

## Biosolid amendment of a calcareous, degraded soil in a semi-arid environment

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### Abstract

Many soils in the Mediterranean region are subjected to progressive degradation as a result of water erosion. A study was carried out to examine the effects of biosolids on a degraded soil. Single doses of 40, 80 and 120 Mg ha<sup>-1</sup> of a biosolid were applied to the surface of soil and their effects on its chemical characteristics and on the native vegetation were assessed. Soil macronutrients, micronutrients and heavy metals were tested one and five years after application. Canopy cover, biomass production and mineral composition of native vegetation were also determined. Biosolids increased the content of most nutrients in the soil, although this effect decreased over time. The soil organic matter content did not increase significantly at the beginning of the experiment, although significant differences were observed after five years. The contents of total and extractable heavy metals did not change with time after the application. The native plant biomass production and the canopy cover significantly increased with all the doses applied at the beginning of the experiment, and remained high five years after biosolids application.

**Additional key words:** canopy cover, heavy metals, native plant biomass, soil chemical properties, soil restoration.

### Resumen

#### Aplicación de un biosólido como enmienda sobre un suelo degradado y calcáreo en un ambiente semiárido

Se ha realizado una experiencia de restauración de un suelo degradado en un ecosistema semiárido. Diferentes dosis de biosólidos (40, 80 and 120 Mg ha<sup>-1</sup>) fueron aplicados una única vez en superficie y se estudiaron sus efectos sobre las principales propiedades químicas del suelo y la cobertura vegetal, producción de biomasa y composición mineral de la vegetación nativa al año y a los cinco años de la aplicación. Las dosis de biosólido incrementaron los contenidos de nutrientes en el suelo, aunque su efecto disminuyó con el tiempo. El contenido de materia orgánica del suelo no experimentó cambio al comienzo de la experiencia, mientras que hubo diferencias significativas entre tratamientos en el último año. El contenido de metales pesados del suelo no varió a lo largo del estudio. La producción de biomasa y cobertura vegetal se incrementaron significativamente con todas las dosis ensayadas y permanecieron así a lo largo de los cinco años del estudio.

**Palabras clave adicionales:** biomasa vegetal, cobertura vegetal, metales pesados, propiedades químicas del suelo, restauración de suelos.

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### Introduction

The need to reduce biosolids waste (stabilized waste water sludge) disposal costs and to increase concentrations of soil nutrients and organic matter has led to the use of biosolids in the amendment and fertilization of both agricultural and natural land.

Generally, correct application of biosolids improves the physical, chemical and biological properties of the soil (García *et al.*, 1998; Lindsay and Logan 1998; Walter *et al.*, 2000; Binder *et al.*, 2002; Martínez *et al.*, 2003a; Ros *et al.* 2003), enlarge the carbon sink, and can reduce greenhouse gas emissions (Akala and Lal, 2001; Swift, 2001; Lal *et al.* 2003).

Many soils in the Mediterranean region are susceptible to water erosion due to loss of the protective vegetation. Adverse climatic conditions and poor land management have led to a reduction of the

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soil organic matter content, its structural degradation and, hence, a loss of fertility. For good plant establishment, the physicochemical and biological soil properties need to be improved (Brofas *et al.*, 2000; García, 2001). The application of biosolids and proper management, can be possible ways for soil restoration. After biosolids application, plant cover is rapidly established, helping to minimize runoff and reduce water erosion (Rostagno and Sosebee, 2001; Martínez *et al.*, 2003b; Ojeda *et al.*, 2003).

Huge amounts of biosolids are produced in the Madrid region, and disposing of them is a problem. However, there are also large areas with marginal, fragile and ecologically sensitive soils around Madrid where excessive human pressure has led to soil degradation. The application of biosolids could alleviate impoverished soil conditions and help to restore this land.

In an attempt to mitigate the loss of soil productivity and restore the vegetation cover, a field experiment was carried out where different rates of biosolids were applied to a degraded native grassland under semi-arid environment. The effects on some soil chemical characteristics and heavy metal concentrations, and on the mineral composition and production of native vegetation, were assessed one and five years after the application.

