JOURNAL OF FUNCTIONAL FOODS XXX (2013) XXX–XXX

Available at www.sciencedirect.com

SciVerse ScienceDirect

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

Pasteurized and sterilized maple sap as functional beverages: Chemical composition and antioxidant activities

*Tao Yuan, Liya Li, Yan Zhang, Navindra P. Seeram**

Bioactive Botanical Research Laboratory, Department of Biomedical and Pharmaceutical Sciences, College of Pharmacy, University of Rhode Island, Kingston, RI 02881, USA

ARTICLE INFO

Article history:
Received 15 March 2013
Received in revised form
24 June 2013
Accepted 25 June 2013
Available online xxxx

Keywords:
Functional beverage
Maple sap
Pasteurized
Sterilized
Phenolics
Antioxidant

ABSTRACT

Maple sap has been consumed for centuries as a tonic by the indigenous peoples of eastern North America but is primarily utilized in this region to produce maple syrup. The natural watery form of maple sap makes its application as a functional beverage appealing but due to microbial growth, sterilization or pasteurization would be necessary before sap could be consumed. This study was designed to investigate the chemical composition (sugars, amino acids, organic acids, minerals, and phenolics) and antioxidant effects of maple sap after undergoing pasteurization and sterilization. After both processes, sugars, amino acids, organic acids, and minerals were preserved in the sap samples and they had similar phenolic contents (0.25–0.27 mg/100 g gallic acid equivalents) and antioxidant activities (IC_{50} ca. 550 μg/mL by DPPH assay). HPLC-DAD analyses revealed over 25 constituents in the sap samples of which 15 were identified using phenolic standards. In addition, one compound, 3′,5′-dimethoxy-4′-hydroxy-(2-hydroxy)acetophenone, not previously reported from maple syrup, was isolated and identified (by NMR) for the first time from maple sap. Therefore, the preservation of chemical constituents and antioxidant activity in maple sap after pasteurization and sterilization warrants its application as a functional beverage beyond its primary use for maple syrup production alone.

1. Introduction

Functional beverages are widely consumed worldwide and are a fast growing segment of the functional foods category. This popularity is largely driven by increasing consumer demand for foods that contain ingredients which may impart health benefits beyond basic nutrition (Hardy, 2000). The functional beverage arena boasts a wide variety of products including performance and energy drinks and vitamin and other fortified waters. However, consumers in developed countries such as the United States, Canada, and Europe are showing increased interest in natural and minimally processed plant-derived beverages (Gruenwald, 2009). A prime example of such beverages includes the liquid endosperm of the coconut (*Cocos nucifera*) fruit, popularly known as coconut water (Yong, Ge, Ng, & Tan, 2009). Coconut water, traditionally consumed in tropical regions for centuries, has recently boomed in sales in the developed world where it can be obtained in bottled and canned forms. Thus, there is great interest by the functional beverage industry to launch new beverage products that would be similarly attractive to consumers.

The sugar maple (*Acer saccharum*) and red maple (*Acer rubrum*) species are native to eastern North America and are

* *Corresponding author*. Tel.: +1 401 874 9367; fax: +1 401 874 5787.
E-mail address: nseeram@uri.edu (N.P. Seeram).
1756-4646/$ - see front matter © 2013 Published by Elsevier Ltd.
http://dx.doi.org/10.1016/j.jff.2013.06.009

widely regarded for their sap which is boiled to produce the natural sweetener, maple syrup. Interestingly, for centuries, the aboriginal peoples of these regions consumed maple sap as a tonic and the virtues of maple products have long been known anecdotally. Nowadays, maple sap is the most consumed product in our food chain (i.e. as maple syrup), which is derived entirely from the sap of deciduous trees. The clear maple sap, which is collected by tapping maple trees, is also commonly known as maple water due to its largely 'watery' composition (ca. 95–99%). The constituents of the natural maple sap include macronutrients, micronutrients, and phytochemicals (so called 'phytonutrients') namely, sucrose and other polysaccharides, vitamins, minerals, amino acids, organic acids, phytohormones, and phenolic compounds (Ball, 2007; Li & Seeram, 2010, 2011a; Perkins & van den Berg, 2009; Potter & Fagerson, 1992). Maple sap is primarily boiled to produce maple syrup (ca. 40 L sap = 1 L syrup) in eastern North America, with the province of Quebec in Canada, leading the world's production of this premium natural sweetener. However its natural watery form and refreshing palatability makes it ideal for consumption. In fact, large quantities of sap collected from the maple tree known as 'gorosoe' or 'tree good for the bones' are consumed by South Koreans for a wide range of health benefits (http://www.nytimes.com/2009/03/06/world/asia/06maple.html) and even used in the place of water for cooking as well as for other commercial purposes. Maple water, obtained from the sugar and red maple species, therefore represents an excellent candidate to explore for functional beverage applications in eastern North America beyond its primary use for maple syrup production alone. However, due to its largely watery nature, it is not surprising that once maple sap is collected, it serves as a rich nutrient medium for the growth of microorganisms (Filteau, Lagacé, Lapointe, & Roy, 2012). Therefore, sap would have to be pasteurized or sterilized to eliminate or reduce microbial growth before it would be appropriate for large scale consumption as a commercial beverage.

