

Effect of understory vegetation management through liming and sewage sludge fertilisation on soil fertility and *Pinus radiata* D. Don growth after reforestation

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Abstract Tree–understory competition is one of the most important aspects that control tree growth after reforestation. The relationship between trees and the understory can be modified by improving acidic soils with lime and by fertilisation. This experiment aims to evaluate the effect of soil improvements on the pasture–tree relationship by liming and fertilisation on different dates in a *Pinus radiata*-reforested area. Both lime and sewage sludge improved soil fertility by increasing Ca and reducing Al in the soil. Initially, tree development was reduced by lime, which improved the establishment of competitive grasses. Tree growth in limed treatments did not initially respond to sludge inputs, likely because both tree and grass roots shared the same soil depth layer. Three years after establishment, the use of high doses of sewage sludge in limed plots caused a growth rate similar to the best treatments of unlimed plots, which grew with a poorly sown grass establishment. After 2 years of the experiment, the presence of *Erica* woody shrub diminished tree development. High doses of sewage sludge with lime, as well as high doses of sewage sludge without lime, applied in April and low doses of sewage sludge without lime added in early February improved tree growth. From a practical point of view, lime and sewage sludge dose close to 100 kg total N ha⁻¹ should be recommended if a silvopastoral system is established, as it enhances pasture production and tree growth.

Keywords Organic fertiliser · Herbaceous · Shrubs · Aluminium · Calcium · Biosolid

Introduction

Most silviculture textbooks (Smith et al. 1997) and reviews (Wagner et al. 1999, 2006) highlight the existence of a critical period when interspecific competition between weeds and trees should be controlled through adequate forest vegetation management to enhance tree development (Wagner et al. 1999, 2006). Herbicide use is one of the most extended techniques to control initial tree–pasture competition, but high costs and important environmental concerns make it necessary and desirable to understand the competition for nutrients between understory plants and trees in order to increase tree growth without herbicide use, if possible. Forest vegetation management also includes the introduction of non-crop species to improve site quality, suppress unwanted species and/or provide forage for grazing animals (Richardson 1993; Wagner et al. 2006) while tree growth is promoted.

Pinus radiata (*P. radiata*) is the most popular species for afforestation in the Province of Lugo, accounting for ~10% of forestland in the Galicia region (MMA 1998; Rigueiro-Rodríguez et al. 2005), which supplies more than 50% of the wood in Spain (one of the largest countries in Europe). The establishment of silvopastoral systems in *P. radiata*-reforested stands would allow the creation of pastures to sustain multiple output systems (e.g., milk and meat) while avoiding costly clearing practices (Rigueiro-Rodríguez et al. 2009). Moreover, the establishment of forest vegetation for use as forage is recommended by the EU COUNCIL REGULATION on support for rural development by the

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European Agricultural Fund for Rural Development (EAFRD), 15 September 2005 (CEE 2005).

P. radiata is broadly distributed around the world in agroforestry systems and it is usually associated with silvopastoral systems (Peri et al. 2007; Benavides et al. 2009). When a pasture is established, the tree radical system is not usually developed, and the herbaceous–tree competition for water and nutrients may be higher than in later stages, mostly when container tree plants are used (Montero et al. 2008). Therefore, increasing soil fertility through practices such as liming or fertilisation should be focussed on to ensuring adequate herbaceous growth without reducing (but perhaps enhancing) tree development (Rigueiro-Rodríguez et al. 2009).

Galician natural soils are very acidic due to the type of rock in the soil and high annual precipitation, which also limits tree growth (Sánchez-Rodríguez et al. 2002). Therefore, it is advisable to carry out liming and fertilisation to improve soil fertility, as well as tree and understory vegetation productivity. In general, liming improves the physical properties of the soil by increasing the pH (Loué 1988; Alloway 1995; Smith 1996; Jonard et al. 2010), cation exchange capacity and microbial activity (Bailey 1995), which cause organic mineralisation and therefore nutrient release (Wheeler 1998). Liming also modifies macronutrient concentrations and storage in the aboveground biomass of Norway spruce trees. Sewage sludge fertiliser is promoted by EU (86/278/CEE) (EU 1986) and Spanish (R.D 1310/1990) (BOE 1990) regulations due to the physical soil improvements it causes and recycling of nutrients from animal or vegetal wastes. European Directive 86/278/CEE states that, for the agricultural use of sludge, it is necessary to take into account the needs of the plants and avoid harming soil quality or productivity. Correspondingly, it is necessary to conduct sampling and environmental monitoring focused on heavy metals due to the higher concentration of heavy metals in sewage sludge than in typical soils. Studies carried out in the area of the present experiment have demonstrated that little environmental damage is caused when adequate doses of sewage sludge are applied to forest soils (Mosquera-Losada et al. 2001; López-Díaz et al. 2007). Apart from the dose, the period of application of the sewage sludge is important because, if the sludge is applied in early winter, nutrients can leach because the rhizosphere from trees and pasture is not able to absorb them due to low root development. If applied too late, however, nutrient use by trees and pasture is delayed, and the growing season could be shortened (Peyraud et al. 2004).

The aim of this study was to evaluate the effect of liming and the application of three doses of sewage sludge at different dates on soil, tree growth and pasture in a silvopastoral system established in an area reforested with *P. radiata* D. Don.

