Shelf Life of Aquaponically-grown Finstar Lettuce in Different Oxygen Transmission Rate Films

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Abstract

The effects of oxygen transmission rate of packaging material on the shelf life of aquaponically-grown Finstar lettuce was studied. Parameters of packaging headspace gas composition (oxygen and carbon dioxide concentrations), lettuce pH, percentage weight loss, total aerobic microorganisms, and color were analyzed every ten days for sixty days. Finstar lettuce was stored at 4°C in four different types of packages (treatments), including a clamshell package and three film bags with oxygen transmission rates (OTR) of 3.0-6.0 cc/(m²/24 hr/1 atm), 80-90 cc/(m²/24 hr/1 atm), and >225 cc/(m²/24 hr/1 atm). The percentage weight loss of the 3.0-6.0 OTR package (-0.76-1.05%) was lowest while the percentage weight loss of the clamshell package was highest (0.81-7.72%) among packaging treatments. Nearly ½ of the panelists rated lettuce as fresh enough to eat as is after 50 days of storage in 80-90 cc/(m²/24 hr/1 atm) films while lettuce packaged in the other treatments had less that 1/3 of the panelists judging the lettuce fresh enough to eat as is. The long shelf-life may be attributed to Finstar having resilient genetic properties along with being greenhouse-grown which lessens the possibility of contamination compared to field-grown lettuces.

Keywords: aquaponics, oxygen transmission rate, packaging, Finstar lettuce, shelf-life

1. Introduction

Minimally processed fresh produce, such as lettuce, has a limited shelf-life since it continues to respire, transpire, and be enzymatically active even after harvest (Del Nobile et al., 2005). According to the United States Department of Agriculture, 30-40% of all available food in the U.S. goes uneaten due to loss before market or as waste after market (United States Department of Agriculture, 2021). Creating better food packaging systems can aid in reducing food waste by extending the quality shelf life of the food. Modified atmosphere packaging (MAP) has been shown to impact lettuce shelf life by controlling the package headspace gas concentrations thereby influencing the rate of respiration (Del Nobile et al., 2006; Escalona et al. 2006; Guo et al., 2019; Kim et al, 2005; Mart fiez-S ánchez et al., 2010; McDonald et al., 1990; Olivera et al., 2010; Peng et al., 2020; Soltani Firouz et al., 2021). Cellular respiration is the process by which glucose and oxygen react to produce carbon dioxide, water, and energy. Understanding the cellular respiration rate of a product can be beneficial in choosing a packaging system.

Another factor that influences product shelf-life is storage temperature. Atmospheres within packages have been shown to change more rapidly at higher temperatures (Oliveira et al., 2010) and high temperatures have led to a faster rate of leafy green deterioration (Peng et al., 2020). MAP and low temperatures along with high relative humidity reduce the rates of release of water through leaves (transpiration) (Soltani Firouz et al., 2021). A lack of moisture in leaves can lead to weight loss, shrinkage and browning (Aguero et al., 2011). Browning is one of the major causes of quality loss in fresh-cut lettuce (Martinez-Sanchez et. al, 2010) mostly caused by enzymatic browning which is the oxidation of phenolic compounds catalyzed by the enzyme polyphenol oxidase (PPO) that results in brown pigments (Araji, 2014). Cutting, peeling, and bruising are common causes of enzymatic browning due to cellular damage allowing enzymes to come in contact with oxygen and substrates. Enzymatic activity can lead to pink discolorations of lettuce as well.

The Finstar lettuce in this study was harvested and packaged as heads and only cut away from the roots. However, it can also be harvested as loose leaves. Head-forming lettuces typically have longer shelf lives than leaf lettuces likely due to the decreased exposure to oxygen.. Browning is considered very important in the lettuce industry since it is easily detected by consumers and this visual defect will affect sales (Degl'Innocenti et

al., 2007).

Created by BASF, Finstar is a hydroponically or aquaponically-grown lettuce developed as a cross between romaine (*Lactuca sativa* var. *longifolia Asterales*) and iceberg lettuces (*Lactuca sativa* var. *capitata*). Lettuce is one of the most common vegetables grown in hydroponic and aquaponic systems. Aquaponic systems are beneficial to both fish and plants because bacteria convert fish waste into nutrients which are used by the plants, thereby improving water quality for the fish (Love et al., 2014). In a coupled aquaponic system, the water is recirculated between the fish and the plants, while in a decoupled system, the water flows only from the fish to the plants (Pattillo et al., 2022).

