The effects of fertilization with anaerobic, composted and pelletized sewage sludge on soil, tree growth, pasture production and biodiversity in a silvopastoral system under ash (Fraxinus excelsior L.)

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Abstract
In silvopastoral systems, tree growth and the composition and productivity of pasture can be modified by management practices such as initial fertilization when tree seedlings are more sensitive to understorey competition. The aim of this study was to compare the effects of fertilization with different types of sewage sludge (anaerobic sludge, composted sludge and pelletized sludge), using different rates of incorporation and mineralization with traditional treatments (with and without mineral fertilizers) on the growth of newly established ash (Fraxinus excelsior L.) and on pasture development, to obtain sustainable management practices that enhance the growth of both components. Soil characteristics, tree growth, sward composition and pasture development were modified differently according to the type of sewage sludge used, and for similar total nitrogen inputs. Anaerobic sludge had a higher initial effect on both tree and pasture productivity. Pelletized sludge sustained better tree and pasture production. Composted sludge was found to be the most appropriate treatment for improving soil characteristics over the long term on sandy soils. It was concluded that pelletized sludge should be promoted because it enhances productivity, allows for better nutrient recovery and is less costly to store and apply compared with anaerobic sludge and composted sludge. No toxic concentrations of Zn or Cu were found in plants or in the soil despite higher concentrations being present in the applied sludge than in soil.

Keywords: agroforestry, Zn, waste, afforestation, land use change, Spain

Introduction
Silvopastoral systems are a type of agroforestry system in which trees and grazing are combined, resulting in benefits for wood production (with long-term economic returns) and livestock (with short-term economic returns) (Rigueiro-Rodríguez et al., 2005). Agroforestry systems are sustainable land management techniques that are promoted by the EU [Council Regulation 1698/2005 (EU, 2005)]. Agroforestry systems have also received favourable evaluations from farmers in Europe; for example, a sample of 214 farmers interviewed in fourteen areas of Europe, half indicated they would attempt silvo-arable agroforestry on their farm (Graves et al., 2008).

In the early stages of the development of silvopastoral systems established through afforestation, competition between trees and pasture can be high (Nair and Graetz, 2004; Rigueiro-Rodríguez et al., 2008a). In newly established systems, adequate management should aim to optimize silvopastoral outputs through the selection of trees and pasture species as well as through fertilization inputs. Shrub development should be avoided through frequent clearing in order to avoid shrub–tree competition and fire risk (Rigueiro et al., 2009). Moreover, in early stages of establishment pasture–tree competition should be avoided, either through mulching or by providing forage for grazing animals (Wagner et al., 2006).

The European Ash (Fraxinus excelsior L.) is a widely distributed tree species that integrates well into silvo-pastoral systems in the Atlantic biogeographic region of Europe (McAdam and Hoppé, 1996; McAdam and Sibbald, 2000). European ash trees possess apical
dominance and deep roots that avoid root competition between trees and pasture; these roots enhance nutrient recovery from the deep soil layer up into the system, making them compatible with silvopastoral systems. Moreover, as a deciduous species, it allows better light penetration than conifers during the autumn and early spring, and provides shading during the summer, thereby reducing evapotranspiration and thus enhancing pasture production when compared with pasture under conifers or on open pasture sites. The open crowns of ash trees also allow light to reach the pasture surface, and they do not intercept much rain (McEvoy, 2004). The most appropriate pasture species for silvopastoral system implementation are those that are well adapted to shading, such as Dactylis glomerata L. (Mosquera-Losada et al., 2001, 2006). However, legume species also enhance pasture quality and production as well as tree growth (Whitehead, 1995; López-Díaz et al., 2009).

In Galicia, the natural soils have low fertility due to their acidity (Zas and Alonso, 2002). This acidity implies a high concentration of saturated aluminium in the exchange complex and low cation and phosphorus availability (Prasad and Power, 1997; Rigueiro-Rodríguez et al., 2007). The EU promotes the use of sewage sludge as a fertilizer because of its specific organic matter and content of macronutrients, particularly N (MMA, 2006). However, a higher concentration of heavy metals (mainly Zn and Cu) in sewage sludge than is normally found in soil, as well as long-term sludge loadings, limits the use of sewage sludge according to the Spanish (R.D. 1310/1990; BOE, 1990) and European Directives (86/278/CEE; DOCE, 1986) in order to prevent harmful effects on soil and vegetation, and on animal and human health.

