

Effects of organic residue management and legume cover on growth of pine seedlings, nutrient leaching and soil properties

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Abstract

- The short-term effect of organic residue management on the growth and nutrition of *Pinus pinaster* Ait. seedlings, and on nutrient leaching and chemical properties of an acid soil was assessed through a lysimeter experiment. Treatments included absence, placement on the soil surface, and incorporation into the soil (with and without legume cover cropping) of organic residues (forest floor litter or forest floor litter plus harvest residues).
- Residues placed on the soil surface enhanced seedling growth. Organic residues reduced nutrient losses (NO_3^- , Ca and Mg) and resulted in nutrient accumulation in the soil. Harvest residues positively affected K seedling nutrition status and enhanced K soil accumulation.
- Legume cover cropping reduced soil nutrient losses (N, Ca, Mg and K) during the early stage of seedling growth; it also improved seedling nutrition status (N and P), but without any effect on growth.
- Harvest residues plus forest floor litter placed on the soil surface was the most appropriate management to both reduce nutrient losses through leaching and increase height of seedlings at the end of the experimental period (two years).

Résumé – Effets de la gestion de résidus organiques et d'un couvert de légumineuses sur la croissance de semis de pin, le lessivage des nutriments et les propriétés du sol.

- L'effet à court terme de gestion des résidus organiques sur la croissance et la nutrition de semis de *Pinus pinaster* Ait. et sur le lessivage des nutriments et sur les propriétés chimiques d'un sol acide a été évalué par une expérimentation en lysimètre. Les traitements ont inclus l'absence, le placement à la surface du sol et l'incorporation dans le sol (avec et sans couverture de légumineuse) de résidus organiques (litière forestière ou litière forestière plus résidus de récolte).
- Les résidus placés à la surface de sol ont augmenté la croissance des jeunes plants. Les résidus organiques ont réduit les pertes en nutriments (NO_3^- , Ca, Mg) et ont abouti à l'accumulation de nutriments dans le sol. Les résidus de récolte ont positivement affecté le statut nutritionnel en K des jeunes plants et ont augmenté l'accumulation de K dans le sol.
- Une couverture de légumineuse réduit les pertes de substances nutritives du sol (N, Ca, Mg et K) pendant la première période de croissance du jeune plant ; elle améliore aussi le statut nutritionnel du jeune plant (N et P), mais sans aucun effet sur la croissance.
- Les résidus de récolte plus la litière forestière placés à la surface de sol ont constitué la gestion la plus appropriée pour à la fois réduire les pertes en nutriments par lessivage et accroître la hauteur des jeunes plants à la fin de la période d'expérimentation (deux ans).

1. INTRODUCTION

Plantations of *Pinus pinaster* represent the largest forest area in Portugal and currently reach 7.1×10^5 ha (IFN, 2006). These plantations are mainly located in the northern and central regions of the country, where they mostly occupy soils

with low nutrient status (Podzols, dystric Cambisols and dystric Regosols), which are highly sensitive to management operations resulting in major losses of nutrients. At the end of the rotation (70–80 years) of pine plantations growing in sandy soils, harvest residues can contain a substantial amount of nutrients (425 and 673 kg ha^{-1} of N and Ca, respectively); the amounts of both nutrients in the forest floor litter layer can reach about 300 kg ha^{-1} (PRAXIS, 2001). Therefore, practices

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of site preparation that include removal of harvest residues and the forest floor layer may considerably reduce soil fertility and can lead to negative nutrient budgets, such as observed in pine plantations in Northern Spain (Dambrine et al., 2000; Merino et al., 1998). In this context, management of organic residues during the inter-rotation period is a crucial factor for maintaining fertility and sustained productivity in those plantations. In fact, the removal of harvest residues to minimize fire risks and to supply future power stations has been encouraged (DGRF, 2006), although little attention has been given to the effects of such practices on the nutrient status of forest soils and on long-term site productivity.

Organic residue management practices could affect environmental soil conditions and alter decomposition rates of organic residues and the subsequent mineralization and availability of nutrients (Smethurst and Nambiar, 1990). Retention of harvest residues at the site, especially branches with high C/N ratios, have a potential capacity for N immobilization during their decomposition. This effect reduces losses of N by leaching in the first years after harvesting (Smethurst and Nambiar, 1995) when mineral N in the soil may be in excess for young tree requirements. Those residues release immobilized N at a later stage of decomposition, offering a greater synchrony between supply and demand by growing trees (Johnson, 1995). Furthermore, management options that increase the degree contact between organic residues and soil can enhance favorable conditions for microbial activity and, thus, accelerate the decomposition process (Carlyle et al., 1998; Pérez-Batallón et al., 2001).

The use of N-fixing leguminous plants may also contribute to improving soil fertility, since they incorporate organic matter and N in the soil (Corbeels et al., 2003) and reduce nutrient losses through leaching during the early stage of plantation development (Aggangan et al., 1999). This may also promote tree growth in forest plantations (Nambiar and Nethercott, 1987) and decrease competition from weeds (Alley et al., 1999; Smethurst and Nambiar, 1990). However, leguminous plants may compete for site resources, and pine growth suppression has also been reported (Alley et al., 1999; Gakis et al., 2004).

In this context, a lysimeter experiment was carried out under controlled conditions, over 24 months, to evaluate the short-term effects of removal, maintenance on the soil surface and incorporation into the soil (with or without legume cover cropping) of organic residues (forest floor litter alone or combined with harvest residues) on the growth and nutrition status of *Pinus pinaster* seedlings, nutrient leaching, and soil chemical properties.

2. MATERIALS AND METHODS

2.1. Lysimeter station

The study was carried out in a lysimeter station located at the Instituto Superior de Agronomia, Lisboa. The area has a Mediterranean climate, tempered by an oceanic influence. The 30-year mean (1971–2000) rainfall monitored in the adjacent meteorological station of Lisboa/Ajuda was 675 mm and the mean annual temperature

was 16.4 °C, ranging from a monthly mean of 11.8 °C in January to 21.0 °C in August (INMG, 2007). During the experimental period (January 1998–February 2000) rainfall was 459, 750 and 44 mm, respectively, in 1998, 1999 and 2000 (January and February).

