The visual quality of minimally processed lettuces stored in air or controlled atmosphere with emphasis on romaine and iceberg types

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Abstract

Five different types of lettuce (iceberg, romaine, butter, green leaf, and red leaf) were prepared as salad pieces and stored for 16 days at 5°C in air or in a controlled atmosphere (CA) (3% $\text{O}_2$ + 10% $\text{CO}_2$). Samples were evaluated for overall visual quality, surface and edge browning, and russet spotting. After 8 days, differences in overall visual quality between air and CA-stored samples were observed; after 12 days, air-stored samples were below the limit of salability, whereas visual quality was maintained in CA-stored pieces from all lettuce types except butter lettuce. Transferring lettuce to air for 12 h at 15°C accentuated the differences between storage atmospheres. An analysis of the various components of overall visual quality showed that surface and edge browning were the defects which most contributed to a decrease in quality. Among the lettuce types studied, the CA benefit for overall visual quality was highest for iceberg lettuce, and some differences between cultivars were observed. There was no CA benefit for butter lettuce, but CA induced surface discoloration and tissue softening. Pre-processing storage of lettuce heads at 5°C for 7 and 14 days negatively affected overall visual quality and leaf edge browning of the prepared salad pieces; this intact head storage effect was more evident on romaine than iceberg pieces.

Keywords: Iceberg; Specialty lettuce; CO$_2$ injury; Visual quality; Surface and edge browning

1. Introduction

The effects of modified atmosphere (MA) or controlled atmosphere (CA) on the quality of minimally processed or salad-cut lettuce have been studied by several researchers (Ballantyne et al., 1988; Bolin and Huxsoll, 1991; Mateos et al., 1993a, 1993b; McDonald et al., 1990). The atmospheres commonly used in the packages of prepared lettuce contain 10–20% CO$_2$ and 1–8% O$_2$. Benefits from MA have been
reported even at optimal storage temperatures of 0 to 2°C for minimally processed lettuce (Krahn, 1977). The main advantage in using MA for minimally processed lettuce is the delay of senescent browning (SB). Ke and Saltveit (1989) and Mateos et al. (1993a) showed that high CO₂ atmospheres delay browning by limiting the production of phenolic compounds.

Using film packages with appropriate permeability, Ballantyne et al. (1988) found that an initial flushing with low O₂ and high CO₂ helped to rapidly establish an equilibrated MA of 1–3% O₂ and 5–6% CO₂ and provided an important extension to storage life compared to bags sealed without flushing. McDonald et al. (1990) reported no discoloration or fermentation of cut lettuce when CO₂ was <20% and O₂ was 1–4%, in naturally developed MA after vacuum sealing. Bolin and Huxsoll (1991) showed that MA with headspace O₂ <2% and CO₂ levels of ~10% resulted in the development of off-flavors in vacuum-packed lettuce, presumably because of the onset of anaerobiosis.

A higher accumulation of anaerobic volatiles, ethanol and acetaldehyde, was found in intact heads as compared to cut lettuce when stored in air + 20% CO₂ (Mateos et al., 1993a). The levels of ethanol and acetaldehyde in intact and cut lettuce stored under 5 or 10% CO₂ were not different from those of the air-stored lettuce. Another possible disadvantage of the use of high CO₂ atmospheres in cut lettuce is the development of brown stain (BS). This disorder has been extensively studied in intact heads (Brecht et al., 1973a; Kader et al., 1974; Siripanich and Kader, 1986) and can be induced by CO₂ levels >2% at temperatures <10°C. The severity of BS is increased by low temperatures (0 to 2.5°C), low O₂ atmospheres, and by transfer after storage to temperatures >10°C (Brecht et al., 1973a, 1973b, 1973c). Mateos et al. (1993b) reported more BS in cut lettuce midribs than in intact heads stored in air + 20% CO₂ for 20 days at 2.5°C and transferred to air for 12 h. For cut lettuce products, however, BS generally has less effect on visual quality than does browning of the cut surface (Mateos et al., 1993b).

