



## Postharvest biology, quality and shelf life of buckwheat microgreens

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### ABSTRACT

Buckwheat microgreens have short shelf life which limits their commercial use. The effects of storage temperature, modified atmosphere packaging (MAP), and wash conditions on quality and shelf life of buckwheat microgreens were assessed. Temperature significantly ( $P < 0.0001$ ) affected package atmospheres and product quality. At the end of storage, microgreens stored at 1, 5 and 10 °C had smaller microbial populations and less tissue electrolyte leakage than those stored at 15, and 20 °C. Package film oxygen transmission rate (OTR) significantly ( $P < 0.05$ ) affected package atmospheres. However, differences in quality and shelf life of microgreens packaged in different OTR films were slight and not evident until day 21 of storage. On day 21, buckwheat microgreens packaged in 16.6 pmol/(m<sup>2</sup> s Pa) oxygen transmission rate package films were observed to have the freshest appearance with lowest tissue electrolyte leakage. Chlorine (100 mg/L) wash significantly ( $P < 0.05$ ) reduced microbial populations, initially; however, after 7 days of storage, all washed microgreens experienced accelerated microbial populations. Our findings suggest that buckwheat microgreens should be stored at 5 °C with moderately high O<sub>2</sub> (14.0–16.5 kPa) and moderately low CO<sub>2</sub> (1.0–1.5 kPa) content to maintain optimal quality and maximal shelf life.

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### 1. Introduction

Microgreens are young and tender cotyledonary leafy greens that are found in a pleasing palette of colors, textures and flavors. They may be used to enhance salads, or as edible garnishes to embellish a wide variety of other dishes. Microgreens, in general, contain higher concentrations of bioactive compounds such as vitamins, minerals, and antioxidants than mature greens (Janovská, Štočková, & Stehno, 2010; Treadwell, Hochmuth, Landrum, & Laughlin, 2010, pp. 1–3; Xiao, Lester, Luo, & Wang, 2012). Buckwheat microgreens specifically are high in antioxidants, flavonoids, carotenoids, and  $\alpha$ -tocopherol contents (Janovská et al., 2010). However, like all microgreens, buckwheat microgreens have only a few days of shelf life (Chandra, Kim, & Kim, 2012).

The two most important storage parameters for postharvest shelf life are storage temperature and atmospheric composition (Hodges & Toivonen, 2008). Numerous studies on postharvest shelf

life of produce emphasize the importance of temperature control (Brecht, 1995; Watada, Ko, & Minott, 1996), which is generally regarded as the most critical factor in prolonging shelf life of fresh-cut produce (Deza-Durand & Petersen, 2011). However, no information could be found on the optimal commercial storage temperature for buckwheat microgreens.

Modified atmosphere packaging (MAP) effectively prolongs the shelf life of fruits and vegetables by decreasing oxygen (O<sub>2</sub>) and increasing carbon dioxide (CO<sub>2</sub>) partial pressures in the package headspace due to the interaction between respiratory O<sub>2</sub> uptake and CO<sub>2</sub> evolution of the packaged plant tissues, and the selective transfer of gases through packaging films (Kim, Luo, & Gross, 2004). Packaging also reduces contamination of the product by bacteria, mold spores, and other environmental pollutants during storage.

No packaged ready-to-eat microgreens are currently found in supermarkets. Microgreens' high price and perishability limit them to a specialty market of upscale restaurants and catering establishments. Thus, there is room for expansion of microgreen availability in the market place if temperature and atmospheric conditions favorable to extension of shelf life are determined. Using buckwheat microgreens as a model, we determined the optimal storage conditions and developed protocols for evaluating the

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effect of package film OTR and temperature on the quality of microgreens.

## 2. Materials and methods

### 2.1. Sample preparation

Buckwheat (*Fagopyrum esculentum* Moench CV. Manner) seeds were purchased from Living Whole Foods Inc. (West Springville, UT, USA), with shell intact. Seeds were soaked in acidified water (pH 5.5–6.0) for 12 h to promote germination according to the supplier's recommendation. Growing medium, Farfard 3B potting soil consisting of 45 g/100 g peat moss, 15 g/100 g vermiculite, 15 g/100 g perlite and 25 g/100 g bark (Griffin Greenhouse & Nursery Supplies, Bridgeton, NJ, USA) was spread evenly in 54 cm × 28 cm × 6 cm trays (vacuum-formed standard 1020 open flats without holes, Growers Supply, Dyersville, IA, USA) containing 2 L water in the bottom. Additional water was sprayed on the surface. The buckwheat seeds were then removed from their acidified water bath and spread evenly over the damp medium, tamping very gently to insure contact with the medium. Seeded trays were kept at 25 °C in the dark for 3–4 days before exposing to light with light irradiance of 9.16 W/m<sup>2</sup> determined by LI-1000 datalogger (LI-COR, Lincoln, NE, USA) for a 12 h photoperiod for an additional 4 days. The trays were sprayed with about 200 mL of water per tray once a day to keep the medium damp. Microgreens were harvested when they reached about 5 cm in height, and the first true leaf was beginning to emerge. The microgreens were cut above the medium line with a pair of sterilized scissors. Damaged microgreens and those with defects were discarded.