Anaerobic digestion and composting are two sewage sludge stabilization processes which are promoted by the EU (EEA, 2000) before the sludge is used as a fertilizer in agriculture. However, sewage sludge stabilized by these processes contains a high proportion of water. Pelletized sewage sludge is derived from the thermic treatment of anaerobic digested sewage sludge in order to reduce water content to 2%, which consequently reduces storage, transport and spreading costs compared with anaerobic or composted sludge (COM) (Mosquera-Losada et al., 2009). Each type of sewage sludge has different characteristics, nutrient contents (Mosquera-Losada et al., 2009) and rates of incorporation into the soil according to the treatment stabilization (EPA, 1994) and the specific local climate.

The aim of this study was to evaluate the effects of municipal sewage sludge that has been stabilized by either anaerobic digestion, composting or pelletization, on changes in soil chemical properties, tree growth, understorey production, biodiversity in terms of sward botanical composition and quality of pasture compared with treatments (receiving either mineral fertilizers or no fertilization) in a silvopastoral system under F. excelsior L. during a 4-year period.

Materials and methods

Characteristics of the study site
The experiment was conducted in A Pastoriza (Lugo, Galicia, NW Spain, European Atlantic Biogeographic Region; t 43° 14’ N, 7° 21’ W; 550 m a.s.l.). Figure 1 shows the mean monthly precipitation and temperatures for 2005, 2006, 2007 and 2008 and the previous 30-year mean. Total annual rainfall was 824·3, 1157·5, 734·4 and 1222·3 mm in 2005, 2006, 2007 and 2008 respectively. Very low precipitation was observed in 2005 and 2007 compared with the 30-year mean. There were periods of drought from April to July 2006 and from June to October 2008, which would have been unfavourable for tree growth and pasture production during these periods. The annual mean temperature was mild (12°C).

The experiment was carried out on abandoned agricultural land. The soil texture at the start of the experiment was sandy (91·81 sand, 4·92 silt and 3·27% clay) and pH (water) was moderately acidic at 5·6. Although the initial soil Mehlich 3-P concentration (35·1 mg kg⁻¹) can be considered high (Sawyer et al., 2008), no risk of phosphorous leaching has been found in the area because of the high soil acidity, which leads to a soil P storage capacity (Mosquera-Losada et al., 2008). All heavy metal concentrations in the soil (Table 1) were below the maximum threshold for using sewage sludge as fertilizer as specified by the European Union Directive 86/278/CEE (DOCE, 1986) and Spanish legislation under R.D. 1310/1990 (BOE, 1990).

Experimental design
At the beginning of the experiment, the soil was double ploughed to a depth of 50 cm, which is traditional practice in the area, and the pasture was sown with a mixture of D. glomerata L. var. Artabro (12·5 kg ha⁻¹), Lolium perenne L. var. Brigantia (12·5 kg ha⁻¹) and Trifolium repens L. var. Huia (4 kg ha⁻¹) in autumn 2004. Bare-rooted 1-year old plants of F. excelsior L. (typical height of 25 cm) were planted at a density of 952 trees ha⁻¹, with a distance between rows of 3·0 × 3·5 m. The experimental design was a randomized block with three replicates and five treatments distributed in experimental units of 168 m² with twenty-five trees arranged in a frame of 5 × 5 trees. The treatments consisted of (i) no fertilization (NF); (ii) mineral fertilization (MIN) with 500 kg ha⁻¹ 8·24:16 compound fertilizer (N-P₂O₅-K₂O) at the beginning of the growing
Figure 1 Monthly precipitation and mean temperatures for the study area in 2005, 2006, 2007 and 2008, and mean data for the last 30 years. T: mean monthly temperature (°C); T30: 30-year mean temperature; P, mean monthly precipitation (mm); and P30: 30-year mean precipitation.

Figure 2 Soil pH in water (a), exchange capacity [cmol (+) kg⁻¹] (b), aluminium saturation percentage in soil exchange complex (%) (c) and amount of Ca extracted by Mehlich (%) (d) in each treatment in the years 2006, 2007 and 2008. NF, no fertilization; MIN, mineral; ANA, anaerobic sludge; COM, composted sludge and PEL, pelletized sludge. Different letters indicate significant differences between treatments. Vertical lines indicate mean standard error.
season and 40 kg N ha\(^{-1}\) after first harvest (MIN); (iii) fertilization with anaerobically digested sludge with an input of 320 kg total N ha\(^{-1}\) before pasture sowing (ANA); (iv) fertilization with composted sewage sludge with an input of 320 kg total N ha\(^{-1}\) before pasture sowing (COM) and (e) application of pelletized sewage sludge, which involved a total input of 320 kg total N ha\(^{-1}\) split into 134 kg total N ha\(^{-1}\) just after pasture sowing in 2004, and 93 kg N ha\(^{-1}\) at the end of 2005 and 2006, which correspond to similar total inputs of 320 kg total N ha\(^{-1}\) (PEL). Based on previous experiments in the area and EPA (1994) recommendations, it was assumed that approximately 0.25 of the total nitrogen would be mineralized in the first year if compost or anaerobic sludge was added, and therefore approximately 80 kg N ha\(^{-1}\) year\(^{-1}\) was applied. There was no available information about the pellet fertilizer, but a total similar input of 320 kg total N ha\(^{-1}\) during the experiment was used.

