

RESEARCH ARTICLE

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Combining innovative mulches and soil conditioners in mountain afforestation with ash (*Fraxinus excelsior* L.) in the Pyrenees (NE Spain)

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Abstract

Aim of study: To assess the effectiveness for improving early seedling performance of the individual and combined application of (i) various doses of an innovative soil conditioner including polyacrylamide-free super-absorbent polymers, fertilizers, root precursors and humic acids; and (ii) innovative mulches based on renewable-biodegradable or recycled raw materials. The assessment was carried out in comparison with reference (commercial) soil conditioners and mulches.

Area of study: Upper montane afforestation site located at 1,430 m altitude in the southern Pyrenees (NE Spain).

Material and methods: We studied the effect of 15 treatments (various combinations of soil conditioners and mulches) on mountain ash (*Fraxinus excelsior* L.), testing survival, diameter and height growth and water and nutrient status during two growing seasons (2014-2015). We also assessed mulch durability during 2014-2016.

Main results: The innovative soil conditioner improved diameter and height seedling growth (92% and 72% respectively) and water and nutrient status. The 40 g/seedling dosage was more cost-effective than the 20 and 80 g/seedling dosas. The new formulation performed better in general than the commercial formulation. Mulches led to slight gains compared to control seedlings, and there were no major differences between the mulch models. The combined application of soil conditioners and mulches was not of particular interest.

Research highlights: Soil conditioners consisting of synergic mixtures of water super-absorbent polymers, fertilizers, root growth precursors and humic acids can improve early seedling performance in coarse-textured, stony soils in montane conditions. Small mulches may be only of limited interest as long as weed competitiveness is poor.

Additional keywords: ecotechnology; groundcovers; reforestation; seedling performance; restoration; water super-absorbent polymer; weed.

Abbreviations used: GSn, growing season number 'n'; leaf water potential, LWP; water super-absorbent polymers, SAP; soil conditioner with water super-absorbent polymers, SCwSAP; Soil Plant Analysis Development, SPAD.

Authors' contributions: Conceived and designed the experiments: JC, PR and MP; data acquisition and management: CF and JC; critical revision of the manuscript for important intellectual content: JC, PR, AA and MP; coordinating the research project: MP.

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Supplementary material (Tables S1 and S2) accompany the paper on FS's website.

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Introduction

The history of land use in the Pyrenees, as in other mountain areas of south Europe, spreads over millennia. In the last century the modernization of farming in lowlands resulted in the abandonment of many traditional agricultural and grassland practices (MacDonald *et al.*, 2000), particularly in the difficult to access, small-sized fields predominant in mountain areas. Consequently, forest has expanded significantly, especially pine forests (Ameztegui *et al.*, 2010). This has homogenized the landscape both in terms of

composition and structure. Afforestation with broadleaf trees helps to decrease the negative consequences of excessive landscape homogenization, such as an increase in fire risk (Palmero-Iniesta *et al.*, 2017) and negative effects on biodiversity (Ameztegui *et al.*, 2018), among others.

In the Pyrenees, European ash (Fraxinus excelsior L.) is a native broadleaf tree with many uses, including timber for tool-making and turnery, fodder (green branches and litter), charcoal and fuelwood (Marie-Pierre et al., 2006; Mottet et al., 2007). Despite these uses, many ash forests were cut down and turned into grasslands in the past (Vigo et al., 2005). However, more recently, its ecological plasticity together with its fast growth and valuable timber has turned ash into a good candidate for afforestation programmes in Europe (Fraxigen, 2005; Weber-Blaschke et al., 2008). The main limiting factor for ash in the southern Pyrenees is its sensitivity to water shortage (Gonin et al., 2013), which is particularly critical during the first years when the root system is not yet well developed (Vallejo et al., 2012). South European montane conditions are characterized by a dry season that coincides with the highest temperatures, and by shallow, coarse-textured soils with high stoniness and steep slopes, resulting in a poor water retention capacity. Soil water content can be increased by support irrigations, which are expensive and often inapplicable in these conditions (Carminati et al., 2010). An alternative option is to mix water superabsorbent polymers (SAPs or hydrogels) with the soil of the planting pit. These polymers can absorb and store up to 400 times their weight in water (Bouranis et al., 1995), which is then available to the plants over an extended time period (Hüttermann et al., 2009). SAPs can increase soil water content by reducing evaporation and percolation losses, which is of particular interest in soils with a poor water retention capacity, i.e. coarsetextured soils (Bhardwaj et al., 2007; Koupai et al., 2008; Del Campo et al., 2011). SAPs have been reported to alleviate soil and plant water potential, and increase plant water use efficiency and the time to reach permanent wilting point, ultimately enhancing plant survival and growth (Sivapalan, 2001; Hüttermann et al., 2009; Del Campo et al., 2011). SAPs are commercialized alone or as synergistic mixtures with other ingredients, especially fertilizers and various organic compounds, with the intention of improving both the physical and chemical soil properties and not only the water-related parameters. Moreover, SCwSAPs aim to prevent some of the limitations of pure SAPs, such as the reduction of NO₃ and NH₄ availability and the risk of being washed away (Rowe et al., 2005). These mixtures are regarded as soil conditioners with SAP (SCwSAP), and have been reported to enhance soil nutrient levels and

