Use of Microbial Consortia for Conversion of Biomass Pyrolysis Liquids into Value-Added Products

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Outline

- Pyrolysis
- Bio-oil
- AD

} Py-AD

- Illumina Sequencing
- Mass Spectrometry
- Relatedness

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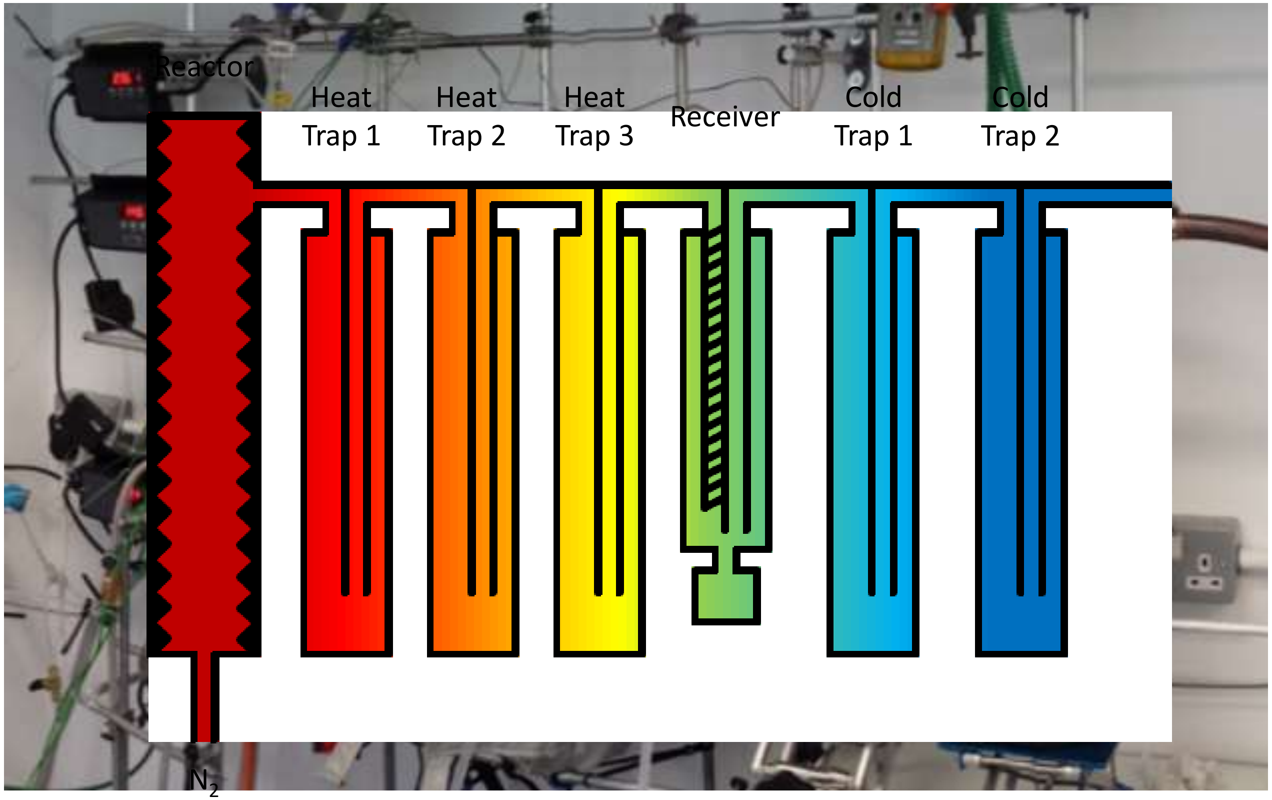
Pyrolysis



- Biomass burned at high temperatures (300°C - 600°C) in the absence of oxygen
- Thermal depolymerisation of lignocellulosic biomass
- Products are char, bio-oil & syngas

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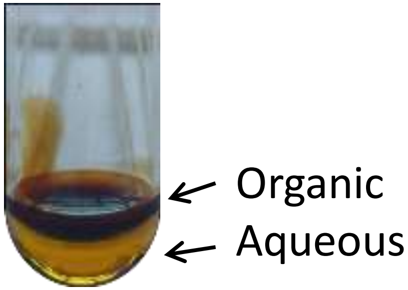
Bio-oil

- Product of pyrolysis of biomass
- Dark brown organic liquid
- High water content (~25 wt %)
    - Extremely high oxygen content
- + 1000s other compounds
- Ages instantly
- Composition dependant
on feedstock
- Low pH & biocatalyst inhibitors



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Anaerobic Digestion

Hydrolysis

- High molecular weight organic polymers split into smaller more bioavailable monomers.
  - Proteins > Amino acids | Carbohydrates > Monosaccharides | Fats > Fatty acids | + H_2

Acidogenesis

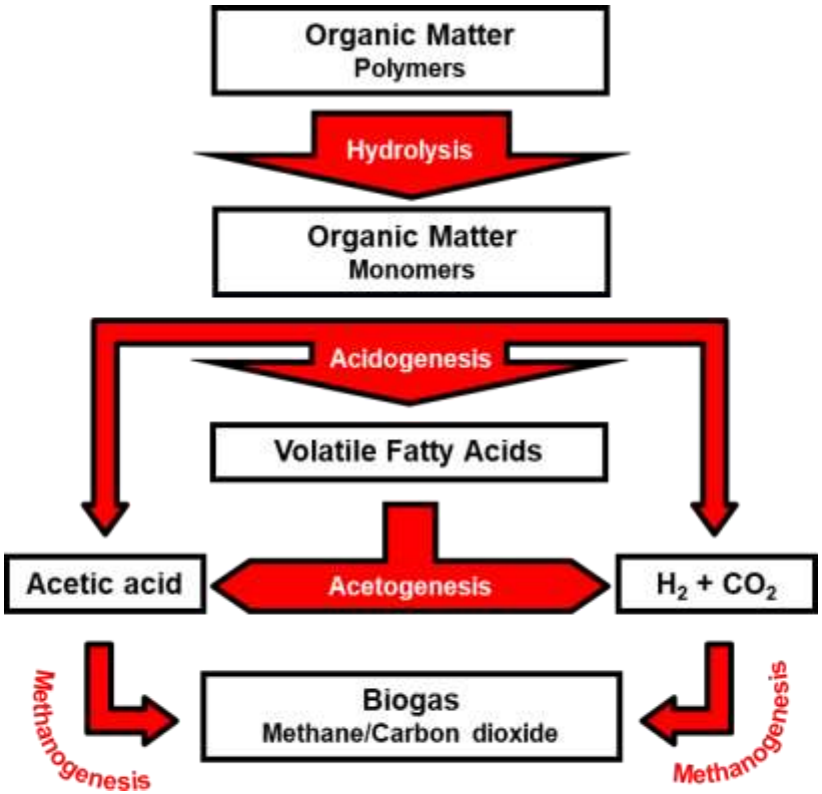
- Acidogenic fermentation of hydrolysed products to short chain…
  - volatile acids (propionic, butyric, acetic, formic, lactic)
  - alcohols (ethanol, methanol)
- $H_2 + CO_2 + NH_3 + H_2S$

Acetogenesis

- Further digestion of acids by acetogens to H_2, CO_2 and acetic acid.

Methanogenesis

- Methanogenic archaea convert H_2 and acetic acid to CH_4, CO_2.



