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Transport of three veterinary antimicrobials from feedlot pens via simulated rainfall runoff

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HIGHLIGHTS

- Antimicrobial concentrations were higher in bedding versus non-bedding material.
- Bulk density rather than water content of pen floor material determined runoff volume.
- More runoff was generated from the non-bedding area compared to the bedding area.
- Physical properties of antimicrobials determined relative mass transported in runoff.
- Order of transport in runoff: sulfamethazine > tylosin > chlortetracycline

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ABSTRACT

Veterinary antimicrobials are introduced to wider environments by manure application to agricultural fields or through leaching or runoff from manure storage areas (feedlots, stockpiles, windrows, lagoons). Detected in manure, manure-treated soils, and surface and ground water near intensive cattle feeding operations, there is a concern that environmental contamination by these chemicals may promote the development of antimicrobial resistance in bacteria. Surface runoff and leaching appear to be major transport pathways by which veterinary antimicrobials eventually contaminate surface and ground water, respectively. A study was conducted to investigate the transport of three veterinary antimicrobials (chlortetracycline, sulfamethazine, tylosin), commonly used in beef cattle production, in simulated rainfall runoff from feedlot pens. Mean concentrations of veterinary antimicrobials were 1.4 to 3.5 times higher in surface material from bedding vs. non-bedding pen areas. Runoff rates and volumetric runoff coefficients were similar across all treatments but both were significantly higher from non-bedding (0.53 L min^{-1}; 0.27) than bedding areas (0.40 L min^{-1}; 0.19). In keeping with concentrations in pen surface material, mean concentrations of veterinary antimicrobials were 1.4 to 2.5 times higher in runoff generated from bedding vs. non-bedding pen areas. Water solubility and sorption coefficient of antimicrobials played a role in their transport in runoff. Estimated amounts of chlortetracycline, sulfamethazine, and tylosin that could potentially be transported to the feedlot catch basin during a one in 100-year precipitation event were 1.3 to 3.6 g $head^{-1}$, 1.9 g $head^{-1}$, and 0.2 g $head^{-1}$, respectively. This study demonstrates the magnitude of veterinary antimicrobial transport in feedlot pen runoff and supports the necessity of catch basins for runoff containment within feedlots.

1. Introduction

Most confined animal feeding operations in western Canada administer veterinary antimicrobials therapeutically to treat disease and sub-therapeutically not only to prevent disease but also as growth promoting agents to enhance feed efficiency. Veterinary antimicrobials commonly administered sub-therapeutically in feed or water to promote feed efficiency and prevent disease include chlortetracycline, sulfamethazine, tylosin, and monensin (Lee et al., 2007). Other growth promoting agents administered to cattle include exogenous hormones

Abbreviations: CTC, *iso*-chlortetracycline treatment; CTCSMZ, chlortetracycline plus sulfamethazine treatment; REC, runoff export coefficient; TYL, tylosin treatment; VRC, volumetric runoff coefficient.

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(Lee et al., 2007) such as 17β-estradiol and testosterone, the non-steroidal phytoestrogen α-zearalanol, and the synthetic steroidal progestin melengesterol acetate. A portion of these administered growth promoting agents may be excreted in urine and or feces either unchanged or as metabolites, some of which may remain biologically active. For example, tylosin administered to cattle is metabolized to macrosin (tylosin C), relomycin (tylosin D), desmethyl tylosin D (all excreted in the feces), and cysteinyl-tylosin A (excreted in the urine) (Lewicki et al., 2009). About 11 to 37% of the sulfamethazine that is administered to cattle is excreted in urine (Nouws et al., 1988).

Beef cattle manure from feedlots has been shown to contain veterinary antimicrobials such as chlortetracycline, sulfamethazine, tylosin (Cessna et al., 2011; Sura et al., 2014), oxytetracycline, methacycline, sulfadimidine, sulfaquanidine, ciprofloxacin, lomefloxacin, danofloxacin and enrofloxacin (Zhao et al., 2010). In addition, commercial feedlot soils (Aust et al., 2008; Netthisinghe et al., 2013), feedlot catch basins (Zhang et al., 2013), runoff from manure-amended soils (Amarakoon et al., 2014; Kuchta and Cessna, 2009; Kuchta et al., 2009), and surface and ground water near intensive cattle feeding operations (Bartelt-Hunt et al., 2011; Watanabe et al., 2008, 2010) have been shown to contain veterinary antimicrobials at various concentrations.

The presence of veterinary antimicrobials in runoff from confined animal feeding operations, manure storage facilities, or manure-amended crop and pasture land is of concern, especially if the runoff enters receiving waters. Relatively high concentrations of these compounds in feedlot manure may promote the development of antimicrobial resistance in bacteria, thus compromising not only the efficacy of antimicrobials used in human medicine but also environmental health (Kim et al., 2010; Lee et al., 2007). Indeed, beef cattle manure and feedlot catch basin water has been shown to contain antimicrobials and resistance genes (Storteboom et al., 2007; Zhang et al., 2013). Consequently, not only veterinary antimicrobials but also nutrients, steroid hormones and antimicrobial resistant bacteria along with the resistance genes they harbor may move from surface runoff to receiving waters. Veterinary antimicrobials have been reported in surface waters in Canada (Forrest et al., 2011; Lissemore et al., 2006; Waiser et al., 2011), the United States (Kolpin et al., 2002; Yang and Carlson, 2003), Europe (Calamari et al., 2003; Carmona et al., 2014; Christian et al., 2003; Gros et al., 2012; Hamscher et al., 2002) and Asia (Chen and Zhou, 2014; Managaki et al., 2007; Zhang et al., 2014).

