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AN RTD DETERMINATION METHOD FOR EXTRUSION COOKING[1]

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ABSTRACT

The residence time distribution (RTD) in an APV Baker twin-screw extruder was studied by injecting a red color during extrusion of rice flour. The redness color value from a Hunter Colorimeter and the red dye concentration of the ground extrudate were used to determine the RTD of rice flour and RTD spread. The redness color values significantly overestimated the mean residence times and RTD spreads ($p < 0.0001$). The redness color values cannot replace the red dye concentrations to determine the residence time distribution in a twin-screw extrusion cooking system.

INTRODUCTION

Extrusion technology has been widely used in plastic, rubber, food, metal, carbon, and ceramic industries (Fellows 1988; Harper 1981; Mercier *et al.* 1989). While single-screw cooking extruders for food and feed applications were initiated in the 1940's, twin-screw extruders were not used for food processing until in the 1970's; however, the number of applications have rapidly expanded throughout the 1980 (Mercier *et al.* 1989). This is because twin-screw extruders have better mixing abilities, more uniform temperature distribution, a better control of residence time, and more positive displacement action than single-screw extruders (Harper 1981; Lin and Armstrong 1990).

The most important factors affecting the extrusion systems are the operating conditions of the extruder and the rheological properties of the food (Fellows

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1988). The residence time distribution (RTD) of the feed material in an extruder is one of the most important parameters characterizing the mixing and chemical kinetics that occur in extruders. It is also a useful tool in determining the optimal operation conditions for blending, dispersing, and polymerization applications (Bruin *et al.* 1978; Lin and Armstrong 1990). Todd (1975a,b) suggested that the RTD data is most useful in diagnosing axial mixing phenomena in twin-screw extruders, since it provides the basis for scale-up and guides in improvements of equipment. Knowledge of RTD in the extruders is also important for considerations in the nutrient degradations, food safety, and control of the product quality.

Residence time distributions of the material in an extruder has be determined by using radioactive-tracer and dye-tracer techniques (Bounie 1988; Davidson *et al.* 1983; Harper 1981; Jager *et al.* 1988, 1989; van Zuilichem *et al.* 1988a,b,c; Wolf *et al.* 1976, 1986) and are generally expressed by E curves and F curves (Levenspiel 1972; Smith 1981). The advantage of the radioactive-tracer technique is the ability to conduct on-line measurements. But it will present problems in the disposal of low level radioactive waste, however, and is more expensive than the dye-tracer technique (Hong *et al.* 1991). The dye-tracer technique can be classified with the fluorescent dye method and color dye method. A colorimeter is usually used in the color dye method for the RTD determination.

Recently, two different color dye methods were used to determine the RTD in the extrusion system: one based on the color values of the extrudate directly from a colorimeter (Altomare and Anelich 1988; Altomare and Ghossi 1986; Likimani, *et al.* 1990; Onwulata 1991) or a color difference meter (Chen *et al.* 1990); the other obtained from the color concentrations (Yeh *et al.* 1992). The comparison between these two methods have never been reported in the literature. Since many researchers used the extrudate color values from a colorimeter for the RTD determination, which is straightforward and less time-consuming than extracting the dye from the extrudate, it is interesting to investigate whether the color values from a colorimeter could also yield acceptable RTD results in a twin-screw extruder.

RESIDENCE TIME DISTRIBUTIONS

The residence time distributions of the material in the extruder are usually described as E and F curves (Altomare and Anelich 1988; Lin and Armstrong 1990). The E curve shows the exit age distribution and is plotted as normalized concentration E(t) vs. residence time (t). The cumulative E(t), i.e., F(t), vs. normalized time (residence time/mean residence time) is plotted as the F curve.

$$E(t) = \frac{c}{\int_0^\infty c\,dt} \approx \frac{c}{\sum_0^\infty c\Delta t} \tag{1}$$

$$F(t) = \int_0^t E(t)\,dt \approx \frac{\sum_0^t c\Delta t}{\sum_0^\infty c\Delta t} \tag{2}$$

where c is concentration and t is time.

The mean residence time ($\bar{t}$), which represents the mean time of the material in the extruder, was given by Smith (1981):

$$\bar{t} = \int_0^\infty tE(t)\,dt = \frac{\sum_0^\infty tc\Delta t}{\sum_0^\infty c\Delta t} \tag{3}$$

The variance (σ^2), which represents the square of the spread of the RTD distributions, was given by Levenspiel (1972):

$$\sigma^2 \approx \frac{\sum_0^\infty (t-\bar{t})^2 c\Delta t}{\sum_0^\infty c\Delta t} = \frac{\sum_0^\infty t^2 c\Delta t}{\sum_0^\infty c\Delta t} - \bar{t}^2 \tag{4}$$

MATERIALS AND METHODS

Materials

Rice flour (RL-100, RIVLAND, Stuttgart, AK), adjusted to a moisture content of 20.0% (wb), was used as the feed material. Red dye (FD&C, #40, Warner Jenkinson, St. Louis, MO) was chosen as the tracer to measure the residence time distributions of the rice flour in a twin-screw extruder.

Extruder

A corotating and intermeshing APV Baker MPF50/25 twin-screw extruder (APV Baker, Inc., Grand Rapids, MI) was used in this study. The power of

this machine is 28.0 kW (kilowatt) and the total barrel length:diameter ratio is 25:1. The barrel diameter is 50 mm. There are nine temperature controlled barrel sections, of which six sections were used (Hsieh *et al.* 1990). The barrel temperature was set at 26.7C (feeding zone), 26.7C, 51.7C, 93.3C, 121.1C, and 121.1C, respectively, throughout the experiments. The screw profile used in this study is shown in Fig. 1. The maximum screw speed of this machine is 500 rpm. Two circular dies, each with one hole, were used. The diameter of each hole was 27.25 mm initially, then tapered to 3.18 mm down stream in a distance of 15.46 mm, and then 3.18 mm in diameter for another 3.08 mm further down stream. Rice flour was fed into the extruder with a K-tron Type T-35 twin-screw volumetric feeder with a Series 6300 controller (K-tron Corp., Pitman, NJ). The adjustable cutter with four blades is operated at 325 rpm. A computer data acquisition system was used to record feed rate, barrel and product temperatures, die pressure and temperature, % torque, screw speed and cutter speed. It takes about 15 min for the extruder to reach the steady state after startup.

Experimental Design

The variables in this study were the feed rate (30, 40, and 50 kg/h) and the screw speed (200, 300, and 400 rpm). The moisture content of feed was adjusted to 20% (wb) by injecting water at ambient temperature into the extruder with a pump. Two replications were used in this 3 × 3 factorial design. For each treatment, 33 samples were collected in a 6 min interval.

Standard Curve Experiment

A standard curve of this study was established by a separate experiment. Rice flour and red dye were mixed in a Hobart N50 mixer (Hobart Canada, Inc., North York, Ontario, Canada) to the red dye concentrations of 0.0%, 0.01%, 0.025%, 0.05%, 0.075%, 0.1%, 0.15%, 0.2%, 0.4%, and 0.6% (w/w). The mixtures were extruded in the same extruder at 20% moisture (wb), 40 kg/h and 300 rpm. When the extruder reached the steady state, a timer was started. Samples for each mixture were collected in 1 min intervals during the 4th to 6th min. The color values (L, a, and b) of each sample were measured according to the method described below, and the concentrations (g color/100 g rice flour) vs. color values were plotted as the standard curve for the experiment.

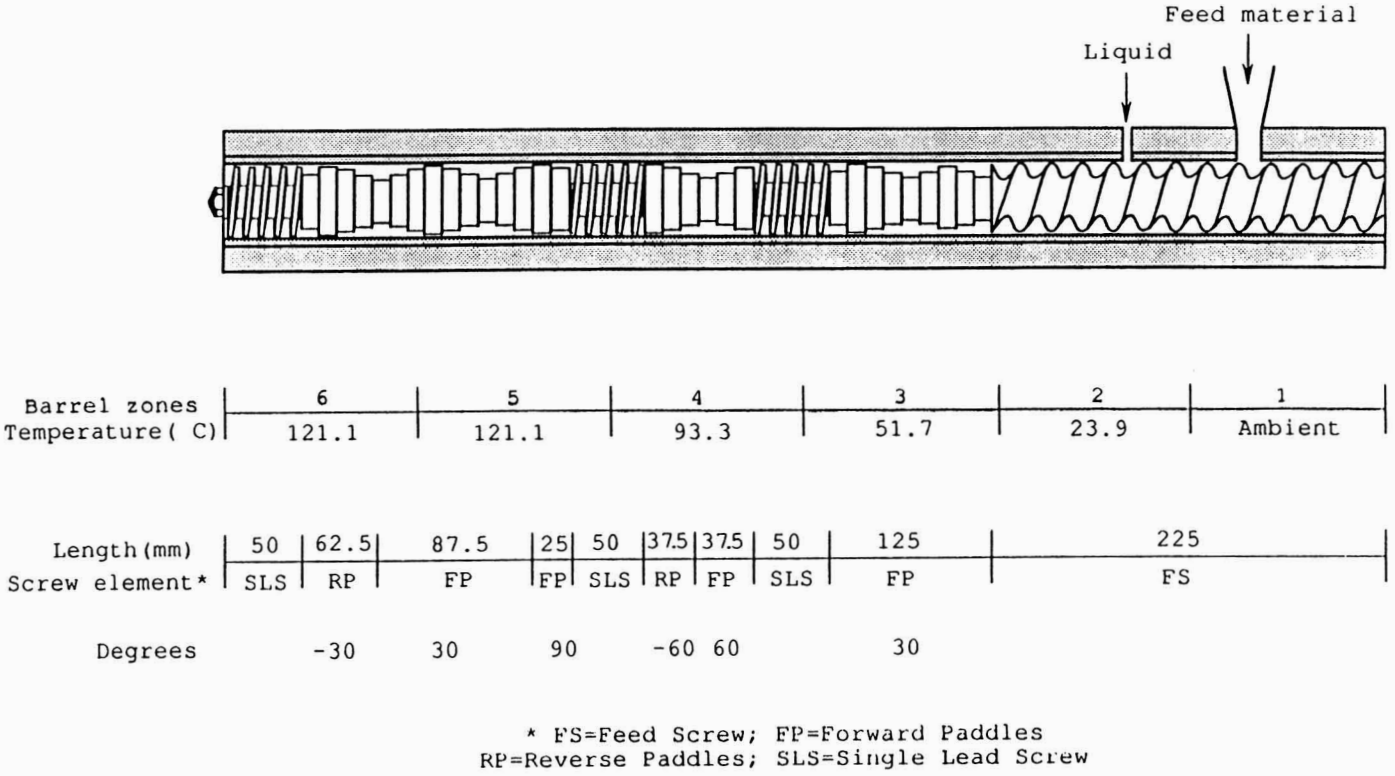

FIG. 1. THE SCREW PROFILE OF AN APV BAKER MPF 50/25 TWIN-SCREW EXTRUDER

Collection of Samples and Color Measurement

Samples of each treatment were collected when the extruder reached the steady state. One gram of red dye was added into the feed port of the extruder along with the rice flour as a tracer, and a timer was started simultaneously. Meanwhile, the extrudate samples were collected for up to 6 min (10 s intervals in the first 30 s, 5 s intervals in the next 60 s, 10 s intervals in the following 90 s, and 20 s intervals in the last 180 s). A total of 33 samples were collected for each treatment. Each sample was ground by a Waring Blendor (New Hartford, CT) and passed through the Taylor Standard Sieve No. 8. The color values (L, a, and b) for each ground sample were measured and recorded using a HunterLab D25 colorimeter (Hunter Associates Lab., Inc., Reston, VA). A white tile (Standard No. C2-28656; L: 91.2, a: −0.9, and b: −0.7) was used to standardize the colorimeter. For each sample, duplicate sets were measured, and two color readings (L, a and b) were recorded for each set. The second reading was obtained after a 90° rotation of the sample. Thus, four readings of color values L, a, and b, respectively, were recorded for each sample.

Data Analysis

Two approaches were used in this research to calculate the mean residence time ($\bar{t}$) and the spread of the residence time distribution (σ) for each treatment. The first approach was based on the redness color values (a) directly from a HunterLab colorimeter. The second approach was arranged by converting the redness color values to color concentrations using the standard curve established.

LOTUS 1-2-3 computer software (Lotus Development Corporation, MA) was used to calculate the mean residence time ($\bar{t}$) and the spread of the residence time distribution (σ) for each treatment in the 3 × 3 factorial design experiment. Significant differences in the mean residence times ($\bar{t}$) and the RTD spreads (σ) between the two approaches were studied using the General Linear Models procedure in SAS (1985). The E curves comparing the redness color value approach with the concentration approach were plotted using SigmaPlot (Jandel Scientific, San Rafael, CA).

RESULTS AND DISCUSSION

Standard Curve

For the standard curve experiment, the color values of L, a, and b versus color concentrations (g color/100 g rice flour) of the mixtures are plotted in

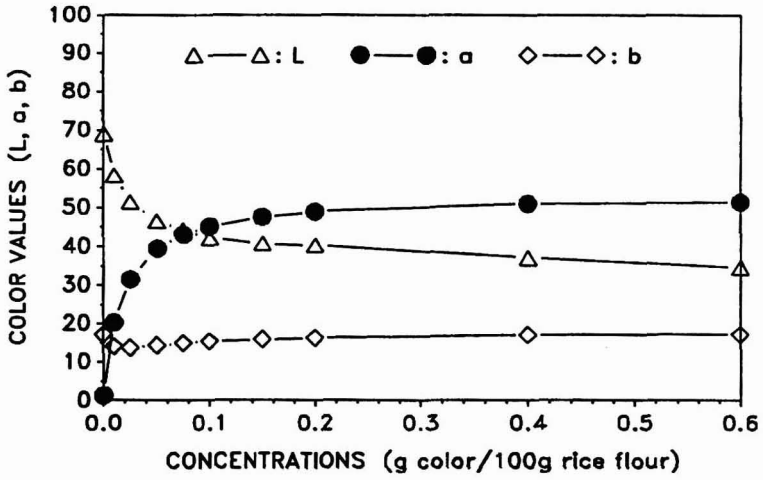

FIG. 2. COLOR VALUES VERSUS CONCENTRATIONS FOR STANDARD CURVE EXPERIMENT

Fig. 2. "L" is a measure of lightness and varies from 100 for perfect white to zero for black; "a" indicates redness when plus, gray when zero, and greenness when minus; and "b" indicates yellowness when plus, gray when zero, and blueness when minus. As shown in Fig. 2, the "L" values decrease from 69 to 35 as the color concentrations increase from 0% to 0.6%. The "a" values increase from 0 to 50 as the color concentration increases. The "b" values vary between 14 and 17. The results indicate that the extrudate became darker and redder as the red dye concentration was increased. Figure 2 also shows that "a" color values changes more siqnificantly with red dye concentration than either "L" or "b" color values. Therefore, the "a" color curve in Fig. 2 was chosen as the standard curve. Second-order polynomial equations were developed and a Fortran 77 program was written to convert the redness "a" color values to concentrations (g color/100 g rice flour).

Deviation of Redness Color Value Approach from Concentration Approach

The colorimetric method is frequently used to measure the residence time distribution in the extrusion cooking system. Based on the definition of the mean residence time and the spread of the residence time distribution, the concentration approach should be used to determine the RTD. However, the redness color value of the extrudate directly from a colorimeter was also used by many researchers (Altomare and Anelich 1988; Altomare and Ghossi 1986; Chen *et*

al. 1990; Likimani *et al.* 1990; Onwulata 1991). Likimani *et al.* (1990) presented RTD E curves directly using the redness color values (a) versus residence time. Some researchers calculate the E(t) in Eq. (1) using redness color values rather than concentration, and the E(t) versus residence time is plotted as the E curve (Altomare and Anelich 1988; Altomare and Ghossi 1986; Chen *et al.* 1990; Onwulata 1991). All the 3 × 3 factorial experiments were used to investigate the deviation.

In the concentration approach, the concentrations of Eq. (3) and (4) were obtained from the standard curve by converting the redness color values (a) to color concentrations for each sample. The "C" of Eq. (3) and (4) was replaced by color value "a" for the redness color value approach.

The mean residence time ($\bar{t}$) and the spread of the residence time distribution (σ) for each treatment using the two approaches are shown in Table 1 and 2.

TABLE 1
THE MEAN RESIDENCE TIMES t (s) OF THE REDNESS COLOR VALUE AND CONCENTRATION APPROACHES

Feed rate (kg/h)	Screw speed (rpm)	Redness color value approach	Concentration approach
30	200	113.19±9.80	82.05±1.44
	300	93.72±9.30	73.15±2.18
	400	81.69±9.33	69.21±0.99
40	200	97.42±0.48	65.75±0.19
	300	87.86±2.16	58.03±2.15
	400	73.40±3.00	53.98±1.43
50	200	85.80±1.77	57.60±0.97
	300	65.46±8.65	49.17±1.13
	400	62.07±10.4	45.97±0.20

The mean residence time ($\bar{t}$) calculated from the concentration approach was lower than that calculated from the redness color value approach. The average mean residence time in the 3 × 3 factorial experiment for redness color value and concentration approaches were 84.50 s and 61.66 s, respectively. There was a significant difference in the mean residence time between the two approaches ($p < 0.0001$). The average RTD spreads in the 3 × 3 factorial

TABLE 2
THE RTD SPREADS σ (s) OF THE REDNESS
COLOR VALUE AND CONCENTRATION APPROACHES

Feed rate (kg/h)	Screw speed (rpm)	Redness color value approach	Concentration approach
30	200	54.78±8.53	24.55±1.98
	300	42.68±12.4	19.76±0.28
	400	37.05±15.2	22.71±5.53
40	200	50.17±0.83	14.12±0.12
	300	51.61±2.85	16.60±2.99
	400	39.33±3.27	16.66±1.10
50	200	46.39±0.50	12.65±0.83
	300	32.12±10.3	14.58±0.27
	400	39.81±16.3	16.97±1.90

experiments were 17.62 s and 43.77 s for the concentration and redness color value approaches, respectively. Also, there was a significant difference in the spread of the residence time distribution (σ) calculated by the two approaches ($p < 0.0001$). When the feed rate was fixed, the concentration approach provided a lower mean residence time ($\bar{t}$) and a lower RTD spread (σ) than the redness color value approach ($p < 0.0001$). The same results were achieved when the screw speed was fixed ($p < 0.0001$). The mean residence time and the spread of the residence time distribution for the redness color value approach were 18–51% and 63–267%, respectively, more than the concentration approach.

The E curves of 30 kg/h feed rate and 200 rpm screw speed, 40 kg/h and 300 rpm, and 50 kg/h and 400 rpm using concentration and redness color value approaches are shown in Fig. 4 through 6. All the E curves plotted by the concentration approach were sharper than these plotted by the redness color value approach, indicating that the spread of the residence time distribution in the concentration approach was smaller than that in the redness color value approach.

