

ANALYSIS OF THE MICROBIOME OF THE HAMSTERS GASTROINTESTINAL TRACT DURING INFECTION WITH THE LIVER FLUKES *OPISTHORCHIS FELINEUS*, *OPISTHORCHIS VIVERRINI* AND *CLONORCHIS SINENSIS*

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INTRODUCTION

The liver flukes *Opisthorchis felineus*, *Opisthorchis viverrini* and *Clonorchis sinensis* are parasites of the mammalian hepatobiliary system. Their geographic areas practically do not overlap.

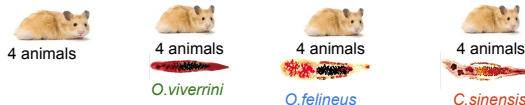
The species have different carcinogenic potential: *O.viverrini* and *C.sinensis* are recognized to be 1A group of biological carcinogens, whereas *O.felineus* belongs to 3A group.

Microbiota of the flukes might contribute to the difference in the carcinogenicity of the species.



METHODS

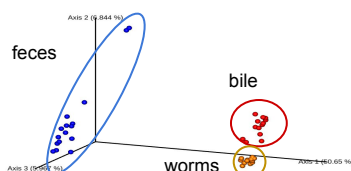
Golden hamsters from the Specific Pathogen Free Animal Facility (Institute of Cytology and Genetics, Novosibirsk, Russia) were used for study.



BILE, FECES and **WORMS** (V3-V4 region of the 16S rRNA gene)

MiSeq Illumina (2 × 300 bp), QIIME2, MEGA (v.10.2.5), R-studio (v.1.4.1106)

RESULTS



There is significant clustering of microbial communities by the source of DNA (the analysis of the main coordinates - the Bray-Curtis distance matrix).

The libraries contained 18,830,015 sequences:

- 427,955 reads per sample (median – 410,272)
- 1% unclassified sequences (maximum 4%) (specified in the Greengenes database)

Taxonomic composition:

A bacterial super-kingdom, 23 different phyla, 54 classes, 82 orders, 143 families, 261 genera.

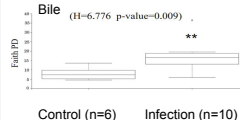
FECES

According to beta diversity analysis there are significant difference between *C. sinensis* and *O. felineus*, *C. sinensis* and *O. viverrini*, however, not between *O. felineus* and *O. viverrini*.

The fecal microbiome of the control animals significantly differed from the feces of animals infected with *C. sinensis* and *O. felineus*, but not from feces infected with *O. viverrini*.

Analysis of variance for samples divided by the type of worm showed a significant difference in 45 OTUs at the genus level.

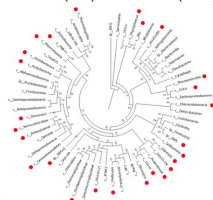
BILE



Analysis of the phylogenetic diversity (Faith PD) showed a significant change in the bile microbiome upon infection.

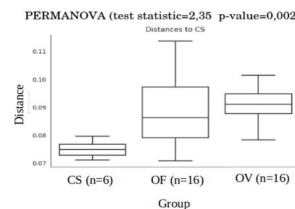
Analysis of variance for samples divided by the type of worm showed a significant difference in 30 OTUs at the genus level.

On the phylogenetic tree, the classes found only in worms and infected bile are marked in red.



WORMS

Permutation analysis of variance showed a difference between the microbiome communities of the three types of worms (a significant difference in 46 OTUs at the genus level).



Classes of bacteria that are unique to worms predominantly inhabit wastewater and activated sludge, fresh water and soil (Planctomycetia, Nitrospira, Gemmatimonadetes, etc.). There are also extremophiles (Epsilonproteobacteria, Deinococci).

OUTCOMES OF THE STUDY

- Beta diversity analysis (Bray-Curtis) shows significant clustering of microbial communities depending on the source of DNA (worms, feces and host bile);
- There are differences between microbiomes of feces during infection *C. sinensis* and *O. felineus*;
- Found bacteria that appear in bile during infection;
- Microbiomes of *Clonorchis sinensis*, *Opisthorchis felineus* and *Opisthorchis viverrini* differ at the class level
- Found unique for worms genus of bacteria (Salmonella, Ochrobactrum, etc.);

ACKNOWLEDGMENT:



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