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Physiology and Prediction of Fruit Tolerance to Low-oxygen Atmospheres

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Abstract. **Fruits of 'Granny Smith' and 'Yellow Newtown' apples *(Malus domestica* Borkh), '20th Century' pear *(Pyrus serotina* L.), and 'Angeleno' plum *(Prunus domestica* L.) were kept in air and in 0.25% or 0.02% O_2 at 0, 5, or 10C for 3, 7, 14, 25, or 35 days to study the effects of low-O_2 atmospheres on their postharvest physiology and quality attributes. Soluble solids content (SSC), pH, and external appearance were not significantly influenced, but resistance to CO_2 diffusion was increased by the low-O_2 treatments. Exposures to the low-O_2 atmospheres inhibited ripening, including reduction in ethylene production rate, retardation of skin color changes and flesh softening, and maintenance of titratable acidity. The most important detrimental effect of the low-O_2 treatments was development of an alcoholic off-flavor that had a logarithmic relation with ethanol content of the fruits. The ethanol content causing slight off-flavor (E_o) increased with SSC of the commodity at the ripe stage, and it could be estimated using the following formula: (Log E_o)/SSC = 0.228. Using SSC of ripe fruits and average ethanol accumulation rate per day (V_E) from each low-O_2 treatment, the tolerance limit (T_l) of fruits to low-O_2 atmospheres could be predicted as follows: $T_l = E_o / V_E = (10^{0.228SSC}) / V_E$.**

Extensive studies have been done to investigate the effects of controlled atmospheres (CA) on postharvest physiology and quality attributes of apples (Bramlage, 1977; Chen et al., 1985, 1989; Couey and Olsen, 1977; Dewey and Bourne, 1982; Johnson and Ertan, 1983; Lau, 1983; Lidster et al., 1983; Little et al., 1982; Liu and Ci, 1986; Porrits and Meheriuk, 1977). Beneficial effects of low-O_2 and/or high-CO_2 atmospheres on extending the storage life of apples include reduction in rates of respiration and ethylene production, color changes, and softening; maintenance of vitamins, sugars, acids, and flavor; and inhibition of some physiological disorders and decay. However, prolonged storage of apples in stress levels of low-O_2 and/or high-CO_2 atmospheres may cause detrimental effects, such as accumulation of ethanol and acetaldehyde, development of off-flavor, failure to ripen after removal to air, and development of low-O_2 and/or high-CO_2 injury (Fidler and North, 1971; Little and Peggie, 1987; Nichols. and Patterson, 1987; Patterson and Nichols, 1988).

Very little work has been done to study the effects of low O_2 and/or high CO_2 on Asian pears. Zagory et al. (1989) found no obvious benefits for CA in long-term storage of 'Early Gold' and 'Shinko' pears. Asian pears are also quite susceptible to low-O_2 and high-CO_2 injuries (such as discolored surface depressions or flesh browning) that were observed after 4 to 6 months of storage in 1% to 3% O_2 or in air enriched with 5% CO_2 (Richardson, 1985).

Low-O_2 and/or high-CO_2 atmospheres reduced respiration rate, softening, decay, and loss of soluble solids and extended the storage life of plums (Claypool and Allen, 1951; Couey, 1960, 1965; Maxie et al., 1958; Sive and Resnizky, 1979). Storage of peaches and nectarines in 1% O_2 + 5% CO_2 with intermittent warming reduced internal breakdown, extended storage life, and maintained levels of sugars, acids, and unsaturated fatty acids (Wang and Anderson, 1982). Treatments with low O_2 and/or high CO_2 were also reported to retard softening and color changes and reduce respiration and ethylene production rates of peaches and nectarines (Anderson, 1982; Kerbel et al., 1989; Smilanick and Fouse, 1989; Wang and Anderson, 1982). Storage of cherries in CA conditions retarded color darkening and reduction in SSC and titratable acidity (Chen et al., 1981; Patterson, 1982). CA treatments were also reported to be beneficial to the storage of apricots (Wankier et al., 1970). However, when stone fruits were stored in stress-inducing O_2 and/or CO_2 concentrations for a period longer than tolerable, detrimental effects, such as abnormal ripening, flesh browning, and large increases in ethanol and acetaldehyde contents, occurred (Kader, 1986; Smilanick and Fouse, 1989).

To meet quarantine requirements for fresh fruits and vegetables, there has been an increased interest in recent years in studying the potential of short-term exposure to O_2 levels below 1% and/or CO_2 levels above 50% for postharvest insect disinfestation (Aharoni et al., 1979; Lidster et al., 1981, 1984; Soderstrom and Brandl, 1987). CA treatments can be used for quarantine procedures only when the fresh commodities can tolerate the low-O_2 and/or high-CO_2 conditions longer than the insects. Through studying the physiological processes that limit the tolerance of fruits to low-O_2 and/or high-CO_2 atmospheres and predicting the tolerance of fruits and insects to CA conditions, the optimum CA treatments that will effectively kill the insects of concern without detrimental effects on the fruits can be selected.

In this paper, we report on the physiological processes and factors that determine the tolerance of fresh fruits to low-O_2 atmospheres and present a mathematical model that predicts the tolerance of fruits to stress-inducing low-O_2 conditions.

Materials and Methods

Materials and treatments. Fruits of 'Granny Smith' and 'Yellow Newtown' apples (both waxed), '20th Century' pear (nonwaxed), and 'Angeleno' plum (waxed) were obtained on the day of harvest from commercial shippers in San Joaquin, Monterey, Solano, and Fresno Counties of California, respectively, and transported in an air-conditioned car to our laboratory at the Univ. of California, Davis, where they were kept overnight at 0C. The experiments were initiated the next morning. Fruits with defects were sorted out and discarded, and six good fruits

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were put into a 4- or 8-liter glass jar as one replicate, with three replicates used per treatment in a flow system. In one set of experiments, the fruit samples were kept in humidified air, 0.25% O_2, or 0.02% O_2 (balance was N_2) at 0, 5, or 10C for 3 days before they were used for the determinations of respiration and ethylene production rates, internal CO_2 concentration, and resistance to gas diffusion. In another set of experiments, the fruit samples were kept in the stated atmospheres at 0, 5, or 10C for 7, 14, 25, or 35 days followed by holding in air for 7 days to allow the ethanol content to decrease at 0C (for 'Granny Smith' apple, '20th Century' pear, and 'Angeleno' plum) or 5C (for 'Yellow Newtown' apple). Then the fruits were transferred to air at 20C for 2 to several days to allow them to ripen before final quality evaluations.