### Sewage sludge

Anaerobically digested sewage sludge, COM and pelletized sludge (PEL) were taken from municipal waste treatment plants at Lugo, Valladolid and Madrid respectively. The calculation of the required amounts of sludge was conducted according to the percentage of the total nitrogen and dry-matter contents EPA (1994), taking into account the European Union Directive 86/278/CEE (DOCE, 1986) and Spanish regulation R.D.1310/1990 (BOE, 1990) regarding heavy metal concentrations for the application of sewage sludge on to soil. The composition of the sewage sludge is summarized in Table 2. The sludge used in the present experiment had a similar composition to the mean composition of the sludge described for plants all over Spain (Mosquera-Losada et al., 2009). The proportion of N in the sludge was higher than the P and K concentrations in the sludge. As the calculations of the required amounts of sludge were based on N and pasture nitrogen, and as phosphorous needs are similar, contamination by phosphorus is not likely to occur. Furthermore, the acidity of the soil would prevent phosphorous from leaching from the soil (Mosquera-Losada et al., 2008).

### Field samplings and laboratory determinations

Soil samples were collected to a depth of 25 cm, as described in the RD 1310/1990 (BOE, 1990) in February 2006, January 2007 and January 2008. In the laboratory, soil pH was determined in water (1:2.5) (Guitián and Carballás, 1976). The aluminium concentration in the exchange complex and the exchangeable cations were determined by extraction with 0.3 M BaCl\(_2\). The K, Ca, Mg and Na exchangeable concentrations were measured with a Varian 220FS Spectrophotometer (Varian, Walnut Creek, CA, USA) using the atomic emissions for K and Na and the absorptions for Ca and Mg. Aluminium concentrations were analysed after valorization with 0.01 N NaOH, using phenolphthalain (1%) in an alcohol-based solution as an indicator (Mosquera and Mombiela, 1986). The effective exchange capacity (EEC) was determined by taking the sum of K\(^+\) Ca\(^+\) Mg\(^+\) Na\(^+\) Al and the aluminium percentage saturation using the quotient Al/EEC. The total soil Ca, Cu and Zn concentrations were determined after microwave digestion (CEM, 1994), and the available Ca, Cu and Zn were measured after extraction with Mehlich (1985) with the Varian 220 FS Spectrophotometer using atomic absorption.

Base tree height and diameter were measured with a graduated ruler and a calliper, respectively, at the beginning of 2005, 2006, 2007 and 2008.

Pasture production was determined randomly by taking four samples of pasture at a height of 2.5 cm per plot (0.3 × 0.3 m) using an electric hand clipper in August and December 2005; June and December 2006; April, June and December 2007 and May and December 2008 before all of the plots were grazed by mature sheep (Galician breed Raza ovella galega) at a stocking rate of fifty sheep over the whole experimental area (2520 m\(^2\)) 1 week after sampling. Two pasture samples were dried for 48 h at 60°C and weighed to estimate pasture production. The other two samples were separated by hand to determine the proportions of the different plant species and the senescent material, and then dried (60°C for 72 h) to determine the botanical composition on a dry weight basis. Annual abundance diagrams (Magurran, 1988) were made which excluded senescent material. The total Zn in the harvested

### Table 1  Heavy metal concentrations in the soil at the beginning of the experiment and legal limits established by European Directive 86/278 and Spain R.D. 1310/1990.

|         | Cd (mg kg\(^{-1}\)) | Cu (mg kg\(^{-1}\)) | Cr (mg kg\(^{-1}\)) | Ni (mg kg\(^{-1}\)) | Pb (mg kg\(^{-1}\)) | Zn (mg kg\(^{-1}\)) |
|---------|---------------------|---------------------|---------------------|---------------------|---------------------|---------------------|
| Initial soil concentration | –                   | 5.8                 | 4.1                 | 2.1                 | –                   | 20.6                |
| Spanish law limits | 1–3                 | 50–210              | 100–150             | 30–112              | 50–300              | 150–450             |

Limits depend on soil pH (minimum: soil pH < 7, maximum: soil pH > 7).

Concentrations of Cd and Pb were below detection limit of the technique used for determination.