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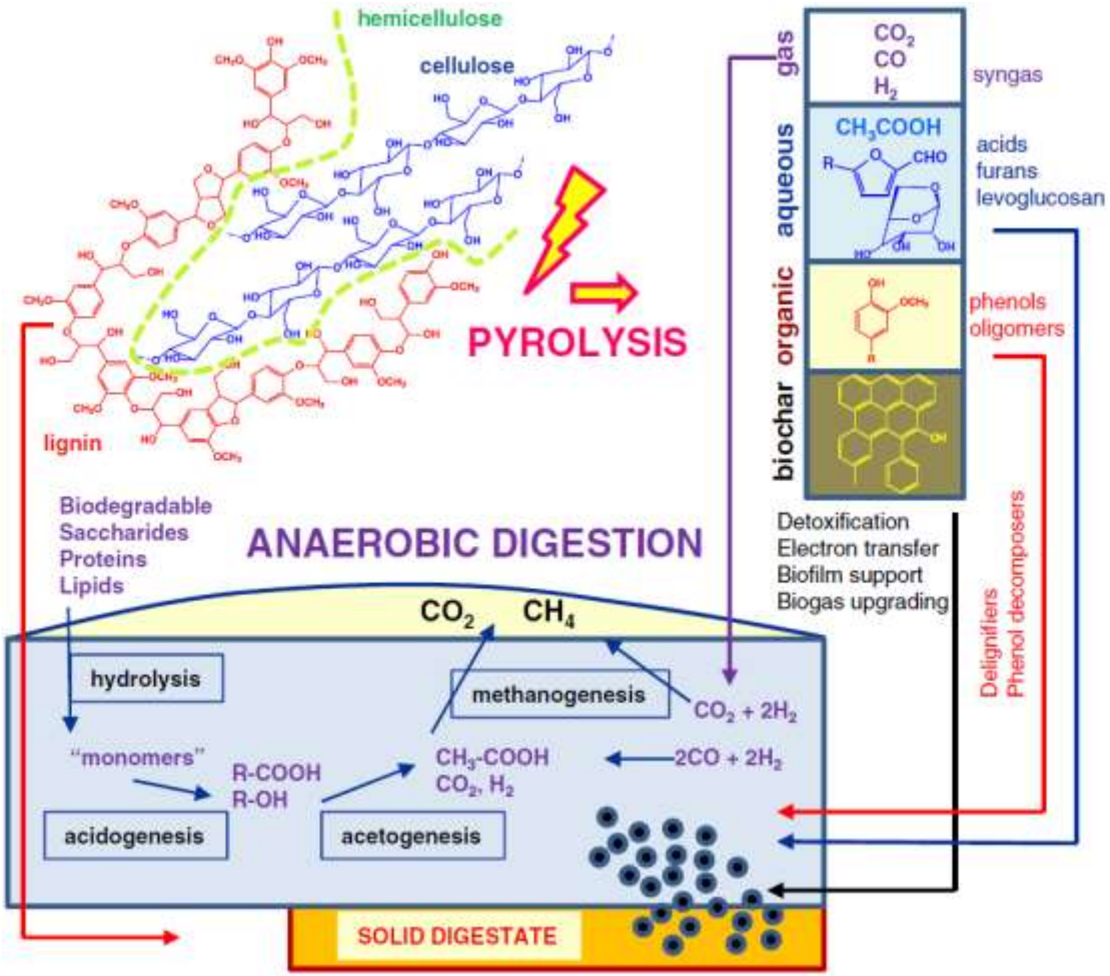
Py-AD

Vast range of microorganisms capable of bioconversion across a spectrum no single species could accomplish.

An ideal platform for the detoxification of complex organic mixtures such as bio-oil.

Enables relevant primary energy savings of non-renewable sources without worsening abiotic resources depletion + a strong reduction of GHGs emissions.



(Fabbri & Torri, 2016)

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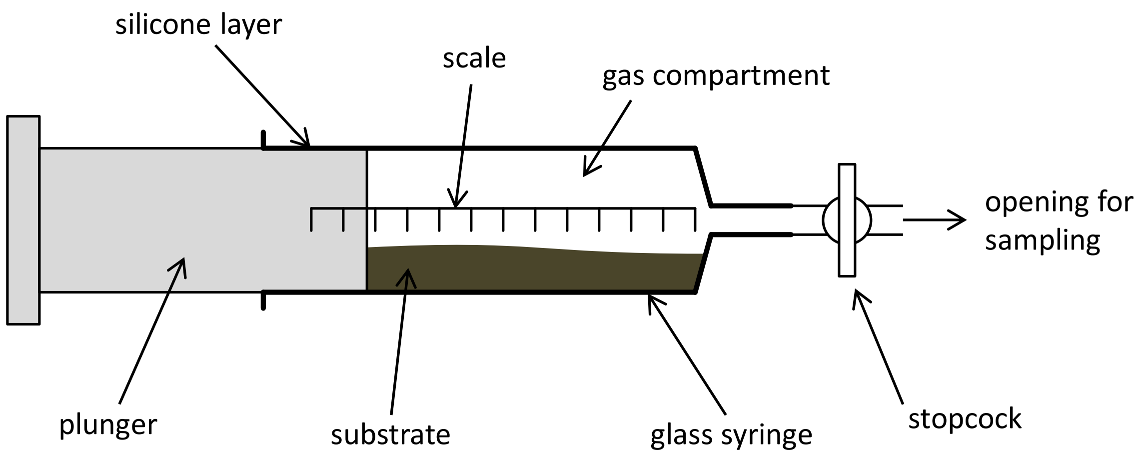
Hohenheim Biogas Yield Test



- 12 × 100 ml glass syringes
- 30 ml Seafield water treatment plant anaerobic digestate
- Supplemented with 10 g/l COD bio-oil, dried anaerobic digestate (AD), wood pellets (WP) or seaweed (SW)
- Mesophilic (~37°C) for 102 days

(adapted Mittweg *et al.*, 2012)

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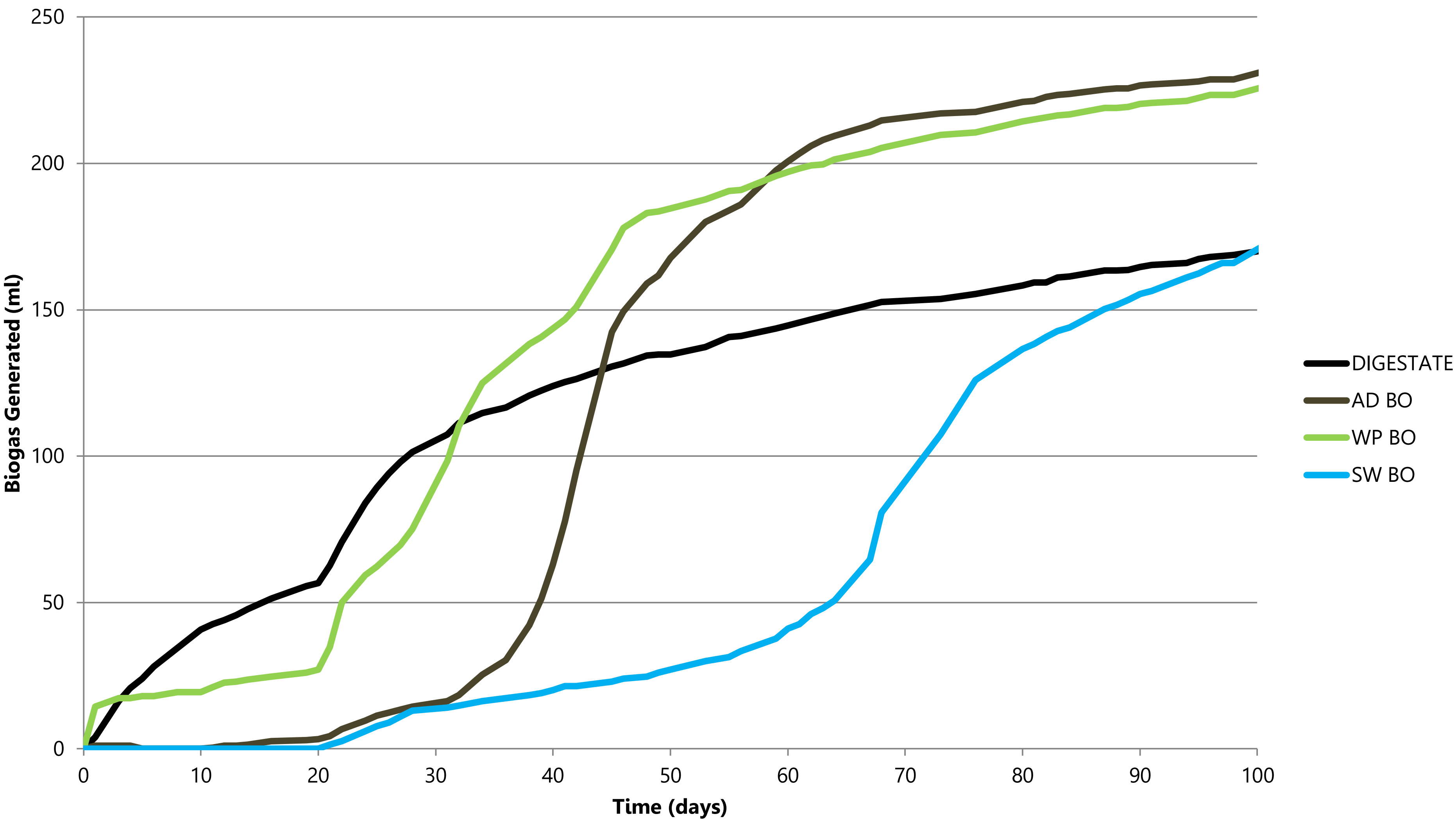
Biogas