For the concentration approach, the concentration from a standard curve is used to calculate the E(t), $\bar{t}$, and σ. For the redness color value approach, the redness color value is used instead to calculate the E(t), $\bar{t}$, and σ. Comparing the two approaches, if the redness color value can be used instead of concentration to calculate the characters of the RTD, it is necessary that no significant differences in the mean residence time and the RTD spread exist between the two approaches. The relationship between color concentration and redness color value must linear as well. However, the results described above showed that

there were significant differences in the mean residence time and the RTD spread between the two approaches. The results in Fig. 3 also show that the relationship between concentration and redness color value is not linear, with the standard curve possessing a concave shape. The standard curve can be studied in two portions: where slopes are greater than 1 and where less than 1. In the portion where slopes of the standard curve are less than 1, i.e., the ratios of concentration to redness color value are less than 1, the redness color value approach will overestimate the E(t). In the portion where slopes of the standard curve are greater than 1, the redness color value approach will underestimate the E(t). The phenomena agree with the shape of E curves shown in Fig. 4 through 6. Both E curves using different approaches show that most of the E(t) in the redness color value approach were greater than those in the concentration approach. Thus, the results of the mean residence time and the RTD spread in the redness color value approach were greater than that in the concentration approach, as shown in Tables 1 and 2. Therefore, it can be seen that the redness color value approach overestimates the mean residence time and the spread of the residence time distribution, and is thus an inaccurate and unacceptable approach for the RTD determination in the extrusion system. According to the results shown in this study, the concentration approach is recommended for the colorimetric RTD determination method instead of redness color value approach for the extrusion cooking system.

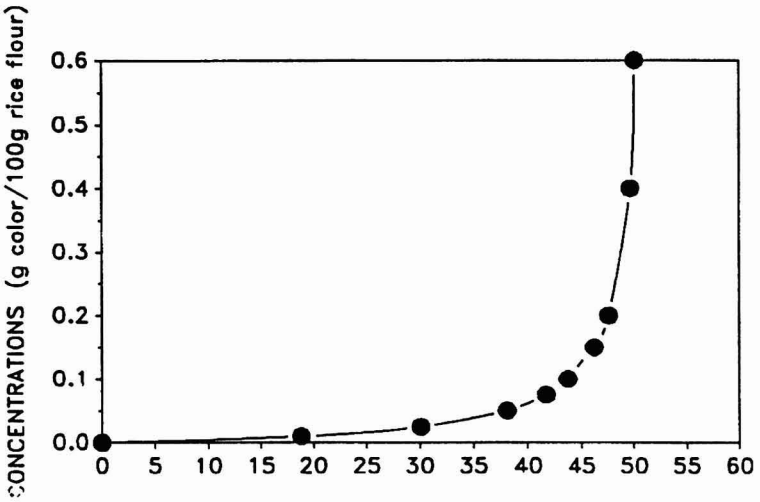

FIG. 3. STANDARD CURVE CONCENTRATIONS VERSUS REDNESS COLOR VALUES

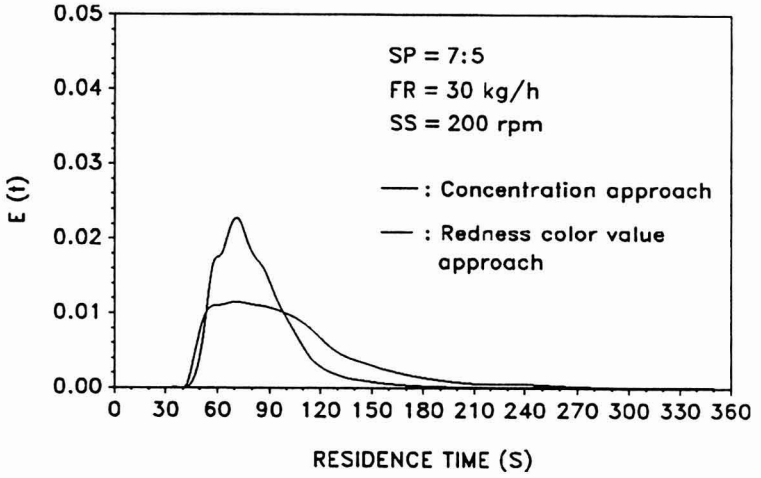

FIG. 4. E CURVES OF DIFFERENT COLORIMETRIC APPROACHES AT 30 kg/h FEED RATE AND 200 RPM SCREW SPEED

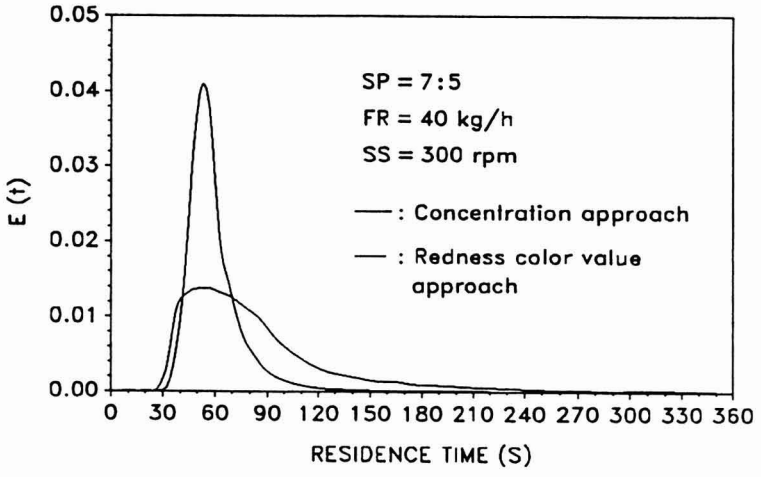

FIG. 5. E CURVES OF DIFFERENT COLORIMETRIC APPROACHES AT 40 kg/h FEED RATE AND 300 RPM SCREW SPEED

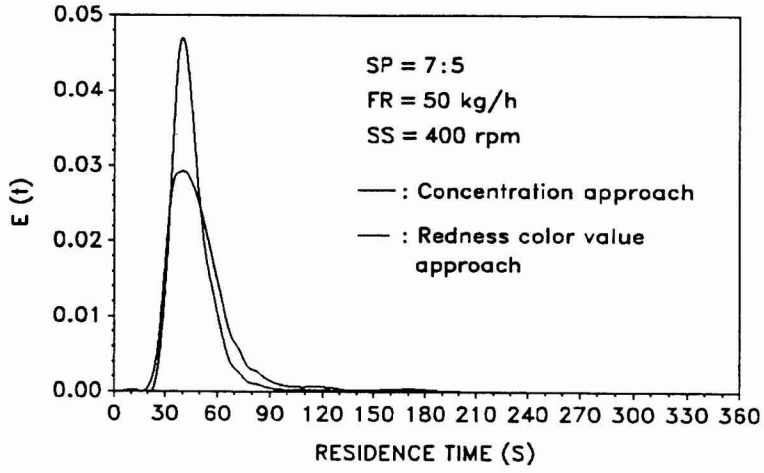

FIG. 6. E CURVES OF DIFFERENT COLORIMETRIC APPROACHES AT 50 kg/h FEED RATE AND 400 RPM SCREW SPEED

It should be noted that the standard curve of the color values versus concentration was established at 20% moisture, 40 kg/h feed rate, and 300 rpm screw speed. One may question the validity of using the same standard curve for extrudates obtained from different moisture contents, feed rates, or screw speeds. While the extrudate color indeed would have varied slightly with the extrusion conditions (Hsieh *et al.* 1990), the redness color value in this study was contributed from the red dye added, not from the corn meal. Thus, all the residence time distribution curves had no redness in the first 20 s or after 360 s where only corn meal was extruded. A random check of the color value at 200 and 400 rpm screw speeds as well as at 30 and 50 kg/h feed rates did not reveal any difference in the standard curve. It appeared that within the feed rate and screw speed ranges studied, the standard curve obtained at the 20% moisture, 40 kg/h feed rate, and 300 rpm screw speed could be used.

CONCLUSION

The colorimetric method is very useful in determining the residence time distributions of rice flour in the twin-screw extrusion system. The redness color value and color concentration approaches were used to determine the RTDs and calculate the mean residence time ($\bar{t}$) and spread of the residence time distribution (σ). There are significant differences between the two approaches, and it

was shown that using the redness color values (a) from a colorimeter directly to study the RTD tend to overestimate the mean residence time and the RTD spread. The concentration approach is the valid and correct method for RTD measurement in a twin-screw extrusion system.

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MODIFIED ATMOSPHERE PACKAGING OF SWEET CORN ON COB

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ABSTRACT

Respiration rates (RR) of two varieties of sweet corn on cob were determined at O_2 concentrations: 5, 10, 15, and 21% and CO_2: 0, 5, and 10%, and temperatures: 2, 12, and 25C. Temperature and O_2 had strong influence on RR. Activation energies for effect of temperature on RR(O_2) and RR(CO_2) were, 44.6 and 104.4 kJ/mole, respectively. At 12C, to maintain 4% O_2 and 6% CO_2 in a package 10.8 cm^2 area, with 1.12 kg of corn, the estimated film permeabilities were 14 ml $O_2/h.cm^2$ and 65 ml $CO_2/h.cm^2$, respectively. Experimental and theoretical transient gas concentrations using microporous films were in reasonable agreement, but the desired O_2 and CO_2 levels were not achieved.

INTRODUCTION

In modified atmosphere packaging (MAP), the desired gas composition in a package is achieved gradually through passive modification of the atmosphere, usually using suitable plastic films; the actively respiring and metabolizing product reduces O_2 and increases CO_2 levels by restricting the exchange of air between the inside of the package and the environment outside (Chinnan 1989).

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MAP can lower respiration and/or delay ripening while avoiding anaerobic processes in fruits and vegetables. MAP is achieved either by reducing the concentration of O_2, which is required for respiration, or by increasing the concentration of an inhibitory gas such as CO_2 (Chinnan 1989; Day 1989a,b; Kader 1987; Kader *et al.* 1989; Labuza and Breene 1989; Shewfelt 1986). However, it is emphasized that in MAP the desired gas concentrations cannot be achieved instantaneously.

The development of MAP has faced several problems (Kader 1987; Kader *et al.* 1989): lack of respiration data at different temperatures and gas concentrations, lack of consistency in respiration data gathered for the same commodity, lack of permeability data of packaging materials at different temperatures and relative humidities, research needed to elucidate the additive effects of reduced oxygen and increased CO_2 in respiration and ethylene production, and more information on the use of gas absorbers and scavengers to sustain a desired atmosphere in packages. At present, design of the best MAP for a certain product is still an empirical process, specific only for that commodity.

For medium and high respiring products like lettuce, cauliflower, broccoli, mushrooms and spinach, commonly available films such as low density polyethylene, plasticized polyvinyl chloride, and polypropylene may not be ideal due to their low gas transmission rates. Microporous and microperforated films with 0.16–0.32 cm dia. holes to allow ventilation also have been employed. Others have developed the idea of using a breathable window in a package made of these films; the size of the window used to control gas exchange is matched to the respiration of the produce (Anon. 1991).

Mathematical models can reduce time and resources in optimizing a MAP system. The main limitation to modeling is the availability of respiration rate data (Kader *et al.* 1989). Another limitation is the availability of these data in the form of equations for the calculation of transient and steady state gas concentrations, and film permeabilities. Usually, respiration rates have been gathered at only one temperature and gas composition, but in order to predict gas compositions in a package, respiration rates are needed at several values of temperatures and gas concentrations. In particular, because O_2 and CO_2 concentrations change from ambient values to substantially different final values, it would be desirable to determine the respiration rates over a wide range of gas concentrations. Mathematical equations that describe respiration as a function of the variables affecting respiration rate are required. Film permeabilities also have been determined at only one temperature, usually, at 23–25C; however, in MAP system, lower temperatures (0–5C) are required for shelf-life extension and permeability values at these temperatures are needed (Zagory and Kader 1989; Zagory *et al.* 1989). Retail packages of produce are subjected to temperatures in the range of about 4–25C and the compositions of O_2 and CO_2

from ambient to the typical recommended 4–6% O_2 and 6–8% CO_2 (Kader *et al.* 1985). Therefore, it would be desirable to obtain produce respiration rate and film permeability data over those ranges of temperatures and gas compositions. Further, mathematical models of the data permit computer calculation of transient gas concentrations and the time required to achieve steady state in the packages (Hayakawa *et al.* 1975; Zagory *et al.* 1989; Morales-Castro 1992).

The objectives of this work were: (1) To determine respiration rates of sweet corn at different gas concentrations and temperatures and to determine the effect of variety of corn. (2) To develop predictive models for respiration rates of sweet corn from the data obtained in objective 1. (3) To compare predicted transient gas concentrations with experimental data obtained in packaged sweet corn.

MATERIALS AND METHODS

Sweet corn was provided by Turek Farms, King Ferry, NY. Two varieties were used: white "Silverado" (sugar enhanced) and "yellow" (super sweet). Ears were selected at fieldside from freshly harvested corn, before hydrocooling, placed in coolers with ice, and brought to the laboratory where they were dehusked and kept at low temperatures until their use, usually the same day.

Respiration Rates

Respiration rates were determined using a combination of pseudo-flow-through and closed system methods (Jurin and Karel 1963) using one gallon glass jars. The metal covers were modified to replace gases and for sampling. One corn ear was placed in a jar, and the jar was weighed and closed tightly. The gas composition inside the chamber was regulated by applying ideal gas law, i.e., assuming partial pressure to be proportional to gas concentration. The chambers were flushed with nitrogen for 5 min in order to ensure an oxygen free atmosphere. A vacuum was drawn on the chamber that was proportional to the required gas concentration using a vacuum pump, and the gas under consideration was admitted into the chamber until its pressure corresponded to the desired volumetric composition. The atmospheric composition inside jars was checked with a chromatograph (Gow Mac, Series 580, Bridgewater, NJ), equipped with a thermal conductivity detector and a CTR 1 column (Alltech, Deerfield, IL) and the jars were stored at different temperatures. Samples were taken right after flushing and at 20 h, intervals for 7–9 days. A complete factorial design with 3 levels was selected. The design included 3 × 3 × 3 level with 3 × 1 control at atmospheric conditions. Gas

concentrations employed were, O_2: 5, 10, 15, and 21% and CO_2: 0, 5, and 10%. Nitrogen was used for balance to 100%. Temperatures employed were: 2, 12, and 25C.

Respiration rates were calculated using the equation:

$$RR = \frac{(C_2 - C_1) * Vol}{W * t} \quad (1)$$

where, RR is respiration rate expressed as ml of O_2 or CO_2/kg. h; C_1 and C_2 are the gas concentrations (fractions) immediately following atmospheric modification in the jars and after about 24 h, respectively, Vol is free volume in the jars, W is weight of commodity, and t is elapsed time between sampling, h.

Statistical analysis of respiration rate data was performed using multiple regression analysis in General Linear Models procedure (GLM) (SAS Institute Inc., Cary, NC) and SYSTAT (Systat Inc., Evanston, IL); SYSTAT (Systat Inc., Evanston, IL) was used to draw three dimensional figures. The optimization program in GENSTAT (Numerical Algorithms Group, Oxford, UK) was used to fit nonlinear equations that included Arrhenius temperature dependence.

Film Permeabilities

Microporous films provided by Cryovac (Duncan, SC): SM-250 and SM-570 were tested. No specifications regarding materials were provided by the manufacturer. A permeability cell was used to measure gas permeabilities using the standard method (ANSVASTM D 1434-75). In order to get permeability data at several fixed temperatures, the experiments were carried out in rooms with controlled temperatures. Because microporous films had high permeabilities, the exposed area of transmission was reduced by placing a metallic plate (area 0.7 cm^2) (Morales-Castro 1992).

Mathematical Modeling

The mass balance differential equations that describe the evolving atmosphere in a modified atmosphere packaging system are:

$$\frac{dc(O_2)}{dt} = \{K_O A[0.21 - c(O_2)] / V\} - R_O W / V \quad (2)$$

$$\frac{dc(CO_2)}{dt} = \{-K_c(CO_2)/V\} + R_C W/V \quad (3)$$

These equations were solved simultaneously using Runge-Kutta-Verner sixth order numerical method (IMSL, Houston, TX) to obtain the concentrations of gases at any time by using an appropriate model for respiration as a function of time (t), initial concentrations of O_2 (0.21) and CO_2 (0.0), permeabilities of film (K_o and K_c), free volume inside the package (V), W is weight of produce, and area of gas transmission (A). The computer programs were tested for their predictive capabilities by solving equations with known solutions.

Packaging Studies

One gallon glass jars were used as the main package. The screw top lids were modified to test different films. Two holes, 2.56 cm in diameter, were cut in the metallic cover and the plastic film under consideration was attached to the cover using silicon adhesive. In some experiments both windows were covered by the film while in others one of the windows was covered with aluminum foil to reduce transmission area. Several ears of corn were placed inside the jars, the jars were weighed and sealed, and stored at different temperatures. Samples of O_2 and CO_2 were taken at time intervals of about 3 h until equilibrium concentrations were reached.

RESULTS AND DISCUSSION

Using GLM procedure in SAS, the effect of O_2 and CO_2, storage time (t), and temperature (T) on respiration rates was investigated. The coefficients were evaluated at $p < 0.001$. The regression equations obtained were:

$$RR\ (O_2) = 111 * O_2 + 5.8 \times 10^{-1} * T + 1.0 \times 10^{-1} * t + 16.34 * 0_2 * T - 8.0 \times 10^{-1} * 0_2 * t - 2.8 \times 10^{-3} * T * t \qquad R^2 = 0.94 \quad (4)$$

$$RR\ (CO_2) = 17.35 + 62.5 * O_2 + 1.84 * T - 8.6 \times 10^{-2} * t + 4.4 \times 10^{-4} * T^2 + 5.5 \times 10^{-4} * t^2 + 10.2 * O_2 * T + 3.14 * CO_2 * T - 1.2 \times 10^{-4} * T * t \qquad R2 = 0.96 \quad (5)$$

where RR is respiration rate in ml O_2/kg.h or ml CO_2/kg.h.

Oxygen Uptake

Effect of O_2. Oxygen concentration had a large influence on respiration. Figures 1 and 2 illustrate data at 2C and 10C after 96 h. It can be seen that as oxygen concentration increased, respiration rate also increased. The effect of oxygen concentration was similar at 2 and 10C; at 2C, respiration rate increased from 10 to 30 ml O_2/kg.h when oxygen was increased from 5 to 21%, while at 10C it increased from 20 ml O_2/kg.h at 5% to 60 ml O_2/kg.h at 21%. These values represent a three-fold increase in respiration rates at the minimum and maximum oxygen concentration values of 5 and 21 %. Generally, lower oxygen atmospheres are recommended to lower respiration rates, but the risk of developing anaerobic respiration at these low concentrations is very high.