Gas measurements. The O_2 and CO_2 concentrations of all gas mixtures were measured by analysis of a 10-ml gas sample using an O_2 analyzer (Model S-3All, Applied Electrochemistry, Sunnyvale, Calif.) in series with an infrared CO_2 analyzer (Model PIR-2000, Horiba Instruments, Irvine, Calif.). Respiration rate was estimated by detecting the CO_2 concentration from the headspace of the jar holding the fruits, the flow rate used, and the fresh weight of the sample. The vacuum extraction method (Saltveit, 1982) was used to determine internal CO_2 concentration and resistance to gas diffusion. The vacuum apparatus consisted of a steel vessel and a heavy-duty funnel with a plastic gas-sampling septum attached to the tip of the funnel. The funnel was placed inside the vessel, which was filled to the top with degassed water. Six fruits were placed under water in the inverted funnel. The vessel was closed and a vacuum of ≈2300 kPa applied. Preliminary experiments showed that the extracted interal gas concentrations did not significantly change over 1 to 5 min under the vacuum and, therefore, a 3-min vacuum time was used in the experiments. The vacuum was then released and the vessel opened. Samples were taken through the septum using a 1-ml syringe for the determination of internal CO_2 concentration. Resistance to CO_2 diffusion was calculated by the ratio: $[(CO_2)_{Int.} - (CO_2)_{Ext.}]/CO_2$ production rate (Trout et al., 1942). Ethylene production rate was estimated by detecting the ethylene concentration of the gas sample from each jar using a Carle gas chromatography (Model 211, Carle; EG & G Chandler Engineering, Tulsa, Okla.) with a flame ionization detector, the flow rate used, and the fresh weight of the sample.

Determinations of quality attributes. Three initial samples of six fruits each were evaluated for external appearance, skin color, flesh firmness, SSC, pH, titratable acidity, ethanol concentration, and flavor score. Similar measurements were done in the final quality evaluations. External appearance was scored using a subjective scale of 1 to 5, where 5 = excellent, 4 = good, 3 = fair, 2 = slight defects, 1 = severe defects. Skin color was measured with a Gardner XL-23 Tristimulus Calorimeter (Gardner Laboratory, Bethesda, Md.) using the color difference meter (CDM) "a" value in which a more negative number means increasing greenness, while a larger positive number indicates increasing redness. Flesh firmness was measured as penetration force with a Univ. of California fruit firmness tester, using an 11-mm plunger tip for apples and an 8-mm tip for pears and plums. Fruit juice was extracted from four flesh sections of each fruit with a hand-press juicer. SSC was measured with an Abbé refractometer (VWR Scientific, San Francisco), and pH and titratable acidity were measured using an automatic titrator with a PHM85 Precision pH meter (Radiometer, Copenhagen, Denmark), an AUB80 autoburette (Radiometer), a PRS12 Alpha printer (Radiometer), and a SAC80 sample changer (Radiometer).

Determination of ethanol content. Frozen fruit juice was thawed and a 2-ml sample was diluted with 18 ml deionized water. The diluted sample was centrifuged at 25,000× *g* for 20 min at 0C, and then ≈2 ml supernatant was filtered through a 0.45-µm microfilter in a 0C room. The filtrate was used for the determination of ethanol concentration using a HP5890A gas chromatography (Hewlett Packard, Palo Alto, Calif.) with a flame ionization detector (at 250C) and a glass column (2 mm × 1.8 m) containing 5% Carbowax on 60/80 Carpoback (at 85C).

Estimation of low-O_2 injury. Low-O_2 injury was estimated by a pretransformed scale of 1 to 5 according to the percentage of brown area in the core and/or flesh of a longitudinal fruit section: 1 = no injury; 2 = slight injury, up to 15% browning; 3 = moderate injury, 16% to 50% browning; 4 = severe injury, 51% to 85% browning; 5 = extreme injury, 86% to 100% browning.

Estimation of flavor score. Flavor score was estimated by tasting using a subjective scale ranging from 1 to 7, where 7 = excellent, 6 = good, 5 = fair, 4 = slight off-flavor, 3 = moderate off-flavor, 2 = severe off-flavor, 1 = extreme off-flavor. Three fruits were tested in each treatment, with one section tasted in each fruit. The evaluation was done in the Postharvest Laboratory, Pomology Dept. The judge rinsed ad-lib with drinking water between tastings to clear his mouth of residual samples and to avoid an influence by prior fruits. Previous studies with strawberries and oranges used an untrained taste panel of ≈100 people and a trained taste panel of four people to conduct sensory evaluations (Ke and Kader, 1990; Ke et al., 1991). D.K. participated in such studies and gained substantial experience in sensory evaluation; due to the limited resources available, only he estimated flavor in this research. This approach reduced the variation in the ability to detect off-flavor, but potentially limited the reproducibility of the results.

Statistical analysis. Data were treated for multiple comparisons by analysis of variance with least significant difference (LSD) between means determined at $P = 0.05$. A computer curve fit program (Cricket Graph, Macintosh SE, Apple Computer, Cupertino, Calif.) was used to analyze the regression and correlation between ethanol content and flavor score.

Results

Storage of 'Granny Smith' apples in 0.25% or 0.02% O_2 at 0 or 10C generally slightly reduced respiration and ethylene production rates but increased internal CO_2 concentration and resistance to CO_2 diffusion (Table 1). Respiration and ethylene production rates and internal CO_2 concentration were higher at 10C than those at 0C, but resistance to CO_2 diffusion was lower at the higher temperature. The low-O_2 treatments did not significantly influence skin color, flesh firmness, and external appearance of 'Granny Smith' apples during the experimental period but slightly retarded the reduction in titratable acidity. As acidity decreased, pH slightly increased in all treatments (Table 1). No visible injury or decay was observed in any of the treatments during the experiment. The low-O_2 treatments did not influence SSC but greatly increased ethanol content (EC) and reduced the flavor score of 'Granny Smith' apples (Fig. 1). The low-O_2 effects were more pronounced at 10C than those at 0C. The effects of 0.029002 were greater than those of 0.25% O_2. Flavor scores of 4 or below indicate that alcoholic off-flavor could be detected in low-O_2-treated fruits.

Exposure of 'Yellow Newtown' apples to 0.25% or 0.02% O_2 at 5 or 10C increased CO_2 production rate, internal CO_2 concentration, and resistance to CO_2 diffusion (Table 2). Res-

Table 1. Effects of O_2 level and temperature on respiration and ethylene production rates, internal CO_2 concentration, resistance to CO_2 diffusion (r_{CO_2}), and quality attributes of 'Granny Smith' apples. Skin color, flesh firmness, external appearance, titratable acidity, and pH were measured after specified days under treatment followed by holding in air at 0C for 7 days and then at 20C for 7 days.

