Deciphering the Fecal Microbiome of Indian Rhinoceros (*Rhinoceros unicornis*) by Metagenomic Approach

Probodh Borah^{1,*}, Rupam Dutta¹, Luit Moni Barkalita¹, Lukumoni Buragohain¹, Pankaj Deka², Samshul Ali³, Bhaskar Choudhury³, Panjit Basumatary³

> ¹Department of Animal Biotechnology, College of Veterinary Science, Assam Agricultural University, Khanapara-22, Assam, India
> ²Department of Veterinary Microbiology, College of Veterinary Science, Assam Agricultural University, Khanapara-22, Assam, India
> ³Center for Wildlife Rehabilitation and Conservation, Wildlife Trust of India, Borjuri, KNP, Assam, India
> *Corresponding Author's E-mail: borahp@vetbifg.ac.in

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ABSTRACT

With the advent of high throughput next generation sequencing (NGS) technology, it has become possible to study the diversity in microbial community of the gut with extraordinary resolution and accuracy. A number of sophisticated bioinformatics tools have also been developed to analyze the enormous data generated by such study. The greater one-horned rhinoceros (*Rhinoceros unicornis*) is native to the Indian subcontinent and majority of its population is found in Assam. The rhinoceros is one of the largest mammalian herbivores and has the ability to utilize fibrous plant matter through microbial fermentation in the hindgut. So far, there has been no report relating to study of the gut microbiota of the one-horned rhinoceros using metagenomic approach. In this study, we extracted genomic DNA from fecal sample of one-horned rhinoceros using commercially available kit as per the manufacturer's protocol. The QC passed DNA sample was used for amplicon generation, targeting V3 -V4 region of 16S rRNA genes. Library was constructed using Nextera XT index kit as per manufacturer's protocol. Further, to obtain an unbiased measure of bacterial diversity in the gut by metagenomic approach, we sequenced the nucleic acid by IllumiaMiseq 2x300 chemistry. Altogether 418,890 sequence reads were generated to characterize the gut microbiome of rhinoceros. The results showed that Firmicutes, Verrucomicrobia, Proteobacteria and Bacteroidetes were the predominant phyla occupying 74.87%, 14.83%, 6.86% and 2.29%, respectively of the microflora. A large proportion was found to comprise with unclassified bacteria. At the genus level, 4.71% Ruminococcus bacteria were detected which is an important member of the microbial community in the hindgut of non-ruminant herbivores, which enabled the host to gain nutrients from fibrous plant materials. The present work provides a framework for understanding the complex microbial community of the one-horned rhinoceros; however, further studies are required to link the distinctive microbiota with their digestive role in the hindgut of the one-horned rhinoceros.

Key words: Metagenomics; Microbiome; Next generation sequencing; Rhinoceros unicornis.