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Air-drying pretreatment effect on soil enzymatic activity

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ABSTRACT

Moreira R.S., Chiba M.K., Nunes S.B., de Maria I.C. (2017): Air-drying pretreatment effect on soil enzymatic activity. Plant Soil Environ., 63: 29–33.

Air-drying of soil samples is a common practice for all-purpose soil testing. However, it may cause the cessation of microbial activity changing the biochemical attributes. For this reason, field-moist samples are commonly used in the assessment of the enzyme activity in soils. This practice may, therefore, discourage the use of enzymes in soil quality evaluations. This study evaluated the effects of air-drying on cellulase, arylsulfatase and acid phosphatase activities in soil; the hypothesis was that the activities of these enzymes determined in air-dried soil samples are similar to those obtained at field-moist samples. Soil samples were collected (0–10 cm) in a long-term experiment that received two rates (10 and 20 t/ha) of sewage sludge and mineral fertilizer and was cropped with maize. Collected soil samples were split into two groups. In the first one, the enzymes were determined at field-moist samples, while in the second one, the samples were air-dried before enzymatic analyses. Acid phosphatase was significantly affected by air-drying while the arylsulfatase activity hardly changed. The results showed that the enzymes determined in air-dried soil samples hold the capacity to identify different organic management and can, potentially, be used as soil quality indicators.

Keywords: soil enzymes; agroecosystems; biochemical parameter; soil fertility; moisture; microbial community

The evaluation of soil quality depends on the integration and synthesis of a large number of soil properties and can be used to enhance sustainable management of agroecosystems. A soil quality indicator should elucidate the ecological processes involved, be sensitive to disturbance and, at least ideally, be easy to measure (Doran and Parkin 1994). Microbiological and biochemical parameters like soil enzymes have those characteristics and respond long before other soil quality indicators (Dick and Tabatabai 1993, Bastida et al. 2008). Also, enzymes are good indicators of soil quality because they play an important role in organic matter decomposition and nutrient cycling, integrating information both about microbiological status and soil chemical conditions (Aon et al. 2001).

There is not a consensus whether the enzyme activity, similar to microbial assays, should be measured immediately after the soil sampling. In most cases, to preserve the original characteristics of the samples they are usually kept refrigerated or frozen until processed (Trasar-Cepeda et al. 2000). However, this practice is more common only for scientific work and less plausible for routine soil fertility assessment. In addition, the air-drying gives the soil samples physical conditions that are ideal to be handled and conserved without refrigeration (Zornoza et al. 2006, Lopes et al. 2015). The enzymatic assays are then easier to be handled and more cost-effective.

Moreover, there are indications that soil enzymatic activity in air-dried samples depends on the type of soil enzyme, sampling location and season (Turner and Romero 2010, Abellan et al. 2011, Lopes et al. 2015). Also, air-drying is likely to eliminate the activity of enzymes more susceptible to denaturation leaving only stabilized enzymes

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that could have an important ecological effect on soil quality (Nannipieri et al. 1990).

In this paper, samples collected from a soil disturbed by the application of sewage sludge (SS) were analysed to evaluate the use of enzymes determined on air-dried samples as soil quality indicators. This approach could enhance the use of biochemical parameters in soil fertility assessments. The aim was to evaluate the effects of air-drying on cellulase, arylsulfatase and acid phosphatase activities in soil. The hypothesis that the activities of these enzymes depend on soil moisture was tested.

MATERIAL AND METHODS

The soil samples were collected in February 2015 in a Ferralsol at a depth of 0–10 cm in a long-term experiment. It was set up in 2001 to study the use of sewage sludge as a source of nitrogen (N) for maize. The trial was located in the city of Campinas, Sao Paulo State, Brazil (approximate coordinates of 22°52'44''S, 47°04'56''W and average altitude of 650 m a.s.l.) and consisted of plots of 4 × 25 m (100 m^2). The field experiment was arranged in a randomized complete block design with three treatments and four replications. The treatments consisted of two sewage sludge rates equal to 10 (SS1) and 20 t/ha (SS2) and a mineral fertilizer treatment (MF). The MF received 6 kg N/ha, 9.2 kg P/ha and 10 kg K/ha. Soil chemical and physical characteristics at the beginning of the study are presented in Table 1.

Soil sampling was performed at five different locations inside each plot. The samples were mixed to obtain two composite samples per plot. A total of 24 samples were collected (3 treatments, each with eight replicate plots). Soil samples were then divided into two groups. In the first group, soil samples were sieved through a 2 mm sieve and immediately stored at 4°C with field moisture. Analyses were performed within a period of 1 week after sampling. In the second group, soil samples were air-dried at room temperature for two weeks, sieved through a 2 mm sieve and stored for 1 month at room temperature (24.8 ± 1.3°C).

