlorite solution (1/100 dilution of 5.25% commercial bleach) and air-dried before sorting (Expt. 2 only).

One fruit each from a group of similar size and appearance was placed into each of two samples per treatment. This distribution of fruit was continued until there were 8 and 9 fruit in each sample for Expts. 1 and 2, respectively. Each sample was held in a 19-liter glass jar that was ventilated with a humidified gas mixture made by mixing a metered flow of air and N_2 in a flow-through system (11). Flow rates were selected and maintained to ensure that CO_2 levels did not exceed 0.25%. In Expt. 1, fruit were held at 12.5° in air or 8%, 4%, 2%, 1%, 0.75%, 0.5%, or 0% O_2. In Expt. 2, fruit were held at 12.5° or 20° in air or 2%, 1%, 0.75%, 0.5%, 0.25%, or 0% O_2. One of the two samples from each treatment was transferred to humidified air after 5 days and the second after 18 days.

Oxygen levels were measured with an S-3A Oxygen Analyzer (Applied Electrochemistry Inst., Sunnyvale, Calif.). Carbon dioxide levels were measured in 1-ml gas samples using an Horiba PIR-2000 infrared gas analyzer. Respiration rates were calculated from the measured differences in O_2 and CO_2 concentrations between the inlet and outlet streams, and respiratory quotient (RQ) values were calculated from these rates expressed in terms of volumes.

Ethylene levels were measured in 1-ml gas samples with a Carle model ACG-211 series S FID gas chromatograph. Relative levels of ethanol and acetaldehyde were determined in 1-ml gas samples using a Hewlett-Packard model 5730A FID gas chromatograph. Volatile separation was performed with a 2.7 m × 6.4 mm Porapak-Q 80/100 mesh column. Difficulties in preparing quantitative standards of ethanol and acetaldehyde necessitated a comparison of their relative levels against a 1.1 $\mu l \cdot liter^{-1}$ C_2H_4 standard (5).

Visual quality and color were scored as previously described (7). Visual quality was rated on a scale where 9 = excellent, 7 = good, 5 = fair, 3 = unsalable at retail, and 1 = not usable. Color was rated from 5 to 1, where 5 = dark green and 1 = yellow.

Results and Discussion

Respiration. The general effect of O_2 levels on respiration rates were similar in both experiments and at both 12.5° and 20°C (data not shown). Except for 0.0% O_2, CO_2 production was lower under reduced O_2 as compared to the air control (Figs. 1A and 2). The rate was lowest at 0.5% and 0.75% in all comparisons and averaged ≈30% to 40% of the control. The respiration rate under 0.25% O_2 was higher than at 0.5% or 0.75% (Fig. 1). This difference suggests that anaerobic respiration was not completely inhibited at this O_2 level. Carbon dioxide production under 0.0% O_2 (100% N_2) tended to be higher than at O_2 levels between 0.5% and 2%, especially at the beginning and end of the holding period (Fig. 2). The rates of CO_2 production at 4% and 8% were similar and, in most cases, were intermediate between the rates at 2% and air for any given day (Fig. 2).

Following transfer to air after 18 days at reduced O_2 levels,

Received for publication 6 Feb. 1987. The cost of publishing this paper was defrayed in part by the payment of page charges. Under postal regulations, this paper therefore must be hereby marked *advertisement* solely to indicate this fact.

[1]Present address: Dept. of Biology, Univ. of Crete, 71110 Iraclion, Crete, Greece.
[2]To whom reprint requests should be addressed.

