Revealing the gut microbiome of orcas (*Orcinus orca*) as indicator of pollution impact

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Orcas (*Orcinus orca*) serve as valuable indicators of ocean health due to their sensitivity to environmental pollution. However, their susceptibility to pollutants poses significant health risks, pushing populations near industrialized regions toward extinction. This grave situation has heightened interest in understanding how pollution impacts the gut microbiome and overall health of orcas. Despite advanced sequencing technologies enabling high-throughput profiling of gut microbial communities in many terrestrial mammals, the orca gut microbiome has remained largely unexplored. This study represents a pioneering effort to explore the gut microbiome of orcas using a comprehensive whole metagenome sequencing approach. The main objectives were to characterize the diversity, composition, and functional gene profiles of the orca gut microbiome, with a particular focus on the diversity and prevalence of microbial genes involved in degradation of persistent organic pollutants (POPs), among both wild (stranded) and captive orcas. Gut content samples were collected from three stranded orcas in Belgium (2023), the Netherlands (2022), and Norway (2021), and from four captive orcas at Loro Parque, Spain (2024). Bacterial DNA was isolated for Nextera XT library preparation. Shotgun sequencing on the Illumina NovaSeq platform yielded 50 million reads per sample. Taxonomic classification was performed using Kraken2 and Bracken, while POP-degrading genes were annotated with Diamond and a custom gene database for microbial POP biodegradation. Results showed that the gut microbiome is dominated by four major bacterial phyla: Proteobacteria, Bacteroidota, Fusobacteriota and Firmicutes. Interestingly, we detected a set of bacterial dehalogenase and dioxygenase enzymes involved in essential degradation steps of POPs (such as PCBs, DDT, HCH and HCB) and facilitating removal of chlorine atoms and break-up of aromatic ring structures. This research provides foundational insights into the orca gut microbiome composition and demonstrates that microbiome functional profiles show potential as indicators for assessing pollutant impacts on cetaceans.

Keywords
Orca; Orcinus Orca; Gut Microbiome; Metagenomics; Pollution; POPs; Shotgun Sequencing; POP-degrading Genes; Pollutant-degrading Genes

PRE-DOC PRESENTATIONS