Short-term effects of different techniques to prepare an organic citrus soil for replanting on its microbiological and biochemical properties

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Abstract

Conversion is a critical period for organic cropping since soil must reach optimal levels of fertility, structure, and biological activity to work properly under organic management. In this work, different management techniques to raise levels of organic matter, to disinfect or to increase soil suppressivity in soil are being evaluated in a replanted organic citrus orchard. These techniques are solarization, use of composted manure (alone or combined with solarization) and incorporation of a disinfectant vegetal cover. Results warn of negative effects of solarization on soil organic matter and biological and enzymatical activity, together with changes in microbial populations mainly because of the temperature increase. Given that the experiment is still ongoing, the duration and final extent of these changes are still being investigated, although it has been seen already that the effects on living organisms are faster to recover than those affecting the organic matrix of soil.

Introduction

Adopting measures to increment soil levels of organic matter and biological activity is key when an agrosystem is to be converted to organic management. Nutrient reserves are increased and more easily mobilized, and soil suppressivity is enhanced. This last aspect is particularly relevant in replantations given that chemical disinfection treatments cannot be used and plantlets are very sensitive.

It is well known that levels of soil organic matter and biological activity are highly correlated, and that soil suppressivity derives from biological mechanisms: ecological competition, release of biocides, and resistance induction in plants. Organic matter is also used directly for disinfection by taking advantage of the volatile compounds released during its degradation, such as ammonia from N-rich residues or isothiocyanates from glucosinolate-rich plants such as brassicaceae. All 'biofumigation' treatments rely on these mechanisms, although relevance of ammonia is limited by the strict limits on N applications in organic farming management. Aiming for higher effectiveness, they are often combined with solarization, although the effects of the heat and derived changes on the biological condition of soil are not fully investigated. A field trial is therefore being carried out to study the effects on soil biochemical and microbiological properties of different techniques to prepare the soil for replanting in a citrus orchard in convertion to organic management. This work discusses the results obtained six months after the application of all treatments was finished.

Material and methods

The field trial is set in a 1.2 ha orchard within an old farm sited at Gandía (Valencia), wherein citrus have been cultivated for more than a century and many *Phytophtora* attacks have been recorded in the last decades of intensive management. Before replanting and converting the orchard to organic management, different techniques to fight against soil-borne pathogens based on disinfection and/or enhancement of soil microbial activity and diversity are being tested:

- untreated control (C).
- solarization (S)
- biodisinfectant vegetal cover (VC): mixture of *Sinapis alba* (at a seed rate of 16.5 kg/ha) and *Brassica carinata*, *B. rapa* and *B. juncea* (at a seed rate of 25 kg/ha). Seed rates were much higher than usual because the experiment schedule imposed a very late seeding and short time to grow before being cut and incorporated to soil.

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- organic amendment (OA): ovine manure at the rate of 16 t/ha (fresh weight)
- solarization + organic amendment (S+OA)

All treatments are evaluated in triplicated experimental subplots, each consisting of eight 1-year old orange trees (*Citrus sinensis* var. *Salustiana*) grafted on Citrange Carrizo rootstock (*C. sinensis* x *Poncius trifoliata*), planted at 6x4 m. Water and nutritional demands of the plantation are being satisfied homogeneously for all trees by means of a fertirrigation system; all fertilizers are certified for use in organic production. For solarization, recently-irrigated soil was covered with 300-gauge transparent polyethylene film and immediately sealed. Soil temperatures reached during the process were monitorized by iButton electronic probes introduced at 15 cm (average sampling depth) just before laying the plastic covers; the data obtained certified successful temperature increases during solarization. All the treatments were finished in early autumn, and a first soil sampling was made; results are discussed in Albiach et al (2013). The following spring 16 subsamples were taken at 0-30 cm depth in each subplot, in both sides of the position the trees would occupy after plantation. All subsamples were carefully mixed into a composite sample per subplot, which was then prepared for biochemical and microbiological analysis. The soil biochemistry parameters studied are: microbial biomass carbon (Vance et al, 1987), alkalyne phosphomonoesterase activity (Tabatabai and Bremner, 1969), phosphodiesterase activity (Browman and Tabatabai, 1978), urease activity (Tabatabai and Bremner, 1972), arylsulphatase activity (Tabatabai and Bremner, 1970), β-D-glucosidase activity (Tabatabai, 1982), N-acetyl-β-D-glucosaminidase (chitinase) activity (Parham and Deng, 2000) and dehydrogenase activity (Casida et al, 1964). The main microbial components in soil determined are: bacteria (YPG agar with cycloheximide, 25ºC, 2 days), fungi (malt agar with antibiotics, 25ºC, 2 days), actinomycetes (Vargas et al, 2009), *Bacillus* sp. (Yi et al., 2012), fluorescent pseudomonas (Simon and Ridge, 1974), denitrifying and nitrifying bacteria (Roux-Michollet et al, 2008), and ammonium oxidation potential (Elsgaard et al, 2001).