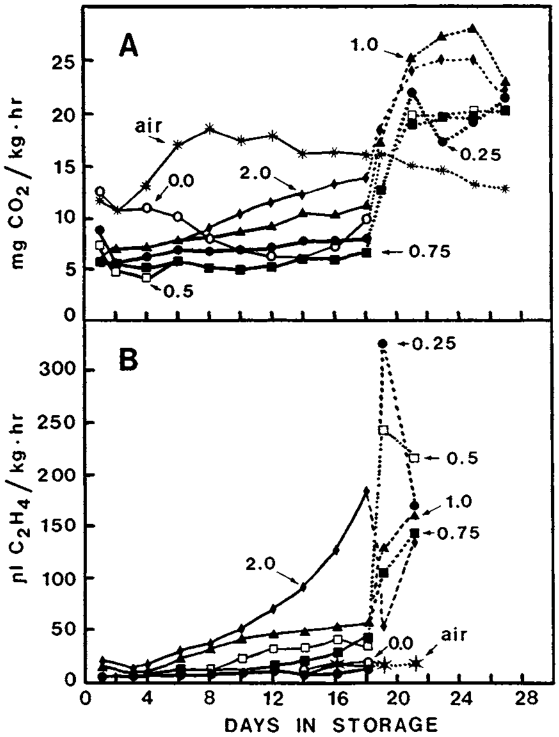

Fig. 1. Effect of O_2 levels at 12.5° on CO_2 (**A**) and C_2H_4 (**B**) production by cucumbers during an 18-day exposure (solid lines) and after transfer to air (broken lines) (Expt. 2). Note that CO_2 values for the 0.5% and 0.75% treatments are the same for days 6 through 12.

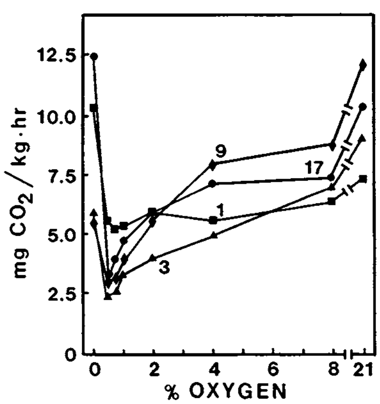

Fig. 2. Carbon dioxide production rates of cucumbers as a function of O_2 level at 12.5°C on days 1, 3, 9, and 17 of holding (Expt. 1).

the respiration rate increased markedly (Fig. 1A). Except for the 8% O_2 treatment in Expt. 1 (data not shown), the rate after transfer exceeded that of the air control. The 0.0% O_2 treatment was discarded at the time of transfer due to physiological breakdown. The increase in respiration in air probably was associated with the increased O_2 level and with the increased production of ethylene after transfer (Fig. 1B). Respiration rates also increased following transfer to air after 5 days under CA, but the responses were less (data not shown).

These data are in general agreement with those of Eaks (4), who used O_2 levels of 0% to 21% and found that CO_2 production was reduced at the lower O_2 levels, except at 0% O_2. Similar observations have been reported for zucchini squash (10). However, the results after transfer to air are not comparable to those of Wills et al. (21), who reported that 2- to 4-day exposures to 2% or 4% O_2 markedly reduced the respiration of zucchini squash following transfer to air at 20°.

Eaks (4) suggested 1% O_2 as the critical level for aerobic respiration in cucumbers, but he did not test lower O_2 concentrations. Our results (Figs. 1A and 2) indicate that 0.5% approximates the critical O_2 level at 12.5°C. This was also true at 20° (results not shown). As stated above, the 0.25% O_2 did not appear to completely suppress anaerobic respiration at 12.5°.

Respiratory quotient. The difficulty of measuring small changes in O_2 concentrations in the gas streams introduced variability into the calculations of O_2 consumption and resulting RQ values. Hence, we averaged the RQ values calculated from daily measurements of CO_2 production and O_2 consumption made during the 18-day holding period (Table 1). The averaged RQ values for all reduced O_2 treatments were greater than the RQ of the appropriate air control. The averaged values for the 0.25% and 0.5% O_2 treatments were in all cases significantly greater than their air controls. The RQ values for 0.75% and 1% were significantly greater than the air control in two of the three comparisons (i.e., in Expt. 1 at 12.5°C and Expt. 2 at 20°, but not in Expt. 2 at 12.5°). These elevated RQ values indicate that anaerobic respiration was occurring. This conclusion applies to parthenocarpic fruit without a film or wax coating. Any increase in diffusive resistance caused by such applications would require an increase in ambient O_2 to maintain the necessary internal O_2 level for aerobic respiration.