## Material and Methods

### Site description and experimental procedures

In April 1997, anaerobically-digested sewage sludge (biosolids) obtained from a domestic wastewater treatment plant in Madrid was applied once to the surface of a calcareous soil 35 km southeast of Madrid. The soil is a Lithic Xerorthent according to the Soil Survey Staff (1998). The native vegetation cover in the area is scarce (30 - 40%), and strongly influenced by climatic limitations, consisting mainly of a mixture of herbaceous plants and slow-growing low bushes (Graminae, *Plantago albicans*, *Helianthemum asperum*, *Teucrium pseudochanmaepitys*, *Thymus zygis*, *Centaurea melitensis*, etc.) (Cuevas *et al.*, 2000). The experimental field was divided into four blocks arranged horizontally on the slope (range: 8-12%). Each block contained four 3 × 20 m plots (with a 1 m buffer zone between plots and 3 m between blocks),

which were randomly assigned to one of the following doses of biosolid (BS): 0, 40, 80 and 120 Mg ha<sup>-1</sup> (dry matter). The climate of the study area is Mediterranean (mean annual precipitation is 380 mm and mean annual temperature 16.8°C).

### Soil sampling and analysis

Nine composite soil samples were taken from each plot at depths of 0 to 15 cm with a 5 cm diameter bucket auger in March 1997 (before treatments), and then again in March 1998 and March 2002 (one and five years after treatment, respectively). Soil samples were air-dried and ground to pass a 2 mm sieve. Some chemical properties of all soil samples from the three sampling dates were determined. Particle size analysis was done with the Bouyoucos method. The pH (1:2.5 soil:water ratio) was measured by glass electrode and the electrical conductivity (EC) was measured with a conductivity cell in a 1:5 (w v<sup>-1</sup>) soil:water extract. Calcium carbonate was measured with the Bernard calcimeter. Soil organic carbon (SOC) was measured by wet oxidation. Total N was determined by the Kjeldahl method and inorganic N forms (NH<sub>4</sub>-N and NO<sub>3</sub>-N) after extraction with 1 M KCl (1:5 soil:extractant ratio) were determined by the indophenol-blue method, and the copperized cadmium reduction method for NH<sub>4</sub>-N and NO<sub>3</sub>-N, respectively. Available P was extracted in 0.5 M NaHCO<sub>3</sub> (Olsen-P) and available K in pH 7, 1 M NH<sub>4</sub>OAc and were later determined by colorimetry and by flame emission spectrometry, respectively. Cation exchange capacity (CEC) and exchangeable Ca<sup>2+</sup> and Mg<sup>2+</sup> were determined by the Na-acetate method. Heavy metal concentrations were determined, after digestion with HNO<sub>3</sub>-HCl 1:3 (McGrath and Cunliffe, 1985), and extracted with diethylenetriaminepentaacetic acid (DTPA) (Lindsay and Norvell, 1978), using a sequential, multi-element inductively coupled plasma (ICP) emission spectrometer. The main chemical properties of the biosolid and the soil before biosolid applications, are shown in Table 1.

### Plant sampling and analysis

The natural vegetation cover growth after the addition of BS was examined and samples were

**Table 1.** Main properties of the biosolids (BS) (on a dry matter basis) and the soil

Properties	BS	Soil ( $X \pm \sigma$ )
pH 1:2.5 soil/water	8.6	8.3 $\pm$ 0.07
EC (dS m <sup>-1</sup> ) at 25°C	1.62	0.14 $\pm$ 0.09
SOC (g kg <sup>-1</sup> )	253	19.5 $\pm$ 2.4
Kjeldahl- N (g kg <sup>-1</sup> )	26.5	2.0 $\pm$ 0.5
C/N	10.3	9.5
CaCO <sub>3</sub> (g kg <sup>-1</sup> )	—	478 $\pm$ 137
Total P (g kg <sup>-1</sup> )	10.6	—
Olsen- P (mg kg <sup>-1</sup> )	613	7.9 $\pm$ 1.23
Total K (g kg <sup>-1</sup> )	2.0	—
Available K (mg kg <sup>-1</sup> )	—	205 $\pm$ 35.1
Texture		Loamy sand
Total Zn (mg kg <sup>-1</sup> )	445	23.2 $\pm$ 5.3
Total Pb (mg kg <sup>-1</sup> )	252	36.6 $\pm$ 3.9
Total Cd (mg kg <sup>-1</sup> )	1.00	0.5 $\pm$ 0.2
Total Ni (mg kg <sup>-1</sup> )	15.3	7.1 $\pm$ 1.8
Total Cr (mg kg <sup>-1</sup> )	48.5	10.2 $\pm$ 2.2
Total Cu (mg kg <sup>-1</sup> )	174	6.4 $\pm$ 1.4

