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Using metagenomics and metatranscriptomics to study specific bacterial species involved in biological phosphorus removal from wastewater

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**Introduction**
Enhanced Biological Phosphorous Removal (EBPR) from wastewater treatment plants is managed by Polyphosphate Accumulating Organisms (PAOs). The model PAO is “Candidatus Accumulibacter phosphatis” (Accumulibacter) and despite a large body of knowledge, their detailed physiology remains elusive.

In this study we combined metagenomics, metatranscriptomics and laboratory scale enrichment in order to examine the gene expression of the uncultured Accumulibacter and the co-enriched associated community.

**Methods**

**Enrichment**
Wastewater treatment plant → Sequencing Batch Reactor

**Genome recovery**
Samples → Metagenomes → Assembly → Genome

**Example**

**Transcriptomics**

**Results**

- **Combining metagenomics, metatranscriptomics and laboratory scale enrichments enables transcriptome studies of most individual species in the community.**

- For the first time we reveal the detailed transcriptome landscape of Accumulibacter during the anaerobic feast and aerobic famine conditions of the EBPR process.

- A new GAO (competitor to PAOs with a negative impact on EBPR) was discovered. Propionivibrio is closely related to Accumulibacter and hit by the current FISH probes used to define PAOs.

**Conclusions**

- Genome extraction from metagenomes. Differential coverage binning is used as the first step in extracting individual genomes from the metagenomes. Each point represents a scaffold, colored by essential single copy genes and scaled by scaffold length. The plot was made using the rmpplot function in the metagenome R package. Supprisingly a bacteria from the genus Propionis vibrio was enriched along with Accumulibacter.

- A GAO identified as a PAO. The putative GAO Propionivibrio (Propionis vibrio) is hit by the probes used to quantify the PAO Accumulibacter (white). However, it does not accumulate massive amounts of polyphosphate. Probes: PA0651 [FL05, Accumulibacter]; PA0462 (Cy3, both); PA0464 (Cy5, both); OI0004 (Cy3, Cy5, both).

- A GAO closely related to Accumulibacter and hit by the current FISH probes used to define PAOs.

- The species Accumulibacter anammoxidatus (white) is enriched during the anaerobic feast (15 min) and aerobic famine (180 min) conditions.