SUPPORTING INFORMATION

Environmental fate of ^{14}C radiolabeled 2,4-dinitroanisole (DNAN) in soil microcosms

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Summary:

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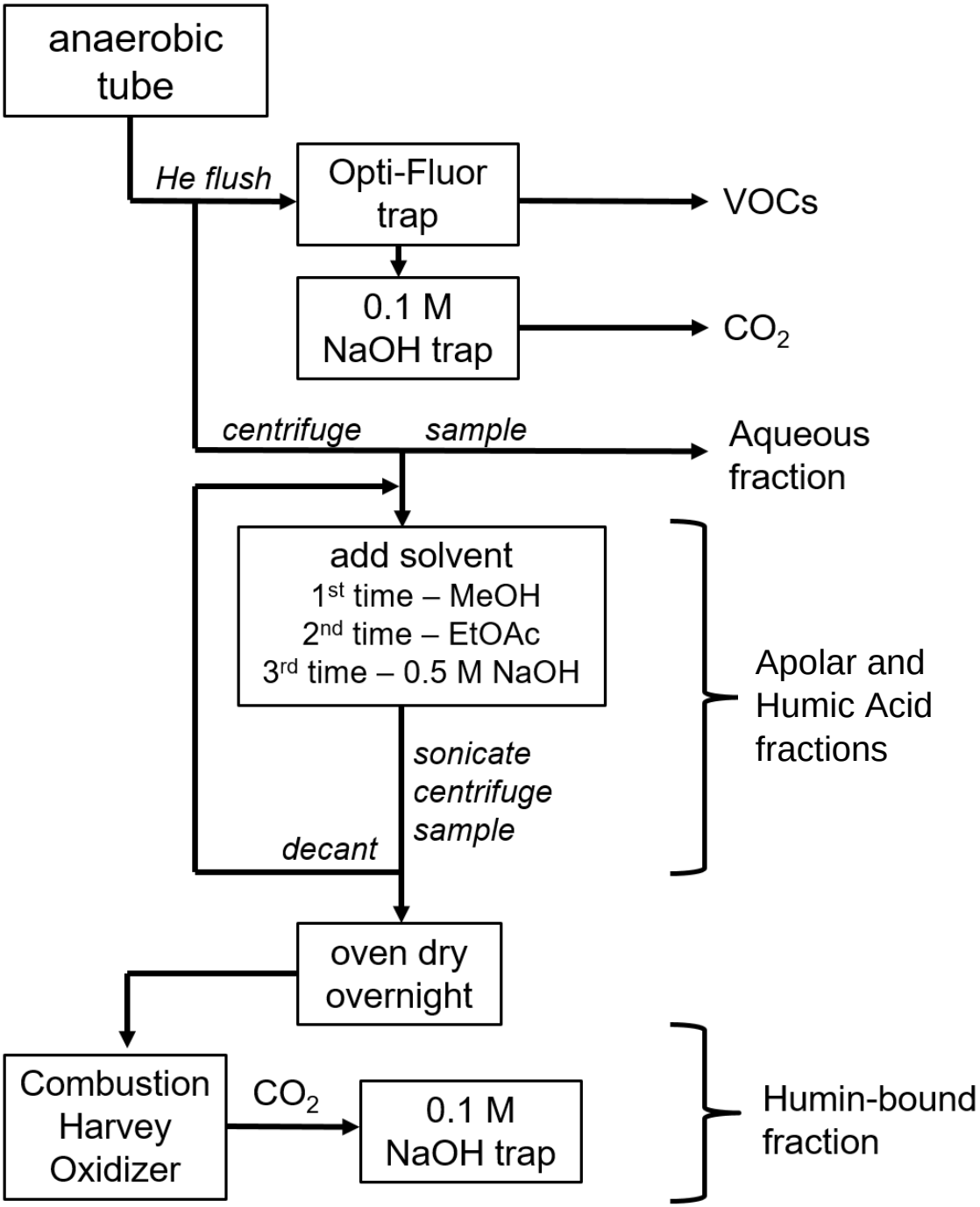

Figure S1. Schematic of ^{14}C label recovery from different fractions after DNAN (bio)transformation: gaseous and volatile species (VOCs, CO_2), aqueous (fulvic acids and water-soluble products), sequential liquid extractions with methanol (MeOH) and ethyl acetate (EtOAc) (apolar fraction), 0.5 M NaOH extraction (humic acids), and combustion in Harvey Oxidizier (unextractable bound residues).

