Gas Exchange in Cut Apples with Bilayer Coatings

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Cut apple pieces coated with double layers of buffered polysaccharide/lipid showed a 50–70% reduction in the rate of CO_2 evolution and about 90% decrease in C_2H_4 as compared with uncoated controls at ~23 °C. This substantial decline in the rates of gas evolution was attributed primarily to the diffusion barrier properties of the lipid layer and secondarily to the inhibitory effect of the ascorbate buffer which contained calcium. Internal CO_2 increased and leveled off at concentrations of ~120 mL/L after 8 h. The internal concentration of C_2H_4 decreased after an initial rise and reached a low level comparable to that observed in the untreated samples after 24 h. There was ~50–75% reduction in the internal O_2 concentration, while the headspace O_2 showed minimal changes. These results suggest that the inhibitory effects of internal CO_2 accumulation and O_2 depletion may be responsible for the reduced gas exchange in the coated fruit.

Keywords: *Edible coating; gas exchange; headspace gases; internal gases; minimal processing*

The market demand for minimally processed fruits and vegetables has undergone a rapid expansion in recent years. Consumers prefer fresh produce that is prepared for convenient consumption without the loss of quality attributes such as texture, flavor, and appearance. Minimal processing includes all operations such as washing, sorting, trimming, peeling, slicing, coring, etc. that would not affect the freshlike quality of the product. Minimally processed products are physiologically active, in that the tissues are living and respiring, with increased perishability. Undesirable changes caused by minimal processing include cellular disruption, increased respiration, increased synthesis of ethylene, induction and acceleration of enzymatic breakdown, and formation of secondary metabolites (Schwimmer, 1981; Rolle and Chism, 1987).

Various approaches have been used to minimize the deteriorative effects caused by minimal processing. These include low-temperature storage, special preparation techniques, use of additives, modified/controlled atmosphere, and application of edible coatings (King and Bolin, 1989; Cantwell, 1992). The rationale for using edible coatings to extend shelf life and improve the quality of minimally processed produce is based on the formation of an artificial barrier that may result in (1) reduction of moisture migration, (2) selective control of gas diffusion, and (3) suppression of undesirable physiological changes (Wong et al., 1994). Most investigations focus primarily on the effect of edible coatings on moisture retention. There has been relatively little attention directed to the other two important factors that may well determine the success of using edible coatings on cut fruits and vegetables.

The objectives of this study were to evaluate the effects of bilayer coatings of polysaccharide/lipid on the gas diffusion pattern and the composition of the internal gases in cut apple pieces.

MATERIALS AND METHODS

Materials. Commercial grade pectin (LM 35) was obtained from TIC Gums Inc., Belcamp, MD. Microcrystalline cellulose (Avicel RC591F) was provided by FMC Corp., Philadelphia, PA. Alginate (Manugel GMB) was obtained from Kelco International Ltd., London. Acetylated monoglyceride (Myvacet 5-07) was from Eastman Chemical Products, Kingsport, TN. All other chemicals and reagents were of analytical grade.

Coating Preparation. Apples were cut into cylinders of 2 cm diameter × 2 cm. The apple pieces were first dipped in a solution containing 1% ascorbic acid, 0.5% citric acid, 0.25% $CaCl_2$, and 2% NaCl. The apple pieces were then double-coated by dipping successively in a 0.5% polysaccharide solution and a melted solution of acetylated monoglyceride.

Water Loss. Coated and untreated samples were stored at 23 ± 1 °C, 50–55% humidity. Percent loss of the initial weight was calculated from the difference in the sample weight taken at 24-h intervals. Rate of moisture loss was calculated by regression analysis of the slope of the curve of weight loss vs time. Water vapor resistance was determined using a modification of Fick's diffusion law (see Mathematical Analysis).

Headspace Gases. The effect of coating on carbon dioxide (CO_2), oxygen (O_2), and ethylene (C_2H_4) was determined by analyzing the headspace gas composition. Typically, three apple pieces, coated or untreated, were stored in a 450-mL sealed glass container at 23 ± 1 °C. Headspace samples were withdrawn at various time intervals and analyzed for CO_2 and C_2H_4 by gas chromatography. For the analysis of CO_2, an HP5890 GC equipped with a thermal conductivity detector and a packed column (183 cm × 0.32 cm ss) of Chromosorb 107 (80–100 mesh) was used. Column temperature was maintained at 60 °C. Ethylene was analyzed with an HP5880 GC equipped with a flame ionization detector and a 183 cm × 0.16 cm glass column packed with 80–100 mesh Porapak N and maintained at 80 °C. For both GC analyses, the injector temperature was 125 °C, the detector temperature was 250 °C, and the He flow rate was 30 mL/min. Concentration of O_2 was analyzed by Mocon HS750 (Minneapolis, MN) oxygen analyzer. Gas rates were calculated by regression analysis of the linear slope of the curve before saturation.

Internal Gases. Concentrations of CO_2, O_2, and C_2H_4 in apple tissues were determined using the evacuation chamber method described by Beyer and Morgan (1970). The main body of the collection flask had a volume slightly larger than the apple piece to minimize the residence time of the evacuated gas in the saturated NaCl solution. A 3-mL calibrated tube with a septum was attached to the top of the flask. Internal gases evacuated from the apple tissue were collected in the calibrated tube and analyzed as described above.

