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Comparison of soil biosolarization with mesophilic and thermophilic solid digestates on soil microbial quantity and diversity

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ABSTRACT

Soil biosolarization (SBS) is a pest control technique that combines passive solar heating and fermentation of amended organic matter. The extreme soil conditions generated during SBS could decrease microbial biomass and restructure the soil microbiome, which could impact soil quality. Digestates from anaerobic digesters may harbor microbial communities tolerant of the oxygen, moisture, and temperature stresses encountered during SBS as these conditions may also occur in digesters. Digestate microbial communities may contribute to soil fermentation during SBS and affect organic matter turnover in soils treated with SBS. The objective of this study was to assess the effect of SBS on soil microbial diversity and quantity when solid digestates from thermophilic (TD) and mesophilic (MD) anaerobic digesters were used as soil amendments. In the soils amended with TD, communities showed the greatest divergence from the initial soil state whereas MD amendment resulted in a microbiome more similar to the non-amended soil. The microbial biomass of the biosolarized soils was significantly greater than the non-amended, solar-heated soil. The microbial biomass in the biosolarized soils was dominated by K-strategic or "native" species. Solar heating of the non-amended soil mainly affected "native" species, leading to conditions where other opportunistic species become more dominant. Further studies are needed to elucidate whether the persistent microbes in the soil are benign or pathogenic and to understand their roles in pest inactivation and nutrient cycling during and following SBS.

1. Introduction

Soil fumigation is an important agronomic practice in the production of many high-value vegetable and fruit crops. Traditional soil fumigants used to eliminate pathogens and weed seeds in agricultural soils, such as methyl bromide, are harmful for the environment and humans. Alternative soil fumigants such as chloropicrin or 1,3-dichloropropene present less risk to the ozone layer (Ajwa et al., 2013), but still present health concerns for humans, making these fumigants undesirable and especially dangerous for urban farms (Sanchez-Moreno et al., 2009). Moreover, these fumigants do not discriminate between undesirable pests and beneficial microorganisms (Momma, 2015). It is therefore necessary to find alternative, sustainable ways of controlling soilborne pests.

Soil biosolarization (SBS) can be a sustainable soil pest control technique as it avoids the use of synthetic pesticides. SBS has successfully inactivated fungal, nematode, insect, and weed pests (Bonanomi et al., 2008; Yao et al., 2016). SBS is a combination of soil solarization, where moist soil is covered with a transparent tarp to increase the soil temperature via passive solar heating (Katan et al., 1976), and anaerobic soil disinfestation (ASD) where soil is amended with organic matter prior to tarping to promote anaerobic microbial activity (Lamers et al., 2010). The addition of organic matter can enhance pest inactivation through several mechanisms. First, the additional microbiota and nutrient source associated with the amendment can enhance soil heating through biological heating. For example, amending soil with

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compost and organic matter yielded temperature increases between 2 and 5 °C greater than non-amended soil when both were solar heated (Achmon et al., 2015; Simmons et al., 2013, 2016). Secondly, anaerobic microbial fermentation of organic matter can result in the production of organic compounds such as volatile fatty acids (VFAs) that accumulate due to the physical barrier of the plastic tarp. VFAs may accumulate to levels that result in a decrease in soil pH and increase in toxicity to soil biota and weed seeds (Achmon et al., 2017; Gamliel and Stapleton, 1997; Huang et al., 2015a; Katase et al., 2009). Thirdly, microbiota may directly affect pest organisms through competition, such as between beneficial fungi and pathogenic fungi, or infection, such as microbial degradation of the seed coat of weed seeds (Huang et al., 2015b; Rokhbakhsh-Zamin et al., 2011). The combined thermal, biochemical and ecological action of the organic amendment in SBS can significantly increase the efficiency of the process, counteracting the need for high solar radiation and long treatment durations of up to 6 weeks, which can infringe on the most productive time of the growing season for farmers.

The soil microbiome plays an essential role in maintaining soil fertility. Soil biosolarization will likely affect soil microbial biomass and diversity although these impacts have not yet been quantified. It has been reported that solar heating of the soil (i.e., solarization) is beneficial to soil microflora because it stimulates fluorescent *Pseudomonas* spp. (Gamliel and Katan, 1991). Soil solarization has also been shown to have favorable effects on soil microbiota as evidenced by increased amino acid synthesis (Chen et al., 2000). Significant changes in the microbial diversity within the soil profile have also been observed after soil solarization or soil biosolarization (Simmons et al., 2014, 2016). Specifically, a significant decrease of the phylum Firmicutes was observed in solarized soils, particularly at greater depths. Additionally, a significant increase of bacteria from the phylum Proteobacteria was observed with increasing soil depth in biosolarized soils. These shifts were attributed to temperature gradients established during solarization and changes in the composition of the soil aqueous and gaseous phases.

