

## THE EFFECT OF COMPOST AND SEWAGE SLUDGE ON SOIL BIOLOGIC ACTIVITIES IN SALT AFFECTED SOIL

Abdelbasset Lakhdar<sup>1,3</sup>, Rosalia Scelza<sup>2</sup>, Riccardo Scotti<sup>2</sup>, Maria A. Rao<sup>2\*</sup>, Naceur Jedidi<sup>3</sup>, Liliana Gianfreda<sup>2</sup>, Chedly Abdely<sup>1</sup>

<sup>1</sup>Laboratoire d'Adaptation des Plantes aux Stress Abiotiques, Centre de Biotechnologies, Technopole Borj Cedria, BP 901, Hammam Lif 2050, Tunisia. <sup>2</sup>Dipartimento di Scienze del Suolo, della Pianta, dell'Ambiente e delle Produzioni Animali, Università di Napoli Federico II, Portici, Italy. <sup>3</sup>Centre de Recherches et Technologies des Eaux, Technopole Borj Cedria, BP 273, Soliman 8020, Tunisia. \*Corresponding author: [maria rao@unina.it](mailto:maria rao@unina.it)

### ABSTRACT

Saline soil was amended with 13.3 and 26.6 g kg<sup>-1</sup> of Municipal Solid Waste (MSW) compost or sewage sludge, and arylsulphatase (ARY), phosphatase (PHO), dehydrogenase (DEH),  $\beta$ -glucosidase ( $\beta$ -GLU), urease (URE) and catalase (CAT) activities as well as physical-chemical properties were determined after 70 day of incubation under laboratory conditions. MSW compost and sewage sludge significantly improved soil physical-chemical properties, especially carbon and nitrogen contents. Accordingly, overall enzyme activities were substantially promoted in presence of both amendments and the higher increases were measured at 13.3 g kg<sup>-1</sup> of MSW compost (increases by 107%, 43%, 20%, 11%, and 148% for, DEH,  $\beta$ -GLU, PHO, URE, and CAT, respectively). Lower beneficial effects occurred at 26.6 g kg<sup>-1</sup> of sewage sludge possibly because of the increased salinity or the presence of trace elements by sewage sludge application. As a general response, MSW compost supplied at 13.3 g kg<sup>-1</sup> seems to be a useful strategy to enhance biologic activities of salt-affected soil.

**Keywords:** Biologic activities, MSW compost, saline soil, sewage sludge.

### INTRODUCTION

Salt toxicity is one of the major edaphic factors limiting crop production and eco-environmental quality in salt-affected soils throughout the world (Hanay *et al.*, 2004; Liang *et al.*, 2005). Excessive amounts of salts have adverse effect on physical and chemical properties and on biologically mediated processes in the soil, such as C and N-mineralization (Tejada *et al.*, 2006). Sal toxicity may also

constitute an important abiotic concern affecting plant yield in arid and semiarid regions (Lakhdar *et al.*, 2008).

Various organic amendments such as Municipal Solid Waste (MSW) compost or Sewage sludge have been investigated for their effectiveness in saline soil remediation (Tejada *et al.*, 2006; Walker and Bernal, 2008). The application of organic matter to saline soils can have

different effects such as speeding up of NaCl leaching, decrease of the exchangeable sodium percentage and electrical conductivity and increase of water infiltration (El-Shakweer *et al.*, 1998). Meanwhile, the application of biosolids increases soil microbial biomass and some soil enzymatic activities such as urease, alkaline phosphatase and  $\beta$ -glucosidase linked to C, N, P and S soil cycles (Liang *et al.*, 2003; Tejada *et al.* 2006). In fact hydrolytic enzymes are sensitive indicators of management induced changes in soil properties due to their strong relationship with soil organic matter content and quality (Masciandaro *et al.*, 2004).

According to Rao and Pathak (1996) and Liang *et al.* (2005), the incorporation of organic amendments to soil stimulate dehydrogenase activity because the added material may contain intra- and extracellular enzymes and may also stimulate microbial activity in the soil. These parameters are the most sensitive to the changes which occur in salt-affected soil, and provide rapid and accurate information on changes in soil quality.

