Impact on Antibiotic Resistance by a Solid-Liquid Separator - Rotary Drum Composter Manure Treatment System on a New York State Dairy Farm

A Case Study

Lauren Ray and Curt A. Gooch

Cornell PRO-DAIRY Dairy Environmental Systems Program

November 2020

Summary

An initial assessment of an on-farm manure treatment system consisting of a screw press solid-liquid separator (SLS) continuously feeding the separated solids to a rotary drum (RD) composting unit was conducted to identify possible impacts on antibiotic resistance (AR) mitigation. The methods and results of the study are published in *On-farm screw press/ rotary drum treatment of dairy manure associated antibiotic residues and resistance* (2020)[1] and this case study provides a summary. The commercial dairy farm located in New York State had about 650 lactating dairy cows and associated replacements. The SLS-RD manure treatment system analyzed was designed to harvest and treat separated manure solids (SS) for use as stall bedding and to remove coarse solids prior to separated liquid (SL) storage in a covered long-term storage. Like all on-farm manure treatment systems used today, the system analyzed was not designed to mitigate antibiotic residues and antibiotic resistance genes (ARGs). Little is known about existing manure treatment systems and their potential impact on AR and this case study provides one example of an on-farm SLS-RD system.

Two of the four antibiotics administered to cows on the study farm, oxytetracycline (OTC) and sulfadimethoxine (SDM), were detected in the excreted manure, along with all four of the ARGs tested. Only manures most likely to contain antibiotic residues from the hospital barn, dry cow barn, and select heifer pens were collected as the influent for the SLS-RD system during the study. The SLS-RD impacts on levels of antibiotic residues and ARGs were compared based on initial and final masses of each constituent, estimated from concentration measurements and mass flow calculations. While most of the ARGs partitioned with the SL, eight times more *tet*O (the ARG associated with tetracycline resistance) partitioned with the SS. In addition, all ARGs and OTC residues in the SS were significantly reduced after the RD unit. Due to the various factors that influence detection of antibiotic residues and ARGs, different results at other farms are expected. For example, the case study farm had lower than typical levels of antibiotic treatment.

Introduction

Antibiotic resistance (AR) is a naturally occurring process where previously susceptible bacteria develop the ability to resist the effects of an antibiotic. AR is gained by the bacteria and not by the host (cow or human). Higher incidence of AR can increase the cost and the effectiveness of treating and preventing bacterial infections in humans, animals, and plants. Antibiotic-resistant bacteria (ARB) are bacteria that can resist the effects of an antibiotic, and antibiotic resistance genes (ARGs) are what give the bacteria this ability. [2,3]

For more information on antibiotics and their use in the dairy industry, see the Antibiotic Resistance and Dairy Production fact sheet. [2]

For more information on antibiotic resistance, ARB and ARG, and the importance to dairy producers, see the Antibiotic Resistance and Dairy Production frequently asked questions resource. [3]

Even with improved herd management, diagnostics, housing, and antibiotic administration, antibiotic residues, ARB, and ARG have been detected in dairy cattle manure. The potential impact that different dairy manure treatment technologies have on mitigating AR is not understood. Naturally occurring ARGs are already present in the environment and more research is needed to determine the full implications of AR when using SS as stall bedding and recycling manure and SL manure to cropland.

This study was conducted on a New York State dairy farm with approximately 650 lactating cows. Blended manures collected from the farm hospital barn, dry cow barn, and select heifer pens were used as system influent. Regular sampling occurred over a two-day period at certain points throughout the manure treatment system. The results provide one example of a field-scale, on-farm assessment of a manure treatment system with primary treatment by a screw press SLS and secondary treatment of SS by a continuously fed in-vessel rotary drum (RD) style composting unit, and the impact on antibiotic residuals and ARGs.

Antibiotics Administered

The antibiotics administered within a two-week period leading up to the study are listed in Table 1 (carrier not included); the relative percentages of each antibiotic shown in Figure 1.

Table 1. Active ingredient mass of antibiotics administered in the two weeks prior to the study.

Antibiotic Drug Class	Antibiotic Name	Active Ingredient – Total Mass Administered (mg)
β-lactam	Ampicillin	125,000
β-lactam	Ceftiofur crystalline-free acid	5,000
β-lactam	Ceftiofur hydrochloride	69,125
Macrolide	Tulathromycin	1,100
Sulfonamide	Sulfadimethoxine (SDM)	315,336
Tetracycline	Oxytetracycline (OTC)	16,500

