

Multi-drug residues and antimicrobial resistance patterns in waste milk from dairy farms in Central California

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Introduction

In the dairy industry, most antimicrobial drugs used to treat lactating cows result in the production of non-saleable milk due to the presence of drug residues above the tolerable concentration established by the U.S. Food and Drug Administration (FDA). To mitigate the economic loss due to waste milk, 33% of dairy farms in the U.S. feed this milk to pre-weaned calves. Though economically sound, this practice may favor increased selection of antimicrobial-resistant bacteria. Currently, there are no reports evaluating the presence and concentration of drug residues in waste milk on California dairies. The aims of this cross-sectional study were to: 1) identify and measure concentrations of antimicrobial residues of waste milk samples from dairy farms in Central California, 2) identify management practices associated with the occurrence of specific antimicrobial residues in waste milk, and 3) characterize the antimicrobial resistance patterns of *E. coli* cultured from waste milk samples.

Materials and Methods

Bulk tank waste milk samples were collected from 25 dairy farms located in California's Central Valley. A questionnaire was used to collect information about farm management practices. Waste milk samples were analyzed for a multi-drug residue panel using liquid chromatography-tandem mass spectrometry (LC-MS/MS). Bacteria were cultured and antimicrobial resistance was assessed with standard techniques; milk quality parameters (fat, protein, lactose, solids non-fat, somatic cell count, coliform count, standard plate count) also were measured.

Results

Of the 25 samples, 11 (44%), 4 (16%), and 1 (4%) had detectable levels of β -lactam, tetracycline, and sulfonamide residues, respectively. Of these, 5 (20%) had β -lactam and

2 (8%) had tetracycline residues above the tolerance/safe limits established by the FDA. Zero samples had sulfonamide residues above the FDA tolerance/safe limit. The most prevalent drug residues were ceftiofur ($n = 7$ (28%); mean \pm SE = 251 ± 177.0 ng/ml), oxytetracycline ($n = 4$ (16%); 16378 ± 308.7) and cephapirin ($n = 3$ (12%); 39 ± 24.5). Of these, ceftiofur (88%) and cephapirin (40%) also were the most commonly used to treat mastitis based on questionnaire answers. *Streptococcus* spp. ($n = 21$ (84%); $51,010 \pm 19,631$ cfu/ml) was the most common bacterium cultured from waste milk samples, followed by *Staphylococcus* spp. ($n = 20$ (80%); $8,625 \pm 4,939$ cfu/ml), *Escherichia coli* ($n = 10$ (40%); $5,870 \pm 2,478$ cfu/ml), non-fermentative gram-negative organisms ($n = 8$ (32%); $9,625 \pm 3,087$ cfu/ml), *Staphylococcus aureus* ($n = 5$ (20%); $121,400 \pm 119,652$ cfu/ml), *Bacillus* spp. ($n = 5$ (20%); 860 ± 563 cfu/ml), and *Klebsiella* spp. ($n = 4$ (16%); $78,750 \pm 73,771$ cfu/ml). *Mycoplasma* spp. was cultured from two waste milk samples (8%). Nine of the 10 *E. coli* isolates were resistant to three or more antimicrobials. In this study, 20% of sampled farms did not pasteurize waste milk prior to feeding to calves.

Significance

Our study findings indicate that waste milk fed to calves on the study dairies in California's Central Valley contained residues of antimicrobial drugs important to veterinary medicine. The presence of multidrug resistant *E. coli* in waste milk urges the need for on-farm practices, such as pasteurization, that reduce calf exposure to resistant bacteria. The presence of drug residues in waste milk at concentrations that have been shown to increase selection of resistant bacteria indicate the need for additional studies targeting on-farm milk treatments that could degrade drug residues in waste milk before feeding to calves.