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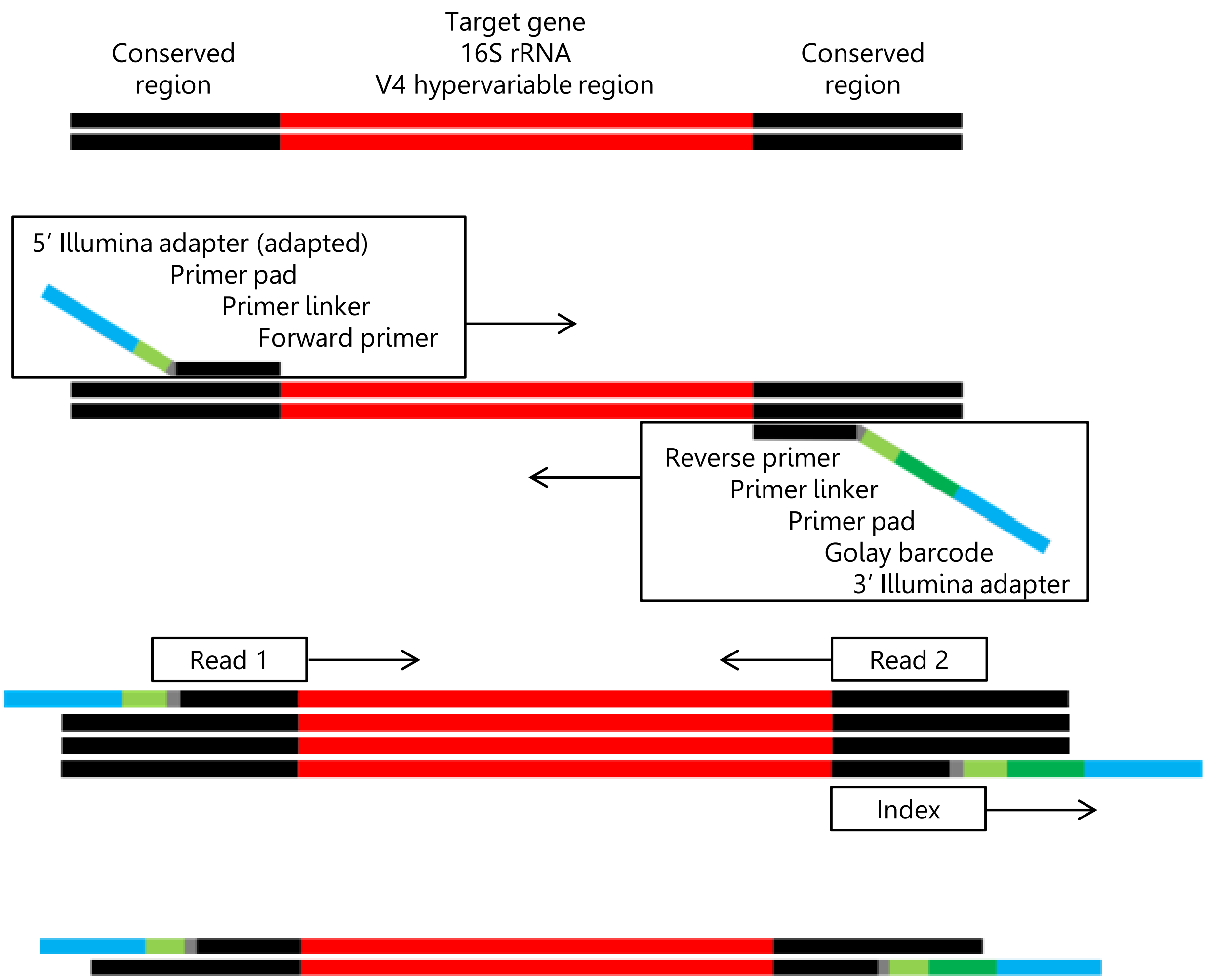
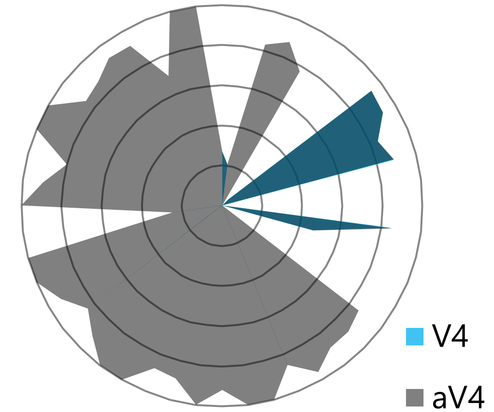
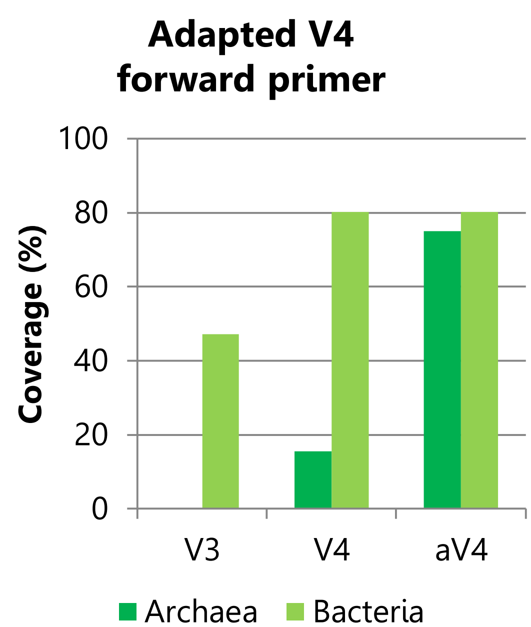
Illumina sequencing







