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# Prokaryotic communities in drinking water biofilters using an alternative filter medium



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## INTRODUCTION

Biofilters are often the heart of drinking water treatment systems in Europe [1]. Research in biofilters has contributed to optimize water treatment. Often filter media consist of washed, dried and sieved quartz sand. The function of alternative filter media for drinking water filters have been investigated [2-4]. These media include carbon products, ceramics, industrial waste products, and synthetic organics such as plastics, expanded clay, coated sands, and metal oxides. The duration of the start-up process has also been investigated [5-6]. However, the effect that alternative filter media may have on the microbial attachment and development of biofilm and therefore in the duration of the start-up period has received little attention.

The overall objective is to investigate the microbial communities on the filter media coating during the start-up of biofilters.

## METHODS

The investigations described in this poster were carried out at Fredensborg waterworks near Skanderborg, Denmark. The waterworks treats anaerobic groundwater using a simple process of aeration and filtration (2 filters in series).

### Setup

After evaluation of several filter media properties, a filter column of calcium carbonate was selected to remove manganese using as inlet water between the waterworks filters. The water was spiked with manganese to achieve a constant concentration of 0.27 mg.L<sup>-1</sup>.

### Water and Filter media samples

Filter medium properties of quartz sand, calcium carbonate, anthracite and manganese oxide were determined using gravimetric methods and a photometric particle analyzer (Camsizer@64, Retsch Technology GmbH).

Water samples of the setup inlet (water between filters from the waterworks) were taken during the experiment and analyzed for standard parameters.

Water samples (water between the filters and clean water from the waterworks) and filter media samples (second filter of the waterworks and filter column) were collected and analyzed DNA extraction, qPCR with broad range bacterial primers, and amplicon sequencing using 16S rRNA primers. The analysis identified the most abundant amplifiable Phyla and Genera in the samples.

