ANALYSIS OF THE UTERINE MICROBIOME IN COWS WITH RETENTIO SECUNDINARUM AND THE DYNAMICS OF ITS CHANGES DURING THERAPY

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Retentio secundinarum (RS) in cows is defined as the failure to expel the placenta 24 hours after calving. This complication is the cause of significant economic losses due to reproductive disorders including: uterine inflammation, prolonged postpartum downtime, reduced mating rates, prolonged inter-partum period and the development of secondary metabolic diseases. These losses add to the cost of treating the animals. The microorganisms most commonly found in RS cases were Escherichia coli, Fusobacterium necrophorum, Clostridium spp. and Trueperella pyogenes, the presence of which in these cases justified the domaic and general use of antibiotics, including third-generation cephalosporins. Their widespread use in the current social context is unjustified. Other antibiotics have been suggested for the treatment of RS. There is now a growing interest in other alternative treatments. Nowadays, precise genetic tools are available to assess the type of microorganisms. The aim of this study was to determine the uterine microbiome of cows with RS. Metagenomic analysis of the popuation of batteries and archaeons was carried out on the basis of the hypo-ergodic V3-V4 region of the S rRNA gene. Specific primer sequences 34IF and 785R (16S analysis) were used to amplify the selected region and prepare the library. PCR was performed using Q5 Hot Start High-Fidelity 2X Master Mix, according to the manufacturer's recommendations. Sequencing was performed on a MiSeq, paired-end (PE), 2×300 nt, using Illumina's v3 kit. Automatic pre-analysis of the data was performed on a Miseq sequencer using Miseq Reporter (MSR) v 2.6 software. The analysis consisted of two steps: (1) automatic demultiplexing of samples and (2) generation of fastq files containing

raw reads. Based on the evaluation of the preliminary results of the NGS sequencing analysis, a predominance of the bacteria Fusobacterium necrophorum, Coxiella burnetii, Esscherichia coli, Streprococcus pluranimalium and Clostridium perfringens was observed in the test specimens immediately after placenta detachment. It is planned to continue the study to confirm the composition of the uterine microbiome in a larger number of animals in order to develop a microbiome typical of RS cases and also to observe it after antibiotic and eubiotic therapy.

Key words: Retentio secundinarum, uterus, microbiome, cows