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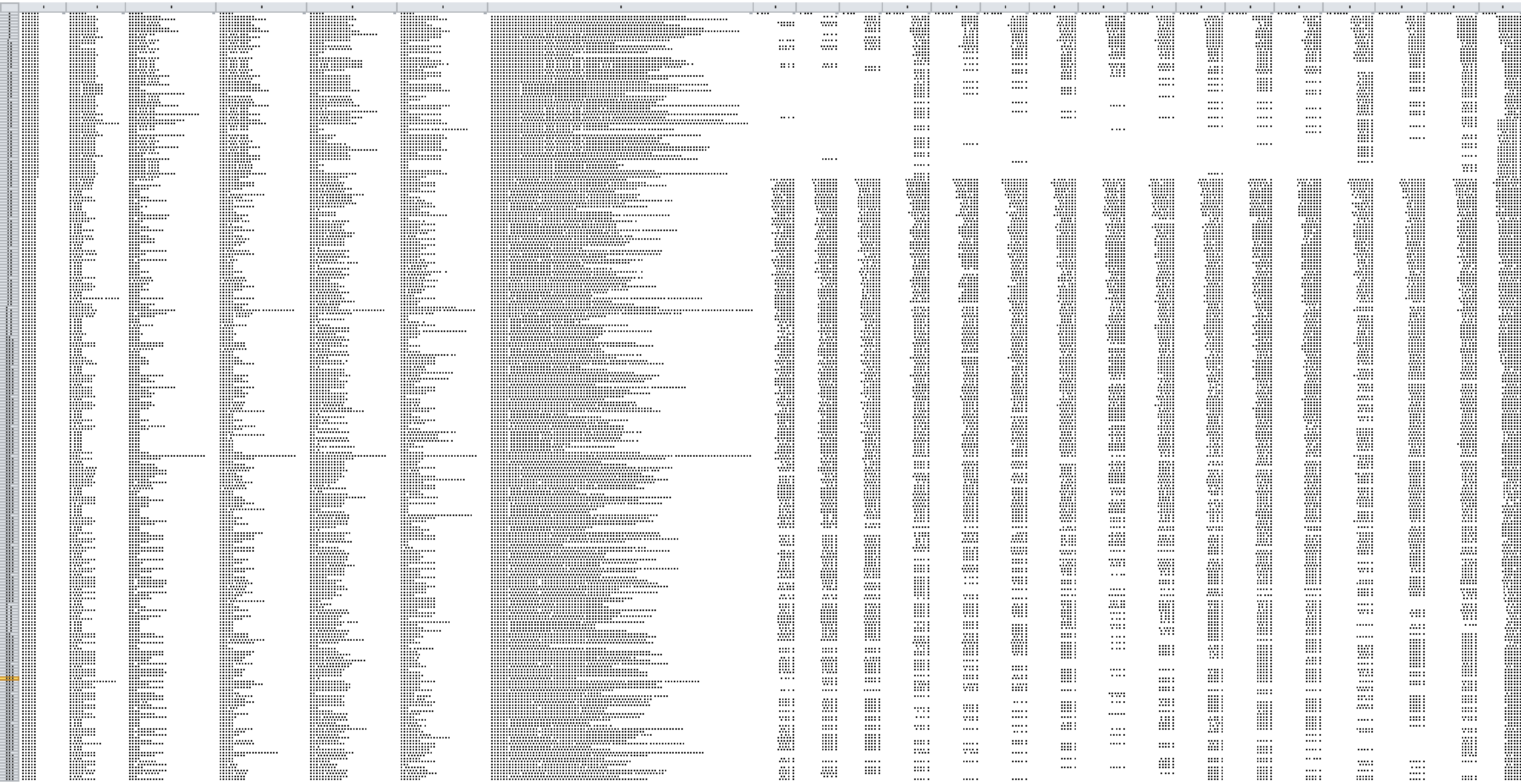

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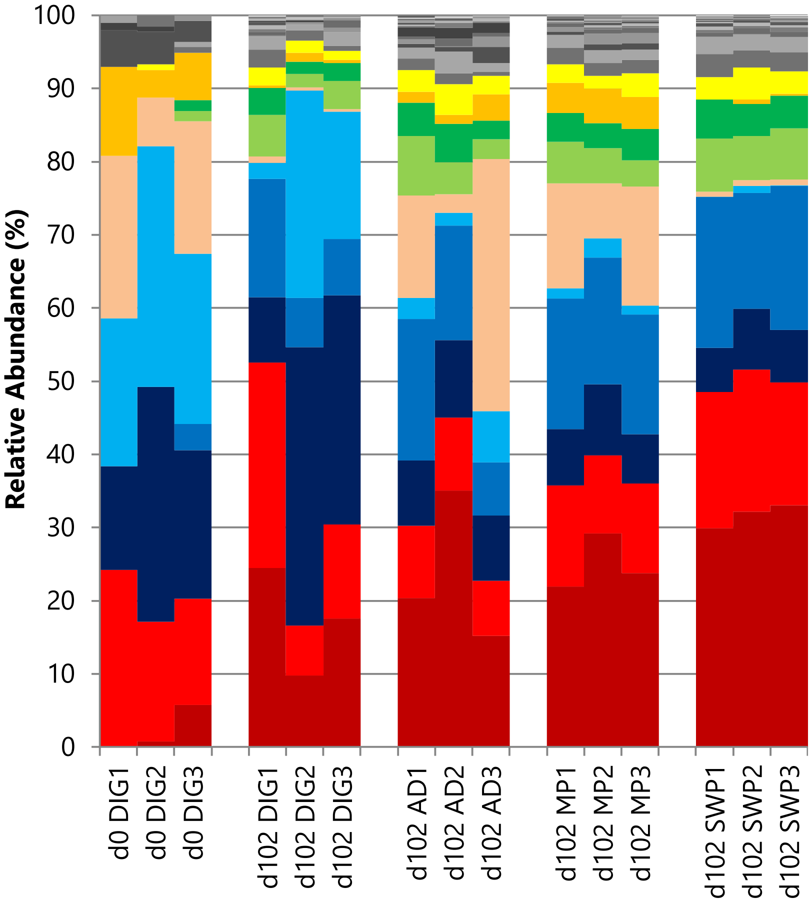

Phylum	Class	Order	Family	Genus
Euryarchaeota	*Methanobacteria*	*Methanobacteriales*	*Methanobacteriaceae*	*Methanobrevibacter*
Euryarchaeota	*Methanomicrobia*	*Methanosarcinales*	*Methanosaetaceae*	*Methanosaeta*
Euryarchaeota	*Methanobacteria*	*Methanobacteriales*	*Methanobacteriaceae*	*Methanobacterium*
Lokiarchaeota	*uncultured*	*uncultured*	*uncultured*	*uncultured*
WSA2	*WCHA1-57*	*uncultured*	*uncultured*	*uncultured*
Euryarchaeota	*Methanomicrobia*	*Methanosarcinales*	*Methanosarcinaceae*	*Methanosarcina*
Lokiarchaeota	*uncultured*	*uncultured*	*uncultured*	*uncultured*
Lokiarchaeota	*uncultured*	*uncultured*	*uncultured*	*uncultured*
Euryarchaeota	*Methanomicrobia*	*Methanomicrobiales*	*Methanomicrobiaceae*	*Methanoculleus*
Euryarchaeota	*Thermoplasmata*	*Thermoplasmatales*	*Marine Benthic Group D and DHVEG-1*	*uncultured*

Archaeal top 10

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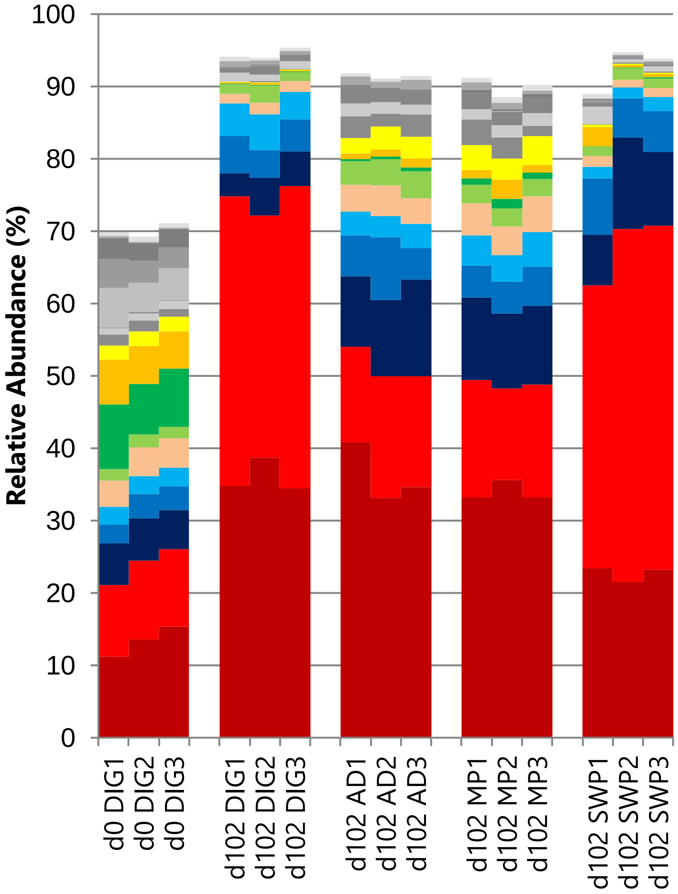