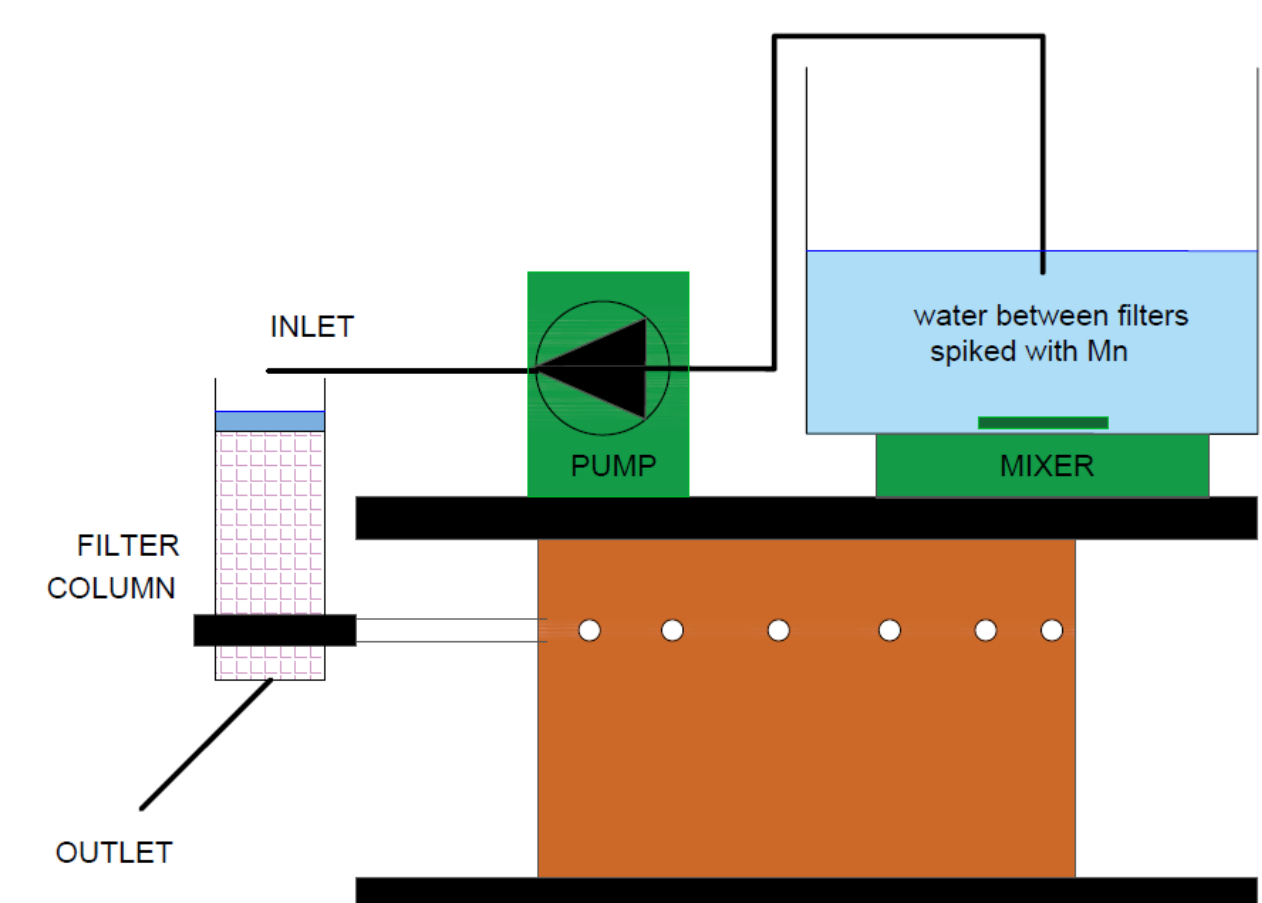


Figure 1 – Setup with a calcium carbonate column.

### At-line measurements

Oxygen concentration, pH, redox and temperature of the column's inlet was frequently measured using Multi 3430 meter (WTW GmbH, Germany).

## DISCUSSION

### DNA extraction, qPCR and pyrosequencing results

Table 3 – Sample overview and qPCR results.

Sample	Ext.DNA [ng/uL]	Lib. DNA [ng/uL]	qPCR [copies/uL]	qPCR [copies/ng DNA]	Reads [#]
FM from Filter 2	0.1	7.53	1.2E+03	1.4E+04	25797
FM from setup	17.3	14.90	2.0E+06	1.2E+05	16122
Water bet. filters	1.4	0.00	-	-	0

Figure 3 shows that microbial analyses detected the presence of commonly reported prokaryotic groups: *Alphaproteobacteria*, *Betaproteobacteria*, *Nitrospirae*, *Acidobacteria* and *Gammaproteobacteria* [7].

Pyrosequencing showed attachment of bacteria present in the inoculator, water between filters of the waterworks, on the filter medium surface (Figure 4).

*Nitrospirae* (AOB's) were much more abundant (2-3 times) on the calcium carbonate column than in the water between filters (used as inlet).

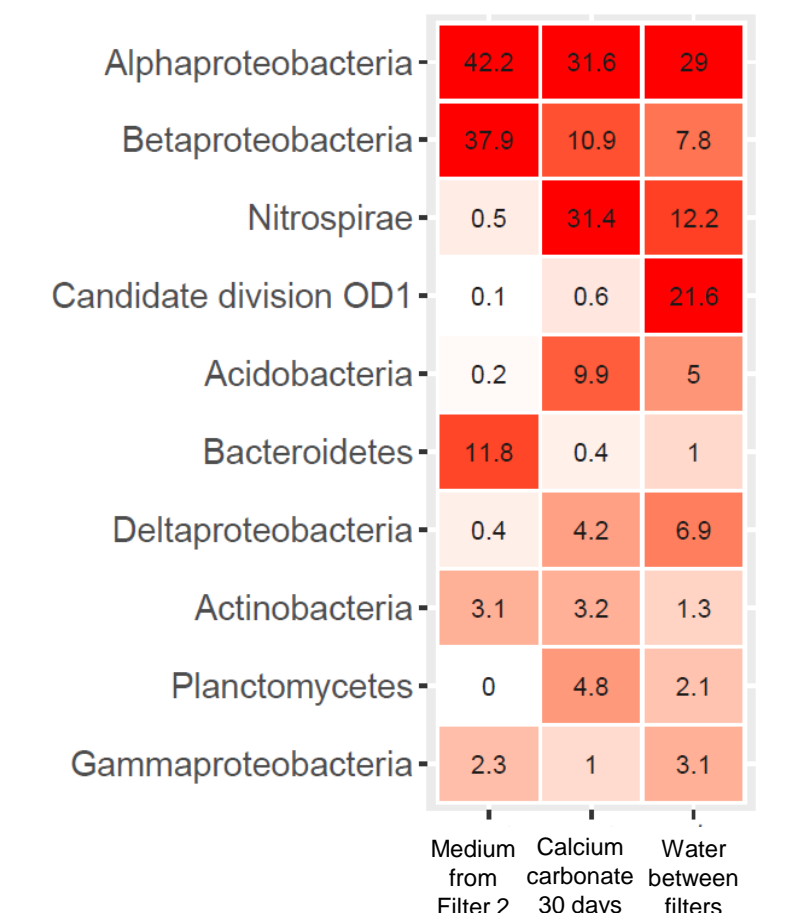


Figure 3 – The 10 most abundant Phyla.

