Applied Soil Ecology xxx (xxxx) xxx–xxx

Contents lists available at ScienceDirect

Applied Soil Ecology

journal homepage: www.elsevier.com/locate/apsoil

Long and short term effects of solarization on soil microbiome and agricultural production

Housam Kanaan[a,b], Sammy Frenk[a,c], Michael Raviv[b], Shlomit Medina[b], Dror Minz[c,*]

[a] Department of Plant Pathology and Microbiology, Robert H. Smith Faculty of Agriculture, Food and Environment, The Hebrew University of Jerusalem, Rehovot 76100, Israel

[b] Institute of Plant Sciences, Agricultural Research Organization, Newe Ya'ar Research Center, Ramat Yishay 30095, Israel

[c] Institute for Soil, Water and Environmental Sciences ARO, Volcani Research Center, 68 HaMacabim Rd. P.O Box 15159, Rishon Lezion 7528809, Israel

ARTICLE INFO

Keywords:
Solarization
Compost
Bacterial community
Fluorescein diacetate (FDA)

ABSTRACT

Soil solarization, a method of chemical-free pest treatment, is a practical and cost-effective way to treat organic farming soil. This method uses polyethylene sheets to capture solar irradiation that heats the soil. Together with heat generated during composting of fresh organic matter, this method can effectively treat many soil-borne pathogens. In this study we examined the effects of solarization with the addition of compost at three maturation levels, on soil microbial community structure and function along with plant performance. Similar hydrolytic activity among the treatments (as measured by FDA hydrolysis) was measured in the first year of solarization. High level of activity was obtained in soil amended with immature compost at the beginning of the second year, suggesting residual (carry-over) effects of previous-year treatments. Amplicon sequencing of 16S rRNA encoding genes was used to study soil bacterial community structure. Diversity and richness of bacterial communities were found to be negatively affected by solarization in all treatments. Interestingly, bacterial communities of solarized soils clustered together, regardless of compost amended and type, separated from the cluster of communities from non-solarized soils of all amendments. Specifically, long and short terms negative effects of solarization on the relative abundance of Alphaproteobacteria, Acidobacteria and Actinobacteria were noticed. In contrast, solarization positively affected Bacilli and Gammaproteobacteria abundance in both long and short term. Furthermore, solarization had long and short term positive effects on productivity of eggplant and wheat plants. This study is the first to describe in high details the combined effects of solarization and amendment of composts of different maturation levels.

1. Introduction

Soil solarization is a method of pest treatment in which polyethylene sheets are used to capture solar irradiation causing soil heat up. Solarization is commonly used as a physical method to eradicate or reduce the pathogenicity level of soil-borne pathogens (SBPs), pests and weeds. This method is considered effective, inexpensive and has few residual side effects (Katan et al., 1976; Candido et al., 2011). Soil solarization is effective for reducing the number of heat-sensitive SBPs, such as *Verticillium dahliae*, *Sclerotium rolfsii* and *Fusarium*. A previous study reported that solarization is an effective method to reduce the rate of disease incidence and improve nutrient balance (Greenberger et al., 1987; Grunzweig et al., 1999).

Application of organic matter to soil, solarization or the combination of both cause physical, chemical and biological changes. Soil-applied compost has more thermophilic bacteria than other soil amendments (Bulluck et al., 2001). Previous studies performed on soil in which lettuce plants were grown, showed that solarization contributed to higher levels of nutrients available to crops (Gamliel and Stapleton, 1993a, 1993b). Furthermore, previous studies described a reduction of microbial activity during solarization of compost-amended soil, resulting from heat production and toxicity of volatile compounds (caused by degradation of organic compounds), leading to the development of partial biological vacuum along with shifts in bacterial and fungal communities (Gamliel and Stapleton, 1993b; Gamliel et al., 2000; Culman et al., 2006). In addition, Gamliel and Stapleton (1993a, 1993b) showed recolonization of the rhizosphere by fluorescent *Pseudomonas* and *Bacillus* spp. after solarization treatment. Combining both solarization and compost amendment may yield a synergistic effect in comparison to each treatment alone. There is existing evidence that increased heat beneath the polyethylene tarps cause higher decomposition rates of organic compounds in soil and a consequential release