Mathematical Analysis. Estimation of the resistance factor was obtained by using Fick's diffusion law: $ds/dt = (A\Delta C)/R$, where ds/dt is the rate of gas exchange ($g \cdot s^{-1}$), R is the resistance ($s \cdot cm^{-1}$) of the coating to gas diffusion, ΔC is the concentration of gas ($mL \cdot cm^{-3}$) inside and outside the apple

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Table 1. Effect of Bilayer Coatings on the Resistance of Cut Apple Pieces to Water Vapor

coating	resistance ($s\cdot cm^{-1}$) 1	2	3	4	av ± SD	% increase after coating
untreated	3.03	3.03	3.30	3.57	3.23 ± 0.22	
Avicel/AMG	40.69	42.95	50.59	50.01	46.06 ± 4.32 a	1326
alginate/AMG	46.07	46.40	42.36	43.54	44.59 ± 1.70 ab	1280
carrageenan/AMG	35.47	40.24	41.87	37.13	38.68 ± 2.52 b	1098
pectin/AMG	35.88	38.63	39.47	38.20	38.04 ± 1.33 b	1078

[a] Means followed by different letters are significantly different ($P > 0.01$) by Duncan's multiple-range test.

piece at time t, and A is the surface area (cm^2) of the apple piece (Burg and Burg, 1965). For water vapor resistance, $\Delta C = (P_i - P_a)/R_cT$, where $(P_i - P_a)$ is the difference in water vapor pressure (mmHg) inside and outside the apple tissue, R_c is the gas constant (3.46 $L\cdot mmHg\cdot K^{-1}\cdot g^{-1}$), and T is in degrees Kelvin.

RESULTS AND DISCUSSION

Four polysaccharide/lipid formulations were used in the bilayer coating of cut apple pieces. The outer layer was acetylated monoglyceride (AMG). For the inner layer, a number of materials, including many commonly used polysaccharide gums and proteins, were screened on the basis of their ability to form a uniform adhesive layer on the cut surfaces, as a protective means for setting an outer layer of AMG (Wong et al., 1994). Four polysaccharides were selected for further studies. Two of these, alginate and pectin, adhered to the cut surface by cross-linking with Ca^{2+} supplied by the dipping buffer. The carrageenan used in the experiment is primarily a kappa type. Avicel exists as a fine suspension of microcrystalline cellulose (cellulose gel) and sodium carboxymethylcellulose (cellulose gum). Both kappa carrageenan and Avicel may be affected by Ca^{2+}, but only to a very limited extent (Wong, 1989).

Water vapor resistance values of the bilayer coatings were determined. All four bilayer coatings had a water vapor resistance of 38–46 $s\cdot cm^{-1}$ (Table 1). Untreated cut apple pieces, in contrast, had a very low resistance of 3.23 $s\cdot cm^{-1}$. These results are comparable to that reported for uncut waxed citrus fruits (Ben-Yehoshua et al., 1985). Waxing has been shown to increase the water resistance of Valencia oranges by 20 $s\cdot cm^{-1}$. Among the coating formulations, Avicel/AMG was significantly better than carrageenan/AMG or pectin/AMG on the basis of Duncan's multiple-range test. However, compared to the lipid layer, the polysaccharide layer in general does not provide substantial protection against water loss (Kester and Fennema, 1986). While a wax coating can result in marked reductions in weight loss, the relative ineffectiveness of hygroscopic coatings in this respect has been well documented (Ben-Yehoshua, 1969; Banks, 1984a; Smith and Stow, 1984).

The effects of these bilayer coatings on respiratory activity (CO_2) and C_2H_4 production were investigated. Physical stresses, such as cutting and slicing, are expected to cause an increase in respiration and C_2H_4 production. These types of physiological response have been well established in the literature (Laties, 1978; Yang and Pratt, 1978; Uritani and Asahi, 1980). The rates of CO_2 and C_2H_4 production were estimated from the linear portion of the slope of a plot of the amount of headspace gas vs time and thus represent the *observed* rate of gas evolved (diffused through the coating) and are not necessarily equal to the actual rate of production. All four coatings tested exhibit a substantial rate reduction in both gases. The results are particularly significant in the case of C_2H_4 gas in that the coated samples showed ≥90% decrease in the rate of evolution, compared with that of the control (Table 2). Reductions in CO_2 and C_2H_4 evolution have been observed in waxed whole fruits, including apples (Meheriuk and Porritt, 1972), avocados (Durand et al., 1984), and citrus fruits (Ben-Yehoshua et al., 1985). Comparison of the percent reduction of the rate of CO_2 that evolved among the four types of coatings (Avicel/AMG, alginate/AMG, carrageenan/AMG, and pectin/AMG) indicates that the Avicel/AMG coating had a significantly greater effect with a difference of 11.9–19.6%. There was no statistically significant differences in the percent reduction of the rate of C_2H_4 gas evolution among the four treatments. Repeating experiments using the same coatings on different commodities confirmed the reduction in the rate of CO_2 evolution in a general range of 50–70% and for C_2H_4 in the 90% range (unpublished results). The rather indiscriminate pattern exhibited by the four bilayer coatings used in the present investigation suggests that the large reduction in the rates of gas evolution could primarily be the effect of the AMG layer acting as a barrier against gas diffusion.