seedling performance (Machado *et al.*, 2016; Coello *et al.*, 2018). However, the application of SAPs and SCwSAPs at afforestation sites has obtained contrasting results depending on their composition, application method, dosage and soil features (Navarro *et al.*, 2005; Del Campo *et al.*, 2011; Coello *et al.*, 2018).

Despite their interest, there are some constraints involved in using SAP or SCwSAP, as most formulations are based on cross-linked polyacrylamide, which causes social concern due to the residual presence of non-polymerized acrylamide. Although the concentrations are within the legal limits (Rowe et al., 2005) and acrylamide has a short half-life in the soil (Lande et al., 1979), SAP manufacturers are currently developing polyacrylamide-free versions that still need to be tested in the field and compared to current commercial formulations. Moreover, a limiting factor of SAPs is that the improvement in conditions at micro-site level is positive for the seedling but also for the competing vegetation, which is a major threat to young afforestations (Willoughby et al., 2009). Extant vegetation can outcompete newly established seedlings in the struggle for water, light and nutrients (Navarro-Cerrillo et al., 2005). The most widespread weeding techniques are to apply herbicide, which is costeffective but is of growing social concern (Willoughby et al., 2009), and mechanical weeding, which is expensive and can only be applied at sites that can be accessed easily.

An alternative option is to use mulches, also known as groundcovers or weeding mats. A mulch is an opaque layer covering the soil around the seedling, impeding weeds from germinating in its vicinity (Maggard et al., 2012). Previous works have demonstrated that mulching has positive effects on seedling survival and growth (Van Sambeek, 2010; Maggard et al., 2012, Coello et al., 2017), increases soil water content because it reduces evaporation and transpiration by weeds (Hueso-González et al., 2015), improves nutrient uptake (Van Sambeek & Garrett, 2004) and buffers soil temperatures (Cregg et al., 2009; Coello et al., 2017). Although plastic mulches are the most widespread (Arentoft et al., 2013), several new environmentally friendly mulch materials are being developed, including biofilms, i.e. plastic-like materials made from renewable sources (Kapanen et al., 2008). Other similar materials include composites made from paper residues (Shogren & Rousseau, 2005) and jute tissues (Debnath, 2014).

Despite the interest in these new materials and techniques for afforestation, the combined application of soil conditioners and groundcovers has rarely been tested in field conditions (Navarro *et al.*, 2005; Coello *et al.*, 2018). This study aims to assess the effectiveness on

early seedling performance (survival, growth and water and nutrient status) of the individual and combined application of (a) new mulches based on renewable or recycled raw materials, either biodegradable or reusable; and (b) polyacrylamide-free SCwSAP, in south European montane conditions. We also studied mulch durability. We compared the performance of these new techniques with commercially available mulches and SCwSAP.

Our working hypotheses were: i) both the use of mulches and SCwSAP will have a positive effect on all seedling parameters compared to their respective controls, while the combined use of the two techniques will lead to a synergistic increase in performance; (ii) the performance of SCwSAP will be proportional to the application dosage; and (iii) the commercial (including polyacrylamide) and the new SCwSAP (polyacrylamide-free) will perform similarly when applied at the same dosage.