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pasture herbage was determined by microwave digestion with nitric acid (CEM, 1994).

**Statistical analysis**

The data were analysed using ANOVA, and the differences between the averages were determined using the LSD test (at the alpha level of 0.05) using the SAS statistical package (SAS, 2001). All soil variables [soil water pH, EEC, Al saturation percentage, total (Cu and Zn) and Mehlich (Ca, Cu and Zn) soil concentrations] were analysed using the ANOVA model ($Y_{ij} = \mu + B_i + T_j + Y_k + BT_{ij} + TY_{jk} + BY_{ik} + \epsilon_{ijk}$), where $\mu$ is the mean; $B_i$ is block (two freedom degrees); $T_j$ is treatment (four freedom degrees); $Y_k$ is year (three freedom degrees), $BT_{ij}$ (block treatment interaction (six freedom degrees), $TY_{jk}$ is treatment year interaction (twelve freedom degrees), $BY_{ik}$ is block year interaction (six freedom degrees) and $\epsilon_{ijk}$ is the error term. All tree and pasture variables were analysed using the ANOVA model ($Y_{ij} = \mu + B_i + T_j + \epsilon_{ij}$), where $\mu$ is the mean; $B_i$ is block (two freedom degrees); $T_j$ is treatment (four freedom degrees); and $\epsilon_{ij}$ is the error term. ANOVA type III errors were taken into account to determine significances.

**Results**

**Soil chemical properties**

$\text{pH in water, effective exchange capacity (EEC), aluminium saturation percentage and Ca extracted by Mehlich}$

Composted sludge (COM) increased the mean soil pH ($P < 0.01$) and the amount of mean Ca extracted by
Mehlich \( (P < 0.05) \), but the effect on ECC was not significant \( (P > 0.05) \). Because of these modifications (Fig 2), the mean percentage of aluminium saturation decreased with this type of sludge application \( (P < 0.01) \). There was a significant effect of year \( (P < 0.001) \) on soil pH \( (2006: 5.6^a; 2007: 5.5^b \text{ and } 2008: 5.2^c) \); on EEC \( (2006*: 8.6^a; 2007: 10^b \text{ and } 2008: 8^c) \) expressed as cmol (+) 100 g \(^{-1}\) soil; on aluminium saturation percentage \( (2006*: 1.7^a; 2007: 2.4^b \text{ and } 2008: 1.1^c) \) and amount of Ca extracted by Mehlich \( (2006: 0.69^a; 2007: 0.25^b \text{ and } 2008: 1.43^a) \) expressed as mg kg \(^{-1}\) (in all cases different superscript letters indicate significant differences between years). The soil pH and aluminium saturation percentage were lower at the end of the study \( (2008) \) than at the beginning of the study \( (2006) \), whereas the EEC and the amount of Ca extracted by Mehlich were higher in 2008 than in 2006.

**Total and Mehlich extracted Cu and Zn**

The total and Mehlich soil levels of Cu and Zn in 2006, 2007 and 2008 are presented in Figure 3. The mean total Cu and Zn were significantly affected by the treatments \( (P < 0.001 \text{ and } P < 0.05 \text{ respectively}) \). There was a significant effect of year on total Cu \( (P < 0.0001) \), total Zn \( (P < 0.0001) \) and on the Zn extracted by Mehlich \( (P < 0.001) \). In the case of the Cu extracted by Mehlich, the interaction of treatment \( \times \) year was significant \( (P < 0.01) \). All of the variables were increased by COM, and total Zn was increased by anaerobic sludge (ANA) compared with the other treatments. The total soil Cu and Zn found in the experiment were below the maximums set by Spanish regulations for the use of sewage sludge in agriculture for acid soils (Cu: 50 mg kg \(^{-1}\) and Zn: 150 mg kg \(^{-1}\) [R.D. 1310/1990, (BOE, 1990)]. The concentrations of total Cu and Zn and Zn extracted by Mehlich were lower in 2008 than in 2006.

**Tree height and diameter**

The average tree heights for each treatment in 2005, 2006, 2007 and 2008 are shown in Table 3. In all 2 years of the study, tree height was significantly modified by fertilization treatment (Table 4). Initially, the tree height was higher in all treatments that received organic fertilizer (ANA, COM and PEL) than the MIN or NF treatments. Tree height in 2005 and 2006 was increased by anaerobic (ANA) and PEL compared with the mineral fertilization (MIN) and no fertilization (NF) treatments. Tree height in 2005 and 2006 was increased by anaerobic (ANA) and PEL compared with the mineral fertilization (MIN) and no fertilization (NF) treatments, but this positive effect was only found in those plots receiving PEL in 2007 and 2008. Trees growing in NF and MIN treatments had the lowest height during the 4 years of the experiment.