To test the above assumption, a series of experiments was conducted to assess individually the effect of each of the three steps employed in the coating of pectin/AMG on apple pieces. The results presented in Table 3 indicate that there was a considerable decrease in the rates of CO_2 and C_2H_4 evolved in samples treated with ascorbate buffer alone as compared with no treatment. Additional coating with pectin (i.e., ascorbate buffer plus pectin coating) did not change the rates of gas evolution for CO_2 and showed no consistent pattern for C_2H_4. The percent gas rate reduction between the ascorbate buffer-treated and the ascorbate buffer/pectin-coated samples was statistically insignificant by Duncan's multiple-range test. A large portion of the decrease in the complete system could therefore be attributed to the AMG layer. The effect of ascorbate buffer is not unexpected, since the Ca^{2+} contained in the buffer is known to suppress respiratory activity and C_2H_4 production (Poovaiah et al., 1988). In addition, Ca^{2+} is involved in the regulatory function of many enzyme actions in cellular and physiological processes. The addition of 100 mM Ca^{2+} has been shown to inhibit ethylene production in apple slices during the first 6 h of incubation (Lieberman and Wang, 1982). A similar reduction was observed when sliced pear and strawberry were dipped in 1% $CaCl_2$ in combination with modified atmosphere storage (Rosen and Kader, 1989). Considering the 0.25% (22.5 mM) concentration of $CaCl_2$ used in the buffer treatment, the observed large decline of gas production may not be entirely due to the action of Ca^{2+}. The possible effects of Ca^{2+} and other additives are currently under investigation.

The large reduction in the rate of gas evolution in these experiments poses two immediate questions that require further consideration. (1) What were the consequences of the reduction of gases diffused through the

Table 2. Observed Rate of Carbon Dioxide and Ethylene Evolved in the Headspace of Untreated and Coated Apple Pieces[a,b]

	carbon dioxide ($mg{\cdot}kg^{-1}{\cdot}h^{-1}$)			ethylene ($mg{\cdot}k^{-1}{\cdot}h^{-1} \times 10^{-4}$)		
exptl set[c]	untreated	coated	% reduction	untreated	coated	% reduction
Avicel + AMG						
A1	27.75	7.13	74.32	305.42	22.32	92.69
A2	25.20	10.58	69.93	592.21	38.55	93.46
A3	50.86	17.38	65.82	902.81	39.48	95.63
A4	48.70	15.66	67.85	514.66	41.93	91.85
A5	40.38	11.23	72.20	842.78	38.61	95.42
A6	29.52	6.69	77.70	93.96	13.47	85.66
A7	54.31	11.77	78.33	335.91	49.78	86.01
A8	42.43	19.33	54.45	295.98	18.84	93.63
			70.08 ± 7.21 a			*91.79 ± 3.64*
pectin + AMG						
B1	45.50	20.53	54.88	533.45	26.11	95.11
		19.68	56.74		25.12	95.29
B2	46.23	24.68	46.60	407.80	23.61	94.21
		22.44	51.47		27.76	93.19
B3	48.97	26.56	45.77	659.98	39.62	93.99
		22.50	54.06		35.03	94.69
B4	40.39	23.39	42.10	755.68	59.94	92.07
		19.77	51.95		37.98	94.97
B5	42.96	21.69	49.50	612.14	52.35	91.45
		20.71	51.80		38.26	93.75
			50.49 ± 4.30 bd			*93.87 ± 1.23*
alginate + AMG						
C1	33.00	14.96	54.67	412.46	34.40	91.66
		12.06	63.45		24.20	94.13
C2	30.92	14.25	53.90	273.94	31.95	88.34
		13.28	57.05		27.10	90.11
C3	34.35	16.02	53.35	339.54	30.97	90.88
		14.61	57.46		35.93	89.42
C4	32.96	14.75	55.25	381.33	24.24	93.64
		13.44	59.22		14.53	96.19
C5	34.38	12.93	62.38	318.19	23.56	92.60
		12.02	65.03		18.11	94.31
			58.18 ± 3.97 cd			*92.13 ± 2.35*
carrageenan + AMG						
D1	38.45	19.73	48.68	363.85	37.27	89.76
		16.65	56.71		25.37	93.03
D2	38.94	17.04	56.24	471.92	24.89	94.73
		16.13	58.57		22.40	95.25
D3	40.21	19.03	53.88	424.82	22.16	94.79
		18.61	53.73		24.19	94.31
D4	33.77	13.95	58.68	460.06	21.74	95.27
		15.40	54.41		29.76	93.53
D5	40.70	15.30	62.41	690.30	35.93	94.79
		15.67	61.50		37.84	94.52
			56.48 ± 1.63 db			*94.00 ± 1.56*