Although iceberg lettuce is predominantly used for prepared salads, other types of lettuce are now used in salad mixes. Information on the postharvest behavior of these other lettuces is lacking, especially with regard to their quality in salad packs. This research compared the visual quality and shelf life of various types of lettuce prepared as minimally processed product and stored under air or CA.

2. Materials and methods

Plant material

Lettuce (Lactuca sativa) grown under commercial conditions in the Salinas Valley (California) from May to August 1994 was used for the experiments. The lettuce was field packed, vacuum cooled and transported to Davis in air-conditioned vehicles and stored overnight at 2.5°C. Commercial cultivars suitable for Salinas growing conditions of 5 types of lettuce (iceberg, romaine, butter, green leaf and red leaf) were used. For one experiment two distinct cultivars of iceberg lettuce, ‘Alpha’ and ‘Legacy’, were used.

Product preparation

Wrapper and core leaves were discarded. Leaves were placed on a cutting board held on crushed ice and tissue segments (5 × 3 cm) containing leaf and midrib were cut using
a sharp knife. These segments approximate the size of pieces used in 'salad packs'. The pieces were rinsed with 5°C water containing 50 µl l⁻¹ NaOCl and centrifuged in a manual salad spinner (Copco® Salad Spinner, Wilton Enterprises, Woodbridge, IL) which removed excess water without causing any visible damage to the lettuce. About 75 g of cut tissue was placed in a 250-ml glass jar, and the top covered with two layers of cheesecloth fixed with a rubber band. Groups of 3 jars were placed into polyethylene bags at 5°C and connected to flow-through systems providing humidified atmospheres of either air or 3% O₂ + 10% CO₂ balanced with nitrogen; gas concentrations were maintained within 10% of the indicated conditions. Previous work demonstrated the effectiveness of this CA in maintaining the quality of cut lettuce (Krahn, 1977; Mateos et al., 1993b). A temperature of 5°C was used as it is a refrigerated condition under which many salad products are commercially handled.

Quality evaluations

Lettuce quality was evaluated using the following parameters and scoring systems:

1. Overall visual quality (OVQ) was rated on a scale from 9 to 1, where 9 = excellent and 1 = unusable
2. Leaf surface browning (LSB) and leaf edge browning (LEB) were scored on a scale from 1 to 5, where 1 = no browning and 5 = severe browning
3. Russet spotting (RS) was scored on a scale from 1 to 9, where 1 = none and 9 = severe.

For each of these defects, an index was calculated by multiplying the scores for severity by the percentage of pieces affected. An OVQ rating of 6 was considered the minimum score required for salability to consumers; this score indicated an overall visual quality between good (score of 7) and fair (score of 5), with defects not exceeding moderate levels.

Surface and edge color of midrib pieces were determined by a color difference meter (Minolta Chroma Meter CR-200) using a white plate for calibration (L = 97.63; a = -0.53; b = 2.38). Color values were correlated with the results of the visual evaluations.

Quality evaluations were conducted every 4 days over the 16-day storage period. The evaluations were carried out immediately after removal of the samples from storage conditions and again after transfer to air at 15°C for 12 h.

Statistical analysis

Three replicates were used per treatment in each experiment. Data were analyzed by ANOVA with mean separation by Duncan's New Multiple Range test. Multiple regression analysis was performed to estimate the contribution of the different quality components to overall visual quality score.

3. Results

Quality of minimally processed lettuces after storage in air or CA

After 4 days of storage, the visual quality of the air-stored lettuces was not different from that of CA-stored lettuces. After 8 days, the visual quality of the air-stored lettuces was significantly less than that of the CA-stored lettuces, and was at the limit of
salability (score 6). After 12 days, the quality of all the air-stored lettuces was below the limit of salability, whereas the 3% O₂ + 10% CO₂ atmosphere maintained high OVQ scores for iceberg, romaine and green leaf salad pieces (Fig. 1). There were important differences in behavior among the types of lettuce after transfer from CA to air. Iceberg lettuce had the lowest quality after air storage, but had the highest quality after CA storage (Fig. 1). In addition, there was no loss of OVQ in iceberg after transfer from CA to air, whereas significant losses were observed for green leaf and romaine lettuces from the same treatments.