Tree diameter (Table 3) was significantly modified by fertilization treatment in all 4 years of the study, with the exception of 2007, (Table 4). In 2005, 2006 and 2007, tree diameter was increased by ANA compared with the other treatments (NF, MIN, ANA, COM and PEL). In 2008, the COM and PEL treatments resulted in larger tree diameters than MIN treatment.

![Figure 3](image_url)
Table 3 Mean values for tree height (cm), tree diameter (mm) and pasture production (t ha\(^{-1}\)) under the different fertilization treatments in the years 2005–2008.

| Year | NF   | MIN  | ANA  | COM  | PEL   | s.e. |
|------|------|------|------|------|-------|------|
| Tree height (cm) |   |      |      |      |       |      |
| 2005 | 32.88 | 32.33 | 38   | 34.85 | 38.05 | 0.69 |
| 2006 | 33.65 | 33.08 | 39.33 | 35.8  | 38.1  | 0.66 |
| 2007 | 41.59b | 42.58b | 48.89ab | 47.65ab | 53.55a | 1.05 |
| 2008 | 114.36ab | 107.56b | 103.63b | 121.84ab | 135a | 3.51 |
| Tree diameter (mm) |   |      |      |      |       |      |
| 2005 | 5b  | 4.83b | 5.89a | 4.7b  | 5b    | 0.14 |
| 2006 | 5.12b | 5.42b | 6.56a | 4.8b  | 5.15b | 0.14 |
| 2007 | 6.29  | 5.92  | 7.54  | 6.45  | 6.09  | 0.14 |
| 2008 | 12.55ab | 10.81b | 12.29ab | 13.68a | 14.04a | 0.37 |
| Pasture Production t ha\(^{-1}\) |   |      |      |      |       |      |
| 2005 | 1.96b | 2.98a | 3.17a | 2.81ab | 3.36a | 0.14 |
| 2006 | 4.91  | 3.98  | 5.15  | 4.82  | 5.24  | 0.18 |
| 2007 | 5.66ab | 5.88ab | 4.81b | 5.33ab | 6.55a | 0.18 |
| 2008 | 3.96  | 4.25  | 3.37  | 3.94  | 4.51  | 0.14 |

Different letters indicate differences between treatments within the same year that were significant at \(P < 0.05\).
s.e., mean standard error; NF, no fertilization; MIN, mineral fertilizer; ANA, anaerobic sludge; COM, composted sludge and PEL, pelletized sludge.

**Pasture understorey**

**Pasture production**

The annual pasture production for the different fertilization treatments in 2005, 2006, 2007 and 2008 is summarized in Table 3. Significant differences were detected between the treatments in all years except 2006 and 2008 (Table 4). The highest levels of pasture production were found in 2007 (4.8–6.5 t ha\(^{-1}\)), whereas the lowest values were recorded in 2005 (1.9–3.3 t ha\(^{-1}\)). In 2005, there was a positive response to organic fertilization (ANA, COM and PEL) and inorganic fertilization (MIN) compared with no fertilization (NF), but this effect was only maintained in the PEL treatment until the end of the experiment. In the final years of the study (2007–2008), the annual pasture production tended to be lower than that of the PEL when the ANA was applied to the soil.

**Table 4** ANOVAs for tree height, diameter and pasture production in 2005, 2006, 2007 and 2008.

| Year | Height | Diameter | Pasture production |
|------|--------|----------|---------------------|
| Treatment effect | 2005 | ns | * | * |
| 2006 | ns | ** | ns |
| 2007 | ** | ns | * |
| 2008 | * | * | ns |

ns, not significant.
* \(P < 0.05\); ** \(P < 0.01\); *** \(P < 0.001\).

**Pasture abundance diagrams**

Figure 4 shows the abundance diagrams for the different fertilization treatments in 2005, 2006, 2007 and 2008. Agrostis capillaris L., D. glomerata L., Holcus lanatus L. and L. perenne L. were present in the sward in all treatments and in all years. The treatments with a high number of species were associated with a higher proportion of dicotyledonous species. Agrostis capillaris L. and D. glomerata L. were the most dominant species throughout the study. The proportion of A. capillaris L. was always over 75% in the NF treatment and approximately 50% in the other treatments and years, with the exception of mineral (MIN) and ANA fertilization in the final year. However, the mineral (MIN) and ANA treatments had higher proportions of D. glomerata L. in the first years of the study. In the PEL treatment, codominance between D. glomerata L. and A. capillaris L. was evident throughout the experiment. In terms of species richness, the ANA treatment had the lowest number of species at the start of the experiment, but the PEL treatment had the lowest number from 2006 onwards. The COM treatment in the second year of the experiment was an exception to this trend, exhibiting the lowest number of species. The PEL treatment was dominated by monocotyledonous species in the final year than the remaining treatments (NF, MIN, ANA and COM).