EC: Electrical conductivity refers to 1:5 soil:water extract; SOC soil organic carbon.  $X \pm \sigma$ : Mean  $\pm$  standard deviation.

collected from each treatment during late May and June of 1998 and 2002 to obtain the total canopy cover percentage and total aboveground plant biomass production. Selected chemical properties were also determined. Total canopy cover was estimated in 16 individual microplots of the different treatment plots (0.22 m<sup>2</sup>), following a random transect. Aboveground plant biomass was measured at the same time by harvesting (hand cutting) the total vegetation of six, randomly selected 0.22 m<sup>2</sup> quadrats per treatment. Plant tissue samples were washed, oven-dried at 60°C for 48 h, weighed, and then ground to 0.1 mm in a mill equipped with a stainless steel screen. Subsamples of dry plant material were ashed at 450°C for 24 h and the ashes dissolved with 2 M HCl. The extracts were analysed for macronutrients, micronutrients and heavy metals by plasma emission spectrometer (ICP). Plant tissue N was determined by the semi-micro Kjeldahl method.

### Statistical analysis

Data were analysed by standard ANOVA for randomised, complete block designs. The means of the main effects were compared using Duncan's test. Significance level was set at  $P \leq 0.05$ . When data were

not normally distributed, the Box and Cox (1964) diagnostic procedure was used to select the most appropriate transformation (Statgraphics Statistical Software, version 4.1).

## Results

### Soil chemical properties and heavy metal concentrations

Table 2 shows the main soil chemical properties. Soil pH did not change significantly as a result of the biosolid application. In the first year, soil EC only increased significantly with the highest dose. Even at the end of the experiment, the EC of the treated plots remained significantly higher than that of the unamended plots, but the values obtained were below toxic levels for plant growth. The SOC contents measured at the start of the experiment did not significantly change with amendment. However, at the end of the experiment, five years after the application, the SOC content of the high rate plots was significantly greater than that of the control plots. Kjeldahl soil N concentration behaved similarly to that of SOC. Significant increases were found only with the highest biosolid rate at the end of the experiment, while NH<sub>4</sub>-N

**Table 2.** Effects of biosolid doses on the main chemical properties, one (1998) and five years (2002) after its application

Variable	Biosolid doses (Mg ha <sup>-1</sup> )							
	0	40	80	120	0	40	80	120
	1998				2002			
pH (1:2.5 soil/water)	8.1	7.9	8.0	7.9	8.1	8.0	8.0	8.0
EC (dS m <sup>-1</sup> ) 25°C	0.15 b	0.15 ab	0.17 ab	0.19 a	0.16 b	0.17 ab	0.17 ab	0.18 a
SOC (g kg <sup>-1</sup> )	24.4	21.2	24.1	21.3	21.3 b	23.4 ab	22.9 ab	25.7 a
Kjeldahl-N (g kg <sup>-1</sup> )	2.42	2.08	2.80	2.25	1.99 b	2.15 ab	2.16 ab	2.40 a
NH <sub>4</sub> -N (mg kg <sup>-1</sup> )	1.41 b	2.57 b	2.63 b	9.11 a	2.78 b	5.01 a	4.20 a	5.22 a
NO <sub>3</sub> -N (mg kg <sup>-1</sup> )	10.0 c	8.27 c	51.6 b	142 a	13.8 c	18.7 bc	22.4 ab	28.6 a
Olsen- P (mg kg <sup>-1</sup> )	11.1 c	42.6 b	80.7 a	80.3 a	11.1 c	35.2 b	45.2 b	71.6 a
NH <sub>4</sub> OAc-K (mg kg <sup>-1</sup> )	271	244	252	271	219	196	200	277
CEC (cmol kg <sup>-1</sup> )	19.8	18.3	19.7	18.7	19.2 b	19.5 b	23.3 a	26.9 a
Ca <sup>+2</sup> + Mg <sup>+2</sup> (cmol kg <sup>-1</sup> )	18.8	17.7	18.5	17.4	18.3 ab	18.5 ab	22.8 a	25.8 a