The aim of this study is to investigate the effects of oxygen transmission rate on the shelf life of aquaponically-grown Finstar lettuce. Quality parameters include percentage weight loss, headspace gas composition (oxygen and carbon dioxide concentrations), color, pH, the enumeration of total aerobic microorganisms. The lack of research on Finstar lettuce provides an exciting opportunity for new findings.

2. Methods

2.1 Aquapnic System

Finstar lettuce seeds from BASF were grown in an aquaponic system in a traditional greenhouse (20-22 $^{\circ}$ C with ambient light) that is maintained and operated by a University Cooperative Extension Service. Heads of Finstar lettuce were harvested after 7-8 weeks, trimmed of any dried leaves, and transported to the laboratory immediately for packaging.

The aquaponic unit consisted of a fish tank, filtration system, and deep well floating raft system. Water was circulated throughout the system by entering the sump, being pumped through the filtration system for nitrification to take place, and then sent to the fish tank and plant growing system. In the coupled aquaponic system used for this study, the flow rate to the fish was approximately 10 gallons/minute while the flow rate to the lettuce was approximately 0.2-0.4 gallons/minute. The water temperature of the system fluctuated between 22-24°C and the fish used in the system were Nile Tilapia (*Oreochromis niloticus*) ranging from 1-2 years old. The same fish were used throughout the duration of the study.

2.2 Packaging and Storage Procedures

Single heads of unwashed lettuce was packaged (one head per package) in four different packaging treatments, which included: a plastic clamshell Z-TUG-LR7 (Plastic Container City, Petersburg, VA) or three types of film bags supplied by Cryovac Division of Sealed Air Corporation (Charlotte, NC). The three different film bags were 1) a barrier bag for bone-in beef and pork with a manufacturer oxygen transmission rate (OTR) range of 3.0-6.0 $cc/(m^2/24 hr/1 atm) @40°F/5°C$; 0% RH and a water vapor transmission rate (MVTR) of 0.4-0.5 $g/(100in^2/24hr/1atm) @100°F/38°C$; 100%RH; 2) a barrier bag for gassing cheeses with a manufacturer OTR of 80-90 cc/(m2/24 hr/1 atm) @40°F/5°C; 0% RH and MVTR of 0.3-1.0 $g/(100in^2/24hr/1atm) @100°F/38°C$; and 3) a non-barrier bag used for produce with a manufacturer OTR of >225 cc/(m2/24 hr/1 atm) @40°F/5°C; 0% RH and MVTR of >225 cc/(m2/24 hr/1 atm) @40°F/5°C; 0% RH and MVTR of >225 cc/(m2/24 hr/1 atm) @40°F/5°C; 0% RH and MVTR of >225 cc/(m2/24 hr/1 atm) @40°F/5°C; 0% RH and MVTR of >225 cc/(m2/24 hr/1 atm) @40°F/5°C; 0% RH and MVTR of >225 cc/(m2/24 hr/1 atm) @40°F/5°C; 0% RH and MVTR of >225 cc/(m2/24 hr/1 atm) @40°F/5°C; 0% RH and MVTR of >225 cc/(m2/24 hr/1 atm) @40°F/5°C; 0% RH and MVTR of 0.6-0.8 $g/(100in^2/24hr/1atm) @100°F/38°C$. A 7.6 x 12.7 cm sample of each packaging film was tested for OTR using the ASTM D3985-02 method with 0% RH, 23°C conditions using a Mocon 2/20 Oxytran tester (Ametek Mocon, Minneapolis, MN). The testing setup was convergent by cycle, with 1 hour conditioning and a 30 minute data point exam time.

Seven packages of each of the four packaging systems were prepared during each of three replications. Bags containing one head of lettuce each were sealed with a double impulse sealer (American International Electric Inc., City of Industry, CA). The packages were stored in refrigerators at 4°C and evaluated after 0, 10, 20, 30, 40, 50, and 60 days. On each testing day, one package of each system was removed from refrigerated storage for analysis.

2.3 Headspace Gas Composition Analysis

Oxygen (O₂) and carbon dioxide (CO₂) percentages of packages containing lettuce were measured with a GS6600 O₂ & CO₂ Headspace Analyzer (Systech Illinois). One empty package of each type was also filled with air as a control. On each testing day, the packages containing and not containing lettuce were tested for headspace gas composition prior to being opened for any other analyses.

2.4 Weight Analysis

The heads of lettuce were weighed on day 0 after being packaged. On each testing day, all the remaining packages not previously used for other analyses were weighed to determine percent weight loss.