**Zn and Cu concentrations in pasture**

The concentration of Zn in the pasture was significantly affected by treatments in April 2007 (\(P < 0.01\), and...
May 2008 ($P < 0.05$) (Figure 5). However, Cu levels in the pasture were not affected by any treatments (data not shown). In April 2007, the concentration of Zn in the pasture was higher when the ANA and PEL treatments were applied. In May 2008 the concentration of Zn in the pasture increased in the ANA treatment compared with the COM treatment. There were no responses to the treatments in terms of the pasture concentrations of Zn after harvests made in 2005, 2006, December 2007 and December 2008.

**Discussion**

This experiment demonstrates that there are very few negative effects of sewage sludge application, either from toxic metal loading or in terms of productivity in the specific edaphoclimatic conditions of this study. The growth of trees and pasture production can be limited by low soil pH, EEC and a high aluminium saturation percentage; however, they can be improved by applications of high pH organic waste and by fertilization (Mosquera-Losada et al., 2009). The soil pH in this experiment ranged from acidic (5.1–5.5) to moderately acidic (5.6–6.0) (Slattery et al., 1999); conditions which usually indicate deficiencies in the availability of cations and, therefore, limit pasture production (Whitehead, 2000). Moreover, the soil EEC was low and usually below 10 cmol (+) kg$^{-1}$ soil, which could be explained by the high sand proportion in the soil of this experimental site (Brady and Weil, 2008). However, the aluminium saturation percentage was below 25%, which indicates the optimum characteristics for pasture and tree growth in Galicia (Mombiela and Mateo, 1984). Aluminium saturation was also found to have a

![Figure 4](image-url)  
*Figure 4* Abundance diagrams for the treatments applied in the years 2005, 2006, 2007 and 2008. Ac: Agrostis capillaris L.; Ac: A. curtisii Kergr. Cd: Cardius spp.; Cap: Capsella bursa-pastoris L.; Cen: Centaurea limbata Hoffmann. /L; Cen: Cerastium glomerata Thull; Di: Digitalis purpurea L.; Dg: Dactylis glomerata L.; Gr: Geranium rotundifolium L.; Hl: Holcus lanatus L.; Hm: Holms mollis L.; J: juncus effusus L.; Lp: Lolium perenne L.; Ma: Matricaria spp.; Mu: Musgo; Or: Ornithopus compressus L.; Pl: Plantago lanceolata L.; Poa: Poa pratensis L.; Ps: Pseudanthematherum longifolium (Thorey) Rouy; Ru: Rumex acetalosa L.; Rnc: Ranunculus repens L.; Ro: Rumex obtusifolius L.; Rp: Raphanus raphanistrum L.; Ru: Rubus spp.; Se: Senecio jacobea L.; So: Sanchus oleraceus L.; St: Stellaria media L. (Vill); Ta: Taraxacum officinale Weber; Tc: Trifolium campestre Schreber; Tr: Trifolium repens L.; Ul: Ulex europaeus L.; Ve: Veronica agrestis L.

![Figure 5](image-url)  
*Figure 5* Concentrations of Zn in pasture (mg kg$^{-1}$) under the different fertilizer treatments in the harvests of April 2007 and May 2008. NF, no fertilization; MIN, mineral; ANA, anaerobic sludge; COM, composted sludge and PEL, pelleted sludge. Different letters indicate significant differences between treatments. Vertical lines indicate mean standard error.
diachronic variation, which usually improved from the start to the end of the experiment. The EEC increase could be explained by the tilling that occurred at the start of the experiment. The soil aggregate structure was probably destroyed by the tilling process (Dexter, 1988), but the structure is likely to have been improved with the addition of fertilizer and with the establishment of pasture and trees, and the potential for increased organic matter input into the soil. The soil pH, however, was lower at the end of the experiment, despite the increase in available Ca and the reduction in available Al caused by sludge incorporation into the soil (Smith, 1996); this could be explained by the increase in the proportion of H+ related to the higher EEC. The soil acidity increase derived from the H+ can be explained by the cation extraction of the crops, the mineralization process (NH4+ is transformed in NO3- and H+ is released in the nitrification process) and the high mean rainfall of the area, which promotes cation leaching (Whitehead, 1995).