[a] Numbers in italics are means of experimental sets A–D. Means followed by different letters are significantly different. [b] Gases were analyzed by GC at 2-h intervals. Rates ($mg{\cdot}k^{-1}{\cdot}h^{-1}$) were calculated on the basis of the linear portion of the slope of the curve before saturation. [c] Experimental sets: A, Avicel + AMG; B, pectin + AMC; C, alginate + AMG; D, carrageenan + AMC. Each of the A sets consisted of one control and one sample. Parts B–D contained one control and two coated samples in each set. Each sample contained three separately processed apple pieces. All samples were treated with ascorbate buffer before coating.

coating? Specifically, did the gas accumulate in the tissue and, if so, to what extent? (2) More importantly, what were the effects of forming a barrier to gas diffusion on the tissue's respiratory activity and C_2H_4 production? To provide answers to these questions, both headspace and internal gases (CO_2, C_2H_4, and O_2) were analyzed separately in several sets of experiments consisting of coated and untreated apple pieces. Since the procedure for the analysis of internal gases was destructive, the analysis of headspace gases did not allow for estimating the rate of gas evolution but did give the actual *amount* of each gas evolved at specific time intervals. Figure 1 shows that there was an increase in the amount of CO_2 evolved in both the coated samples and the controls, as determined by headspace analysis. The bilayer coating caused a decrease in the amount of CO_2 evolved throughout the time course. The evolution of C_2H_4, however, followed a very different pattern (Figure 2). The C_2H_4 evolved in the control showed a steep increase, while that of the coated samples remained at a fairly constant low level. The headspace O_2, shown in Figure 3, showed a very slight decrease by ~0.2% and 0.4% in the coated and untreated samples after 24 h, respectively.

The percent reduction of the amount of CO_2 and C_2H_4 that evolved over time was then plotted in Figure 4. The percent reduction of the amount of CO_2 from the tissue decreased with time from ~56% at the 2-h interval to ~26% after 24 h. In contrast, C_2H_4 reduction follows a positive correlation, reaching ~95% reduction after 24 h. The coating therefore was at first sight more effective in forming a barrier to C_2H_4 than to CO_2. The results also seem to reaffirm the data described in the above section indicating that the percent rate reduction of C_2H_4 was in the ~90% range, compared with the lower percent range for CO_2 (Table 2).

However, considering that the concentration of CO_2 was in the range of milliliters per liter while the C_2H_4

Table 3. Effect of Ascorbate Buffer, Pectin, and Acetylated Monoglyceride on the Reduction of the Rate of Carbon Dioxide and Ethylene Production

	% reduction of gas production[b]					
	ascorbate buffer		+ pectin		+ acetylated monoglyceride	
exptl set[a]	CO_2	C_2H_4	CO_2	C_2H_4	CO_2	C_2H_4
1	35.46	15.21	26.75		53.19	87.39
2	5.18	5.46	8.97	3.94	33.48	83.23
3	28.20	16.63	20.94	45.35	44.51	86.99
4	18.52	26.46	19.54	53.76	50.21	91.85
5	35.32	37.71	45.83	40.55	52.54	89.11
6	32.00	53.25	28.72	38.37	47.92	91.62
7	19.78	33.83	40.83	41.51	49.58	81.57
8	27.07	34.32	22.04	25.93	57.62	88.62
9	24.19	26.13	27.08	37.09	47.05	86.62
10	31.27	32.84	24.01	26.86	50.99	89.40

[a] Each experimental set consisted of one control and two treated samples. Each sample contained three separately processed apple pieces. [b] Gases were analyzed by GC at 2-h intervals. Rates were calculated on the basis of the linear portion of the slope of the curve before saturation. Percent reduction represents an average for an individual experimental set.

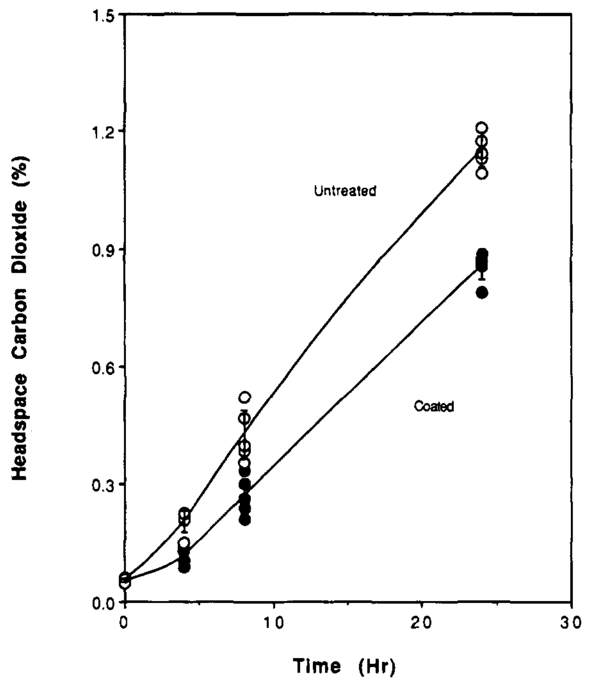

Figure 1. Amount of CO_2 evolved in coated (pectin/acetylated monoglyceride) and untreated apple pieces. Each datum represents an average based on the measurement of headspace gases in two sets of coated samples and two sets of control (untreated samples). Each set contained three separately processed apple pieces. Vertical bars are standard deviations of all data points at each time interval. The curves are interpolation of the average of all data points. The x-axis represents time intervals after the coating procedure described in the text.

