Gut Mycobiome and Phytobiome Diversity and Composition in Coprolites from Huecoid and Saladoid Ancient Cultures

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The study of the forgotten fungal component of the gut microbiome may help us to understand the microbiota role in human biology and cultural factors. We are studying coprolites from Huecoid and Saladoid ancient cultures to characterize their mycobiome and phytobiome and assess their functional potential to infer their diet, health and lifestyles. Also, we compare their mycobiome with those of extant Amazonia. Ancient DNA extraction from coprolites followed by shotgun metagenomics sequencing with Illumina Miseq allows to detect plants and fungi diversity in the gut. Bioinformatic analyses have showed a higher Shannon index in Saladoids in comparison to Huecoids and Amazonia suggesting higher species richness in the Saladoid culture. Differences have been found in fungal communities and these results support the hypothesis of two different pre-Columbian cultures. We identified sequences with homology to edible mushrooms such as *Tuber spp., Agaricus spp.,* and Trametes spp. The former group were rare in Huecoids and abundant in Saladoids. Trametes species, which were slightly more frequent in Saladoids, have been used during years for medicinal purposes. Plant sequence alignment showed Ipomoea spp., Phaseolus spp., and Zea mays sequences suggesting food plants introduction from South America. Functional annotation revealed cellular functions including protein metabolism and stress response. This study allows an expanded vision concerning ancient human microbiomes, particularly pre-Columbian cultures that contribute to Puerto Rican history. DNA sequences from coprolites will also allow us to compare with extant gut microbiome and explore how it may have change over millennia.