Supporting Information

Biomass Composting with Gaseous Carbon Dioxide Capture

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Materials and Methods

Composting Feedstock

A representative compost feedstock with a C:N ratio of approximately 20:1 and moisture content of 62% is used in the composting experiments. The compost recipe for each bioreactor includes active compost from a commercial composting operation consisting of food waste, animal bedding, and landscaping waste (35.55g), rice (37.01g), fish meal (1.81g), soybean meal (1.82g), lard (1.98g), and wood shavings (4.82g); masses provided are on a dry weight basis. This particular feedstock is derived and adapted from prior published research on composting experiments and is used to ensure comparisons between conditions can be made with confidence.[1] The bioavailability of the composting recipe used in this study is similar to that of published studies on small and large scale composting.[2-4]

Composting Reaction and Gas Analysis

The 500mL composting reactors are custom-made with a series of sensors and controls. Conditions are run in duplicate. For each reactor, a computer continuously records the CO_2 concentration, O_2 concentration, pressure, temperature, and relative humidity of each reactor headspace. Due to the small scale of the reactor, temperature control is required to maintain 35°C during the composting reaction. When the O_2 concentration decreases below the low set point, a solenoid valve automatically opens to capture high purity CO_2 gas from the headspace and inject fresh oxidant into the reactor until the high set point is achieved. The intermittent, brief cycling of gas ensures the reactors do not lose much moisture. Each 500ml composting reactor is loaded with 205 grams of wet biomass feedstock and is allowed to compost for 15 days. A portion of the reactors operate under pressurized conditions (3 psig), while the remainder operate at atmospheric pressure (0 psig). The temperature, pressure, and relative humidity are measured using an Ankom RF sensor. The oxygen concentration is measured using a DFRobot electrochemical oxygen sensor. The CO_2 concentration is measured using a Sensirion thermal conductivity CO_2 sensor. The oxidant is either air, oxy-fuel (30 vol% CO_2 and 70 vol% oxygen O_2), or oxygen (100 vol% O_2).

16S and Internal Transcribed Spacer (ITS) rRNA sequencing

Samples are processed and analyzed targeting the 16S/ITS Amplicon sequence.[5,6] Full service including DNA extraction, library preparation, sequencing and bioinformatics pipeline is performed by Zymo Research's Microbiomics Services (Irvine, CA). DNA extraction is performed using the ZymoBIOMICS-96 MagBead DNA Kit®. Libraries are prepared with the Quick 16S/ITS NGS Library Prep Kit® targeting the V3-V4 region and ITS2, respectively. PCR reactions are performed with real-time PCR to control cycles and limit PCR chimera formation. Final library is cleaned up with the kit Select-a-Size DNA Clean & Concentrator®, then quantified with TapeStation (Agilent Technologies, Santa Clara, CA) and Qubit (Thermo Fisher Scientific, Waltman, WA). A microbial community standard (ZymoBIOMICS Microbial Community®) is used as the positive control for each DNA extraction and each library preparation. Negative controls as blank extraction control, blank library preparation control are included. Final library is

sequenced using Illumina NextSeq 2000® with 30% PhiX spike in. The 16S rRNA sequencing data were used to conduct a Phylogenetic Investigation of Communities by Reconstruction of Unobserved States (PICRUSt2) analysis, which allowed us to understand whether or not the proper enzymes were present for CH4 and N2O production.

Results and Discussion

Minimum thermodynamic work

Table S1. Minimum thermodynamic work for CO_2 separation from three gaseous streams. Calculations assume final concentration of 99.9%, capture rate of 99%, and gas temperature of 25C.[7]

	Feed CO_2 concentration (mol%)	Minimum Energy Demand (kJ/kgCO2)
Direct air capture	0.04%	494
Air-fed composting + CO2 capture	18%	144
Oxy-fuel composting + CO2 capture	95%	10

