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Influence of microbial community composition on activated sludge floc properties

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Background

The activated sludge process is key in modern wastewater treatment. The activated sludge ecosystem contains a core community of abundant organisms and it must be stable in order to maintain the desired nutrient removal. In addition, bacterial morphology, mode of growth and extracellular polymeric substances (EPS) composition determine floc size, shape and strength, which in turn influence sludge properties that are important for plant operation. A few organisms have been correlated with different sludge properties, however, the picture is far from complete.

Conclusions

- Microbial community composition relates to different activated sludge fractions.
- Acrobacter and Sulfuritalea are enriched in the bulk water fraction and in the \bullet fraction loosely bound to the floc and could therefore be associated as poor floc formers.
- Dechloromonas and Tetrasphaera abundances decreased in supernatants and \bullet could therefore be associated with potential strong floc formers.
- Sulfuritalea and Tetrasphaera can be correlated to sludge characteristics that are important for the proper plant operation (SVI and shear sensitivity).

Aim

To investigate whether the microbial community composition differs between the flocs and the supernatant (bulk water) and whether certain bacterial species correlate to floc strength.

Methods

Sludge fractions



Activated sludge was collected from 23 wastewater treatment plants (WWTP) and physiochemical sludge characteristics were determined. Each sludge sample was then split in three

Results

Community composition relates to different activated sludge fractions from 23 WWTPs



Multivariate analysis visualized the relationship between all samples analyzed from 23 WWTPs.

Samples grouping into individual sludge fractions (total sludge – bulk water – loosely bound fraction) can be clearly observed. WWTP city names are presented.

- Total sludge a
- Supernatant before shear
- Supernatant after shear

Total sludge

fractions.

16S amplicon sequencing



Correlation between sludge parameters and bacteria in the sludge fractions

SLUDGE	VOLUME INDEX								
Total sludge				Superna	Supernatant after shear				
OTU	taxonomic classification	abundance [%]	R	OTU	taxonomic classification	abundance [%]	R		
276	g_Lactococcus	0.1	0.8	11683	fCaldilineaceae	0.9	0.6		
9794	gArcobacter	0.2	0.8	11886	gCaldilinea	0.2	0.7		
11683	fCaldilineaceae	3.2	0.7	12307	gPseudorhodobacter	0.2	0.7		
11886	gCaldilinea	0.1	0.7	4117	gGordonia	0.2	0.9		
12307	gPseudorhodobacter	0.3	0.7	8679	gFodinibacter	0.2	0.9		
4117	gGordonia	0.2	0.7	5480	gNocardioides	0.1	0.7		
8679	gFodinibacter	0.3	0.7	10334	gThermomonas	0.1	0.7		
13221	gStreptococcus	0.1	0.7	8804	gAlicycliphilus	4.6	0.8		
5480	gNocardioides	0.1	0.6	13375	gParabacteroides	0.2	0.6		
5643	gFerruginibacter	0.2	0.6	975	gMycobacterium	0.3	0.6		
7904	fChitinophagaceae	0.3	0.6	15219	gBrevundimonas	0.1	0.6		
6268	gLeucobacter	0.2	0.9	2634	gEnterococcus	0.1	0.7		
49	gBS063	0.3	0.7	13221	gStreptococcus	0.3	0.7		
5387	gGeothrix	0.4	-0.8	6930	gHF_BF39	0.4	0.7		
13371	gNannocystis	0.2	-0.7	2741	gB1-K2-141	0.5	0.6		
1858	gA0837	0.3	-0.7	2681	gMK04	0.2	0.6		
14179	gSulfuritalea	0.4	-0.7	5384	fRhodobacteraceae	0.3	0.7		
6532	gFerruginibacter	0.4	-0.7	954	fPhyllobacteriaceae	0.3	0.6		
9778	gSulfuritalea	0.5	-0.7	7086	f_Lachnospiraceae	0.1	0.8		
13898	gCandidatusEpiflobacter	0.7	-0.6	4378	f_Lachnospiraceae	0.1	0.6		
15957	fCaulobacteraceae	0.1	-0.8	5387	gGeothrix	0.1	-0.8		
5957	fAKYH767	0.1	-0.7	12821	cBetaproteobacteria	7.6	-0.8		
6267	oRhizobiales	0.3	-0.7	196	fenv.OPS_17	0.3	-0.6		
				10601	f 0319-6G20	0.1	-0.7		

Spearman correlation of bacteria present in different sludge fractions with important sludge characteristics: Sludge Volume Index (SVI) and shear sensitivity. Bacteria

PC1 [20.3 %]

Comparison of relative abundance between the different activated sludge fractions

Relative abundance of 10 most frequently occurring genera compared between the different sludge fractions. The variation between the WWTPs is captured by the boxplot width. Acrobacter seems to be linked to the loosely bound sludge fraction, whereas especially *Dechloromonas* and *Tetrasphaera* could be potential strong floc formers.

Enrichment of bacteria in different activated sludge fractions



SHEAR SENSITIVITY

Total sludge			Supernatant after shear						
0	TU	taxonomic classification	abundance [%]	R	OTU	taxonomic classification	abundance [%]	R	
59	954	gTrichococcus	2.1	0.6	14179	gSulfuritalea	3.2	0.7	
12	837	gPseudacidovorax	0.3	0.7	7201	gBacteriovorax	0.1	0.6	
104	410	gProteiniclasticum	0.2	0.7	6275	gTetrasphaera	1.5	-0.8	
8	31	gThauera	0.2	0.8	13677	pWCHB1-60	0.2	-0.7	
143	300	fPropionibacteriaceae	0.1	-0.6	6532	gFerruginibacter	0.2	-0.7	
19	94	gHyphomicrobium	0.9	-0.6	11480	grJ14	0.2	-0.7	
102	225	fKineosporiaceae	0.2	-0.6					
21	L01	gQEDR3BF09	0.3	-0.7					
65	532	gFerruginibacter	0.4	-0.6	negative influence on parameter				
13	898	gCandidatusEpiflobacter	0.7	-0.6	positive influence on parameter				
30	186	g Tessaracoccus	0 1	-0.7					

that were strongly correlated (-0.6>R>0.6) with the mentioned parameters are listed above. One abundant OTU, belonging to the class of Betaproteobacteria, seems to have a positive impact on SVI.

log2 re

High abundance of Sulfuritalea correlates with high shear sensitivity, whereas the presence of *Tetrasphaera* correlates to low shear sensitivity.

Bacteria enriched in the supernatant compared to total sludge. Highlighted bacteria seem to contribute to poor floc properties.



Bacteria enriched in the supernatant after shear compared to **supernatant**. Highlighted bacteria seem to be loosely attracted to the sludge flocs.







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