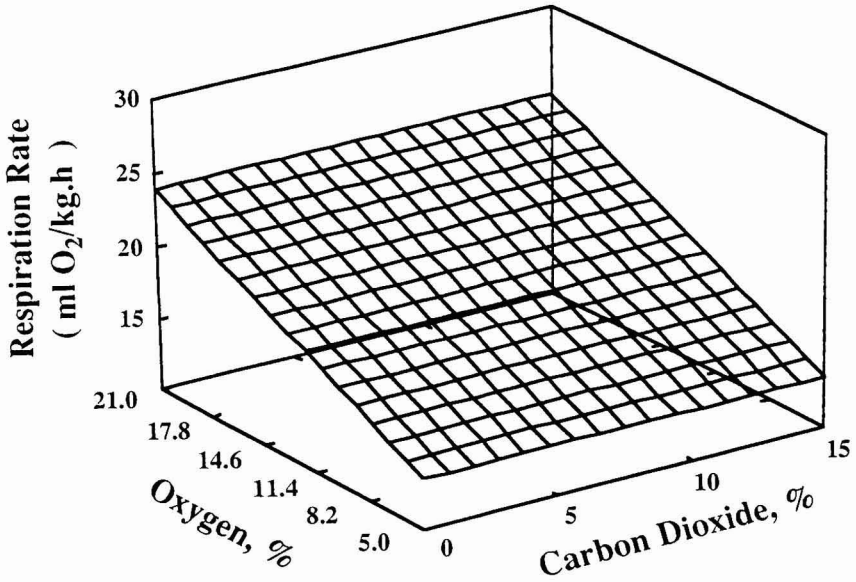

FIG. 1. EFFECT OF OXYGEN AND CARBON DIOXIDE ON RESPIRATION RATE (ml O_2/kg.h) OF SWEET CORN AT 2C AFTER 96 h

Effect of CO_2. The influence of concentration of CO_2 on $RR(O_2)$ was found to be negligible (Fig. 1 and 2). Cameron (1989) suggested that CO_2 does not have an effect on oxygen uptake of red tomato fruit. However, Jurin and Karel (1963) reported a slight decrease in the respiration rate of apples with an increase in CO_2. Hayakawa *et al.* (1975) concluded that the rate of oxygen

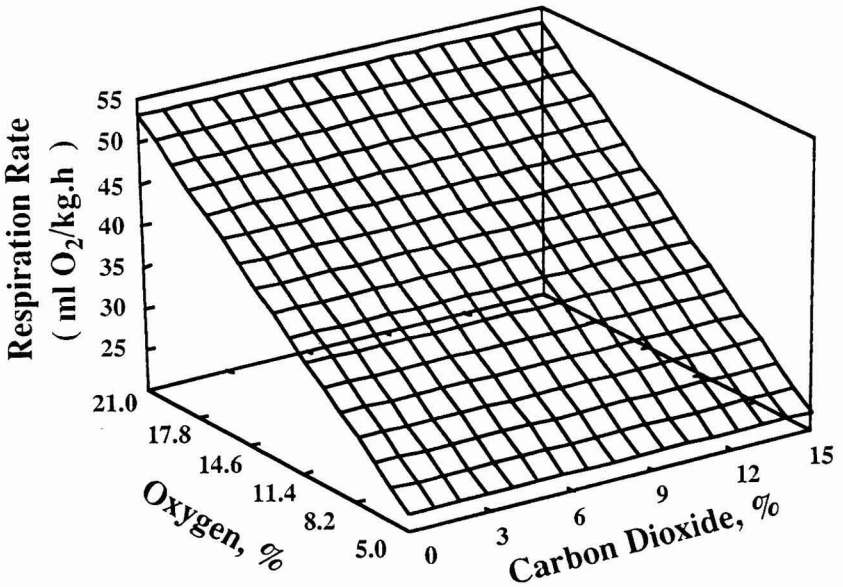

FIG. 2. EFFECT OF OXYGEN AND CARBON DIOXIDE ON RESPIRATION RATE (ml O_2/kg.h) OF SWEET CORN AT 10C AFTER 96 h

consumption was not greatly influenced by CO_2 concentration. Henig and Gilbert (1975) found a decrease in the respiration rate (CO_2 generated) when CO_2 was increased above 10%. Lee *et al.* (1991) found that the respiration rate of broccoli was inversely proportional to CO_2 concentration.

Effect of Storage Time. As Eq. (4) and (5) indicate, storage time affected respiration rates, especially at high temperatures and high oxygen concentrations (not shown). At 2C, respiration at 96 h was slightly higher than at 24 h. Similar results were obtained at 10C. Chinnan and Pendalwar (1989) observed a steady decline in oxygen consumption and CO_2 evolution rates with increasing storage time at high oxygen concentrations (20% oxygen) where RR dropped from 25 ml/kg.h to about 10 ml/kg.h.

Effect of Temperature. Figures 1 and 2 also indicate the effect of temperature on respiration rate. Our results are in agreement that for every 10C rise in temperature, respiration rates are doubled (Q_{10} value) (Kader *et al.* 1989). Arrhenius activation energy for RR(O_2) was estimated by nonlinear regression analysis of experimental data to be 44.6 kJ/mole. Relatively few respiration

studies have been conducted at different temperatures: Gagnon *et al.* (1991) obtained respiration rates for mushrooms at three temperatures: 0, 7 and 14C and Kader (1989) reported respiration studies for several commodities (green beans, broccoli, chili pepper, mango, and tomato) at different temperatures and gas compositions.

Carbon Dioxide Released

Effect of Oxygen. Again, oxygen had a major impact on respiration as CO_2 generated. Values for respiration at 2C were 23 and 36 ml CO_2/kg.h at 5 and 21% O_2, respectively; at 10C, the values were 44 and 70 ml CO_2/kg.h for the same oxygen concentrations (Fig. 3 and 4). Because oxygen provides the energetic requirements for metabolic reactions, if its concentration is lowered, all the processes taking place in the living cells slow down, since aerobic respiration is affected.

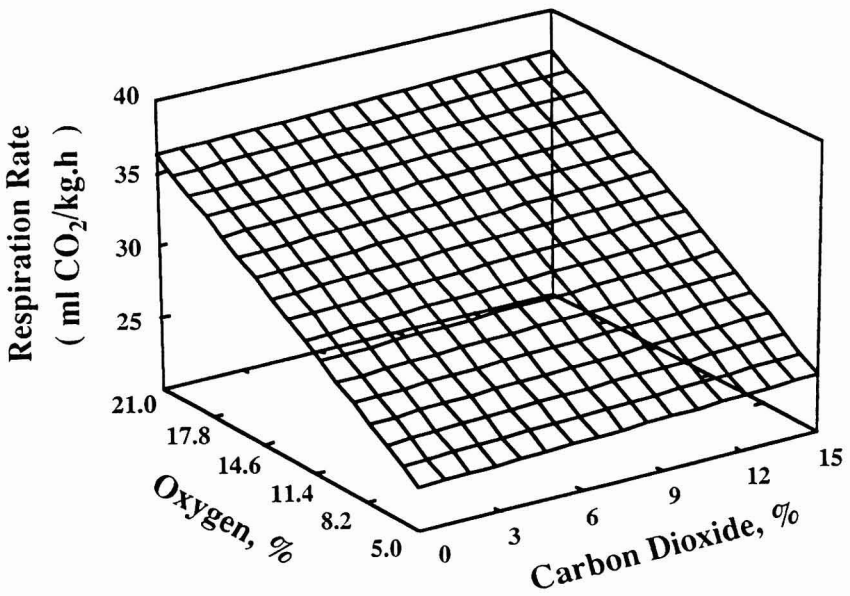

FIG. 3. EFFECT OF OXYGEN AND CARBON DIOXIDE ON RESPIRATION RATE (ml CO_2/kg.h) OF SWEET CORN AT 2C AFTER 24 h

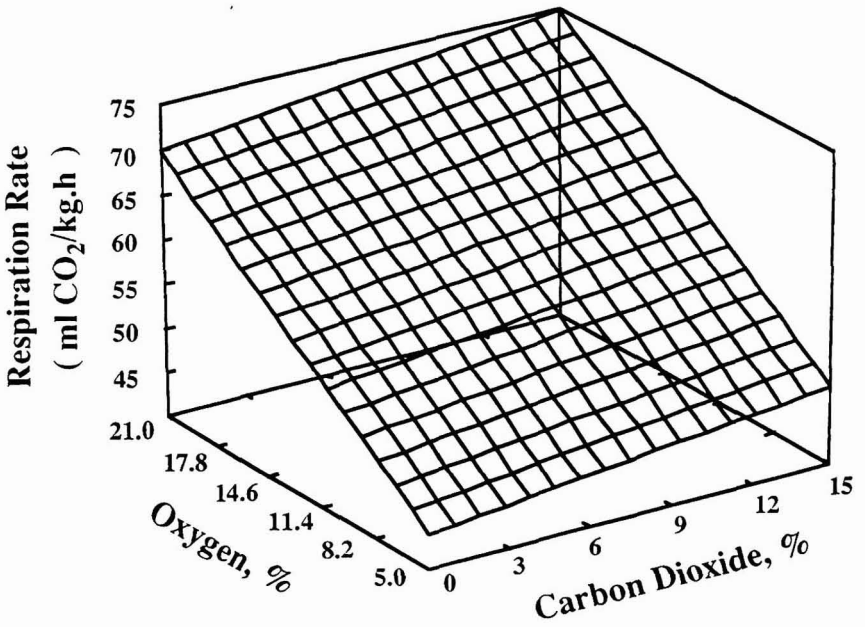

FIG. 4. EFFECT OF OXYGEN AND CARBON DIOXIDE ON RESPIRATION RATE (ml CO_2/kg.h) OF SWEET CORN AT 10C AFTER 24 h

Effect of CO_2. Carbon dioxide concentration had a negligible effect on respiration, as indicated in Eq. 5. RR(CO_2) increased one unit when CO_2 was increased from 0 to 15% at 2C. At 10C, RR(CO_2) increased five units. In contrast to our study, some studies in other commodities showed that increasing CO_2 decreased the respiration rate.

Effect of Storage Time. Storage time was more important for RR(CO_2) than for RR(O_2). There was a 20% decrease in respiration (from 114 to 86 ml CO_2/kg.h) at 10% O_2 concentration, between 24 h and 120 h at 25C. A similar effect was observed at 21% O_2 where respiration decreased from 149 to 120 CO_2/kg.h at 25C. The effect of storage time on respiration at low temperatures (2C) was found to be negligible at 10% and 21% O_2 concentrations. The decrease in respiration rate with storage time could be attributed to the depletion of nutrients so that as the reserves are being consumed limited amounts of nutrients are available.

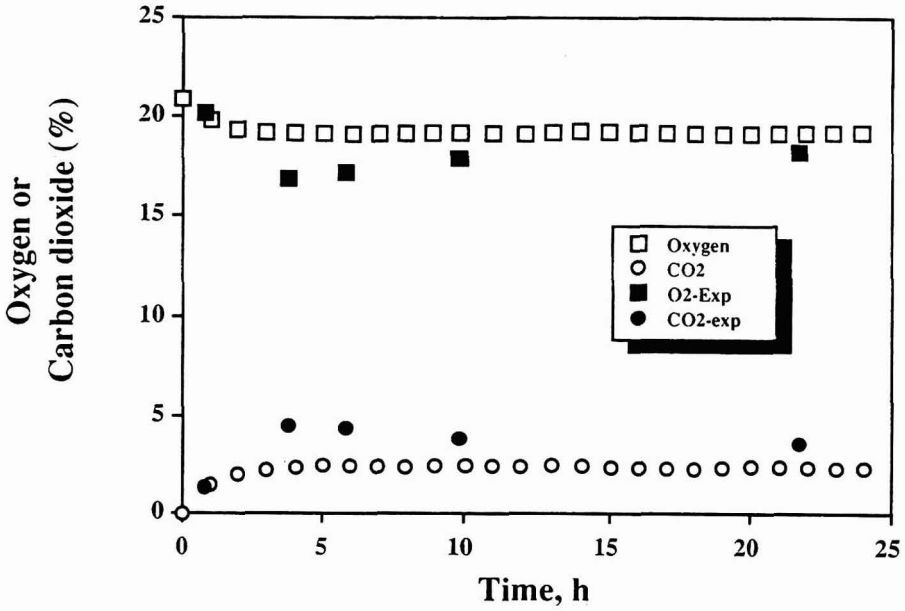

FIG. 5. PACKAGING STUDIES WITH SWEET CORN AT 2C SIMULATING A PACKAGE WITH 6 CORN EARS (1.05 kg), SM-250 MICROPOROUS FILM, 5.4 cm^2 AREA, AND 1.92 L VOLUME
Open symbols represent computer simulations and closed symbols represent experimental data

Effect of Temperature. The effect of temperature followed the rule that for every 10C increase, respiration rate doubled. At 2C and storage time of 24 h, respiration rate was 24 ml CO_2/kg.h while at 25C the respiration rate was 114 ml CO_2/kg.h. The same trend was observed after storage for 120 h where respiration rates were 24 ml CO_2/kg.h at 2C and 86 ml CO_2/kg.h at 25C. King and Bolin (1989) and Labuza and Breene (1989) reported that Q_{10} is about 2–3 for fruits and vegetables so the values obtained here are in agreement. The Arrhenius activation energy for $RR(CO_2)$ was determined by nonlinear regression analysis to be 104.4 kJ/mol, a value more than twice the activation energy for $RR(0_2)$. Therefore, increasing storage temperature affected rate of release of CO_2 more than rate of consumption of O_2.

Effect of Variety of Sweet Corn on Respiration Rate

Two different varieties were tested: white sugar enhanced sweet corn and yellow super sweet referred to as white and yellow, respectively. Respiration

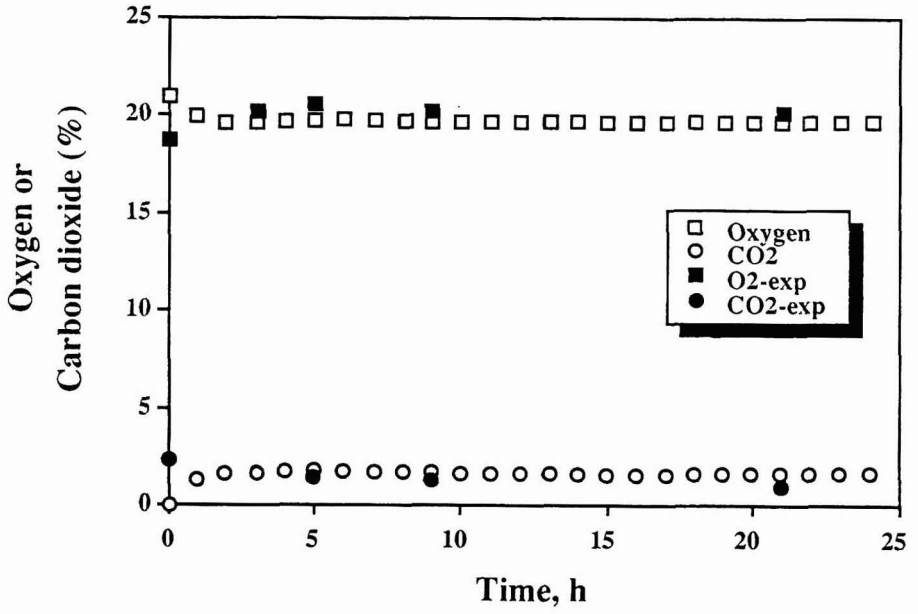

FIG. 6. PACKAGING STUDIES WITH SWEET CORN AT 2C SIMULATING A PACKAGE WITH 6 CORN EARS (1.14 kg), SM-570 MICROPOROUS FILM, 5.4 cm^2 AREA, AND 1.92 L VOLUME
Open symbols represent computer simulations and closed symbols represent experimental data.

rates were measured at 21% O_2, at 2, 4, 12, and 25C over a period of four days. Multiple regression analysis using SYSTAT indicated that there was no significant difference between the respiration rates of the two varieties. Variations in respiration with variety of a vegetable are due to differences among the plant parts, in the surface area-to-volume ratio, and in the nature of their surface coatings that influence their gas diffusion characteristics (Kader 1987).

Unsteady State Gas Composition and Packaging Studies with Sweet Corn

Assuming steady state conditions in Eq. (2) and (3), film permeabilities were estimated for specific weights of corn, temperature, and desired O_2 and CO_2 concentrations. Because respiration rates depended on time and gas

concentrations, a range of permeability values were obtained (Morales-Castro 1992). As an example, at 12C, to maintain 4% O_2 and 6% CO_2 in a package 10.8 cm^2 area, with 1.12 kg of corn, the estimated film permeabilities were 14 ml $O_2/h.cm^2$ and 65 ml $CO_2/h.cm^2$, respectively.

Recommended gas compositions for maximizing shelf-life of vegetables are typically 4–6% O_2 and 6–8% $C0_2$ (Kader *et al.* 1985). Based on permeability calculations and earlier studies (Day 1989a; Rowan 1989; McLachlan and Stark 1985), plastic films with high permeabilities were necessary to obtain the suggested gas composition. Because microporous films SM-250 and SM-570 provided by Cryovac (Duncan, SC) had high permeabilities, they were used for packaging studies; their O_2 and CO_2 permeabilities were determined to be 300 $ml/cm^2.h$ and 465 $ml/cm^2.h$; there was negligible difference in film permeabilities for O_2 and CO_2.

Equations (2) and (3) were solved numerically to obtain the concentrations of O_2 and CO_2 at any time by using appropriate models for $RR(O_2)$ and $RR(CO_2)$, and the initial concentrations of oxygen (0.21) and carbon dioxide (0.0). Input variables were: temperature, weight of produce, free volume of the package, area, and permeabilities of the film to oxygen and carbon dioxide.

The predicted and experimental values of gas concentrations using the films SM-250 and SM-570 with six ears of corn at 2C are shown graphically in Fig. 5 and 6, respectively. In Fig. 5, concentration of O_2 decreased at the beginning and then increased until an equilibrium concentration was reached; an opposite behavior could be observed for carbon dioxide. These deviations are attributed to the limits of applicability of the mathematical model. In general, there was a close agreement between experimental and predicted values. However, using the microporous films, concentration of O_2 in the desired range of 4–6% could not be obtained.

CONCLUSION

Respiration rates of sweet corn on cob were very high and strongly depended on O_2 and CO_2 concentrations, and temperature; to a lesser extent they were also influenced by storage time. For MAP of corn, films with high O_2 and CO_2 permeabilities would be necessary. Experimental and theoretical gas concentrations in microporous film packages containing corn were in reasonable agreement. However, because of the very high film permeabilities, the desired steady state O_2 and CO_2 concentrations could not be obtained.

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MODIFIED ATMOSPHERE PACKAGING OF HEAD LETTUCE

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ABSTRACT

*Magnitudes of respiration rates of lettuce heads at O_2 levels: 5, 10, 15, and 21%, and three temperatures: 2, 12, and 25C ranged between 3 to 30 ml O_2/kg.h. Oxygen concentration (O_2). temperature (T), storage time (t), and the interactions between temperature and O_2 ($T*O_2$) and temperature and time (T*t), affected respiration rates. Predicted and experimental O_2 and CO_2 concentrations in packages using two plastic films were in good agreement with each other. However, steady state gas concentrations were achieved after about 90 h.*

INTRODUCTION

There are several categories into which fresh items are classified on the basis of respiration rates (RR): low, medium and high respirers. For example, products with long shelf-lives are usually low respirers, as in the case of potatoes, dry onions, cabbage and carrots. Medium respirers include products such as cauliflower and lettuce (c.v. romaine), while high respiring products correspond to the most perishable category such as mushrooms, broccoli and spinach. Head lettuce (*Lactuca sativa* L) is considered to be a low-respiring product but after slicing, its RR increases considerably and enzymatic reactions such as browning are accelerated with considerable reduction in shelf-life (McDonald *et al.* 1990). Ballantyne (1989) described a successful modified atmosphere packaging (MAP) of shredded lettuce where shelf-life was doubled

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with an equilibrium atmosphere in the package of 2–3% O_2 and 4–6% CO_2. Zagory and Kader (1989) described the factors affecting quality of vegetables in controlled atmosphere and Zagory *et al.* (1989) described the use of computer models to design MAP of vegetables. However, it is emphasized that the desired gas concentrations in MAP cannot be achieved instantaneously, and it is important to know the time required to achieve steady state.

At present, design of the best MAP of a given product is still an empirical process, specific only for that commodity. Respiration rate data is essential for the design of MAP, especially in the form of equations that can be used in the calculation of transient and steady state gas concentrations, and film permeabilities. Earlier, we (Morales-Castro *et al.* 1994) reported RR and MAP of sweet corn on cob, and reviewed studies on MAP of vegetables. In order to predict transient gas compositions in a package, respiration rates are needed at several values of temperatures and gas concentrations. In particular, because O_2 and CO_2 concentrations change from ambient values to substantially different final values, it would be desirable to determine the respiration rates over a wide range of gas concentrations. The objectives of this work were: to determine RR of lettuce at different conditions of gas concentrations and temperatures, to develop predictive models for RR of lettuce, and to compare predicted transient gas concentrations with experimental data obtained in packaged head lettuce.

