Impact of pastures composition on calves microbiota in saliva, rumen and feces

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Rearing dairy male calves on underused pasture offers the opportunity to solve economic, sustainable and welfare associated tradeoffs. This study deals with assessing how different pasture compositions affect the calves microbiota across time. Understanding the influence on gastrointestinal settlement and development helps to understand ruminal fermentation efficiency. Three contrasting pasture compositions and one indoor system were compared by the effects on bacterial community composition. Buccal swab and feces sampling allowed repeated sampling over calves age (three- to six-month-old). Additionally, rumen fluid was collected once at the end of the experiment via esophageal probe. It is to be assumed that increase of composition and diversity of the pasture lead to higher microbial diversity. Male calves (n=72) were raised on three different composed pastures (intensive, permanent, alpine) and one hay based indoor system in Switzerland. Eighteen animals were examined per feeding system, every six were of the same genotype (Brown Swiss, Swiss Fleckvieh, Crossbreed = Limousin x Brown Swiss). Pasture composition was described by grass type and alongsides like herbs (%). Bacterial communities of buccal swabs (bioinformatically separated in oral- and rumen associated communities), rumen fluid, and feces were characterized by 16S rRNA amplicon sequencing, processed in QIIME 2 (DADA2), and taxonomically assigned using GTDB (v226). Diversity metrics (Shannon index and Bray–Curtis dissimilarities) and statistical tests (Kruskal–Wallis, PERMANOVA) were calculated in R (vegan package). Buccal swabs enabled us to collect rumen-associated samples at three time points on all locations. This challenging experimental design wouldn't allow an intensive sampling via the esophageal probe. In total 3,427 ASVs were revealed from buccal swab samples and 3,937 ASVs from rumen fluid at the end of the trial, with 1,726 ASVs overlapping in both sample sets and defined as rumen-associated ASVs in buccal swab samples. Thus, an impressive recovery of rumen bacteria could be shown using buccal swabs. As this method is less invasive and seems to be a robust method for future ruminant experiments. Microbial diversity analysis revealed the highest alpha diversity (Shannon) on almost all samples from animals at alpine pastures, followed by the permanent pasture. Beta diversity analysis showed a clustering of the indoor samples with intensive, and permanent with alpine. Changes according to the age of animals were minor while showing only no or minor significance for the diversity structure. Calves grazing alpine and permanent pastures developed richer, more diverse microbiota compared to indoor or intensive systems. In conclusion, our results support the hypothesis that greater diversity in pasture composition is associated with higher diversity and complexity of microbial communities in calves. In addition, we assume that buccal swab sampling provides a good approximation on rumen fluid microbial community.