FREQUENCY OF ANTIBIOTIC RESIDUES IN A CENTRAL WISCONSIN DAIRY

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ABSTRACT

Antibiotics, used to maintain healthy dairy animals, persist in milk produced by treated animals for several days after therapy. Antibiotic residues, if present in the milk supply, negatively impact suitability for processing and consumption. This is an area of concern in the dairy industry and for the general public. This study explores the incidence of antibiotic residues in milk provided to a Central Wisconsin dairy. Random, unidentified samples were obtained over a four month period from producers located within a 100–mile radius of a dairy located in Granton, Wisconsin. All samples were tested within one week of their receipt, and maintained at refrigerated temperatures at all times prior to testing. Following testing, the origin of the samples was revealed. Samples were derived from two milk populations: those intended for human consumption (bulk milk), and those from cows under surveillance following antibiotic treatment (milk removed from the commercial supply). All bulk milk samples tested negative for over 55 different antibiotic residues, while all samples from the surveillance population tested positive for antibiotic residues. Our results are consistent with those observed nationally, and suggest that dairy producers recognize the ongoing concern of antibiotic contamination in the milk supply and are taking steps to prevent antibiotic contamination in milk. Our findings also suggest the public should have minimal concern with respect to antibiotic residues in the commercial milk supply.

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KEYWORDS

- Antibiotics
- Drug residues
- Dairy foods
- Food safety
- *Bacillus stearothermophilus* var. *calidolactis*

INTRODUCTION

Antibiotic use on dairy farms is imperative to treating ill cows. Use of antibiotics is most common in the treatment of mastitis, a painful inflammation of the cow's udder (8). There are approximately 40 drugs approved for use in lactating dairy cows (14). Once an ill cow is treated, the antibiotics may be retained in the cow body for several days. This makes it important for producers to test regularly for drug residues in treated cows. Generally, all antibiotics should be excreted from milk within 72 hours of treatment (7). However, if not used properly, antibiotics can be retained in the cow for longer periods. The Food and Drug Administration surveys indicate that improper use of drugs in the

control of mastitis is the major source of antibiotic residues found in milk supply (10).

If present, antibiotic drug residues in our milk supply pose a public health problem. Specifically, the consumption of milk contaminated with antibiotics can detrimentally influence our ability to treat human infections, because infectious agents adapt to the continual exposure of drugs (12). Further, for those individuals who are already sensitized to antibiotics, overuse of these antibiotics in dairy cows could result in allergies if these products make their way to our local grocery stores. For example, penicillin is the most frequent human drug allergy and is common – affecting approximately 10% of the population with anaphylaxis in 0.01% and fatal reactions in 0.0015% of cases (1).

Most concern with antibiotic persistence is placed on the development and spread of virulent and resistant bacteria within the dairy animal. These antimicrobial-resistant pathogens may emerge in the food production chain, and can be transmitted to humans, and cause infection. Salyers and Shoemaker outline the potential concern of the ability of bacteria to exchange resistance genes through their exposure to the intestinal tracts of consumers, as well as their potential to interact with any pathogenic bacteria in passage through the human colon (13).

Human exposure to resistant bacteria from non-human usage of antibiotics can create new, more severe, infections that would not have otherwise occurred, and cause an increased frequency of treatment failures (5).

Because the foodborne route is a major transmission pathway for bacteria and resistance genes (5), as well as antibiotic residues themselves, understanding farming practices relating to managing and treating ill cows becomes relevant to our everyday lives, especially for those purchasing commercial milk and milk products.

Consequently, educating the public on milk sanitation practices and the safety of our nation's milk supply is an important area of research. Our investigation was aimed at exploring the possible public health issue of antibiotic persistence in milk products through the collection and testing of raw milk samples collected from a dairy in Granton, WI. Current national findings place the frequency of antibiotic persistence in the commercial milk supply below 0.015% (14–16), and suggest that the public, at present, should have minimal concern regarding antibiotic residues when consuming commercial milk products. We hypothesize that the commercial milk supply contains very little (and likely undetectable levels) antibiotic residues.

MATERIALS AND METHODS

Milk samples were obtained from Lynn Dairy Inc. (located in Granton, Wisconsin), between March 09th, 2015 – June 07th, 2015. At the time of collection, samples were identified through a six-digit number with no indication of milk supplier or type of milk. Milk suppliers for these samples spanned a 100-mile radius from the dairy plant where milk samples were processed. Samples were received in batches of 25 and each batch was tested within a week of receipt. Prior to testing, samples were stored in a refrigerator below 6°C.

A total of 268 samples were tested for antibiotic residues using DSM's Delvotest SP NT, a simple and reliable test that detects over 55 different types of antibiotics including beta lactams, sulphonamides, aminoglycosides, quinolones, macrolides and tetracyclines (4). The Delvotest incubator allows for 10 ampoules to be tested at a time. In this study, 9 milk samples and 1 control sample of penicillin were tested each time. Separate pipettes were used to add 0.1 ml of each milk sample into the designated ampoules. Each ampoule was marked with the six-digit sample number provided at time of collection for sample identification. Individual pipettes were dipped 1 cm into the sample which was then transferred onto the agar medium in the designated ampoule. The incubation temperature was set at 64 degrees Celsius and testing took approximately 3 hours (2).