The lysimeter station consisted of 28 lysimeters constructed in PVC, with a surface area of 0.28 m², an inner diameter of 0.6 m and a depth of 1 m. Each lysimeter had, at its bottom, a drainage system 10 cm thick constituting, from the bottom to the top, a double plastic mesh (2 mm), a gravel layer washed with deionized water, a double layer of filtering material (Terram) and a washed sand layer, which was directly in contact with the experimental soil substrate. In the lysimeter base, a hole was connected, through a plastic tube, to a sampling bottle (5 L), where leachates were continuously collected. Lysimeters were exposed to weather conditions in an open fenced area, to avoid animal access, and adjacent to the aforementioned meteorological station. They were placed 1 m apart on a concrete base with openings for the drainage tubes, the surrounding space between them having been filled with sand material covered by pine bark. The lysimeters' top edges were 15 cm above the covering bark surface.

2.2. Materials

The mineral soil and the organic residues used in the experiment were collected from an 80-year-old maritime pine plantation (*Pinus pinaster*) in the Leiria National Forest (lat. 39° 42' N; long. 8° 03' W; 40 m a.s.l.), located in central coastal Portugal. The plantation density was about 200 tree ha⁻¹ and its productivity (commercial timber with bark) was 6 m³ ha⁻¹ y⁻¹ (PRAXIS, 2001).

Soils are mostly Haplic Arenosols (WRN, 2006) developed on coastal sand dunes. Soil substrates were collected from an Ah horizon and a C horizon. Each soil substrate was sieved (< 2 mm) and six subsamples were taken for analysis. Both horizons were sandy and their characteristics are shown in Tab. I. The Ah horizon had lower pH (4.8), and a greater organic C content (22.2 g kg⁻¹) and C/N ratio (35.8) than the C horizon.

The forest floor litter layer (L and F layers) and harvest residues (green needles, 0.5–5.0 cm diameter branches and cones) were collected at harvesting time (November, 1997), by randomized sampling of 1 m² areas for a total of five samples for each material. Samples were dried (80 °C), and subsamples were taken for chemical analysis.

2.3. Treatments

Effects of organic residue management were assessed through three main options: absence of organic residues, presence of forest floor and presence of forest floor plus harvest residues. Options related to residue placement (on the surface or incorporated into the soil) and legume cover cropping were also considered. Specifically, seven treatments were installed as described in Tab. II. Treatments were replicated four times and were established at random.

In each lysimeter, over the respective drainage system, were placed 180 kg (105 °C dry weight) of sieved homogenized mineral soil of the C horizon (about 0.45 m depth), packed at a bulk density of about 1.4 g cm⁻³; over this material were placed 130 kg of the A horizon (about 0.35 m depth, bulk density of about 1.3 g cm⁻³). Except in the treatment NR, 2.5 kg (dry weight) of forest floor litter were applied in each lysimeter. In the treatments LHS, LHI and LHL, 0.5 kg of needles, 2.4 kg of branches (cut into 20-cm long pieces) and

Table I. Particle size, pH values and contents of organic C, total N, extractable bases, Al and P of soil mineral substrates (< 2 mm, 105 °C dry weight) used in the lysimeters.

Horizon	Particle size			C	N	pH H ₂ O	Extractable bases				Al ⁺³	P
	Sand	Silt	Clay				Ca ²⁺	Mg ²⁺	Na ⁺	K ⁺		
Ah	974 ±9	20 ±3	6 ±6	22.2 ±9.3	0.62 ±0.27	4.83 ±0.43	2.51 ±1.12	0.56 ±0.28	0.15 ±0.04	0.09 ±0.04	0.15 ±0.10	5.6 ±1.9
C	983 ±9	10 ±11	7 ±7	0.2 ±0.1	0.02 ±0.01	5.84 ±0.14	0.06 ±0.03	0.01 ±0.00	0.12 ±0.05	0.01 ±0.00	0.03 ±0.01	3.0 ±1.0

Values are means ($n = 6$) ± standard deviation (S.D.).

Table II. Description of treatments.

Treatment	Forest litter	Harvest residue	Legume
NR	–	–	–
LS	Surface	–	–
LI	Incorporated	–	–
LL	Incorporated	–	Yes
LHS	Surface	Surface	–
LHI	Incorporated	Incorporated	–
LHL	Incorporated	Incorporated	Yes

0.5 kg of cones were also applied in each lysimeter. The amounts of harvest residues were about two times greater than those measured in an 80-year-old pine plantation (PRAXIS, 2001).

In the LI, LL, LHI and LHL treatments, residues were mixed with the Ah horizon and introduced into the top 30–35 cm depth of lysimeters. One seedling of *P. pinaster* was planted on 27 January 1998 in each lysimeter. A uniform set of seedling plants was used with similar vigor and growth (15.9 cm height). Each seedling received 10 g of NP fertilizer (18:21). In the treatments LL and LHL, a legume (*Lupinus luteus*) was seeded twice (January, 1998 and 1999) at a rate of 107 kg ha⁻¹. Treatment installation was concluded in 29 January 1998. Herbaceous plants other than *L. luteus* in lysimeters were controlled by hand.

Irrigation water was applied in lysimeters (depending on the frequency of rainwater) to maintain soil moisture above 60% of available water capacity (difference between water content at 6.2 and 1500 kPa), moisture being measured by Time Domain Reflectometry (6050X1 Trase System, Soil Moisture Equipment Corp., CA, USA). In the NR treatment, a pair of stainless steel wave guides 60 cm long was permanently inserted into the soil of each lysimeter and measurements were carried out daily. On 9 and 16 March 1998, (NH₄)₆Mo₇O₂₄ was added in treatments LL and LHL through irrigation water to promote *L. luteus* development. Over the experimental period, the total amount of water applied was 414 mm in all treatments. Amounts of nutrients supplied in the lysimeters of each treatment are shown in Table III.