Mean values within the same line and year followed by the same letter are not significantly different at  $P \leq 0.05$ ; EC: electrical conductivity; SOC: Soil organic carbon; CEC: cation exchange capacity.

and NO<sub>3</sub>-N contents were significantly higher than in the control soil in the three treatments at both sampling dates. The NO<sub>3</sub>-N concentrations were higher than those of NH<sub>4</sub>-N at both sampling times.

At the end of the experiment, the available P content was significantly higher in the amended plots than in the untreated plots, but lower than it was in the first year after the biosolid was applied. After one and five years, soil-available K was low in most of the amended plots; differences between treatments were not significant. The results obtained agree with the low K content of the biosolids applied (Table 1).

At the end of the experiment, the CEC of the intermediate and high rate (80 and 120 Mg ha<sup>-1</sup>) plots

was significantly higher than the unamended plots. No significant differences were found, however, in the CEC values after the first year of treatment, even for the highest sludge rate. The concentration of extractable Mg<sup>+2</sup> and Ca<sup>+2</sup> did not change significantly with any treatment.

Soil total heavy metal concentration increased as result of the biosolid amendment but the differences among treatments were not significant ( $P > 0.05$ ) at either sampling date. The concentration of DTPA-extractable metals in the soil did not change significantly with the BS treatments (Table 3) after one year of the application, while significant differences were found for most extractable heavy metals at the end

**Table 3.** Effects of biosolid doses on soil extractable (DTPA) heavy metal content one (1998) and five (2002) years after its application

DTPA-extractable heavy metals (mg kg <sup>-1</sup> )	Biosolid doses (Mg ha <sup>-1</sup> )							
	0	40	80	120	0	40	80	120
	1998				2002			
Zn	0.92	1.04	1.32	1.40	0.47 c	2.12 b	2.56 b	3.30 a
Pb	2.03	1.86	2.26	1.97	1.46 b	1.74 b	1.93 ab	2.41 a
Cd	nd	nd	nd	nd	0.02	0.02	0.02	0.02
Ni	0.18	0.16	0.17	0.18	0.10 b	0.14 b	0.19 ab	0.23 a
Cr	nd	nd	nd	nd	nd	nd	nd	nd
Cu	0.37	0.42	0.63	0.71	0.26c	0.80 b	1.16 a	1.41 a

Mean values within the same line and year followed by the same letter are not significantly different at  $P \leq 0.05$ ; nd: not detected. DL (detection limit) for Cd < 0.08 mg kg<sup>-1</sup> and for Cr < 0.1 mg kg<sup>-1</sup>.

of the experiment. DPTA-Cd and Cr levels were below detection limit ( $< 0.08$  mg Cd kg<sup>-1</sup> and  $< 0.1$  mg Cr kg<sup>-1</sup>) in all the treatments and at the two sampling dates.

### Characteristics of native vegetation

The BS application had a favourable effect on the establishment of native vegetation, an influence that persisted until the end of the experiment. With the exception of the highest rate, one year after the application, total plant cover percentage increased significantly ( $P \leq 0.05$ ) with all treatments (Table 4). Compared to the control plots, canopy cover was higher by 83%, 70% and 12% for the low, intermediate and high rate, respectively. Total aerial plant biomass production was significantly greater than in the control

( $P \leq 0.5$ ) for all three biosolid-amended soils in the first year, but only for the highest rate at the end of the experiment (Table 4).