Observation	Days under treatment	0C Air	0C 0.25% O_2	0C 0.02% O_2	10C Air	10C 0.25% O_2	10C 0.02% O_2	LSD at 5%
CO_2 production rate ($ml \cdot h^{-1} \cdot kg^{-1}$)	3	1.6	1.2	1.4	4.8	4.4	4.2	0.3
C_2H_4 production rate ($\mu l \cdot h^{-1} \cdot kg^{-1}$)	3	0.1	ND[z]	ND	0.4	ND	ND	0.1
Internal CO_2 (%)	3	2.0	2.0	2.4	3.8	5.1	4.8	0.8
r_{CO_2} [$\%(ml \cdot h^{-1} \cdot kg^{-1})^{-1}$]	3	1.2	1.6	1.7	0.8	1.1	1.1	0.3
Skin color (CDM[y] "a" value)	7	−14	−14	−14	−14	−14	−14	
	35	−14	−13	−13	−12	−13	−14	1
Flesh firmness (N)	7	69	73	80	72	73	76	
	35	76	72	68	65	71	70	6
External appearance[x]	7	5.0	5.0	4.9	4.9	4.8	4.9	
	35	4.5	4.8	4.4	5.0	5.0	4.8	0.5
Titratable acidity (%)	7	0.57	0.57	0.65	0.52	0.56	0.58	
	35	0.46	0.55	0.51	0.41	0.54	0.52	0.03
pH	7	3.5	3.5	3.5	3.5	3.5	3.5	
	35	3.6	3.6	3.6	3.6	3.6	3.6	0.1

[z]ND = Not detected.
[y]CDM = color difference meter; a more negative "a" value indicates a greener appearance.
[x]Estimated using a scale of 1 to 5, where 5 = excellent, 4 = good, 3 = fair, 2 = slight defects, and 1 = severe defects.

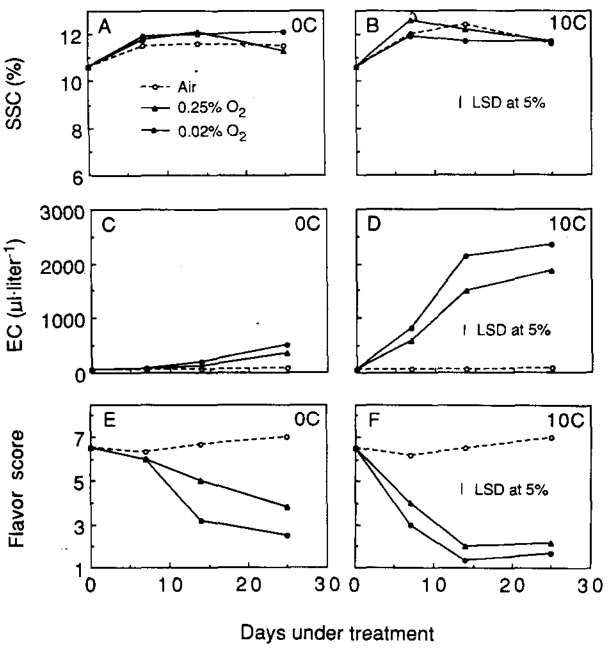

Fig. 1. Effects of O_2 level and temperature on the soluble solids content (SSC), ethanol content (EC), and flavor score of 'Granny Smith' apples kept in air, 0.25% O_2, or 0.02% O_2 at 0 or 10C for 7, 14, or 25 days followed by holding in air at 0C for 7 days and then at 20C for 7 days. Flavor score was estimated using a scale of 1 to 7, where 7 = excellent, 6 = good, 5 = fair, 4 = slight off-flavor, 3 = moderate off-flavor, 1 = extreme off-flavor.

piration rate and internal CO_2 concentration were higher in fruits at 10C than in those at 5C, while resistance to CO_2 diffusion was lower at 10C. No ethylene production was detected in 'Yellow Newtown' apples from any treatment in this experiment. The low-O_2 treatments slightly retarded skin yellowing, flesh softening, and decreases in titratable acidity of the fruits, but external appearance and pH were not significantly influenced (Table 2). No visible injury or decay was observed during the experiment. Treatments with 0.25% or 0.02% O_2 did not significantly affect SSC but greatly increased the ethanol content and reduced the flavor score of 'Yellow Newtown' apples (Fig. 2). The effects at 10C were greater than those at 5C.

Storage of '20th Century' pears in 0.25% or 0.02% O_2 at 0 or 5C generally decreased respiration rate but decreased internal CO_2 concentration consistently only at 0C and increased resistance to CO_2 diffusion (Table 3). No ethylene production was detected in '20th Century' pears from any treatment. Skin color, flesh firmness, and external appearance of the fruits were not significantly influenced by the low-O_2 treatments. Exposure of '20th Century' pears to 0.25% or 0.02% O_2 at 0 or 5C for 7 days did not cause any visible injury; but longer exposures (14 to 35 days) resulted in core browning. This result indicates that '20th Century' pears were sensitive to low-O_2 injury. The low-O_2 atmospheres did not have consistent effects on titratable acidity and pH of the fruits (Table 3). No decay was observed during the experiment. The low-O_2 treatments at 0 or 5C did not significantly influence SSC but greatly increased ethanol content and decreased the flavor score of '20th Century' pears (Fig. 3). Generally, the effects of low-O_2 treatments were more pronounced at 5C than at 0C; and the effects of 0.02% O_2 were greater than those of 0.25% O_2.

Exposure to 0.25% or 0.02% O_2 at 5 or 10C reduced respiration and ethylene production rates but increased resistance to CO_2 diffusion of 'Angeleno' plums (Table 4); internal CO_2 concentration was not significantly influenced by the low-O_2 treatments. Respiration and ethylene production rates and internal CO_2 concentration were higher at 10C than at 5C, while resistance to CO_2 diffusion was lower at the higher temperature. As the plums ripened, skin color changed from red to dark red (lower "a" values) and flesh firmness decreased. The low-O_2

Table 2. Effects of O_2 level and temperature on respiration rate, internal CO_2 concentration, resistance to CO_2 diffusion (r_{CO_2}), and quality attributes of 'Yellow Newtown' apples. Skin color, flesh firmness, external appearance, titratable acidity, and pH were measured after specified days under treatment followed by holding in air at 5C for 7 days and then at 20C for 14 days.