2.4. Measurements and sampling

The height of seedlings was measured monthly for 24 months. From 27 September 1998, basal stem diameter at 5 cm above ground level was measured using a digital caliper. To assess seedling nutrition status, eight pairs of needles were sampled from the upper

Table III. Amounts of nutrients (g) supplied per lysimeter as fertilizer, irrigation, rainfall, forest floor litter and harvest residues.

	N	Ca	Mg	K	P
Fertilizer	1.800	0.000	0.000	0.000	0.916
Irrigation	0.060	1.250	0.217	0.068	< 0.001
Rainfall	0.194	0.916	0.098	0.087	< 0.001
Forest floor litter	8.075	10.600	1.500	2.325	0.375
Harvest residues	13.282	19.358	2.914	4.642	0.719

half of the canopy of each seedling on two occasions (November 1998 and 1999).

Tree seedling sampling to determine biomass and respective nutrient contents was performed in March 2000. All seedlings were cut at the base and root systems were pulled out from the soil. Dry weight of each component was calculated from total fresh weight and moisture content, and a subsample was taken for subsequent chemical analysis.

Samplings to assess legume biomass and nutrient content took place in May 1998 and 1999, when plants reached the flowering stage. Legume plants were pulled out from the soil, and above- and below-ground components were separated, weighed and dried. A subsample was used for chemical analysis. Later, plant components were spread over the soil surface in the LL and LHL treatments.

Samples of rainwater, according to rainfall events, were collected for analysis using an open collector placed in the adjacent meteorological station. The volume of lysimeter leachates was recorded and subsamples were collected for analysis according to rainfall events. Both rainwater and leachate subsamples were stored at –15 °C immediately after collection.

Soil was sampled at the end of the experiment (March 2000), before destructive tree sampling. In each lysimeter, after removal of organic residues remaining on the soil surface, four soil cores (65 mm diameter) were collected from three depths (0–10, 10–20 and 20–30 cm), and combined in a composite sample for each lysimeter, which was dried (45 °C) and sieved (< 2 mm).

2.5. Laboratory procedures

Organic residues were dried at 45 °C and ground in a laboratory mill to a particle size of 0.5 mm. The mineral elements (Ca, Mg, K and P) were determined after ashing (6 h at 450 °C) which was taken up in HCl. Total N was determined using Kjeldhal digestion. The carbon content was measured by wet oxidation. The Ca, Mg and K

Table IV. Height (cm) and diameter (mm) of seedlings after 12 and 24 months of study (February 1999 and 2000), and total biomass (g/lysimeter, dry weight) at the end of the experiment (March 2000). Values are means ($n = 4$) \pm S.D.

Treatment	Height		Diameter		Biomass
	Feb 1999	Feb 2000	Feb 1999	Feb 2000	March 2000
NR	5.3 \pm 10.1	8.3 \pm 8.3 b	0.0 \pm 0.8 ac	7.8 \pm 0.7 ab	805.6 \pm 100.7 ab
LS	81.0 \pm 4.1	113.3 \pm 2.1 a	19.3 \pm 0.4 abc	28.5 \pm 3.2 ab	849.0 \pm 64.0 ab
LI	70.3 \pm 4.2	94.7 \pm 4.6 b	17.7 \pm 0.5 b	26.6 \pm 2.3 ab	738.6 \pm 59.8 ab
LL	65.3 \pm 4.1	96.0 \pm 2.9 b	20.2 \pm 0.8 c	26.0 \pm 3.2 ab	674.0 \pm 161.2 b
LHS	85.3 \pm 15.9	116.0 \pm 9.6 a	18.5 \pm 0.2 abc	30.2 \pm 1.3 a	960.5 \pm 135.7 a
LHI	63.7 \pm 19.4	91.3 \pm 9.5 b	18.2 \pm 1.8 abc	25.2 \pm 1.8 b	782.3 \pm 93.5 ab
LHL	67.3 \pm 4.5	94.3 \pm 2.5 b	19.3 \pm 0.8 abc	26.2 \pm 0.7 ab	851.6 \pm 91.8 ab

Different letters in the same column denote significant differences ($p < 0.05$) among treatments by the Tukey multiple range test.

Table V. Results of Multifactor ANOVAs showing the effects of residue type (forest floor litter or harvest residues), residue placement (soil surface or incorporation into the soil) and legumes (presence or absence) on height (cm) and diameter (mm) (February 2000) and total biomass (g/lysimeter) of tree seedlings (March 2000), and on nutrient content in green needles (mg g^{-1} , November 1998 and 1999) and nutrient content in tree seedling biomass (g/lysimeter) (March 2000). Levels of significance: *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$ and n.s. not significant.

Factors	Seedlings			Green needles						Seedling biomass				
	Height	Diameter	Biomass	N 1998	N 1999	K 1998	K 1999	P 1998	P 1999	N	Ca	Mg	K	P
Residue	n.s.	n.s.	*	n.s.	n.s.	n.s.	**	n.s.	n.s.	**	n.s.	n.s.	***	***
Placement	***	**	*	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.	*	**	**	n.s.	*
Legume	n.s.	n.s.	n.s.	***	**	n.s.	*	**	**	*	n.s.	n.s.	n.s.	n.s.
Residue \times Placement	n.s.	n.s.	n.s.	n.s.	n.s.	**	n.s.	n.s.	n.s.	n.s.	*	n.s.	*	n.s.
Residue \times Legume	n.s.	n.s.	n.s.	n.s.	n.s.	*	n.s.	n.s.	n.s.	n.s.	**	n.s.	n.s.	n.s.

were measured by atomic absorption spectrometry (AAS) and P by colorimetry.

Samples of rainwater and leachates were centrifuged and filtered through a 0.45- μm membrane and pH was measured. Concentrations of NO_3^- -N and NH_4^+ -N (limit of detection 0.01 mg L^{-1}) were determined with a segmented flow autoanalyzer, using the hydrazinium reduction and the modified Berthelot method, respectively (Houba et al., 1994). The Ca, Mg and K were measured by AAS, the limit of detection being 0.001 mg L^{-1} . The accuracy of determinations was checked by using certified reference samples.