2.5 Microbial Analysis

A 0.1% sterile peptone solution was prepared by combining 1g of peptone with 1L of distilled water, stirring the solution until dissolved, and autoclaving it at 121° C for 20 minutes. Twenty grams of lettuce were mixed with 180 ml of 0.1% sterile peptone and stomached for 90 seconds at 230 RPM in a stomacher filter bag (Seward, UK) in a Stomacher 400 Circulator (Seward, UK), and the rinse solution was used to make serial dilutions for enumeration of total aerobic microorganisms. One ml of the serial dilutions was sterilely pipetted onto petrifilm (3M Corporation, Saint Paul, MN). The Aerobic Count Petrifilms were incubated for 24 hours at 37°C and then petrifilms with 20-250 cells were counted and numbers converted to colony forming units (CFU) and log CFU.

2.6 pH Analysis

An Accumet Model 10 pH Meter (Thermo Fisher Scientific, Waltham, MA) was used to measure the pH of the samples. Two probes were used to take pH measurements of the microbiological rinse solution and the values were averaged.

2.7 Pictures

Digicam Control software was used to capture the pictures taken with a Nikon D3400 camera (Nikon Corp., Tokyo, Japan). Pictures were taken of the top and bottom of each head of lettuce on day 0 prior to packaging and again on each testing day.

2.8 Color Analysis

L*, a*, and b* values were generated from the pictures of the Finstar lettuce using MATLAB Software (Mathworks, Natick, MA). Total color difference, chroma, and hue were calculated from the L*, a*, and b* values as

$$\Delta E = \sqrt{(L - L0)^2 + (a - a0)^2 + (b - b0)^2}$$

where L0, a0, and b0 are the day 0 are the values taken on day 0
Chroma = $\sqrt{a^2 + b^2}$

$$Hue = 180 + \tan^{-1}(\frac{b}{a})$$

2.9 Shelf Life Sensory Analysis

A panel of nineteen students, alumni, and staff from an University's Department of Food Nutrition and Packaging Sciences rated unlabeled photos of days 30, 40, 50, and 60 Finstar lettuce on a 3-point scale with 1=eat as is, 2=trim then eat or 3=discard. Panelists were untrained and asked to evaluate based on their personal experience.

2.10 Statistical Analysis

The experiment was replicated three times on different days with different crops of Finstar lettuce. The data were subjected to analysis of variance (ANOVA) using SAS. Tukey's Studentized Range Significant Difference (HSD) test was used to determine if data was significantly different ($p \le 0.5$).

3. Results and Discussion

The manufacturer's and measured OTR values differed however this is likely due to the fact that measurements were taken at different temperatures. Mo et al. (2004) reported that OTR decreased exponentially with increases in temperature between 10 $^{\circ}$ and 40 $^{\circ}$. (Mo et al., 2004).

Table 1. Manufacturer and measured oxygen transmission rates of films used to package Finstar lettuce.

Relative oxygen	Manufacturer oxygen transmission	Measured oxygen transmission			
transmission rate	rate (cc/[m2 -day]) @ 4.4 ℃	rate (cc/[m2 -day]) @ 23 ℃			
High	>225	111.9			
Medium	80-90	33.8			
Low	3-6	11.4			

3.1 Headspace Gas Composition

There were no significant differences between both the O_2 and CO_2 percentages of the different empty packaging treatments (data not shown) however packages containing Finstar lettuce had significant differences in package O_2 concentration, package CO_2 concentration, and lettuce weight loss (Table 1).

treatment	O2	CO2	pН	weight	L*	a*	b*	hue	chroma	ΔΕ
	(%)	(%)		loss (%)				angle ()		
clamshell	22.65 ^a	0.17 ^c	6.43 ^b	3.52 ^a	50.65	-17.81	40.21	178.84	44.02	7.53
>225	18.96 ^b	1.41 ^c	6.56 ^a	2.40^{b}	52.14	-17.86	42.10	178.83	45.80	5.35
80-90	12.77 ^c	6.46 ^b	6.67^{a}	1.82 ^b	51.46	-18.91	42.01	178.85	46.10	4.82
3.0-6.0	10.75 ^c	10.15^{a}	6.66 ^a	0.09 ^c	51.44	-18.58	41.78	178.85	45.77	4.41

Table 2. Overall averaged effect of package type (OTR) on package gas composition, pH, weight loss, L*, a*, b*, hue, chroma and ΔE of Finstar lettuce stored under refrigeration for 60 days sampled every 10 days

Means with different superscripts are significantly different (P≤0.05). n=21.

