Food Control 47 (2015) 114–119

Contents lists available at ScienceDirect

Food Control

journal homepage: www.elsevier.com/locate/foodcont

Efficacy of chlorine dioxide gas and freezing rate on the microbiological quality of frozen blueberries

Lei Zhang [a], Zhinong Yan [a], Eric J. Hanson [b], Elliot T. Ryser [a, *]

[a] Department of Food Science and Human Nutrition, Michigan State University, East Lansing, MI 48824, USA
[b] Department of Crop and Soil Sciences, Michigan State University, East Lansing, MI 48824, USA

ARTICLE INFO

Article history:
Received 18 February 2014
Received in revised form
5 June 2014
Accepted 9 June 2014
Available online 24 June 2014

Keywords:
Blueberry
Freezing
Chlorine dioxide
Microbial quality

ABSTRACT

Blueberries are prone to microbial contamination, with growth of bacteria, yeasts and molds during bulk freezing negatively impacting quality and marketability. As a follow-up to our previous work, the combined impact of ClO_2 gassing and freezing rate on the microbiological quality of frozen blueberries was examined. Sixteen lugs of blueberries (~9.1 kg/lug) were stacked inside a large plastic container at a commercial blueberry processing facility. In each of four trials, one container was exposed to ClO_2 gas (4 ppm) using three 3-kg sachets while one ungassed container remained untarped. Before and after commercial processing, 50-g samples of gassed and ungassed blueberries were quantitatively examined for mesophilic aerobic bacteria (MAB), yeasts, and molds. After processing, additional 50-g samples were placed in a −20 °C freezer under different conditions where the berries reached a temperature of −3 °C after 3 h (quick-frozen), 2 days (intermediate-frozen) and 5 days (slow-frozen). Fruit was sampled periodically during 6 months of frozen storage at −20 °C. MAB yeast and mold populations decreased ~2 and 1 log CFU/g, respectively, in ClO_2-gassed and ungassed fruit, with MAB, yeast and mold populations increasing ~1 log CFU/g during quick freezing to −3 °C and ~2 log CFU/g during intermediate and slow freezing to −3 °C. Based on these findings, ClO_2 gassing followed by quick freezing provides an effective means for meeting the current microbiological standards being imposed by buyers of frozen blueberries.

1. Introduction

The United States is the world's leading producer of blueberries with 55% of total production. In 2012, a total of 473 million pounds of cultivated high-bush blueberries was grown in the United States, approximately 18% coming from Michigan (United States Department of Agriculture, 2012). The blueberry industry is composed of two market categories: fresh and processed. About 70% of the U.S. blueberry crop is processed fruit with the remainder sold to the fresh market. Most of these processed blueberries are frozen for use as an ingredient in other foods such as baked goods, yogurt, and ice cream.

Blueberries are perishable and prone to microbial contamination with both spoilage and pathogenic microorganisms during growing, harvesting and further processing (Tournas & Katsoudas, 2005). Microbial growth on blueberries at inappropriate storage temperatures is the primary cause for decreased quality and shelf life after harvest. In addition, blueberries have also been identified as a vehicle for foodborne illness in several outbreaks (Calder et al., 2003; Center for Science in the Public Interest, 2008; Miller, Rigdon, Robinson, Hedberg, & Smith, 2010; Ryser, 1999). In order to address increasing concerns surrounding microbiological spoilage, many buyers of frozen blueberries have now established arbitrary limits for mesophilic aerobic bacteria, *Escherichia coli*, coliforms, yeasts and molds. However, such microbial limits are often difficult to meet since the types and levels of microbial contaminants vary widely, depending on the field, season (e.g., temperature, rainfall), plant health, and harvest management practices with microbial populations peaking at the end of the season.

Blueberry processing typically begins 12–24 h after harvest with dumping of the berries onto a conveyor belt followed by removal of leaves, sticks and other debris. After de-stemming and passage through a sanitizer-containing wash/flume tank to decrease the microbial load and remove any unripe green fruit, the berries are mechanically inspected, sorted and either individually quick frozen or boxed for the freezer. Current commercial use of chlorine-based sanitizers containing 50–100 ppm free chlorine, coupled with a typical contact time of 30–60 s, generally reduces the microbial load on blueberries ≤1 log CFU/g (Crowe, Bushway, Bushway, Davis-Dentici, & Hazen, 2007) with substantial growth

* Corresponding author. Tel.: +1 517 355 8474x185; fax: +1 517 353 8963.
E-mail address: ryser@msu.edu (E.T. Ryser).

http://dx.doi.org/10.1016/j.foodcont.2014.06.008
0956-7135/

of survivors possible during slow bulk freezing over several days. Subsequent use of such frozen blueberries in baked goods may adversely affect end product quality due to enzymatic breakdown of starch and other stiffening agents (Crowe et al., 2007).

Freezing is one means of prolonging the shelf-life of blueberries by preventing microbial growth, with an additional lethal effect on microorganisms related to temperature shock, concentration of extracellular solutes, toxicity of intracellular solutes, dehydration and the formation of large ice crystals during freezing (Hazen, Bushway, & Davis-Dentici, 2001; Zaritzky, 2005). Additional factors affecting the microbiological quality of frozen blueberries relate to the degree of ripeness and temperature fluctuations during storage that can lead to partial thawing and refreezing.