The decline in OVQ for air-stored iceberg and romaine lettuces followed a concave pattern, with the highest loss between days 4 and 8 (Fig. 2). The loss of OVQ in the CA-stored lettuces was very slow until after day 12. The visual quality of the air-stored iceberg pieces declined more rapidly than that of the romaine pieces, whereas under CA, the opposite was observed.

The main effect of CA on quality of minimally processed lettuce was to prevent senescent browning, especially on the cut leaf edges (Table 1). After 8 days, CA storage
significantly reduced leaf edge discoloration in comparison to air-stored tissue from all lettuce types. The highest LEB indices were observed in air-stored romaine lettuce pieces after 12 days. A score of 3 indicates moderate browning, sufficient to seriously reduce the marketability of the product.

Symptoms of BS were observed in CO₂-treated samples, while in air-stored product discoloration was due to SB. BS appears first on the midrib surfaces as a variable-sized sunken or yellowed area which later can become more delineated and brown. Severe CO₂ injury was observed in butter lettuce; symptoms were tissue softening and generalized discoloration. Despite differences in the etiology of surface discoloration, SB, BS and other CO₂ discolorations were scored under the category of LSB. Incipient BS was observed on the midribs of the 10% CO₂ treated salad pieces within 4 days for butter lettuce, 8 days for romaine, green and red leaf lettuces, and 12 days for iceberg lettuce. To confirm the accuracy of the subjective scales used for scoring LSB and LEB, these scores were compared with \( L, a, \) and \( b \) color values determined on the surface and an angled cut edge of midrib tissues. The LSB scores on air-stored tissue had correlation coefficients of \(-0.58, 0.97\) and \(0.90\) for the \(L, a\) and \(b\) color values, respectively. The LEB scores had correlation coefficients of \(-0.87, 0.98\) and \(0.92\) for the same respective color values. The \(a\) color value was most highly correlated with visual evaluations.

The benefit of using CA to preserve a specific quality parameter was estimated by calculating the percentage difference of the CA-stored versus the air-stored values; this was called ‘CA benefit’. The greater the calculated benefit, the greater is the expected effectiveness of an atmosphere in maintaining a specific quality attribute. The use of CA (3% O₂ + 10% CO₂) was beneficial to the OVQ in 4 of the 5 types of lettuce studied, and its importance generally increased with time in storage (Fig. 3). This CA, however,
Table 1
Effects of air and CA (3% O₂ + 10% CO₂) on leaf edge browning (LEB) and leaf surface browning (LSB) of 4 types of minimally processed lettuce evaluated after 4, 8 and 12 days at 5°C and after transfer to air at 15°C for 12 h. LEB and LSB were scored on a scale of 1 to 5, where 1 = no browning and 5 = severe browning. The browning index was calculated by multiplying the scores for severity by the percentage of pieces affected. Within a column for a given parameter, different letters indicate significant differences at the 5% level.