Materials and Methods

Description of the study area

The experiment was conducted in Fontanals de Cerdanya, in the Pyrenean mountains of Catalonia, NE Spain (42°23'9.11N; 1°55'53.90E). The plot is located at a mean altitude of 1,430 m a.s.l, on a north aspect with an average slope inclination of 30%. The mean annual temperature is 7.5°C while the mean annual precipitation

is 887 mm, 272 mm of which occur in summer. According to the Köppen classification, the climate is between Cfc (Temperate) and Dfb (Continental). The soil has a loamy-sandy texture (22% clay, 21% silt, 57% sand), the pH is 5.2 and organic matter content is 2.56%. Most soils in this area are Humic Dystrudepts (USDA, 1999) with a mesic temperature regime (Poch & Boixadera, 2008).

This plot was used for cattle grazing until it was abandoned in 2013. The main woody vegetation species, with a very low density, are the trees ash (*Fraxinus excelsior*) and wild pear (*Pyrus communis* L.), and the shrubs *Rosa canina* L. and *Rubus idaeus* L.

The annual summer precipitation was monitored continually with a weather station (Davis Instruments, USA). The summer of the first growing season (2014, GS1 hereinafter) was rather wet (404 mm) compared to the historical reference (272 mm, Ninyerola *et al.*, 2005), while the summer of the second growing season (2015, GS2 hereinafter) was drier (220 mm).

Experimental design and treatments

We planted 450 seedlings following a randomized incomplete block design with six blocks. In each block we planted 75 seedlings that were randomly assigned, in groups of 5 seedlings, to the 15 experimental treatments or combinations of various mulching and soil conditioning techniques. In total each treatment was applied to 30 seedlings. Table 1 shows the details of the techniques applied (mulch and soil conditioner).

Table 1. Description of the experimental techniques.

Technique type	Technique code	Description			
Mulch	Control	No mulch applied.			
	Com_Plastic	Commercial black polyethylene film, anti-UV treated, 80 µm thick.			
	Com_Biofilm	Ökolys®, a commercial green biodegradable woven mat.			
	New_Biofilm	Prototype of black biodegradable frame (biopolymer), fused to a black commercial biodegradable film based on PHA (polyhydroxyalkanoate), $80~\mu m$ thick, manufactured by Groencreatie and DTC. The frame is to make installation easier.			
	New_Jute	Prototype of biodegradable woven jute mat treated with furan bio-based resin for increased durability, manufactured by La Zeloise NV.			
	New_Rubber	Prototype of black layer made of recycled rubber, anti-UV treated, 1.5 mm thick to make fixation unnecessary, manufactured by EcoRub BVBA.			
Soil conditioner	SC-	No soil conditioner applied.			
	New_SC20; New_ SC40; New_SC80	TerraCottem Arbor®, at the prototype stage when tested. Product developed for tree an shrub planting. Its formulation includes a new generation of polyacrylamide-free water super-absorbent polymers (36.25% of total weight), fertilizers (14.5%; NPK 3-1-7), hu acids (0.75%), growth precursors (0.25%) and volcanic rock (48.25%). The numbers 20 and 80 indicate the dosage (g/seedling).			
	Com_SC40	TerraCottem Universal®, a commercially available soil conditioner with cross-linked polyacrylamide and polyacrylic acid polymers (39.50%), fertilizers (10.50%; NPK 9-2-11), growth precursors (0.25%) and volcanic rock (49.75%). The dosage was 40 g/seedling.			

The 15 treatments were organized into three sub-experiments:

- (i) Sub-experiment 1: a full factorial design combining the 6 different mulch treatments with a new SCwSAP applied at a dose of 40 g/seedling (New_SC40) as well as without it (SC-). Overall there were 12 treatments with 30 seedlings per treatment, thus 360 seedlings in total. The soil conditioner dose of 40 g/seedling corresponds to the manufacturer's recommendation for the most similar commercial product available.
- (ii) Sub-experiment 2: a study of the effect of four different doses of New_SC (0, 20, 40 and 80 g/seedling), combined with a reference mulch (Com_Plastic) in all cases. Each treatment comprised 30 seedlings, with a total of 120 seedlings in this sub-experiment.
- (iii) Sub-experiment 3: a study comparing a commercial and a new SCwSAP (Com_SC vs. New_SC), both applied at a dosage of 40 g/seedling and combined with a reference mulch (Com_Plastic). Each treatment comprised 30 seedlings, with a total of 60 seedlings.