Biomass production for all biosolid treatments at the end of the experiment was less than that, in the first year after biosolid application. Moreover, no significant differences in aerial biomass were found among the 0, 40 and 80 Mg ha<sup>-1</sup> treatments, or between the intermediate and high doses. Significant differences were found between the highest and lowest rate.

Biosolid application increased the macro- and micronutrient concentrations of the native vegetation to levels adequate for plant growth (Table 5). After the first year of application the N, P and K contents of the aerial components of the plants in the treated plots were significantly higher than those in the control plots, while at five years after treatment, the P content was the only nutrient with significantly higher levels. Plant

**Table 4.** Effects of biosolid doses on vegetation properties one (1998) and five (2002) years after its application

Biosolid doses (Mg ha <sup>-1</sup> )	Aerial biomass yield (g m <sup>-2</sup> )		Canopy cover (%)		Δ Canopy cover <sup>1</sup> (%)	
	1998	2002	1998	2002	1998	2002
0	153 c	169 bc	43 b	58 b	–	–
40	977 a	252 b	78 a	71 a	+81	+23
80	923 a	313 ab	73 a	69 a	+70	+19
120	701 b	385 a	48 b	72 a	+12	+24

Mean values within the same column and year followed by the same letter are not significantly different at  $P \leq 0.05$ . <sup>1</sup>: Percentage differences in canopy cover between the treated and unamended soil.

**Table 5.** Effects of biosolid doses on the main macronutrient, micronutrient and heavy metal concentrations of plant aerial tissues one (1998) and five (2002) years after its application

Elements	Biosolid doses (Mg ha <sup>-1</sup> )							
	1998				2002			
	0	40	80	120	0	40	80	120
N (g kg <sup>-1</sup> )	15.6 b	17.6 b	22.4 a	23.7 a	10.4	13.7	13.2	14.3
P (g kg <sup>-1</sup> )	0.93 c	2.17 b	2.40 a	2.09 b	0.94 c	2.15 ab	2.16 ab	2.76 a
K (g kg <sup>-1</sup> )	9.22 b	11.4 a	11.6 a	12.4 a	9.98	12.2	10.8	11.1
Zn (mg kg <sup>-1</sup> )	19.6 b	18.7 b	31.8 a	22.0 b	26.9	25.3	21.8	26.3
Cu (mg kg <sup>-1</sup> )	8.0	9.14	6.19	8.68	10.5	9.78	7.19	8.39
Cd (mg kg <sup>-1</sup> )	< 0.1	< 0.1	< 0.1	< 0.1	< 0.1	< 0.1	< 0.1	< 0.1
Ni (mg kg <sup>-1</sup> )	0.20	0.21	0.28	0.28	0.29	0.31	0.43	0.42
Cr (mg kg <sup>-1</sup> )	nd	nd	nd	nd	nd	nd	nd	nd
Pb (mg kg <sup>-1</sup> )	2.02	2.54	3.00	2.26	3.41	3.38	2.99	3.17

Mean values within the same line and year followed by the same letter are not significantly different at  $P \leq 0.05$ ; nd: not detected.

uptake of Cr, Cd, Ni and Pb was not influenced by treatments. The concentration of Zn in aerial plant tissues was only significantly different at the 80 Mg ha<sup>-1</sup> dose one year after biosolid application. No differences were observed between treatments with respect to Cu concentrations on the two sampling dates, or Zn concentrations at the end of the experiment. Metal contents did not correlate with the amount of BS applied. Zn and Cu concentrations of plants in the treated plots were low and close to deficiency levels.

## Discussion

Generally, plant nutrient concentrations in soil were higher in treated plots than in the control plots (Table 2). The slight EC increase was not sufficient to inhibit plant growth (Porta *et al.*, 1999). A greater increase in SOC was expected because the biosolids contained 253 g kg<sup>-1</sup> of organic carbon (Table 1). The addition of biosolids stimulates soil microbial activity and much of the added organic matter and also the indigenous soil organic matter may be rapidly mineralised (Woods *et al.*, 1987). On the other hand, the increases in nutrient levels in the treated plots accelerated plant establishment since within a few months after amendment, they were covered by spontaneous vegetation. This would have a feedback effect, leading to the addition of more organic carbon from plant residues (Nortcliff, 1998), which could contribute to the slight increase in SOC observed for the highest dose at the end of the experiment.