Bioreactor Gas Composition

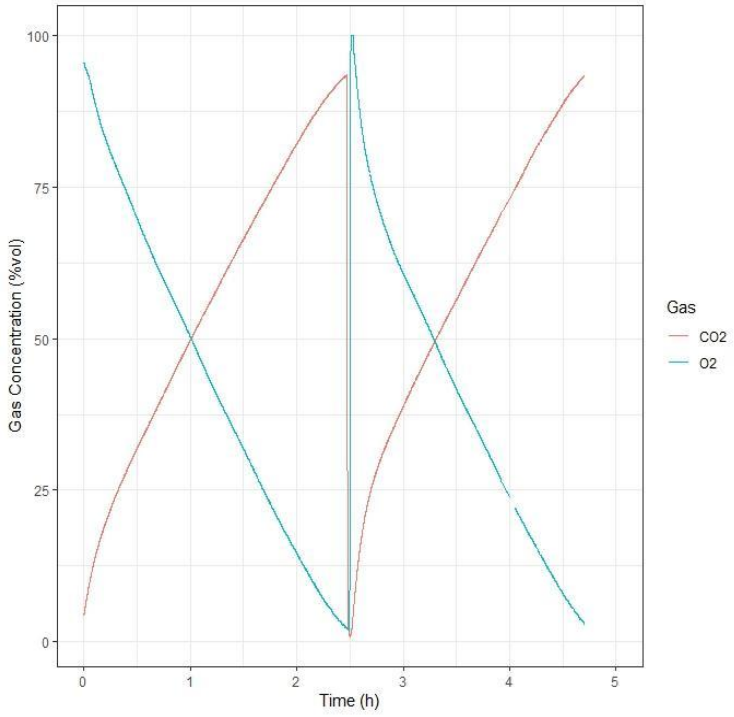

Figure S1. Experimental bioreactor gas composition data demonstrating the process of achieving high purity biogenic CO_2 using pure oxygen. Data collected over a 5-hour period once reactor is at steady state.

Phylogenetic Investigation of Communities by Reconstruction of Unobserved States (PICRUSt2)

16S rRNA sequencing data were used to conduct a Phylogenetic Investigation of Communities by Reconstruction of Unobserved States (PICRUSt2) analysis, which allowed us to understand whether or not the proper enzymes were present for CH4 and N2O production.[8,9] More specifically, we quantified the relative abundance of genes mcrA and pmoA for the CH4 synthesis pathways and amoA, narG, nirK, nirS, norB, and nosZ for the N2O synthesis pathways. Annotation showed missing genes for both pathways, thereby indicating a lack of CH4 and N2O-producing microorganisms, as shown in Figure S2. The GC data corroborated lack of CH4-producing microorganisms, as discussed in the main text. Figure S3 shows the functional genes involved in denitrification to form nitrous oxide, of which only 1 gene was idenstied as present via the PICRUSt2 analysis.

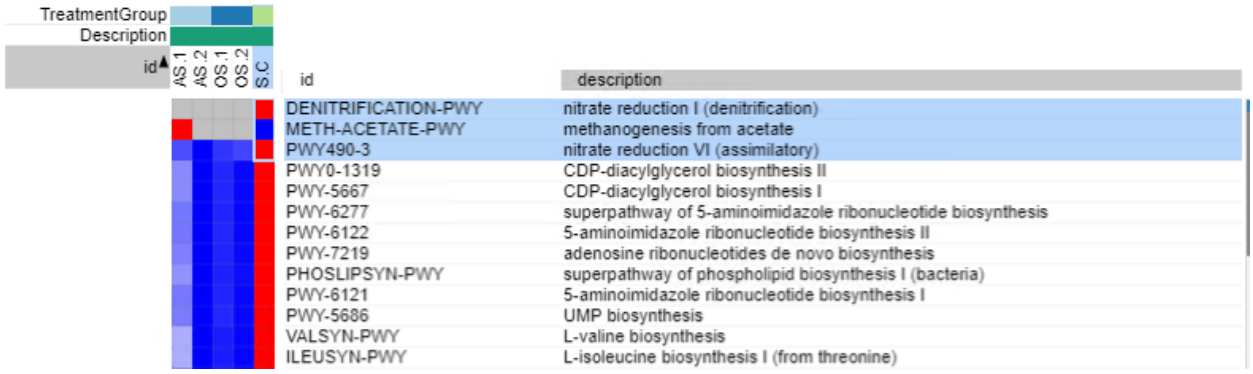

Figure S2: Predicted Pathways heatmap with PICRUSt2 (Phylogenetic Investigation of Communities by Reconstruction of Unobserved States) from 16S rRNA**.**

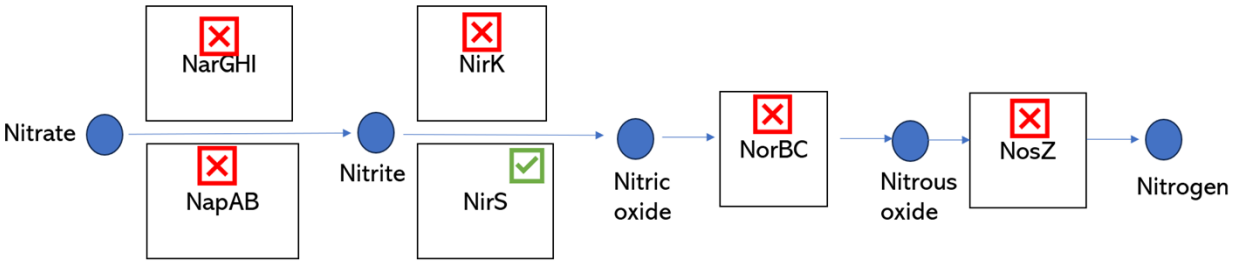

Figure S3: Functional genes involved in denitrification to form nitrous oxide.[10]