### 2.2. Temperature and package atmosphere treatments

Microgreens used for temperature studies were packaged (10 g/bag) unwashed in sealed 15 cm × 15 cm bags prepared with polyethylene films of 16.6 pmol/(m<sup>2</sup> s Pa) oxygen transmission rate (OTR). The microgreen samples were stored at 1, 5, 10, 15, or 20 °C for 14 days with evaluations performed on days 0, 3, 6, 10 and 14. Microgreens prepared for package atmosphere treatment were sealed (10 g/bag) unwashed in polyethylene bags (15 cm × 15 cm) with film OTRs of 8.0, 16.6, 21.4 and 29.5 pmol/(m<sup>2</sup> s Pa) and stored at 5 °C for 21 days with evaluations performed on days 0, 4, 7, 14 and 21.

### 2.3. Wash conditions

Wash solutions containing 100 mg/L and 50 mg/L free chlorine were prepared with concentrated (6 g/100 g) sodium hypochlorite with pH adjusted to 6.5 using citric acid. A control water wash was prepared without added sodium hypochlorite. Microgreens were placed in mesh bags, then washed in one of the 3 wash solutions with gentle agitation, for 30 s. The microgreens (while in the mesh bags) were centrifuged at 20.5 × g for 200 s with a commercial salad centrifugal dryer (model T-304; Garrouite Spin Dryer, Meyer Machine CO, USA) to remove excess water. The washed microgreens, along with unwashed controls (no wash) were packaged in sealed 15 cm × 15 cm bags of 16.6 pmol/(m<sup>2</sup> s Pa) film OTR. Washed and unwashed packaged microgreens were stored at 5 °C for 21 days and evaluations were performed on days 0, 4, 7, 14 and 21.

### 2.4. Analysis of packaging headspace atmospheric composition

The CO<sub>2</sub> and O<sub>2</sub> in the headspace of packaged buckwheat microgreens were measured using a gas analyzer (Check mate II, PBI Dansensor Co., Denmark) by inserting the needle of the

measuring assembly through a septum adhered to the packaging film.

### 2.5. Total aerobic mesophilic bacteria

Microgreens (3 g) per replicate were macerated in 27 mL phosphate buffered saline (PBS), using a model 80 Lab Stomacher (Seward Medical, London, UK) for 2 min at high speed in filtered stomacher bags. A 50 µL sample of each filtrate or its appropriate dilution was logarithmically spread on agar plates with an automatic spiral plater (Wasp II, Don Whitley Scientific Ltd., West Yorkshire, UK). The total aerobic mesophilic bacteria were plated on tryptic soy agar (TSA, Difco Lab, Sparks, MD, USA) and incubated at 30 °C for 24–48 h. Microbial colonies were counted using an automated colony counter (ProtoCOL SR, Synoptics, Cambridge, UK) and reported as log CFU/g of tissue.

### 2.6. Tissue electrolyte leakage

Tissue electrolyte leakage was measured following a modified procedure from Kim, Luo, Tao, Saftner, and Gross (2005). Buckwheat microgreens (3 g) were submerged in 150 mL of distilled water for 30 min. The electrical conductivity of the solution was measured using a conductivity meter (model 135A; Orion Research Inc., Beverly City, MA, USA). Total electrolytes of the microgreen samples were determined after freezing at –20 °C for 24 h and thawing at room temperature. Tissue electrolyte leakage was expressed as a percentage of total electrolytes.

### 2.7. Experimental design and statistical analysis

Package atmospheres, tissue electrolyte leakage, and microbial data were analyzed as two-factor linear models using the PROC MIXED procedure (SAS Institute Inc 1999; Cary, NC). The two factors were storage time and treatment type, which depended on the experiment: storage temperature (1, 5, 10, 15 and 20 °C), package film OTR (8.0, 16.6, 21.4, 29.5 pmol/(m<sup>2</sup> s Pa)), or wash treatment (no wash, tap water wash, 50 mg/L chlorine, 100 mg/L chlorine) and each had four or five levels. Microbial data were log transformed prior to analysis. Different samples were analyzed on each evaluation day for all studies. Four replications (four bags) per treatment per evaluation period were examined. Assumptions of normality and variance homogeneity of the linear model were checked and the variance grouping technique was used to correct for variance heterogeneity. When effects were statistically significant, means were compared using Sidak adjusted *P*-values to maintain experiment-wise error ≤0.05.

## 3. Results and discussion

### 3.1. Effects of temperature on the quality of buckwheat microgreens

Storage temperature and time significantly (*P* < 0.0001) affected the O<sub>2</sub> and CO<sub>2</sub> concentrations within the packages (Fig. 1A and B). The O<sub>2</sub> concentration in microgreen packages stored at 1, 5 and 10 °C declined gradually and reached equilibrium on day 10 (around 16 kPa), but in microgreen packages stored at 15 and 20 °C, O<sub>2</sub> concentrations decreased more rapidly from 21 kPa on day 0–8.2 and 6.1 kPa, respectively by day 14. The changes in CO<sub>2</sub> partial pressure followed a reverse trend compared to O<sub>2</sub> (Fig. 1B). The CO<sub>2</sub> concentrations at the different storage temperatures showed similar trends, except at 20 °C which increased sharply at the end of storage.

The initial total aerobic mesophilic bacteria plate count for buckwheat microgreens was 7.2 log which is relatively high, similar

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