Results and Discussion

The effects of the treatments on the soil biochemical parameters (Table 1) were consistent with those found in the first sampling (Albiach et al, 2013), showing a clear, although not always statistically significant, depressive effect of solarization on all parameters but chitinase.

This negative effect is very relevant because it shows that not only living organisms but also the activity of organic components stabilized within the soil organic matrix have been affected, not only by the heat but also by the increased degradation caused by the increment in temperature and moisture at the initial stages of the solarization. Enzyme activity may be very important in soils under organic cropping and its depression may be slower o more difficult to recover from, as the small differences between results obtained after the treatments and six months later show.

Table 1: Effect of the treatments on soil biochemical parameters

Treat	MB	DHA	PA	β -GA	PDA	CHA	UA	ASA
S	151	1,15 a	1,26 a	0,426	0,459 ab	0,105	0,0522 a	0,113 a
CV	311	2,00 b	1,67 b	0,497	0,586 c	0,0955	0,114 b	0,222 b
EO	272	2,24 b	1,64 b	0,613	0,574 bc	0,106	0,122 b	0,229 b
S+EO	112	1,05 a	1,14 a	0,388	0,404 a	0,0965	0,0546 a	0,100 a
C	277	2,11 b	1,75 b	0,651	0,593 c	0,108	0,125 b	0,249 b

MB: microbial biomass (µg C/g), DHA: dehydrogenase activity (µg TPF/g · h), PA: Phosphomonoesterase activity (µmol PNF/g . h), β –GA: β-glucosidase activity (µmol PNF/g . h), PDA: phosphodiesterase activity (µmol PNF/g . h), CHA: chitinase activity (µmol PNF/g . h), UA: urease activity (µmol N-NH4/g . h), ASA: arylsulphatase activity (µmol PNF/g . h). Values followed by the same letter are not significantly different at the 0.05 probability level according to the LSD test. Absence of letters indicates no significant effects.

Regarding the effects on the microbial populations studied in soil (Table 2), the statistical significance of the results was negatively affected by the usually high variability of biological parameters in agricultural soils.

Nevertheless, the changes found immediately after the treatment application (Albiach et al, 2013) have disappeared, suggesting a faster recovery of populations from disturbance, and showing how soil enzymes may be very useful as indicators of changes in soil quality after different management alternatives, since they are more sensitive than modifications in organic matter content but less variable and volatile than purely microbiological parameters.

Table 2. Effect of the treatments on soil microbial populations

Treat	Bacteria (UFC/g)	Fungi (UFC/g)	AM (UFC/g)	FP (UFC/g)	*Bacillus* sp (UFC/g)	AOB (MPN/g)	NOB (MPN/g)
S	$5{,}11 \cdot 10^6$	$1{,}24 \cdot 10^5$	$1{,}42 \cdot 10^6$	$1{,}63 \cdot 10^3$	$1{,}32 \cdot 10^6$ a	$3{,}56 \cdot 10^4$	$4{,}67 \cdot 10^3$
CV	$8{,}00 \cdot 10^6$	$7{,}59 \cdot 10^4$	$1{,}69 \cdot 10^6$	$6{,}54 \cdot 10^2$	$3{,}47 \cdot 10^4$ b	$4{,}05 \cdot 10^4$	$3{,}28 \cdot 10^3$
EO	$5{,}47 \cdot 10^6$	$1{,}06 \cdot 10^5$	$1{,}93 \cdot 10^6$	$1{,}23 \cdot 10^3$	$5{,}97 \cdot 10^4$ b	$3{,}43 \cdot 10^4$	$1{,}33 \cdot 10^4$
S+EO	$5{,}85 \cdot 10^6$	$7{,}67 \cdot 10^4$	$2{,}33 \cdot 10^6$	$8{,}52 \cdot 10^2$	$1{,}65 \cdot 10^6$ a	$7{,}55 \cdot 10^4$	$2{,}20 \cdot 10^3$
C	$5{,}77 \cdot 10^6$	$1{,}39 \cdot 10^5$	$2{,}07 \cdot 10^6$	$1{,}90 \cdot 10^3$	$3{,}20 \cdot 10^4$ b	$3{,}52 \cdot 10^4$	$1{,}23 \cdot 10^4$

AM: actinomycetes, FP: fluorescent pseudomonas, AOB: ammonia-oxidizing bacteria, NOB: nitrite-oxiding bacteria. Values followed by the same letter are not significantly different at the 0.05 probability level according to the LSD test. Absence of letters indicates no significant effects.

Conclusions

Given that soil plays such a central role in organic cropping, every management technique should be checked as for its effects on soil quality. Solarization has been shown to be successful in the fight against soil pathogens, mainly nematodes, but our results suggest that there may be potentially long-lasting negative effects that should be taken into account. Soil organic matter and biological activity are extremely important for soil resilience, nutrient cycling and pathogen suppression, so any negative impact on them is expected to be harmful and should be avoided, especiallly during the conversion period, when their levels are the lowest. Techniques based on active vegetal covers and application or organic amendments are therefore preferable to increase organic matter and biological activity and diversity to build up fertility and resistance to pathogen in new organic farms.

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