Soil properties were determined on the fine fraction (< 2 mm), following the methodology described by Póvoas and Barral (1992). Soil pH was determined potentiometrically in water (soil:solution ratio 1:2.5). Organic C was determined by wet oxidation following the method described by De Leenheer and Van Hove (1958). Total N was determined using Kjeldhal digestion. Extractable P was determined using the Egnér-Riehm method (Egnér et al., 1960). The extractable base cations were assessed by 1 M NH_4OAc , adjusted at pH 7.0, and the extractable Al was determined after extraction with 1 M KCl. The Ca, Mg, Na, K and Al were measured by AAS and P by colorimetry.

2.6. Statistical analysis

Treatment effects on growth and nutrition status of seedlings, nutrient leaching, and soil chemical properties were tested by analysis of variance (ANOVA) and comparison among treatment means using the Tukey multiple range test. In order to discriminate the rela-

tive importance of the key factors that were established (residue type, residue placement and legumes), a 3-factor analysis of variance was carried out. Statistical procedures were performed using Statgraphics plus 5.1 (Manugistics Inc.).

3. RESULTS

3.1. Seedling growth and biomass

Over the experimental period, seedling growth parameters (height, diameter and total biomass) of the treatment without organic residues (control) were mostly of the same magnitude as those observed in the other treatments (Tab. IV). However, at the end of the study, the seedling height in the treatments with organic residues placed on the soil surface (LS and LHS) was significantly greater than in control. Residues placed on the soil surface significantly increased seedling height ($p < 0.001$) and diameter ($p < 0.01$) by 23 and 19%, respectively, in comparison with their incorporation (Tab. V). The growth increase was irrespective of the residue type (no significant residue \times placement interaction). Tree seedling total biomass was significantly affected ($p < 0.05$) by both residue type and placement. Legume cover cropping did not lead to significant differences in height, diameter or seedling biomass.

Table VI. Aboveground and belowground biomass (g/lysimeter, dry weight) and contents of nutrients (g/lysimeter) in legumes, in the LL and LHL treatments. Values are means \pm SD ($n = 4$). Differences between treatments were not statistically significant ($p > 0.05$, ANOVA procedure).

Treat	Above-ground	Below-ground	N	Ca	Mg	K	P
May 1998							
LL	74.6 \pm 34.4	19.4 \pm 3.8	1.88 \pm 0.40	0.57 \pm 0.10	0.26 \pm 0.05	1.26 \pm 0.42	0.16 \pm 0.02
LHL	74.4 \pm 43.1	20.1 \pm 4.1	2.30 \pm 0.48	0.58 \pm 0.12	0.31 \pm 0.08	1.34 \pm 0.65	0.19 \pm 0.05
May 1999							
LL	31.0 \pm 10.2	7.9 \pm 2.6	0.79 \pm 0.24	0.21 \pm 0.06	0.08 \pm 0.02	0.38 \pm 0.10	0.06 \pm 0.01
LHL	17.1 \pm 6.0	4.4 \pm 1.5	0.43 \pm 0.09	0.12 \pm 0.04	0.05 \pm 0.01	0.29 \pm 0.09	0.02 \pm 0.01

Table VII. Contents (mg g⁻¹) of N, K and P in green needles over the experiment period (November 1998 and 1999), and in tree seedling biomass (g/lysimeter) at the end of the experiment (March 2000).

Treatments	Green needles						Seedling biomass				
	N		K		P		N	Ca	Mg	K	P
	1998	1999	1998	1999	1998	1999					
NR	14.2	5.9	8.2	3.4	1.1	0.5	3.37	2.49	0.66	1.27	0.33
	± 1.3 a	± 0.4 a	± 0.4 ab	± 0.5 a	± 0.1	± 0.0 a	± 0.51 ab	± 0.22 ab	± 0.10 ab	± 0.24 a	± 0.05 ab
LS	11.0	5.3	8.6	3.2	1.0	0.5	3.72	3.20	0.81	1.36	0.37
	± 0.7 b	± 0.2 a	± 0.8 ab	± 0.4 a	± 0.0	± 0.0 a	± 0.23 ab	± 0.21 a	± 0.01 a	± 0.21 a	± 0.03 ab
LI	12.0	5.9	8.4	4.1	1.0	0.6	3.17	2.32	0.68	1.43	0.29
	± 1.0 ab	± 0.6 a	± 1.3 ab	± 0.4 a	± 0.0	± 0.1 a	± 0.29 a	± 0.38 b	± 0.05 ab	± 0.11 a	± 0.04 a
LL	14.8	9.5	7.9	4.7	1.2	0.8	3.45	1.52	0.51	1.32	0.32
	± 2.2 a	± 3.3 b	± 0.6 ab	± 1.1 ab	± 0.1	± 0.2 b	± 0.39 ab	± 0.43 c	± 0.15 b	± 0.36 a	± 0.07 a
LHS	12.0	5.6	7.1	4.9	1.0	0.6	4.03	2.72	0.84	2.79	0.44
	± 1.9 ab	± 0.1 a	± 0.9 a	± 0.1 ab	± 0.1	± 0.0 a	± 0.51 ab	± 0.30 ab	± 0.12 a	± 0.39 b	± 0.08 b
LHI	10.5	6.1	8.8	4.3	1.0	0.6	3.63	2.29	0.64	2.36	0.38
	± 0.4 b	± 0.2 a	± 0.4 ab	± 1.3 ab	± 0.2	± 0.2 a	± 0.37 ab	± 0.31 b	± 0.11 ab	± 0.20 bc	± 0.04 ab
LHL	14.1	7.7	9.3	6.0	1.2	0.7	4.26	2.34	0.68	2.06	0.39
	± 0.8 a	± 0.9 ab	± 0.5 b	± 1.1 b	± 0.1	± 0.1 ab	± 0.72 b	± 0.27 b	± 0.09 ab	± 0.26 c	± 0.02 ab

Values are means ($n = 4$) \pm S.D. Different letters in the same column denote significant differences ($p < 0.05$) among treatments by the Tukey multiple range test.

