Insights into Archaeological Human Sample Microbiome Using 16S rRNA Gene Sequencing

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Abstract: Human body is inhabited by a vast number of microorganisms, collectively known as the human microbiome, and there is a tremendous interest in evolutionary changes in human microbial ecology, diversity and function. The field of paleomicrobiology, study of ancient human microbiome, is powered by modern techniques of Next Generation Sequencing (NGS), which allows extracting microbial genomic data directly from archaeological sample of interest. One of the major techniques is 16S rRNA gene sequencing, by which certain 16S rRNA gene hypervariable regions are being amplified and sequenced. However, some limitations of this method exist including the taxonomic precision and efficacy of different regions used. The aim of this study was to evaluate the phylogenetic sensitivity of different 16S rRNA gene hypervariable regions for microbiome studies in the archaeological samples. Towards this aim, archaeological bone samples and corresponding soil samples from each burial environment were collected in Medieval cemeteries in Latvia. The Ion 16S™ Metagenomics Kit targeting different 16S rRNA gene hypervariable regions was used for library construction (Ion Torrent technologies). Sequenced data were analysed by using appropriate bioinformatic techniques; alignment and taxonomic representation was done using Mothur program. Sequences of most abundant genus were further aligned to E. coli 16S rRNA gene reference sequence using MEGA7 in order to identify the hypervariable region of the segment of interest. Our results showed that different hypervariable regions had different discriminatory power depending on the groups of microbes, as well as the nature of samples. On the basis of our results, we suggest that wider range of primers used can provide more accurate recapitulation of microbial communities in archaeological samples. Acknowledgements. This work was supported by the ERAF grant Nr. 1.1.1.1/16/A/101.

Keywords: 16S rRNA gene, ancient human microbiome, archaeology, bioinformatics, genomics, microbiome, molecular biology, next-generation sequencing

Conference Title: ICBBN 2017: International Conference on Biomechatronics, Bioinformatics and Neuroengineering

Conference Location : New York, United States **Conference Dates :** October 05-06, 2017