A simulated salinized experiment was performed in this study to examine the effects of MSW compost and sewage sludge incorporated in salt affected soil on the activity of some key enzymes related to nutrient cycling (arylsulphatase, phosphatase, dehydrogenase,  $\beta$ -glucosidase, urease and catalase).

## **MATERIALS AND METHODS**

### **Experimental design**

A loam-silt, saline soil was collected from salt-affected ecosystem Sebkhia Soliman (North-East Tunisia), dried and sieved at

2 mm. The main soil properties were: 23% clay, 55% silt, 22% sand, and pH (1:25) 8.2, total CaCO<sub>3</sub> 23%, EC 75 dS m<sup>-1</sup>.

Municipal solid waste (MSW) compost was collected from composting station of Beja (North Tunisia). It was mechanically produced by mixing weekly the waste heap under aerobic conditions by fast fermentation and aged 8 months before use. Urban secondary sewage sludge was obtained from the activated sludge waste water treatment plant of Radès near Tunis, Tunisia. The aerobic digested sludge was previously air dried. Table 1 shows the main chemical properties of MSW compost and S sludge. In a completely randomized experimental design with three replications, *Hordeum maritimum* seeds were sown in pots containing 1 kg of soil in a growth chamber with 14/10 h light/dark cycle, 22/18°C day/night temperature, Photosynthetic Active Radiations (PAR) of 200 mmol m<sup>-2</sup> s<sup>-1</sup> PAR, and a relative humidity of 60-80%.

The soil was previously amended with two amounts of MSW compost (C1 and C2) or sewage sludge (S1 and S2), 13.3 and 26.6 g kg<sup>-1</sup> corresponding respectively to 40 and 80 t ha<sup>-1</sup> soil. A non-amended soil served as control soil. After 70 days incubation, moistened soil samples were collected for enzymatic assays. Plant responses were previously studied by Lakhdar *et al.* (2008).

### **Enzymatic activity assays**

Enzyme activities were determined on fresh, moist, sieved (<2mm) soils. Substrates, i.e., *p*-nitrophenyl- $\beta$ -D-glucoside and *p*-nitrophenyl-phosphate, for  $\beta$ -glucosidase ( $\beta$ -GLU) (E.C. 3.2.1.21) and phosphatase (PHO) (E.C. 3.1.3.2) were used, respectively.

**Table 1.** MSW compost and sewage sludge characteristics (means and standards calculated on three replication basis).

	MSW compost	Sewage sludge
pH	7.90±0.04	7.07±0.50
EC ( $\mu\text{S cm}^{-1}$ )	$8 \cdot 10^3 \pm 40$	$1.01 \cdot 10^3 \pm 50$
C ( $\text{g kg}^{-1}$ )	130±22	272±20
N ( $\text{g kg}^{-1}$ )	11.4±0.6	38.7±8.0
C/N	11.4±1.8	7.0±0.8
P <sub>2</sub> O <sub>5</sub> ( $\text{g kg}^{-1}$ )	0.24±0.01	0.10±0.01
K <sup>+</sup> ( $\text{g kg}^{-1}$ )	1.80±0.09	0.85±0.05
Cu <sup>2+</sup> ( $\text{mg kg}^{-1}$ )	91.6±13.2	284.0±2.3
Pb <sup>2+</sup> ( $\text{mg kg}^{-1}$ )	251.6±12.1	101.7±2.9
Zn <sup>2+</sup> ( $\text{mg kg}^{-1}$ )	290.2±11.1	592.7±21.7

Briefly, 1 g of soil was incubated with 5 ml of buffered substrate in reaction flasks for 1 h at 30°C, under continuous stirring. Specific buffers and pHs were used as reported in Sannino and Gianfreda (2001) and Eivazi and Tabatabai (1990). Enzymatic reactions were stopped by rapidly transferring the mixtures to a freezer and holding them there for 10 min. Concentrations of *p*-nitrophenol were determined at 400 nm after addition of NaOH and CaCl<sub>2</sub> for PHO and Tris/NaOH buffer (pH 10.0) and CaCl<sub>2</sub> for  $\beta$ -GLU.