MATERIALS AND METHODS

Iceberg lettuce grown in Florida was provided by the J.C. Brock Corp. (Buffalo, NY). The samples were brought to the laboratory in containers packed with ice. Respiration rates were determined employing a combination of pseudo-flow-through and closed system methods (Jurin and Karel 1963) using glass jars at four different oxygen levels: 5, 10, 15, and 21% and at three temperatures: 2, 12, and 25C as described elsewhere (Morales-Castro 1992). The initial concentration of CO_2 was 0%. The gas composition inside jars was measured with a chromatograph (Gow Mac, Series 580, Bridgewater, NJ), equipped with a thermal conductivity detector and a CTR 1 column (Alltech, Deerfield, IL) (Morales-Castro 1992).

Respiration rates were calculated using the equation:

$$RR = \frac{(C_2 - C_1) * Vol}{W * t} \quad (1)$$

where, RR is respiration rate expressed as ml of O_2 or CO_2/kg. h; C_1 and C_2 are the gas concentrations (fractions) immediately following atmospheric modification in the jars and after about 24 h, respectively, Vol is free volume in the jars, W is weight of commodity, and t is elapsed time between sampling, h.

Statistical analysis of RR data was performed using multiple regression analysis in General Linear Models procedure (GLM) in SAS (SAS Institute Inc., Cary, NC) and SYSTAT (Systat Inc., Evanston, IL) was used to fit RR data and to draw the three dimensional figures. The optimization program in GENSTAT (Numerical Algorithms Group, Oxford, UK) was used to fit nonlinear equations that included Arrhenius temperature dependence.

Film Permeabilities

For MAP studies the films used were: Cryovac PD-941 of high oxygen and carbon dioxide permeabilities that was made from a multilayered polyolefin of 75 gauge, and a linear low density polyethylene (LLDPE) of 125 gauge. A permeability cell was used to measure gas permeabilities using the standard method (ANSI/ASTM D 1434-75) (Morales-Castro 1992). In order to get permeability data at several fixed temperatures, the experiments were carried out in temperature controlled rooms.

Mathematical Modeling

The mass balance differential equations that describe the evolving atmosphere in a modified atmosphere packaging system are:

$$\frac{dc(O_2)}{dt} = \{K_O A[0.21 - c(O_2)] / V\} - R_O W / V \quad (2)$$

$$\frac{dc(CO_2)}{dt} = \{-K_c(CO_2) / V\} + R_C W / V \quad (3)$$

These equations were solved simultaneously using Runge-Kutta-Verner sixth order numerical method (IMSL, Houston, TX) to obtain the concentrations of gases at any time by using an appropriate model for respiration as a function of time (t), initial concentrations of O_2 (0.21) and CO_2 (0.0), permeabilities of film (K_o and K_c), free volume inside the package (V), weight of produce (W), and area of gas transmission (A). The computer programs were tested for their predictive capabilities by solving equations with known solutions.

Packaging Studies

One gallon glass jars were used as the main package. The screw top lids were modified to test the two films. Two holes, 2.56 cm in diameter, were cut in the cover and the plastic film under consideration was attached to it using silicon grease.

RESULTS AND DISCUSSION

Respiration studies on lettuce heads were conducted at: 5, 10, 15, and 21% and at three temperatures: 2, 12, and 25C. The measured respiration rates were in the region of 3–30 ml O_2/kg.h and lower than values obtained earlier for sweet corn (Morales-Castro 1992). The following mathematical equations were fitted to the respiration data using the General Linear Model procedure of SAS and SYSTAT; the coefficients were evaluated at $p<0.001$.:

$$RRO2 = -2.28 + 19.34*02 + 7.3 \times 10^{-1}*T + 1.7 \times 10^{-2}*t + 1.70*02*T \times 2.0 \times 10^{-3} * T * t \qquad R2 = 0.96 \tag{4}$$

$$RRCO_2 = 4.48 \times 10^{-1} * O_2 + 6.62 \times 10^{-1}* T 5.0 \times 10^{-3} *t + 1.47*0_2 *T - 1.0 \times 10^{-3} * T * t \qquad R2 = 096 \tag{5}$$

where, RRO_2 is rate of uptake of O_2 and $RRCO_2$ is the rate of release of CO_2. In Eq. (4) and (5), it is readily seen that the variables that affected RRO_2 and $RRCO_2$ were oxygen concentration (O_2), temperature (T), storage time (t), and the interactions between temperature and O_2 ($T*O_2$) and temperature and time (T*t).

Response of Respiration Rate in ml O_2/kg.h to Different Variables

Effect of Oxygen and Temperature. Respiration rates were affected by both O_2 concentration and temperature. Figure 1 shows the effect of O_2 concentration and temperature on RRO_2. The influence of O_2 concentration on RRO_2 was higher at 25C than at 0C. After 24 h storage, at 25C, RR increased from about 20 to 30 ml of O_2/kg.h (50% increase) when O_2 concentration was

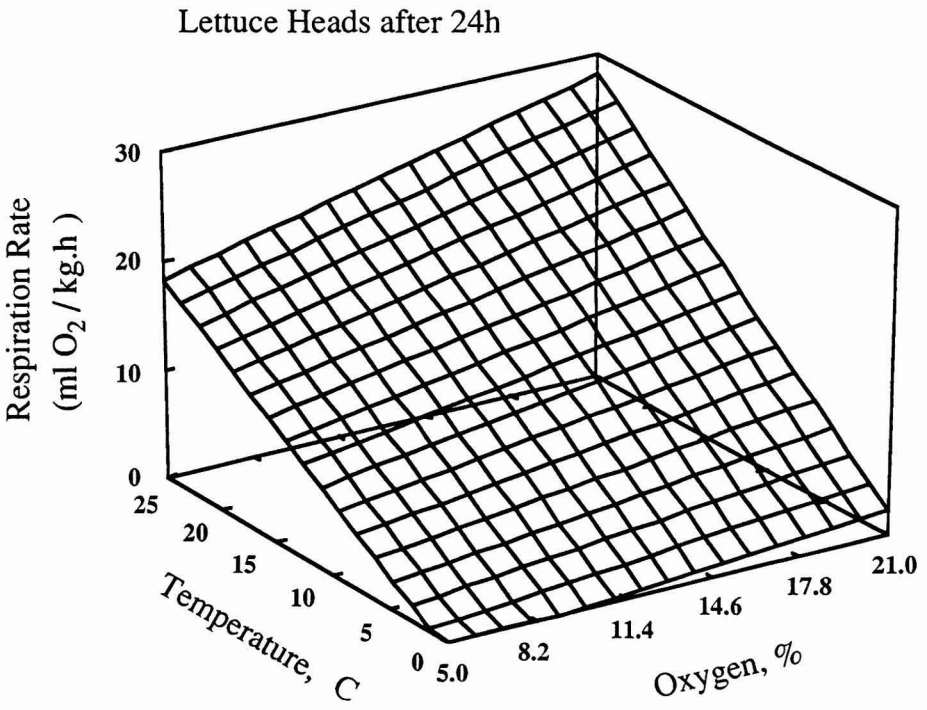

FIG. 1. EFFECT OF O_2 CONCENTRATION AND TEMPERATURE ON RRO_2 OF LETTUCE HEADS AFTER 24 h

increased from 5% to 21%. In contrast, at 0C, the increase in RR was 3 or 4 units only. Similar increase in RR with O_2 concentration was also observed after 144 h storage (not shown).

At 5% oxygen, when temperature was increased from 12 to 25C, RR increased from 8 to 18 ml O_2/kg.h (about a 125% increase) (Fig. 1). For the same increase in temperature but at an oxygen concentration of 21%, respiration increased from 11 to 28 ml O_2/kg.h (155% increase), so that Q_{10} was slightly above 2.55. The magnitude of activation energy was estimated to be 44.6 kJ/mole.

Effect of Time. RR decreased slightly with storage time at high temperatures while at low temperatures it seemed to increase. However, RR values at a temperature of 0C ranged from 1–5 RR units (not shown), so that this effect can be considered to be negligible; the same effect was observed at 10 and 21% oxygen concentration.

Response of Respiration Rate in ml CO_2/kg.h to Different Variables

Effect of Oxygen and Temperature. Oxygen had an insignificant effect on $RRCO_2$ at low temperatures; at higher temperatures, it caused only a slight increase in respiration rates. For example, at 5% oxygen concentration and at a temperature of 12C, RR was 9 ml CO_2/kg.h and at 21% oxygen, respiration increased by 3 units only. When temperature was increased to 25C, the values of respiration were 18 and 24 ml CO_2/kg.h for 5 and 21% oxygen concentration, respectively (not shown).

RR increased from 9 to 18 ml CO_2/kg.h when temperature was increased from 12 to 25C at 5% oxygen concentration. When oxygen was increased to 21%, RR increased from 12 to 24 ml CO_2/kg.h in the same temperature range (Fig. 2). These values represented a Q_{10} of 2 which agreed with values reported elsewhere (Kader *et al.* 1989). The magnitude of activation energy was estimated to be 57.1 kJ/mole.

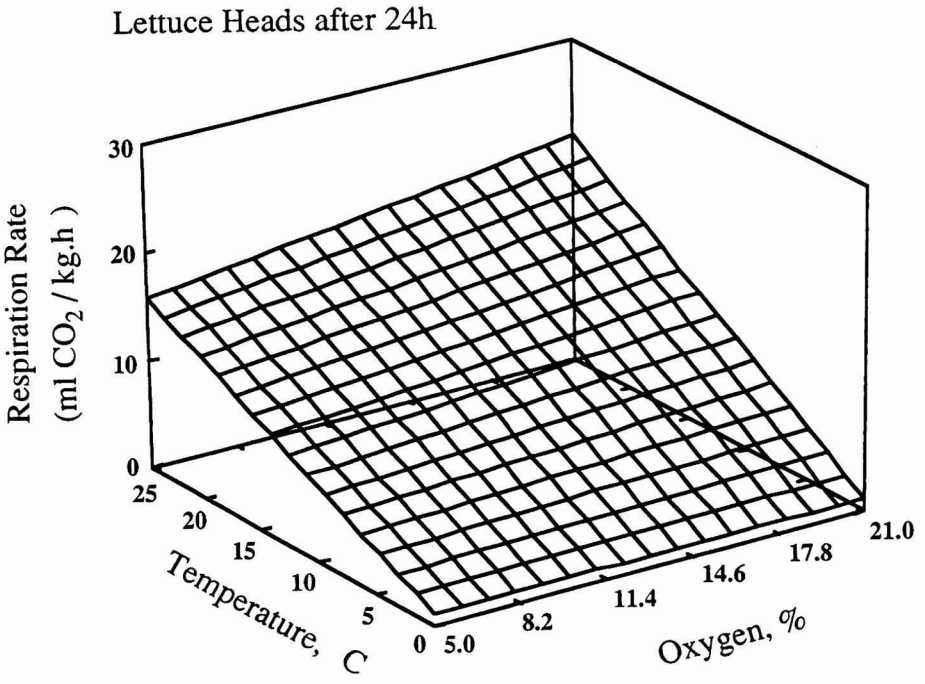

FIG. 2. EFFECT OF O_2 CONCENTRATION AND TEMPERATURE ON $RRCO_2$ OF LETTUCE HEADS AFTER 24 h

Effect of Storage Time. Storage time had no effect on RR at low and high temperatures (not shown). Even when oxygen was increased from 10 to 21%, the effect of time on respiration was negligible.

Packaging Studies with Lettuce

Packaging studies with lettuce heads were carried out in a similar way as done with sweet corn. Glass jars with modified lids were used as the package model and the lettuce heads were placed inside. The film to be tested was attached to the mouth of the jar with a septum attached to it to withdraw samples for head space gas analysis. The two films used in these experiments had the following transmission rates for oxygen and carbon dioxide at 10C: (1) LLDPE, K_o = 0.024 ml O_2/h.cm^2 and K_C = 0.044 ml CO_2/h.cm^2, and (2) film PD941 supplied by Cryovac (Duncan, SC), K_o = 0.060 ml O_2/h.cm^2 and K_c = 0.160 ml CO_2/h.cm^2.

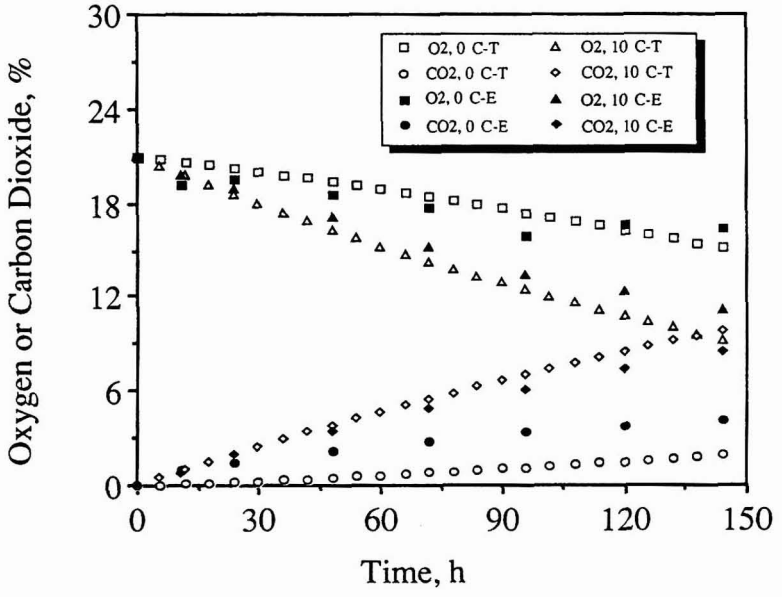

FIG. 3. PREDICTED AND EXPERIMENTAL TRANSIENT O_2 AND CO_2 CONCENTRATIONS USING A LINEAR LOW-DENSITY POLYETHYLENE FILM AT 0 AND 10C. Area of gas transmission was 91.6 cm^2. Weight of lettuce head in the experiment at 0C was 374 g and in that at 10C was 294 g.

Figure 3 shows predicted and experimental transient gas concentrations using LLDPE film at 0 and 10C. At 0C, experimental concentrations of O_2 and CO_2 were in good agreement with computer generated values, although CO_2 concentration differed by about 1%. At 10C, experimental results were very close to predicted values. For film PD941, at 0C (Fig. 4), O_2 concentrations matched predicted values very closely while for CO_2 there was a difference of about 1%. When temperature was increased to 10C (Fig. 4), the difference between the two sets of values was larger. More importantly, with both films, although significant reduction in O_2 concentration and increase in CO_2 concentration in the MAP were achieved that should contribute to increase in shelf-life of lettuce heads, steady state gas concentrations were not achieved for about 90 h. Therefore, a lettuce head in a MAP will be subjected to gas compositions other than those desired for long periods of time.

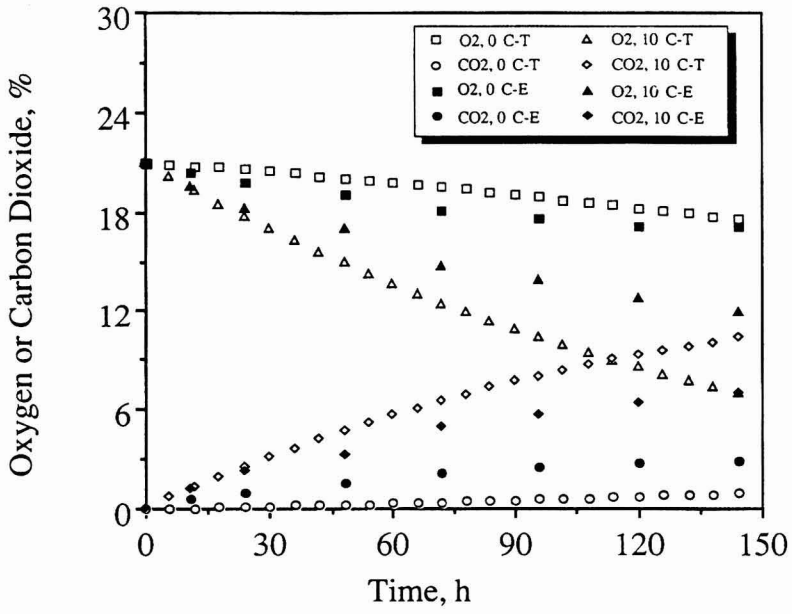

FIG. 4. PREDICTED AND EXPERIMENTAL TRANSIENT O_2 AND CO_2 CONCENTRATIONS USING PD941 FILM AT 0 AND 10C
Area of gas transmission was 91.6 cm^2. Weight of lettuce head in the experiment at 0C was 334g and in that at 10C was 388 g.

We believe that the computer values were sufficiently close to the experimental values and the differences have several explanations. As in the case with sweet corn (Morales-Castro 1992), respiration studies were conducted

before the packaging studies. The variation due to harvest time can account for some of the differences between experimental and predicted values. The discrepancy may also indicate the limits of applicability of the mathematical models for respiration rates. Further work is needed to answer this question. In addition, small errors in permeability values can also contribute to differences in experimental and predicted values.

CONCLUSIONS

Magnitudes of respiration rates of lettuce heads ranged between 3 to 30 ml O_2/kg.h, so that lettuce heads can be classified as low respirers. Oxygen concentration (O_2). temperature (T), storage time (t), and the interactions between temperature and O_2 ($T*O_2$) and time (T*t), affected respiration rates. The two films tested, one a low-density linear polyethylene and the other known as PD941, caused significant reduction in O_2 concentration and increase in CO_2 concentration in the MAP that should contribute to increase in shelf-life of lettuce heads. However, steady state gas concentrations were not achieved for about 90 h. Therefore, a lettuce head in a MAP will be subjected to gas compositions other than those desired for long periods of time.

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THERMAL DEATH PARAMETERS OF ORANGE JUICE AND EFFECT OF MINIMAL HEAT TREATMENT AND CARBON DIOXIDE ON SHELF-LIFE

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ABSTRACT

Freshly squeezed, refrigerated orange juice has a relatively short shelf-life, which could be extended by minimal processing. D and z values of orange juice microflora were obtained using the capillary method, as well as a plate heat exchanger. The effect of low levels of CO_2 on the shelf-life of the juice was also evaluated. The D_{60} value of typical orange juice flora was about 5 s and the z was 4–5C. A combination of minimal heat treatment (15 s at 60C) and 6 mM CO_2 extended the storage life of orange juice to 35 days at 4C.

Carbon dioxide flushed into a 10% headspace of 350 ml jars resulted in 6 mM dissolved CO_2 in the juice at 4C. This level of CO_2 extended the shelf-life of unpasteurized juice to 25 days at 4C and 10 days at 10C, as compared to 17 and 5 days without CO_2, respectively. No significant difference in organoleptic evaluations was found between minimally heat treated juices with or without CO_2 and fresh untreated juices without CO_2 during the first week of storage.

INTRODUCTION

Untreated fresh orange juice has become a well-accepted product in certain markets. However, since its shelf-life is only 1 to 2 weeks when held near 0C, or a few days if stored at 8–10C, its market potential is rather limited. Spoilage of such fresh untreated orange juice is due to the proliferation of its endogenic microflora. This flora is composed of acid tolerant bacteria, yeasts and molds. The major spoilage bacteria are of the *Lactobacillus* and *Leuconostoc* species (Berry *et al.* 1956; Parish and Higgins 1988). Yeasts of the *Saccharomyces* and

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Candida genera were also found in spoiled orange juice (Parish and Higgins 1989). Pasteurization of citrus juices renders them microbially, as well as enzymatically stable, but notably impairs their organoleptic properties.

