Genome-centric metagenomic investigation of 132 samples collected from biogas reactors revealed important functional roles for microbial species belonging to underexplored taxa.

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Anaerobic digestion

- Similarities in the biochemical process
- Complex microbiome and many uncultured
- Anaerobic digestion of organic matter is the largest biogenic source of methane on Earth
Anaerobic digestion

- Analyzing Anaerobic Digestion considering the microbial composition as a black box is like playing a chess match without knowing the pieces
Functional representation of the AD microbiome

Metagenome Assembled Genome (MAG)

Functional analysis

PG assigned to the functional role in the AD food chain

Gene expression profiles (metatranscriptomics)

Campanaro et al., Biotech for Biof 2016
Beta diversity of 132 samples revealed temperature and feedstock as main drivers of the microbial community composition.
Genomes reconstruction

- Importance of reconstructing genomes...
- a puzzling view of the subject is making difficult to clarify his identity...
Genomes reconstruction

• Once reconstructed the full picture...
• you can have a more clear view of the subject...
Genomic reconstruction

• This is why reconstruction of **Metagenome Assembled Genomes (MAGs)** can help to define microbiome composition and functional roles...
Taxonomy of 1,635 MAGs of the AD microbiome
MiGA database

- A dedicated MiGA database was developed for making all the information available to the scientific community
- were [http://microbial-genomes.org/projects/biogasmicrobiome](http://microbial-genomes.org/projects/biogasmicrobiome)
- taxonomy and genomic characteristics of each species
Relative abundance

- In most samples these taxa represent **2-3% of the AD microbiome**, but in specific samples they can represent **more than 10%**.
Metabolic reconstruction

Evaluation of their gene content suggests:

Ability to **synthesize nucleotides (not all the phyla)**;

Relevant number of glycoside hydrolases and glycoside transferases (particularly in *Candidatus* Dojkabacteria and *Candidatus* Magasanikbacteria) -> **degradation of some polymeric substances**

The presence in some species of the «phosphate acetyltransferase-acetate kinase pathway, involved in the conversion of acetyl-CoA to acetate» -> **production of acetate**

Absence of pathways involved in amino acids and lipids biosynthesis -> **proteins and lipids scavenging from the environment**

Presence of numerous proteases and transaminases -> **salvage amino acids from environmental sources**
Relative abundance of functional pathways in different reactors

- Relative abundance of MAGs associated to the encoded metabolic pathways allowed to estimate the relevance of different pathways in each sample:
  - (A) methanogenesis
  - (B) WL-pathway
  - (C) polysaccharides degradation.
New Results

The anaerobic digestion microbiome: a collection of 1600 metagenome-assembled genomes shows high species diversity related to methane production

Stefano Campanaro, Laura Treu, Luis M Rodríguez Rojas, Adam Kovalovszki, Ryan Ziels, Irena Maus, Xinyu Zhu, Panagiotis G Kougias, Arianna Basile, Gang Luo, Andreas Schlüter, Konstantinos T Konstantinidis, Irini Angelidaki

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A network of laboratories to study the AD system

To deal with the complexity of this metagenomic study we are forming a network of laboratories from Denmark, Greece, Spain, Germany, China, USA, Canada…

…and more collaborations are welcome!
Aknowledgments

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• Department of Environmental Science and Engineering, Fudan University: Gang Luo
CRIBI Biotechnology Center

- CRIBI is an interdepartmental Biotechnology Center located at Padova University
- 22 different research groups of 14 different departments
- 4 facilities dedicated to research
  - Next generation sequencing;
  - Proteomics
  - Microarrays, real-time PCR, digital PCR
  - Peptide synthesis

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Thank you!

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Genome size

MAGs genome size according to taxonomic assignment

The low completeness associated to MAGs belonging to some phyla suggests that essential gene sets has to be revisited