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Genetic diversity of Treponemes in dairy herds and their surrounding environment

Martin W. Nielsen, Tim K. Jensen, Hans-Christian Ingerslev, Kirstine Klitgaard

Introduction
The causative agents of the highly contagious inflammatory skin disease digital dermatitis (DD), which has reached endemic levels in parts of the world, have been identified as mainly governed by Treponema species. Of the 20 different phenotypes identified from DD biopsies, Treponema phagedenis-like, T. medium/T. vincentii-like and T. denticola/T. pedis-like phenotypes appear to be highly associated with the disease progression [1,2]. Specific niches of infection reservoirs and transmission routes, however, still remains to be clarified in order to target and control the spread of the infection.

To address the issue of infection reservoirs of DD, we surveyed for evidence of treponemes in the content of the bovine gastrointestinal tract (feces), in DD lesions, and in the dairy farm environment (slurry).

Conclusion
We surveyed for the presence of DD Treponema in environmental samples and individual scrapings from (DD- and mammary) lesions. We were not able to identify DD Treponema in the environmental samples (field, slurry and individual feces samples).

The phylogenetic compositions of the environmental samples were remarkably alike and significantly different from the DD-associated Treponema, figure 2.

The DD lesions and the ulcerative mammary dermatitis sample proved to be much more diverse and dominated by species previously identified in DD biopsies [1,2].

The question remains if this is a sensitivity issue – or if the reservoir of these bacteria is actually to be found elsewhere in the dairy herd environment.

Methods
Samples from potential reservoirs were collected and subsequently DNA was purified by QIAamp®DNA stool mini kit. The hyper-variable regions V3 and V4 of 16S rRNA were PCR amplified as described in [1], and subsequently sequenced on a MiSeq, Illumina. Sequences were analyzed by BION-meta (https://app.box.com/BION)

Figure 1. Healthy and digital dermatitis hooves
Severe lesions are easily identified (b,c) in comparison to a healthy hoof (a). Typical DD lesions characterized by irregular, raw raised areas that often develop keratinaceous hairlike projections (b, c).

Figure 2. Phylogenetic composition within the samples
(a) Phylogenetic composition of individual environmental and lesion samples, * denotes the ulcerative mammary dermatitis. (b) Representation of the average phylogenetic contribution in each sample category. Data represent 34 samples of feces (field, slurry and individual cows) and 9 scrapings (8 DD lesions and 1 ulcerative mammary lesion)*