Materials and methods

Characteristics of the study site

The experiment was carried out in Lugo (NW Spain: 7°20'N; 43°0.92'W; 450 m asl), San Breixo Mount, between 1999 and 2003. The area belongs to the Atlantic biogeographic region of Europe (EEA 2006). Mean monthly precipitation and temperatures for 2000, 2001, 2002 and 2003 and the mean of the previous 10 years are shown in Fig. 1. Total mean annual precipitation and mean annual temperatures for the last 10 years were approximately 1,300 mm and 11.5°C, respectively. There is generally a drought period during July and August and a low mean temperature during January and February (below 7°C), which limits pasture and tree development. Total annual mean precipitation was above the 10-year mean in 2000 (1,505 mm), lower in 2001 (925 mm) and 2003 (1,038 mm) and similar in 2002 (1,351 mm). The temperature and precipitation during the months of the study were lower than those calculated for each corresponding month of the last 10 years (1999–2009). Compared with the last 10 years (except for 2001), monthly mean temperatures during the study period were less than 1°C below the corresponding monthly mean temperatures in 7 of 12 months of the year. Monthly mean precipitation of the 4 years of the study was lower than the monthly mean temperature for the last 10 years in 9 of the 12 months of the year. Mean temperatures in March and April were lower in 2000 (7.6°C) than in 2001 (8.6°C), 2002 (8.32°C) and 2003 (10.02°C). Summer precipitation was lower in 2000 (59 mm) and 2001 (102 mm) than in 2002 (203 mm) or 2003 (150 mm). The soil texture is sandy clay loam (USDA 2001). Initial soil analyses revealed a low water (4.28) and KCl (3.87) pH and high organic matter content (8.74%). The initial C/N ratio was 22, which has a slow mineralisation rate, and therefore, the nitrification rate is not enough to provide an adequate supply of mineral nitrogen to plants. All heavy metal concentrations in the soil were below the maximum thresholds for using sewage sludge as fertiliser, as specified by the European Union Directive 86/278/CEE (DOCE 1986) and Spanish legislation under R.D. 1310/1990 (BOE 1990).

Experimental design

The experimental design was a randomised block with three replicates. After a 30-year-old *P. radiata* plantation was harvested, a plantation of *P. radiata* was established at a density of 1,667 trees ha⁻¹ in October 1998. Scrubland was the main understory vegetation before planting, which included species such as *Ulex europaeus* L., *Erica cinerea* L., *Calluna vulgaris* L., *Genistella tridentata* L., *Rubus*

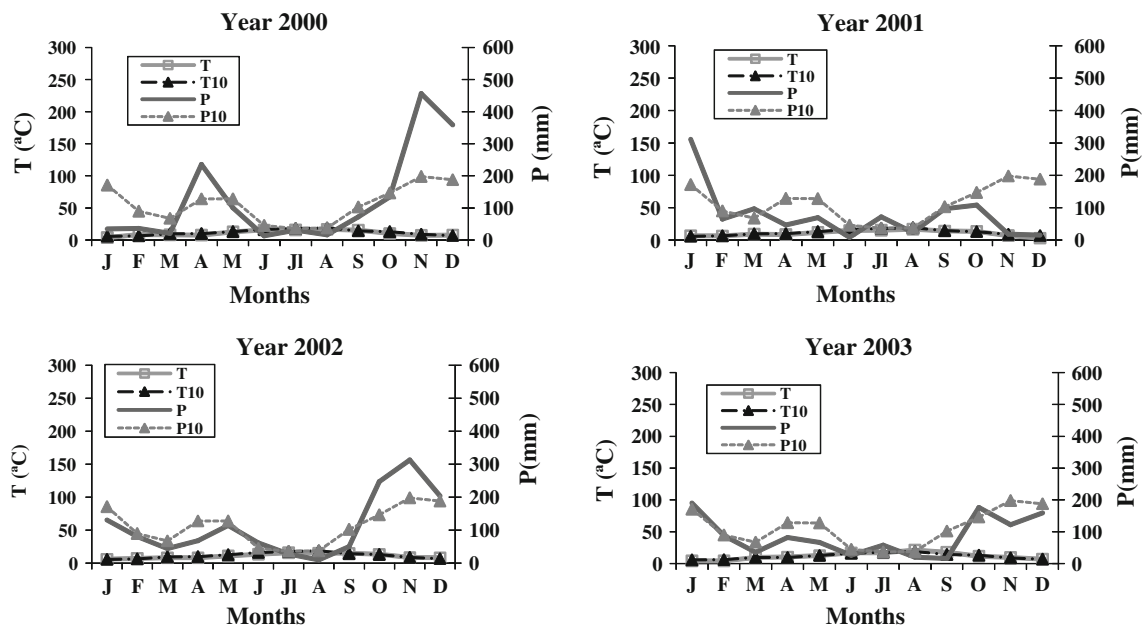


Fig. 1 Precipitation (mm) and temperature (°C) during the years of study and the average of these parameters in the last 10 years. T10 average 10-year temperature (1993–2003), T average temperature of

the year studied, P10 average 10-year precipitation (1993–2003), P average precipitation of the year studied

spp., *Avenula sulcata* J. Gay ex Delastre, *Agrostis curtisii* Kerguelen and *Holcus mollis* L.

In October 1999, when forestry plants were 1 year old, the experiment was established in 39 (13 treatments \times 3 replicates) experimental units of 12 \times 8 m, each consisting of 25 trees arranged in a 5 \times 5 frame with a distance of 3 m between rows and 2 m between lines. An untreated buffer area of 3 m between rows and 2 m between lines were established between experimental units. Half of the plots were limed in October 1999 and half were left unlimed. After liming, a sowing mixture of 25 kg ha⁻¹ *Lolium perenne* L. var. Brigantia (ryegrass), 4 kg ha⁻¹ *Trifolium repens* L. var. Huia (clover) and 10 kg ha⁻¹ *Dactylis glomerata* L. var. Artabro (cocksfoot) was applied in all plots in early November 1999. Fertilisation treatments consisted of two doses of sewage sludge containing 50 (L: low) and 100 (H: high) kg ha⁻¹ total N, applied on three different dates (at the beginning of February (1), March (2) and April (3)) in limed and unlimed plots. Sewage sludge fertilisation was superficially applied during 2000, 2001, 2002, 2003 and 2004. Sewage sludge doses were applied based on previous experimental results following the recommendations of the EPA (1994), which considers that \sim 25% of the total N is available in the year of application of the sewage sludge. Sewage sludge doses above those used in this experiment demonstrated to increase nitrate leaching without tree growth benefits (Egiarte et al. 2005). A “no fertilisation treatment without liming (NF)” was used as a control treatment.

Sewage sludge

Anaerobically digested municipal sewage sludge was produced in Lugo city. The composition of the sewage sludge used in this study is shown in Table 1. Heavy metal concentrations in the soil at the beginning of the experiment were below the Spanish legally permitted limits for use in agriculture (BOE 1990).