Observation	Days under treatment	5C Air	5C 0.25% O_2	5C 0.02% O_2	10C Air	10C 0.25% O_2	10C 0.02% O_2	LSD at 5%
CO_2 production rate ($ml \cdot h^{-1} \cdot kg^{-1}$)	3	3.1	3.7	3.6	5.2	6.2	6.2	0.2
Internal CO_2 (%)	3	5.3	7.6	8.0	6.1	9.6	11.4	1.0
r_{CO_2} [$\% \cdot (ml \cdot h^{-1} \cdot kg^{-1})^{-1}$]	3	1.7	2.0	2.2	1.1	1.5	1.8	0.2
Skin color (CDM[z] "a" value)	7	−14	−13	−13	−13	−13	−13	
	35	−11	−12	−13	−9	−12	−11	1
Flesh firmness (N)	7	99	102	100	104	99	103	
	35	73	81	79	70	92	89	9
External appearance[y]	7	4.6	4.2	4.1	4.2	4.6	4.5	
	35	4.2	4.4	4.3	4.3	4.6	4.0	0.8
Titratable acidity (%)	7	0.56	0.52	0.54	0.54	0.52	0.55	
	35	0.39	0.42	0.42	0.36	0.42	0.38	0.03
pH	7	3.6	3.6	3.6	3.6	3.6	3.6	
	35	3.7	3.7	3.7	3.8	3.7	3.8	0.1

[z]CDM = color difference meter; a more negative "a" value indicates a greener appearance.
[y]Estimated using a scale of 1 to 5, where 5 = excellent, 4 = good, 3 = fair, 2 = slight defects, and 1 = severe defects.

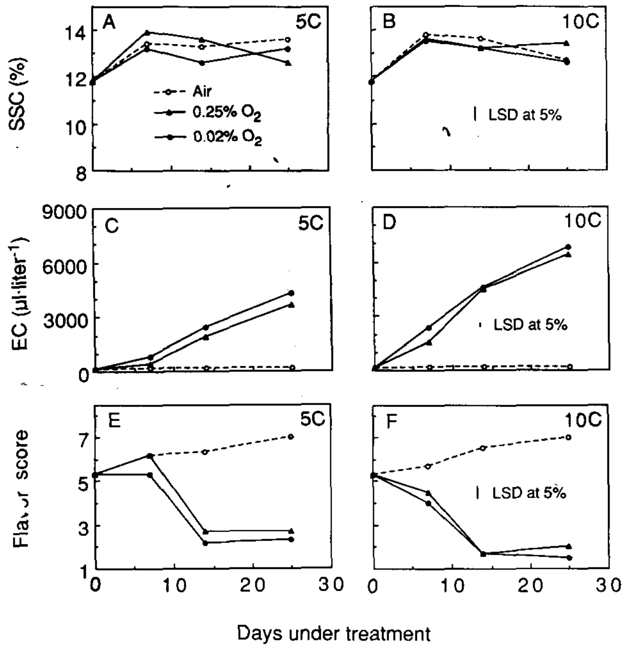

Fig. 2. Effects of O_2 level and temperature on soluble solids content (SSC), ethanol content (EC), and flavor score of 'Yellow Newtown' apples kept in air, 0.25% O_2, or 0.02% O_2 at 5 or 10C for 7, 14, or 25 days followed by holding in air at 5C for 7 days and then at 20C for 14 days. Flavor score was estimated using a scale of 1 to 7 (see legend of Fig. 1 for details).

treatments retarded skin color changes at both 5 and 10C, but the retardation effect on flesh softening was noted only at 10C (Table 4). External appearance, titratable acidity, and pH of the plums were not consistently influenced by the low-O_2 treatments. No visual low-O_2 injury was observed and decay was negligible during the experiment. The low-O_2 treatments did not significantly influence SSC but dramatically increased ethanol content and decreased flavor score of the plums (Fig. 4). The effects were more pronounced at 10C or in 0.02% O_2 than at 5C or in 0.25% O_2.

Discussion

The major beneficial effects of short-term exposure to 0.25% or 0.02% O_2 were the inhibition or delay of ripening processes, including reduction in respiration and/or ethylene production rates; retardation of skin color changes and flesh softening; and maintenance of acidity in the fruits. External appearance, pH, and SSC were not significantly influenced by the low-O_2 treatments. For all the fruits tested thus far, including apples, Asian pears, 'Bartlett' pears (Ke et al., 1990), nectarines (Kerbel et al., 1989), peaches, plums, strawberries (Ke et al., 1991), and 'Valencia' oranges (Ke and Kader, 1990), '20th Century' pear is the only one in which visible low-O_2 injury was observed during short-term exposure to low O_2 levels. However, alcoholic off-flavor was detected in the low-O_2-treated fruits of all the commodities after various durations during the experiments. Therefore, the occurrence of an alcoholic off-flavor is the most important detrimental effect with short-term low-O_2 treatments. A low-O_2-treated fruit may look good but taste bad. Thus, the tolerance of fresh fruits to short-term low-O_2 atmospheres is largely limited by the occurrence of off-flavor.

After the flavor score decreased to 4 (slight off-flavor) or a lower value, the flavor was not acceptable. Therefore, the number of days to reach a flavor score of 4 was used as the tolerance limit of fruits to low-O_2 atmospheres from the experiments (T_l^e). The T_l^e values for all treatments of the four commodities were obtained from Figs. 1-4 by linear extrapolation and are presented in Table 5. For each commodity, T_l^e was higher at the higher O_2 concentration and lower at the higher temperature.

The accumulation of ethanol was accompanied by a decrease in the flavor score of the four commodities tested in this study (Figs. 1-4). A computer curve fit program (including linear, polynomial, logarithmic, and exponential regressions) was used to analzye the correlation between ethanol content and flavor score; the best curve fit was obtained by the logarithmic regression (Fig. 5). The correlation coefficients *(r)* were 0.93 to 0.94 for the four commodities. When the ethanol content data were transformed to a Log scale, straight lines were obtained from the correlation analysis. Therefore, ethanol content and flavor score, as related to ethanol content, have a logarithmic relation,

Table 3. Effects of O_2 level and temperature on respiration rate, internal CO_2 concentration, resistance to CO_2 diffusion (r_{CO_2}), and quality attributes of '20th Century' pears. Skin color, flesh firmness, external appearance, titratable acidity, and pH were measured after specified days under treatment followed by holding in air at 0C for 7 days and then at 20C for 3 days.