Composted sludge (COM) increased soil fertility compared with other treatments. The pH and mean amount of available Ca were improved by the COM treatment, which was expected and is explained by the different composition of the sludge and the rate of mineralization (Mosquera-Losada et al., 2009). Applied doses of sewage sludge with compost were increased in order to meet the nitrogen required by EPA (Environmental Protection Agency) (1994) recommendations because the COM composition revealed a lower concentration of nitrogen than the anaerobic and PEL. The COM also had a low mineralization rate, as indicated by the EPA (Environmental Protection Agency) (1994) (the mineralization rate is approximately 20% for anaerobic compared with 10% for COM in the first year). Moreover, the pH and available Ca were increased in the COM treatment because of the higher concentrations of Ca, K and Mg compared with anaerobic and PEL (excepting Ca). The COM treatment applied approximately 1835.8 kg Ca ha⁻¹, 99.53 kg K ha⁻¹ and 541.9 kg Mg ha⁻¹; meanwhile, only 90.51 kg Ca ha⁻¹, 28.66 kg K ha⁻¹ and 67.88 kg Mg ha⁻¹ were added with the ANA treatment, and 776.85 kg Ca ha⁻¹, 23.87 kg K ha⁻¹ and 158.96 kg Mg ha⁻¹ were the soil inputs with the PEL treatment. The higher rate of Ca and Mg with the COM also explains the reduction in the aluminium saturation percentage for this treatment (Smith, 1996; Prasad and Power, 1997; Speir et al., 2004). In previous studies, the anaerobic sewage sludge effect on the soil pH and EEC was found to depend on the previous initial soil pH. When the initial soil pH was high, anaerobic sludge inputs increased acidity as extraction was promoted (Mosquera-Losada et al., 2006), but when the soil pH was very low (4.5), as described by López-Díaz et al. (2007), a positive effect of sewage sludge application on soil pH was found. In this study, although the soil fertility was enhanced by COM treatment, tree growth and pasture production were not promoted. The lack of enhancement in tree growth and pasture production was probably due to the lower mineralization rate and the N availability of COM treatment compared with that of the anaerobic (ANA) or PEL as described by the EPA (1994). Warman and Termeer (2005) found better crop production in response to ANA than to COM sludge. The mineralization rate depends on the local climate and on the mineralization rates (EPA, 1994), and in our case, the COM treatment seems to have an initial lower mineralization rate compared with the ANA treatment. As a result, the effect of COM on pasture production and tree growth could become apparent over a longer period, as was seen with tree diameter. The soil fertility is differently affected by sewage sludge application, and the effect is dependent on the initial soil pH and on the type of the sewage sludge applied.

At the end of the 4-year study, the heights of ash trees varied from 104 to 135 cm and stem diameters from 10.81 to 14.04 mm. The tree heights were within the range (23–275 cm) reported in a study carried out in UK after 3 years of experimentation (Mwase et al., 2008). The initial tree diameters in our study were also similar to those found by the same authors (15.3 mm). A positive effect was found for the ANA treatment in the two first years of the experiment; this trend has also been described for Populus × euroamericana (Rigueiro-Rodríguez et al., 2008b) and Pinus radiata D. Don, under soil conditions that were initially either very acid (López-Díaz et al., 2007) or neutral (Mosquera-Losada et al., 2006). In other work, Muys et al. (2004) and Weber-Blaschke and Rehfüess (2002) demonstrated that soil fertility improvements led to higher F. excelsior growth, relative to the control, on sites with sandy loam and loam soils respectively. In both the present experiment and in these previous studies, the positive effect of the ANA treatment could be attributed to the soil fertility and water retention improvements caused by anaerobic fertilizer applied at the establishment of the plantation (Wolsteholme et al., 1992). However, at the end of the experiment, only the PEL treatment showed a significant increase in tree height diameter and the COM treatment in tree diameter compared with the MIN treatment, which could be explained by the enhancement of pasture growth and increased competition between trees and pasture caused by the MIN treatment as compared with the NF treatment.

Annual pasture production was below the common levels in this region due to the droughts in 2005, 2006, 2007 and 2008 (mean monthly precipitation of these years were lower than mean precipitation over the previous 30 years) and the low temperatures at the
beginning of the year. Pasture production was significantly increased by organic or inorganic fertilization as found by Mosquera-Losada et al. (2006) and by López-Díaz et al. (2009) in agrarian soils in Galicia afforested with P. radiata D. Don with a pH close to neutral (pH 6.8) and a slightly acidic pH (pH 6.3) respectively. At the end of this study, only the PEL treatment was found to have positively affected annual pasture production by the PEL treatment because of the annual application; however, no residual effect was found as a result of the COM or ANA treatments (Rigueiro-Rodríguez et al., 2000; López-Díaz et al., 2007).

