

Comparison of Rumen Microbial Analysis Pipelines Based on 16s rRNA Gene Sequencing

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Abstract : To investigate complex rumen microbial communities, 16S ribosomal RNA (rRNA) sequencing is widely used. Here, we evaluated the impact of bioinformatics pipelines on the observation of OTUs and taxonomic classification of 750 cattle rumen microbial samples by comparing three commonly used pipelines (LotuS, UPARSE, and QIIME) with Usearch. In LotuS-based analyses, 189 archaeal and 3894 bacterial OTUs were observed. The observed OTUs for the Usearch analysis were significantly larger than the LotuS results. We discovered 1495 OTUs for archaea and 92665 OTUs for bacteria using Usearch analysis. In addition, taxonomic assignments were made for the rumen microbial samples. All pipelines had consistent taxonomic annotations from the phylum to the genus level. A difference in relative abundance was calculated for all microbial levels, including Bacteroidetes (QIIME: 72.2%, Usearch: 74.09%), Firmicutes (QIIME: 18.3%, Usearch: 20.20%) for the bacterial phylum, Methanobacteriales (QIIME: 64.2%, Usearch: 45.7%) for the archaeal class, Methanobacteriaceae (QIIME: 35%, Usearch: 45.7%) and Methanomassiliococcaceae (QIIME: 35%, Usearch: 31.13%) for archaeal family. However, the most prevalent archaeal class varied between these two annotation pipelines. The Thermoplasmata was the top class according to the QIIME annotation, whereas Methanobacteria was the top class according to Usearch.

Keywords : cattle rumen, rumen microbial, 16S rRNA gene sequencing, bioinformatics pipeline

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