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**FECAL MICROBIOTA COMPOSITION OF KRSKOPOLJE PIG BREED RAISED IN TWO DIFFERENT PRODUCTION SYSTEMS**B. Papic<sup>1</sup>, J. Plut<sup>2</sup>, M. Stukelj<sup>2</sup><sup>1</sup>*Institute of Microbiology and Parasitology, Veterinary Faculty, University of Ljubljana, Ljubljana, Slovenia*<sup>2</sup>*Clinic for Reproduction and Large Animals, Clinic for Ruminants and Pigs, Veterinary faculty, University of Ljubljana, Ljubljana, Slovenia***Background and Objectives**

Krskopolje breed is the only preserved Slovenian autochthonous pig breed known for its robustness and meat quality. The aim of this study was to evaluate the fecal microbiome of Krskopolje breed, which has not yet been investigated.

**Material and Methods**

A total of 18 pigs kept at three different locations and fed organic commercial feed were included into the study: (i) pigs kept on pasture together with small ruminants (group L1), (ii) pigs kept indoors and without other farm animals and fed only commercial feed (group L2), and (iii) pigs kept on pasture with cattle (group L3). Fecal samples were collected rectally in all four seasons of 2022, from grower to finisher phase. Groups L2 and L3 were raised on the same organic farm and maintained by the same livestock workers, whereas group L3 was reared on a different organic farm and were maintained by other livestock workers. A total of 72 DNA samples underwent 16S rRNA gene (V3–V4 region) sequencing using Illumina paired-end (2x250 bp) technology. Operational taxonomic unit (OTU)-based microbiome analysis was performed using QIIME 1.9.1.

**Results**

Alpha diversity was comparable between study groups; the average number of observed OTUs was 1746. The two predominant bacterial phyla in all three groups were Firmicutes and Bacteroidota. Significant seasonal changes in microbiome composition were observed between all seasons (nonparametric MANOVA,  $p < 0.008$ ). The microbiome composition differed significantly between study groups L1 and L2–L3, as shown by principal coordinate analysis of weighted UniFrac distances and nonparametric MANOVA ( $p = 0.001$ ), whereas groups L2 and L3 did not differ significantly ( $p = 0.2$ ). Linear discriminant analysis Effect Size (LefSe) identified several bacterial taxa that were most differentially abundant between the different study groups and the different seasons.

**Discussion and Conclusion**

Pig microbiota composition is not stable over time and is influenced by external (rearing-related) factors. In this study, the production system or co-rearing with other animal species had no significant influence on the pig microbiome composition. Rather, the main observed differences in microbiota composition could be explained by the farm and livestock workers.