Field sampling and laboratory processing

Soil samples were taken within the area occupied by the 9 inner trees at the beginning of 2004 at a depth of 25 cm. Samples were taken to the laboratory, air-dried and sieved through a 2-mm screen. After this preparation, we determined calcium (Ca²⁺) concentrations extracted by the Mehlich method (M3) (Mehlich 1985), as well as saturation percentages of aluminium (Al³⁺) and calcium (Ca²⁺) after soil extraction with Ba₂Cl (Gutián and Carballás 1976).

Tree heights (h) were measured using a graduated ruler in 2000 and pole in 2004. Base diameter (d) (year 2000) and diameter at breast height (dbh, 1.30 m) (year 2004) were estimated using a calliper. Tree variables were measured in October 2000 and August 2004 for the nine inner trees of each experimental plot in order to avoid the “border effect”.

Pasture production was estimated by collecting four random samples (1 \times 1 m) per plot within the area

Table 1 Chemical properties of the sewage sludge applied and law limits established by European Directive 86/278 and the Spanish R.D. 1310/1990

Parameters	Year 2002	Year 2003	Law limits
Dry matters (%)	22.78	25.20	
pH	7.47	6.51	
N (%)	3.62	2.10	
P (%)	1.83	1.91	
K (%)	0.41	0.21	
Ca (%)	0.37	0.2	
Mg (%)	0.63	0.34	
Na (%)	0.13	0.03	
Fe (%)	0.76	1.17	
Cr (mg kg ⁻¹)	71.05	28.20	1000–1500
Cu (mg kg ⁻¹)	218.55	77.80	1000–1750
Ni (mg kg ⁻¹)	91.95	47.90	300–400
Zn (mg kg ⁻¹)	61.55	23.50	2500–4000
Cd (mg kg ⁻¹)	7.05	8.20	20–40
Pb (mg kg ⁻¹)	120.70	77.70	750–1200
Mn (mg kg ⁻¹)	323.60	130.20	

The first and the last number of the range indicate the limits for the sewage sludge to be applied in acid and basic soils, respectively

occupied by the 9 inner trees using battery-driven hand shears. Samples were collected in July and November 2000, in May, July and December 2001 and 2002, and in June and November 2003. The samples were transported to the laboratory and weighed while fresh. Then, 100 g was separated from each sample and oven-dried to determine the dry weight (60°C × 48 h) and later dry matter production. Seasonal pasture production in May and July 2001 and 2002 were summed to allow comparisons between years as spring pasture production was obtained. Annual pasture production was calculated by summing all of the harvests within the year. The proportion of sown (ryegrass, cocksfoot and clover) and weed species was estimated by hand separation of 100 g of fresh pasture in the laboratory, and their corresponding percentages were later estimated after oven drying (60°C × 48 h) and weighing.

Statistical analyses

The results obtained were analysed by ANOVA (randomised block design). The following models were used: $P_{ijkl} = \mu + L_i + S_j + D_l + B_k + (LS)_{ij} + (LD)_{il} + (LB)_{ik} + (SD)_{jl} + (SB)_{jk} + (DB)_{lk} + (LSD)_{ijl} + (LDB)_{ilk} + (SDB)_{jlk} + \varepsilon_{ijkl}$, where P_{ijkl} is the dependent variable of soil, pasture production and tree variables, μ is the variable mean, L_i is the i lime fertilisation effect, S_j is the j sludge dose fertilisation effect, B_k is the k block effect and their double and triple interactions, and ε_{ijklm} is the error component.

Significant differences between treatment levels were determined using the SLICE option of SAS. The Bonferoni test was used for subsequent pair-wise comparisons ($P < 0.05$; $\alpha = 0.05$) only if the ANOVA was significant. The SAS statistical software package (SAS 2001) was used for all analyses.

Results

Soil

ANOVA results show that saturation percentages of Al³⁺ and Ca²⁺, as well as the Mehlich Ca²⁺ concentrations, were significantly affected by our treatments. The Al³⁺ saturation percentage was significantly influenced by lime ($P < 0.001$) and sewage sludge ($P < 0.01$). Al³⁺ saturation values for lime treatments were less than 31% (Table 2), while unlimed treatment values were greater than 42%, reaching 55% in the control treatment (NF). No fertilisation (55^a%) and unlimed treatments (49^a and 43^a% for low and high dose of sewage sludge) had higher Al³⁺ saturation values than limed treatments that received a high (17^b%) or low (28^b%) dose of sewage sludge (different superscript letters indicate significant differences between treatments), independent of the date of application (Table 2).

There was also a positive effect of lime ($P < 0.01$) and sewage sludge dose ($P < 0.01$) on the saturation percentage of Ca²⁺ (Table 2). All limed treatments (60^a% for low and 72^a% and high dose of sewage sludge) had a higher saturation percentage of Ca²⁺ in the soil than the unlimed treatments (34^b, 43^b and 37^b% for 0, 50 and 100 kg N per ha⁻¹ applied as sewage sludge). The levels of Ca²⁺ were greater than 55% in all limed plots, but less than 43% in unlimed plots. The results indicate that Al³⁺ was replaced by Ca²⁺ when lime was applied.

The concentration of Ca²⁺ extracted by the Mehlich method was significantly affected by lime*date ($P < 0.01$) and lime*sewage sludge dose ($P < 0.05$) interactions. As with the saturation percentage of Ca²⁺, lime significantly increased the Ca²⁺ concentration extracted by the Mehlich method, but only when low or high sewage sludge dose was applied in early April. No effects of application doses on the saturation percentage of Al³⁺, Ca²⁺ and Ca²⁺ extracted by the Mehlich method were detected in either limed or unlimed plots, with the exception of a positive effect of sewage sludge increasing the Ca²⁺ saturation percentage and Ca²⁺ extracted by the Mehlich method and a negative effect of sewage sludge reducing the Al³⁺ saturation ratio when applied in March in limed treatments. Ca extracted by the Mehlich method was increased in those limed plots receiving a low dose of sewage sludge in April compared with March.