Observation	Days under treatment	0C Air	0C 0.25% O_2	0C 0.02% O_2	5C Air	5C 0.25% O_2	5C 0.02% O_2	LSD at 5%
CO_2 production rate ($ml \cdot h^{-1} \cdot kg^{-1}$)	3	1.8	0.8	1.0	2.7	1.6	1.6	0.2
Internal CO_2 (%)	3	0.35	0.23	0.26	0.48	0.39	0.46	0.12
r_{CO_2} [$\% \cdot (ml \cdot h^{-1} \cdot kg^{-1})^{-1}$]	3	0.14	0.23	0.21	0.14	0.21	0.24	0.05
Skin color (CDM[z] "a" value)	7	−2.1	−2.3	−2.6	−2.4	−1.2	−1.8	
	35	−0.7	−2.4	−1.0	−0.2	−1.9	−2.6	2.2
Flesh firmness (N)	7	32	37	37	34	32	34	
	35	32	30	31	31	32	31	4
Internal injury severity[y]	7	1.0	1.2	1.0	1.0	1.2	1.0	
	14	1.0	1.8	1.9	1.2	1.5	2.0	0.7
	35	1.0	3.1	3.2	1.0	1.8	2.3	
External appearance[x]	7	5.0	5.0	5.0	5.0	5.0	5.0	
	35	4.8	4.9	4.3	4.9	4.8	4.9	0.6
Titratable acidity (%)	7	0.22	0.25	0.25	0.24	0.21	0.21	
	35	0.16	0.14	0.14	0.16	0.18	0.17	0.03
pH	7	4.4	4.3	4.4	4.3	4.4	4.4	
	35	4.6	4.6	4.7	4.5	4.5	4.6	0.1

[z]CDM = color difference meter; a more negative "a" value indicates a greener appearance.
[y]Estimated using a pretransformed scale of 1 to 5 according to the percentage of brown area in the longitudinal section: 1 = no injury; 2 = slight injury, 1% to 15% brown area; 3 = moderate injury, 16% to 50% brown area; 4 = severe injury, 51% to 85% brown area; 5 = extreme injury, 86% to 100% brown area.
[x]Estimated using a scale of 1 to 5, where 5 = excellent, 4 = good, 3 = fair, 2 = slight defects, and 1 = severe defects.

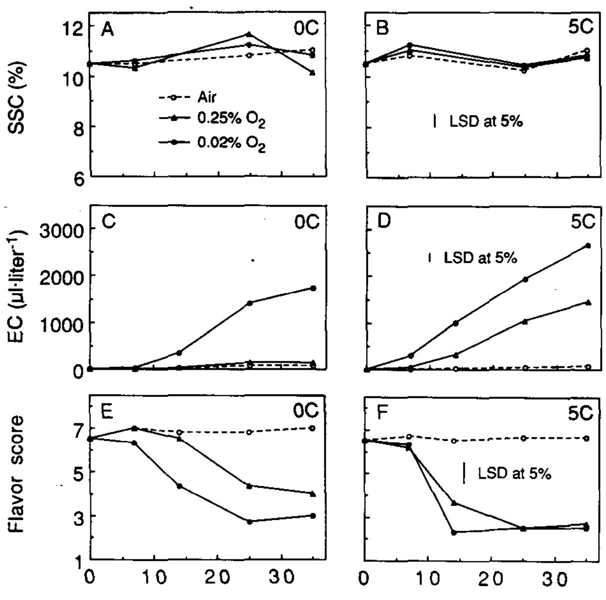

Fig. 3. Effects of O_2 level and temperature on soluble solids content (SSC), ethanol content (EC), and flavor score of '20th Century' pears kept in air, 0.25% O_2, or 0.02% O_2 at 0 or 5C for 7, 14, 25, or 35 days followed by holding in air at 0C for 7 days and then at 20C for 3 days. Flavor score was estimated using a scale of 1 to 7 (see legend of Fig. 1 for details).

While it is well-known that the accumulation of ethanol may cause off-flavor in fruits and vegetables, the ethanol concentrations that cause alcoholic off-flavors varied greatly among commodities. For example, an ethanol content >200 $\mu l \cdot liter^{-1}$ caused a slight off-flavor (a flavor score of 4) in '20th Century' pears (Fig. 3), but it took >1000 and 2000 µl ethanol/liter to cause a slight off-flavor in 'Yellow Newtown' apples (Fig. 2) and 'Angeleno' plums (Fig. 4), respectively.

The variation in the threshold ethanol concentration causing off-flavor among commodities may be due to the interaction between ethanol and other compounds (such as sugars and organic acids). Using the data from Figs. 1–4 to analyze the relationship between SSC and the ethanol content causing slight off-flavor (E_o) in apples, 'Asian' pears, and plums, we found higher SSC to generally require higher ethanol concentrations to cause slight off-flavor (Fig. 6A). Since the relationship between ethanol content and flavor score is logarithmic (Fig. 5), the E_o data were transformed to a Log scale. The ratio of (Log E_o)/SSC is very constant (with a standard deviation of 0.019) for all low-O_2 treatments of the four commodities (Fig. 6B):

$$(\text{Log } E_o)/\text{SSC} = 0.228 \quad [1]$$

Thus, SSC plays a very important role in determining the ethanol level that causes off-flavor. SSC probably provides a background flavor for the interaction with ethanol; the higher the SSC, the higher the ethanol content required to cause off-flavor in the fruits. It is still not clear if this effect is due to a specific component of SSC, since SSC represents all the compounds from the fruits that are soluble in water.

From Eq. [1], the ethanol content causing slight off-flavor could be estimated from SSC of ripe fruits for all the commodities tested as follows:

$$E_o = 10^{0.228\ \text{SSC}} \quad [2]$$

From the estimated ethanol content causing slight off-flavor in Eq. [2] and the calculated average ethanol accumulation rate per day (V_E) of fruits under a low-O_2 treatment, the tolerance

Table 4. Effects of O_2 level and temperature on respiration and ethylene production rates, internal CO_2 concentration, resistance to CO_2 diffusion (r_{CO_2}), and quality attributes of 'Angeleno' plum. Skin color, flesh firmness, external appearance, titratable acidity, and pH were measured after specified days under treatment followed by holding in air at 0C for 7 days and then at 20C for 2 days.

Observation	Days under treatment	5C Air	5C 0.25% O_2	5C 0.02% O_2	10C Air	10C 0.25% O_2	10C 0.02% O_2	LSD at 5%
CO_2 production rate ($ml \cdot h^{-1} \cdot kg^{-1}$)	3	4.0	2.2	3.2	7.5	3.7	4.5	0.5
C_2H_4 production rate ($\mu l \cdot h^{-1} \cdot kg^{-1}$)	3	6.0	0.6	0.8	10.0	2.2	1.6	0.8
Internal CO_2 (%)	3	1.5	1.4	1.5	1.9	1.6	1.9	0.3
r_{CO_2} [$\% \cdot (ml \cdot h^{-1} \cdot kg^{-1})^{-1}$]	3	0.34	0.63	0.46	0.24	0.40	0.41	0.08
Skin color (CDM[z] "a" value)	25	10	14	15	5	12	13	
	35	6	14	12	3	14	14	4
Flesh firmness (N)	25	33	34	35	5	37	32	
	35	31	28	33	7	28	32	11
External appearance[y]	25	4.3	4.0	4.8	4.9	4.8	4.8	
	35	4.5	5.0	4.8	4.2	4.6	4.5	0.9
Titratable acidity (%)	25	0.41	0.40	0.43	0.32	0.36	0.34	
	35	0.37	0.35	0.40	0.30	0.34	0.32	0.03
pH	25	4.1	4.0	4.0	4.4	4.1	4.1	
	35	4.1	4.2	4.2	4.0	4.1	4.3	0.1

[z]CDM = color difference meter; a darker red resulted in a lower "a" value.
[y]Estimated using a scale of 1 to 5, where 5 = excellent, 4 = good, 3 = fair, 2 = slight defects, and 1 = severe defects.