concentration was found in the range of microliters per liter (i.e., 3 orders of magnitude less), it could be that the decrease in the percent reduction of the amount of CO_2 evolved (with respect to the increasing CO_2 production in time) was the result of a pressure gradient due to a concentration effect instead of a preferential diffusibility of the CO_2 across the coating. The limiting factor therefore could well be the amount of gas produced rather than the gas resistance of the coating. This assumption could be first tested by determining the resistance of the coating to the two gases. To this end, experiments were designed to measure the gas evolution

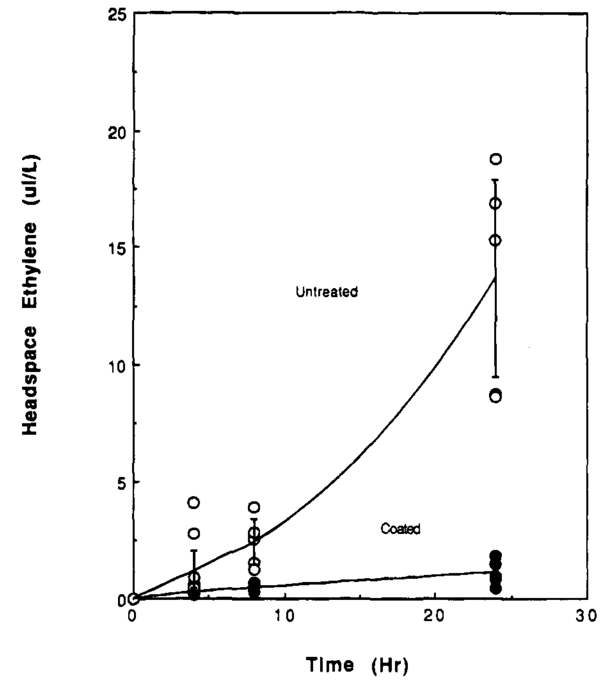

Figure 2. Amount of C_2H_4 evolved in coated and untreated apple pieces. Details are given in Figure 1 legend.

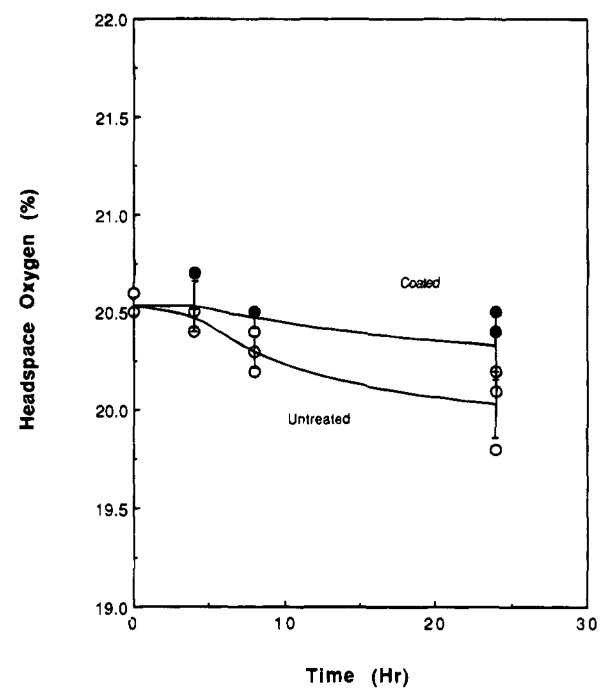

Figure 3. Amount of O_2 in the headspace of coated and untreated apple pieces. Details are given in Figure 1 legend.

rate by monitoring the headspace composition of the gases, after which the samples were removed and subjected to internal gas analysis. The R value for CO_2 calculated from the measurements was found to be approximately 30 times that for C_2H_4 (Table 4). Thus, there is a strong indication that the decrease and increase in the percent reduction of the amount of CO_2 and C_2H_4 diffused across the coating with time were definitely not consequential to the differences in the resistance of the coating toward the two gases. The remaining question then is whether or not the reduction was caused by a concentration effect.

A further insight into the process can be derived by a parallel comparison between the percent reduction of

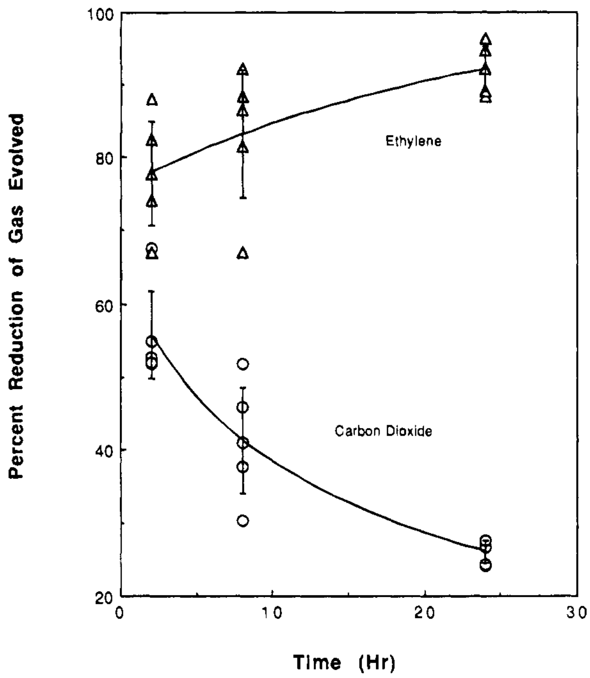

Figure 4. Percent reduction of CO_2 and ethylene in coated apple pieces. Details are given in Figure 1 legend.

