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## Effect of high-pressure processing of bovine colostrum on immunoglobulin G concentration, pathogens, viscosity, and transfer of passive immunity to calves

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### ABSTRACT

This study aimed to determine the effects of high-pressure processing on the immunoglobulin concentration, microbial load, viscosity, and transfer of passive immunity to calves when applied to bovine colostrum as an alternative to thermal pasteurization. A pilot study using *Staphylococcus aureus* was conducted to determine which pressure-time treatments are most appropriate for use with bovine colostrum, with the goals of maximizing bacterial inactivation while minimizing IgG content and viscosity changes. Following the pilot study, an inoculation study was conducted in which first-milking colostrum samples from Holstein-Friesian cows were inoculated with known concentrations of various bacteria or viruses and pressure processed at either 300 MPa for up to 60 min or at 400 MPa for up to 30 min. The recovery of total native aerobic bacteria, *Escherichia coli*, *Salmonella enterica* ssp. *enterica* serovar Dublin, *Mycobacterium avium* ssp. *paratuberculosis*, bovine herpesvirus type 1, and feline calicivirus were determined after processing. Colostrum IgG content was measured before and after pressure processing. Shear stress and viscosity for each treatment was determined over shear rates encompassing those found during calf feeding and at normal bovine body temperature (37.8°C). Following a calf trial, serum IgG concentration was measured in 14 calves fed 4 L of colostrum pressure processed at 400 MPa for 15 min. In the pilot study, *S. aureus* was effectively reduced with pressure treatment at 300 and 400 MPa (0, 5, 10, 15, 30, and 45 min), with 2 treatments at 400 MPa (30,

45 min) determined to be inappropriate for use with bovine colostrum due to viscosity and IgG changes. High-pressure processing at 300 MPa (30, 45, and 60 min) and 400 MPa (10, 15, and 20 min) was shown to effectively reduce total native aerobic bacteria, *E. coli*, *Salmonella* Dublin, bovine herpesvirus type 1, and feline calicivirus populations in bovine colostrum, but no decrease occurred in *Mycobacterium avium* ssp. *paratuberculosis*. All inoculation study pressure treatments insignificantly decreased IgG content of colostrum. Treatment of colostrum at 400 MPa for 15 min during the calf trial decreased IgG content of colostrum. Treatment at 400 MPa for 15 min increased colostrum viscosity, with 2 of 14 samples requiring dilution with water for calf feeding. Calves fed pressure-processed colostrum had similar serum IgG but lower efficiency of absorption than calves fed heat-treated colostrum. The results of this study suggest that high-pressure processing of bovine colostrum maintains an acceptable IgG level while decreasing bacterial and viral counts. Changes in viscosity sometimes made calf feeding more difficult, but still feasible. Additional research to optimize this technology for on-farm use is necessary.

**Key words:** high-pressure processing, bovine colostrum, pasteurization, *Mycobacterium avium* ssp. *paratuberculosis*

### INTRODUCTION

Transfer of maternally derived antibodies via ingestion of an adequate quantity of good-quality colostrum has long been understood to be fundamentally important for health and future productivity of replacement heifers (Robison et al., 1988; Weaver et al., 2000). Inadequate absorption of maternal antibodies, specifically IgG1, is associated with significant economic burden for the dairy industry due to increased morbidity and mortality rates in preweaned calves (Wittum and Perino, 1995; Weaver et al., 2000). Though essential for provid-

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ing protective immunity until calves' immune response matures, colostrum feeding can be a potential risk factor for the direct transmission of specific pathogenic bacteria and viruses shed by the dam, or environmental pathogens acquired during handling (McGuirk and Collins, 2004). Microbial contamination of colostrum may also decrease IgG absorption, and thereby contribute to failure of transfer of passive immunity (James et al., 1981; Poulsen et al., 2002). Despite emphasis on clean colostrum protocols, bacterial loads exceeding industry goals (less than  $10^5$  cfu/mL total bacterial plate count, and less than  $10^3$  cfu/mL coliform plate count) are consistently found in colostrum (McGuirk and Collins, 2004), with reports of 92% (Swan et al., 2007) and 82% (Poulsen et al., 2002) of colostrum samples from dairies located in the Midwestern United States having total bacterial loads of more than  $10^5$  cfu/mL.

