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Enhancing sustainable biodegradation of organic micropollutants by methanotrophic microbiomes: insights into the interaction between heterotrophic and methane oxidizing bacteria

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Introduction: Danish monitoring programs show that organic micropollutants (OMPs) occurrence in water works wells has increased in the last decade (Thorling & Geus, 2021). Most of the current technologies employed for OMPs removal in raw drinking water facilities have shown either limited removal efficiencies or very high operational costs (Shahid et al., 2021). It is therefore necessary to find sustainable and economical solutions to implement at drinking water facilities. Organic micropollutants can be biologically degraded through primary metabolism or co-metabolism. In co-metabolic processes, microorganisms transform micropollutants by unspecific enzymatic activity without assimilating carbon or obtaining energy (Benner et al., 2013). Efficient biodegradation of OMPs through co-metabolic activity of methane oxidizing bacteria (MOB) has been reported (Hedegaard et al., 2018), (Papadopoulou et al., 2019). Previous studies have shown that aerobic heterotrophs present in methanotrophic microbiomes play a significant role in the biodegradation of OMPs (Mortensen et al., 2023). In the latter study, the effect of CH_4 loading rates was investigated in a hybrid membrane biofilm reactor (hMBfR), leading to 100 % and 88 % removal efficiency of sulfamethoxazole (SMX) and 1H-benzotriazole (BZT), respectively (Fig. 1). Strikingly, BZT was mostly biodegraded by the suspended biomass, poor on MOB. Little information is available on the effect of operating conditions on interactions of heterotrophs and MOB, and biodegradation potential of mixed enrichments of those communities. In the present study we investigated effect of solid retention time (SRT), nitrogen source, methane loading, O_2/CH_4 ratios and preadaptation to OMPs on biodegradation potential of frequently occurring OMPs in Danish groundwater systems.

Methods: Three different sets of inoculums were sampled from a fermenter, a groundwater treatment plant, and a hospital wastewater treatment plant. Enrichments were cultivated in 250 mL serum bottles in batch mode. Cultures were exposed to diluted nitrate and ammonium (dNMS and dAMS) solutions, continuously shaken at 25°C. They were exposed to two different SRTs (5 and 15 days), two different methane initial concentrations (6.07 and 3.03 mg CH_4/L), and different O_2/CH_4 ratios. Preadaptation of cultures to OMPs was ensured by spiking a stock solution containing seventeen (17) parent compounds and transformation products. Three batch experiments in sequence were run for culture adaptation and characterization (growth rates, methane, and oxygen yields). At the end of the third growth curve, a 48h batch test was run to characterize OMP removal.

Results: Indigenous cultures from the three different sources were successfully enriched under methanotrophic conditions and utilized to investigate effect of operating parameters on co-metabolic activity of heterotrophs and MOB. OMPs biokinetics are not included due to space limitations.

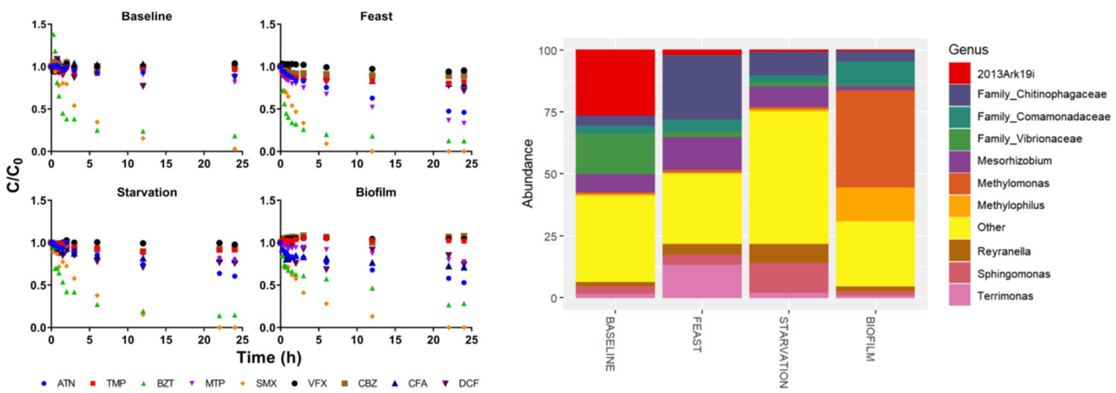

Figure 1. Degradation of OMPs and relative abundance of bacteria at genus level during batch tests at three CH_4 feeding regimes, and biofilm (Mortensen et al., 2023).