Phylum	Class	Order	Family	Genus
Thermotogae	*Thermotogae*	*Petrotogales*	*Petrotogaceae*	*Defluviitoga*
Cloacimonetes	*W5*	*uncultured*	*uncultured*	*uncultured*
Bacteroidetes	*Sphingobacteriia*	*Sphingobacteriales*	*Lentimicrobiaceae*	*uncultured*
Bacteroidetes	*Bacteroidia*	*Bacteroidales*	*Porphyromonadaceae*	*Proteiniphilum*
Firmicutes	*Clostridia*	*D8A-2*	*uncultured*	*uncultured*
Firmicutes	*Clostridia*	*Thermoanaerobacterales*	*Thermoanaerobacteraceae*	*Gelria*
Bacteroidetes	*Bacteroidia*	*Bacteroidales*	*Porphyromonadaceae*	*uncultured*
Proteobacteria	*Gammaproteobacteria*	*Pseudomonadales*	*Pseudomonadaceae*	*Pseudomonas*
Firmicutes	*Clostridia*	*Clostridiales*	*Caldicoprobacteraceae*	*Caldicoprobacter*
Firmicutes	*BSA1B-03*	*uncultured*	*uncultured*	*uncultured*

Bacterial top 10

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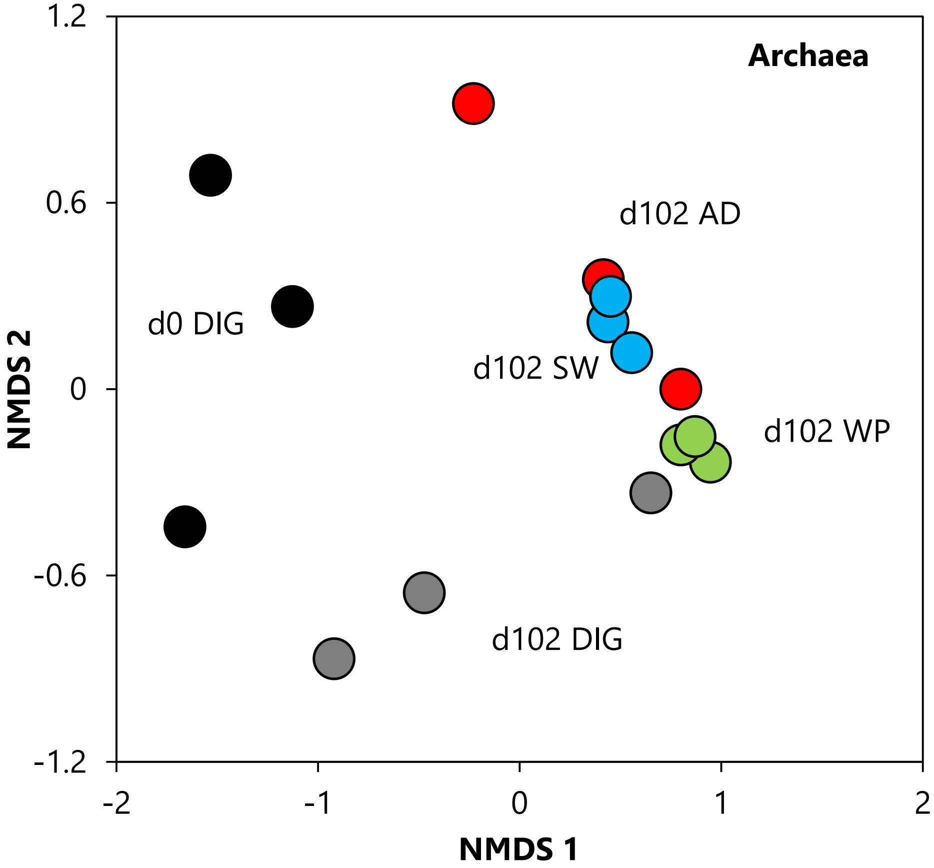