The activity of urease (URE) (E.C. 3.5.1.5) and arylsulphatase (ARY, E.C. 3.1.6.1) were determined as described by Kandeler and Gerber (1988) and Tabatabai and Bremner (1970), respectively. Dehydrogenase (DEH) (E.C. 1.1. Activity was measured by mixing 1 g of soil with 1 ml of buffered

tetrazolium salts (TTC) solution, according to Trevors (1984). Catalase (CAT, E.C. 1.11.1.6) activity was determined according to Rodriguez-Kabana and Truelove, 1982.

One unit of enzyme activity was defined as the number of  $\mu\text{moles}$  (for  $\beta$ -GLU) or  $\mu\text{grams}$  (for DEH, FDAH, NR and URE) of product released at 30°C h<sup>-1</sup> by 1 g of dried soil. Control tests with autoclaved soils were carried out to evaluate the spontaneous or abiotic transformation of substrates.

All determinations were made in triplicate and data were corrected to oven-dry (105°C) moisture content.

#### Statistical analysis

Statistical analysis was performed by Analysis of variance (ANOVA) and Duncan's multiple range test was used to

evaluate significant differences between means at  $P \leq 0.05$ . All statistical analyses were carried out with the program SPSS 13.0.

## RESULTS

### Soil physical and chemical characteristics

The addition of two doses of MSW compost and Sewage sludge had different effects on some of physico-chemical properties of the soil. They generally increased the values of all measured properties, as respect to the control soil. Different patterns were, however, observed according to the dose and type of used amendment (Table 2).

Different patterns were, however, observed according to the dose and type of used amendment (Table 2). For instance, no significant change of pH was observed under MSW compost, while a slight decrease was recorded with sewage sludge treatment. Both amendments induced an increase of EC especially at 26.6 g kg<sup>-1</sup>. EC increased by 30% and 33% in C2 and S2, respectively, while only 13-14% was detected at 13.3 g kg<sup>-1</sup>.

Increases were also measured for C and N contents. Indeed, carbon content increased up to 4.0-4.4-fold the value of control soil in C1 and S1, respectively, and much more (5.0-5.5 fold) in the presence of 26.6 g kg<sup>-1</sup> of MSW compost and sewage sludge. A more moderate increase was measured for N that was on average 50% higher than that measured in

**Table 2.** Soil physical and chemical characteristics after the culture. Control: soil without amendment. C1: soil amended with 13.3 g kg<sup>-1</sup> of MSW compost; C2: soil amended with 26.6 g kg<sup>-1</sup> of MSW compost; S1: soil amended with 13.3 g kg<sup>-1</sup> of sewage sludge; S2: soil amended with 26.6 g kg<sup>-1</sup> of sewage sludge. Data are the means of 3 replicates. Means followed by the same letters are not significantly different according to the Duncan's Multiple Range Test at  $P \leq 0.05$ .

	Control	C1	C2	S1	S2
pH	8.27±0.01ab	8.28±0.04ab	8.31±0.01a	8.26±0.01b	8.22±0.01c
EC (µS cm <sup>-1</sup> )	647±29c	741±10b	843±60a	731±61b	863±100a
C (g kg <sup>-1</sup> )	2.5±0.1c	10.9±0.2b	12.7±0.1a	10.1±0.1b	13.7±0.3a
N (g kg <sup>-1</sup> )	1.2±0.1c	1.8±0.1b	1.9±0.3a	1.8±0.1b	2.1±0.1a
C/N	2.1±0.1c	6.1±0.3ab	6.7±0.3a	5.6±0.1b	6.5±0.2a
K <sup>+</sup> (mg g <sup>-1</sup> )	0.25±0.005b	0.27±0.004a	0.24±0.01b	0.23±0.004c	0.26±0.003a
Ca <sup>2+</sup> (mg g <sup>-1</sup> )	0.16±0.01d	0.17±0.01c	0.17±0.01c	0.22±0.005b	0.34±0.01a
Cu <sup>2+</sup> (mg kg <sup>-1</sup> )	15.65±0.35d	17.30±0.14c	19.05±0.35b	18.75±0.3b	22.40±0.28a
Pb <sup>2+</sup> (mg kg <sup>-1</sup> )	96.8±1.6b	103.6±6.4b	113.1±13.5a	114.7±8.9a	114.0±5.0a
Zn <sup>2+</sup> (mg kg <sup>-1</sup> )	83.9±7.1d	86.6±8.0c	89.9±14.0b	86.9±3c	95.6±9.9a

the control soil in C1, C2 and S1 and only with the highest amount of sewage sludge reached a 1.75-fold increase.