Mild (minimal) heat treatment ofjuices, will reduce their microbial load, cause minimal damage to their organoleptic properties and extend shelf-life. Such heat treatment will not inactivate the pectolytic enzymes; however, cloud separation, caused by the enzymatic activity, is today accepted by customers and seen as a sign of freshness. Thermal resistance of microorganisms in citrus juices, as well as kinetics of microbial spoilage were reported by several authors (Murdock *et al.* 1953; Kopelman and Schayer 1976). However, there is very little information concerning thermal death parameters at relatively low temperatures.

Another option to increase microbial stability of juices is to use carbon dioxide, which is a well-known microstatic agent for numerous microorganisms (Daniels *et al.* 1985). Some of these microorganisms are affected even by small amounts (5–20mM) of dissolved CO_2 (Jones and Greenfield 1982). Ultra-high pressure (58,000 psig for 10 min), for cold pasteurization, has been reported to stabilize orange juice for 2–3 months at room temperature (Parish 1993). However, the equipment required to apply this method is very costly.

The objectives of this study were to obtain D and z values of some orange juice spoilage organisms and define conditions of minimal heat treatment, with or without the addition of CO_2, in order to extend the shelf-life of the juice without affecting its "fresh like" aroma and taste.

MATERIALS AND METHODS

Evaluation of Heat Resistance of Microorganisms in Capillaries

Streptococcus lactis was isolated from spoiled orange juice and *Saccharomyces cerevisiae* was obtained from the collection of the American Type Culture Collection (ATCC) #7753. Petri plates with Lactobacilli agar (MRS) (Difco) were inoculated with *Str. lactis* and Petri plates with yeast extract (YE) agar (Difco) were inoculated with *S. cerevisiae*. Plates were incubated for 48 h at 3C, rinsed with saline solution, which was added to pasteurized orange juice (11° Brix, pH 3.5) to obtain an inoculum of about 10^7–10^8/ml.

The heat resistance of these microorganisms in orange juice was determined using the capillary tube method (Lawrence and Block 1968). The dimensions of the tubes were: internal diameter: 2.4 mm, external diameter: 4 mm, length: 150 mm. The tubes were filled with 0.6 ml inoculated orange juice and sealed with special plugs. A thermostated ($\pm$ 0.5C) water bath was used for heating.

The inoculated capillaries were put into the bath in a basket, held for the desired time, and cooled immediately in ice water. The come-up time of the orange juice in the tubes was determined by inserting a thin thermocouple, connected to a digital readout, into a capillary tube filled with juice. The come up time was taken into account in calculating the heat resistance parameters. The thermal resistance of the microorganisms was determined at 50, 55, 60, 65, 70, and 75C. For each heating condition, at least 4 tubes were prepared, and all experiments were repeated at least 7 times on different days. Survivors were recovered on Orange Serum Agar (OSA), which was incubated at 30C for 48h, and the number of colony forming units (CFU) was counted. The data obtained from these experiments were used to calculate the D and z values.

Thermal Resistance of *S. cerevisiae* in a Plate Heat Exchanger

An inoculum of *S. cerevisiae* was prepared in flasks containing YE broth and then shaken at 30C for 15–20 h. The broth was then centrifuged and the cells were added to pasteurized orange juice to obtain a level of 10^6–10^7 cfu/ml. The inoculated juices were heated in an APV (APV Co., England) model JHE laboratory plate heat exchanger.

The temperature in the heat exchanger was controlled by means of a pneumatic proportional controller ($\pm$ 0.5C). The residence time in the holding tube of the heat exchanger, was monitored by adjusting the flow rate of the positive feed pump (Reliance Tri Clover model PRED-3, Ladish Co., Kenosha, WI). The thermal treatments were carried out at 51.5–61.5C for 10–15 s. Numbers of survivors were estimated on OSA, which was incubated for 48 h at 30C and survival curves were obtained. Experiments were repeated at least 3 times.

Juice Preparation for Evaluation of Shelf-Life

Shamuti and Valencia oranges, depending on season, were used for the minimal heat treatment studies. The oranges were stored overnight at 4C, washed in water containing about 5 ppm Cl_2, extracted in a FMC In Line Juice extractor, and filtered through a 1 mm rotary screen, and then divided into 2 equal parts. One part of the juice was filled without heating into 350 ml glass jars and the other part was heated, in the APV plate heat exchanger at 60C for 16 or 21 s, and then filled into 350 ml glass jars. Head spaces of 10 and 20%, 35 and 70 ml respectively, were created by means of plastic plungers, and were flushed with pure N_2, pure $C0_2$, or with air. The jars were closed and stored at 4 and 10C. The juices were examined periodically for microbial load (on OSA

as above), dissolved $C0_2$, and by sensory analysis. All determinations were carried out in at least 2 replications.

Dissolved carbon dioxide was determined by a similar method as the one described for milk by King and Mabbit (1982). Orange juice in 5-ml portions of was transferred to 100 ml vials equipped with magnetic stirrers. These vials were sealed with teflon septa and aluminium rings, and 1 ml of 1N H_2S0_4 was injected into the vials. After shaking for 2 min, the concentration of CO_2 in the head space was measured by gas chromatography, using a Becker-Packard (Holland) model 406 gas chromatograph equipped with a thermal conductivity detector, and a Poropack Q (60/80 mesh) 0.25 in × 6 ft column. Helium was used as the carrier gas, at 50 ml/min, and column temperature was 40C. A standard curve was prepared, using standard solutions of $NaHC0_3$, from which the CO_2 content in the samples was calculated.

Sensory evaluations of the juices were carried out by means of triangular difference tests and when difference were found on a hedonic scale (Larmond 1977). The taste panel consisted of at least 10 judges. Samples were presented at room temperature, in partitioned booths, in a special taste facility. In the hedonic test the panel was asked to score samples on a scale from 5 (best) to 1 (worst).

Statistical evaluations of hedonic sensory test were carried out by analysis of variance according to Larmond (1997). Results of shelf-life data were subjected to a two way analysis of variance to determine the effects of treatment, storage time and interaction between them.

RESULTS AND DISCUSSION

Thermal Resistance of *S. cerevisiae*

A typical heat resistance plot of *S. cerevisiae* at 70C is shown in Fig. 1. The decrease in CFU followed an apparent first order behavior to about 10^2 /ml, after which the number of organisms was not reduced further. This "tailing" phenomenon was observed at all temperatures used in this study, and with both microorganisms (i.e., *S. cerevisiae* and *Str. lactis*). The number of survivors remained constant even after prolonged exposure to heat, e.g., 400 s at 60C, or 250 s at 70C. This type of tailing curve is mentioned in the literature in connection with spore forming bacteria or for conditions of nonlogarithmic death curves (Stumbo 1949; Pflug 1979). The tailing phenomenon in connection with the use of thermal death tubes for resistance studies was also shown by Weissman (1975). The tailing phenomenon was observed in the present study only in heat resistance experiments using capillaries, and not in the plate heat

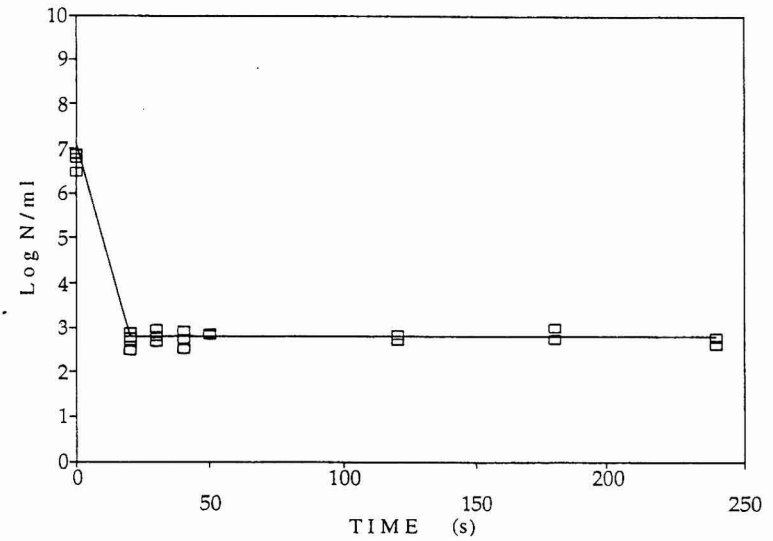

FIG. 1. THERMAL DEATH CURVE OF *S. CEREVISIAE* AT 70C WITH TAILING

exchanger. The linear line portion of the thermal resistance curves obtained using capillaries and in the heat exchanger were similar. Therefore, for the calculation of D and z values only the straight line section of the curves was employed. All survival curves of *S. cerevisiae* followed first order kinetics above 10_2 cfu/ml, with r^2 values above 0.82. The D_T values ranged from 568 s at 50C to 4.1 s at 60C, and the z value was 5.4C. The results of some of the experiments with *S. cerevisiae* are summarized in Table 1. The values obtained were comparable to those reported in the literature. Kopelman and Schayer (1976) found, for yeasts in reconstituted orange juice, D_{55} = 165.6 s and z = 5.35C. Parish (1991) found D_{53} = 21–31 s for *S. cerevisiae* inoculated into grapefruit serum.

Heat Resistance of *Str. lactis* in Capillaries

All survival curves of *Str. lactis* fitted first order kinetics, above 10^2 cfu/ml, with r^2 values above 0.8. The D_T values obtained ranged from D_{50} = 138 s to D_{60} = 6.3 s, and z = 7.2C. The results for *Str. lactis* are summarized in Table 2. Parish (1991) reported D_{53} = 110–115 s for *Lactobacillus plantarum* and D_{53} = 89–94s for *Leuconostoc mesenteroides*.

TABLE 1.
D VALUES OF *S. CEREVISIAE* AT 3 TEMPERATURES OBTAINED IN CAPILLARIES

Temperature (C)	Number of samples	D value (s)	r^2
50	10	568	0.93
55	15	62.6	0.94
60	12	4.1	0.94

TABLE 2.
D VALUES OF *STR. LACTIS* AT 3 TEMPERATURES OBTAINED IN CAPILLARIES

Temperature (C)	Number of samples	D value (s)	r^2
50	11	138	0.92
55	10	86	0.94
60	14	6.3	0.98

Heat Resistance of *S. cerevisiae* in Plate Heat Exchanger

A typical survival curve obtained for *S.cerevisiae* in a plate heat exchanger at 58C is shown in Fig. 2. D values obtained in the temperature range of 51.5C to 63C are summarized in Table 3. The D_T values ranged between $D_{51.5}$ = 111

s to D_{63} =v 1.7 s and the z value was 5.8C. When holding times were extended, at all temperatures used, sterile juices were obtained and no "tailing" phenomenon was observed. The heat resistance values obtained for *S. cerevisiae* in the plate heat exchanger were comparable to those obtained from the straight line portion of the capillary tube method. This indicates that one can use the linear portion of the D curves, obtained by the capillary tube method, for calculating pasteurization times.

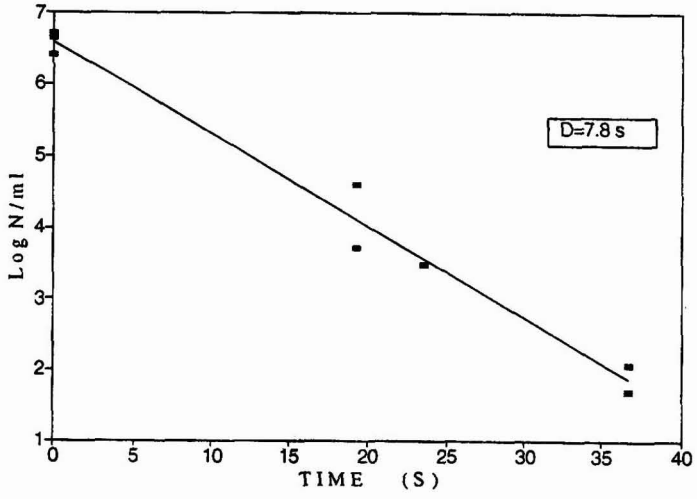

FIG. 2. THERMAL DEATH CURVE OF *S. CEREVISIAE* AT 58C OBTAINED IN A PLATE HEAT EXCHANGER

Effect of CO_2 on Fresh Orange Juice in Glass Jars

The head spaces of 10 and 20% of jar volumes flushed with CO_2 resulted in the dissolution of 6 and 10 mM CO_2, respectively, in fresh orange juices stored at 4C. These quantities of CO_2 extended the lag phase of microbial growth, at 4C, to about 21 days as compared to 15–16 days without CO_2 (Fig. 3). The inhibition of microbial growth is also demonstrated in Fig. 4, which shows the amounts of dissolved CO_2 in the different juices. The effect of CO_2 on microbial growth was highly significant ($p = 0.001$). Since a major end product of microbial metabolism is CO_2, the concentration of dissolved CO_2 remained constant as long as the microflora did not proliferate.

TABLE 3.
D VALUES OF *S. CEREVISIAE* OBTAINED
IN A PLATE HEAT EXCHANGER

Temperature (C)	Number of samples	D value (s)	r^2
51.5	11	111	0.77
57.5	8	10.8	0.98
59	10	8.3	0.90
61.5	5	2.8	0.99
63	7	1.7	0.99

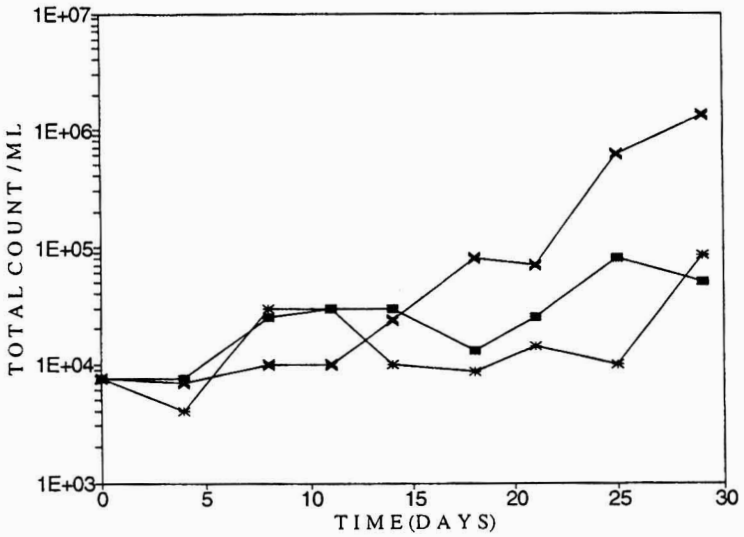

FIG. 3. TOTAL COUNTS IN ORANGE JUICES WITH AND WITHOUT MODIFIED ATMOSPHERES STORED AT 4C
■ 10 mM CO_2, * 6 mM CO_2, × air

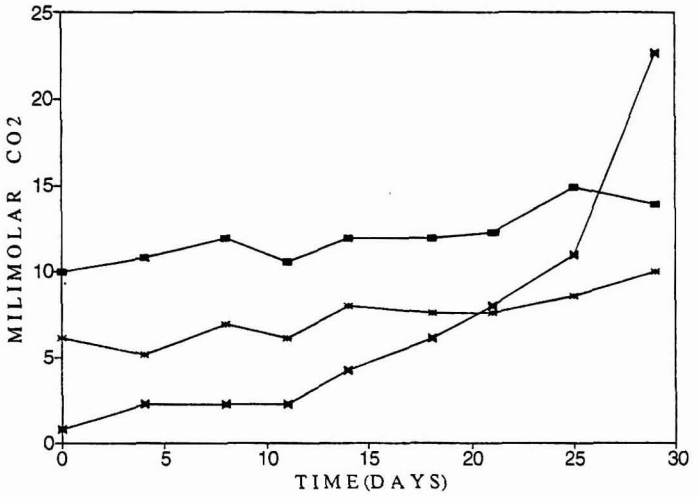

FIG. 4. DISSOLVED CO_2 CONCENTRATION IN ORANGE JUICES WITH AND WITHOUT MODIFIED ATMOSPHERES STORED AT 4C
■ 10 mM CO_2 * 6 mM, × air.

There was a significant difference ($p = 0.05$) in the triangle sensory evaluation between juices with 6 and 10 mM CO_2. The latter juices were judged unacceptable in hedonic evaluations. However, there was no difference between juices with 6 mM CO_2 and juices without CO_2. Based on these results, only the smaller 10% (35 ml) headspace was used in subsequent experiments.

End of shelf-life of the juices was determined when the juices received hedonic sensory scores of 3 or below. The juices with 6 mM CO_2 had a shelf-life of 25 days at 4C as compared with 15–16 days for juices with air in the headspace. Juices stored at 10C showed a similar trend to those stored at 4C as regards microbial growth and dissolved CO_2. The shelf-life of these juices with and without $C0_2$ in the headspace was 10–11 days and 4–6 days, respectively.

Effect of Minimal Heat Treatment

The effect of minimal heat treatment (60C, 16 s) and 6 mM CO_2 on microbial growth is shown in Fig. 5. While the heat treatment reduced the microbial load by 2 log cycles, to less than 10^2 cfu/ml, the lag phase was essentially unaffected by this treatment. The shelf-life of the heat treated juices without CO_2, at 4C, was 35 days as compared to 17 days for fresh juices with

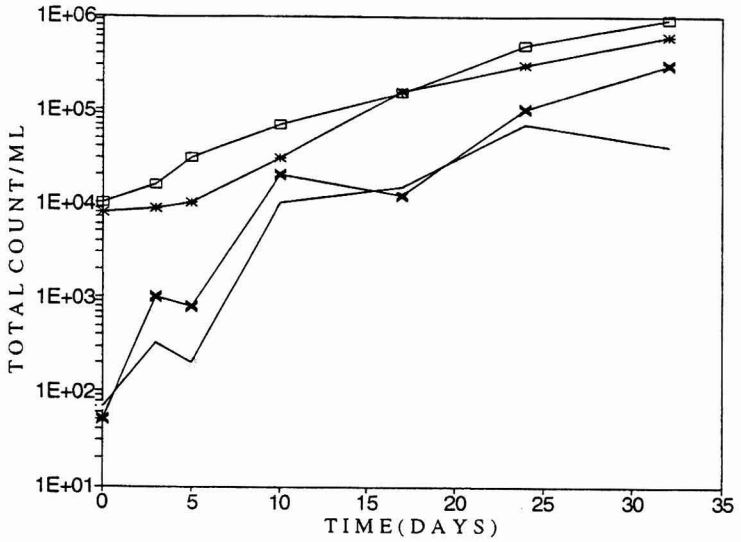

FIG. 5. TOTAL COUNTS IN DIFFERENTLY TREATED ORANGE JUICES STORED AT 4C
□ Unpasteurized, × unpasteurized + 6 mM CO_2,
× pasteurized, ⁘ pasteurized + 6 mM CO_2.

6 mM CO_2. The addition of CO_2 to the heat processed juices had only a small effect, and extended shelf-life by an additional day or two. Apparently, the flora of the heat treated juices was different from that of the fresh juice and was not affected by CO_2.

CONCLUSIONS

In heat resistance studies conducted in capillary tubes surviving microorganisms remained at a level of 10^2/ml, even after prolonged heating. However, in a plate heat exchanger commercial sterility was obtained after prolonged heating. The D and z values obtained for *Str. lactis* and *S. cerevisiae*, from the logarithmic portion of the thermal resistance curves, in both the capillary tube method and the plate heat exchanger, were comparable. The addition of CO_2 into a 10 headspace of fresh orange juice in glass jars as well as a minimal heat treatment (60C for 15 s) extended the shelf-life of orange juice significantly with no detectable effect on taste.