<table>
<thead>
<tr>
<th>Browning index</th>
<th>Iceberg</th>
<th>Romaine</th>
<th>Green Leaf</th>
<th>Red Leaf</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>4 d</td>
<td>8 d</td>
<td>12 d</td>
<td>4 d</td>
</tr>
<tr>
<td>LEB Air</td>
<td>1.23 a</td>
<td>1.65 b</td>
<td>2.38 b</td>
<td>1.05 a</td>
</tr>
<tr>
<td>Air + 12 h</td>
<td>1.23 a</td>
<td>2.83 a</td>
<td>3.63 a</td>
<td>1.05 a</td>
</tr>
<tr>
<td>CA</td>
<td>1.03 b</td>
<td>1.01 c</td>
<td>1.33 c</td>
<td>1.00 a</td>
</tr>
<tr>
<td>CA + 12 h</td>
<td>1.03 b</td>
<td>1.01 c</td>
<td>1.33 c</td>
<td>1.00 a</td>
</tr>
<tr>
<td>LSB Air</td>
<td>1.04 a</td>
<td>1.89 b</td>
<td>2.04 a</td>
<td>1.03 a</td>
</tr>
<tr>
<td>Air + 12 h</td>
<td>1.04 a</td>
<td>3.13 a</td>
<td>1.96 a</td>
<td>1.03 ab</td>
</tr>
<tr>
<td>CA</td>
<td>1.00 a</td>
<td>1.01 c</td>
<td>1.33 a</td>
<td>1.00 a</td>
</tr>
<tr>
<td>CA + 12 h</td>
<td>1.00 a</td>
<td>1.01 c</td>
<td>1.33 a</td>
<td>1.06 b</td>
</tr>
</tbody>
</table>
Fig. 3. Benefit of CA (3% O₂ + 10% CO₂) to the overall visual quality (OVQ) of 5 types of minimally processed lettuce. Lettuce samples were evaluated after 12 days at 5°C plus 12 h in air at 15°C. Benefit was determined from the differences in the scores of air-stored and CA-stored lettuce and that value was transformed as a percentage of the score in air. For a given time of evaluation, different letters indicate significant differences among types of lettuce at the 5% level.

was not beneficial to the OVQ of butter lettuce, having a negative effect even in the early days of the experiment. For the other lettuce types, the benefit of CA became notable by day 8. At day 12, CA provided a significantly greater benefit for iceberg lettuce than it did for romaine, green leaf and red leaf lettuces. At day 17, however, the CA benefit was similar for iceberg, green leaf and red leaf lettuces, and greater than for romaine lettuce. The CA used in this study was developed for cut iceberg lettuce and although it maintained the overall visual quality of this particular type of lettuce, it was less effective for the other types.

The calculation of CA benefits for LEB showed no differences among the cut lettuces tested (data not shown). Over the 16-day period, the average CA benefit for LEB was estimated at 72%; for this defect, CA benefit was similar before and after transfer to air at 15°C for 12 h. The average CA benefit for OVQ showed significant differences between butter lettuce and the other types of lettuce studied (Fig. 4).

Effect of storage of intact lettuces on quality of the minimally processed products.

Heads of iceberg and romaine lettuces were stored for up to 2 weeks at 5°C in air before processing. The salad pieces were then stored at 5°C in CA (3% O₂ + 10% CO₂). Salad pieces prepared from both lettuce types differed in quality at day 12 (Table 2). For iceberg lettuce, storage did not affect the OVQ and LEB values of the salad pieces at day 12. If the salad pieces were evaluated after transfer to air at 15°C for 12 h, however, LEB indices increased as storage time of the intact heads increased. The negative effect of intact head storage was more evident on the quality of the romaine salad pieces. A 7 or 14 day storage period resulted in a substantial increase in LEB of the romaine salad pieces, both before and after transfer to 15°C.
After storage + 12 h

Fig. 4. Average benefit of CA (3% O₂ + 10% CO₂) to the overall visual quality (OVQ) of 5 types of minimally processed lettuce over a 17-day storage period at 5°C. Lettuce pieces were evaluated after storage and again after transfer to air at 15°C for 12 h. The percentage benefit was calculated as described in Fig. 3. For a given evaluation, different letters indicate significant differences at the 5% level.

Table 2
Differences in overall visual quality (OVQ) and leaf edge browning (LEB) of minimally processed iceberg and romaine lettuce prepared from heads stored in air for 0, 7 and 14 days at 5°C. The lettuce pieces were evaluated after storage for 12 days in 3% O₂ + 10% CO₂ and again after transfer to air at 15°C for 12 h. OVQ was scored on a scale of 9 to 1, where 9 = excellent and 1 = unusable. LEB was scored on a scale of 1 to 5, where 1 = no browning and 5 = severe browning. The LEB index was calculated by multiplying the scores for severity by the percentage of pieces affected. For a given parameter and lettuce type, different letters indicate significant differences at the 5% level.

