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Capture of methane by biofilter fungi - A chromatographic isotherm study

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Extended Abstract.

Methane (CH_4) is the second most potent and relevant anthropogenic greenhouse gas (GHG). Livestock operations are a significant source of anthropogenic CH_4. Biofilters, low-cost bioreactors that use microbes to capture and degrade emissions from livestock operations, have GHG mitigation potential, but farm exhausts which require high-ventilation rates limit CH_4 capture. Biofilter microbiota likely affects CH_4 sorption and we hypothesize that fungi, due to their unique physiology, can improve CH_4 capture in livestock emission biofilters. To test this, a chromatographic isotherm approach was used, where a column packed with ratios of inert sand and fungal material were plumbed in a gas chromatogram oven to the injection and detection ports. Methane spikes were injected into a field relevant flow and sorption characteristics were studied. Results show increased sorption with fungal augmentation for both tested materials. These results suggest increasing fungal biomass in biofilters may enhance CH_4 biofiltration. Optimizing biofilters to reduce GHGs in addition to odors has low-cost potential to simultaneously protect local air quality and the global environment.

Problem Statement

Methane, a potent greenhouse gas emitted largely by animal agriculture in the US (US-EPA, 2014), is technically and financially difficult to manage on farms. Biofilters are low-cost pollution control bioreactors that use microbial growths to capture and degrade pollutants from contaminated farm effluents. Biofilters have the potential to mitigate CH_4 emissions from animal houses and manure storages (Veillette et al., 2012) in addition to odor (Chen and Hoff, 2009), but as currently deployed capture little CH_4. Unlike passive flow biofilters successfully used on landfills to remove CH_4 (Ménard et al., 2012), exhausts from animal agriculture are high-volume and forced quickly through a biofilter, creating capture, not CH_4 biodegradation rate limits. Fungi have unique physiology that has been utilized in industrial biofiltration systems to improve mitigation of hard to capture, hydrophobic emissions (Kennes and Veiga, 2004). While fungi are present in livestock emission biofilters, their biocatalytic potential has been poorly characterized despite their potential to enhance emissions control (Ralebitso-Senior et al., 2012). If fungi can be shown to improve CH_4 capture as they have for other emissions it is possible they can be harnessed, and by selection of their growths biofilters currently managing odorous livestock emissions can be optimized for CH_4 mitigation.

Objective

The aim of this work was to use a flow-through chromatographic isotherm column to test and explore the ability of fungal materials to capture CH_4.

Methods

To test the influence of fungal materials on CH_4 sorption, a flow-through chromatographic isotherm approach was used (Figure 1), similar to the experimental set-up used by Goss (1992) to study vapor partitioning in soils. The isotherm test column consisted of a 10 cm X 1.6 cm ID stainless steel tube capped at both ends with 2 mm thick, 0.5 um pore stainless steel fritted metal disks. Stainless steel compression fittings secured the disks in place, enabled quick exchanges of test media and were fitted with denatured capillary gas chromatography (GC) columns (60 cm X 0.3 mm ID). In an up-flow orientation, the test column was plumped to the injection port and FID detector in the oven of a Shimadzu GC (2010 Plus, Shimadzu Corp., Kyoto, Japan). The oven was maintained near ambient (25°C), an inert carrier gas was used (He), and CH_4 injections (10 µL 100 ppm) were made with a gas-tight syringe fitted with a Chaney adapter (Hamilton Co., Reno, NV, USA). Ultrapure sand (40-100 mesh, manufacturer), muffle furnaced to remove any residual organics, was used as inert control media, while uniform fungal materials were homogenized with sand in various ratios for the treatments.

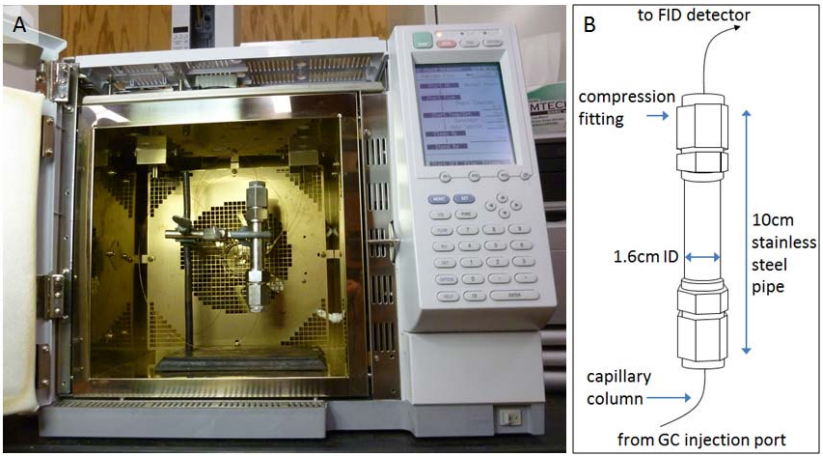

Figure 1.A Photograph and **B** schematic of isotherm test column.