Table 4. Gas Resistance of Bilayer Coatings[a]

exptl set[b]	resistance (s·cm^{-1})		
	CO_2	C_2H_4	O_2
1	4.9×10^4	1.9×10^3	4.2×10^7
2	3.4×10^4	0.6×10^3	4.4×10^7
3	3.7×10^4	1.8×10^3	5.4×10^7
4	3.8×10^4	1.5×10^3	

[a] Headspace gases were analyzed by GC at 2-h intervals. Internal gases were determined at the 6-h interval. Internal O_2 was analyzed at the 24-h interval. [b] Each experimental set consisted of two samples. Each sample contained three separately processed apple pieces. For O_2 analysis, each set consisted of three apple pieces, and the internal O_2 was taken as the total amount in the three apple pieces.

gas evolved and the internal gases and the amount evolved in the headspace for both the coated and the control (untreated) samples. The differences in the amount of CO_2 evolved, as indicated in Figure 1, remained fairly constant with the two curves following a similar slope, in particular, after 8 h of coating. These results are consistent with the conclusion drawn from Figure 4. Since the resistance to CO_2 diffusion was constant, the amount of gas diffused across the coating had to be proportional to the increased amount of gas produced. Consequently, the percent reduction of gas evolution decreased with time, because the gas production increased with time. The positive correlation of the percent reduction of C_2H_4 evolution with time, as indicated in Figure 4, is also consistent with the comparison presented in Figure 2. The fact that the two curves representing the untreated and the coated samples did not run parallel to one another also suggests a possible inhibition of C_2H_4 production in the coated sample in addition to simple diffusion resistance (see following discussion on internal C_2H_4).

Further evidence can be revealed by taking a closer look at the internal concentrations of these two gases (Figures 5 and 6). In the case of CO_2, the concentration in the untreated apple tissues was unchanged, while a substantial overall increase could be observed in the coated samples (Figure 5). The internal concentration

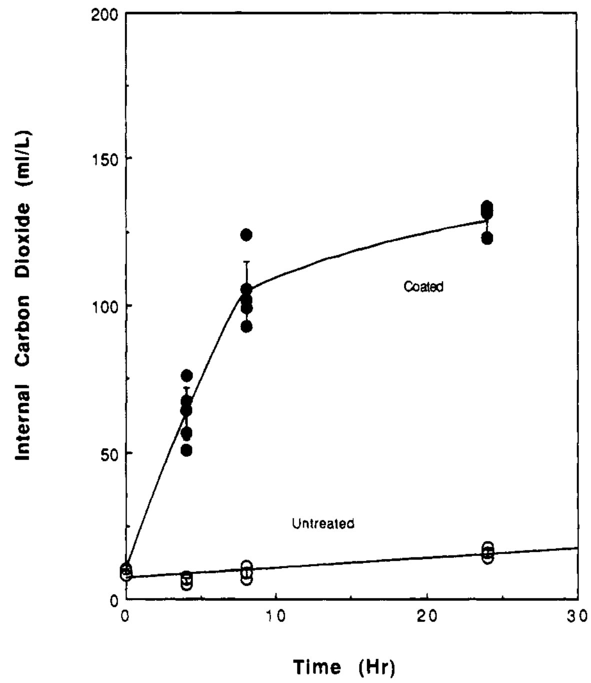

Figure 5. Internal CO_2 in coated and untreated apple pieces. Each datum represents an average based on the measurement of internal gases in three sample sets and two controls. Each set contained three separately processed apple pieces. Vertical bars are standard deviations of all data points at each time interval. The curves are interpolation of the average of all data points.

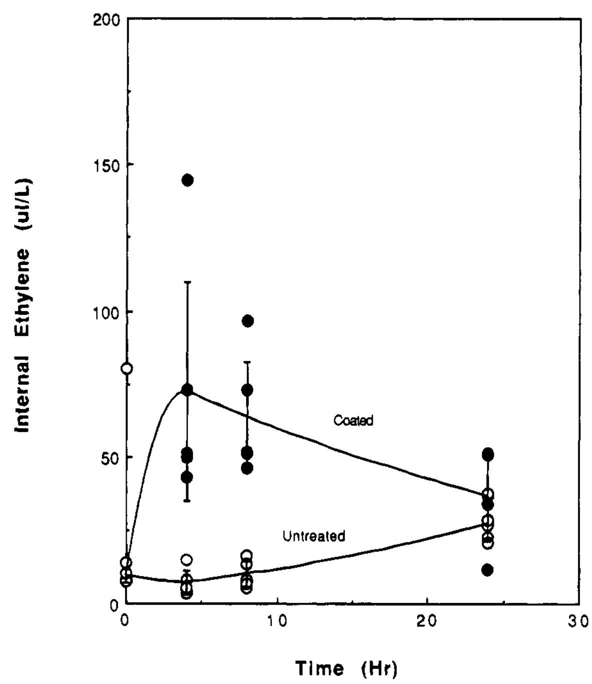

Figure 6. Internal C_2H_4 in coated and untreated apple pieces. Details are given in Figure 5 legend.