ACKNOWLEDGMENT

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EDIBLE CORN-ZEIN FILM COATINGS TO EXTEND STORAGE LIFE OF TOMATOES

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ABSTRACT

Tomatoes at the turning stage of maturity were coated with corn-zein (CZ) protein film of 5 (0.20), 15 (0.61) and 66 μm (2.60 mil) thickness. O_2 transmission rate at 0% RH and 30C and CO_2 transmission rate at 0% RH and 21C of protein film were much lower than those of a typical shrink wrap film at 0% RH and 23C. Whereas the water vapor transmission rate of CZ film at 15C and 85% RH was much higher than that of the shrink wrap film at 37C and 100% RH. Uncoated tomatoes took 6 days to turn red. A 6-day delay in ripening was observed with coatings of 5 and 15 μm thick without adverse effects. The 66 μm coating markedly delayed color development, while showing the greatest weight loss and alcohol fermentation due to anaerobic fermentation.

INTRODUCTION

Edible film coatings can provide an alternative for extending postharvest life of fruits and vegetables. For example, sucrose fatty acid ester coatings have

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been employed for banana and apples (Cox's Orange Pippin, McIntosh and Golden Delicious), and hydrophobic emulsion such as beeswax-coconut oil emulsion was applied to peach (Banks 1983, 1984; Chu 1986; Erbil and Muftugil 1986; Santerre *et al.* 1989; Smith and Stow 1984). A lipid mixture of hydrogenated vegetable oil, emulsified propylene glycol esters of fat, beeswax and sucrose fatty acid esters have been applied to retain volatile flavor components in pineapple oranges during storage (Nisperos-Carriedo *et al.* 1990).

Controlled atmosphere (CA) storage in which the environmental conditions are closely monitored and precisely controlled is commercially used in extending the shelf-life of many fruits and vegetables (Smock 1979; Barrett *et al.* 1991). However, such a technique often involves high capital and maintenance costs and requires the attention of relatively skilled operators. Modified atmosphere (MA) storage involving the use of flexible films is a viable alternative for providing the desirable conditions. Due to advances in manufacturing of flexible films it is possible to produce films having physical characteristics to result in desired gaseous compositions around a respiring product (Kader *et al.* 1989). In view of environmental concerns of nonbiodegradability of plastic films, coating fruits and vegetables with edible films shows potential to maintain quality and extend shelf-life.

Edible film coatings can be made from food materials regarded as generally recognized as safe (GRAS). Compounds that can be used for making edible coatings include cellulose derivatives and proteins. These coatings provide a good barrier to oxygen and carbon dioxide transmission, but a poor barrier to water vapor (Guilbert 1986; Kester and Fennema 1986). These are desirable characteristics for fruit and vegetable packaging. The oxygen and carbon dioxide barrier leads to a reduction in respiration rate by limiting exposure to ambient oxygen and increasing internal carbon dioxide. The poor water vapor barrier allows movement of water vapor across the coating, thus preventing water condensation, which can be a potential source of microbial spoilage (Ben-Yehoshua 1985). Controlling the lipid content in cellulose and protein coatings can modify the water vapor barrier properties (Kamper and Fennema 1984; Kester and Fennema 1989).

Corn-zein is an alcohol-soluble protein with excellent film- and fiber-forming properties (Balmaceda and Rha 1974). Corn-zein coating has been used on confectionery products and nuts because it is a good barrier to oxygen and lipid (Cosler 1958). The commercial edible coating, Cozeen™ (Zumbro Inc., Hayfield, MN), which has been used for candy and nuts, contains corn-zein as a major component. Corn-zein coating is a good barrier to oxygen (Aydt *et al.* 1991), but its water vapor permeability is about 800 times higher than a typical shrink wrapping film.

Edible coatings applied to the surfaces of fruits and vegetables change the internal gas composition, which in turn influences the product respiration rate.

Proper edible coatings should maintain an optimal range of internal oxygen concentration of the fruits and vegetables (Kader 1985). Knowledge of gas transmission rates of the coatings and the internal gas composition of fruits and vegetables affected by application of coatings is required for suitable selection of edible film coatings. Most research on edible film coatings for horticultural products has been by trial and error methods without systematic studies. In our study, corn-zein film coating was selected to be used on tomatoes. The objectives of this study were (1) to measure the transmission rates of oxygen, carbon dioxide and water vapor of corn-zein film developed as a coating in our laboratory; and (2) to determine the effect of coating tomatoes with corn-zein edible films on their color change, alcohol formation, weight loss and internal gas compositions (oxygen and carbon dioxide) of the fruit during storage.

MATERIALS AND METHODS

Fruit

Tomatoes, *Lycopersicon esculentum* (cv. 'Mountain Pride'), at turning stage were sorted for uniform size, color and physical damage and dipped in 100 ppm of Benlate® (Dupont, Wilmington, DE), Benomyl [methyl 1-(butylcarbomyl)-2-benzimidazole carbamate] for 30 s. A total of 240 tomatoes were divided into four groups, control, A, B and C.

Treatment

A commercial corn-zein product (Regular Grade F 4000, INC Biomedicals, Inc., Cleveland, Ohio) was used in preparing edible film solutions. Corn-zein is alcohol soluble prolamine protein of corn gluten and has the characteristic of forming clear, tasteless and almost invisible edible film. Three solutions (A, B, C) were prepared using 54 g corn-zein, 14 g glycerin and 1 g citric acid dissolved in 520 g, 260 g and 130 g ethanol (95%), respectively. Tomatoes from groups A, B and C were dipped for 10–15 s in the solutions A, B, and C, respectively. After dipping they were held by fingers or tongs for about 5 s and then placed on waxed paper for drying. They were dried at room temperature by blowing air with a table fan. All the samples (control and coated) were held under ambient conditions (21C, 55–65% RH).

Measurements

Film Gas Transmission Rates. It was not possible to determine gas transmission rates of the film coating on the fruit itself. Therefore, a separate

flat film was prepared. Two primarily known methods of preparing flat films are those by Kamper and Fennema (1984) and Aydt *et al.* (1991). We followed the method described by Kamper and Fennema (1984). The edible film solution was poured onto a glass plate, dried and carefully detached from the plate after drying. It was then kept in desiccator until measuring the gaseous transmission rates. Film thickness measurements were recorded at several locations using a micrometer to an accuracy of 0.001 mm. The carbon dioxide transmission rate was measured at 21C and 0% RH using a permeability cell described by Gilbert and Pegas (1969). An OX-Tran 100 oxygen transmission tester (Mocon Modern Controls, Inc., Minneapolis, MN) was utilized to measure the oxygen transmission rate at 30C and 0% RH. To determine the transmission rate of water vapor, small cups filled with desiccant and covered with the corn-zein film to be tested were held in 85% RH and 15C conditions using a saturated solution of lithium sulfate. The weight change of the desiccant was recorded periodically (Kamper and Fennema 1984; Labuza and Contreras-Medellin 1981).

Coating Thickness. The thickness of the corn-zein film coating was measured by taking a thin section of the coated fruit and observing it under a light microscope (Olympus Light Microscope, Model BHS).

Weight Loss. The weight loss of the fruit during storage was measured by weighing to the nearest 0.001 g.

Apparent Internal Gas Composition. Open-ended glass tubes (1.2 cm I.D. and 2.0 cm long) were attached to the stem scar of four tomatoes of each group and fitted with noncorrosive rubber stoppers every two days during storage. The stopper was placed on the glass tube for 2 h prior to sampling gas for measuring O_2 and CO_2 using gas chromatography (GC) equipped with a thermal conductivity detector (Hewlett Packard, Model 5890). Then, the stopper was removed for free exchange of gas between the environment and fruit until the next measurement. The column specifications and GC operating conditions are as follows: column CRT-I, 182.9 cm long, (Alltech Associates, Deerfield, IL); oven, 25C; injector, 100C; detector, 250C; N_2 flow rate, 20 l/h. The procedure for measuring internal gas composition was adopted from Banks (1983) and Banks and Kays (1988).

Alcohol Content. Every two days, two tomatoes from each group were ground in a blender, transferred to a bottle containing 10 ml of distilled water and sealed with a septum cap. The bottle was shaken vigorously and immersed in a water bath at 95C for 30 min and then cooled and held at 50C for 10 min. A headspace sample (1 ml) was withdrawn by a gas-tight syringe and injected into a GC equipped with a flame ionization detector (Hewlett Packard, Model

5790). The column specifications and GC operating conditions are as follows: column Microtek 2000-R with Carbowax 20 M (270 cm × 6.4 mm); oven, 25C; injector, 150C; detector, 180C; N_2 flow rate, 36 l/h. The procedure described here was adapted from Banks (1984) and Davis and Chace (1969).

Color Measurement. Five coated tomatoes from each group were monitored for their surface color change using a Gardner XL-845 colorimeter (Pacific Scientific, Bethesda, MD) (Table 1). Instrumental color readings (L, a and b) were obtained every two days and were taken as an average of 8 different points on the circumference and blossom end of the tomatoes using a standard pink calibration tile (L=69.1, a=23.4 and b=9.3) (Yang and Chinnan 1987). It is not practical to take off the edible coating from tomatoes every time for measuring color change during storage, thus a correction factor to offset the effect of the film coating on the instrumental color readings was obtained. A separate group of 54 tomatoes was selected where the surface color change was recorded on the same tomato with and without the coating, using a modification of the method developed by Pendalwar and Chinnan (1991). Average hue angle of nine tomatoes at each stage was calculated before coating. Fruit from each stage was divided into three groups of three tomatoes, each of which were coated with one of the three solutions, and color measurements were performed.

RESULTS AND DISCUSSION

Gas Transmission Rate

Gas and water vapor transmission rates (WVTR) of corn-zein (CZ) protein film, which was prepared on the glass plate, are presented in Table 2 and compared with those of a typical shrink-wrap film for individual seal packaging of tomatoes (Yang *et al.* 1987). Thickness of this CZ film was 310 ± 8 μm (12.2 ± 0.3 mil). Data for films of unit thickness given in Table 2 show that O_2 and CO_2 transmission rates of the corn-zein film prepared in our laboratory were much lower than the D-955™ film, whereas the water vapor transmission rate (WVTR) was much higher. Cozeen™ (Zumbro Inc., Hayfield, MN), commercial corn-zein based film coating primarily developed for coating nuts, provides a poorer barrier to carbon dioxide, oxygen and water vapor transmission than those of corn-zein film prepared in this study (Aydt *et al.* 1991). High O_2 and CO_2 barrier and low water vapor barrier properties of corn-zein film are favorable characteristics for application to coating fruits and vegetables to prevent condensation of water vapor. Although a film with greater WVTR is desirable for packaging fruits and vegetables, too high WVTR can result in

TABLE 1.
HUNTER COLOR VALUES (L, a and b) OF NONCOATED (NT) AND COATED TOMATOES (CT) WITH CORN-ZEIN (CZ) AT VARIOUS MATURITY STAGES AND ASSOCIATED CORRECTION FACTOR (CF)

Maturity stage	CZ Soln.		Hunter color values[1]			Hue angle	CF[2]
			L	a	b		
Green	A	CT	41.3	-9.5	21.3	114.0	0.009
		NT	40.7	-9.0	21.2	113.0	
	B	CT	41.4	-9.7	21.4	114.4	0.012
		NT	40.6	-9.2	21.3	113.3	
	C	CT	40.5	-9.4	21.8	113.3	0.015
		NT	39.7	-8.5	21.5	111.6	
Breaker	A	CT	38.2	-7.4	19.3	110.9	0.026
		NT	37.2	-6.3	19.2	108.1	
	B	CT	38.4	-7.6	19.2	111.6	0.016
		NT	37.7	-6.9	19.2	109.8	
	C	CT	39.5	-7.9	19.4	112.2	0.025
		NT	38.9	-6.8	19.3	109.5	
Turning	A	CT	37.1	-5.8	17.8	108.0	0.027
		NT	36.5	-4.8	17.5	105.2	
	B	CT	37.3	-5.5	17.6	107.8	0.041
		NT	36.1	-4.3	17.5	103.6	
	C	CT	37.4	-5.6	18.0	107.9	0.033
		NT	37.4	-4.6	17.7	104.5	
Pink	A	CT	36.5	0.04	16.7	89.7	0.029
		NT	36.1	0.7	16.3	87.2	
	B	CT	36.3	-0.2	16.8	90.7	0.031
		NT	35.0	0.6	16.4	88.0	
	C	CT	36.8	-0.3	16.8	91.3	0.042
		NT	35.8	0.7	16.4	87.6	
Light red	A	CT	35.5	7.4	15.0	63.9	0.076
		NT	34.8	8.9	15.0	59.4	
	B	CT	33.3	7.3	15.8	65.4	0.045
		NT	32.3	8.1	15.7	62.6	
	C	CT	34.9	6.7	15.7	66.8	0.055
		NT	33.2	7.8	15.5	63.3	
Red	A	CT	26.4	23.1	13.9	31.0	0.040
		NT	25.4	24.0	13.7	29.8	
	B	CT	26.4	22.9	13.6	30.9	0.044
		NT	25.2	23.8	13.4	29.6	
	C	CT	27.4	23.2	12.2	27.8	0.041
		NT	26.6	24.2	12.2	26.7	

[1]Hunter color measures refer to mean of three tomatoes.

[2]Correction factor was calculated by taking the difference of hue angle of noncoated and coated tomatoes and dividing the difference by the noncoated tomato value.

TABLE 2.
GAS TRANSMISSION RATES OF CORN ZEIN (CZ) FILMS AND A TYPICAL SHRINK WRAPPING FILM

Item #	Film type	Thickness		Gas transmission rate[1]		
		μm	(mil)	CO_2	O2	H_2O vapor
	Film					
1	CZ[2]	310	(12.2)	0.0311	0.0041	73.45
2	D-955[3]	15	(0.6)	14.60	6.80	14.57
3	CZ[4]	25	(1.0)	0.38	0.05	896.15
4	D-955[4]	25	(1.0)	8.76	4.08	8.74
5	Cozeen[5] (303 NF)	25	(1.0)	6.35	1.07	6938.28
	Coatings[6]					
6	CZ-A	5	(0.20)	1.90	0.25	4480.75
7	CZ-B	15	(0.61)	0.62	0.08	1469.10
8	CZ-C	66	(2.60)	0.15	0.02	344.67

[1]Units for O_2 and CO_2 are $L/m^2 \cdot day$; unit for H_2O vapor is $g/m^2 \cdot day$. O_2 values at 21 C, 0% RH and 1 atm; CO_2 at 30 C, 0% RH and 1 atm; H_2O vapor at 15 C at 85% RH.

[2]Values in this row were experimentally determined in this study.

[3]D-955™ film (Cryovac Division of W.R. Grace Co., Duncan, SC); values in this row were taken from manufacturer's data (Yang et al. 1987). O_2 and CO_2 values at 23 C, 0% RH and 1 atm; H_2O vapor at 37 C and 100% RH .

[4]Values calculated for unit film thickness for comparison of corn zein and D-955™ films.

[5]Cozeen™ (303 NF) is a commercial product available from Zumbro Inc. (Hayfield, MN); values in this row were taken form Aydt et al. (1991) and were calculated for unit thickness for comparison.

[6]Gas transmission values for the coatings were calculated with various thicknesses from the experimental values in item # 1, these can also be calculated from item # 3.

excessive weight loss of fruit when stored at low humidity. Thicknesses of the corn-zein coating of tomato groups, A, B and C, were 5 ± 1 μm (0.20 ± 0.038 mil), 15 ± 3 μm (0.61 ± 0.112 mil) and 66 ± 4 μm (2.60 ± 0.155 mil) (mean of ten replicates ± standard deviation), respectively. Gas transmission rate of the coating decreases as the coating thickness increases. It may be noted that the gas transmission rates of coatings were calculated from the gas transmission of flat films.

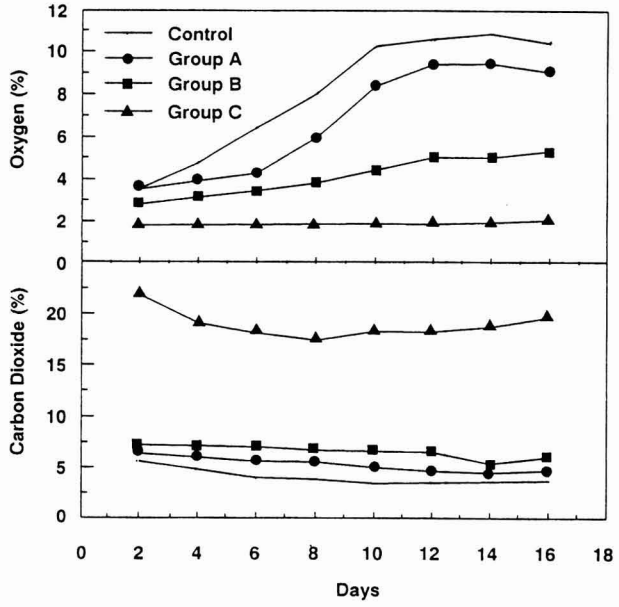

FIG. 1. APPARENT INTERNAL GAS COMPOSITIONS OF FOUR GROUPS OF TOMATOES. CONTROL IS WITHOUT ANY COATING. GROUPS A, B AND C REFER TO TOMATOES COATED WITH CORN-ZEIN FILM OF THICKNESS 5, 15 AND 66 μm, RESPECTIVELY

Internal Gas Composition

Apparent O_2 and CO_2 compositions of tomatoes during storage are given in Fig. 1. Since the method used in our study does not necessarily give the true internal gas composition, but is an indicator of the internal gas composition, this measurement is referred to as apparent internal gas composition. Internal O_2 composition (IOC) of control, group A and group B did not differ significantly ($\alpha = 0.05$) beyond 10 days of storage. Other statistical results ($\alpha = 0.05$) for mean values of IOC were as follows: for control, no difference between 2 and

4 days, 4 and 6 days, or 6 and 8 days; for group A, no difference was found between 2, 4 and 6 days, or 6 and 8 days; for group B no difference was observed between 2, 4 and 6 days, or 4, 6 and 8 days, or 8 and 10 days. For group C, IOC values were not significantly different from 2 to 14 days, and between 14 and 16 days. Internal CO_2 (ICC) of control, group A and group B were not statistically different after 6, 4 and 8 days of storage, respectively. Other statistical results for ICC were: no difference between 2 and 4 or 4 and 6 days for control; no difference between 2 to 10 days for group A; and no difference between 2 to 12 days or then between 12 to 16 days for group B.

In our preliminary studies for measuring internal gas composition, it was observed that 2 h was sufficient to achieve equilibrium conditions between the tomato tissue and headspace in glass vials attached to the fruit. However, the preliminary studies were done on mature green fruit respiring under ambient conditions. Due to reduction in respiration rate after applying the coating, it is probable that the 2 h period was not sufficient to result in equilibrium internal gas composition (Banks and Kays 1988). It is to be noted that the initial O_2 concentration in the glass vial is 21% and it continues to decrease until an equilibrium with the internal gas composition is reached. Thus, if the equilibrium is not reached, the measured value will be greater than the actual internal oxygen concentration. An opposite trend is expected in case of CO_2 level.

