Evaluation of heat and pH treatments on degradation of ceftiofur in whole milk

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Introduction

Waste milk feeding practices have been implicated as a potential source for disseminating antimicrobial resistant bacteria among animals and the environment. Feeding dairy heifers raw milk with low concentrations of antimicrobials increases the proportion of antimicrobial resistance. Two potential interventions to degrade antimicrobial drugs in milk are heat and pH treatment. The aim of this study was to evaluate the effect of heat and pH treatments on the degradation of ceftiofur and ceftiofur free acid equivalents (CFAE) in milk at concentrations previously found in waste milk on dairy farms.

Materials and Methods

Saleable whole milk was spiked with chemical grade ceftiofur sodium to a final concentration of 200 or 400 ppb for heat or pH trials, respectively. Four replicates were performed for each set of treatments. Three temperature treatments were evaluated: low-temperature long-time (LTLT, 63°C for 30 minutes), high temperature short time (HTST, 72°C for 15 seconds) and high-temperature long-time (HTLT, 92°C for 20 minutes). A control group stored at room temperature was used. Two pH treatments were evaluated: low pH group (LpH), using formic acid to achieve a pH of 4.0, and high pH group (HpH), by adding sodium hydroxide until a pH of 10.0 was achieved as measured using a pH meter. A control milk group (pH ~6.5-6.7) was also used. Samples were stored at -80°C until tested for drug quantification, immediately after the treatment was evaluated. Ceftriaxone was quantified using liquid chromatography mass spectrometry (LC-MS/MS) at the California Animal Health & Food Safety Toxicology laboratory (Davis, CA). The concentration of CFAE was measured using high-performance liquid chromatography (HPLC) at the UC Davis Veterinary Drug Residue Laboratory. CFAE was quantified only for samples from the high pH treatment group because it was the only pH group for which ceftiofur degradation was observed. A linear regression analysis was performed in SAS (SAS Institute, Cary, NC), controlling for repetitive measures.

Results

The initial concentrations of CFAE for heat and pH treatments were 128.5 ± 7.44 and 234 ± 17.8 ppb, respectively. There was a significant effect on the degradation of CFAE for HTLT treatment, which represented a 35.24% degradation of the initial concentration of CFAE (83.2 ± 6.59 ppb). The degradation of CFAE did not differ between the control and the remaining two heat treatment groups (control: 129.9 ± 5.69, HTST: 127.56 ± 6.59, HTLT: 124.78 ± 6.59 ppb). Concentration of CFAE remained stable at normal (213.75 ± 17.80 ppb) and low pH (240.33 ± 10.28 ppb) but declined significantly immediately after sodium hydroxide was added and pH 10 was achieved (10 ± 10.28 ppm). Initial concentrations of CFAE (269.5 ± 4.16 ppb) in samples decreased after milk was alkalized to pH 10 (10 ± 4.16 ppb). Increasing milk pH resulted in the degradation of the 95.72 and 96.28% of the initial concentration of CFAE and CFAE, respectively.

Significance

Alkalinizing milk to pH 10 using sodium hydroxide increased the degradation of ceftiofur and CFAE in milk. Heating milk to 92°C for 20 minutes also decreased ceftiofur concentrations in milk samples. pH and heat treatments are cost effective on-farm strategies that could decrease antimicrobial concentrations in waste milk below Food and Drug Administration approved tolerances, without causing milk coagulation. Further studies to evaluate the possibility of using these approaches on farms are needed, including normalization of the milk pH to allow safe consumption of milk by calves, effect of treatment on milk nutrients, and evaluate the treatment’s effect on calf intake.