of CO_2 increased initially and gradually leveled off upon further storage. The internal CO_2 reaches saturation at a concentration range of ~120 mL/L. Consequently, the actual amount of CO_2 produced by the tissue and initiated by cutting was detected largely as gas evolved. This leveling plateau in the curve therefore represents an equilibrium that is reached when the CO_2 produced is proportional to the CO_2 evolved (diffused across the coating). It has been reported that wax treatment of whole Mandarin fruit caused an increase in the concentration of internal CO_2, reaching a maximum in 7 h, after which time it leveled off (Hasegawa and Iba, 1980). This reported observation on whole fruit surprisingly matches closely the results obtained in the present investigation. It is quite possible that the observed leveling of CO_2 in coated fruits reflects, at least in part, a self-inhibition effect of the tissue's respiratory activity by the accumulated CO_2. Burg and Burg (1965) proposed such an inhibition effect to explain the 20–30% differences observed in the initial and final rate of respiration in green pepper and tomato having the pedicel sealed with lanolin. The inhibition of respiration by a high concentration of CO_2 exposure, which has been well demonstrated in numerous studies, forms the basis for modified (or controlled) atmosphere storage of whole fruits and vegetables (Smock, 1979; Kader, 1992). The mode of action of elevated CO_2 concentration is not clear. The effect has been generally attributed to the inhibitory action on the various enzyme systems involved in mitochondrial activity (Shipway and Bramlage, 1973; Monning, 1983; Chaves and Tomas, 1984). Although we cannot distinguish the saturation effect from the inhibition factor in the present investigation, it is likely that both played important roles in reaching the equilibrium state.

For internal C_2H_4, the control showed only a slight rise in gas concentration (Figure 6), apparently because a large portion of the C_2H_4 produced was evolved freely through the pulp with little restriction and was detected as a large increase of headspace C_2H_4 (Figure 2). For the coated samples, there was a continuous drop in the amount of internal C_2H_4 after an initial high concentration in the tissue (relative to that of the control). Taking into account the observation that the increase of headspace C_2H_4 of the coated samples was minute, it becomes evident that the restricted diffusion across the coating had a suppressive action on C_2H_4 production which was likely related to the alteration of the internal atmosphere of the fruit tissue (increase in internal CO_2 and decrease in O_2 as discussed below). The initial rise in internal C_2H_4 could also enhance the respiratory activity as shown in Figure 1 (Tucker and Grierson, 1987). However, while the CO_2 production continued to rise (Figure 1), the C_2H_4 concentration actually decreased after an initial burst (Figure 6). It could be that the initial rise in C_2H_4 had a "triggering" effect sufficient to cause a continued increase in respiration.

Various studies have reported an increase in internal CO_2 and a reduction of C_2H_4 in whole apples coated with sucrose polyester (Banks, 1984b; Drake et al., 1987; Smith et al., 1987). Accumulated CO_2 may function as a natural ethylene antagonist and delay many responses of fruit tissue to ethylene (Yang, 1985). The direct effect of elevated CO_2 on ethylene production is uncertain, although CO_2 has been shown to act as a competitive inhibitor of C_2H_4, causing a delay in ripening (Burg and Burg, 1967; Beyer, 1979). Certain studies seem to suggest a direct effect of CO_2 on ethylene synthesis, since an increase in endogenous 1-aminocyclopropane-1-carboxylic acid (ACC) of the treated tissue was detected (Chaves and Tomas, 1984). The effect of CO_2 on ethylene action would also affect ethylene synthesis in the case of autocatalytic ethylene production (where the effect or action of ethylene is to increase its own synthesis). The exact action has not been established. Oxygen, however, has been unequivocally demonstrated to be an essential component in the conversion of ACC to C_2H_4 in the biosynthetic pathway (Adams and Young,

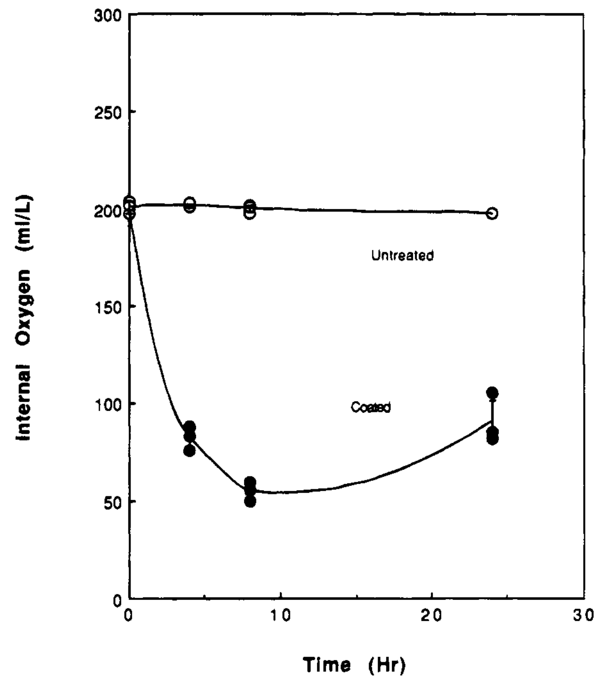

Figure 7. Internal O_2 in coated and untreated apple pieces. Details are given in Figure 5 legend.

