

College of Veterinary Medicine, *Department of Infectious Diseases*

*Institute of Bioinformatics*

Athens, Georgia, March 17th, 2022

Dear Dr. Bingle,

Thank you for the constructive feedback on our manuscript entitled “Selection of software and database for metagenomics sequence analysis impacts the outcome of microbial profiling and pathogen detection”. We have amended the manuscript extensively according to the reviewer’s comments. Specifically, we

1) increased the number of software used for comparisons from 3 (Kraken2, CLARK, and CLARK-S) to 9 (BLASTN, Kraken2, Bracken, CLARK, CLARK-s, Centrifuge, Metaphlan3, Diamond, and Kaiju);

2) we added analyses on differences at the phylum and species-level microbial profiling;

3) we edited the manuscript sections to remove repetitive sentences and to better emphasize the conclusion of our study in the discussion section; and

4) we changed the manuscript title from “The selection of shotgun metagenomics software introduces biases in microbial profiling and pathogen detection” to “Selection of software and database for metagenomics sequence analysis impacts the outcome of microbial profiling and pathogen detection”.

We believe the changes and additions address all of the reviewers’ suggestions and the manuscript has greatly improved as a result of the review process.

Thank you again for taking the time to edit our manuscript.

Sincerely,

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