<table>
<thead>
<tr>
<th>Overall visual quality</th>
<th>Leaf edge browning index</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Iceberg</td>
</tr>
<tr>
<td></td>
<td>12 d</td>
</tr>
<tr>
<td>0</td>
<td>5.2 a</td>
</tr>
<tr>
<td>7</td>
<td>6.50 a</td>
</tr>
<tr>
<td>14</td>
<td>6.50 a</td>
</tr>
</tbody>
</table>

The quality of salad pieces prepared from different iceberg cultivars

Minimally processed iceberg lettuce cultivars Alpha and Legacy, known to differ in their susceptibility to RS, were compared. Important differences were found in the quality of the salad pieces prepared from the two cultivars during storage. The calculated CA benefits for OVQ and RS were higher for 'Alpha', the cultivar more resistant to RS (Fig. 5). The CA benefit for LEB, however, was similar between the two cultivars.

Factors affecting the overall visual quality of minimally processed lettuces

The contribution of specific defects (LEB, LSB, RS) to the OVQ was calculated by multiple regression analysis. From all the experiments conducted, which included 5 types of lettuce, the following equation was derived: $OVQ = -1.10 \times LSB - 0.966$
Fig. 5. Average benefit of CA (3% O₂ + 10% CO₂) to (A) overall visual quality (OVQ), (B) leaf edge browning (LEB), and (C) russet spotting (RS) of minimally processed iceberg lettuce from cvs. Alpha and Legacy. Lettuce pieces were evaluated over a 16-day storage period at 5°C before and after transfer to air at 15°C for 12 h. The percentage benefit was calculated as described in Fig. 3. For a given parameter and evaluation period, different letters indicate significant differences at the 5% level.

\[ \text{LEB} - 0.26 \text{RS} + 10.58 \quad (R^2 = 0.76) \]. All coefficients were significant at the 5% level. The contribution of LSB and LEB to OVQ is negative and about the same magnitude. In this general case, RS contributed relatively little to a decreased OVQ score because its appearance in prepared salad product is uncommon. However, if salad pieces are prepared from RS sensitive cultivars, RS can be a significant contributor to a decreased OVQ score. The equation calculated from ‘Alpha’ and ‘Legacy’ data is \[ \text{OVQ} = -2.12 \text{LSB} + 0.043 \text{LEB} - 0.501 \text{RS} + 10.64 \quad (R^2 = 0.83) \].
4. Discussion

Browning is the major cause of quality loss in minimally processed lettuce. The onset of SB varied among the lettuce types, but an atmosphere of 3% O₂ + 10% CO₂ effectively controlled SB. Differences in browning potential among the lettuces may be attributed to variations in phenolic metabolism as reported for other disorders such as RS (Hyodo et al., 1978). López-Gálvez et al. (1995) showed that higher levels of PAL (phenylalanine ammonia lyase) activity could be induced in cut butter and romaine pieces than in pieces of other types of lettuce.

High CO₂ atmospheres are beneficial for the control of SB, but can be injurious (Ke and Saltveit, 1989). In addition to BS, CO₂ injury may include tissue softening and a generalized surface discoloration. Mateos et al. (1993b) reported that midribs of iceberg salad pieces were more affected by BS than were green tissue or midribs of intact heads. BS was observed in cut midribs 10 days after exposure to air + 10% CO₂ at 2.5°C. In the present study conducted at 5°C, BS was initiated in CA-treated midribs of butter lettuce within 4 days, romaine within 8 days and iceberg within 12 days. Among the 5 types of cut lettuces, butter and romaine were the most severely injured by the CA. The extreme sensitivity of butter lettuce to CO₂ was also reported by Tataru and Weichmann (1974) on intact heads. They found that storage in air + 2% CO₂ resulted in better quality than storage in air + 4% CO₂ or air. Lipton (1987) reported BS on midribs of intact 'Parris Island' romaine heads stored at 0 to 5°C for 2 weeks in 3% O₂ + 7.5% CO₂, but not in air + 5% CO₂. Lipton concluded that intact romaine is more tolerant to CO₂ than iceberg lettuce, but this appears not to be the case for the minimally processed product. Product preparation may increase sensitivity to CO₂ as reported for minimally processed iceberg lettuce (Mateos et al., 1993a). In addition, there is considerable variation in iceberg cultivar susceptibility to CO₂ (Brecht et al., 1973c) and that may also be the case for romaine cultivars.

