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Evaluation of microbial community composition of dairy cows manure and soil before and after its fertilization.

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Abstract

Microbiota is the entire collection of microorganisms in a specific environment and microbiome is all the genetic material within it. This is a complex system influenced by variations in its ecosystem itself, i.e. soil and gut. Bovine manure is commonly used for the fertilization of agricultural soils and it comprises the intestinal microbiota influenced both by feed and possible residues of drugs used for therapy. The excessive use of antibiotics in animal husbandry and subsequent land application of animal wastes may introduce massive quantities of antimicrobial drugs (AMD) and resistant bacteria into the soil environment. A research project has been granted by the Italian Ministry of School Education, University and Research (PRIN 2015KA3XFH) to evaluate the role of intensive animal farming as potential source of environmental antimicrobial contamination and resistance. To this aim samples of manure and soil (before and 30 days after fertilization) were collected from intensive dairy cow farms located in Veneto and Lombardia regions, where intensive animal farming is widespread. All samples were screened for detecting and quantifying the commonly used AMDs (i.e. beta-lactams, fluoroquinolones, polymyxins and macrolides) by HPLC-MS methods. The microbiome of the samples was determined by employing a culture independent approach based on metagenomics and NGS-sequencing. All samples were negative for AMDs selected. The manure microbiome was homogenous among the different farms and regions, but very different from the soil one. In manure a higher presence of Porphyromonadaceae, Lachnospiraceae, Clostridiaceae, Pseudomonadaceae and Ruminococcaceae families was observed, whereas in soil the Chitinophagaceae family was the most represented. After fertilization, the microbiome composition was not modified in soil, however the absence of drugs concentration in manure may have had a role in this process. The research project will be completed by assessing the prevalence and the diversity of antimicrobial resistance genes to multiple antimicrobial classes by quantitative real time PCR. This is one of the first studies that evaluates the modifications in the microbial communities of manure and soil before and after fertilization. Thanks to the metagenomics analysis, this research project may elucidate the role of intensive animal farming in the diffusion of antimicrobial resistance in the environment.