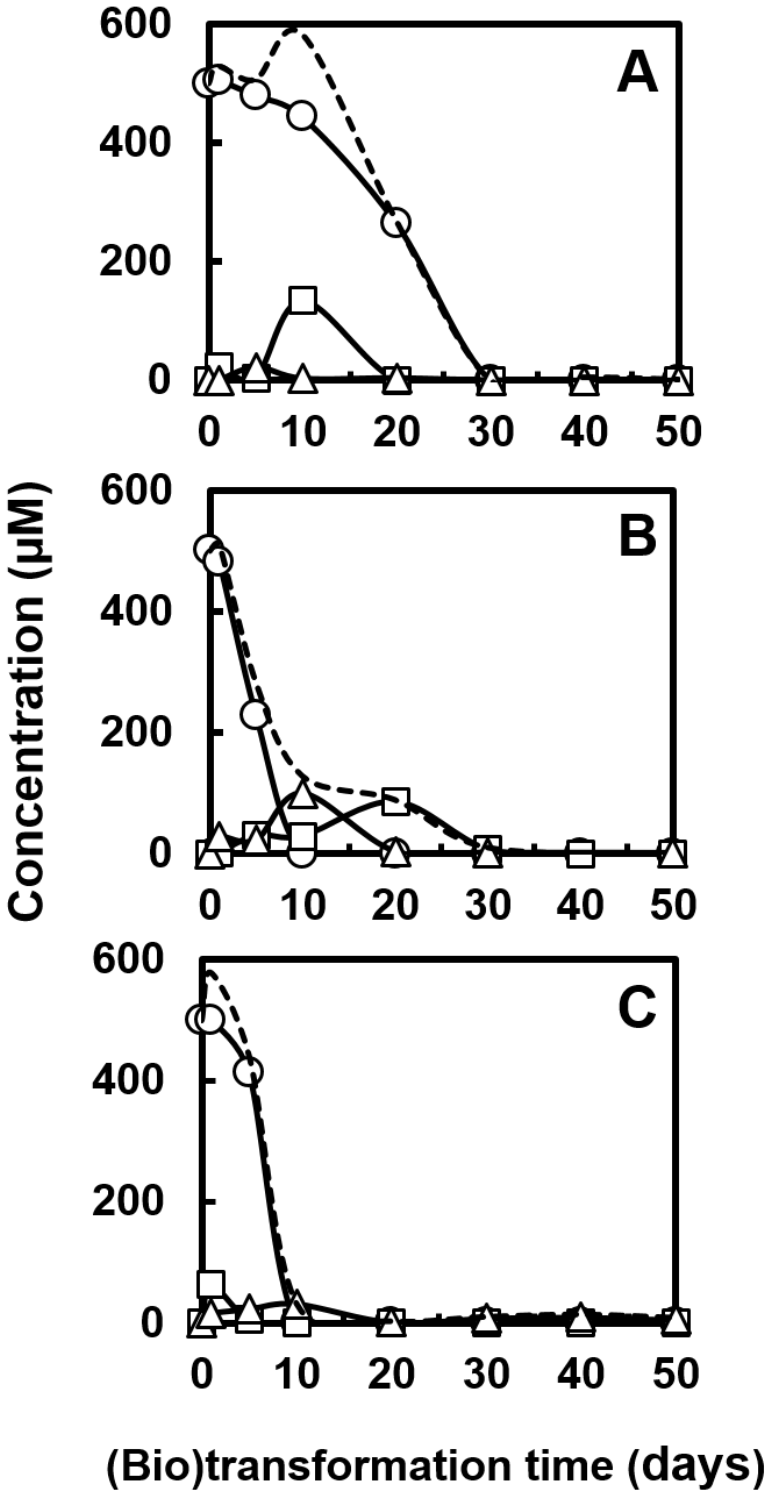

Figure S2. DNAN (○) anaerobic (bio)transformation to MENA (□) and DAAN (Δ) in Camp Butner soil (A), Camp Navajo soil (B), and Mahou anaerobic sludge (C) in parallel non-labeled microcosms. The incubation media, inocula, and conditions were the same as the labeled experiments (Section 2.2.1.)

