Inactivation of Shiga toxin-producing *Escherichia coli* O157: H7 and mesophilic background microbiota of meat homogenate using elevated hydrostatic pressure, mild heat, and thymol

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Abstract: A six-strain mixture of *E. coli* O157:H7 was exposed to 0 to 9 min of six treatments: (i) hydrostatic pressure (400 MPa) at 4 °C; (ii) hydrostatic pressure and thymol at 4 °C; (iii) thymol at 4 °C; (iv) heat at 40 °C; (v) hydrostatic pressure at 40 °C; and (vi) hydrostatic pressure and thymol at 40 °C. Pressure intensity level of 400 MPa and thymol concentration of 0.15% (w/v) were used for the experiments of inoculated pathogen (4.0 to 5.0 log CFU/mL) in a homogenate (10% nonsterilized beef in 90% sterilized distilled water). Temperature was precisely monitored by stainless steel water jacket surrounding pressure chamber (16 mL volume), mechanically linked to a refrigerated circulating water bath. Analyses of variance were conducted followed by Tukey- and Dunnett's-adjusted mean separations. Pathogen counts before treatment were 4.08 ± 0.7 log CFU/mL and were reduced ($P < 0.05$) to 0.67 ± 0.2 log CFU/mL after 6 min of pressure treatment. Thymol and mild heat (40 °C) further augmented decontamination efficacy of pressure treatments where in their presence, the mesophilic background microbiota counts of pressure-treated samples after 3, 6, and 9 min were reduced ($P < 0.05$) by 2.1, 2.5, and 3.1 log CFU/mL, respectively. Results of the current study indicate that thymol and mild heat could enhance decontamination efficacy of elevated hydrostatic pressure for pasteurization of food commodities. This could be of great significance for industry practitioners to assure microbiological safety of a product and cost optimization by benefiting from synergism of antimicrobials, mild heat, and elevated hydrostatic pressure.

Keywords: *Escherichia coli* O157: H7, high-pressure processing, meat homogenate, mesophilic background microbiota

Practical Application: Thymol and mild heat could enhance decontamination efficacy of pressure-based pasteurizer that could be of great significance for practitioners. Application of pressure coupled with antimicrobial and mild heat could assure microbiological safety of a product, lead to cost optimization, and assist in meeting regulatory requirements of food commerce such as Hazard Analysis and Critical Control Point and Preventive Control for Human Food rule of Food Safety Modernization Act. Addition of an antimicrobial could have further co-benefits for the product due to residual protective effects during shelf-life and minimizing potential undesirable organoleptic changes associated with pressure treatments of >400 MPa.

1. INTRODUCTION

As a commensal bacterium, *Escherichia coli* resides within the gastrointestinal area of many warm-blooded animals and humans. While vast majority of *E. coli* serotypes do not cause any health complication, exposure to limited serogroups of this bacterium could lead to intestinal and extraintestinal health complications in human (Fouladkhah, Geornaras, & Sofos, 2013; Fouladkhah, Geornaras, Yang, & Sofos, 2013; Kabir et al., 2019). In the United States, vast majority of foodborne illness episodes associated with pathogenic *E. coli* belong to Enterohaemorrhagic pathotype of this pathogen. *Escherichia coli* O157: H7 is the most epidemiologically significant serogroup of this pathotype and is capable of encoding Shiga toxin that could lead to hemorrhagic colitis and in rare cases (particularly for children under the age of 5) could lead to potentially lethal kidney complication in form of hemolytic uremic syndrome (Kaper, Nataro, & Mobley, 2004). As a pathogen, *E. coli* O157: H7 had been first reported in 1980s (Lim, Yoon, & Hovde, 2010; Paton & Paton, 1999; Wells et al., 1983) and gained importance in food commerce and regulatory affairs after involvement in a highly publicized hemorrhagic colitis outbreak of hamburgers in a fast food chain in 1992–1993 in pacific northwest of the United States (CDC, 1993).

According to active surveillance data of the U.S. Centers for Disease Control and Prevention (CDC), in a typical year in the United States, more than 63,000 individuals experience illness caused by O157 serogroup of Shiga toxin-producing *E. coli*. Around 68% of these illnesses are foodborne in nature, leading to hospitalization in 46.2% of the illness episodes with 0.5% death rate (Scallan et al., 2011). In addition to the above-mentioned 1992–1993 pacific northwest outbreak and an array of sporadic episodes, based on data from National Outbreak Reporting System (NORS) of CDC, there have been at least 715 (of which 68.8% were foodborne in nature) single and/or multistate outbreaks from 1998 to

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