The combination Com_Plastic x New_SC40 was present in all three sub-experiments, while the combination Com_Plastic x SC- was present in sub-experiments 1 and 2.

Field trial establishment

We planted the seedlings in late March 2014, during vegetative dormancy. Soil preparation consisted in mechanical soil digging (40 x 40 x 40 cm) with a backhoe spider excavator, which was used to make micro-basins for runoff collection. The plantation frame was 3 x 3 m, for a density of 1,100 seedlings · ha-1. The tree species chosen was mountain ash (Fraxinus excelsior L.) from the local provenance Central Pyrenees. The seedlings were one-year old and provided in 300 cm³ containers. They were 15-20 cm high and met the general seedling quality criteria (Cortina et al., 2006). We applied the soil conditioner right before planting following the manufacturer's indications: we dug a sub-pit sized 30 x 30 x 30 cm, put half of the dosage at the bottom of the pit and mixed the other half with the soil used for filling up the pit when the seedlings were planted. We installed the mulches manually right after planting. We chose a small mulch size (40 x 40 cm, similar to the area of the planting pits), to limit costs and because we predicted that there would be low to intermediate weed proliferation during the first years. To protect the afforestation from browsing damage by roe deer (Capreolus capreolus L.) we installed a 1.3 m high perimeter fence consisting of four lines of barbed wire.

Assessment of seedling survival and growth

We assessed all seedlings at the time of planting (March 2014) and at the end of the first two growing seasons (October 2014 and 2015) to determine their survivorship, diameter and height. We conducted an additional visual assessment of survival eight weeks after the seedlings had been planted to remove any seedlings from the study that had died soon after planting as a result of poor seedling quality or careless planting (two seedlings in total). We measured seedling diameter at a painted, constant point, 4-5 cm above the ground, with a precision of 0.1 mm using a digital calliper. Seedling height was measured from the ground up to the highest bud, with a cm precision, using a measuring tape. Annual growth rates of alive seedlings were calculated as the difference between the measurement at the end of each growing season and the previous measurement.

Assessment of physiological traits

We measured two traits related to seedling performance: midday leaf water potential (LWP, hereinafter), which is a proxy of seedling water status; and leaf SPAD (Soil Plant Analysis Development), which is a proxy of seedling nutrient status (Djumaeva et al., 2012). We measured LWP using a pressure chamber (Solfranc Technologies, Spain) with N₂ as the pressure gas. We measured the pressure (bars) at which the water within the leaf was ejected through the petiole. In both 2014 and 2015 we conducted 4 fortnightly measurements in July and August. On each of these 8 dates we sampled one leaf from one randomly chosen seedling per treatment and block (n = 6; 90 measures in total). These measurements were taken between 10:00 and 14:00 solar time, i.e., at the highest sun angle. We used a Minolta SPAD-502 instrument (Minolta Camera Co, Japan) to measure SPAD. The LWP and SPAD were measured on the same dates, except for the two August measurements in GS1 when SPAD could not be measured. In each SPAD sampling we measured one randomly chosen alive seedling per treatment and block (n = 6; 72 measurements in total). We calculated the average SPAD value of three leaves from each sampled seedling. Both LWP and SPAD values were obtained from sun-exposed, fully elongated and healthy leaves located in the upper third of the sampled seedling.

Mulch durability

The physical integrity of mulches was assessed visually in October 2016 after 30 months in the field. The mulches were assigned to a damage category

depending on the proportion of their surface that was damaged, either physically (torn) or due to weeds growing through and on the mulch layer: (i) intact (no damage); (ii) slight damage (1-25% damage); (iii) intermediate damage (26-50%); severe damage (>50%).

Statistical analyses

We analysed the data independently for each subexperiment. We considered treatment as a fixed factor and block as a random factor. The LWP and SPAD data collected on different measuring dates were combined to build a more robust dataset.