3.2. Biomass and nutrient content in legumes

Aboveground biomass was much greater in 1998 (about 74 g) than in 1999 (31.0 and 17.1 g in the D and G treatments, respectively, Tab. VI). Belowground biomass ranged from 4.4 g in 1999 to 20.1 g in 1998, being about 25% of the total biomass. Differences among treatments were not statistically significant. Nutrient contents were also greater in the first year of the study. The content of N (1.88–2.30 g) in legumes, in 1998, was close to that applied as fertilizer, but much smaller than that applied as residues (Tab. III).

3.3. Nutrient content in seedling needles

In the first year of the experiment, contents of N in seedling needles (Tab. VII) of treatments with organic residues (10.5–12.0 mg g⁻¹) were lower than in their absence

(14.2 mg g⁻¹), but similar contents were observed in the second year (5.3–6.1 and 5.9 mg g⁻¹, respectively).

The effect of residue type on nutrient contents of seedling needles was only detected for K ($p < 0.01$) at the end of the study period (Tab. V), the values being greater in the treatments including forest floor litter plus harvest residues than in those with forest floor litter only (5.21 and 4.13 mg g⁻¹, respectively). In the first year, harvest residues increased K needle content when applied on the soil surface (interaction residue \times placement, $p < 0.01$), and when combined with legume cover cropping (interaction residue \times legume, $p < 0.05$). Residue placement options did not show significant differences in any of the nutrients analyzed in green needles. Legume cover cropping led to significantly greater N ($p < 0.001$ and $p < 0.01$, respectively, in 1998 and 1999) and P ($p < 0.01$) needle contents in both years (Tab. V), and K ($p < 0.05$) contents in November 1999. No significant differences for Ca and Mg contents were detected among treatments.

3.4. Nutrient content in tree seedling biomass

Similar N seedling contents were observed in the treatments with organic residues (3.17–3.46 g) and in the control (3.37 g) (Tab. VII). Contents of N, K and P were significantly greater in the treatments with forest floor litter plus harvest residues (4.13, 2.40 and 0.42 g, respectively) than in those with forest floor litter only (3.60, 1.36 and 0.34 g, respectively, Tab. V). The interaction residue \times placement showed that differences in K contents were greater in the soil surface placement.

Residues placed on the soil surface showed higher biomass N (3.87 g), Ca (2.96 g), Mg (0.83 g) and P (0.41 g) contents than when they were incorporated (3.40, 2.11, 0.63 and 0.35 g, respectively) ($p < 0.05$, $p < 0.01$, $p < 0.01$ and $p < 0.05$, respectively; Tab. V). Differences in Ca biomass content between surface and incorporation placement were greater for forest floor litter than for forest floor litter plus harvest residues (interaction residue \times placement, $p < 0.05$, Tab. V). Legume cover cropping positively affected N contents ($p < 0.05$) in biomass (Tabs. V and VII), but reduced biomass Ca content when only the forest floor litter layer was applied (interaction residue \times legume, $p < 0.01$).

3.5. Leachate volume and nutrient leaching

Rainfall and irrigation water amounted to 1611 mm during the experiment. The NR treatment had the largest leachate volume (430 mm, Fig. 1). Legume cover cropping reduced the accumulated volume leached by 15% ($p = 0.018$) when compared with its absence. The greatest volume of leachate was measured during the early experimental period, that is, from January to June of 1998.

Mineral N was mostly leached during the first six months, with 47–93% of the NO_3^- -N and 84–91% of the NH_4^+ -N being leached during the experimental period (Fig. 1). Nitrate was the dominant form of the mineral N (about 85%). The largest mineral N- NO_3^- , Ca and Mg losses (Fig. 1) were observed in the absence of organic residues (treatment NR), although significant differences were only detected relatively to N- NO_3^- in treatments with legume (LL and LHL) and with harvest residues incorporated (LHI).

Residue type and placement and legume cover did not significantly affect leaching of NO_3^- , NH_4^+ , Ca or Mg. Accumulated loss of K was significantly affected by residue type ($p = 0.0006$), the values being greater in lysimeters with forest floor litter plus harvest residues (547.4 mg/lysimeter) than in those with forest floor litter only (369.0 mg/lysimeter). Over the first year of study, legume cover cropping reduced accumulated losses of N, Ca, Mg and K (40, 25, 28 and 50%, respectively) in relation to its absence.

3.6. Soil chemical properties

At the end of the experiment, values of pH- H_2O (Tab. VIII) in the 10 cm topsoil layer were significantly greater in the NR (5.67) than in the other treatments (4.45–5.18). In the 0–30 cm

soil layer, values in lysimeters with forest floor litter plus harvest residues were significantly reduced in relation to those with forest floor litter only ($p < 0.001$). A significant reduction was also observed for soil surface placement ($p < 0.05$).

Down to 30 cm topsoil depth, the presence of organic residues increased organic C and nutrient contents (total N, extractable Ca, Mg, K and P) when compared with their absence (NR treatment), but significant differences were only observed for some treatments. Organic C contents in the LS (in the 0–10 cm topsoil layer) and in the LHI treatments (at 20–30 cm) were significantly greater than those observed in the NR. Total N contents in the NR were significantly lower than in the other treatments, except in the treatment LHS, at least in one layer down to 30 cm depth. Nevertheless, it is noted that C and N amounts corresponding to organic residues on the soil surface (LHS treatment) were not taken into account and, therefore, differences observed could be more pronounced in the long term.

In the 10–30 cm soil layer, contents of extractable Ca in treatments LL and LHI (and at least in one layer in treatments LS, LI and LHL) were significantly greater than in the NR. Extractable Mg contents were significantly greater than in the NR treatment (except LHS) at least in one layer down to 30 cm depth. Contents of extractable K in the NR were significantly lower than those observed in several layers of the LHI and LHL treatments. The LHI treatment showed mostly significant greater extractable K contents than in the treatments with forest floor litter only (LS, LI and LL). Extractable P contents in the control (10–30 cm soil layer) were significantly smaller than in the treatments LHI and LHL.