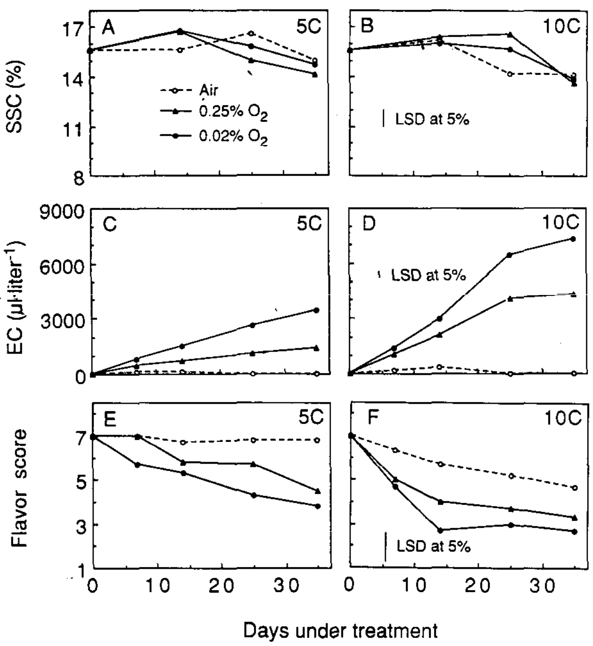

Fig. 4. Effects of O_2 level and temperature on soluble solids content (SSC), ethanol content (EC), and flavor score of 'Angeleno' plums kept in air, 0.25% O_2, or 0.02% O_2 at 5 or 10C for 7, 14, 25, or 35 days followed by holding in air at 0C for 7 days and then at 20C for 2 days. Flavor score was estimated using a scale of 1 to 7 (see legend of Fig. 1 for details).

limit (T_I) of fruits to low-O_2 atmospheres can be predicted by:

$$T_I = E_O / V_E = (10^{0.228\ SSC}) / V_E, \qquad [3]$$

where T_I = tolerance limit predicted, i.e., the number of days to cause slight off-flavor; E_O = the ethanol content ($\mu l \cdot liter^{-1}$) of the fruit that causes slight off-flavor; V_E = the average ethanol accumulation rate ($\mu l \cdot liter^{-1} \cdot day^{-1}$) of the fruit under the low-O_2 treatment; and SSC = soluble solids content (%) of the fruit when ripe.

According to Eq. [3], two primary factors determine the tolerance of fruits to low-O_2 atmospheres: SSC of ripe fruits and average ethanol accumulation rate of the fruits under the low-O_2 treatment. Other factors may affect fruit tolerance to low O_2 through influencing these two primary factors. SSC of harvested ripe fruits is not greatly influenced by the O_2 level or temperature used during these short storage treatments (Figs. 1-4). Cultural conditions before harvest and fruit maturity at harvest may influence SSC of the fruit when ripe. However, ethanol accumulation rate can be greatly influenced by several factors including the following:

1) Respiration rate of the fruits under a low-O_2 treatment: V_E generally increases as respiration rate increases since, under the low O_2 levels (0.25% to 0.02%), respiration is largely anaerobic, which produces much ethanol. Respiration rate is commodity-specific and may be influenced by the O_2 level and temperature used.

2) O_2 level used: ethanol accumulates at a higher rate at 0.02% than at 0.25% O_2 since anaerobic respiration rate is higher at the lower concentration.

3) Temperature used: as temperature increases, O_2 consumption rate and respiration rate increase, which cause an increase in V_E.

4) Resistance to gas diffusion: this is largely commodity-specific, but it maybe influenced by postharvest treatment, such as waxing, which causes a great increase in resistance to gas diffusion. Under the same O_2 level and temperature, a commodity with a higher resistance to gas diffusion will limit the efficiency of O_2 use and the diffusion of ethanol out of the fruit, causing a higher V_E.

Equation [3] was used as a model to predict the tolerance of fruits to low-O_2 atmospheres. Using the average SSC of the ripe fruits, E_O was estimated by Eq. [2]. V_E values for all treatments

Table 5. Comparison of tolerance limit from experiments (T_l^e) and predicted tolerance limit (T_l) of 'Granny Smith' and 'Yellow Newtown' apples, '20th Century' pears, and 'Angeleno' plums to 0.25% or 0.02% O_2 at several temperatures. T_l is predicted by the model $T_l = E_o/V_E$ or $T_l = (10^{0.228\ SSC})/V_E$, where E_o is the predicted ethanol content causing slight off-flavor by using the average soluble solids content (SSC) of the commodity at the ripe stage and V_E is the average ethanol accumulation rate of the fruits in the low-O_2 treatment.

Commodity	Temp (°C)	O_2 (%)	SSC (%)	E_o (μl·liter⁻¹)	V_E (μl·liter⁻¹·day⁻¹)	T_l^e (days)	T_l (days)	$T_l^e - T_l$ (days)
'Granny Smith' apple	0	0.25	11.9	517	19	23.4	27.2	−3.8
	0	0.02	11.9	517	29	11.9	17.8	−5.9
	10	0.25	11.9	517	74	7.0	7.0	0.0
	10	0.02	11.9	517	94	5.0	5.5	−0.5
'Yellow Newtown' apple	5	0.25	13.4	1136	147	11.3	7.7	3.6
	5	0.02	13.4	1136	173	9.9	6.6	3.3
	10	0.25	13.4	1136	256	8.2	4.4	3.8
	10	0.02	13.4	1136	272	7.0	4.2	2.8
'20th Century' pear	0	0.25	10.7	275	8	35.0	34.4	0.6
	0	0.02	10.7	275	24	16.2	11.7	4.5
	5	0.25	10.7	275	22	13.1	12.4	0.7
	5	0.02	10.7	275	40	11.0	6.8	4.2
'Angeleno' plum	5	0.25	15.3	3079	68	40.6	45.3	−4.7
	5	0.02	15.3	3079	96	31.6	32.1	−0.5
	10	0.25	15.3	3079	164	14.0	18.8	−4.8
	10	0.02	15.3	3079	259	9.4	11.9	−2.5