In the salad product form, butter and romaine were more sensitive to the CA than iceberg lettuce. The physiological basis for this variable response among cultivars or types of lettuce is unknown (Lipton, 1977). Variations in response to CO₂ among cultivars and developmental stages has been reported in numerous leafy vegetables and fruits (Ke et al., 1993; Paull, 1992). The benefit obtained by using CA (3% O₂ + 10% CO₂) to maintain the quality of salad pieces differed depending on lettuce type. Iceberg salad pieces showed the greatest benefit from this CA. Such differences among minimally processed lettuces in response to CA may result in differential quality changes in salad mixes. Therefore, the optimum atmosphere composition for salad mixes may need to be adjusted depending on the components of the mix.

The effects of storage prior to salad preparation was studied with iceberg and romaine lettuces. Iceberg lettuce heads can be stored for up to 14 days at 5°C without a decrease in shelf-life of the salad product as compared to product prepared from nonstored lettuce. For romaine lettuce, however, quality of the salad product decreased if the heads had been stored 7 days. This reduction in quality could be mitigated by using lower temperatures and/or CA during storage. Understanding the effects of storage on subsequent quality is important because large quantities of lettuce are shipped to
When present, RS greatly diminished the overall quality of minimally processed lettuce. Piece size may affect the development of RS. For example, when whole (15 x 2.5 cm), salad size (5 x 3 cm) and finely chopped (1.0 x 0.5 cm) midribs were stored at 5°C under 5 ppm ethylene, the smallest pieces developed little RS (unpublished data). This is important since salad pieces of different size are commercially prepared. Low O₂ and high CO₂ inhibit RS, while maximum susceptibility occurs near 5°C (Ke and Saltveit, 1989). The CA used in the present study should inhibit RS development even if product is held near 5°C. However, in highly RS sensitive cultivars (‘Legacy’), RS occurred even under CA. Therefore, RS sensitive cultivars should not be used for minimally processed lettuce.

High quality cut lettuce must be fresh looking, bright green, crisp, and have no off-odors. Since there are different components of OVQ, it would be informative to establish the relative importance of each defect. We have demonstrated that in the absence of RS, both LSB and LEB are the most serious defects leading to a decrease in OVQ of salad products. This detailed quality assessment can be instructive for evaluating different types and cultivars of lettuce prepared as salad products.

5. Summary

The quality of CA-stored salad pieces, prepared from intact iceberg heads stored up to 14 days at 5°C, was similar to that of salad pieces prepared from freshly harvested lettuce. The quality of minimally processed romaine lettuce was detrimentally affected by a 7 day storage period of the intact heads.

The use of a CA containing 3% O₂ + 10% CO₂ was more effective in maintaining the quality of iceberg pieces than it was for quality retention in the other minimally processed lettuce types. The magnitude of the benefits derived from CA needs more critical assessment in relation to lettuce types and cultivars, atmosphere–temperature combinations, and the shelf-life required commercially.

In general, both LSB and LEB are the defects which most contribute to a decreased OVQ in minimally processed lettuce. Romaine salad pieces had the highest LEB index after 12 days of storage in air at 5°C; under CA, LEB was reduced in pieces from all lettuce types. LSB, including BS, was an important defect on CA-stored pieces from romaine lettuce. On butter lettuce, CO₂ injury resulted in severe discoloration and tissue softening. If RS sensitive cultivars were used, this disorder could seriously affect the OVQ of salad pieces.

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References