Ethylene production. Production of C_2H_4 in air ranged from ≈5 to 15 $nl \cdot kg^{-1} \cdot hr^{-1}$ at 12.5°C in Expt. 1, and from ≈10 to 20 $nl \cdot kg^{-1} \cdot hr^{-1}$ at both 12.5° and 20° in Expt. 2. These values are similar to those previously published for cucumber fruit (8, 14). Considering all tests, C_2H_4 production in CA did not correlate well with O_2 levels, except that it was near the limits of detectability in 0.0% O_2. However, at 12.5° in Expt. 2, the C_2H_4 production rate increased with time at 0.5% to 2.0% O_2; this increase appeared to correlate with the O_2 levels (Fig. 1B). Mencarelli et al. (10) found that C_2H_4 production of zucchini squash at 5° and 10° was a function of O_2 concentration. However, stress from chilling injury, which has been shown to induce C_2H_4 production (20), may have been a factor in their study.

After transfer to air from reduced O_2, the C_2H_4 production rate usually exceeded that of the air control. The increase over the air control was usually related inversely to the prior O_2 level (Fig. 1B). The above result applies to both the 5- and 18-day exposures. Production of C_2H_4 from its immediate precursor,

Table 1. Average RQ values of parthenocarpic cucumber fruit as calculated from daily measurements of CO_2 production and O_2 consumption during the 18 days of holding.

	Respiratory quotient[z]		
		Expt. 2	
O_2 concn (%)	Expt. 1, 12.5°C	12.5°C	20°C
21 (air)	0.90 d	0.94 c	0.88 d
8	1.02 bc	---[y]	---[y]
4	0.99 c	---[y]	---[y]
2	1.02 c	1.00 c	0.99 cd
1	0.97 c	1.02 bc	1.04 c
0.75	1.10 b	0.97 c	1.08 c
0.5	1.16 a	1.10 b	1.22 b
0.25	---[y]	2.04 a	1.57 a

[z]Mean separation within columns by LSD at the 5% level.
[y]Treatments not included in the experiment.

1-aminocyclopropane-1-carboxylic acid (ACC), is dependent on the O_2 level in the tissue (1). Anaerobic atmospheres inhibit C_2H_4 production from ACC and often results in the accumulation of ACC. Upon transfer to air, this accumulated ACC is converted to C_2H_4 within 1 to 2 days (1). This expected elevated rate of C_2H_4 production was measured 1 day after transfer to air. The continued production of C_2H_4 at rates greater than the air control 3 days after transfer to air suggests that the lowest O_2 levels, which caused physiological injury, also may have resulted in the production of stress C_2H_4 (Fig. 1B).

Low-O_2 injury. Physiological injury occurred at 0.25% O_2 and is reflected by reduced visual quality scores (Fig. 3). All fruit held in 0.0% O_2 developed injury symptoms that resembled the pitting observed in cucumbers following chilling. Pitting first appeared at both ends and gradually covered the fruit. Another low-O_2 injury symptom was shriveling near the stem end. Fruit in 0.0% and 0.25% O_2 accumulated exudate on their surface. Along with pitting, this favored decay development. Li et al. (9) mentioned that O_2 levels between 0.0% and 2% caused shrinking of the stem end of cucumbers and increased susceptibility to infection by microorganisms. In this study, such effects were observed only in 0.0% or 0.25% O_2. During 18 days in 0.0% O_2, almost all the fruit showed decay; however the surface color remained dark green. A similar retention of chlorophyll was observed in oat leaves kept in N_2 (15).

Pitting had developed in 30% of the fruit in 0.25% O_2 after 5 days, and in 70% of the fruit after 18 days. When transferred to air, all fruit from this treatment deteriorated rapidly. The occurrence of symptoms typical of low-O_2 injury was variable, even affecting some fruit in 2% O_2. The internal tissue of affected fruit had a watery appearance and off-odors were detected.

Eaks (4) observed that pitting developed in standard (i.e., nonparthenocarpic) cucumbers only after transfer to air from 8 days in 0.0% O_2 at 5° or 15°C. In this study, however, parthenocarpic fruit developed pitting while in a humidified CA of 0.0% or 0.25% O_2. Since pitting is a secondary symptom of physiological injury, its development could depend on many internal and external factors. Mencarelli et al. (10) observed that zucchini squash held in 0.5% O_2 at 7.5° accumulated exudate on the surface; they concluded that 0.5% O_2 was insufficient for aerobic respiration. This apparent difference between cucumbers and squash in susceptibility to low-O_2 injury might be an inherent characteristic of the fruit. Also, chilling injury at 7.5° may have contributed to symptom development on the squash.