The lower proportion of A. capillaris L. and the higher proportion of D. glomerata L. could be associated with low and high soil fertility, as demonstrated in soils with a pH below 4.97 (Mosquera-Losada et al., 2001). Modifications to soil fertility also caused variations in the dominance of these two species, with a high proportion of D. glomerata L. initially associated with the MIN and ANA treatments followed by a switch in the botanical composition to over 75% A. capillaris L. by the end of the experimental period.

Fertilizer treatment effects on soil fertility explain tree and pasture production, and affect biodiversity. The ANA treatment initially increased soil fertility, as demonstrated by higher tree growth, greater pasture production, a lower number of species and a higher proportion of D. glomerata L. (Mosquera-Losada et al., 2001) compared with the other treatments (NF, MIN, COM and PEL). These results are of particular interest in areas in which the initial tree development is important to guarantee tree survival. However, there is a reduction in tree and pasture growth when soil fertility is depleted, probably because the nutrient requirements of trees are greater than in the other treatments and because the capacity for nutrient retention in the ANA treatment is lower than in the other treatments because nutrient leaching occurs. On the contrary, the PEL treatments sustained better tree and pasture production over the long term as the nutrient inputs were supplied as split doses through time. The higher fertility of soils fertilized with PEL probably explains the dominance of monocotyledonous species and the low number of species throughout the experiment.

It is important to be aware of the effects of sewage sludge on the concentrations of Cu and Zn in the soil in plants, because Cu and Zn are commonly present in higher proportions in the municipal sewage sludge (Smith, 1996; Mosquera-Losada et al., 2009) relative to soil concentrations. As seen with the macronutrients, the COM treatment resulted in higher input rates of Cu and Zn (4.46 kg Cu ha⁻¹ and 27.76 kg Zn ha⁻¹) into the soil than the ANA (3.59 kg Cu ha⁻¹ and 26.43 kg Zn ha⁻¹) or PEL treatment (1.74 kg Cu ha⁻¹ and 14.47 kg Zn ha⁻¹). The COM treatment increased the soil pH compared with the other treatments, which usually implies a reduction of Cu and Zn solubility because of the CEC increment (Prasad and Power, 1997). In China, Miao-Miao et al. (2007) also found that the use of COM as fertilizer increased the concentrations of Zn and Cu in the soil, but in that study the COM contained higher concentrations of Cu and Zn due to the effects of local industry (Cu: 2316 mg kg⁻¹ and Zn: 2971 mg kg⁻¹) compared with the sludges used in our experiment. ANA also increases the total soil Zn because this type of sludge has a higher concentration of Zn than the COM or the PEL (anaerobic sludge: 1752.3 mg Zn kg⁻¹; soil: COM: 478.7 mg Zn kg⁻¹ soil and PEL: 753.1 mg Zn kg⁻¹). An increment of Cu and Zn in soil as a result of sewage sludge applications was also found by Yuan (2009). In contrast, in this study the concentrations of Zn and Cu in the soil were lower in 2008 than at the beginning of the experiment. This could be explained by leaching, but is more likely due to pasture and tree extractions.

The range of Zn concentrations in the pasture in this experiment (18.63–49.31 mg kg⁻¹) was at the low end of the concentrations commonly found in pastures (27–150 mg kg⁻¹) and below the levels of 100 and 400 mg kg⁻¹ considered excessive or toxic for plants (Kabata-Pendias and Pendias, 1985; Smith, 1996). ANA tended to increase the soil and pasture Zn concentrations in the acidic soils of the Galician region, as described by Mosquera-Losada et al. (2001). In spite of the higher Zn inputs with the COM treatment, which increased the total and the available Zn in the soil, no effect was detected in the pastures because of the increased soil pH compared with other treatments, which could in turn reduce the availability of absorbable Zn in the soil (Prasad and Power, 1997). A Zn concentration of 500 mg kg⁻¹ is considered toxic for cattle, sheep and horses (Smith, 1996), but this value was not reached or exceeded in the pasture in this experiment.

**Conclusion**

Soil characteristics, tree growth, pasture species biodiversity and pasture development are modified by the type of sewage sludge used when similar nitrogen inputs are applied. The ANA treatment has a higher initial effect on tree and pasture productivity, but PEL treatment sustains better production as it is applied in several times and the COM treatment improved soil characteristics over the long term in sandy soils. The PEL treatment should be promoted because this treatment enhances productivity, allows for better nutrient recovery and is less costly to apply than the other two treatments. No toxic Zn or Cu concentrations were found in the plants or in the soil in spite of the higher concentrations in sewage sludge than in the soil.
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