Soil temperatures above 50 °C are considered to be lethal to most soil-borne pathogens and most mesophilic microbes (Stapleton, 1996) and temperatures around 60 °C have been shown to significantly decrease microbial metabolism and survival in general (Palese et al., 2004). For instance, it has been observed that soil solarization decreased microbial activity as well as the activities of phosphatase and β-glucosidase enzymes in different solarized plots compared to non-solarized controls (Scopa et al., 2009).

Elucidating the impact of SBS on soil biological activity and microbial community structure is important for understanding post-treatment implications for agriculture. The effects of biotic and abiotic stress on soil organisms can be assessed by measuring changes in biological activity, microbial biomass, soil respiration and enzyme activities (Scopa and Dumontet, 2007). In addition, active microbial soil biomass and diversity can be used to understand microbial decomposition of soil organic matter (SOM), a critical element of the soil phytonutrient cycle (Stenstrom et al., 1998).

The objective of this study was to assess the composition and activity of microbial communities in biosolarized soils amended with two different solid digestates from anaerobic digestion of mixed organic wastes. Like compost, which has been shown to be an effective inoculum for biosolarization (Simmons et al., 2013), digestates contain robust, anaerobic, organic matter-degrading microbial communities that may also tolerate biosolarization. Studies have shown that SBS can increase the proportion of facultative and obligate anaerobic microorganisms in the soil (Yao et al., 2016). As a result, digestate amendment could influence soil microbial community restructuring during biosolarization and help prime the soil with active bacteria following treatment. This could be important for occupying soil niches that pathogens may otherwise recolonize. Furthermore, enriching the soil with biomass-degrading bacteria from digestate could benefit nutrient cycling in the soil. In this study, soil microbial biomass following biosolarization was estimated using substrate-induced respiration (SIR) with measurement of the respiration response kinetics (Anderson and Domsch, 1978; Panikov and Sizova, 1996; Stenstrom et al., 1998). These data can provide information on the physiological state of the microbial biomass by estimating the ratio of growing (r-strategic) versus non-growing (K-strategic) (Chen et al., 2012b). The taxonomic diversity of microbial communities was also analyzed via next generation 16S rRNA gene sequencing. These results will help gauge the valorization potential of digestates in agricultural to improve soil microbial activity and diversity as well as provide guidelines for application of digestate in biosolarization.

2. Materials and methods

2.1. Soil and digestate description

Dry topsoil (Hanford sandy loam) was collected from the 0–15 cm depth range at UC Kearney Agricultural Research and Extension Center (KARE) in Parlier, CA (36.6_N; 119.5_W; elevation 97 m a.s.l.), sieved through a 2 mm screen and stored at room temperature (S). The contents of organic matter, sand, silt and clay were 0.015 g g^{-1}, 0.41 g g^{-1}, 0.37 g g^{-1} and 0.22 g g^{-1}, respectively.

Two solid digestates from two anaerobic digesters with different operational conditions and ofeedstocks were used in the experiment. A thermophilic digestate (TD) was acquired from an anaerobic digester located on the University of California, Davis campus in Davis, CA. The UC Davis digester processes mixed organic waste (food, agriculture, and green wastes). The digester utilizes sequential thermophilic hydrolysis and methanogenesis (55 °C) with low solids loading (5–10% total solids in the methanogenesis phase). The solid digestate was periodically separated from the liquid phase of the methanogenic sludge and dewatered by pressing. The Yolo County Landfill (Woodland, CA) provided a mesophilic digestate (MD) from anaerobic digestion of food, manure and green wastes. Digestion occurred under high solids loading (40–60% of moisture content) and mesophilic conditions (35 °C). Both digestates were air-dried, ground and sieved (< 2 mm) prior to mixing with the sampled soil. The total N of the soil, the TD and the MD amendments was 0.04, 1.48 and 1.03%, respectively. The total C of the soil, the TD and the MD amendments was 0.38, 47.10 and 41.53%, respectively.

2.2. Soil mesocosm preparation

Soil mesocosms served as experimental units in field studies as described in previous studies (Achmon et al., 2017; Simmons et al., 2013). Soil mixtures for mesocosms were prepared by amending dry soil with dry thermophilic (STD) or mesophilic digestate (SMD) to achieve 1.5% loading (dry weight basis). Soil without amendment was used as a control (S). Soil mixtures were wetted to their respective field capacities and allowed to incubate overnight at 4 °C so that moisture could equilibrate between the various soil components. Equilibrated soil mixtures were packed into 3.8 l black plastic grow bags (neHydro, Southampton, MA). The bags contained drainage holes to facilitate moisture and gas exchange with the surrounding soil. Compact temperature sensors and data loggers (Thermochron iButtons model 1922L, Embedded Data Systems, Lawrenceburg, KY) were embedded in the center of each microcosm at 15 cm depth. The diameter and height of the filled mesocosms were 17.8 cm and 22.5 cm, respectively.

2.3. Field experiment

The field site was also located at KARE and it was prepared as previously described (Achmon et al., 2017; Simmons et al., 2013). Each field plot measured 1.8 × 8.5 m and contained one mesocosm from each treatment and the arrangement of mesocosms was randomized.