For the packages containing Finstar lettuce over the sixty days of refrigerated storage, O_2 percentages ranged from 0.034%-24.1% while carbon dioxide percentages ranged from 0%-20.5%. The mean O_2 percentages of the clamshell and >225 OTR packages were higher than the 3.0-6.0 OTR and 80-90 OTR packages (Table 2, Figure 1). The mean oxygen percentages of the clamshell packages were significantly higher than all the film packages throughout storage (Figure 1). The mean O_2 percentages of the >225 OTR packages (Figure 1).

The mean carbon dioxide percentages of the clamshell and >225 OTR packages were lower than the 3.0-6.0 OTR and 80-90 OTR packages with the clamshell barely having any CO₂ throughout the study (Figure 2). The mean CO₂ percentages of the 3.0-6.0 OTR packages were significantly higher than the other packaging treatments (Figure 2). The mean CO₂ percentages of the 80-90 OTR packages were significantly higher than the clamshell and >225 OTR packages (Figure 2).

The atmospheres of the 3.0-6.0 and 80-90 OTR packages changed more over the 60-day duration than the clamshell and >225 OTR packages. When the means of all the packaging treatments from each testing day were averaged together, the mean day 0 O_2 percentage was significantly higher than days 20-60. The mean day 50 and 60 O_2 percentages were significantly lower than days 0-40. The mean day 0 CO_2 percentage was significantly lower than days 10-60.

Packages that had higher oxygen percentages had lower carbon dioxide percentages (Figure 1, Figure 2). In a study investigating the respiration rate of iceberg lettuce, a decrease in the percent oxygen concentration and an increase in the percent carbon dioxide concentration were shown in all three films (two were polyolefinic and one was a biodegradable) over ten days (Del Nobile et al., 2005). Atmospheres of packaged shredded romaine lettuce showed to be modified more rapidly at 25°C than 5°C (Oliveira eta al., 2010). Figures 1 and 2 show the effect of oxygen transmission rate on the mean oxygen and carbon dioxide percentages over the duration of the study.



Figure 1. Effect of package oxygen transmission rate on oxygen headspace composition of Finstar lettuce packages over storage time (days). The OTR are in units of cc/(m²/24 hr/1 atm) @40°F/5°C; 0% RH. n=3. a-d data points with different superscripts are significantly different (p≤0.05)



Figure 2. Effect of package oxygen transmission rate on carbon dioxide headspace composition of Finstar lettuce packages over storage time (days). The OTR are in units of cc/(m²/24 hr/1 atm) @40°F/5°C; 0% RH. n=3. a-c data points with different superscripts are significantly different (p≤0.05)

Oxygen (O₂) comprises approximately 21% oxygen of Earth's atmosphere while carbon dioxide (CO₂) comprises 0.04%. Modifying a package's atmosphere has been shown to affect the shelf life of lettuce (Del Nobile et al., 2006; Escalona et al. 2006; Guo et al., 2019; Kim et al, 2005; Mart nez-S nchez et al., 2010; McDonald et al., 1990; Olivera et al., 2010; Soltani Firouz et al., 2021). Respiration rates can be reduced in low O_2 and elevated CO_2 atmospheres (Del Nobile et al., 2006). In a study on the changes in respiration of fresh-cut butterhead lettuce, the lettuce packaged in low O2 and moderate to high CO2 atmospheres had higher respiration rates compared to lettuce packaged in high O_2 and moderate CO_2 atmospheres (Escalona et al., 2006). The increased respiration rates in high CO_2 atmospheres could be attributed to the high CO_2 levels causing physiological stress compared to air atmosphere (Escalona et al., 2006). Excessive CO_2 levels along with O_2 depletion can be injurious (Kim et al., 2005). A study analyzing the effects on quality maintenance and shelf life of fresh-cut romaine lettuce due to delayed modified atmosphere packaging found that strong off odors, accumulated ethanol and acetaldehyde, and CO_2 injury due to package atmospheres being low in O_2 and high in CO₂ could be reduced by delayed packaging (Kim et al., 2005). Discoloration and off-flavors were detected when the atmospheres of 'Salinas' crisp head lettuce packages had CO₂ percentages above 20% (McDonald et al., 1990). Air leakage due to poor seals of 'Salinas' crisp head lettuce packages led to browning and discoloration likely due to elevated O_2 levels (McDonald et al., 1990). In a study on the effects of low oxygen levels and light exposure on fresh-cut romaine lettuce, atmospheres with low O₂ partial pressures (pO₂) had a positive effect on shelf life due to controlling browning and preventing off-odors and off-flavors (Mart nez-S ánchez et al., 2010). Overall, according to the literature, too much O_2 or too much CO_2 can both have negative effects on the shelf life of lettuce.