Specific pathogens shed in colostrum can be due in part to endemic herd infection and include *Salmonella enterica* ssp. *enterica* serovar Dublin, *Mycobacterium avium* ssp. *paratuberculosis* (MAP), *Mycoplasma* spp., and bovine leukemia virus (BLV; Ferrer and Piper, 1981; Streeter et al., 1995; Gonzalez and Wilson, 2003; Houser et al., 2008). To reduce the risk for perpetuation of infectious disease in calves from contaminated colostrum, veterinarians and dairy replacement heifer raisers have adopted multiple management strategies. Some farms are able to avoid feeding colostrum from specific disease positive (e.g., BLV or MAP) cows, but this can create shortages in colostrum supply (Poulsen et al., 2010). Such shortages may be compensated for with the use of commercially available colostrum replacement products derived from freeze-dried bovine colostrum or serum, reported effective at providing passive immunity (Jones et al., 2004; Foster et al., 2006; Godden et al., 2009; Pithua et al., 2009, 2010; Poulsen et al., 2010). Colostrum may also be treated to reduce microbial contamination on farm, with heat treatment currently the most common method (reviewed in Donahue et al., 2012).

Initial experiments using high temperature consistent with the Pasteurized Milk Ordinance showed temperatures greater than  $62.5^\circ\text{C}$  resulted in increases in viscosity and denaturation of maternal IgG, which made colostrum unacceptable to feed to calves (Meylan et al., 1996; Tyler et al., 2000; Godden et al., 2003). To avoid these negative side effects, heat pasteurization techniques have since been modified to significantly decrease microbial pathogens while maintaining sufficient IgG concentration to feed to replacement dairy heifer calves (Godden et al., 2006; McMMartin et al., 2006; Johnson et al., 2007; Elizondo-Salazar and Heinrichs, 2009; Donahue et al., 2012). When this was implemented on commercial dairies, Donahue et al. (2012)

reported heat treatment of colostrum at  $60^\circ\text{C}$  for 60 min resulted in significant reductions in total bacteria and coliform counts and no change in IgG concentration. Yet, clinically significant bacterial concentrations remained in treated colostrum fed to calves, suggesting room for improvement.

High-pressure processing (HPP) is a nonthermal technology that provides a promising alternative to traditional thermal pasteurization. It was first applied to bovine milk by Hite (1899) to effectively extend shelf life. High-pressure processing is most commonly applied to foods packaged in flexible pouches that are immersed in a pressure vessel. The pressure inside the sealed vessel is increased by pumping pressure transmitting fluid (commonly an oil-in-water emulsion) into the vessel (San Martín et al., 2002; Balasubramaniam and Farkas, 2008). During the process, some adiabatic heating may occur due to pressure buildup, but heating is moderate compared with thermal processes. High-pressure processing also offers process uniformity since pressure is applied instantaneously in all directions (Cheftel, 1995; San Martín et al., 2002; Balasubramaniam and Farkas, 2008). Like thermal treatments, HPP is able to inactivate pathogens and spoilage microorganisms; however, unlike some thermal treatments, it preserves compounds that are sensitive to elevated temperatures. This combined microbial inactivation and preservation of quality results in a food or other biological product that is both safe and of a higher quality (Cheftel, 1995; San Martín et al., 2002; Balasubramaniam and Farkas, 2008). Although expensive for large-scale production, HPP provides a viable processing alternative for high-value, low-volume products (San Martín et al., 2002). Currently, the availability of HPP is predominantly limited to large-scale human food operations, yet discussions with manufacturers of this equipment suggest that small units for on-farm use could be obtained for approximately \$25,000.

Few studies have evaluated the use of HPP for maintaining high levels of immunoglobulins while reducing microbial contamination in colostrum. Masuda et al. (2000) concluded that HPP could effectively extend shelf-life of bovine colostrum while maintaining immunoglobulins, and Viazis et al. (2007) showed HPP-treated human milk had higher immunological activity than heat-treated milk. Additionally, Viazis et al. (2008) determined that HPP inactivated pathogens of concern in human milk. They determined that while HPP showed great promise as a suitable alternative technology to thermal pasteurization of human milk, further study was required on the effects of HPP on viruses such as cytomegalovirus, HIV, and hepatitis serotypes. Similar to these observations, in the current study it was hypothesized that effective microbial re-

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