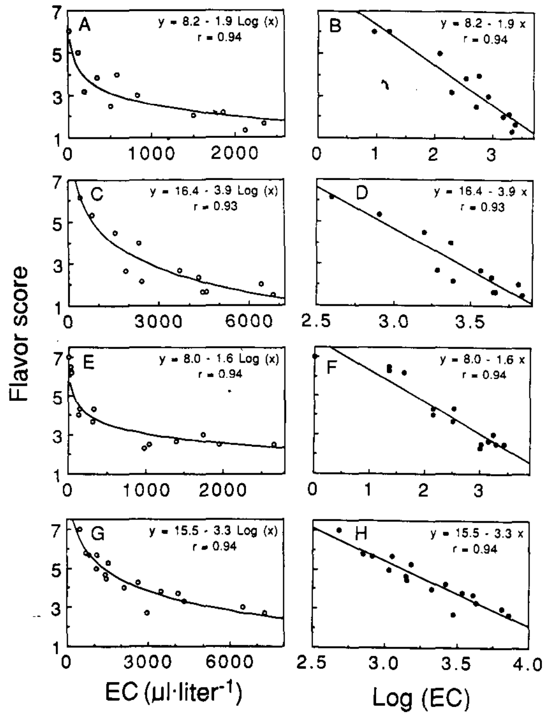

Fig. 5. Correlation between ethanol content (EC) and flavor score of low-O_2-treated fruits of 'Granny Smith' (**A** and **B**) and 'Yellow Newtown' apples (**C** and **D**), '20th Century' pears (**E** and **F**), and 'Angeleno' plums (**G** and **H**). Flavor score was estimated using a scale of 1 to 7 (see legend of Fig. 1 for details).

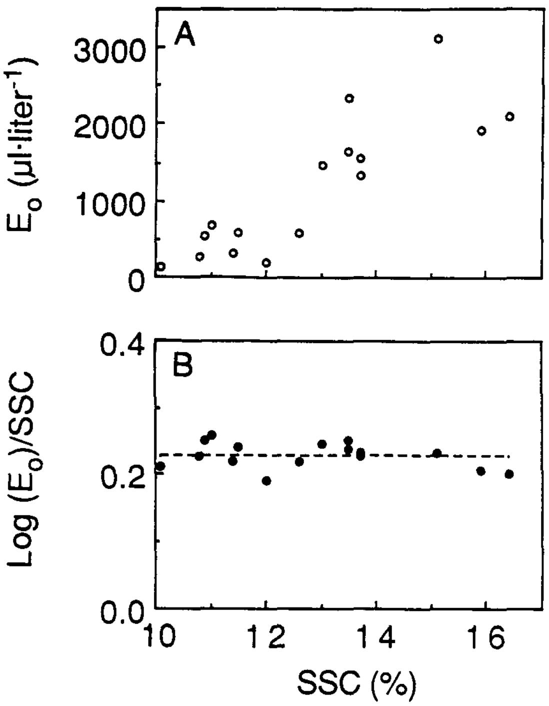

Fig. 6. Relationship between soluble solids content (SSC) and ethanol content causing slight off-flavor (E_O) in low-O_2-treated 'Granny Smith' and 'Yellow Newtown' apples, '20th Century' pears, and 'Angeleno' plums.

of the four commodities during the first 1 to 2 weeks were calculated from Figs. 1-4. The tolerance limit (T_l^e) obtained from Figs. 1–4 of the experiments and the tolerance limit predicted by Eq. [3] (T_l) are quite close for most treatments (Table 5); the average deviation equals 2.9 days. Our later study on 'Bing' cherries (unpublished data) also indicated that Eq. [3] could reasonably predict fruit tolerance to low-O_2 atmospheres with a deviation of 1 to 2 days between the predicted and observed tolerance limits. For the practical application of the model to postharvest insect disinfestation, fruit tolerance to low-O_2 atmospheres appears to be predictable; with additional information from entomologists, optimum combinations of O_2 level and temperature could be selected so that the insects of concern could be killed without detrimental effects on the commodity.

Literature Cited

Aharoni, Y., P. Hartsell, J.K. Stewart, and D.K. Young. 1979. Control of western flower thrips on harvested strawberries with acetaldehyde in air, 50% carbon dioxide or 1% oxygen. J. Econ. Entomol. 72:819-822.

Anderson, R.E. 1982. Long-term storage of peaches and nectarines intermittently warmed during controlled-atmosphere storage. J. Amer. Soc. Hort. Sci. 107:214-216.

Bramlage, W.J. 1977. Pretreatment of CA 'McIntosh' with high CO_2, p. 182-188. In: D.H. Dewey (ed.). Controlled atmospheres for the storage and transport of perishable agricultural commodities. Hort. Rpt. 28, Michigan State Univ., East Lansing.

Chen, P.M., W.M. Mellenthin, S.B. Kelly, and T.J. Facteau. 1981. Effects of low oxygen and temperature on quality retention of 'Bing' cherries during prolonged storage. J. Amer. Soc. Hort. Sci. 106:533-535.

Chen, P.M., K.L. Olsen, and M. Meheriuk. 1985. Effect of low-oxygen atmosphere on storage scald and quality preservation of 'Delicious' apples. J. Amer. Soc. Hort. Sci. 110:16-20.

Chen, P.M., D.M. Varga, E.A. Mielke, and S.R. Drake. 1989. Post-storage behavior of apple fruit after low oxygen storage as influenced by temperature during storage and in transit. J. Food Sci. 54:993-996.

Claypool, L.L. and F.W. Allen. 1951. The influence of temperature and oxygen level on the respiration and ripening of Wickson plums. Hilgardia 21:129-160.

Couey, H.M. 1960. Effects of temperature and modified atmosphere on the storage life, ripening behavior, and dessert quality of Eldorado plums. Proc. Amer. Soc. Hort. Sci. 75:207-215.

Couey, H.M. 1965. Modified atmosphere storage of Nubiana plums. Proc. Amer. Soc. Hort. Sci. 86:166-168.

Couey, M. and K. Olsen. 1977. Commercial use of prestorage carbon dioxide treatment to retain quality in Golden Delicious apples, p. 165-169. In: D.H. Dewey (ed.). Controlled atmospheres for the storage and transport of perishable agricultural commodities. Hort. Rpt. 28, Michigan State Univ., East Lansing.