Available phosphorus (P) and available sulfur (S) were quantified as described by Raij et al. (2001). The soil organic carbon (SOC) was quantified according to the Walkley-Black method (Nelson and Sommers 1982). The activity of the enzymes acid phosphatase and arylsulfatase was determined according to Tabatabai (1994). The activity of cellulase was determined according to Schinner and von Mersi (1990).

Data of soil enzymes from air-dried samples (X) were plotted against field-moist samples (Y) and linear relationships were fitted to quantify the deviation from the 1:1 slope. The change of enzyme activities (%) due to air-drying was calculated for each treatment using the Eq. 1:

$$Enz_{act} = [(B–A)/A] \times 100 \quad (1)$$

Where: A – amount of enzyme activity in the field-moist samples; B – amount of enzyme activity in the air-dried stored samples.

To assess the effects of the air-drying on the enzymatic activities the data were analysed by the analysis of variance (ANOVA – *F*-test) and means were compared by the Tukey's test. Both, *F*-test and Tukey comparison were carried out at $P < 0.05$. All assumptions required by the ANOVA were verified and confirmed before calculations.

RESULTS AND DISCUSSION

The effect of air-drying on soil enzymes activities is variable (Zornoza et al. 2006, Turner and

Table 1. Chemicals and physical properties of soil

Parameter	Unit	Treatment		
		MF	SS1	SS2
C_{org}	(g/L)	3.98	20.38	25.15
pH		5.0	5.1	4.8
P	(mg/L)	42.25	173.25	225.75
S	(mg/L)	19.5	53.0	84.0
Ca		37.00	54.75	48.00
K		2.78	3.32	2.60
H	($mmol_+$/L)	26.00	35.75	61.25
CEC		88.28	117.32	131.1
Sum of bases		62.28	81.58	69.85
Base saturation		70.25	69.25	53.50
Clay	(%)	28.0	28.0	28.0
Silt		16.3	16.3	16.3
Sandy		55.6	55.6	55.6

SS1 – sewage sludge 10 t/ha; SS2 – sewage sludge 20 t/ha; MF – mineral fertilizer; CEC – cation exchange capacity

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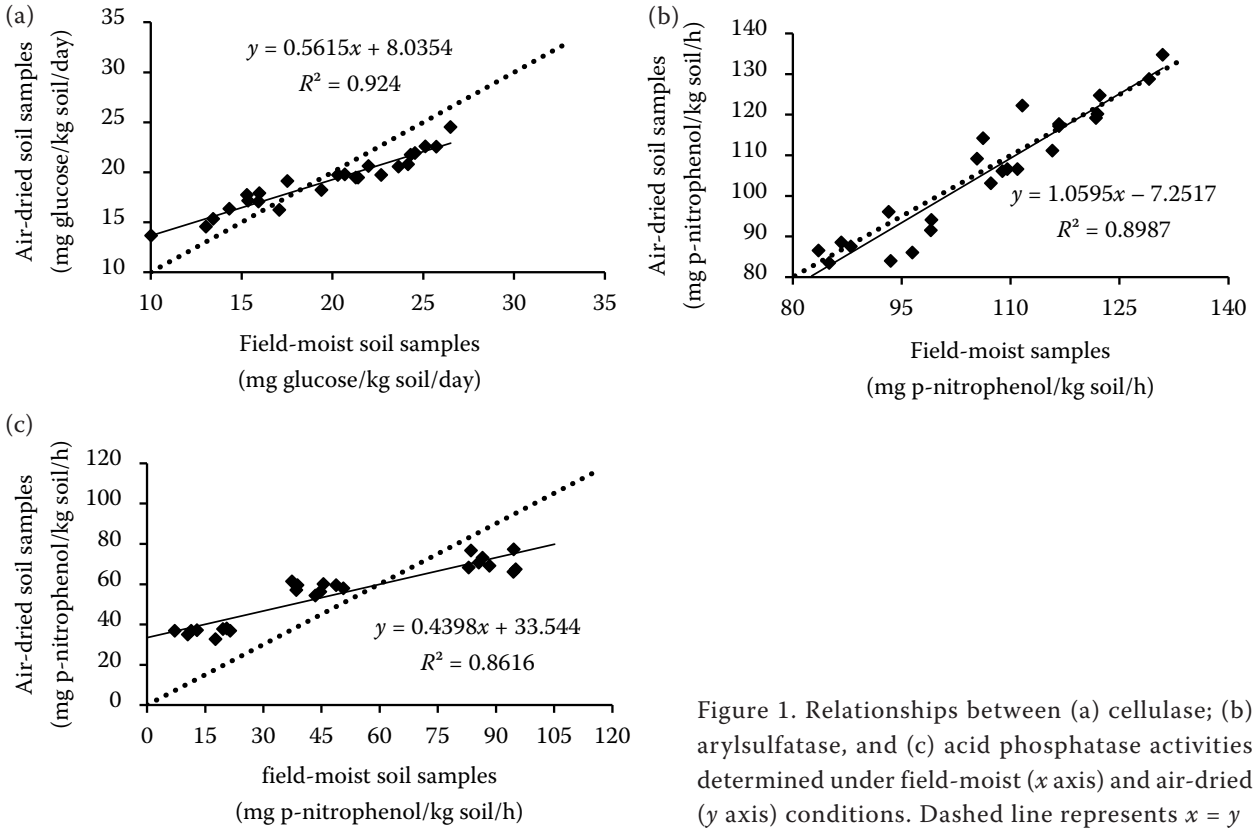

