Bacterial topography of the bovine respiratory tract: analysis of upper respiratory tract microbiotas as the source of the lung microbiota in healthy cattle

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Introduction

It is generally accepted that the nasopharynx is the primary source of bovine respiratory disease (BRD) pathogens for the lower respiratory tract (LRT) of cattle. Indeed, it has long been held that the nasopharynx plays a causative role in the development of BRD. However, it has been shown that there is poor agreement between the microbiota of the nasopharynx and the LRT in cattle. Conflicting results from recent studies suggest that BRD pathogens in the nasopharynx may not always significantly differ between healthy cattle and those diagnosed with BRD. These observations make sense in light of recent research in human medicine, which suggests that bacterial pathogens that cause lung infections don't originate from the nasopharynx and that the nasopharyngeal microbiota contribute very little to composition of lung microbiota. There was surprisingly more similarity between the mouth/oropharyngeal microbiotas and the lung microbiota. In the pursuit of finding alternative strategies for controlling BRD at the feedlot such as probiotics or local antimicrobials, it is crucial to understand which upper respiratory tract (URT) microbiotas contribute the most to the LRT microbiotas so that we can modulate the correct bacterial communities. Therefore, the objective of this study is to map the bacterial microbiotas present along the entire beef cattle respiratory tract and determine the relative contributions of these communities to the microbiota of the lung.

Materials and Methods

A total of 18 healthy feedlot steers were enrolled to the study at a "natural" feedlot located near Strathmore, Alberta. Health status was assessed by veterinary clinical examination, including a visual assessment of each animal for signs associated with clinical BRD, rectal temperature measurement and lung ultrasonography. Animals had not received antibiotics at any point in their lives prior to sampling. For each animal, the following respiratory tract locations were sampled: nasal cavity (left and right nostril), nasopharynx (left and right), bottom of the mouth, hard palate, oropharynx, tonsils (left and right), trachea (proximal and distal), right cranial bronchus (proximal and distal), left caudal bronchus (proximal and distal), and right caudal bronchus (proximal and distal). Total DNA was extracted from all samples and sent for 16S rRNA metagenomic sequencing. Sequencing data will be processed using DADA2 to infer exact sequence variants, and the resulting variants will be used to characterize the different microbiota and determine the relative contributions of these communities to the microbiota of the lung. This will be accomplished using a variety of different statistical approaches, including comparisons of relative pathogen abundances, PERMANOVAs to compare community structure, and redundancy analyses to visualize how the microbiota at each sampling site relate to each other.

Results

Results are pending; sequencing data will be available for analysis before the end of May 2019. We hypothesize that alpha diversity will decrease from the URT to the LRT, with the lowest diversity observed in distal bronchi. Bacterial populations colonizing the lung will likely differ from the URT, with nevertheless numerous genera present at both the URT and the LRT.

Significance

This study will map for the first time the bacterial microbiotas present along the entire respiratory tract of beef cattle and determine the relative contributions of URT communities to the microbiota of the lung. Ultimately, the results of this study will hopefully provide a better idea of which URT microbiotas should be the primary focus of future microbiota research in beef cattle and the primary target for microbiota modulation in the effort to find alternatives to antimicrobials.