Dewey, D.H. and M.L. Bourne. 1982. Low oxygen CA storage of McIntosh apples, p. 101-107. In: D.G. Richardson and M. Meheriuk (eds.). Controlled atmospheres for storage and transport of perishable agricultural commodities. Timber Press, Beaverton, Ore.

Fidler, J.C. and C.J. North. 1971. The effect of periods of anaerobiosis on the storage of apples. J. Hort. Sci. 46:213-221.

Johnson, D.S. and U. Ertan. 1983. Interaction of temperature and oxygen level on the respiration rate and storage quality of Idared apples. J. Hort. Sci. 58:527-533.

Kader, A.A. 1986. Biochemical and physiological basis for effects of controlled and modified atmospheres on fruits and vegetables. Food Technol. 40(5):99-100, 102-104.

Ke, D. and A.A. Kader. 1990. Tolerance of 'Valencia' oranges to controlled atmospheres as determined by physiological responses and quality attributes. J. Amer. Soc. Hort. Sci. 115:779-783.

Ke, D., L. Goldstein, M. O'Mahony, and A.A. Kader. 1991. Effects of short-term exposure to low O_2 and high CO_2 atmospheres on quality attributes of strawberries. J. Food Sci. 56:50-54.

Ke, D., H. van Gorsel, and A.A. Kader. 1990. Physiological and quality responses of 'Bartlett' pears to reduced-O_2 and enhanced-CO_2 atmospheres and storage temperature. J. Amer. Soc. Hort. Sci. 115:435-439.

Kerbel, E., D. Ke, and A.A. Kader. 1989. Tolerance of 'Fantasia' nectarines to low O_2 and high CO_2 atmospheres, p. 196-202. In: D. Reid (ed.). Proc. Intl. Inst. Refrigeration Conf. Techn. Innovations Freezing Refrigeration Fruit Veg., Univ. of California, Davis.

Lau, O.L. 1983. Effects of storage procedures and low oxygen and carbon dioxide atmospheres on storage quality of 'Spartan' apples. J. Amer. Soc. Hort. Sci. 108:953-957.

Lidster, P.D., H.J. Lightfoot, and K.B. McRae. 1983. Fruit quality and respiration of 'McIntosh' apples in response to ethylene, very low oxygen and carbon dioxide storage atmospheres. Scientia Hort. 20:71-83.

Lidster, P.D., K.H. Sanford, and K.B. McRae. 1981. Effects of modified atmosphere storage on overwintering populations of the apple rust mite and European red mite eggs. HortScience 16:328-329.

Lidster, P.D., K.H. Sanford, and K.B. McRae. 1984. Effects of temperature and modified atmosphere on survival of overwintering populations of European red mite eggs on stored 'McIntosh' apple. HortScience 19:257-258.

Little, C.R., J.D. Faragher, and H.J. Taylor. 1982. Effects of initial oxygen stress treatments in low oxygen modified atmosphere storage of 'Granny Smith' apples. J. Amer. Soc. Hort. Sci. 107:320-323.

Little, C.R. and I.D. Peggie. 1987. Storage injury of pome fruit caused by stress levels of oxygen, carbon dioxide, temperature, and ethylene. HortScience 22:783-790.

Liu, F.W. and L.J. Ci. 1986. Responses of daminozide-sprayed 'McIntosh' apples to various concentrations of oxygen and ethylene in simulated CA storage. J. Amer. Soc. Hort. Sci. 111:400-403.

Maxie, E.C., B.J. Robinson, and P.B. Catlin. 1958. Effect of various oxygen concentrations on the respiration of Wickson plum fruit and fruit tissues. Proc. Amer. Soc. Hort. Sci. 71:145-156.

Nichols, W.C. and M.E. Patterson. 1987. Ethanol accumulation and poststorage quality of 'Delicious' apples during short-term, low-O_2, CA storage. HortScience 22:89-92.

Patterson, M.E. 1982. CA storage of cherries, p. 149-154. In: D.G. Richardson and M. Meheriuk (eds.). Controlled atmospheres for storage and transport of perishable agricultural commodities. Timber Press, Beaverton, Ore.

Patterson, M.E. and W.C. Nichols. 1988. Metabolic response of 'Delicious' apples to carbon dioxide in anoxic and low-oxygen environments. HortScience 23:866-868.

Porritt, S.W. and M.M. Meheriuk. 1977. Effect of CO_2 treatment on storage behavior of apples and pears, p. 170-174. In: D.H. Dewey (ed.). Controlled atmospheres for the storage and transport of perishable agricultural commodities. Hort. Rpt. 28, Michigan State Univ., East Lansing.

Richardson, D.G. 1985. CA recommendations for pears (including Asian pears), p. 422-444. In: S.M. Blankenship (ed.). Controlled atmospheres for storage and transport of perishable agricultural commodities. Hort. Rpt. 128, North Carolina State Univ., Raleigh.

Saltveit, M.E. 1982. Procedure for extracting and analyzing internal gas samples from plant tissue by gas chromatography. HortScience 17:878-881.

Sive, A. and D. Resnizky. 1979. Extension of the storage life of 'Red Rosa' plums with controlled atmosphere storage. Bul. Intl. Inst. Refrig. 59:1148.

Smilanick, J.L. and D.C. Fouse. 1989. Quality of nectarines stored in insecticidal low-O_2 atmospheres at 5 and 15C. J. Amer. Soc. Hort. Sci. 114:431-436.

Soderstrom, E.L. and D.G. Brandl. 1987. Controlled atmospheres for postharvest control of codling moth on fresh tree fruits. 1986 Res. Rpt., California Tree Fruit Agreement, Sacramento.

Trout, S.A., E.G. Hall, R.N. Robertson, F.M.V. Hackney, and S.M. Sykes. 1942. Studies in the metabolism of apples. 1. Preliminary investigation on internal gas composition and its relation to changes in stored Granny Smith apples. Austral. J. Expt. Biol. Med. Sci. 20:219-223.

Wang, C.Y. and R.E. Anderson. 1982. Progress of controlled atmosphere storage and intermittent warming of peaches and nectarines, p. 221-228. In: D.G. Richardson and M. Meheriuk (eds.). Controlled atmospheres for storage and transport of perishable agricultural commodities. Timber Press, Beaverton, Ore.

Wankier, B.N., D.K, Salunkhe, and W.F. Campbell. 1970. Effects of controlled atmosphere storage on biochemical changes in apricot and peach fruits. J. Amer. Soc. Hort. Sci. 95:604-609.

Zagory, D., D. Ke, and A.A. Kader. 1989. Long term storage of 'Early Gold' and 'Shinko' Asian pears in low oxygen atmospheres p. 353-357. In. J.K. Fellman (ed.). Proc. Fifth Intl. Controlled Atmospheres Res. Conf., Wenatchee, Wash.