Moderate increases or no statistically variations occurred for soil  $K^+$  at both applied doses of either MSW compost or sewage sludge. The addition of the lower amount of MSW compost slightly raised  $Ca^{2+}$  value and no additional increase was monitored by doubling its quantity.

On the contrary, sewage sludge elevated the level of the cation from  $0.16 \pm 0.01 \text{ mg g}^{-1}$  of the control soil up to  $0.22 \pm 0.005 \text{ mg g}^{-1}$  and  $0.34 \pm 0.01 \text{ mg g}^{-1}$  at  $13.3 \text{ g kg}^{-1}$  and  $26.6 \text{ g kg}^{-1}$ , respectively.

Heavy metal concentration at some extent increased or did not change in

concomitance with MSW compost and sewage sludge supply. However with the higher dose of sewage sludge an increase by 50% of  $Cu^{2+}$  and by 14% of  $Zn^{2+}$  occurred as respect to their respective controls (Table 2).

### Enzymatic activities

According to the substantial increase of C amount (and in turn of soil organic matter content), the levels of measured enzyme activities were generally enhanced by the application of the two amendments as respect to the non-amended soil (Table 3).

The highest pronounced increase was measured for DEH (by 107%) and CAT

**Table 3.** Soil arylsulphatase (ARY,  $\mu\text{mol } p\text{-NP g}^{-1} \text{ h}^{-1}$ ), phosphatase (PHO,  $\mu\text{mol } p\text{-NP g}^{-1} \text{ h}^{-1}$ ), dehydrogenase (DEH,  $\mu\text{g TPF g}^{-1} \text{ h}^{-1}$ ),  $\beta$ -glucosidase ( $\beta$ -GLU,  $\mu\text{mol } p\text{-NP g}^{-1} \text{ h}^{-1}$ ), urease (URE,  $\text{mg urea g}^{-1} \text{ h}^{-1}$ ) and catalase (CAT,  $\mu\text{mol O}_2 \text{ g}^{-1} \text{ min}^{-1}$ ) activities after 70 days incubation. Control: soil without amendment. C1: soil amended with  $13.3 \text{ g kg}^{-1}$  of MSW compost; C2: soil amended with  $26.6 \text{ g kg}^{-1}$  of MSW compost; S1: soil amended with  $13.3 \text{ g kg}^{-1}$  of sewage sludge; S2: soil amended with  $26.6 \text{ g kg}^{-1}$  of sewage sludge. Data are the means of 3 replicates. Means followed by the same letters are not significantly different according to the Duncan's Multiple Range Test at  $P \leq 0.05$ .

	Control	C1	C2	S1	S2
DEH	2.355±0.201d	4.875±0.653a	3.053±0.541c	2.885±0.153c	3.257±0.161b
$\beta$ -GLU	0.917±0.012c	1.309±0.011a	0.963±0.082c	0.997±0.051bc	1.063±0.042b
PHO	6.108±0.151c	7.353±0.122b	6.020±0.234c	7.787±0.270a	5.901±0.234c
ARY	0.257±0.014e	0.278±0.013d	0.304±0.015c	0.420±0.013b	0.538±0.021a
URE	3.814±0.193b	4.237±0.101a	3.806±0.164b	2.697±0.201c	2.451±0.192d
CAT	2.111±0.521d	5.240±0.453a	4.674±0.171b	3.506±0.173c	3.503±0.504c

(by 148%) activities in the presence of the lower MSW compost dose. Minor rises were detected for both enzymes at a double MSW quantity.

Sewage sludge also increased DEH and CAT activities but at a less extent as respect to compost and no variation was observed between the two applied doses.

Among the four hydrolases, soil urease was the most affected by amendment and contrasting responses occurred with compost and sludge. As respect to the urease value of control soil, sludge decreases URE activity and much more at the higher applied dose (Table 3). By contrast, a 1.1-fold increment or no change was measured for URE activity in C1 and C2, respectively.