One particularly promising microbial reduction strategy is the use of chlorine dioxide (ClO_2) gas, which has been shown to effectively reduce foodborne pathogens on a wide range of products including blueberries, raspberries and strawberries (Sy, McWatters, & Beuchat, 2005) as well as on inoculated apples (Du, Han, & Linton, 2002, 2003) under laboratory conditions. When our research team previously assessed the use of ClO_2 gas-generating sachets as a novel microbial reduction strategy for blueberries, bacterial populations decreased $\geq$100-fold on gassed fruit compared to the ungassed controls (Popa, Hanson, Todd, Schilder, & Ryser, 2007). Nevertheless, microbial growth remained problematic in 272-kg pallets of blueberries since up to 7 days were required to completely bulk-freeze these berries in a large commercial freezing room.

The overall goal of this study was to develop a microbial reduction and control strategy for frozen blueberries that would help satisfy the microbial specifications being imposed by buyers. In this study, we examined the impact of ClO_2 gas and three different freezing rates on the microbiological quality and enzyme levels of frozen blueberries.

2. Materials and methods

2.1. Source of blueberries

Fresh, mechanically harvested highbush blueberries were obtained from the same grower at different field locations through the Michigan Blueberry Growers Association (MBG, Grand Junction, MI) during the second half of the harvest season (mid-August to mid-September). Sixteen lugs of blueberries (~9.1 kg/lug) were placed in a 1.2 × 1.2-m plastic container provided by Eka Chemical (Marietta, GA) at a MBG processing facility with four lugs per level and four levels per pallet (~146 kg of fruit per pallet).

2.2. Chlorine dioxide gas exposure

One pallet (16 lugs of blueberries) was tarped with a plastic sheet, sealed, and exposed to ClO_2 gas (4 mg/L) for 12 h at ~12–14 °C. ClO_2 was generated using three 3-kg sachets placed at the bottom of the container and mechanically pump-circulated throughout the container. A second pallet (16 lugs of blueberries) sat near the blueberries being treated and served as the ungassed control. After treatment, six gassed and six ungassed blueberry samples (>50 g each) were randomly taken from each lug, placed into Whirl-Pak bags, and transported to the laboratory in an insulated chest containing cold packs.

2.3. Blueberry processing

Ungassed followed by gassed berries were commercially processed at the MBG facility as follows: (1) dumped onto the conveyor, (2) conveyed to the spreader/shaker for pneumatic removal of twigs and leaves, (3) de-stemmed, (4) sanitized in a water flotation tank containing 100 ppm free chlorine with simultaneous removal of unripe green fruit, (5) mechanically sorted based on color and size, and (6) boxed in 4.5-kg corrugated cardboard boxes for freezing.

2.4. Blueberry freezing rates

The targeted rates for quick (−3 °C after 3 h), intermediate (−3 °C after 2 d) and slow freezing (−3 °C after 5 d) were based on temperature data previously collected from MBG at the start of the season using TEMP 101 continuous temperature recorders (Ertco Precision, Dubuque, IA) that were inserted into the geometric center, midpoint and outer edge of 4.5-kg boxes of berries on randomly selected pallets in their −20 °C commercial bulk freezer. In each of the four freezing trials, two randomly selected 4.5-kg boxes of processed blueberries (one gassed, one ungassed) were immediately transported to Michigan State University, dispensed into Whirl-Pak™ bags (50 g each), and frozen to −20 °C at three different rates, all of which were continuously monitored using TEMP 101 recorders. Quick freezing was accomplished by placing the blueberries (14 bags per trial) on open trays in a −20 °C forced air walk-in freezer. For intermediate freezing, 18 bags per trial were placed in an insulated ice chest that was then held in a walk-in cooler at 12 °C. After incrementally decreasing the cooler temperature to −2 °C during a 16-h period, the temperature within the ice chest fell below 0 °C after 24 h, at which time the ice chest was transferred to the −20 °C walk-in freezer. Slow freezing was accomplished similarly to intermediate freezing except that the temperature of the walk-in cooler was reduced incrementally to −2 °C during 96 h after which the blueberries were transferred to the −20 °C walk-in freezer.

2.5. Microbiological analysis

Gassed and ungassed blueberry samples taken before processing, after processing when freezing began (time 0), and at various times during freezing and continuous frozen storage were examined for populations of mesophilic aerobic bacteria (MAB), yeasts and molds. Unprocessed berries were analyzed immediately upon arrival. Samples frozen to −3 °C at different rates were microbiologically analyzed at 0, 2 and 3 h (quick freezing), 0, 6, 12, 24, and 48 h (intermediate freezing), and 0, 1, 2, 3, 4, and 5 days (slow freezing), with all blueberries analyzed after 1, 3 and 6 months of frozen storage. Blueberry samples (50 g) were placed in sterile 24-oz Whirl-Pak bags containing 50 ml of sterile phosphate buffer solution (PBS) and processed for 2 min in a Pulsifier (Filataflex Ltd., Ontario, Canada). Thereafter, samples were serially diluted in PBS and plated in duplicate on tryptic soy agar (Difco-Becton Dickinson, Sparks, MD) containing 0.6% yeast extract (Difco-Becton Dickinson) and 100 ppm cycloheximide (Sigma, St. Louis, MO) (TSAYE-C) for enumeration of mesophilic aerobic bacteria (MAB), and on potato dextrose agar (Difco-Becton Dickinson) containing 20 ppm streptomycin (Sigma) and 50 ppm ampicillin (Sigma) (PDA-SA) for enumeration of yeasts and molds. Colonies on TSAYE-C plates were counted after 48 h of incubation at 37 °C, whereas colonies on PDA-SA plates were counted after 72–96 h of ambient incubation (~22 °C).

2.6. Data analysis

Analysis of Variance (ANOVA) was conducted on all microbial count data. The Statistical Analysis System (SAS, Version 8, SAS© Institute Inc., Cary, NC) and Statgraphics Plus® (Statpoint Technologies, Warrenton, VA) programs were used to analyze the microbial