

# Investigation of the relationship between manure processing method and presence of *M. avium* subsp. paratuberculosis and *Salmonella* spp. in recycled manure solids bedding on Midwest dairy farms

F. Peña Mosca,<sup>1</sup> DVM, MS; S. M. Godden,<sup>1</sup> DVM, DVSc; E. Royster,<sup>1</sup> DVM, MS; D. Albrecht,<sup>1</sup> BS; S. Wells,<sup>1</sup> DVM, PhD; B. Crooker,<sup>2</sup> PhD; N. Aulik,<sup>3</sup> PhD

<sup>1</sup>Department of Veterinary Population Medicine, University of Minnesota, St. Paul, MN 55108

<sup>2</sup>Department of Animal Science, University of Minnesota, St. Paul, MN 55108

<sup>3</sup>Wisconsin Veterinary Diagnostic Laboratory, University of Wisconsin, Madison, WI 53706

## Introduction

While many dairies use green (GRN) recycled manure solids (RMS) bedding, some first process slurry through an anaerobic digester (DIG), while others have adopted secondary (SEC) processing methods such as mechanical composters (COM), hot air dryers (DRY), or, more recently, infrared drying (IR), in an effort to lower mastitis pathogen counts in ready to use (RTU) solids. However, these processing methods could also potentially reduce levels of other important pathogens in RTU solids. Our objective was to investigate the relationship between use of DIG and other SEC processing methods on *M. avium* subsp. paratuberculosis (MAP) and *Salmonella* spp. (SAL) in RTU RMS on Midwest farms.

## Materials and methods

Twenty-seven dairy premises in MN and WI were recruited to achieve a sample of different processing methods including GRN (n = 6), COM (n = 3), DIG (n = 9), DIG-DRY (n = 6), DIG-IR (n = 1), and DRY (n = 2). Premises were visited once in summer 2021 to collect slurry and bedding samples before and after each processing step within the system (e.g. raw or post-digested slurry, post-pressed solids and post-COM, -DRY- or -IR RTU solids). Slurry and solids samples were submitted to the Wisconsin Veterinary Diagnostic Lab to test for the presence of MAP (liquid culture with PCR confirmation) and *Salmonella* spp. (culture). Farms were categorized into one of 4 system types: GRN (n = 6), DIG only (n = 9), SEC only (n = 5) or DIG+SEC (n = 7). Logistic regression was used to compare the odds for a positive MAP or SAL test in initial raw slurry samples versus in final RTU RMS samples within each of the 4 system categories. Logistic regression was then used to describe the odds of a positive MAP or SAL test before and after each distinct processing step within the system. Models offered to control for breed and herd size, but they were not retained in final models. Multiple comparisons were accounted for by using Tukey adjustment.

## Results

For MAP testing, 80%, 67%, 60%, and 57% of initial raw slurry samples, and 40%, 0%, 20% and 0% of finished RTU solids samples were MAP-positive, for GRN, DIG, SEC, or DIG+SEC systems, respectively. When evaluating individual processing steps within a system, the odds (95% CI) for a positive MAP test in a post- (vs pre-processed) sample varied in magnitude and significance depending on the step evaluated (e.g. pre- vs post-digested slurry, 0.09(0.01-0.52),  $P = 0.007$ ; ready-to-press slurry vs post-pressed solids, 0.32(0.09-1.12),  $P = 0.075$ ; post-pressed solids vs RTU solids after SEC processing, 0.45(0.03-5.84),  $P = 0.54$ ). It was notable that MAP was recovered in 2 post-DIG slurry samples, suggesting that DIG alone cannot be counted upon to completely eliminate MAP.

For SAL testing, 100%, 89%, 100% and 86% of initial raw slurry samples, and 80%, 33%, 20% and 0% of finished RTU solids samples were SAL-positive, for GRN, DIG, SEC or DIG+SEC systems, respectively. When evaluating individual steps within a system, the odds (95% CI) for a positive SAL test in a post- (vs pre-processed) sample varied in magnitude and significance depending on the step evaluated (e.g. pre- vs post-digested slurry, 0.04 (0.004-0.39),  $P = 0.005$ ; ready-to-press slurry vs post-pressed solids, 1.38 (0.45-4.25),  $P = 0.56$ ; post-pressed solids vs RTU solids after SEC processing, 0.08(0.01-0.90),  $P = 0.04$ ).

## Significance

A high proportion of raw slurry and GRN RTU RMS samples were positive for MAP and SAL. Despite small sample sizes for some systems, results show that either DIG or SEC processing (e.g. DRY or COM) can result in a substantial numerical, if not statistical, reduction in risk for a positive MAP or SAL test. However, DIG alone cannot be counted upon to eliminate either MAP or SAL. No MAP or SAL was detected in RTU solids when a combination of DIG plus SEC processing was used. However, the latter results should be interpreted with caution given the very small sample size. Larger studies are needed to more extensively evaluate the biological and economic impacts for all of these RMS processing systems, and the newer IR systems in particular.