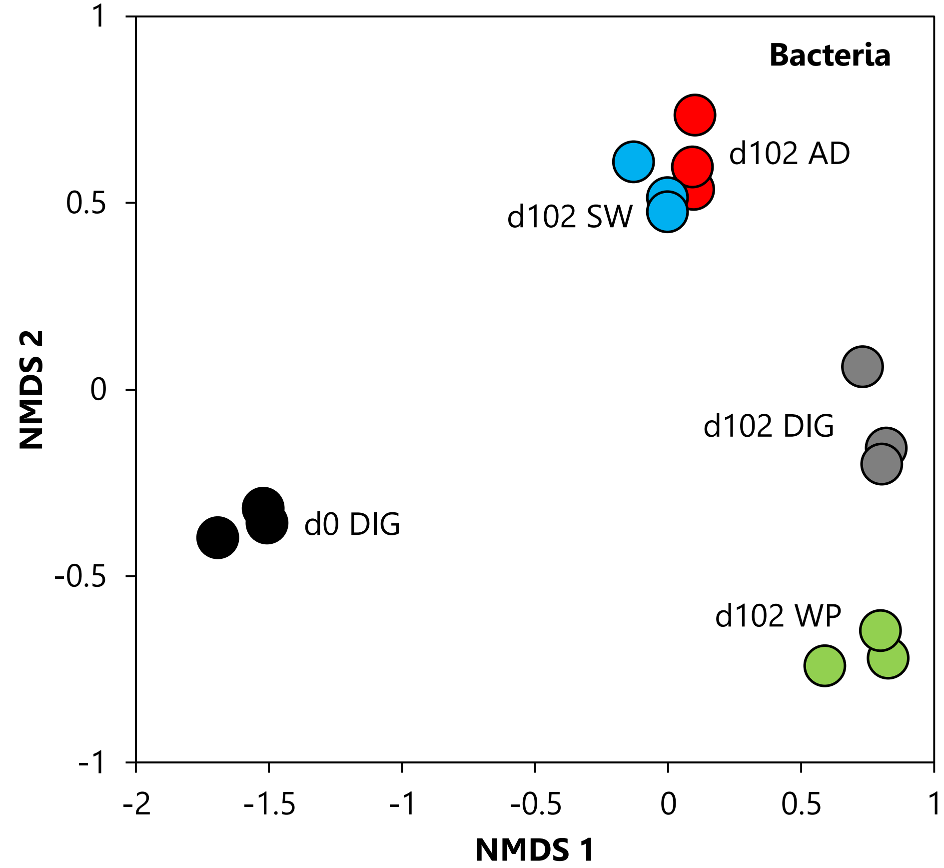

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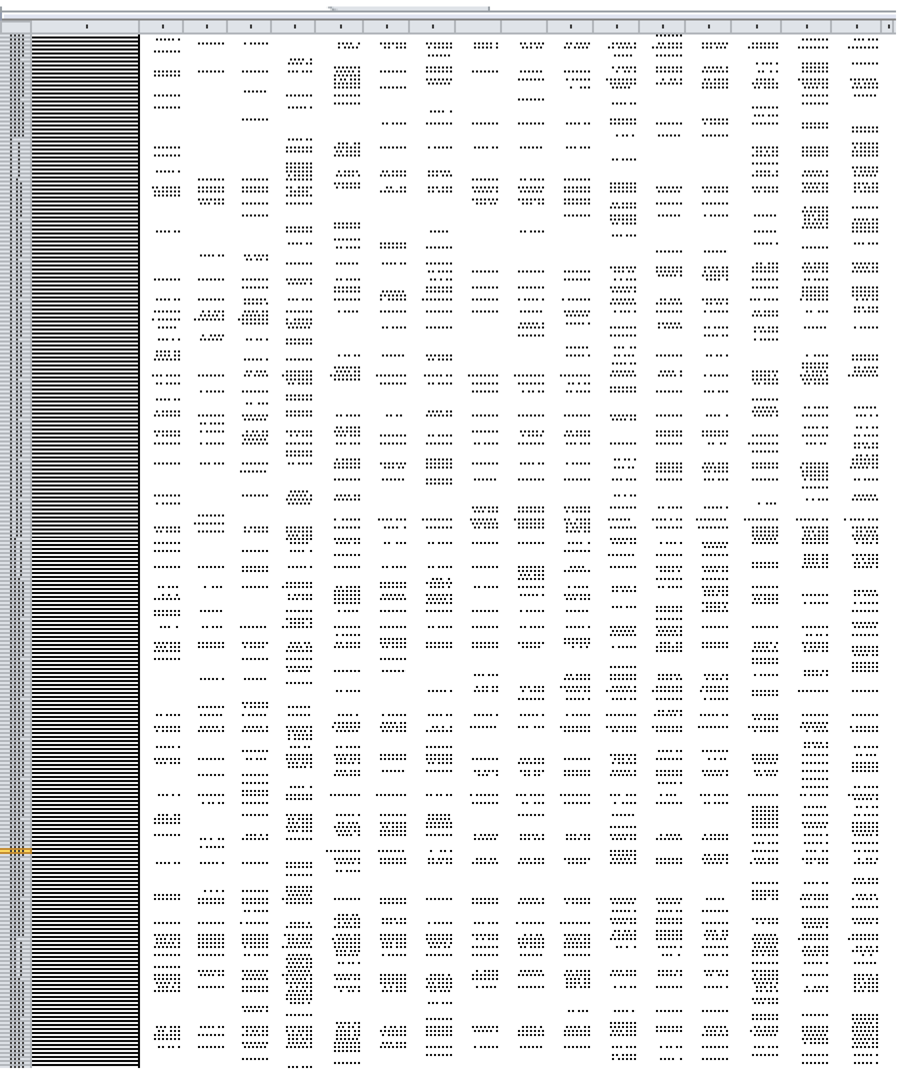
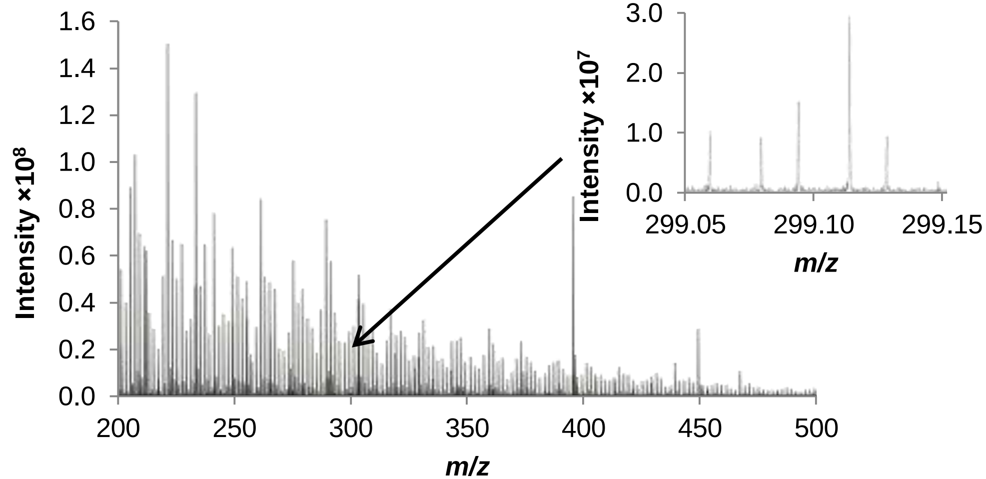
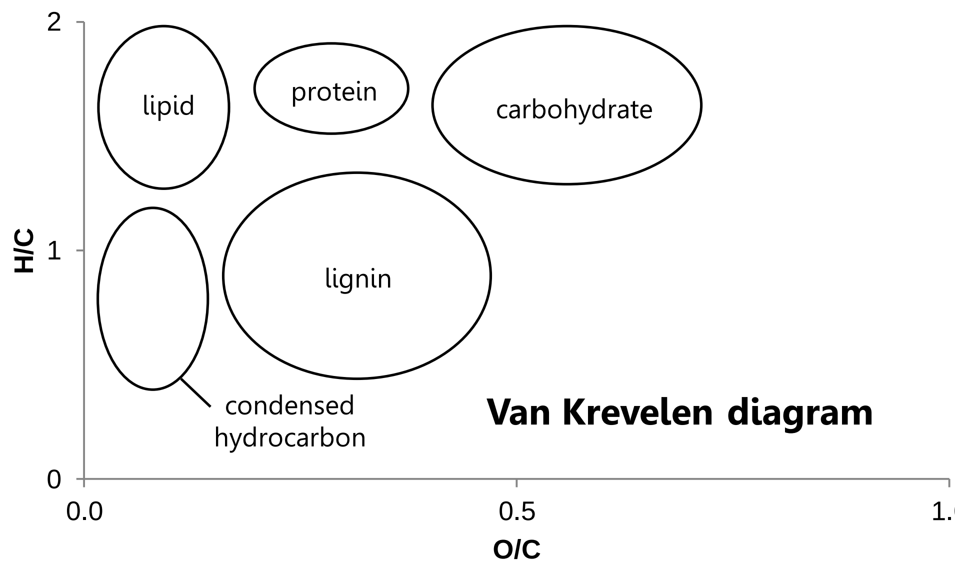
ESI FT-ICR MS DCNC CJℏ⊙Щ M CNIO RW∞







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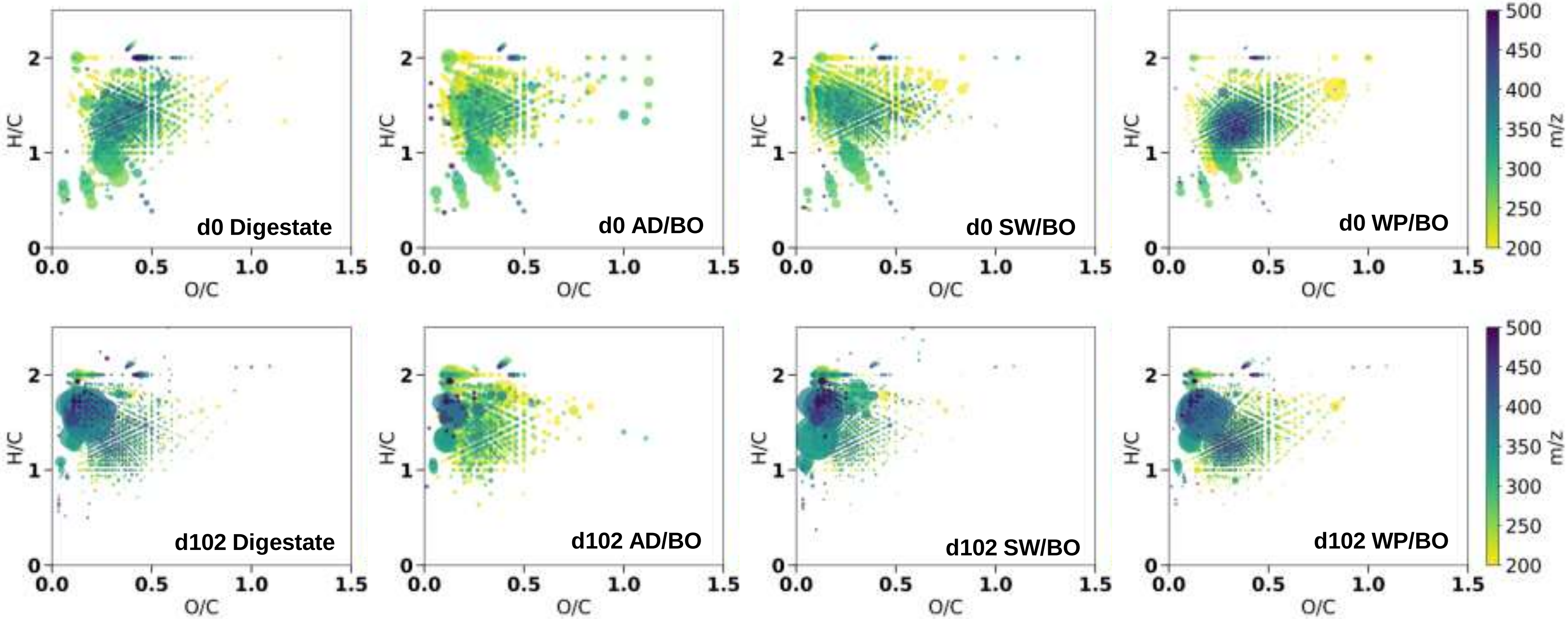