The effect of residue type on extractable K contents was observed in the 0–30 cm soil layer, being 38, 64 and 33% ($p = 0.0009$, $p = 0.0004$ and $p < 0.05$ in 0–10, 10–20 and 20–30 cm, respectively) greater in lysimeters with forest floor litter plus harvest residues. Forest floor litter placed on the soil surface increased contents of Ca, Mg, K and P in the 0–10 cm topsoil layer as compared with their incorporation (interaction residue \times placement, $p < 0.05$). Incorporation of residues increased Ca, Mg and K contents in the 10–20 and 20–30 cm soil layers (about 22, 26 and 40%, respectively). Legume cover did not significantly affect any of the elements for the analyzed depths. Legume cropping increased N content in the topsoil surface layer when combined with forest floor litter plus harvest residues (interaction residue \times legume, $p < 0.05$).

4. DISCUSSION

The methodology used in this study presents some limitations, as the soil structure and water transfers are disturbed in the lysimeter installation and the amounts of residues used are about two times greater than at the harvest of the first rotation of *Pinus pinaster* plantations (PROBRAVO, 2007). Although experimental conditions have a limited value in assessing the complexity of field situations, they give insight into the effect of harvest residues regarding growth and nutrition of *P. pinaster*, nutrient leaching and soil chemical properties.

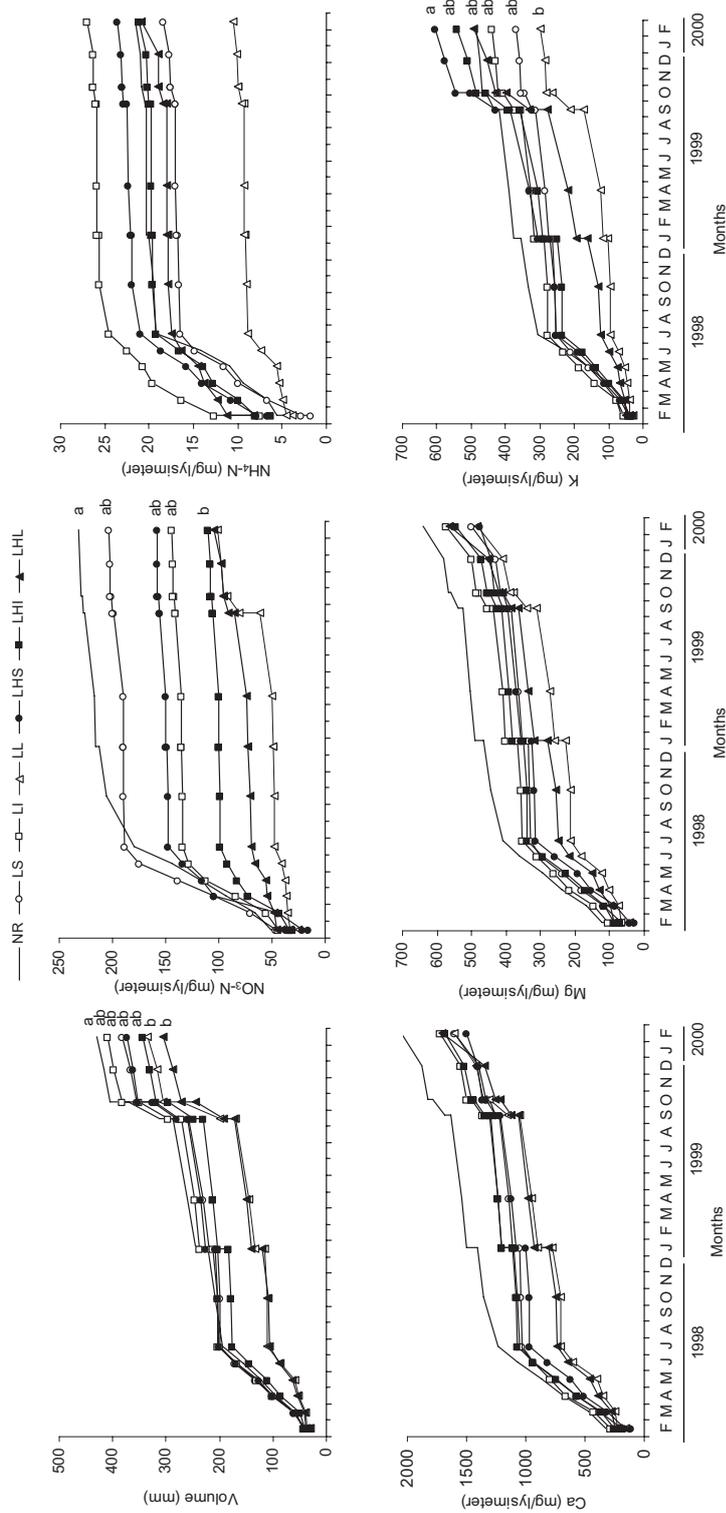


Figure 1. Volume of accumulated leached (mm) and accumulated amounts (mg/lysimeter) of NO₃-N, NH₄-N, Ca, Mg and K collected in leachates of lysimeters over the experimental period (*n* = 4). Different letters for data at the end of the study denote significant differences (*p* < 0.05) among treatments by the Tukey multiple range test.

Table VIII. Values of pH and contents of organic C and total N (g kg^{-1}), extractable Ca, Mg, K ($\text{cmol}_c \text{ kg}^{-1}$) and P (mg kg^{-1}) according to soil depth.