Figure 1. Relationships between (a) cellulase; (b) arylsulfatase, and (c) acid phosphatase activities determined under field-moist (x axis) and air-dried (y axis) conditions. Dashed line represents $x = y$

Romero 2010, Abellan et al. 2011, Lopes et al. 2015). Based on the deviation from a 1:1 relationship for cellulase (0.56), arylsulfatase (1.06) and acid phosphatase (0.44), it is possible to verify that the arylsulfatase was less affected by air-drying followed by cellulase and acid phosphatase (Figure 1). The greatest deviation was observed for the acid phosphatase activity, with overestimations of the field-moist values.

Acid phosphatase in the MF treatment was the most affected by air-drying with an increase of 139.6%. The increase was less expressive in the SS1 treatment (33.8%) and in the SS2 treatment a decrease of 20.1% was observed (Table 2). These results disagree with Speir and Ross (1975) and Sparling et al. (1986) who observed a decline in acid phosphatase activity in air-dried samples. Lopes et al. (2015) also found a decrease of 72% of acid phosphatase activity in air-dried samples. However, our results agree with Eivazi and Tabatabai (1977) who found an increase in acid phosphatase activity with air-dried soil samples. It can be postulated that drying the soil caused the death of microorganisms and consequently, resulted in the release of intracellular enzyme into the soil. Increasing levels of sewage sludge decrease the change in acid phosphatase activity caused by air-drying. This may be due to the increase of organic matter content in SS treatments that provided better conditions of living for microbial cells.

Higher rates of sewage sludge increased cellulase and acid phosphatase activity (Figure 2). Cellulase

Table 2. Mean percentage change (+ increase; – decrease) of soil enzymes activities after air-drying samples (%)*

Treatment	Cellulase	Arylsulfatase	Acid phosphatase
MF	+14.5	–3.2	+139.6
SS1	–4.8	+3.1	+33.8
SS2	–11.4	–3.1	–20.1

*[(B–A)/A] × 100. A – amount of enzyme activity in field-moist samples; B – amount of the enzyme activity in the air-dried store samples; MF – mineral fertilizer; SS1 – sewage sludge 10 t/ha; SS2 – sewage sludge 20 t/ha