The respiration rate is usually very high during the early stages of development and decreases as fruits and vegetables mature. Climacteric fruits, such as apple, banana and tomato are characterized by a preclimacteric stage characterized by low respiration, an increase of respiration to the climacteric peak followed by a decline at the postclimacteric stage (Kays 1991). In case of tomatoes it is during the climactic period that the color changes rapidly from 'breaker' or 'turning' stage to 'red'. Thus, the approach used in measuring O_2 concentration is probably responsible for the observed increase in measured O_2 concentration in the control samples and those in groups A and B over 16 days. However, O_2 concentration of group C having a thicker coating was not affected, probably because O_2 for respiration was not available through the surface of the fruit but only from that in the glass tube causing the equilibrium conditions to be established much faster than in other groups. Initial CO_2 concentration in the glass tube was zero; thus, it is believed 2 h were probably adequate to establish equilibrium condition for CO_2. Figure 1 indicates that CO_2 concentration of group C was much higher than the other three groups.

Alcohol Production

Fruits and vegetables are susceptible to a wide range of physiological disorders, the incidence of which depends on a number of factors, including

atmosphere concentration of O_2 and CO_2. Modification of internal atmosphere by the use of coating can increase disorders associated with high CO_2 and low O_2 concentration, such as accumulation of ethanol and alcoholic off-flavors (Smith *et al.* 1987). Extremely low O_2 contents for broccoli and cauliflower result in off-flavors associated with a change from aerobic to anaerobic metabolism (Weichmann 1987). Alcohol production was observed only in group C (Fig. 2) and was attributed to low O_2 and high CO_2. Mean values presented in Fig. 2 were significant ($\alpha = 0.05$) for all days except between 2 and 4 days.

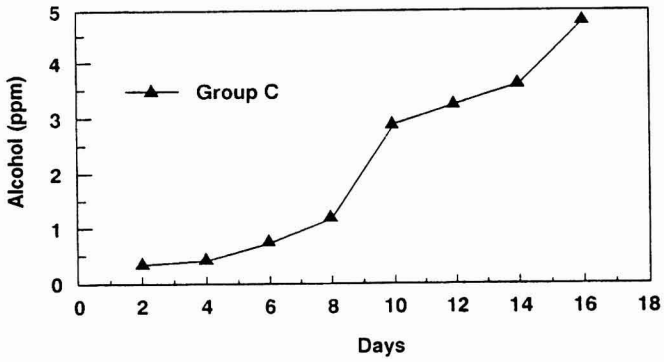

FIG. 2. CHANGE IN ALCOHOL PRODUCTION OF TOMATOES IN GROUP C
Refer to legend for Fig. 1 for treatment explanation.

Color Change

The L, a, b values of coated and noncoated tomatoes used in calculating correction factors and the corresponding corrections factors (CF) are presented in Table 1. An example of the use of correction factor is presented here. If hue angle of a tomato with a coating from solution A is 113, then it was assumed that the maturity stage of the tomato was between 'green' and 'breaker' (see Table 1). Appropriate correction factor, CF, was calculated from

$$CF = 0.009 + (0.026 - 0.009)/(114 - 110.9) = 0.0145$$

Following which the corrected hue angle, CHA, was estimated from

$$CHA = 113 - (113 \times 0.0145) = 111.4$$

Color changes during tomato ripening are characterized by a loss of chlorophyll and a rapid accumulation of carotenoids (lycopene), as chloroplasts are converted to chromoplasts (Khudairi 1972). Color is an important factor in the consumer acceptability of tomatoes (Beattie *et al.* 1983). Color development of tomato is influenced by the gas composition of its environment (Yang and Chinnan 1987). In ripening of tomatoes, high CO_2 level decreases ethylene synthesis, which can delay the color change (Buescher 1979). The tomatoes coated with corn-zein film delayed the color change; and the degree of color change was mainly dependent on the thickness of coating, which increased the CO_2 level and decreased the O_2 level (Fig. 3). The color change, represented by hue angle, of group C was delayed to pink stage (hue angle = 91) after 12 storage days. In the group B, it took 10 days to attain a red color (hue angle = 30). On the other hand, it took only 6 days for the control group to become red.

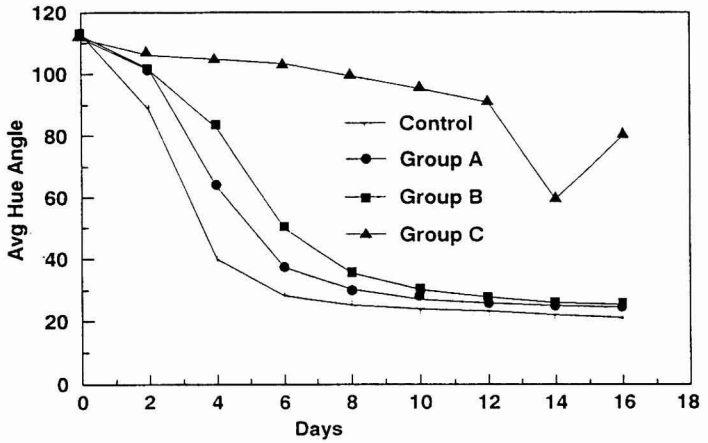

FIG. 3. CHANGE IN HUE ANGLE OF FOUR GROUPS OF TOMATOES
Refer to legend for Fig. 1 for treatment explanation.

Hue angles were not significantly different (α = 0.05), after 8 days of storage except for group C. Other results not statistically different were: no difference between 6 and 8 days for control; no difference between 6 to 10 days for group A; and, no difference between 0 and 2 days for group B. For group C hue angles were not statistically different for up to 12 days of storage; also there was no difference between 12 and 16 days, or 14 and 16 days of storage due to large variation in data after 12 days of storage.

Weight Loss

One purpose for applying edible coatings to fruits and vegetables is to reduce weight loss during storage. For example, Ben-Yehoshua (1969) reported marked reductions in weight loss of wax coated oranges during storage. Slight reductions in weight loss in bananas and apples have been found with sucrose ester-based coating (Banks 1984; Smith and Stow 1984). In our study all groups of tomatoes showed loss of weight during storage (Fig. 4). After 14 days of storage, the weight loss in the control group was 5.3% (w/w), whereas in groups A and B weight losses were 4.3% and 4.5% (w/w), respectively; however, no statistical difference ($\alpha = 0.05$) was observed between these means. Group C showed the greatest weight loss compared with the other groups. Group C had the thickest coating of corn-zein film resulting in reduced availability of O_2 reduced dissipation of CO_2 and increased production of alcohol (Fig. 2). The weight loss of group C was also visually apparent during storage, as indicated by shrivelling of the fruit and fermented odor detected when tomatoes were cut for alcohol content determinations. We attribute the increased weight loss to accelerated senescence and anaerobic fermentation of the tomato tissue, resulting in the loss of sugars, generation of heat and the production of end products of anaerobic fermentation (Weichmann 1987).

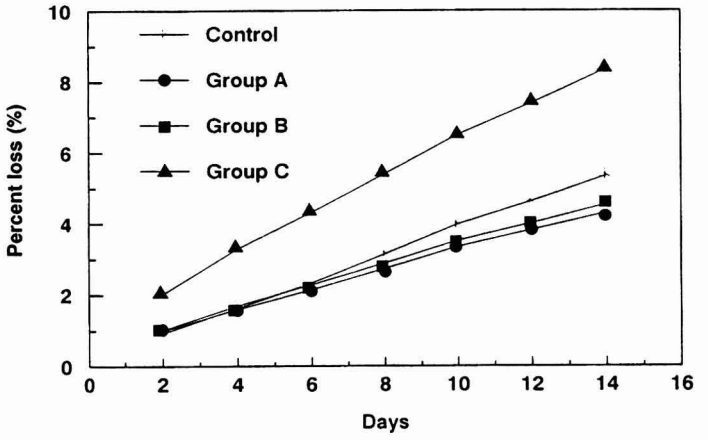

FIG. 4. CHANGE IN WEIGHT LOSS OF FOUR GROUPS OF TOMATOES
Refer to legend for Fig. 1 for treatment explanation.

SUMMARY AND CONCLUSIONS

The success of edible film coatings for fruits and vegetables will mainly depend on the control of coating thickness because the transmission rates of O_2, CO_2 and water vapor depend on the thickness and compositions of films. A thick coating with good gas barrier properties causes detrimental effects as it reduces internal O_2 concentration below a desirable and beneficial level and increases the CO_2 concentration above critical tolerable limits. This leads to anaerobic fermentation. O_2 and CO_2 transmission rates of corn-zein (CZ) protein film were much lower than those of a typical shrink wrap film (SWF), whereas WVTR of the film was much higher than a typical SWF. CZ film had an effect of delaying color change of tomatoes during the storage, and the degree of color change was mainly dependent on the coating thickness.

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PACKAGING, HANDLING AND STORAGE OF LIPOXYGENASE-FREE FULL-FAT SOY FLOUR

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ABSTRACT

Lipoxygenase-free full-fat soy flour was prepared by hot water blanching and hot-air-oven heat treatment to raw soybeans. Whole soybeans, splits and flours of raw and treated soybeans were used to evaluate their packaging and handling characteristics in polythene bags up to 400 μm thickness. Storage quality of differently processed soy flours was judged in polythene bags of 100 μm. Polythene bags of 50 μm film thickness were judged suitable by stack load test for packaging of whole soybean, soy-splits and soy flours. For safe handling, however, the minimum film thickness of 200 μm was found to be safe as decided by the drop test. 100 μm film thickness of polythene bags was considered appropriate for safe storage up to 45 days in ambient conditions (5–36C; 6–97% rh) on account of FFA rise and change in moisture content. The lipoxygenase-free full-fat soy flour produced by dry heat was found to have better storage quality over raw and blanched soy flour. Overall, the polythene bag film thickness of 200 μm is considered for safe packaging, handling and storage of full-fat soy flour up to 45 days under ambient conditions.

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INTRODUCTION

Prior to being subjected to processing operations, soybeans need to be stored so as to maintain requisite quality. After processing, the products are more difficult to store. So proper packaging is a must for powdered products. Various techniques and packaging materials have been used for storage of soybeans and soy products. A brief account of the storage aspects and the quality criteria is presented.

Polythene bags (175 μm) and metal containers were reported to be the best for maintaining quality of full-fat soy flour obtained by hot water blanching, throughout the storage period of 10 months, as evidenced by no change in free fatty acids (FFA), moisture content and insect infestation (Gandhi *et al.* 1985). Raw soy flour absorbed relatively more moisture as compared to soy flour produced by blanching when stored in polythene bags ($<$ 15 μm) at different temperatures (5–40C) and relative humidities (45–92% rh). A slight increase from 0.4 to 0.6% in FFA was observed during 4 months storage at 25–40C and 40% relative humidity (Mishra 1987). The FFA rise was negligible up to 45 days of storage of full-fat soy flour (Mishra 1987; Seth and Nath 1989). Polythene film of 4.8 μm thickness was reported best for storage of lipoxygenase-free flour with 0.02% antioxidant—butylated hydroxytoluene (BHT)—incorporation at 40C, 92% rh (Seth and Nath 1989). Only about 1.5–2% decrease in nitrogen solubility index (NSI) was reported for storage of full-fat soy flour for 45 days (Mishra 1987; Seth and Nath 1989). In the case of soybeans, dry heat is effective in improving oxidative stability. Dry-heat-treated soy flour samples showed low peroxide formation, relatively low FFA and excellent flavor stability after two years storage (Mustakas *et al.* 1969).

Earlier studies revealed that dry heat treatment does not have any deleterious effect on product quality and helps to retain it during storage. The objective of this work was to identify the proper polythene film thickness for preparing suitable bags for safe packaging and handling of whole soybeans, splits and flour, and judge the storage quality of lipoxygenase free full-fat soy flour produced by different methods. This study focuses on the effect of heat treatment mode on storage and handling quality characteristics of different soy products investigated.

MATERIALS AND METHODS

Material

Raw whole soybeans (cv. JS-7244) and soy flour, hot water blanched soy

splits and dry-heat-treated whole soybeans and splits were used for the study. Lipoxygenase-free full-fat soy flours were produced by hot water blanching (Gandhi *et al.* 1985) and dry heat treatment (Kulkarni 1992). For production of soy flour by blanching process, the raw soybeans were dehusked and split in dehuller. Soy splits were blanched in boiling water for 25 min, dried to about 10% moisture content dry basis (d.b.), milled in laboratory hammer mill (Fritsch Make, Germany) and sieved to pass through Indian Standard Sieve (ISS) No. 25. The dry- heat-treated lipoxygenase-free soy flour was produced by heat treatment of whole soybeans at 11% d.b. in a hot air oven at 200C for 10 min (Kulkarni 1992). Dry-heat-treated samples were cooled in desiccator and milled in laboratory hammer mill (Fritsch Make, Germany) and sieved to pass through ISS No. 25. Residual lipoxygenase activity of full-fat soy flour was estimated by a standard method (Axelrod *et al.* 1981). Lipoxygenase-free full-fat soy flours thus produced were used for storage quality evaluation and also for experiments on packaging and handling.

Packaging Quality

Polythene bags with different sheet thicknesses were used for packaging the soy products. The polythene bags were subjected to impact and stack load tests (Surya Nath and Ganguly 1983) to judge their suitability for safe packaging, storage and handling for different soy products, namely, whole soybeans, soy splits and flour. Polythene bags with film thickness ranging from 50 μm to 400 μm thickness were used for the tests.

Impact Test. Five bags each of 50, 100, 150, 200 and 400 μm were selected. They were filled with different soy products (Table 1), sealed and dimensions measured. Quantity of product filled in bag varied due to particle size and density. Each bag was dropped up to 15 times on a flat cemented floor from a height of 1.25 m and examined for intactness.

Stack Load Test. Five bags, each having a specific thickness from 50 to 400 μm were filled with soy products and sealed. Each bag was placed in between two smooth stainless steel plates. The upper plate was then loaded with 49, 98, 147, 196, 245, 294, 343 and 373 Newtons (N) static load and observed for failure at the sealing line. In one set, five bags of 100 μm thickness, containing different soy products (Table 1), were stacked in one stack, loaded with 373 N and observed for 24 h.

TABLE 1.
QUANTITIES OF SOY PRODUCTS AT FILLING OF POLYTHENE BAGS

Product	Sample mass, g	Bag size, mm x mm
Whole soybeans	480	175.0 x 155.0
Soy-splits (raw & heat treated)	450	175.0 x 155.5
Soy-splits (blanched & dried)	440	177.5 x 155.0
Soy flour (- 25 ISS)	280	175.5 x 154.5

ISS : Indian Standard Sieve (Opening 0.25mm)

Storage

The soy flour samples (25 ISS) were stored under ambient conditions in polythene bags of 100 μm film thickness for 45 days. Daily temperature and relative humidity data were recorded. Samples were drawn from the packets to simulate the practice normally followed in a domestic kitchen. The first sample was drawn after 10 days and subsequently an interval of seven days was maintained (Table 3). Samples were analyzed for moisture content and FFA. The FFA were estimated by following the standard procedure (AOAC 1975) and expressed as percent of oleic acid. Initial and final moisture contents were determined by hot air oven method (AACC 1969) by exposing 2 g of powdered sample to a temperature of 130C for 2 h.

RESULTS AND DISCUSSION

Packaging Material Quality Evaluation

Drop and stack load tests were used to judge the suitability of polythene bags with sheet thickness ranging from 50 μm to 400 μm for safe storage and handling of soy products.

Drop Test. It was found that 50 and 100 μm sheet thicknesses were not suitable (Table 2) for packaging of any of the soy products investigated.

TABLE 2.
DROP TEST PERFORMANCE OF PACKAGING MATERIAL FOR VARIOUS SOY PRODUCTS
(Height of drop 1.25 m; maximum no. of drops: 15)

Polythene sheet thickness μm	Performance parameter	Soy products											
		Whole soybean			Soy splits ppp			Soy splits blanched			Soy flour		
	Replication --	1	2	3	1	2	3	1	2	3	1	2	3
50	Burst,n th drop (B)	1	1	1	1	1	1	1	1	1	1	1	1
	Location (L)												
	(Side / top)	-	-	-	-	-	-	-	-	-	-	-	-
	(Seal)	+	+	+	+	+	+	+	+	+	+	+	+
100	B	1	1	1	1	1	1	1	1	1	1	4	5
	L (Side / top)	-	-	-	-	-	-	-	-	-	-	+	+
	(Seal)	+	+	+	+	+	+	+	+	-	+	-	-
150	B	+	+	+	10	4	8	7	9	+	5	+	+
	L (Side / top)	+	+	+	-	+	-	-	-	+	+	+	+
	(Seal)	+	+	+	+	-	+	+	+	+	-	+	+
200	B	+	+	+	+	6	12	+	+	+	+	+	+
	L (Side / top)	+	+	+	+	+	+	+	+	+	+	+	+
	(Seal)	+	+	+	+	-	-	+	+	+	+	+	+
400	B	+	+	+	+	+	+	+	+	+	+	+	+
	L (Side / top)	+	+	+	+	+	+	+	+	+	+	+	+
	(Seal)	+	+	+	+	+	+	+	+	+	+	+	+

- : failure, + : intact, B : burts, L : location of burst
ppp: produced by dry heat treatment

Considering the criteria of 5 drops suggested earlier for flour (Surya Nath and Ganguly 1983), 150 μm polythene bags may be regarded as safe, at least for soy flour. However, the results indicate that the number of drops should be increased to 15 in order to make the criteria widely applicable. Following the modified criteria and results obtained (Table 2), it can be concluded that 200 μm thick polythene bag is preferable for packaging of soy products.

Stack Load Test. Tests designed to ascertain the performance of packaging material during stacking showed that all the bags with different thickness withstood the maximum force of 373 N. Stacking with 373 N load, 50 μm thick polythene bags containing any one of the soy products revealed the strength of the packaging material. Another test of multiple stacking with different soy

products also indicated the material suitability even after 24 h. For stacking alone, the polythene film thickness of even 50 μm could be considered safe.

Storage Quality

Earlier work on storage of soy flour, produced by hot-water blanching, revealed that the product can be safely stored in polythene bags (175 μm) for a duration up to 10 months (Gandhi *et al.* 1985). A shelf-life of 45 days would be adequate in view of the domestic utilization pattern suggested. Therefore, the soy flour was stored up to 45 days for evaluation of shelf-life under ambient conditions.

Storage Conditions. The soy flour was stored in polythene bags having 100 μm sheet thickness. During the investigation, the ambient temperature and relative humidity varied in the range of 5–36C and 6–97% rh, respectively (Fig. 1). Initial moisture content of the soy flours, namely, raw soy flour, soy flour produced by hot water blanching and dry heat treatment was 11.5, 10.43 and 5.43% d.b., respectively (Table 3).

Moisture Content. At the end of storage period of 45 days, around one percentage point increase in moisture content was found in soy flour produced by dry heat treatment. However, maximum increase of 2.37 percentage point was observed in blanched soy flour sample as against 1.41 percentage point in soy flour produced by blanching (Table 3). Some earlier studies reported no change in moisture content and FFA when full-fat soy flour was stored in 175 μm polythene bags for 10 months under ambient conditions (Gandhi *et al.* 1985). However, more than 1% increase in moisture content of full-fat soy flour filled in < 15 μm polythene bags and stored at 15–40C and 40–92% relative humidity was reported (Mishra 1987; Seth and Nath 1989). The outcome of this study, conducted with a temperature variation of 5–36C and 6–97% rh, is to provide confirmation to the earlier conclusions. The results show less increase in moisture content of lipoxygenase-free soy flour produced by dry heat compared to other flours stored under similar conditions and indicate a possible longer shelf-life and a possibility of less spoilage due to high moisture rise during storage.