3.2 pH

Throughout the sixty days of refrigerated storage, pH values ranged from 6.00-7.37 and generally increased from day 0 to day 10, decreased till day 30, increased till day 50, and then decreased again at day 60. The pH values of the clamshell packages were significantly lower than the 3.0-6.0 OTR and 80-90 OTR packages but not the >225 OTR packages (Figure 3).



Figure 3. Effect of oxygen transmission rate on the pH of Finstar lettuce over storage time (days). The OTR are in units of cc/(m²/24 hr/1 atm) @40°F/5°C; 0% RH. n=3. a-c data points with different superscripts are significantly different (p≤0.05)

In a study done on the effects of packaging type and storage temperature on the growth of foodborne pathogens on shredded Romaine lettuce, there were no significant effects of modified atmosphere on pH; the pH values ranged from 6.0-6.6 at 25°C and 6.3-6.5 at 5°C (Oliveira et al., 2010). Lower growth temperatures along with lower pH values lead to decreased growth rates of S. Entertitidis on hydroponic lettuce (Xylia et al., 2022). Increasing pH values were shown to increase the radical scavenging effects of lettuce extract (Altunkaya et al., 2016).

The packages with the higher carbon dioxide percentages and lower oxygen percentages had higher pH values (Figure 1, Figure 2, Figure 3). Reduction in respiration rate of lettuce by moderate CO_2 levels can be attributed to intracellular pH change with pH variations influencing enzymatic activity or respiratory pathways (Escalona et al., 2006).

3.3 Percent Weight Loss

The percent weight loss of the clamshell packaged lettuce was significantly higher than other treatments (Figure 4) while the percent weight loss of the 3.0-6.0 OTR packaged lettuce was significantly lower than the other treatments (Figure 4). When the means of all the packaging treatments from each testing day were averaged together, the mean day 30, 50, and 60 percent weight loss was significantly higher than days 0, 10, 20, and 40. Mean day 0 and 10 percent weight loss was significantly lower than days 30-60.



Figure 4. Effect of package oxygen transmission rate on percentage weight loss of Finstar lettuce packages over storage time (days). The OTR are in units of cc/(m²/24 hr/1 atm) @40°F/5°C; 0% RH. n=3. a-c data points with different superscripts are significantly different (p≤0.05)

The lowest percent weight loss of lettuce packaged in the 2.0-6.0 OTR film was likely due to a slower lettuce respiration rate by the high CO₂ headspace compositions and low O₂ headspace compositions (Figure 1, Figure 2). The significantly higher percentage weight loss of lettuce packaged in the clamshell package was likely due to moisture escaping through the small opening between the top and bottom of the clamshell creating a high respiration rate due to high O₂ and low CO₂ headspace gas compositions. Elevated levels of respiration can lead to increased weight loss due to transpiration. In a study of the effects of modified atmosphere packaging on the quality of whole lettuce (*Lactuca sativa L. Grand Rapid*), the percentage weight loss by the sixth day of storage was 7.3% for the control and 4.2% for the MAP-treated package (Guo et al., 2019). It is worth noting the heads of Finstar lettuce were weighed while still in their packages which included some water respired or transpired from the lettuce. Percentage weight loss values would likely be higher if lettuce is weighed outside of the package.

3.4 Microbiological Results

The total aerobic CFU/g of lettuce for all OTR packaging treatments ranged from ~ 2 logs/ml to ~3.5 logs/ml (Figure 5). The OTR of the packaging did not have a significant effect on total aerobic bacteria over the 60-days of refrigerated storage.



Figure 5. Effect of package oxygen transmission rate on log CFU/g of Finstar lettuce packages over storage time (days). The OTR are in units of $cc/(m^2/24 \text{ hr/1 atm}) @40^\circ\text{F/5}^\circ\text{C}; 0\%$ RH. n=6

3.5 Color

There were no significant differences between the L*, a*, and b* values of the Finstar lettuce from the different packaging treatments. No significant color variation between films was also reported in a study on romaine and iceberg lettuces (Del Nobile et al., 2005). Dielectric barrier discharge atmospheric cold plasma (DACP) treatment on the inactivation of Escherichia coli O157:H7 and aerobic microorganisms in romaine lettuce did not affect L, a, b values or total color difference (Min et al., 2016). The L*, a*, and C* (color saturation) values of romaine lettuce decreased with elevated NaCl concentrations in irrigation water indicating a possible effect of osmotic pressure on leaf integrity (Kim et al., 2008).