* Corresponding author. Tel.: 972-3-9683316, 972-50-6220316; fax: 972-39604017.
E-mail address: minz@volcani.agri.gov.il (D. Minz).

http://dx.doi.org/10.1016/j.apsoil.2017.10.026
Received 15 March 2017; Received in revised form 17 October 2017; Accepted 22 October 2017

of volatile chemicals such as alcohols, aldehydes, sulfides and isothiocyanates that may play a role in eradicating various microorganisms (Gamliel and Stapleton, 1997; Gamliel et al., 2000). A previous study comparing bacterial communities in solarized and non-solarized soil reported that clusters of bacteria were grouped according to these treatments (Gelsomino and Cacco, 2006). The main effect of solarization on microbial communities is the reduction of bacterial density and increased abundance of thermo-tolerant bacteria (Simmons et al., 2014). In addition to the main effect of solarization, it was also evident that the density of fluorescent *Pseudomonas* increased by up to 130 fold (Gamliel and Katan, 1991). van der Voort et al. (2016) recently reported reduction in soil disease suppressiveness as a result of heat treatment. Another recent study documented a sharp reduction in microbial activities, measured as respiration rate, heat generation and dehydrogenase activity, as a result of soil solarization (Kanaan et al., 2016). Although thermophilic organisms may act within heated soil and degrade incorporated organic matter (Simmons et al., 2013), the majority of soil microorganisms optimally operate at lower temperatures. Microorganisms inhabit different niches in the soil resulting in high diversity of approximately 10^4 species per gram of soil (Curtis et al., 2002; Griffiths and Philippot, 2012) with diverse functional capabilities and optimal growth conditions. The high diversity and richness were shown to be affected by ecosystem type; soil texture, pH and nutrient balance were found to be the main factors determining these variables (Li et al., 2014). Further environmental conditions determining community compositions include soil disturbance by high temperatures caused during the solarization process. These processes modify the taxonomic composition of bacterial communities, whereas richness and evenness were similar (Simmons et al., 2014). Despite soil microbiology being the focus of vast research, particularly regarding consequences of disturbances, the combined effect of solarization with compost amendment has received less attention. The present study assessed changes in bacterial community composition in solarized soil combined with compost at three maturation levels (relative to non-solarized and unamended soil) by measuring the distribution and abundance of 16S rRNA gene amplicons. This study provides new insights into the dynamic changes occurring in the soil microbiome as a result of the described solarization process.

2. Materials and methods

2.1. Experimental system

The field experiment was conducted at Newe Ya'ar Research Center, in the Jezre'el Valley, Israel, starting 2010 and sampled in the summer of 2011. The experiment was conducted in Chromic Haploxerert (fine-clayey, montmorillonitic, thermic) soil under typical Mediterranean region climate: cool winters (average minimum of 8–9 °C) and warm dry summers (average maximum of 28–29 °C). Physicochemical characteristics of the studied soil (0–30 cm depth) were determined as described by Sharma et al. (2017): cation exchange capacity (CEC) was 65 cmol kg^{-1}, electrical conductivity (EC) was 0.18 dS m^{-1}, pH 7.36, soil organic carbon (SOC) was 1.23% and total nitrogen (TN) was 0.11%. The contents of $CaCO_3$ was 13%, sand – 19%, silt – 18% and clay – 64%. The average annual precipitation is about 530 mm, concentrated mostly between November and March. The experiment duration was eight weeks in total, four weeks of solarization followed by an additional recovery period of four weeks. Eight treatments in three replicates were arranged in a randomized blocks design. The total area (main plot) was 600 m^2, whereas every sub-plot (one replicate) was 18 m^2 to a total of 24 sub-plots.