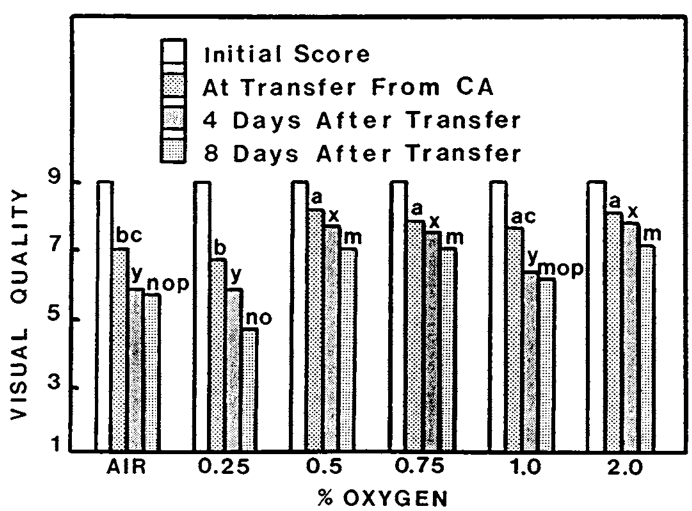

Fig. 3. Effect of CO_2 levels on visual quality of cucumbers held at 12.5°C for 18 days in CA and then transferred to air for 4 or 8 days. Mean separation at each evaluation time by LSD test at the 5% level. Values assigned the same letter did not differ significantly. Visual quality was rated on a scale of 9 to 1, where 9 = excellent, 7 = good, 5 = fair, 3 = unsalable at retail, and 1 = not usable.

Visual quality and shelf-life. Cucumbers held in 0.0% O_2 at either 12.5° or 20°C deteriorated to an unsalable condition during the 18-day exposure. Quality was still acceptable after a 5-day exposure to 0.0% O_2 at either temperature, but the fruit deteriorated rapidly following transfer to air. The visual score of fruit held at 0.25% O_2 for either 5 or 18 days did not vary from that of the air controls at 12.5°, but was reduced at 20°. In contrast, O_2 levels of 0.5% to 8% in Expt. 1 and 0.5% to 2.0% in Expt. 2 resulted in higher visual scores than the air control after 18 days at 12.5°, and at most of the subsequent inspections (Fig. 3 for 18 days in CA at 12.5°). For the most part, the differences between treatments were not significant after 5 days in CA at 12.5° (data not shown). At 20°, differences were not significant following either 5 or 18 days in CA for the 0.5% to 2.0% O_2 treatments (data not shown). These results are in general agreement with Eaks (4), who found that holding for 8 days in reduced O_2 at 15° did not prolong subsequent life of cucumbers after transfer to air at 25°.

Blisters, a physiological disorder of cucumbers (7, 12, 13), appeared during storage. Blisters took longer to appear on fruit transferred to air from lower O_2 levels. All air control fruit developed blisters during the 18 days at 20°C, yet no blisters were observed in fruit held in low O_2 for the same period. Upon transfer to air, however, blisters developed much faster in fruit held at 20° than in those held in 12.5°.

Greeness was retained better by fruit held 18 days in low O_2 than by fruit held in air (Table 2). Similarly, Apeland (2) observed that fruit held for 23 days in either 5% O_2 or 5% CO_2 retained good color; however, after transfer to air, fruit previously held in high CO_2 yellowed faster than those from the low-O_2 treatment. Both Apeland (2) and Wang (19) observed reduced yellowing of fruit held in 1% O_2.

Ethanol and acetaldehyde production. The effects of O_2 levels <2% on ethanol and on acetaldehyde production were generally similar (data not shown). Fruit produced relatively large amounts of ethanol and acetaldehyde when held in 0.0% O_2,

Table 2. Effect of previous O_2 level on color of parthenocarpic cucumber fruit after transfer to air following an 18-day low-O_2 exposure (9 days in Expt. 1 and 8 days in Expt. 2).