Table 2 Percentage of saturation ratio of aluminium (%Al), calcium extracted in BaCl₂ (% Ca) and calcium extracted by the Mehlich method (mg kg⁻¹ Ca Mehlich) in soil in January 2004 for each of the treatments

Dose	Date	Lime								
		% Al			% Ca			mg kg ⁻¹ Ca Mehlich		
		0 Mg	2.5 Mg	Sig	0 Mg	2.5 Mg	Sig	0 Mg	2.5 Mg	Sig
50	F	48a	27b	**	40b	61a	**	–	–	–
50	M	51a	31b	**	35b	56a	**	–	–	–
50	A	47a	26b	**	37b	65a	**	1.06b	2.65a	**
100	F	42a	17b	**	43b	71a	***	–	–	–
100	M	45a	14b	***	41b	75a	***	–	–	–
100	A	42a	19ba	**	43b	72a	***	1.42b	3.47a	**
Lime	Date	Dose								
		% Al			% Ca			mg kg ⁻¹ Ca Mehlich		
		50	100	Sig	50	100	Sig	50	100	Sig
2.5	M	31a	14b	**	56b	75a	**	1.04b	2.62a	**
Lime	Dose	Date								
		mg kg ⁻¹ Ca Mehlich								
		F	M	A	Sig					
2.5	50	1.40ab	1.04b	2.65a	*					

50: dose of 50 kg ha⁻¹ total N; 100: dose of 100 kg ha⁻¹ total N; and F, M and A: different sewage sludge application dates for February, March and April, respectively. Letters indicate significantly different averages

Trees

Tree results (base diameter, diameter at breast height and height) for each treatment in 2000 and 2004 are shown in Table 3. Tree diameter (d ($P < 0.01$ in 2000)), dbh ($P < 0.01$ in 2004)) and height ($P < 0.01$ in 2000) were significantly affected by the lime*sludge*date interaction during the study. Moreover, height was significantly affected by limed*sludge dose ($P < 0.001$), sludge dose*date ($P < 0.001$) and date*lime ($P < 0.01$) in 2004.

Tree height in 2004 was significantly higher in unlimed plots receiving low (4.21^a) or high (4.06^a) dose of sewage sludge, which significantly differed from no fertilisation treatment (3.16^b). The same response of tree height in 2004 was found in limed plots when the high dose of sewage sludge was used (4.22^a), but not when the low dose (3.31^b) of sewage sludge treatment in unlimed plots is taken into account. Lime inputs reduced tree height or diameter in all samplings when low or high dose of sewage sludge were added in February or April, respectively, with the exception of the diameter in 2004, when no differences were detected between lime treatments if high dose of sewage sludge was added in April (Table 3). The same negative effect of lime on tree height was

detected in 2004 when the low dose of sewage sludge was applied in March and April. On the contrary, lime inputs improved tree height and diameter in 2000, when the high dose of sewage sludge was added in March, but no differences were found in 2004.

Tree height in 2000 was reduced by the increase in the sewage sludge dose applied in April in limed plots, as happened in unlimed plots fertilised in February with respect to the height of 2004 and the diameter in the both years. However, the contrary was found in tree height in 2004, when the increase in the dose of sewage sludge applied in April improved tree height, independently if it had been previously limed. Similarly, tree height in 2004 was higher in those plots fertilised with high dose of sewage sludge applied in March in limed plots. The date of application factor was only relevant when no lime had been previously applied, with the exception of the diameter in 2004. In unlimed plots, February applications of low doses of sewage sludge increased all tree variables studied in all the years, but the opposite effect was found when the high dose of sewage sludge was used in early April in all variables. Finally, in limed plots, tree diameter in 2004 was enhanced by the inputs of the low dose of sewage sludge in April.

Table 3 Diameter (cm) and average height (m) of trees for each treatment applied in 2000 and 2004

Dose	Date	Lime											
		H2000			H2004			D2000			D2004		
		0 Mg	2.5 mg	Sig	0 Mg	2.5 mg	Sig	0 Mg	2.5 mg	Sig	0 Mg	2.5 mg	Sig
50	F	0.87	0.64	**	4.6	3.6	**	2.00	1.54	*	11.11	8.0	*
50	M	–	–	–	3.53	3.01	*	–	–	–	–	–	–
50	A	–	–	–	4.2	3.29	*	–	–	–	–	–	–
100	M	0.71	0.75	*	–	–	–	1.24	1.74	*	–	–	–
100	A	0.92	0.69	**	5.0	4.14	*	1.90	1.35	*	–	–	–

Date	Lime	Date	Dose											
			H2000			H2004			D2000			D2004		
			50	100	Sig	50	100	Sig	50	100	Sig	50	100	Sig
	0 Mg	F	0.87a	0.71b	ns	4.80a	3.42b	***	2.04a	1.37b	**	11.11a	8.28b	***
		M	0.64b	0.61b	ns	3.53b	3.54b	ns	1.32b	1.37b	ns	8.23b	8.34b	ns
		A	0.66b	0.92a	ns	4.22ab	4.97a	*	1.33b	1.90a	*	8.23b	10.60a	ns
		Sig	*	***		**	*		**	*		***	*	
	2.5 Mg	F	0.65	0.77	ns	3.66	4.33	ns	1.56	1.48	ns	8.09ab	10.10	ns
		M	0.65	0.79	ns	3.01	4.09	**	1.37	1.74	–	6.79b	9.71	**
		A	0.78	0.70	**	3.29	4.15	*	1.81	1.39	–	10.14a	9.27	–
		Sig	ns	ns		ns	ns		ns	ns		*	ns	

D2000 = basal diameter in 2000, D2004 = diameter breast height in 2004, H2000 = average height in 2000 and H2004 = average height in 2004. Different letters indicate significantly different averages between treatments (between dates of application per each lime and sludge treatments). 50: dose of 50 kg ha⁻¹ total N; 100: dose of 100 kg ha⁻¹ total N; and F, M and A: different sewage sludge application dates for February, March and April, respectively. Differences between the two values of lime or sewage sludge doses are indicated by asterisks

*** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$

Pasture production

Annual pasture production in each treatment is shown in Table 4. Only annual pasture production in 2001 and 2002 was significantly affected by the lime*sludge dose*date ($P < 0.05$) and the sludge dose*date ($P < 0.05$) interactions, respectively. Annual pasture production was lower in 2000 (between 1.79–2.73 Mg DM ha⁻¹ and 1.75–2.65 Mg DM ha⁻¹) and 2001 (0.67–1.10 Mg DM ha⁻¹ and 0.53–0.78 Mg DM ha⁻¹) than in 2002 (3.93–6.21 Mg DM ha⁻¹ and 1.29–3.50 Mg DM ha⁻¹) and 2003 (3.85–5.54 Mg DM ha⁻¹ and 2.33–4.40 Mg DM ha⁻¹). Pasture production was higher in 2002 and 2003 than in the other years that were studied due to increased summer precipitation and initial higher spring temperatures than average (Fig. 1).

There were no lime effects on annual pasture production in 2001 and 2002 (Table 4). However, high doses of sewage sludge applied in early February or April improved annual pasture production if lime had been previously applied in 2001 and 2002, respectively. Annual pasture production was increased if high dose of sewage sludge was added in early February in limed plots in 2001 compared with early April. However, annual pasture production

was reduced when high doses of sewage sludge were applied in early March compared with the other dates of application in both limed and unlimed plots.

Seasonal pasture production was significantly affected by sludge dose in the spring of 2001 ($P < 0.05$) and by the interactions of lime*sludge dose*date ($P < 0.05$) in autumn 2001 and sludge dose*date ($P < 0.01$) in spring 2002. Maximal seasonal pasture production was found in the spring of 2002, followed by 2003, 2000 and 2001, as with annual pasture production. Lime inputs reduced autumn production in 2001 when compared with the low dose of sewage sludge applied in early February. Sewage sludge dose increase positively affected pasture production depending on the date of application and on previous lime inputs. The positive effect of the high dose of sewage sludge on pasture production was observed in early sludge applications (February) in limed plots in spring 2001 and in late applications (April) in unlimed plots in autumn 2001 and in limed plots in spring 2002. Regarding the date of sludge application, pasture production was positively affected when the low dose of sewage sludge was applied in limed plots in April than in March or February in autumn 2001. The high dose of sewage sludge applied in early

Table 4 Annual and seasonal (spring: sum of the production of May and July) pasture production expressed in kg ha⁻¹ for each treatment in those significant harvests

Annual pasture production																	
Dose	Date	Dose						Lime	Date	Dose							
		Prod 2001			Prod 2002					Prod 2001				Prod 2002			
		50	100	Sig	50	100	Sig			F	M	A	Sig	F	M	A	Sig
2.5 Mg	F	671	1,102	**	–	–	–	50	2.5	671b	827ab	1056a	*	–	–	–	*
2.5 Mg	M	–	–	–	–	–	–	100	2.5	–	–	–	–	4,545a	2,896b	6,009a	*
2.5 Mg	A	–	–	–	4,090	6,009	**	100	0	–	–	–	–	5038a	2,913b	4,365ab	**

Seasonal pasture production																	
Sampling	Dose	Date	Lime			Lime	Date	Dose			Lime	Dose	Date				
			0 mg	2.5 Mg	Sig			50	100	Sig			F	M	A	Sig	
			Spring 2001	–	–			–	–	–			2.5	F	509	837	*
Autumn 2001	50	F	339	162	**	0	A	222	359	*	2.5	50	162b	252b	327a	*	*
Spring 2002	–	–	–	–	–	2.5	A	2,227	4589	**	2.5	100	4,016a	2,916b	4,589a	**	**
	–	–	–	–	–	–	–	–	–	–	0	100	4,554a	3,061ab	3,804b	**	**

Prod: production for each treatment. 50: dose of 50 kg ha⁻¹ total N; 100: dose of 100 kg ha⁻¹ total N; and F, M and A: different sewage sludge application dates for February, March and April, respectively. Different letters indicate significantly different averages between treatments (between dates of application per each lime and sludge treatments). Differences between the two values of lime or sewage sludge doses are indicated by asterisks

*** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$

March reduced pasture production in spring 2002 in limed plots compared with the same dose inputs on the other two dates. However, early inputs (February) of high dose of sewage sludge in unlimed plots caused an increase in pasture production in spring 2002 compared with late sewage sludge applications (April).

Botanical composition

The relative proportion of sown species as a result of the type of treatment in each harvest is presented in Table 5. The proportion of sown species, mainly cocksfoot, was more adequate one year after establishment (2002) in most of the limed treatments. Ryegrass and clover establishment hardly occurred in the first year, and the percentages of these species were low within the whole study (Table 5). Sown species were significantly affected by treatments in July 2000 (lime: $P < 0.05$), in May (lime*sludge dose*date: $P < 0.05$), in July (sludge: $P < 0.05$) and December (lime*sludge dose*date: $P < 0.05$) 2002, and in June (sludge*date: $P < 0.001$) and November (lime: $P < 0.01$ and sludge: $P < 0.01$) 2003. Sown species establishment was usually better if lime had been previously added to the soil (Table 5). Regarding the effect of sewage sludge in limed plots, it was found that high doses of sewage sludge applied in April significantly improved the proportion of sown species in May 2002 and December 2002 as well as

in unlimed plots in November 2003. However, early applications (February) of high doses of sewage sludge reduced the proportion of sown species in limed plots in July and December 2002 and in July 2003. March inputs of high doses of sewage sludge also initially reduced the proportion of sown species in limed plots in July 2002, but the effect was reverted in the harvests of June 2003 in limed plots and in unlimed plots in December 2002 and June 2003.

Sown species proportion in unlimed plots was usually reduced when the low dose of sewage sludge was applied in March, July and December of 2002 and in June 2003 compared with February inputs and in limed plots in June 2003. However, the proportion of sown species was increased when high doses of sewage sludge were added in March in limed plots in the harvest of D2002.