Composts were prepared in insulated 5 m^3 bins equipped with forced aeration at 400 m^3 h^{-1} air per bin, used to prevent overheating and occurrence of anaerobic conditions, as described previously (Raviv et al., 1998, 2005). Compost feedstock mixtures consisted of 90% cattle manure and 10% wheat straw (on a dry weight basis). The temperature set point was 60 °C at a depth of 50 cm, assuming that these temperatures are sufficient to kill all human pathogens (Berry et al., 2013).

The composts were mechanically mixed several times in order to homogenize the temperature and aerate. Mature, partially mature, and immature composts, were taken at 182, 70, and 32 days of composting, respectively. More details about compost characteristics can be found in Kanaan et al. (2016).

The resulting composts were applied (6 kg DW/m^2) according to their degree of maturity and incorporated at a depth of 20 cm at a volumetric ratio of 3%. The solarized soil was covered using a 0.1 mm thick polyethylene sheets (Anti drip, "Ginegar products", Israel) on four treatments which included an unamended control and three maturation levels of compost. Accordingly, non-solarized soil included four treatments as for the solarized soil. In both solarized and non-solarized soil, moisture was maintained at 70% of field capacity by weekly measurements and adjustments during the entire experimental period. The required soil moisture was achieved by using two irrigation systems; one for the solarized and another for non-solarized plots.

Solarized soil containing compost from the previous year was grinded using a mechanical grinder and sieved through a 2 mm mesh. Samples of 120 gr soil were mixed with fresh compost of different maturation levels according to the experimental design as described by Kanaan et al. (2015). These samples were put in nylon mesh bags, buried and incubated in the field at 10 cm depth (Kanaan et al., 2015). Samples were taken at week zero for pre solarization samples and weeks four and eight for post solarization samples.

2.2. DNA extraction and molecular analysis

Genomic DNA was isolated from duplicate soil samples using the MO BIO Laboratories, power soil DNA kit, Inc. (West Carlsbad, CA, USA), catalog number 12888-50/100. DNA extracts were stored at −80 °C until further analysis.

2.3. PCR amplification and sequencing of 16S rRNA gene fragments

Genomic DNA was amplified and sequenced using the Earth Microbiome Project guidelines. The V4 region of the 16S rRNA gene was amplified with region-specific primers that included the Illumina flow-cell adapter sequences, CS1 and CS2 (Green et al., 2015). The primer set, CS1_515 (ACACTGACGACATGGTTCTACAGTGCCAGCMGCCGCGGTAA) and CS2_806R (TACGGTAGCAGAGACTTGGTCTGGACTACHVGGGTWTCTAAT) were used. Amplicons were quantified, normalized and then sequenced in a MiSeq (Illumina, San Diego, CA, USA) run according to procedures described in the supplementary methods of Caporaso et al., 2012. Paired-end sequences were approximately 250 bp in size.

2.4. Sequence analysis

Approximately 11.98 million paired-ends reads were obtained and analyzed using the MOTHUR package version 1.33.3 according to the standard operating procedure (SOP) described at the software package website (http://www.mothur.org/wiki/MiSeq_SOP, 01.08.16). Shortly, paired reads were first merged to contigs and duplicate reads were grouped. The resulting ~6.13 million unique sequences were aligned to the Silva based reference alignment database provided in the software web site. Sequences were then clustered using "pre.cluster" function (1 mismatch for each 100 bp) to reduce sequencing errors (Huse et al., 2010). Suspected chimeric sequences were removed using the Uchime algorithm (Edgar et al., 2011) where frequent reads served as reference for rare reads. The remaining 762,451 unique sequences (representing a total of 5.21 million replicated sequences) were assigned a taxonomy using the greengenes database reference file (DeSantis et al., 2006). Archaeal, Eukaryotic, mitochondria, chloroplast and unknown sequences were removed from the analysis. The sequences were randomly