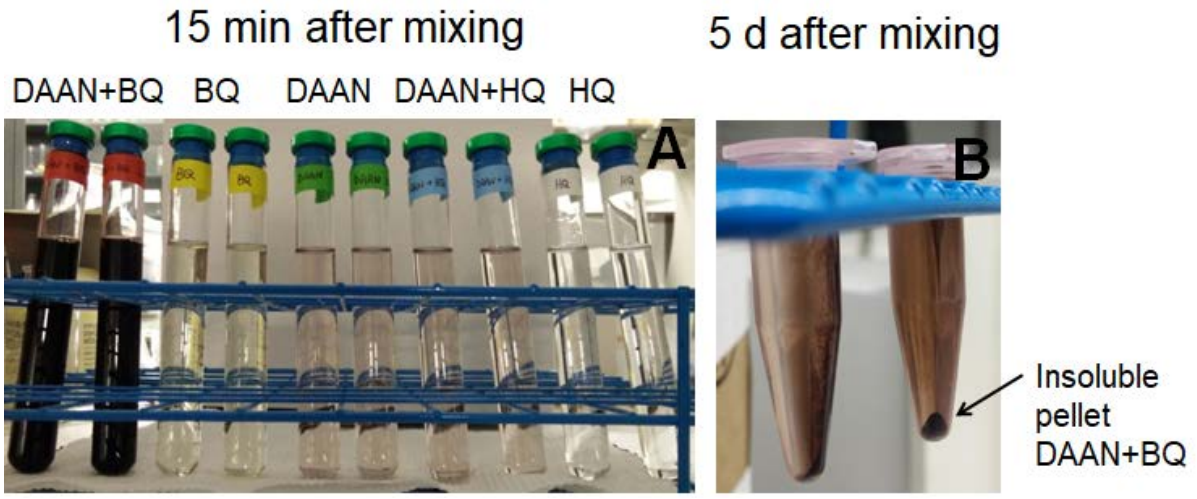

Figure S3 Visual appearance of treatments and controls in the DAAN+BQ pairing experiment. **A)** Dark color is formed immediately after reacting DAAN with BQ, the photo is taken 15 minutes after mixing. **B)** Insoluble pellet can be readily seen after 5 d in the DAAN+BQ treatment. Neither color nor insoluble pellet was observed in the controls. **Abbreviations**: DAAN = 2,4-diaminoanisole; BQ = 1,4-benzoquinone; HQ = 1,4-hydroquinone. **Legend:** DAAN+Q = incubation of DAAN and BQ together (paired); DAAN+HQ = incubation of DAAN and HQ together (paired); DAAN, BQ and HQ indicate each chemical incubated alone, respectively.

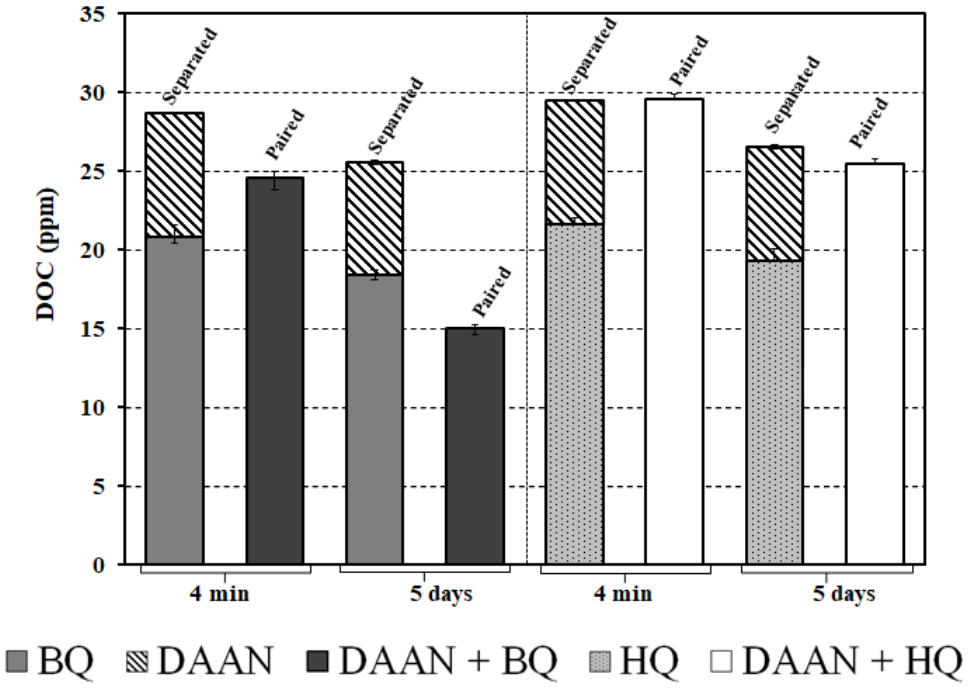

Figure S4 Dissolved organic carbon (DOC) after 4 min. and 5 d of incubation of DAAN+Q treatment in comparison to control experiments (DAAN only, BQ only HQ only and DAAN+HQ). Paired means the two chemicals were incubated together and DOC was measured in the mixture. Separated means the two chemicals were incubated separately and DOC of the two were measured separately and are summed in the stack bar. The results indicate that pairing DAAN+BQ caused loss of DOC.