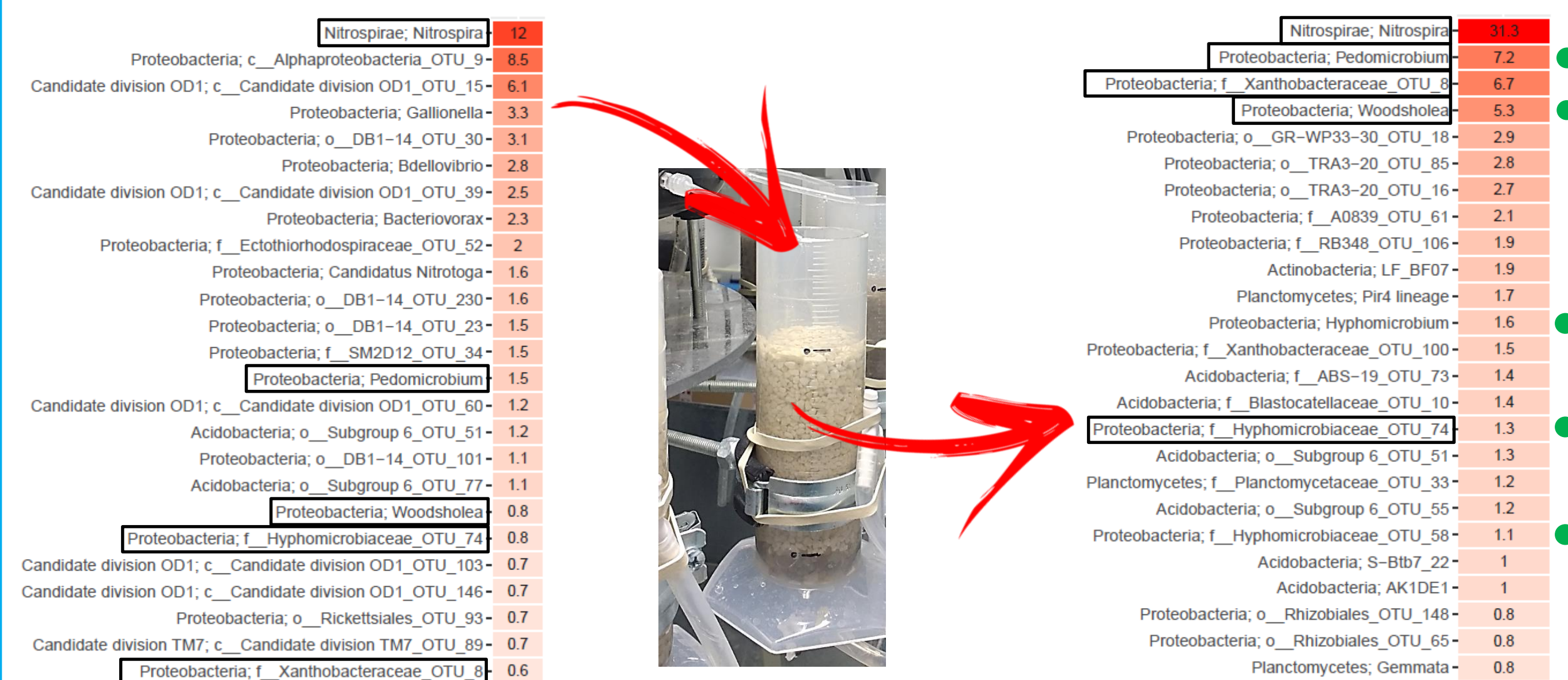


Figure 4 – Comparison of microbial diversity in the inlet (water between filters of the waterworks) and the filter medium coating - after 30 days, 20% of manganese removal (20 minutes of contact time). □ Detected in both inlet and filter medium samples. ● Commonly connected to biological oxidation of manganese.