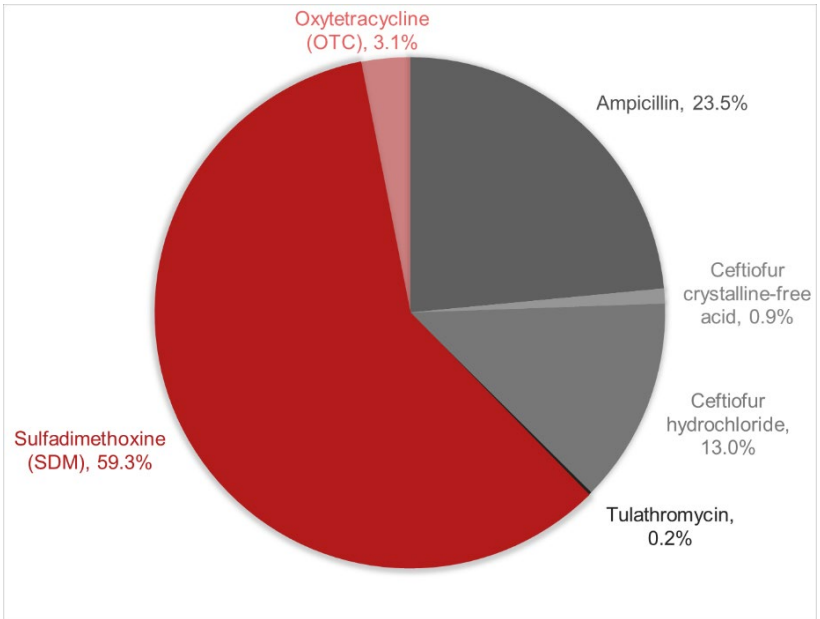

Figure 1. Percentage of active ingredient mass of each antibiotic administered during the two weeks prior to the study. Carrier not included.

Manure Treatment System Analyzed

A schematic of the SLS-RD system analyzed is shown in Figure 2. A propeller agitator semi-continuously mixed raw influent in a 27,740-gallon pit prior to and as it was pumped to the screw press SLS outfitted with a 0.5 mm screen opening. The SLS was located above the RD vessel (35.4 ft. long by 6.2 ft. diameter) that is designed for bedding recovery (Bedding Recovery Unit, FAN Separator GmbH, Marktschorgast, Germany). SS fall into the RD that has a retention time of approximately 24 hours. During the study, the RD unit operating temperature averaged 120°F at the inlet and 162°F near the outlet based on manufacturer-installed and calibrated monitoring equipment.

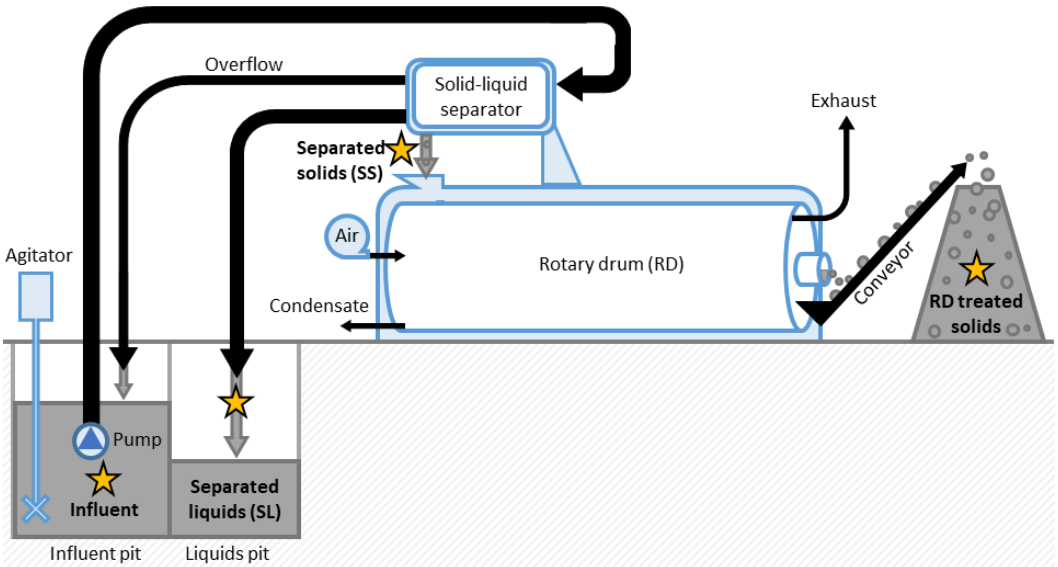

Figure 2. Schematic of SLS and RD composting system (blue), plumbing and conveyors (black arrows), and material flows (grey). Yellow stars indicate sample locations.[1]

Samples from four sample locations on the SLS-RD manure treatment system (indicated in Figure 2 with yellow stars) were analyzed for nine sulfonamides, seven tetracyclines, and nine macrolide antibiotic residues using liquid chromatography with a tandem mass spectrometry. β-lactam residues were not analyzed based on previous studies that showed non-detection in manure, attributed to their instability in the environment and rapid degradation to penicilloic acid.