Prof. cm	Treatments						
	NR	LS	LI	LL	LHS	LHI	LHL
	pH H ₂ O						
0–10	5.67 ± 0.16a	5.02 ± 0.13b	5.16 ± 0.10be	5.18 ± 0.11 b	4.45 ± 0.12 c	4.87 ± 0.11 ef	4.79 ± 0.13 df
10–20	4.77 ± 0.11 a	4.59 ± 0.03 ac	4.63 ± 0.05 ac	4.68 ± 0.02 ac	4.23 ± 0.13 b	4.40 ± 0.09 bd	4.54 ± 0.05 cd
20–30	5.13 ± 1.02 a	4.48 ± 0.14 ab	4.64 ± 0.12 ab	4.71 ± 0.11 ab	4.09 ± 0.07 b	4.41 ± 0.07 ab	4.40 ± 0.17 ab
	Organic C						
0–10	19.1 ± 2.6 a	30.1 ± 4.7 b	27.7 ± 2.7 ab	29.3 ± 2.4 ab	28.7 ± 7.8 ab	32.3 ± 8.8 ab	32.9 ± 3.4 ab
10–20	23.4 ± 5.8	33.3 ± 11.3	31.0 ± 4.3a	30.5 ± 2.7	26.0 ± 1.8	30.5 ± 3.3	30.1 ± 12.8
20–30	22.6 ± 1.3a	24.4 ± 4.6ab	28.5 ± 2.5ab	26.4 ± 4.3ab	30.2 ± 4.6ab	32.0 ± 4.4b	30.3 ± 2.5ab
	Total N						
0–10	0.49 ± 0.07 a	0.77 ± 0.08 b	0.76 ± 0.16 b	0.70 ± 0.07 ab	0.59 ± 0.05 ab	0.68 ± 0.17 ab	0.77 ± 0.10 b
10–20	0.43 ± 0.03 a	0.63 ± 0.05 bc	0.71 ± 0.08 bc	0.77 ± 0.12 b	0.54 ± 0.10 ac	0.63 ± 0.06 bc	0.74 ± 0.12 b
20–30	0.49 ± 0.04 a	0.61 ± 0.12 ab	0.67 ± 0.07 ab	0.68 ± 0.03 ab	0.62 ± 0.11 ab	0.71 ± 0.13 ab	0.73 ± 0.15 b
	Extractable Ca						
0–10	2.94 ± 0.37 ab	4.11 ± 1.71 a	3.48 ± 0.05 ab	3.12 ± 0.69 ab	3.24 ± 0.28 b	3.46 ± 0.35 ab	3.22 ± 0.21 ab
10–20	1.68 ± 0.16 a	2.28 ± 0.21 bc	2.47 ± 0.06 b	2.40 ± 0.25 bc	1.87 ± 0.17 ac	2.66 ± 0.45 b	2.40 ± 0.27 bc
20–30	1.82 ± 0.27 a	1.97 ± 0.19 ac	2.33 ± 0.10 ab	2.53 ± 0.37 bc	2.04 ± 0.20 ac	2.80 ± 0.30 b	2.30 ± 0.32 ab
	Extractable Mg						
0–10	0.32 ± 0.02 a	0.69 ± 0.21 b	0.53 ± 0.04 ab	0.54 ± 0.12 ab	0.50 ± 0.08 ab	0.64 ± 0.24 b	0.63 ± 0.08 ab
10–20	0.27 ± 0.05 a	0.43 ± 0.07 ab	0.50 ± 0.02 b	0.49 ± 0.04 ab	0.40 ± 0.05 ab	0.63 ± 0.16 b	0.62 ± 0.18 b
20–30	0.33 ± 0.06 a	0.43 ± 0.04 ab	0.52 ± 0.01 bc	0.53 ± 0.06 bc	0.48 ± 0.06 ab	0.66 ± 0.15 c	0.49 ± 0.06 ab
	Extractable K						
0–10	0.06 ± 0.01 a	0.12 ± 0.03 ab	0.09 ± 0.01 ac	0.06 ± 0.02 a	0.13 ± 0.02 bc	0.14 ± 0.04 b	0.13 ± 0.03 bc
10–20	0.03 ± 0.00 a	0.06 ± 0.01 a	0.06 ± 0.01 ac	0.06 ± 0.01 a	0.08 ± 0.00 ac	0.15 ± 0.08 b	0.13 ± 0.03 bc
20–30	0.05 ± 0.01 a	0.06 ± 0.01 a	0.10 ± 0.07 ab	0.07 ± 0.01 ab	0.09 ± 0.01 ab	0.13 ± 0.02 b	0.09 ± 0.01 ab
	Extractable P						
0–10	5.5 ± 1.7	8.2 ± 2.3	5.0 ± 0.8	6.6 ± 1.8	6.6 ± 2.9	9.9 ± 3.3	9.7 ± 1.3
10–20	3.7 ± 1.1 a	4.9 ± 0.9 ac	5.1 ± 0.8 ac	5.6 ± 1.1 ac	3.4 ± 1.2 a	8.8 ± 2.5 b	7.4 ± 0.9 bc
20–30	1.5 ± 1.1 a	3.4 ± 0.7 ac	2.6 ± 1.4 a	2.8 ± 1.3 ac	4.8 ± 0.6 ab	8.7 ± 3.1 b	6.9 ± 2.8 bc

Different letters in the same row denote significant differences ($p < 0.05$) among treatments by the Tukey multiple range test. Values are means ($n = 4$) ± S.D.

In the present study, organic residues on the soil surface improved seedling growth in comparison with their incorporation. Other authors have also reported enhanced growth of pine (Egnell and Valinger, 2003) and eucalypt (Mendham et al., 2003; Nzila et al., 2001) in low fertility soils with harvest residues maintained on the soil surface. However, limited effect has also been reported in Douglas-fir (Roberts et al., 2005) and eucalypt plantations (Mendham et al., 2001) with relatively high fertility, suggesting that the effect of residue management on tree growth may vary with site quality. Our results are in line with those observed in a similar experiment with *Eucalyptus globulus* by Soares et al. (2002), who suggest that improvement of seedling growth through harvest residue placement on the soil surface could be related to greater root development. However, other factors might be involved, such as the lower N and P immobilization reported by Carlyle et al. (1998) for this placement in relation to their incorporation, which is also in agreement with the lower N and P influxes

to roots as reported by Soares et al. (2002). Residues placed on the soil surface may also reduce soil water evaporation, as observed in a lysimetric (Gómez-Rey et al., 2007) and a field (Pérez-Batallón et al., 2001) experiment. Therefore, as already suggested for pine seedlings planted under Mediterranean conditions (Valdecantos et al., 2006), water availability may have positively influenced seedling growth.