To minimize transport of veterinary antimicrobials, antimicrobial resistant pathogens and resistance genes, steroid hormones, and nutrients from feedlots and manure storage sites via rainfall/snowmelt runoff, catch basins can be installed within these facilities to contain runoff. Catch basins (also known as retention ponds or runoff holding ponds) are designed as temporary storage facilities for runoff. In the province of Alberta (Canada), catch basins for feedlots and manure storage facilities are required to hold runoff resulting from a 1 in 30 years 24-h rainstorm and to have a protective liner to prevent infiltration through the bottom of the catch basin (Province of Alberta, 2013). The contents of catch basins must be managed so that contaminants do not leave the feedlot or manure storage property, and do not enter surface waters. Options for managing the contents of the catch basins include application of basin contents to adjacent cropland either by tanker truck, sprinkler irrigation (excluding crops for human consumption which are intended to be consumed raw/uncooked), or slow flood release using flow rates that ensure infiltration into the soil (Alberta Agriculture and Rural Development, 2012).

In an Alberta study, Miller et al. (2004) measured inorganic nutrients (nitrogen and phosphorus) in simulated rainfall runoff from pens in a research feedlot, and catch basin water for total heterotrophs, total coliforms and *Escherichia coli* bacteria. High total phosphorus concentrations in the runoff and continual high populations of bacteria in catch basin water demonstrated the need for containment of feedlot runoff. In another study, Miller et al. (2006) used the same feedlot to assess the effect of bedding material and location within feedlot pens on nutrient concentrations and bacterial loads. Nutrient concentrations and bacterial populations were higher in the bedding than the non-bedding areas of the pens, making bedding areas a greater contributor of nutrients and bacteria in runoff. Using the same research feedlot, the objectives of the current study were to determine how the concentrations of three veterinary antimicrobials (chlortetracycline, sulfamethazine, and tylosin) commonly administered to beef cattle in Canada, differed among bedding and non-bedding areas in feedlot pens, to determine how the concentrations of these veterinary antimicrobials varied with time in simulated rainfall runoff from the pens, and to quantify the amount of each antimicrobial that could potentially be transported to the catch basin.

2. Materials and methods

2.1. Study design, setup and antimicrobial treatments

The study was conducted in a research feedlot facility (described in detail by Miller et al., 2004) at the Agriculture and Agri-Food Canada Research Centre, Lethbridge, AB, Canada (49°38′ N, 112°48′ W). On 14 Dec. 2010, twelve pens (4 antimicrobial treatments × 3 replicates) in the research feedlot were each assigned 10 steers. Pen dimensions were 14 m × 19.5 m with a stocking density of 27 m^2 $head^{-1}$, which is somewhat lower than that of commercial feedlots (17 m^2 $head^{-1}$) (Kennedy et al., 1999). Cattle in each pen were bedded with equal amounts of barley (*Hordeum vulgare* L.) straw as required for winter comfort and in accordance with animal care guidelines for proper housing (CCAC, 2009). Because of the small size of the pens, relative to those in commercial feedlots, the bedding area in each pen occupied a much greater proportion (~65%) of the pen floor area (Fig. 1) compared to that in commercial feedlots. The remaining portion of the pen floor was categorized as non-bedding area.

The four antimicrobial treatments were established by fortifying feed with antimicrobials, commonly used on beef cattle in Canada, on dry-weight basis at recommended sub-therapeutic levels (Compendium of Veterinary Products, 2003): (1) CTC consisted of chlortetracycline at 44 mg kg^{-1} feed (Aureomycin, Alpharma Inc., Bridgewater, NJ), equivalent to 350 mg $head^{-1}$ day^{-1}; (2) CTCSMZ consisted of a combination of chlortetracycline at 44 mg kg^{-1} feed and sulfamethazine at 44 mg kg^{-1} feed (Aureo S-700 G, Alpharma Inc., Bridgewater, NJ), equivalent to 350 mg $head^{-1}$ day^{-1} each; TYL consisted of tylosin at 11 mg kg^{-1} feed (Tylan, Elanco Animal Health, Calgary, AB, Canada), equivalent to 87.5 mg $head^{-1}$ day^{-1}; and CON (control) consisted of feed without antimicrobials. Cattle were fed a 70% barley silage, 25% barley grain, 5% supplement diet for the first 80 days and then transitioned over 21 days to an 85% barley grain, 10% barley silage, 5% supplement diet which was fed until the cattle were ready for market. The manure was not removed from pens and had accumulated for a period of 153 days (manure accumulation period) when the simulated rainfall runoff experiments were initiated.

2.2. Simulated rainfall runoff experiments

2.2.1. Rainfall simulation, runoff rates, and Pen floor sample collection

After 153 days (14 Dec. 2010 to 16 May 2011) of the feeding and antibiotic administration period, cattle were temporarily re-located to allow rainfall simulation experiments in the pens. The bedding and non-bedding areas of the feedlot pens within the research facility would have been similar to those in commercial feedlots. The profile of a feedlot pen floor generally consists of three distinct layers: a manure/bedding pack, a black interface layer, and the underlying original soil (Miller et al., 2008). The black interface layer is a compact mixture of manure and mineral soil and is thought to be responsible for restricting infiltration and leaching (Mielke et al., 1974). One location in the bedding area and one in the non-bedding area were randomly selected in each of the 12 pens for simulated rainfall runoff