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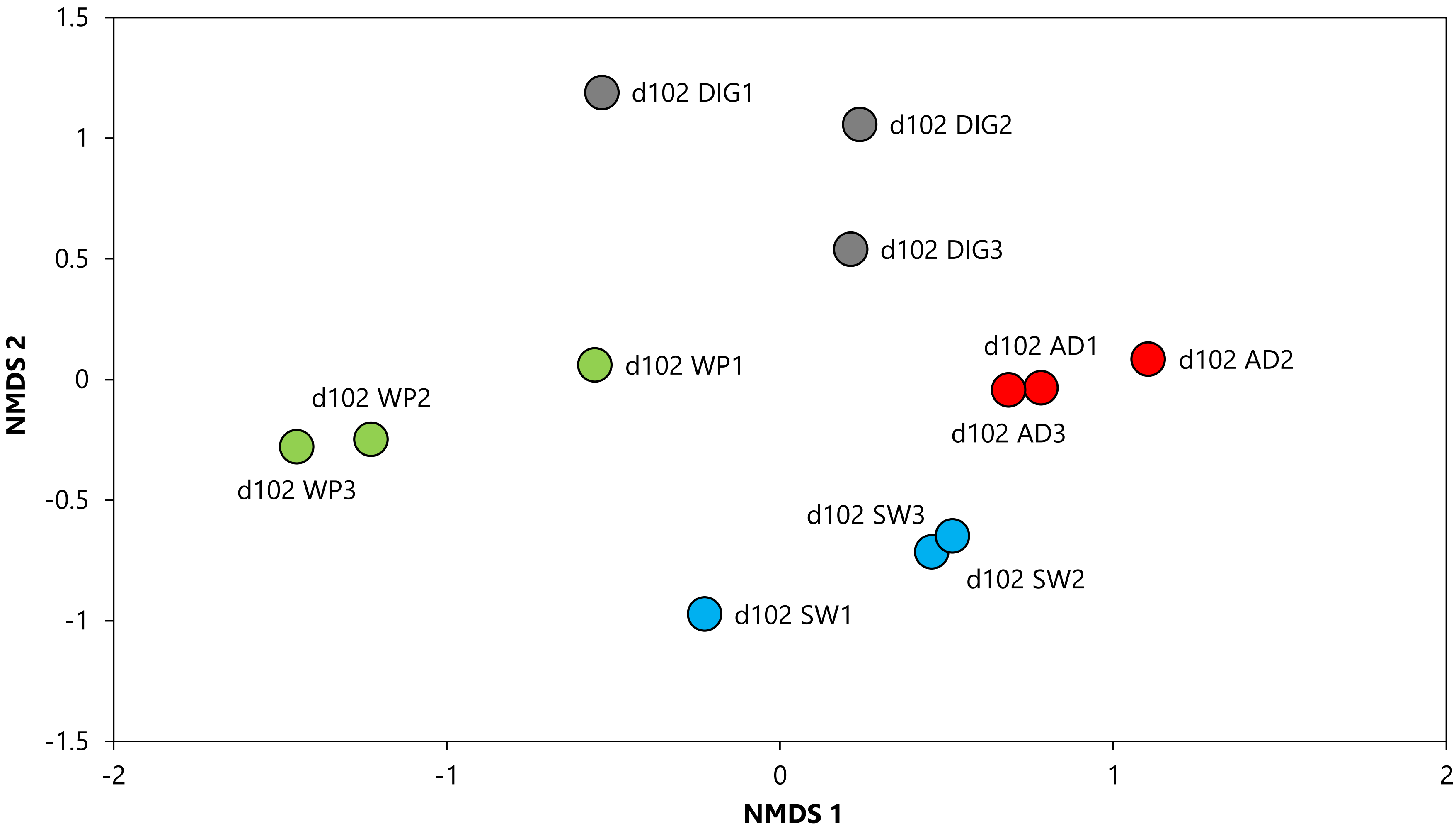

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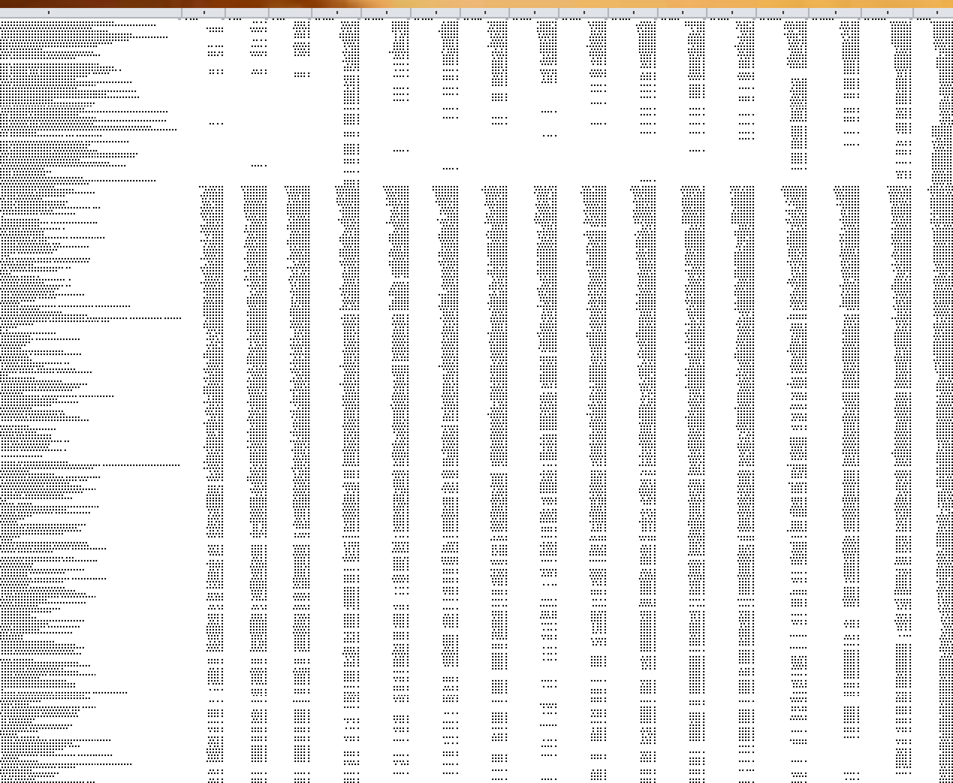
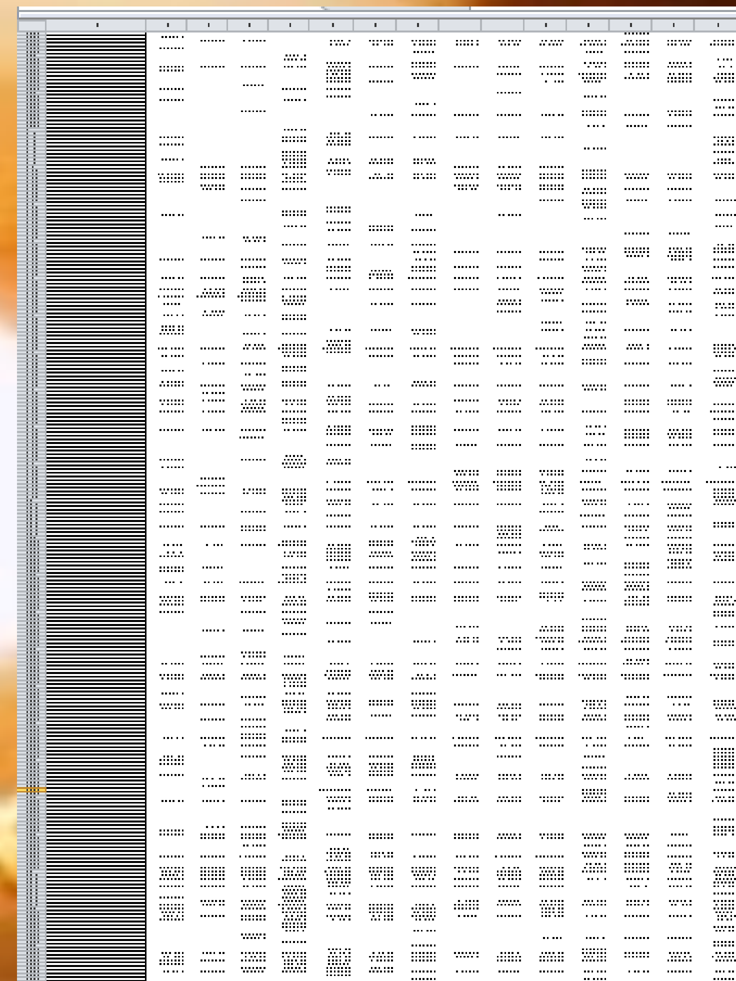
Relatedness





