Deep sequencing metagenomic analysis of experimentally induced bovine digital dermatitis lesions

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Introduction

Bovine digital dermatitis (DD) is a leading cause of lameness in both dairy and beef cattle in the US and much of the world. Previous work by our laboratory and others has demonstrated that metagenomic analysis of naturally occurring digital dermatitis biopsies yields evidence of a complex polymicrobial disease process. We have demonstrated that lesions develop through a systematic progression of morphologic changes that are each associated with significantly different microbial communities. While these studies have provided insights into the pathogenesis of the disease process, they are often complicated by confounding associated with temporal differences in the time or season of biopsy collection, chronicity of the lesion, and the lack of baseline metagenomic data for each foot prior to lesion development. The goal of this study was to utilize a recently developed and highly reliable experimental lesion induction model of digital dermatitis to collect a large number of metagenomic samples with minimal confounding.

Materials and Methods

For the study, 36 Holstein steer calves with known DD-naïve status were utilized. Skin biopsies were obtained from each of the calves prior to induction of DD. Digital dermatitis was induced using macerated digital dermatitis lesions obtained from naturally infected dairy cattle. Following experimental induction, the feet of each calf were again biopsied for histopathology and 16S metagenomic analysis. In total, 298 biopsies were multiplexed onto a single lane of Illumina MiSeq.

Results

The sequencing run yielded 15,104,019 reads, of which 13,685,124 passed initial quality filtering and had barcodes that matched 1 of the 298 samples. Nine samples were excluded due to low sequencing depth, resulting in 289 samples analyzed with a mean depth of 37,630 sequences. Evaluation of the phylogenomics of the induced lesions compared to the naturally occurring lesions demonstrated significant overlap and grouping of the lesions by principal component analysis.

Significance

Data analysis revealed similar changes in bacterial microbiota to those previously observed in 16S-based sequencing of naturally occurring DD lesions and provides additional insights into the pathogenesis and etiology of bovine digital dermatitis. These results are significant for 2 reasons. First, the induced lesions provide a much more refined metagomic profile and assist with determining a core microbiome for digital dermatitis. Second, these results validate that the induced lesions mirror the bacterial composition of naturally occurring digital dermatitis. These results provide a strong rationale for use of the induction model to study the disease pathogenesis and treatment. These results will be of interest to commercial companies interested in testing the efficacy of therapy against DD.