Soil NO<sub>3</sub>-N levels were relatively high, particularly one year after biosolid applications. Rates of more than 80 Mg ha<sup>-1</sup> could, therefore, pose a potential surface water contamination hazard at times of significant runoff. All the treated plots had soil test P (Olsen) levels above optimum levels for plant growth. Even at the end of the experiment, available P was about 3.5 and 6-fold greater in the treated plots than in the control plot. The increase in soil P concentration for the low BS rates could alleviate an apparent P deficiency in the study site soil, mainly due to its high CaCO<sub>3</sub> content. Bofras *et al.* (2000) also reported an increase in available soil P when biosolids were applied. However, surface water quality could be seriously impaired by excessive NO<sub>3</sub>-N and P levels. Therefore, the intermediate and high dose of biosolids is not recommended owing to a potential contamination

hazard for surface waters, particularly during rainfall periods. In semi-arid soils, water tables are generally deep and the probability of leached nutrient or toxic elements reaching them is low. This factor reduces the risk of groundwater pollution. On average, treated plots had lower concentrations of soil K than control plots. Biosolid application had a negligible effect on extractable soil K, because K is water-soluble and removed from the biosolids during dewatering (Shober *et al.*, 2003).

The increase in the content of DTPA-extractable metals may have been due to organic matter decomposition and to the incorporation of soluble organic matter into the soil (MacBride, 1995). In spite of the high doses of biosolids applied, extractable heavy metal concentrations were below the levels considered toxic to plants (Madejón *et al.*, 2003). The high pH and the CaCO<sub>3</sub> content of the soil justify the low levels of the DTPA-extractable heavy metals found. This is important since concerns surrounding heavy metals frequently limit the use of biosolid applications. Therefore, on the basis of the heavy metal contents, all three biosolids rates could be safely applied to this soil with no fear of phytotoxic effects.

A remarkable change of native plants has been observed after the application of BS, with a reduction of perennial species and an increase in annual plants. These findings are similar to those obtained by Biondini and Redente (1986), who reported that plant diversity decreases in the presence of high nutrient levels. No similar pattern was found by Pierce *et al.* (1998), who reported that perennial grasses and shrubs remain the dominant species in a semiarid shrubland after biosolid treatment.

The total canopy cover for the 120 Mg ha<sup>-1</sup> treatment was not significantly different from that of the control plots one year after the application. Partial biosolid decomposition was observed at the first sampling time, possibly indicating that some biosolids remained on the soil surface, acting as a physical barrier to plant emergence.

Despite the total canopy cover increasing significantly for all treatments, a significant decrease in the percentage of relative canopy cover was observed in all biosolid-treated plots between the first and the fifth year. Although the highest rate produced a total biomass greater than the control one year after the application, a combination of different (physical and chemical) factors associated with this application may



have partially limited the plant yield compared to the other BS rates. No significant relationship was observed between biomass production and biosolid rate. However, the increases obtained could be attributed directly to increased soil fertility (Table 2). Despite the supply of N, P and other essential nutrients — possibly, the major factors causing the increase in biomass — an improvement of soil physical and biological properties cannot be discounted. Plant production in the last year increased slightly as BS rates increased, probably because the inhibitor effects observed in the first year decreased over time. Apparently, even after five years, sufficient quantities of nutrients from the biosolid-amended soils were still available for plant use. The development and maintenance of a vegetation cover is an important factor in soil quality since it protects it from erosive processes and supplies organic C as an energy source for microorganisms through root exudates and plant remains (Brockway *et al.*, 1998).

The degraded soil used in this study required biosolid doses of up to 80 Mg ha<sup>-1</sup> to improve its fertility. Therefore, one initial large application would facilitate the rapid establishment of native vegetation and, consequently, could contribute to reducing soil erosion. Higher rates may result in a significant improvement in many soil properties as well as in plant production, but they pose a potential P and N-NO<sub>3</sub> contamination hazard to runoff, especially shortly after application.

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