Amounts of N, Ca and Mg leached from the soil were low when compared with the amount supplied. For example, the amount of N leached (254 mg) in the absence of organic residues over the study period was of the same magnitude as the mean annual input through rainfall (194 mg), and it was only 14% of N applied as fertilizers. This low leaching may be explained by the high soil C/N ratio (35.8) which is above the critical condition for N release. In addition, the low N leaching observed in treatments with residues may be related to the high C/N ratio of forest floor litter (72) and harvest residues (67 and 185 for needles and branches, respectively), which may act as

a sink for N during their decomposition (Ganjegunte et al., 2004; Hyvönen et al., 2000) and contribute to maintaining N in the system. This is also in agreement with the fact that N assimilated by tree seedlings was of the same magnitude as that observed in the absence of residues. Therefore, the maintenance of residues in the site may contribute to enhancing soil N fertility in the long term, as losses through leaching were negligible. However, it has been reported that the maintenance of organic residues may induce N deficiency during the early stages of tree growth in N-poor sites (Mendham et al., 2003). In fact, during the first year of the experiment, contents of N in seedling needles of treatments with organic residues were lower than in their absence, but this difference was not associated with reduced tree growth. In addition, the similarity in needle N contents, N amounts accumulated by seedlings and N losses through leaching, at the end of the study, suggest that maintenance of residues did not enhance N availability during the study period. Nitrogen contents observed in needles are of the same magnitude as those measured in similar treatments applied over two years in the field (PROBRAVO, 2007), that is, in the site from which the present experimental soil and residues were collected.

Harvest residues have a positive influence on K nutrition, as two years after planting a significant increase in green needle K contents and pine seedlings was observed in the treatments where they were applied. Also, greater K losses by leaching and extractable K accumulation in the soil were observed. This may be explained by the high mobility of K, which is readily leached from decomposing pine residues (Ganjegunte et al., 2004; Xu, 2006). In addition, this trend might also be increased by the double amount of residues applied in treatments with harvest residues. In fact, contents of K in needle seedlings were slightly greater than those reported for similar seedlings under field conditions, where the amount of experimental residues was not doubled (PROBRAVO, 2007).

An enhancement of organic C and N contents was observed at the end of the study in the mineral substrate, due to the presence of organic residues. This was noticeable along soil depth in treatments with both forest floor and harvest residues, and it was independent of their placement. In the top 0–30 cm soil layer, an increase in organic C (33–51%) and total N (24–60%) contents was observed in the presence of organic residues relatively to their absence. These results are in agreement with those reported by Chen and Xu (2005), who observed a favorable effect of residue maintenance on soil C and N status in a pine plantation grown in sandy soils, and also by Madeira and Ribeiro (1995) in a lysimeter experiment with needle litter applied on a sandstone substrate. However, our results did not agree with those by Olsson et al. (1996a) and Mendham et al. (2003), for sandy soils, as soil C and N status were not affected by harvest residue management. Such a difference may be related to the low amounts of C in the soils of the present study.

The small response to residue management practices observed for nutrient losses, through leaching, suggests that nutrients released from residues should have been accumulated in the soil. As organic residues contained substantial amounts of nutrients, their presence also improved the pool of ex-

tractable base cations in the topsoil layer (0–10 cm), following the trend observed by Olsson et al. (1996b) in podsolized soils of pine stands, and by Johnson and Todd (1998) in a temperate mixed oak forest. However, Ca and Mg contents decreased in the 10–30 cm soil layer in all treatments (especially in the absence of organic residues), which may be explained through transfer to deeper soil layers; this transfer was more pronounced by residue incorporation into the soil. Although a net Ca and Mg loss from the system was not observed, their allocation in soil deeper layers could negatively affect the availability for young trees' roots. Moreover, the incorporation of forest floor plus harvest residues also favored the transference of K and P to a soil depth beyond 30 cm.

During the first year of study, legume cover reduced N, Ca, Mg and K losses, which may be related to nutrient uptake by legumes, and to the decrease in water drainage, which was the lowest in the respective treatments. As these differences were not observed at the end of the study period, the results suggest that legume cover does not significantly affect nutrient losses in the long term. Treatment with legumes led to an improvement in N status in seedlings and needles, as reported by Nambiar and Nethercott (1987) in radiate pine plantations on sandy soils with low fertility. However, soil N fertility was not affected by legume cover, as N content in the topsoil layer was only enhanced when it was combined with harvest residues. This may be ascribed to the low N content in the legume plants used, which was not sufficient to counteract the effect of organic residues on N immobilization.

Despite the potential for legumes to compete with seedlings for site resources, seedling growth was not affected by their use, which supports data reported on the effect of legume cover cropping during the early growth of pine (Simpson et al., 2004) and eucalyptus plantations (Mendham et al., 2004). Although, in the short term, the growth of pine seedlings and soil N fertility were not influenced by this practice, the benefits observed on N loss reduction through leaching during early stages of plantation development and on N availability improvement may contribute to enhancing forest plantation productivity in the next rotations.

5. CONCLUSIONS

The maintenance of forest floor litter and harvest residues during the inter-rotation period may contribute, in the short term, to reducing nutrient losses from leaching and to improving soil nutrient status in the first years of pine plantations. In addition, seedling growth was dependent on the placement method, as it was improved by the maintenance of residues on the soil surface, but not by their incorporation into the soil, with or without legumes. Although seedling growth improvement was not associated with its nutrient status, positive effects observed on soil fertility through residue maintenance and legumes may contribute to long-term nutrient supply to pine plantations and, thus, to ensuring sustained productivity. However, appropriate field studies are needed to evaluate these effects over a longer period.

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