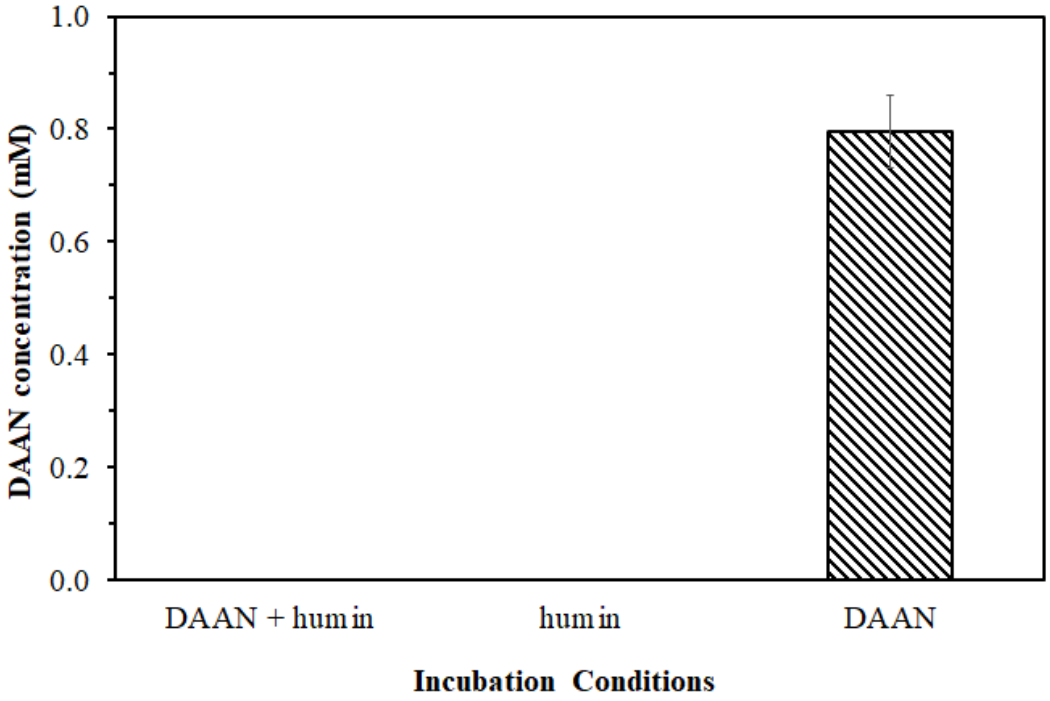

Figure S5. DAAN concentration 4 minutes after incubating it with 20 mg of humin in 20 mL. Controls with DAAN lacking humin and humin lacking DAAN were also measured. DAAN was non-detectable in the DAAN+humin treatment and the humin only control.

Methods.

UHPLC-DAD for Detection of DAAN, BQ, and HQ in DAAN+BQ pairing Experiment
Samples were analyzed using an ultra-high performance liquid chromatograph coupled to a diode-array detector (UHPLC-DAD, Agilent 1290 Infinity, Santa Clara, CA, USA). 10 µL sample injections were separated using an Inertsil ODS-SP column (4.6 mm x 250 mm, 5 µm; GL Sciences, Torrance, CA) at room temperature. An inline guard column was used to remove any particles remaining after centrifugation and supernatant harvest.

To analyze samples from the experiment pairing DAAN and BQ, the mobile phase was run at a flow rate of 1 mL/min for 25 min and was isocratic (85/15 %v/v H_2O/acetonitrile). The compounds were analyzed at the following wavelengths and had the following retention times: 2DAAN (300 nm, 16 min), BQ (290 nm, 9.8 min), and HQ (290 nm, 6.5 min). All analyzed compounds compared with standards with respect to retention times and UV-Vis spectra.

DOC
DOC analysis were performed using a Shimadzu total carbon analyzer V_{CSH} (Columbia, MD, USA). Solids were removed from the samples by centrifugation and the pH was adjusted to 2 using concentrated hydrochloric acid (HCl). The dissolved carbon was then combusted at 680°C and analyzed. A calibration curve using potassium hydrogen phthalate concentrations in the appropriate range was generated prior to each session of sample measurement.