The four ARGs listed below were also analyzed in the samples collected over the two-day sampling period. Total amounts of antibiotic residues and ARGs were calculated from the measured concentrations and mass flows. For additional information on methods of measuring antibiotic residues, ARB, and ARG, see the fact sheet series on Antibiotic Residues in Dairy Manure and Antimicrobial Resistance and Dairy Manure. [4]

- *sul1*: associated with sulfonamide resistance
- bla_{OXA-1}: associated with β-lactam resistance
- *intI1*: associated with gene transfer
- *tet*O: associated with tetracycline resistance

Results

Impact on Antibiotic Residues

Only OTC and SDM antibiotic residues were positively detected among the 25 antibiotics analyzed, and the average total mass of each at the sample locations is reported in Table 2. The calculated mass of both OTC and SDM decreased after the SLS, but only the OTC mass reduction was significant at 39%. Approximately 80% of the OTC and over 90% of the SDM partitioned with the SL after SLS. Of the OTC and SDM mass detected in the SS, a 50% reduction of OTC was estimated after the RD composter while the SDM mass increased by approximately 20%.

Table 2. Average total mass ± standard deviation of OTC and SDM residues at the four sample locations on the SLS-RD manure treatment system (n = 15).

Sample location	Antibiotic residue average total mass (mg)	
	OTC	SDM
Influent	14,099 ± 1,674	24,778 ± 5,671
Separated liquid	6,735 ± 2,484	18,486 ± 6,147
Separated solids	1,915 ± 1,006	1,409 ± 226
RD treated solids	1,049 ± 132	1,804 ± 113

Measured Impact on Antibiotic Resistance Genes

All four ARGs analyzed were detected at each sample location, and the calculated total ARG copies are shown in Table 3. The fate of *tet*O was unique with approximately 8 times more partitioning into the SS than into the SL. In contrast, 70% or more of the *sul1*, bla_{OXA-1}, and *intI1* gene copies partitioned into the SL. While no significant effect on the amounts of these

three gene copies after the SLS were observed, there was a small, measurable reduction in *tet*O. Each of the ARGs were significantly reduced after the RD process with measured reductions of 98% in *tet*O ($p < 0.001$), 75% in *sul1* ($p < 0.001$), 87% in bla_{OXA-1} ($p < 0.001$), and 60% in *intI1* ($p = 0.01$).

Table 3. Average total number of ARG copies ± standard deviation at the four sample locations on the SLS-RD manure treatment system, normalized to 16S rRNA gene copies (n = 12).

Sample location	*tet*O	*sul1*	bla_{OXA-1}	*intI1*
Influent	247,000 ± 150,000	49,500 ± 3,680	12,000 ± 2,640	41,000 ± 10,300
Separated liquids	12,100 ± 14,700	41,000 ± 9,280	12,800 ± 6,240	31,100 ± 7,310
Separated solids	94,700 ± 28,400	10,700 ± 1,840	2,050 ± 752	13,000 ± 4,770
RD treated solids	1,620 ± 1,500	2,640 ± 600	256 ± 215	5,180 ± 2,560

Key Takeaways

- This case study provides one example of the impact of both screw press SLS of dairy manure and RD composting treatment of SS on the antibiotic residues and ARGs measured in the manure.
- The estimated mass of OTC residues was significantly reduced (by 39%) after the SLS, but not the SDM residue, *sul1*, bla_{OXA-1}, or *intI1*. The latter 3 ARGs partitioned with the SL, while *tet*O primarily partitioned with the SS and was minimally reduced after SLS.
- All 4 ARGs and OTC residues detected in the SS were found to be significantly reduced after the RD unit, with the estimated mass of the OTC residues reduced by 50%.

Authors

Lauren Ray Email: ler25@cornell.edu
Curt Gooch Email: cag26@cornell.edu

[1] Oliver, J.P., J.J. Hurst, C.A. Gooch, A. Stappenbeck, L. Sassoubre, and D.S. Aga. 2020. *On-farm screw-press/rotary drum treatment of dairy manure associated antibiotic residues and resistance.* J. Environ. Qual. https://doi.org/10.1002/jeq2.20161.
[2] PRO-DAIRY AR and Dairy Production – An Overview. https://prodairy.cals.cornell.edu/sites/prodairy.cals.cornell.edu/files/shared/documents/DES_AMR_Overview.pdf.
[3] PRO-DAIRY AR and Dairy Production – FAQs. https://prodairy.cals.cornell.edu/sites/prodairy.cals.cornell.edu/files/shared/documents/DES_FAQ-AMR%26Dairy.pdf.
[4] PRO-DAIRY DES Fact Sheet Series': https://prodairy.cals.cornell.edu/environmental-systems-mgt/environmental-considerations/amr/.

This material is based upon work that is supported by the National Institute of Food and Agriculture, U.S. Department of Agriculture, under award number 2016-68003-24601. Any opinions, findings, conclusions, or recommendations expressed in this publication are those of the author(s) and do not necessarily reflect the view of the U.S. Department of Agriculture.