Similarly, the cocksfoot proportion was higher in limed than in unlimed treatments in July and in May 2002 in low doses applied in February and high doses added in March or April (Table 5). No differences between treatments were found within unlimed or limed plots in the first year of the experiment. However, since 2002, the effects of the dose and date of sludge application on sown species were more relevant, mainly and almost exclusively linked to cocksfoot proportion. In limed plots, the increase in sewage sludge dose applied in February reduced the proportion of cocksfoot in July 2002, December 2002 and July 2003.

Table 5 Proportion of sown species and *Erica* spp. by harvest in study plots in 2000, 2002 and 2003. 50: dose of 50 kg ha⁻¹ total N; 100: dose of 100 kg ha⁻¹ total N; and F, M and A: different sewage sludge application dates for February, March and April, respectively

Sampling	Dose	Date	Lime			Lime	Date	Dose			Lime	Dose	Date			Sig
			0 Mg	2.5 Mg	Sig			50	100	Sig			F	M	A	
<i>Dactylis glomerata</i>																
Jl 2000	50	F	0	16.86	**	–	–	–	–	–	–	–	–	–	–	–
Jl 2000	100	M	11.67	18.87	**	–	–	–	–	–	–	–	–	–	–	–
Jl 2000	100	A	3.09	16.59	*	–	–	–	–	–	–	–	–	–	–	–
My 2002	50	F	13.73	63.33	**	2.5	A	28.14	81.58	***	2.5	50	63.33a	34.15ab	28.15b	*
My 2002	100	M	22.84	63.52	**	0	F	13.73	43.78	*	–	–	–	–	–	–
My 2002	100	A	27.18	81.58	***	–	–	–	–	–	–	–	–	–	–	–
Jl 2002	–	–	–	–	–	2.5	F	57.19	13.96	**	0	50	66.45a	14.31b	37.70ab	*
Jl 2002	–	–	–	–	–	2.5	M	41.65	6.42	*	–	–	–	–	–	–
D 2002	–	–	–	–	–	2.5	F	46.07	4.11	**	2.5	100	4.11b	29.19ab	50.59a	**
D 2002	–	–	–	–	–	2.5	A	15.78	50.59	*	0	50	39.41a	0b	39.62a	**
D 2002	–	–	–	–	–	0	M	0	46.16	**	–	–	–	–	–	–
Jn 2003	–	–	–	–	–	2.5	F	57.82	8.19	***	2.5	50	57.82a	7.50b	27.61ab	***
Jn 2003	–	–	–	–	–	2.5	M	7.5	31.59	*	0	50	36.42a	0b	23.95ab	**
Jn 2003	–	–	–	–	–	0	M	0	23.47	*	–	–	–	–	–	–
N 2003	50	A	5.65	37.42	**	0	A	5.65	39.36	**	–	–	–	–	–	–
<i>Lolium perenne</i>																
Jl 2000	100	M	0	18.98	**	–	–	–	–	–	–	–	–	–	–	–
N 2000	50	F	0	12.50	*	–	–	–	–	–	–	–	–	–	–	–
My 2002	–	–	–	–	–	0	F	0	1.25	*	0	100	1.25a	0b	0b	*
Jl 2002	50	M	0	0.25	*	2.5	M	0.25	0	*	2.5	50	0b	0.25a	0b	*
D 2002	50	A	3.9	0	**	0	A	3.9	0	*	0	50	0b	0b	3.9a	*
Jn 2003	100	M	0.65	0	*	0	M	0	0.65	*	0	100	0b	0.65a	0b	*
N 2003	50	A	0.65	0	*	2.5	A	0.65	0	*	–	–	–	–	–	–
<i>Trifolium repens</i>																
N 2000	100	F	0	1.93	*	2.5	F	0	0.01	*	2.5	100	1.93a	0.00b	0.00b	*
My 2002	50	M	0	7.02	*	–	–	–	–	–	–	–	–	–	–	–
D 2002	50	M	0	1.66	*	2.5	M	1.66	0	*	2.5	50	0	1.66	0	*
Jn 2003	50	F	0	0.08	*	2.5	F	0.08	0	*	2.5	50	0.08a	0b	0b	*
<i>Erica</i> spp.																
N 2000	50	M	24.40	3.97	*	0	M	0.24	0.02	*	0	50	7.34ab	24.40a	0.00b	*
My 2002	100	A	26.63	0	**	0	A	0	26.63	*	0	100	0.12b	0.17b	26.63a	*
Jl 2002	50	M	34.23	0	***	0	M	34.23	3.74	***	0	50	0.51b	34.23a	0.00b	***
D 2002	50	M	31.27	0	**	0	M	31.27	0	**	0	50	6.09ab	31.27a	0.67b	**
Jn 2003	50	M	52.00	1.77	***	0	M	52.00	0	***	0	50	15.74ab	52.00a	0.94b	***
N 2003	–	–	–	–	–	0	M	12.17	0	*	–	–	–	–	–	–
<i>Sown species</i>																
Jl 2000	50	F	0	25.94	*	–	–	–	–	–	–	–	–	–	–	–
Jl 2000	100	M	0	34.70	*	–	–	–	–	–	–	–	–	–	–	–
My 2002	50	F	13.73	66.12	***	2.5	A	30.76	82.29	**	–	–	–	–	–	–
My 2002	50	M	15.93	41.17	*	0	F	13.73	45.03	*	–	–	–	–	–	–
My 2002	100	M	22.84	64.95	**	–	–	–	–	–	–	–	–	–	–	–
My 2002	100	A	27.18	82.29	***	–	–	–	–	–	–	–	–	–	–	–
Jl 2002	100	M	14.31	41.91	*	2.5	F	57.19	13.96	**	0	50	66.45a	14.31b	37.7ab0	**
Jl 2002	–	–	–	–	–	2.5	M	41.91	6.42	*	–	–	–	–	–	–
D 2002	–	–	–	–	*	2.5	F	46.07	4.11	**	2.5	100	4.1b	29.22ab	50.63a	**