Free Fatty Acids (FFA). At the beginning of storage, the FFA content in the raw soy flour sample was 0.49% against 0.46% in blanched and dry-heat-treated soy flour. A higher initial level of FFA in raw soy flour was reported (Mishra 1987) as compared to blanched soy flour. The FFA level in all soy flours processed differently, increased with the duration of storage. Increase in

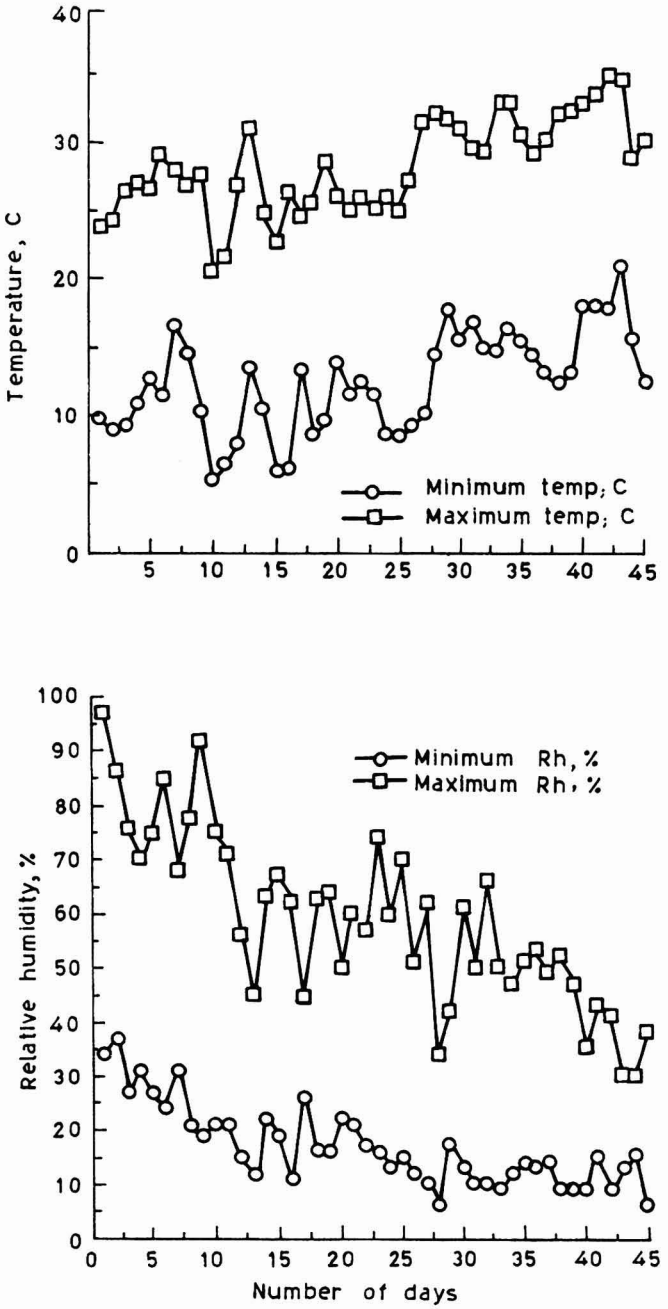

FIG. 1. VARIATION OF AMBIENT TEMPERATURE AND RELATIVE HUMIDITY DURING STORAGE PERIOD

TABLE 3.
MOISTURE CONTENT OF VARIOUS SOY FLOUR SAMPLES BEFORE AND AFTER 45 DAYS STORAGE

Storage condition			Moisture content, % d. b.	
Temp., C	rh, %	Soy flour type	Before	After
5 - 36	6 - 97	Raw	11.49	12.90
		Blanched	10.43	12.80
		ppp	5.60	6.64

ppp : produced by dry heat treatment

TABLE 4.
VARIATION OF FREE FATTY ACIDS WITH DURATION OF STORAGE FOR DIFFERENT SOY FLOURS
(% of oleic acids)

Soy flour	Duration of storage, days						
	0	10	17	24	31	38	45
Raw	0.49	0.49	0.59	0.60	0.60	0.64	0.78
Blanched	0.46	0.46	0.56	0.56	0.56	0.56	0.64
PPP	0.46	0.46	0.56	0.56	0.56	0.56	0.56

PPP : produced by dry heat treatment.

variation of free FFA was observed to be maximum for raw soy flour followed by blanched and dry-heat-treated soy flours (Table 4).

This observation reflects on the potential shelf-life of the product under any storage conditions. An increase of 0.29, 0.18 and 0.10 percentage points in FFA of raw, blanched and dry-heat-treated soy flour, respectively, depicts better storability on the basis of low FFA and moisture rise in soy flour produced by dry heat. A safe storage of blanched soy flour in 175 μm thick polythene bags

without any rise in FFA was reported earlier (Gandhi *et al.* 1985). Using them, dry heat treated soy flour could be expected to have a still longer shelf-life. The storage quality of soy flour produced by dry heat treatment, as ascertained, was not found inferior, on account of moisture and FFA rise, to the blanched or raw soy flour. Also, on FFA rise basis, our study revealed a longer shelf-life for dry-heat-treated soy flour than blanched or raw, as it would require longer duration to attain same level of FFA or moisture than other soy flours studied. It is also evident that no appreciable deterioration in the quality of this soy flour, judged on FFA rise basis, takes place during 45 days of storage. This would be adequate in view of the normal utilization pattern followed in domestic kitchens. The 100 μm thick polythene can therefore, be considered as an appropriate packaging material for storage of all the types of soy flour that were studied for at least 45 days storage.

CONCLUSIONS

Packaging and handling of different soy products, namely, whole soybean, raw and heat-treated soy splits and soy flour require consideration of the possibility of failures of packaging material and the safety of processed material. For example, 50 μm thick polythene film is safe for safe packaging of the products for stacking. However, it is clear from the studies that 200 μm thick polythene film is suitable for safe handling of the product. Polythene film that is 100 μm thick is suitable for safe storage of lipoxygenase-free full-fat soy flour up to 45 days. To take care of all the three aspects, i.e., packaging, handling and storage, 200 μm polythene film is considered appropriate for lipoxygenase-free full-fat soy flour produced either by hot water blanching or by dry heat. However, the lipoxygenase-free full-fat soy flour produced by dry heat possessed relatively better storage quality characteristics compared to raw soy flour or soy flour produced by hot water blanching. The polythene bags with 200 μm thickness should be preferred for small size packaging of soy products namely, whole soybean, soy splits and soy flour to withstand the load during stacking, ensure safe handling and guarantee good quality of the stored product.

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EFFECT OF LOW TEMPERATURE BLANCHING, CYSTEINE-HCl, N-ACETYL-L-CYSTEINE, Na METABISULPHITE AND DRYING TEMPERATURES ON THE FIRMNESS AND NUTRIENT CONTENT OF DRIED CARROTS

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ABSTRACT

Low temperature long time (LTLT) blanching (70C for 20 min) together with calcium treatment can be used to significantly improve the texture of rehydrated dried carrots when compared to high temperature short time (HTST) blanching (100C for 3 min). LTLT blanching allows pectin methyl esterase to deesterify pectin, which can then react with calcium to form salt bridges. 0.3% L-cysteine-HCl was found to be most effective in preventing ascorbic acid loss and obtaining a product with the highest rehydration ability, compared to pretreatments with 0.3% N-acetyl-L-cysteine and 0.1% sodium metabisulphite. On the other hand 0.1% sodium metabisulphite was most effective in preserving the carotenoids content of dried carrots. Ascorbic acid and rehydration ability were more adversely affected by long drying time than high drying temperature, while carotenoids were more sensitive to high drying temperature than drying time. Hence, 60C drying temperature was good for ascorbic acid and rehydration ability, while 40C drying temperature was good for carotenoid and color of dried carrots.

INTRODUCTION

Color, texture and nutrient loss occurs during processing, drying and storage of dehydrated products. Low-temperature long-time (LTLT) blanching in the range of 60–75C was found to be optimum for maintaining the texture of cooked, canned or frozen vegetables (Van Buren *et al.* 1960; Canet and Hill 1987), including canned carrots (Lee *et al.* 1979). This was because at this

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relatively low blanching temperature, pectin methyl esterase (PME) was active to deesterify and increase the free carboxyl groups on pectin, which could then form salt bridges with divalent cations to produce a firmer textured product (Van Buren *et al.* 1960). Quintero-Ramos *et al.* (1992) found that the texture and rehydration of dehydrated carrots are improved LTLT blanching (60–65C for > 30 min). Seow *et al.* (1993) also reported that LTLT followed by high-temperature short-time (HTST) blanching improved the dehydration, rehydration and texture of rehydrated vegetables.

Loss of ascorbic acid during processing and storage can be reduced by pretreatment with SO_2 (Bender 1958; Bolin and Stafford 1974) and cysteine-HCl (Mohamed *et al.* 1993). Molnar-Perl and Friedman (1990) reported that N-acetyl-L-cysteine and reduced glutathione were more effective than L-cysteine and almost as effective as sodium sulfite in preventing browning in apples and potatoes. This study attempted to compare the effects LTLT blanching, drying temperatures and various additives (N-acetyl-L-cysteine, cysteine-HCl, metabisulfite and calcium) on the texture, ascorbic acid and carotenoids content of dried carrots.

MATERIALS AND METHODS

Carrots *(Daucas carota)* purchased from the local market, were trimmed, scraped, washed, cut into 1 cm cubes and thoroughly mixed. Blanching was done (using a 3 × 3 factorial experimental design) in a water bath containing distilled water at (1) 70C for 20 min (LTLT), (2) 100C for 3 min (HTST), (3) 80C for 20 min (LTLT) and containing the following additives: (1) control, (2) 2% glycerol, and (3) 2% glycerol and 1% $CaCl_2$. The carrots were dried at 40C (Memmert-natural ventilation oven of air speed 0.1m/s at 40% relative humidity) to constant weight.

The second set of experiments were carried out on diced carrots, which had been blanched at 70C for 20 min, containing the following additives:

A. 2% glycerol and 1% $CaCl_2$
B. 2% glycerol, 1% $CaCl_2$ and 0.1% Na metabisulfite
C. 2% glycerol, 1% $CaCl_2$ and 0.3% L-cysteine-HCl
D. 2% glycerol, 1% $CaCl_2$ and 0.3% N-acetyl-L-cysteine

and dried at 40C, 50C and 60C to constant weight (4 × 3 factorial design).

Rehydration were done by placing 25 g samples in boiling water for 15 min, then drained for 5 min, weighed and expressed as the amount of water gained per gram of dry solids. Moisture content was measured by oven drying at 105C

overnight (AOAC 1980). Ascorbic acid was determined by titrating with a standardized 2,6-dichlorophenolindophenol in the presence of HPO_3 (AOAC 1980). Carotenoids were analyzed by a modified AOAC (1980) method whereby 2 g dried samples were soxhlet extracted with petroleum ether containing 30% hexane. The extract was made to volume with petroleum ether and the solution read at 436 nm against a standard curve. Hardness was taken as the average yield stress of at least six individual cubes, using an Instron 1140 Universal testing machine with an 8 mm diameter plunger, at 50 mm min^{-1} crosshead speed to 50% deformation.

Sensory evaluations were done by 15 taste panelists for color, shape, flavor, texture and overall acceptability of fresh and rehydrated carrots served in the form of beef stew, on a hedonic scale of 1–9 (9 = most liked). The carrots used for sensory evaluation were those dried at 40C because they had the most desirable appearance compared to those dried at the higher temperatures. The beef stew was prepared by combining the dried or fresh diced carrots with water, diced beef and other ingredients (tomato puree, celery, onions, cornstarch, salt, sugar and monosodium glutamate) into cans, which were sealed, retorted at 121C for 20 min, then cooled and stored at room temperature.

Data were statistically evaluated for Analysis of Variance (ANOVA) and Duncan's Multiple Range Test (DMRT) on SAS statistical program, using an IBM compatible PC.

RESULTS AND DISCUSSION

The texture of rehydrated dried carrots, which were blanched at 70C, was significantly firmer than those blanched at 80C or 100C (Table 1). Those given 1% $CaCl_2$ and 2% glycerol were much firmer than the glycerol treated or control samples. These results show that LTLT could be used to improve not only the texture of frozen and canned vegetables (Steinbuch 1976; Lee *et al.* 1979) but also dehydrated vegetables.

Samples treated only with glycerol and $CaCl_2$ rehydrate poorly at all the drying temperatures studied (Table 1). As a trend samples containing S-compounds dried at the higher temperature (60C) rehydrate better than those dried at the lower temperatures (40C or 50C). At the lower drying temperatures samples with L-cysteine-HCl treatment showed significantly better rehydration ability compared to those treated with Na metabisulphite or N-acetyl-L-cysteine. The improved rehydration characteristics may be because these treatments helped protect the amino groups on the protein from undergoing Maillard reactions, facilitating rehydration.

TABLE 1.
CHARACTERISTICS OF DEHYDRATED CARROTS WITH DIFFERENT PRETREATMENTS AND DRYING TEMPERATURES

PRETREATMENTS \ BLANCHING TEMPERATURE	FIRMNESS OF REHYDRATED CARROTS (Kg) 70 C	80 C	100 C
CONTROL UNTREATED	1.99^{A}	1.24^{B}	0.46^{C}
GLYCEROL	2.11^{A}	1.12^{B}	0.58^{C}
GLYCEROL AND $CaCl_2$	3.22^{A}	2.46^{A}	1.15^{B}

PRETREATMENT	REHYDRATION ABILITY (g/g dry solids)	ASCORBIC ACID (mg/100g)	CAROTENOIDS CONTENT (µg/g)
40 C			
Glycerol and $CaCl_2$ (Control)	2.13^{e}	8.63^{de}	1350.08^{c}
Glycerol, $CaCl_2$ and Na metabisulphite	2.15^{de}	9.78^{cde}	2616.83^{a}
Glycerol, $CaCl_2$ and Cys-HCl	2.25^{bc}	10.93^{bc}	1427.36^{bc}
Glycerol, $CaCl_2$ and acetyl cysteine	2.12^{e}	10.93^{bc}	1633.27^{b}
50 C			
Glycerol and $CaCl_2$ (Control)	2.10^{e}	8.34^{e}	529.03^{f}
Glycerol, $CaCl_2$ and Na metabisulphite	2.10^{e}	10.64^{bcd}	1194.99^{cd}
Glycerol, $CaCl_2$ and Cys-HCl	2.39^{a}	12.36^{ab}	778.51^{e}
Glycerol, $CaCl_2$ and acetyl cysteine	2.21^{cd}	11.21^{bc}	684.63^{ef}
60 C			
Glycerol and $CaCl_2$ (Control)	2.11^{e}	8.63^{e}	582.45^{ef}
Glycerol, $CaCl_2$ and Na metabisulphite	2.32^{ab}	8.91^{de}	1101.64^{d}
Glycerol, $CaCl_2$ and Cys-HCl	2.31^{b}	14.09^{a}	584.00^{ef}
Glycerol, $CaCl_2$ and acetyl cysteine	2.25^{bc}	12.26^{ab}	657.14^{ef}

	SENSORY HEDONIC SCORES color	shape	texture	flavor	overall
40 C					
Fresh carrot	6.60^{a}	8.27^{a}	7.60^{a}	7.20^{a}	7.67^{a}
Glycerol and $CaCl_2$ (Control)	6.67^{a}	6.20^{b}	5.67^{b}	6.73^{a}	6.20^{b}
Glycerol, $CaCl_2$ & Na metabisulphite	7.00^{a}	6.47^{b}	5.93^{b}	6.80^{a}	6.53^{b}
Glycerol, $CaCl_2$ and Cys-HCl	7.13^{a}	6.33^{b}	5.87^{b}	6.87^{a}	6.47^{b}
Glycerol, $CaCl_2$ & acetyl cysteine	7.20^{a}	7.20^{a}	5.73^{b}	6.80^{a}	6.80^{b}

values followed by the same letters are not significantly different at 5% level for each product.

The ascorbic acid content of dried carrots showed an almost similar trend to that of rehydration ability, i.e., samples that were dried at the fastest rate and treated with L-cysteine-HCl had the highest content (Table 1). However although cysteine-HCl treated dried carrots had the highest ascorbic acid content,

the values were not significantly different from those treated with N-acetyl-L-cysteine. The mechanism for protection of ascorbic acid by cysteine-HCl was tkought to be due to the reduction of o-quinones to a colorless complex (possibly hydroquinones), forstalling quinones reaction with ascorbic acid. Cysteine-HCl was only effective in inhibiting enzymic browning but not nonenzymic browning (Mohamed and Leong 1987).

The rate of drying of the diced carrots at the various temperatures is shown in Fig 1. At 60C, it took 22 h to dry the carrots to constant weight (about 9.8% moisture), whereas at 40C it took 42 h. Ascorbic acid, which is easily destroyed, is adversely affected by the long drying time.

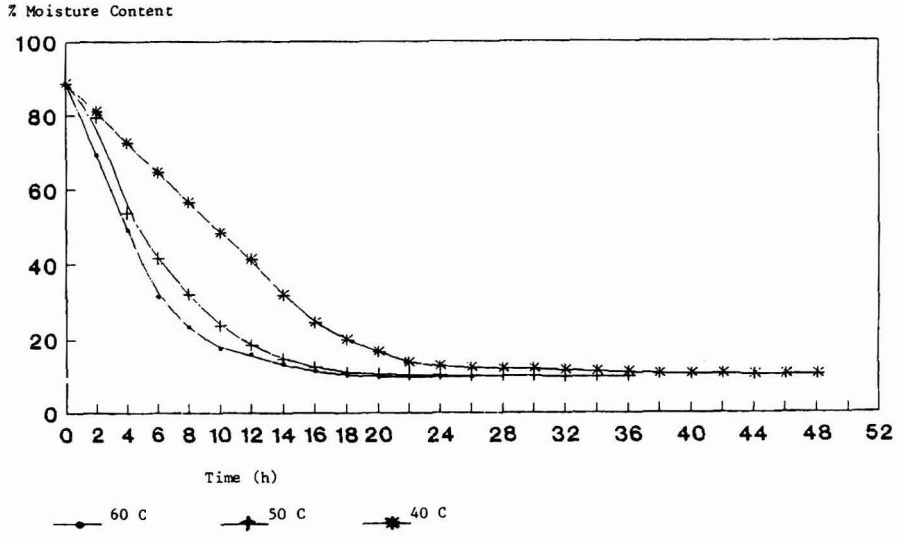

FIG. 1. RATE OF MOISTURE LOSS IN CARROTS DRIED AT DIFFERENT TEMPERATURES

A somewhat different pattern was observed for the carotenoids content where samples containing the highest carotenoids were those dried at 40C and given Na metabisulphite treatment (Table 1), showing that carotenoids were sensitive to drying temperature more than drying time. The oxidation of carotenoids could be retarded by Na metabisulphite but not cysteine. A significant correlation between the development of off-flavor and carotenoid destruction has been reported by Falconer *et al.* (1964) for freeze-dried carrot powder stored at 18C. Visual assessment also showed that the samples dried at 40C had the most desirable color compared to those dried at the higher temperatures.

The rehydrated 40C dried carrots given different treatments were not significantly different in color compared to fresh carrots when served in the form of beef stew, although their scores were generally higher than the fresh carrots. N-acetyl-L-cysteine treated dried carrots were similar in appearance to the fresh carrots. The scores were high for texture, flavor, and overall acceptability for all the rehydrated 40C dried carrots, although significantly lower than the fresh carrots (Table 1).

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