Unlike batch isotherms used to study the biofiltration phenomena, this system is flow-through and may better model actual biofilter dynamics. Furthermore, by retrofitting a GC, abiotic conditions are tightly controlled and detection is real-time and highly sensitive. Under isothermic conditions, the observed CH_4 retention is a measure of its sorption patterns. By comparing sorption patterns of a treatment to the sorption patterns of the control, the ability of fungal materials to sorb CH_4 can be tested (*i.e.* smaller peak areas with treatments compared to controls suggest CH_4 is sorbed to the fungal material being tested).

Statistical analyses were conducted with R 3.0.1 (GNU Project). Due to outlier points, normal distribution was not verified by a Shapiro-Wilk test, and therefore, Krustal-Wallis was used to test for differences in mean peak areas. Tukey HSD was used post-hoc to explore differences in these means. The alpha level was set to 0.05 for all analyses.

Results

Preliminary results suggest fungal materials can sorb CH_4. When the inert sand control chromatographs were compared to chromatographs from a treatment, sorption of low concentration CH_4 was clearly observed (Figure 2). Evident by the consistent peak area of the treatments (*i.e.* the area did not increase with subsequent injections); the sorption sites appear to be un-saturated even when small fungal levels were present. The lack of significant peak tailing suggests the fungal sorption of CH_4 is irreversible or at least long-lasting.

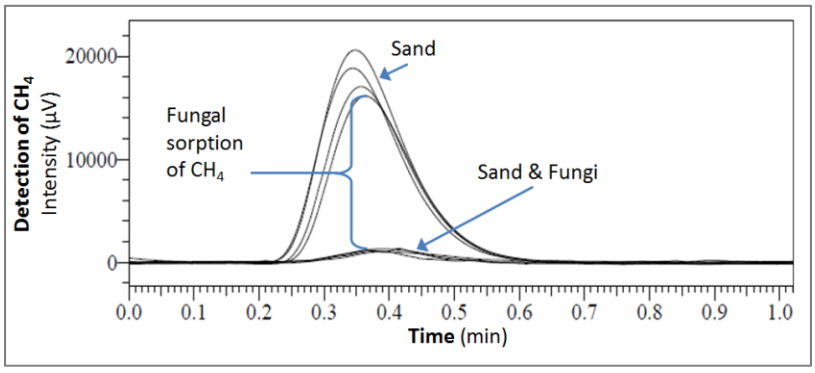

Figure 2 Overlay of chromatographs showing ability of fungal material to capture low concentration CH_4.

Fungal materials from both major phyla (Ascomycota & Basidiomycota) showed an ability to sorb CH_4 (Figure 3). Increasing the proportion of fungal material relative to the inert sand increased the ability of the isotherm column to sorb injected CH_4 (*i.e.* peak area was negatively correlated to the fungal ratio). While this pattern was not significant for the ascomycete material for the ratios tested, results for the basidiomycete materials for the ratios tested were statistically significant. Interestingly, the ascomycete material tested apparently sorbed more of the spiked CH_4 than the basidiomycete materials. More data and repeated runs are needed to accurately quantify sorption capacities.

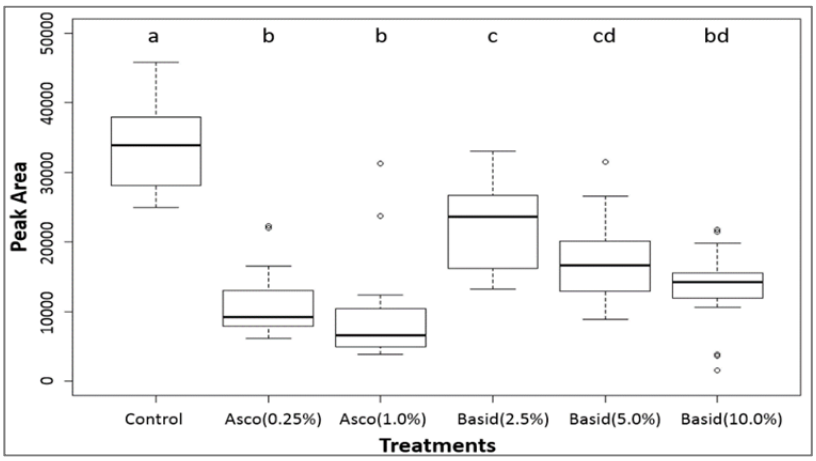

Figure 3 Boxplots of peak area for control and treatments containing various levels (w/w) of ascomycete (Asco) or basidiomycete (Basid) fungal materials (n=20). Different letters denote significantly different mean values (α=0.5)

Conclusion & Significance

To our knowledge, this is the first work demonstrating the ability of fungi to sorb the greenhouse gas CH_4. While this work is preliminary, and undoubtedly more tests must be run to verify and expand on the results, these findings suggest microbes other than CH_4 oxidizing methanotrophs may govern rate limits effecting the ability of biofilters to mitigate CH_4. If fungi can improve CH_4 capture in a biofilter, thus facilitating subsequent oxidation, it is possible that by selecting for specific fungi in a biofilter a potent greenhouse gas can be mitigated simultaneously with odorous livestock production emissions.

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