Table 5 continued

Sampling	Dose	Date	Lime			Lime	Date	Dose			Lime	Dose	Date			
			0 Mg	2.5 Mg	Sig			50	100	Sig			F	M	A	Sig
D 2002	–	–	–	–	–	2.5	A	15.78	50.63	*	0	50	39.41a	0b	43.52a	**
D 2002	–	–	–	–	–	0	M	0	46.28	**	–	–	–	–	–	–
Jn 2003	–	–	–	–	–	2.5	F	57.9	8.19	***	2.5	50	57.9a	7.5b	27.6b	***
Jn 2003	–	–	–	–	–	2.5	M	7.5	31.59	*	0	50	36.42a	0b	24.04ab	**
Jn 2003	–	–	–	–	–	0	M	0	24.12	*	–	–	–	–	–	–
N 2003	50	A	5.65	38.08	**	0	A	5.65	39.37	***	–	–	–	–	–	–

Different letters indicate significantly different averages between treatments (between dates of application per each lime and sludge treatments). Differences between the two values of lime or sewage sludge doses are indicated by asterisks

*** $P < 0.001$, ** $P < 0.01$, * $P < 0.05$

March inputs of high doses of sewage sludge reduced the proportion of cocksfoot in limed plots in July 2002, but increased it in June 2003. In unlimed plots, the higher sewage sludge doses improved the proportion of cocksfoot when it was applied in February in the harvest of May 2002, in March in the harvests of December 2002 and June 2003 and finally in April in the harvest of November 2003. The proportion of cocksfoot was usually low in March applications of low dose of sewage sludge either if lime was applied (June 2003) or not (July 2002, December 2002 and June 2003).

The proportion of ryegrass and clover was usually below 8%. Ryegrass was initially increased by liming before July 2002 harvests, when significant, but the reverse answer was found since this harvest. However, clover was improved by lime, when significant, during the whole study. The effect of the dose and the date of application on the percentage of clover or ryegrass was not consistent.

Unsown *Erica* spp. species (Table 5) proportion was lower in the limed treatments when low dose of sewage sludge was applied in March in the harvests of November 2000, May 2002, July 2002, December 2002 and June 2003 or high dose was added in April when the harvest was carried out in May 2002. The low dose of sewage sludge also improved the proportion of unsown *Erica* spp, when lime was not previously applied, in March in all harvests of 2002 and 2003, although it was favoured by the high dose of sewage sludge when applied in April and the harvest was done in May 2002. There was an increase in *Erica* spp when the low dose of sewage sludge was applied in March compared with the other dates in the previous mentioned harvests.

Discussion

The reduction of Al^{3+} saturation percentage and, consequently, the increase in Ca^{2+} availability as a result of

adding lime to very acidic soils have been broadly described in the literature (Benbi and Nieder 2003; Jonard et al. 2010). The replacement of Al^{3+} by Ca^{2+} in the soil exchange cation capacity explains the increase in initial establishment of sown herbaceous species during the first year of this study in limed plots (López-Díaz et al. 2007, 2009). Ryegrass established first, followed by cocksfoot, due to their different establishment rates (higher in ryegrass than cocksfoot) (Mosquera-Losada et al. 2006). However, cocksfoot was more persistent than ryegrass because it is better adapted to low soil fertility, shading, and summer drought in this area (López-Díaz et al. 2007, 2009).

Soil improvements caused by lime application and the subsequent better establishment of sown species in limed plots compared with unlimed plots caused a reduction in tree height and diameter when low dose of sewage sludge was added in February, March and April. Sown species are more able to uptake nutrients than young trees, which suffered from sown species competition. Tree–pasture competition enhancement in limed plots was not overcome by the addition of low doses of sewage sludge in February or April. However, tree growth was initially (year 2000) enhanced when lime and high dose of sewage sludge was added to the soil in February probably due to the poor establishment of sown species and the higher nutrient availability for trees, which benefited tree nutrient uptake. Tree growth improvement as a result of liming in very acid soils was also described by Jonard et al. (2010) in beech plantations. Annual production was not consistently affected by treatments until the spring of 2001 due to adverse climatic conditions for pasture production. Therefore, initial high competition of tree-sown species in limed plots is likely due to the fact that sown species are likely more competitive with trees for nutrients than unsown species, mostly when tree-pasture roots share the same rhizosphere depth (Wagner et al. 1999). The increase in tree growth as a result of understory biomass reduction when the botanical composition is similar has been

extensively described (Zutter and Miller 1998; Wagner et al. 2006). This experiment shows that the composition of the herbaceous understory also determines tree growth. López-Díaz et al. (2009) observed a similar result in young *P. radiata* stands established in agronomic soils when legumes are involved. Lupine legume introduction also resulted in long-term *Pinus sylvestris* growth acceleration in Germany (Prietz et al. 2008). The negative effect of gramineous sown species on tree development has been described in agronomic soils (Mosquera-Losada et al. 2006) but not in very acidic forest soils when pasture establishment, lime and fertilisation were performed in a five-year-old *P. radiata* stand, likely due to the deeper roots of older trees used in the López-Díaz et al. (2007) experiment.

Sewage sludge affected tree parameters in unlimed plots from the first year of the experiment because unsown understory development was not able to compete with trees in unlimed plots as it had in limed treatments, and this affected tree growth differently. The effect of sewage sludge, which has a significant amount of Ca^{2+} , on the reduction of Al^{3+} saturation percentage is important (Rodríguez-Barreira 2007), as seen in very acidic (López-Díaz et al. 2007) and acidic (Mosquera-Losada et al. 2011) soils, when higher sewage sludge doses than those applied in this experiment were used with limed treatments. The more important effect on soil parameters of lime compared with sewage sludge that was found in this experiment could be explained by the lower inputs of Ca^{2+} made through the sludge (López-Díaz et al. 2007; Mosquera-Losada et al. 2011). In limed plots, high doses of sewage sludge only significantly reduced the Al^{3+} soil saturation percentage when they were applied in March but not in February or April. This reduction may explain the lower tree growth and the lower proportion of cocksfoot when low doses of sewage sludge were added compared with the high dose applied in March, at the end of the experiment, in limed plots.