Significantly detectable increases also occurred for ARY activity in the presence of sewage sludge, whereas moderate effects were observed with compost (Table 3). Enhancements by 20% and 27% were monitored for PHO in C1 and S1, while no variation was observed in C2 and S2.

## **DISCUSSION**

The low productivity of saline soils may be ascribed not only to their salt toxicity or damage caused by excess amounts of soluble salts but also to their low soil fertility (Liang *et al.*, 2003). As expected, the added organic amendment (MSW compost and Sewage sludge) improved properties of salt-affected soil (Table 2).

No variation of soil pH was observed under C1 and C2, likely because MSW compost has a pH similar to that of the soil. On the contrary and in accordance with the findings of Tsadilas and Samaras (1998), Sewage sludge reduced slightly soil pH probably because its pH is close to neutrality. EC was similarly increased at two doses of both amendments.

Hargreaves *et al.* (2008) explained this fact by the presence of supra-optimal salt concentrations in both amendments. In addition, the application of organic matter promotes flocculation of clay minerals, which is an essential condition for the aggregation of soil particles allowing the increase of oxygen diffusion rate for soil

micro-organisms. In addition enrichment of the rhizosphere with  $\text{Ca}^{2+}$ , mainly observed in S2 treatment (Table 2), tends to replace exchangeable  $\text{Na}^+$  on the adsorption sites accelerating saline water leaching (Garcia *et al.*, 2000; Tejada *et al.*, 2006).

In consistence with the findings of Madejon *et al.* (2001) and Marschner *et al.* (2003) both amendments improved soil C and N contents (Table 2).

Meanwhile, enzymatic activities were significantly stimulated (Table 3). This could possibly be related to a shift in microbial community towards the dominance of salt tolerable micro-organisms (Li *et al.*, 2006).

Oxidative enzymes, especially DEH, were proposed as a measure of overall microbial activity. DEH being an intracellular enzyme related to oxidative phosphorylation processes that occurs in all intact, viable microbial cells (Tejada *et al.*, 2006).

Incorporation of both MSW compost and Sewage sludge stimulated DEH activity because the added material may contain intra- and extracellular enzymes and may also stimulate microbial activity in the soil (Liang *et al.*, 2005). DEH was markedly increased at C1 treatment (Table 3). Accordingly,  $\beta$ -GLU, URE, PHO and CAT mostly increased in C1, thus supporting a general increment of the soil enzymatic activities upon compost addition. Pathak and Rao (1998), found a steady evolution of  $\text{CO}_2$  throughout 3 months of high salinity treatment showing a high activity of the heterotrophic microflora. However, C2 and S2 treatments seem to be supra-optimal doses, likely due to salinity or heavy metal increment. In fact, some studies indicated that high doses of some organic materials can introduce into the soil toxic compounds such as heavy metals which could have a negative effect on enzyme activities (Garcia *et al.*, 1994).

The only exception was soil ARY that was enhanced at high dose of both organic amendments.

The demand for P by soil microorganisms and plants may be responsible for the stimulation in the synthesis of this enzyme (Tejada *et al.*, 2006). In addition, added organic matter (high increase of C content) likely may alleviate the 'salting-out' process involved in the decrease of enzyme solubility through alteration of the enzyme 'catalytic site'. Furthermore, Rasul *et al.* (2006) stated that these microorganisms would likely have low substrate use efficiency. Thus, by directing organic substrate utilization for the maintenance of metabolic activity, micro-organisms could overcome temporary salinity stress (Tate, 1995; Li *et al.* 2006).

## CONCLUSIONS

In conclusion, both MSW compost and Sewage sludge affected soil physical and chemical properties and biological activities. The positive effect of the organic amendments on soil biological quality is due to the stimulation of microbial growth and/or to the addition of microbial cells or enzymes with the amendment, which can counteract the negative effect of salinity. This hypothesis seems to be reliably supported by the increase observed for more than one soil enzyme activity that very likely behaved as valid indicators of soil biological activity. However, a balance between adequate fertilization and the possible environmental risks caused by over fertilization must be considered.

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