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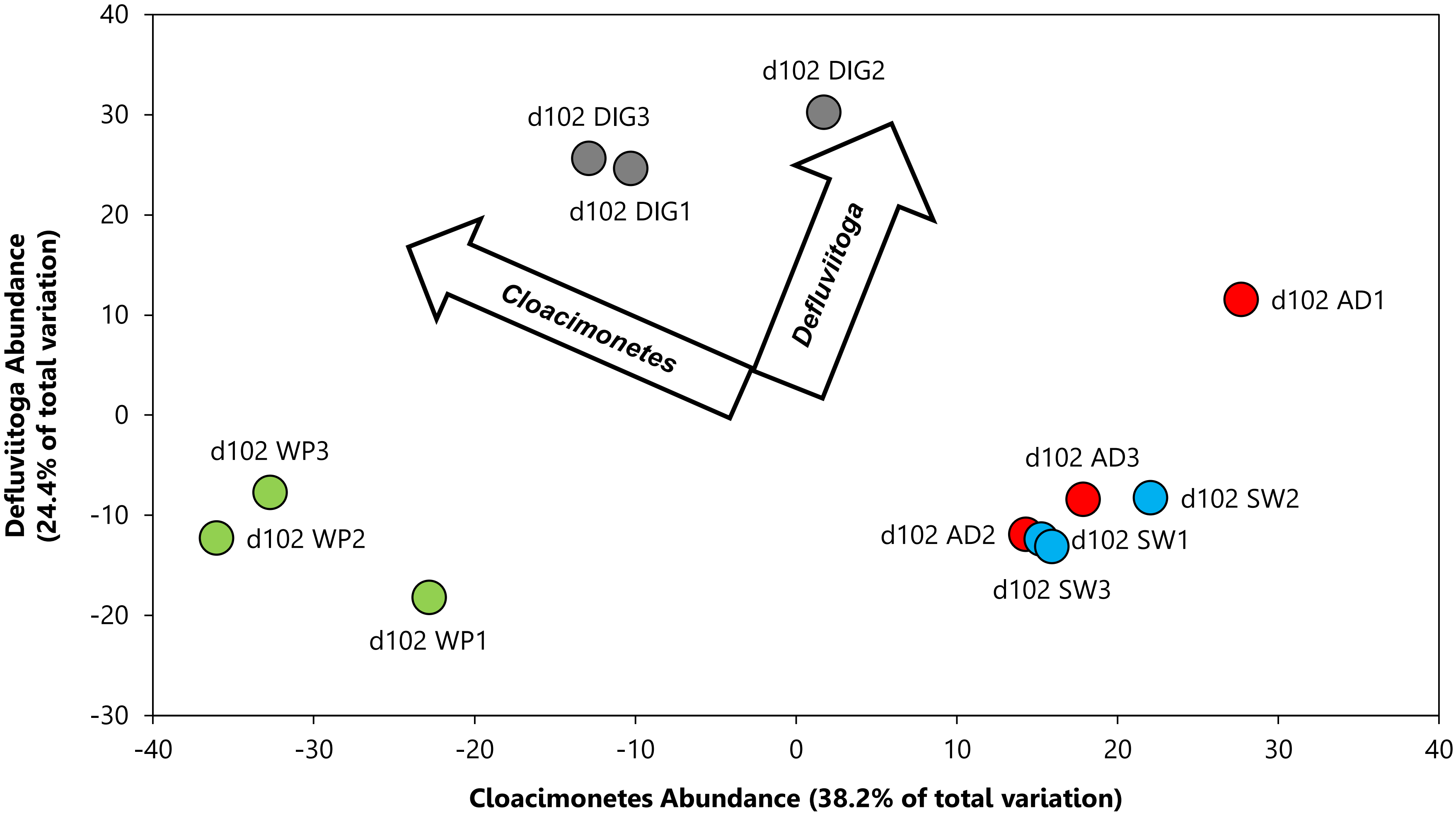

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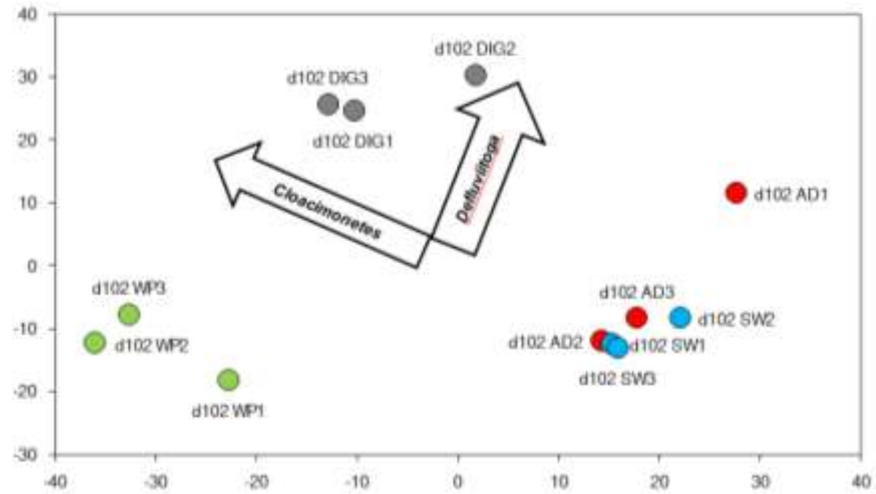
Relatedness

Analysis:

- Distance-based linear model (DistLM).
- Multivariate chemical data matrix using the abundance profiles of *Candidatus. Cloacimonetes* and the *Defluviitoga* as the predictor variables.
- The predictor variables are additionally plotted as vectors (annotated arrows).
- The abundance profiles of these two microorganisms are cumulatively able to explain 62.61% of the chemical variation observed.



- *Candidatus Cloacimonetes* phylum abundance correlates with the chemical pattern separating reactor conditions – propionate degradation?

- *Defluviitoga* suggests that increases in its abundance are related to the chemistry observed for digestate-only control reactors – specific inhibition by bio-oil?

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Importance

- Patterns in reactor chemistry can be correlated to fluxes in microbial community
- Understanding the microbial players involved at each stage of AD
- Longitudinal studies: continuous sampling of both the chemical and biological species involved to identify process bottlenecks
- Strategies to overcome inhibition

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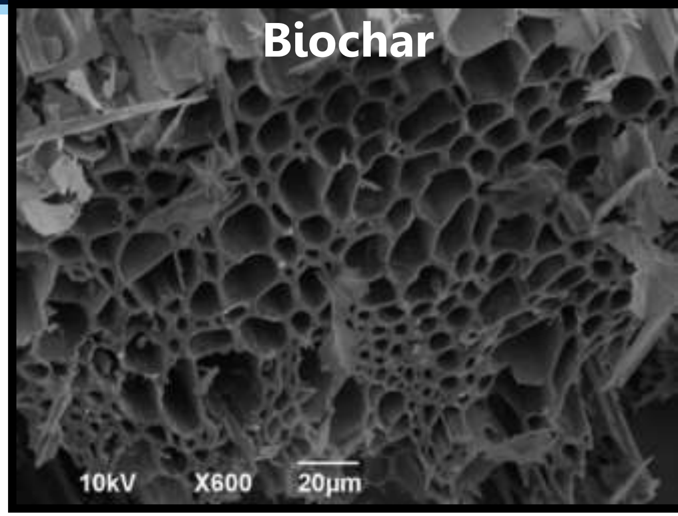
What next?

Biochar supplementation aids AD by the adsorption of inhibitory compounds and via the adherence of microbial cells in biofilms.

- High surface area, biofilm formation
- Biofilms show increased resistance to environmental stresses
- Partially conductive to the flow of electrons, capable of supporting direct interspecies electron transfer (DIET)



Scanning electron microscope image of a biochar surface. From Schulz, H. & Glaser, B. (2012).

carbogenics

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Thank you!

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