It is important to highlight that even though pasture production was not significantly affected by treatments in the first year of the experiment, there was an initial tree response to treatments due to the botanical composition response to lime and fertilisation inputs. The annual pasture production's lack of response to treatments in 2000 and of seasonal pasture production in 2000 were due to low temperatures in 2000 (7.6°C) between March and April compared with the same months in 2001 (8.6°C), 2002 (8.32°C) and 2003 (10.02°C). Understory herbaceous production usually depends on soil fertility and climatic conditions. The lack of response of pasture production in the first year to treatments could be explained because sludge incorporation and, therefore, nutrient availability were reduced due to low temperatures (mean temperatures in the

first 4 months of 2000 were always less than 8°C) and precipitation (less than 90 mm in the first 3 months of 2000). In the second year of the study, even though precipitation was very high, the temperature in the first 3 months was very low. Later, when the temperature increased, precipitation was low. In 2002, the mean monthly temperatures were always greater than the mean of the last 10 years, and precipitation was high enough to allow pasture development. Subsequently, we saw an influence on pasture production in response to treatments. Finally in 2003, temperatures at the beginning of the year were less than 4°C in the first 2 months. When temperature increased, precipitation was much lower than the mean.

Initial tree–pasture competition usually has an important impact on subsequent tree growth because relative tree growth is the fastest in this initial time period. However, a recovery of tree growth can be obtained later if adequate management is provided. Continuous high doses of sewage sludge in limed plots caused an increase in tree growth compared with low doses of sewage sludge, likely because excess nutrients were not taken up by grasses and could be used by tree roots in deeper zones in spite of the similar proportion of sown species found in high doses of sewage sludge in limed treatments. If higher doses of sewage sludge than those used in the present experiment are used, an increase in nitrate leaching and potential water contamination is observed (Egiarte et al. 2005). Therefore, a better use of nutrients by trees and pasture was observed in our experiment, and the global productivity increased when lime was combined with high doses of sewage sludge in March and April, but not in February. Early applications of sewage sludge (February) in limed plots significantly increased spring and annual pasture production in 2001, which probably reduced tree growth.

Treatments with levels of Ca^{2+} saturation percentage greater than 56% reduced tree dbh. The effect of treatments in the last year on tree growth was more dependent on the initial tree response after reforestation in unlimed plots than in limed plots, likely due to lower nutrient availability (low Ca^{2+} and high Al^{2+}), which made tree recovery more difficult.

The response of pasture production to treatments was not clear due to climatic conditions and the differences in botanical composition caused by lime and sludge. When sewage sludge caused a significant effect on pasture production, it was usually found that high doses of sludge increased pasture production if applied earlier (February) in spring 2001 in limed plots, but this effect occurred in later applications (April) in autumn 2001 and spring 2002 in unlimed and limed plots, respectively. The response of pasture to high doses of sewage sludge applied earlier (February) in the first significant harvest may be linked to the longer period of growth between fertilisation and

harvesting of the same year. However, the response seen in the last years of study is due to the already established sown species. This indicates that early applications provide more time to use nutrients for growth, but on the contrary, once sown, herbaceous species of pasture are established, nutrients are used more efficiently if the sludge is applied when the pasture production response is higher (late April).

Regarding the effect of the sludge application dates on pasture production, when climatic conditions allowed pasture response, a clear response was not detected, likely due to the different botanical composition in the plots. A similar scenario was observed in an experiment developed in a forested area with *P. radiata* that had previously been used for dairy and meat production (Rigueiro-Rodríguez et al. 2010). In agronomic lands, the response of pasture production to the date of sludge application usually depends on climatic conditions. The response is higher if the sludge is applied early when initial temperatures allow the pasture to use the nutrients but lower when the initial temperature does not allow pasture growth because of leaching (Peyraud et al. 2004). In the last case, the presence of a tree with deep roots makes possible the use of leached nutrients and therefore reduces nitrate contamination (Nair et al. 2008).

Tree growth was usually better in unlimed plots that received inputs of sewage sludge in early February, likely due to the longer period of nutrient availability, which is used by sown species to enhance their proportion in limed plots and by trees in unlimed plots. The initial higher presence of shrubs in the treatment receiving high sewage sludge doses in early April in unlimed plots reduced grass–tree competition at this stage in the superficial soil layers (i.e., where tree roots are) and caused an initial increase in tree growth production. Initially, greater competition between tree and herbaceous compared with tree and woody understories has been described by Wagner et al. (1999) and by Zutter and Miller (1998), who found higher tree–grass competition at initial ages of the tree plantation but a higher shrubby–tree competition at mid-rotation in loblolly pine. This could be explained by the fact that younger trees initially share the herbaceous rhizosphere area, as shown in this experiment, but when older, woody understory could have a higher effect on tree growth. In this experiment, as trees developed, tree growth was reduced in the unlimed treatment that received low doses of sewage sludge in March (i.e., the treatment with the highest *Erica* shrub proportion). Root development of *Erica* is deeper than the herbaceous layer, so when trees age, there may be competition at greater depths. Once tree roots are deep enough to avoid competition with the herbaceous layer, and a good establishment of herbaceous layer is obtained (limed plots), tree growth depends on soil fertility. Thus, if high doses of sewage sludge are applied (enough in

this case for tree and pasture growth), tree development is enhanced compared with low doses of sewage sludge, as is pasture growth (e.g., high dose compared to low dose applied in February in limed treatments in spring 2001, high-dose treatment compared with low dose applied in April in unlimed plots in Autumn 2001 or in limed plots receiving high dose of sewage sludge in Spring 2002).

Conclusions

Better establishment of highly competitive grasses caused by initial soil improvements from the application of lime and fertilisation resulted in a reduction in tree growth. However, the negative effect was overcome after trees developed and if adequate fertilisation practices were performed. Moreover, the presence of a shrubby understory can compete with trees and reduce their growth when trees get older. From a practical point of view, lime and sewage sludge dose close to 100 kg total N ha⁻¹ should be recommended if a silvopastoral system is established, as it enhances pasture production and tree growth.

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