There were no significant changes in the L* values of Finstar lettuce over time in refrigerated storage and a* values generally increased over time with days 30, 50, and 60 being significantly higher than day 0. Conversely the b* values generally decreased over time with day 0 b* values significantly higher than day 50 b* values but not day 60.

In defining color functions, hue angle represents the angle within the color globe and moves through the color spectrum from red-yellow, green, and blue while chroma is the intensity of color (Clydesdale and Francis, 1975). Both hue and chroma have been found to decrease as percentage loss of chlorophyll increases (Clydesdale and Francis, 1975). ΔE is the measure of total color difference between day 0 and testing day calculated as $\sqrt{(L1-L2)^2 + (a1-a2)^2 + (b1-b2)^2}$ where L0, a0, and b0 are values taken on day 0.

The hue values from the lettuce packaged in the 80-90 OTR bags were significantly higher than the lettuce packaged in the >225 OTR bags. Days 0, 10, 20, and 40 hue values were significantly higher than days 30, 50, and 60. The hue values for all four packaging treatments ranged from $178.65^{\circ}-178.91^{\circ}$ which puts them in

quadrant II of the Hunter L, a, b System Diagram near the green hue angle of 180°. All a* values were negative indicating a green color and all b* values were positive indicating a yellowness. There were no significant differences between the chroma, or total color difference (ΔE) of the lettuce packaged in the different treatments. Chroma decreased over time in refrigerated storage but not significantly until day 50. ΔE increased over time; days 0 and 10 were significantly lowest.

The color of the lettuce remained bright green throughout the duration of the study except for some browning of the outer leaves. No pinking was detected, this may be because of Finstar's vibrant green color. To the human eye, the inside leaves of the heads of Finstar are a slightly lighter green color than the outer leaves and the bottom part of the leaves is a lighter shade of green compared to the top of the leaves.

3.6 Sensory Shelf-Life

3.6.1 Shelf-Life Analysis

Figure 6 shows the percentage of 19 panelists rating the lettuce by examining the high resolution pictures of the top and bottom sides of each lettuce head. It is likely hue values influenced the ratings since lettuce packaged in 80-90 OTR bags had significantly higher hue values than lettuce packaged in >225 OTR bags. Day 50 ratings were significantly higher than day 30 and day 40 ratings which shows the shelf life of aquaponically-grown heads of Finstar lettuce in this study was mostly between 40 and 50 days. About a third of the panelists still rated the lettuce to "eat as is" after 50 days of storage and nearly $\frac{1}{2}$ of the panelists stated they would eat the lettuce for samples stored in the 80-90 OTR bags after 50 days.





4. Conclusions

Oxygen headspace percentages generally were clamshell >225 OTR >3.0-6.0 OTR and 80-90 OTR packages. The carbon dioxide package headspace levels followed the opposite pattern of 3.0-6.0 OTR packages >80-90 OTR >>225 packages and clamshell packages. Both the 3.0-6.0 OTR and 80-90 OTR packages had decreasing oxygen and increasing carbon dioxide percentages while the clamshell and >255 OTR packages had very small percentages of carbon dioxide throughout the study. The pH values of the clamshell packaged lettuce were significantly lower than 3.0-6.0 OTR and 80-90 OTR but not the >225 OTR packaged lettuce. Packages with lower carbon dioxide headspace percentages and higher oxygen headspace percentages had lower pH values. The percent weight loss of the clamshell packaged lettuce was significantly lower than the other treatments while the percent weight loss of the 3.0-6.0 OTR packaged lettuce was significantly lower than the other treatments. On a 3-point scale with 1=eat as is, 2=trim then eat or 3=discard, the shelf-life ratings of the 80-90 OTR packaged lettuce packaged in clamshells had the lowest. Packaging treatments with higher percentages of CO₂ and lower percentages of O₂ had lower ratings, lower percentage weight loss, and higher hue angles.

Even with the headspace compositions of the packages varying so widely, there were no significant differences of colony forming units (CFU), log CFU, L*, a*, or b* color parameters. Finstar lettuce maintained a relatively long shelf life in all four packages regardless of oxygen transmission rate.

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