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High-pressure and thermal-assisted pasteurization of habituated, wild-type, and pressure-stressed *Listeria monocytogenes*, *Listeria innocua,* and *Staphylococcus aureus*

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ABSTRACT

Current study investigated decontamination efficacy of elevated hydrostatic pressure against wild-type, 72-h habituated, and pressure-stressed phenotypes of *Listeria monocytogenes*, *Listeria innocua,* and *Staphylococcus aureus* in milk and orange juice. Pressure intensity levels of 350 and 500 MPa were utilized at 4.4 °C and 60.0 °C for up to 7 min. Temperature was controlled using a stainless steel water jacket surrounding reaction chamber connected to a refrigerated circulating water bath. Results were statistically analyzed using ANOVA. While <2 log reduction of *L. monocytogenes*, *L. innocua,* and *S. aureus* were observed after treatments of up to 7 min at 500 MPa at 4.4 °C, same treatment at 60.0 °C resulted in ≥5 log reductions ($p < 0.05$) of pathogens in milk. Similar trends were observed when the pathogens were inoculated in acidic environment of orange juice and treated at 350 MPa at 4.4 and 60 °C. As such, under the conditions of our experiments, we observed that mild heat could to great extend augment the efficacy of a pressure-based pasteurization treatment. The three phenotypes (habituated, wild-type, and pressure-stressed) of these Gram-positive pathogens exhibited comparable sensitivity to treatments, indicating a validated treatment against wild-type cells could be as efficacious against other phenotypes as well.

1. Introduction

In the last few years, several new vegetable and fruit juices, purees, and milk products have been appeared in the North America and European market because of their antioxidant capacity, presence of elevated bioactive compounds, high nutritional value, and high consumer acceptance (Andlauer & Fürst, 2002; Heckman et al., 2010; Tumpanuvatr & Jittanit, 2012). In the food industry, one of the most important concerns is the multiplication and survival of foodborne pathogens affecting consumers safety by causing infections, intoxication, and/or toxicoinfections (Patrignani et al., 2015). A wide spectrum of foodborne pathogens could cause an array of illnesses in contaminated raw and processed agricultural commodities (Orden et al., 2002; Tauxe, 2002). These contamination episodes could be a concern for individuals of all ages and particularly around 30% of the United States population are considering as at-risk individuals which include pregnant women, the young children, the immunocompromised, and the elderly (Bajpai et al., 2012; Fouladkhah, 2017).

The *Listeria* genus contains non-spore forming Gram-positive bacteria, among them *L. monocytogenes* is recognized as a ubiquitous foodborne pathogenic species (mainly, 1/2a, 1/2b, and 4b serotypes) which causes listeriosis in general and at-risk populations (Rodrigo Tarté et al., 1996; Tauxe, 2002). Listeriosis outbreaks were linked with an array of food products including vegetables, fluid raw milk and dairy products, ready-to-eat commodities, and juices (Ho et al., 2007; McLauchlin et al., 2004; Tauxe, 2002). *L. monocytogenes* was identified and isolated from apple juice (unpasteurized) and an array of raw and processed agricultural commodities (Martinez-Gonzales et al., 2003; Sado et al., 1998). As there is risk involved using pathogenic bacteria in different laboratory studies, *L. innocua* had been suggested and used as a potential surrogate for *L. monocytogenes* (Fairchild & Foegeding, 1993; MacDonald & Sutherland, 1993) in microbiological validation studies.

In addition, *Staphylococci* is a widely distributed Gram-positive bacterium in the environment, which can live on human and animal mucosal and skin surfaces (Soares et al., 2011). Among 49 *Staphylococci* species, *Staphylococcus aureus* is recognized as the most pathogenic

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