1979). Therefore, it may well be that the increase in respiratory activity caused a depletion of O_2 which indirectly slowed C_2H_4 production. This assumption is indeed supported by the analysis of the internal concentration of O_2 in the untreated and coated samples (Figure 7). The internal O_2 concentration in the untreated samples did not change considerably, while that of the coated samples showed a significant drop of ~50–75% of the original concentration. Thus, one may suggest that the diffusion of headspace O_2 to the tissue was inhibited by high O_2 resistance of the bilayer coating. The coating in this experiment is calculated to have an R value of 4.7×10^7 s·cm^{-1} for oxygen (Table 4). In fact, dried films of acetylated monoglyceride (the same type of materials used for the lipid layer in the present investigation) have been reported to have a similar O_2 resistance of 1.11×10^7 s·cm^{-1} (Kester and Fennema, 1989). It is, therefore, reasonable to conclude that as the respiratory activity increased in the cut apple tissues and the internal O_2 was depleted, the high O_2 resistance of the coating could cause a high O_2 concentration gradient between the tissue and the atmosphere and, consequently, a reduction of C_2H_4 synthesis.

Another factor that may contribute to the decrease of internal C_2H_4 after an initial rise in concentration is the possible autoinhibitory effect of ethylene. Application of exogenous ethylene has been shown in some fruits to suppress ACC synthesis and also to increase malonylation of ACC; both processes can result in the reduction of ACC convertion to ethylene (Vendrell and McGlasson, 1971; Liu et al., 1985a,b). It should also be noted, however, that autocatalytic ethylene synthesis is commonly found in many fruits (Tucker and Grieson, 1987). Ethylene has been demonstrated to enhance wound-induced ethylene production by promoting the enzymatic conversion of ACC to the gas (Hoffman and Yang, 1982). The results obtained in this work do not enable us to distinguish the two opposite effects exerted by ethylene. Furthermore, most of the studies from which conclusions were made on the autocatalytic or autoinhibitory action of ethylene were done by external treatments of the gas. It is uncertain how well these results can be correlated to the effect of high internal ethylene concentration.

It is obvious that the application of an artificial barrier to diffusion by coating inevitably causes a substantial modification of the atmosphere of the internal tissue as well as of the interfacial layer surrounding the tissue. Desirable effects associated with coating whole fruits include decrease in the incidence of certain physiological disorders, retardation of color development, softening, ripening, and extension of postharvest life (Rohrbach and Paull, 1982; Dhalla and Hanson, 1988; Erbil and Muftugil, 1986). On the other hand, excessive restriction of gaseous diffusion has been known to cause off-flavor development and storage injury (Little and Peggie, 1987; Cohen et al., 1990). Caution, therefore, must be exercised in the use of coatings. Each coating must be individually tested and assessed under its own set of conditions.

CONCLUSION

The results obtained in the present investigation allow the following gas exchange pattern to be proposed. In apple pieces coated with polysaccharide/lipid bilayer, increased respiration causes a buildup of internal CO_2 in the tissue, which approaches a plateau ~8 h after coating. This internal accumulation of CO_2 along with the continuous production of the gas creates a concentration gradient across the coating. The increasing CO_2 production after saturation contributes to a continuous sharp increase in the gas evolved in the headspace, resulting in an apparent decrease in the percent reduction of CO_2 evolved (coated vs untreated as observed). The increase in respiratory activity caused a depletion in internal O_2 due to the partial permeability of the coatings to this gas which cannot be readily replenished by inward diffusion of atmospheric O_2 due to the restriction by the coating. The lowering of O_2 concentration contributes to the suppression of C_2H_4 production. The decrease in C_2H_4 production is supported by the observation that the amounts of C_2H_4 evolved and present in the tissue remain relatively unchanged, with the latter actually showing a continuous reduction with time. These observations may provide a model for optimizing the use of edible coatings and eventually help to improve the shelf life of lightly processed fruits and vegetables.

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Received for review February 18, 1994. Revised manuscript received July 14, 1994. Accepted August 4, 1994.[⊗] Reference to a company and/or product is only for purposes of information and does not imply approval or recommendation of the product to the exclusion of others which may also be suitable. All programs and services of the U.S. Department of Agriculture are offered on a nondiscriminatory basis without regard to race, color, national origin, religion, sex, age, marital status, or handicap.

[⊗] Abstract published in *Advance ACS Abstracts*, September 15, 1994.