Relative abundance

The 16S data were assessed for relative abundance of bacteria, as shown in Figure S4. As can be seen in Figure S4, Leuconostocaceae (genus Bacillus) is the most abundant in the initial mix of food waste and compost obtained from a commercial composting system, and Bacillaceae (genus Bacillus) is the 2nd most abundant. Clostridiaceae (genus Clostridia) is the third most abundant in the initial mix and its relative abundance decreases under higher CO_2 environments in the air and oxy-fuel reactors. However, the relative abundance of Ruminococcacae (genus Clostridia)

increases and is the 2nd most abundant in the air and oxy-fuel reactors, with Bacillaceae (genus Bacillus) being the most abundant. And Leuconostocaceae (genus Bacillus) is not present in the air and oxy-fuel reactors. In summary, various strains of Bacillus and Clostridia are abundant under all conditions with varying levels of abundance, and future work should aim to better understand the mechanistic differences.

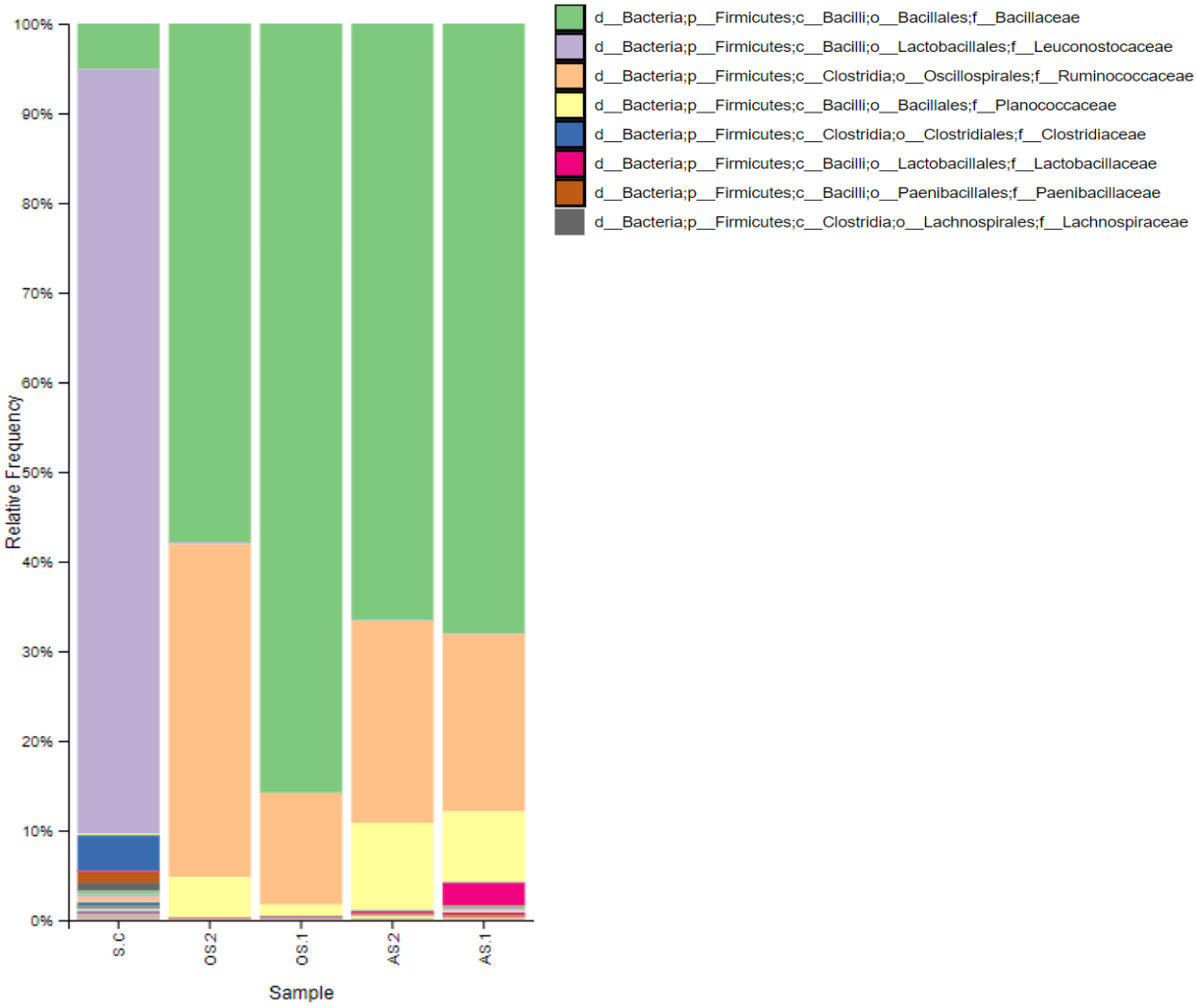

Figure S4: Relative abundance of bacteria at taxonomic level for compost samples: SC (untreated initial mix of food waste and compost from traditional composting operation), OS (compost after 15 days treated via oxy-fuel as oxidant), and AS (compost after 15 days treated via air as oxidant)

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