The identification of phenolic compounds in maple sap and maple syrup, as well as their antioxidant activities, has been previously reported (Kermasha, Goetghebeur, & Dumont, 1995; Li & Seeram, 2010, 2011a, 2011b; Thériault et al., 2006). However, the phenolic content and antioxidant activities of pasteurized and sterilized maple sap in relation to its functional beverage applications have not been studied. In the past few years, our laboratory has been involved in a research program to identify bioactive phytochemicals present in maple syrup and maple plant parts (Li & Seeram, 2010, 2011a, 2011b; Wan et al., 2012; Yuan, Wan, Liu, & Seeram, 2012; Yuan et al., 2011). During these works, we have isolated and identified (by NMR) ca. 100 phenolic compounds from maple and have established qualitative HPLC-DAD profiles using these chemical markers to chemically characterize and standardize various maple products. Thus, given our group's interest in maple chemistry, in the current project we sought to evaluate the effects of pasteurization (at 78 °C) and sterilization (at 140 °C) on the chemical composition (sugars, amino acids, organic acids, minerals and phenolics) and antioxidant activities of maple sap. To the best of our knowledge, this is the first report to characterize the chemical composition and evaluate the antioxidant activities of maple sap after undergoing sterilization and pasteurization in relation to its functional beverage applications.

2. Materials and methods

2.1. Chemicals and general experimental procedures

Nuclear magnetic resonance (NMR) data were recorded on a Varian 500 MHz instrument with TMS as internal standard. Inductively coupled plasma mass spectrometry (ICP-MS) data were acquired on a Thermo Scientific iCAP Qa instrument coupled to an ASX-510 HS High Speed autosampler. All high performance liquid chromatography (HPLC) was performed on a Hitachi Elite LaChrom system (Pleasanton, CA, USA) consisting of a L2130 pump, L-2200 autosampler, equipped with a L-2455 Diode Array Detector (DAD), L-2485 Fluorescence (FL) Detector and L-2490 Refractive Index (RI) Detector all operated by EZChrom Elite software. All semi-preparative scale was conducted on the Hitachi Elite LaChrom system using a Phenomenex Luna C18 column (250 × 10 mm, S-5 μm). Medium pressure liquid chromatography (MPLC) separations were carried out on pre-packed C18 columns (4 × 37 cm) connected to a DLC-10/11 isocratic liquid chromatography pump (D-Star Instruments, Manassas, VA) with a fixed-wavelength detector. All solvents were HPLC grade and were obtained from Pharmco-AAPER through Wilkem Scientific (Pawcatuck, RI, USA). Unless otherwise stated, all reagents including diphenylpicrylhydrazyl (DPPH), the Folin–Ciocalteau reagent as well as sugars (sucrose, glucose, fructose) and organic acid (oxalic acid, L-malic acid, fumaric acid, citric acid, D-tartaric acid, succinic acid) standards were purchased from Sigma–Aldrich (St. Louis, MO, USA). The AccQ Fluor Reagent Kit for evaluation of the amino acid contents was purchased from Waters Corp (Milford, MA, USA). Standards of phenolic compounds were previously isolated by our group from maple syrup and identified using nuclear magnetic resonance (NMR) and other spectroscopic methods (Li & Seeram, 2010, 2011a, 2011b).

2.2. Maple sap samples

Over the past years our laboratory has collaborated with the Federation of Maple Syrup Producers of Quebec (Quebec, Canada) where the vast majority of the world's supply of maple syrup is produced (Li & Seeram, 2010, 2011a, 2011b). Similar to our previous studies on maple syrup and maple plant parts, maple sap samples were provided as original ($n = 34$) pasteurized ($n = 35$) and sterilized ($n = 14$) forms by the Federation of Maple Syrup Producers of Quebec. The sap samples were pasteurized or sterilized using an industrial plate heat exchanger (Alfa-Laval Sterilab, van Den Heuvel, Netherlands; http://www.dairyandfoodequipment.com/en/product/1464) commonly used in the dairy industry for pasteurization of milk. The maple sap samples were pasteurised by HTST (high temperature short time) for 18 s at 78 °C and sterilised by UHT (ultra high temperature) for 4 s at 140 °C, respectively. The sap samples (2 L each) were shipped frozen to our laboratory and immediately stored at −20 °C on receipt. Aliquots (20 mL) of each maple sap sample were freeze-dried for subsequent polyphenol assay (by the

Please cite this article in press as: Yuan, T. et al., Pasteurized and sterilized maple sap as functional beverages: Chemical composition and antioxidant activities, *Journal of Functional Foods* (2013), http://dx.doi.org/10.1016/j.jff.2013.06.009