	Color scores[zy]		
		Expt. 2	
O_2 concn (%)	Expt 1, 12.5°C	12.5°C	20°C
21 (air)	3.5 b	4.0 b	3.9 b
8	4.2 a	---[x]	---[x]
4	4.2 a	---[x]	---[x]
2	3.9 a	4.9 a	4.7 a
1	3.9 a	4.8 a	4.7 a
0.75	4.5 a	4.8 a	4.6 a
0.5	4.4 a	4.7 a	4.6 a
0.25	---[x]	4.6 ab	---[x]

[z]Color scale: 5 = dark green, 4 = green, 3 = yellowish green, 2 = greenish yellow, and 1 = yellow.
[y]Mean separation by LSD at the 5% level.
[x]Treatments not included in the experiment.

while production of both of these volatiles was dramatically lower in fruit held in 0.5% O_2. The small amounts of both volatiles produced by the fruit in 0.5% and 0.75% O_2 did not result in reduced visual quality or color retention. Only trace amounts were measured at 2% or 4% O_2, and none were detected at 8% or 21% O_2 at either 12.5° or 20°C. For example, acetaldehyde production at 12.5° was measurable at 1% O_2; slightly higher at 0.75%; increased 2-fold at 0.5%, 9-fold at 0.25%, and 14-fold at 0%.

Based on respiration rates, RQ values, development of symptoms of low-O_2 injury, and visual deterioration, it appears that 0.25% O_2 resulted in anaerobiosis at 12.5° and 20°C. The effect of O_2 level on ethanol and acetaldehyde production are in agreement with this. Upon transfer to air, evolution of ethanol ceased in 1 day for fruit previously held in 0.75% O_2, but this decline took 11, 9, and 3 days for fruit held in 0%, 0.25%, or 0.5% O_2, respectively. With acetaldehyde, the time for production to cease for fruit previously held in 0%, 0.25%, 0.5%, and 0.75% O_2 was 9, 7, 1, and <1 day, respectively. The evolution of these volatiles after transfer could be due either to continued production and/or to continued diffusion of previously produced volatiles.

Segall et al. (16) detected acetaldehyde, but not ethanol, in waxed cucumbers. Acetaldehyde accumulation depended on the number of wax coatings, which presumably created reduced O_2 levels inside the tissue. Francis et al. (6) found that 1% O_2 + 16% CO_2 promoted anaerobic respiration and alcohol accumulation in segmented squash of different types. A taste panel could detect 50 mg of alcohol per 100 g of raw squash. Since 0.5% or 0.75% O_2 retained visual quality and color in our study, it would be interesting to determing acceptable levels of these volatiles in cucumbers stored in low O_2.

The fact that CO_2 and C_2H_4 production rates were nearly the same at 12.5° as at 20°C suggests that 12.5° may be at the threshold for chilling injury for parthenocarpic cucumber fruit. This observation warrants further study.

We conclude that the critical O_2 concentration for aerobic respiration of parthenocarpic cucumbers held at 12.5° or 20°C was 0.5%. This critical level did not change over the 18-day exposure. The 0.5% and 0.75% O_2 treatments for 18 days resulted in less deterioration, based on visual quality, than the air control. This was true at the time of transfer from the controlled atmosphere to air, and after 4 to 8 days in air at 20°. However, small amounts of ethanol and acetaldehyde were produced at these O_2 levels. The effect of increased CO_2 levels in association with low O_2 levels should be investigated, since this would be the situation resulting from the use of film wraps.

Since 2% O_2 maintained visual quality as effectively as lower O_2 levels without the danger of anaerobic respiration, we suggest that O_2 levels be maintained no lower than 2% in commercial CA. The margin of benefit to be derived from reduced O_2 levels does not appear to be great.