## RESULTS

### Filter media properties

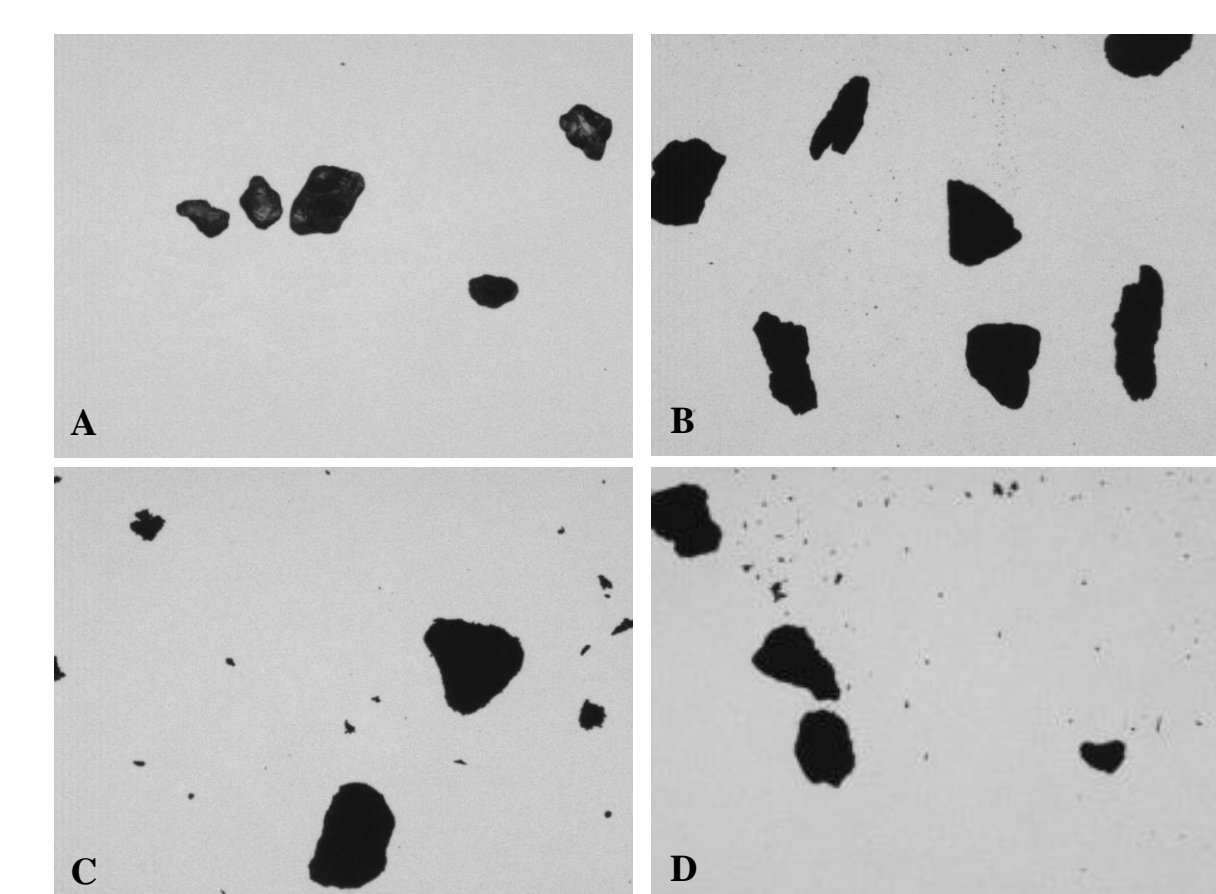


Figure 2 – Photometric particle analyzer of different media.

### Water quality before filtration

Table 1 – Water quality before column.

Water quality before filters		
Parameter	Average	Unit
Treatment parameters		
Oxygen	11.1	mg.L <sup>-1</sup>
Iron	0.010	mg.L <sup>-1</sup>
Manganese	0.27	mg.L <sup>-1</sup>
Ammonium	<0.02	mg.L <sup>-1</sup>
Others		
pH	7.6	
Redox	230	mV
Temperature	12.2	°C

Table 2 – Properties of the different filter media.

Filter media	Shape	Grain Size (mm)		Porosity	Density gcm <sup>-3</sup>
		(10%)	(90%)		
A. Quartz	0.86	0.50	0.80	0.46	1.48
B. Calcium carbonate	0.85	2.30	4.10	0.46	1.40
C. Anthracite	0.81	1.40	2.50	0.50	0.95
D. Manganese oxide	0.82	1.60	3.20	0.49	1.68

## CONCLUSIONS

- After 30 days of start-up, 20% of the manganese in the inlet was removed (contact time of 20 min). Even with low manganese removal, there was a selection for some taxonomic groups on the filter material relative to the inlet.
- 16S rRNA amplicon sequencing showed attachment of bacteria commonly reported as MnOB's and AOB's on the medium coating. These included bacteria present in the inoculator (water between filters). Further, bacteria undetected in the inoculator suggest that other MnOB's were formed in the coating of the medium, such as: *Hyphomicrobium* and *Hyphomicrobiaceae*.
- Further investigations on microbial communities evolving on different filter media can have great importance for start up and management of biofilters for drinking water treatment.

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