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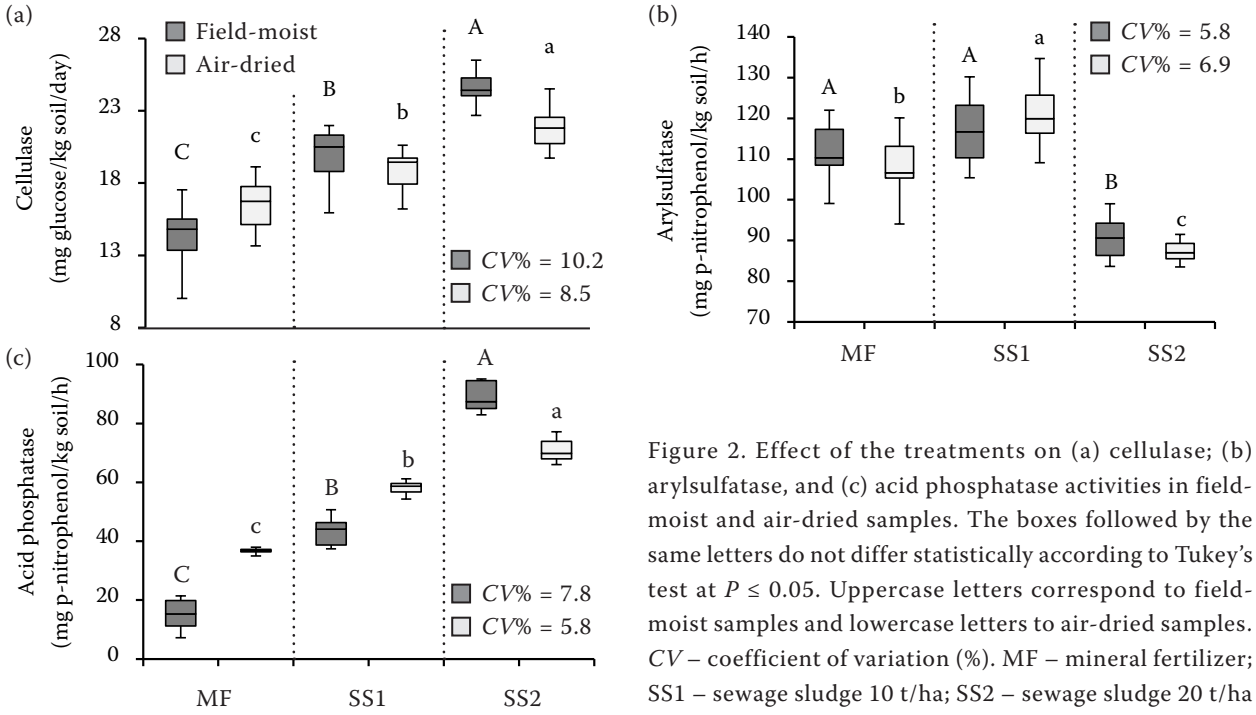

Figure 2. Effect of the treatments on (a) cellulase; (b) arylsulfatase, and (c) acid phosphatase activities in field-moist and air-dried samples. The boxes followed by the same letters do not differ statistically according to Tukey's test at $P \leq 0.05$. Uppercase letters correspond to field-moist samples and lowercase letters to air-dried samples. *CV* – coefficient of variation (%). MF – mineral fertilizer; SS1 – sewage sludge 10 t/ha; SS2 – sewage sludge 20 t/ha

raised by 71% and 32.4% and acid phosphatase by 486.1% and 95.3% in field-moist and air-dried samples, respectively. This may be correlated to the higher soil C and P contents with the sewage sludge application (Table 1).

Arylsulfatase activity decreased with increasing rates of sewage sludge by circa 20% in the same way for both moisture conditions (Figure 2). Yada et al. (2015) also obtained a reduction of arylsulfatase activity with the application of sewage sludge in comparison to the control. The authors attributed this result to the greater sensitivity of the enzyme to some components of the SS. In our work, the decrease of arylsulfatase with sewage sludge can be explained, in part, by the increase of available S in soil of 172% and 331% in comparison to the MF (Table 1). A significant and negative correlation of enzyme activity and the amount of available nutrient, the product of the enzyme, was expected due to the inhibition of the activity by feed-back processes (Olander and Vitousek 2000, Zornoza et al. 2006). In the same way, it can be argued that SS application increased soil sulfate to a level in which moisture conditions were not causing changes to arylsulfatase activity due to the amount of available S present in the soil.

Ranking positions of treatments based on their enzymatic activity did not change due to soil moisture (Figure 2). These results showed that for our experimental conditions, the enzymatic activity could be useful to distinguish soil disturbed by residue addition with no interference of moisture.

Li and Sarah (2003) examined the effect of air-drying on arylsulfatase and acid and alkaline phosphatases activity and did not find significant differences with field-moist samples. Bandick and Dick (1999) compared the activity of enzymes in different crops and agricultural practices and did not obtain a difference in the ranking of treatments between both dried and non-dried samples. These evidences support our expectation that the activities of these enzymes determined in air-dried samples are representative of those obtained from field-moist samples.

The use of air-dried soil samples enables eliminating the associated activity with free enzymes, which are more susceptible to denaturation (Nannipieri et al. 1990). Evaluation of stable enzymes reflects important aspects of the soil ecosystem functioning (Zornoza et al. 2006). They could have an important ecological effect on soil quality because biochemical activity could remain in soil despite the rapid reduction of the microbial community (Zornoza et al. 2006).

In conclusion, it can be stated that cellulase, arylsulfatase and acid phosphatase determined in air-dried soil samples hold the capacity to identify different organic management, although this

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pre-treatment significantly increased the acid phosphatase activities. It is also important to encourage more investigations concerning different amounts and types of organic material (urban, industrial or crop residues) to ensure consistency to the results reported in this paper.

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