Literature Cited

1. Adam's, D.O. and S.F. Yang. 1979. Ethylene biosynthesis: Identification of 1-aminocyclopropane-1-carboxylic acid as a intermediate in the conversion of methionine to ethylene. Proc. Natl. Acad. Sci. USA 76:170–174.
2. Apeland, J. 1961. Factors affecting the keeping quality of cucumbers. Intl. Inst. Refrig. Bul. Suppl. 1:45–58.
3. Apeland, J. 1966. Factors affecting the sensitivity of cucumbers to chilling temperatures. Intl. Inst. Refrig. Bul. 46. Annexe 1:325–333.
4. Eaks, I.L. 1956. Effect of modified atmospheres on cucumbers at chilling and non-chilling temperatures. Proc. Amer. Soc. Hort. Sci. 67:473–478.
5. Eaks, I.L. 1980. Effect of chilling on respiration and volatiles of California lemon fruit. J. Amer. Soc. Hort. Sci. 105:865–869.
6. Francis, F.J., M.A. Jimenez, and E.M. Sanna. 1961. Alcohol content and atmospheric changes in prepackaged squash. Proc. Amer. Soc. Hort. Sci. 78:438–444.
7. Kanellis, A.K. 1984. Physiological responses of cucumber fruit to stage of development at harvest and to oxygen levels in the storage atmosphere. MS Thesis, Univ. of California, Davis.
8. Kanellis, A.K., L.L. Morris, and M.E. Saltveit, Jr. 1986. Effect of stage development on postharvest behavior of cucumber fruit. HortScience 21:1165–1167.
9. Li, Y., V.H. Wang, C.H. Duan, and C.Y. Mao. 1973. Preliminary observation of various oxygen partial pressures on morphological and constitutional changes in cucumber during storage. Acta Bot. Sinica 15:140–144.
10. Mencarelli, F., W.J. Lipton, and S.J. Peterson. 1983. Responses of 'zucchini' squash to storage in low-O_2 atmospheres at chilling and nonchilling temperatures. J. Amer. Soc. Hort. Sci. 108:884–890.
11. Morris, L.L. 1969. A two-stage, flow-through system for vegetables, p. 13–16. In: D.H. Dewey, R.C. Herner, and D.R. Dilley (eds.). Controlled atmospheres for the storage and transport of horticultural crops. Proc. Natl. Controlled Atmosphere Res. Conf., Mich. State Univ. Hort. Rpt. 9.
12. Papadakis, C.M. 1981. Effect of nitrogen and temperature on the performance of European cucumber, *Cucumis sativus* L., using soil-less culture. MS Thesis, Univ. of California, Davis.
13. Robinson, J.E., K.M. Browne, and W.G. Burton. 1975. Storage characteristics of some vegetable and soft fruits. Ann. Applied Biol. 81:399–408.
14. Saltveit, M.E., Jr. and R.F. McFeeters. 1980. Polygalacturonase activity and ethylene synthesis during cucumber fruit development and maturation. Plant Physiol. 66:1019–1023.
15. Satler, S.O. and K.V. Thimman. 1983. Relation between respiration and senescence in oat leaves. Plant Physiol. 72:540–546.
16. Segall, R.H., A. Dow, and P.L. Davis. 1974. Effect of waxing on decay, weight loss, and volatile pattern of cucumbers. Proc. Fla. State Hort. Soc. 87:250–251.
17. Stoll, K. 1974. Storage of vegetables in modified atmospheres (CA). Acta Hort. 38:13–22.
18. Uffelen, J.A.M. 1975. The keeping quality of cucumbers. Landbouwkundig Tijdschrift 87:295–299.
19. Wang, C.Y. 1985. Effect of low O_2 atmosphere on postharvest quality of Chinese cabbage, cucumbers, and eggplants, p. 142–149. In: S.M Blankenship (ed.). Controlled atmospheres for storage and transport of perishable agricultural commodities. Proc. Fourth Natl. Controlled Atmosphere Res. Conf. North Carolina State Univ. Hort Rpt. 126.
20. Wang, C.Y. and D.O. Adams. 1980. Ethylene production by chilled cucumbers (*Cucumis sativus* L.). Plant Physiol. 66:841–843.
21. Wills, R.B.H. and P. Wimalasiri. 1979. Short pre-storage exposures to high carbon dioxide or low oxygen atmospheres for the storage of some vegetables. HortScience 14:528–530.