The test was read through analysis of the solid and buffered agar medium containing a pH indicator and a test organism, *Bacillus stearothermophilus* var. *calidolactis* (3). A purple/blue reading implies the reduced growth of the test organism, and therefore a positive test for antibiotics in the raw milk. A green/yellow test implies the growth of the test organism, and therefore a negative test result for antibiotics in the sample.

Table 1. Antibiotic residue analysis of milk samples in this study

Source of Sample	Total Samples	Number Positive	Percent Positive	Disposition per Pounds
Bulk Milk Pickup Tanker	3,147,302	429	0.014%	17,754,000
Pasteurized Fluid Milk and Milk Products	37,707	0	0.000%	0
Producer	445,223	266	0.060%	240,000
Other	49,953	8	0.016%	99,000

RESULTS

Following testing, milk sample numbers were identified as originating from bulk milk samples or milk from animals under surveillance for antibiotic residues. The bulk milk samples total 264 of the 268 samples tested negative for antibiotic residues. The remaining four samples were obtained from animals under surveillance for antibiotic residues and all four of these samples tested positive for antibiotic residues (Table 1).

DISCUSSION

BULK TANK SAMPLES

All 264 samples originating from bulk milk samples were those that were deemed fit for bulk tank inclusion by the dairy producer, and thus added to the truckload. However, prior to acceptance, dairy processors will test the truckload once it arrives at the processing establishment as a precautionary measure. If the bulk tank is negative for antibiotic residues, it is presumed that the tank is antibiotic free and safe to use. If a bulk tank is positive for antibiotic residues, the truckload will be dumped. Additionally, individual bulk, raw milk samples from the truckload will be tested in order to determine the cause of contamination. In our study, all samples deemed fit for bulk tank inclusion were completely antibiotic free.

NON-BULK TANK SAMPLES FROM ANIMALS UNDER SURVEILLANCE

Next, it is important to note that the four positive test results observed in our study were not considered "true positives" for antibiotics as these milk samples were not intended to be included into the bulk tank by the dairy producer. Specifically, milk producers may send individual milk samples from treated cows to milk processors for antibiotic testing. This precautionary routine is used to see if the drug still persists within the treated cow's system (6). As it is unlawful to place unfit milk on the market for human consumption, this primary screening method is integral in determining whether the milk is fit for inclusion in the bulk tank (6). Importantly, these samples are relevant to our data as it provides evidence that milk producers are exercising safe milk sanitation practices by monitoring treated cows and separating their milk from the central milk supply.

CONCLUSIONS

Our findings are consistent with surveys from the Food and Drug Administration's annual report on national milk drug residues. The national survey under the FDA has been conducted since 1994, and illustrated the findings of bulk milk antibiotic testing over the course of 20 years (16). These surveys show a peak in 1996 with 0.1% of bulk milk samples testing positive for antibiotic residues to its current low of 0.014% in 2014 (16).

The most recent FDA survey was conducted from October 1, 2013 to September 30, 2014 (16). A total of 4,008,662 tests were conducted, consisting of nine different groups of individual drugs or different drug families. A total of 3,680,185 samples were collected and analyzed for animal drug residues. Of those samples, only 703 were positive for a drug residue. The majority of samples (3,147,302) were samples from bulk milk pickup tankers of raw milk. Only 429 samples, or 0.014%, tested positive for a drug residue. Importantly, no antibiotic residues were found in samples that came from pasteurized fluid milk, or samples considered "retail-ready" (16). This would suggest that for our study, over 7000 bulk milk samples from our dairy would need to be surveyed before a single positive test would be likely.

The National Milk Drug Residue Database is a voluntary industry reporting program that identifies the following: the extent of national testing activities, the analytical methods used, the kind and extent of the animal drug residue identified, and the amount of contaminated milk (16). Mandatory reporting of drug residues in milk is required under the Pasteurized Milk Ordinance; a strict set of requirements

for milk production, transportation, pasteurization, equipment sanitation, and labeling (15). Dairy farmers and processors work closely with the Food and Drug Administration, the U.S. Department of Agriculture (USDA), and state regulatory agencies to promote the highest safety standards for milk (9). As such, it is not surprising that we found no bulk milk samples testing positive for antibiotic residues in our study given its sample size and these control measures.

Reducing the use of antibiotics minimizes the likelihood of antibiotic resistant bacteria to develop, as well as drug residues to exist in the milk supply. Maintaining antibiotic free milk begins with the efforts of dairy producers in implementing effective milk sanitation practices. Further, it is essential that producers and milk processors build trusting partnerships that allow for effective and economically sound business relationships. Preliminary screening methods for drug residues at milk processing establishments, previously discussed in this study, are critical in the continued prevention of contaminating truckloads of milk. In conclusion, our study is in agreement with national surveys, which have demonstrated that these efforts are being maintained, as our results, along with results on a national level, show extremely low incidences of antibiotic persistence in milk. Ultimately, these findings should provide reassurance that milk products will continue to be safe for consumption, and that efforts in reducing antibiotic contamination are ongoing and well implemented by the industry.

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