Seedling survival and mulch durability were analysed with descriptive statistics. Seedling annual diameter and height growth, LWP and SPAD were assessed with an ANOVA, which was two-way in sub-experiment 1 (factors: mulch, soil conditioner, and their interaction) and one-way in sub-experiments 2 and 3. We used a significance level of 0.05 and assessed pairwise differences between treatments with the post-hoc Duncan's multiple range test. When necessary (seedling growth and LWP), values were log or root transformed to meet the ANOVA assumptions of normality and homoscedasticity. Tables and figures show untransformed data. The ANOVAs were run with SPSS v19.0 software.

Results

Seedling survival

The overall survival rate at the end of the first growing season (GS1) was 99%, dropping to 93% at the end of the second growing season (GS2). In sub-experiment 1 the lowest survival rate after two growing seasons corresponded to New Rubber (82%) and the Control (90%), while the rest of the mulches showed survival rates above 93%. For the soil conditioner, New SC40 showed a similar survival rate at the end of GS2 as SC- (93% and 90%, respectively). In sub-experiment 2 the treatments leading to the lowest survival rates after two growing seasons were SC- (90%) and New SC80 (93%), while New SC20 and New SC40 resulted in 97% and 100%, respectively. In sub-experiment 3, both Com SC40 and New SC40 led to high survival rates at the end of GS2 (97% and 100%, respectively). Table S1 [suppl.] provides the annual survival of all treatments and sub-experiments.

Seedling growth

Table 2 shows the outcomes of the ANOVAs conducted in each sub-experiment. In sub-experiment 1 the mulches did not significantly affect annual diameter growth in GS1 or GS2; however, for height growth

Table 2. Summary of outcomes of the ANOVAs of annual diameter (DG) and height (HG) growth during the first (GS1, 2014) and the second (GS2, 2015) growing seasons; seedling water status (midday Leaf Water Potential, LWP) and seedling nutrient status (Soil Plant Analysis Development, SPAD).

	DG GS1	DG GS2	HG GS1	HG GS2	LWP	SPAD
Sub-experime	ent 1					
			Mulch (dF=5)			
F	1.02	1.13	2.07	2.92	1.25	1.46
p-value	0.409	0.344	0.072	0.020	0.284	0.203
		Soil	conditioner (dF=1))		
F	201.41	8.88	30.592	1.29	6.10	4.49
p-value	< 0.001	0.003	< 0.001	0.261	0.014	0.035
		Interaction m	ulch * soil condition	ner (dF=5)		
F	2.04	1.78	0.247	1.12	1.66	2.51
p-value	0.072	0.117	0.941	0.361	0.142	0.030
Sub-experime	ent 2					
		Soil co	onditioner dose (dF=	=3)		
F	28.67	4.79	7.73	1.76	0.06	3.71
p-value	< 0.001	0.004	< 0.001	0.185	0.979	0.013
Sub-experime	ent 3					
		Soil condi	tioner formulation ((dF=1)		
F	6.85	8.63	3.02	0.07	0.85	8.05
p-value	0.011	0.005	0.092	0.799	0.360	0.006

New_Biofilm induced significantly higher growth rates than the Control in GS2 (Figure 1A). The use of soil conditioner resulted in a higher diameter and height growth than in the Control, although differences were greater for GS1 than for GS2, where differences between treatments were significant for diameter growth but not for height growth (Figure 1B). No significant effect of the interaction between mulch and soil conditioner on diameter growth or height growth was found (Table 2).

In sub-experiment 2 we observed that the diameter growth during GS1 was larger with increasing doses of soil conditioner, although the two highest doses (40 and 80 g/seedling) were not significantly different (Table 2). However, although the addition of soil conditioner also increased diameter growth during GS2, we did not observe significant differences between the tested doses (Figure 2A). In the case of height growth the pattern was unclear in GS1, with New_SC40 leading to higher values than New_SC20, which in turn was not significantly different to New_SC80. In GS2 there were no significant differences between treatments for height growth.

In sub-experiment 3 we observed significantly higher diameter growth when New_SC40 was used compared to Com_SC40 in both GS1 and GS2, while height growth rates were not significantly different (Figure 2B). Table S2 [suppl.] provides the annual growth values for each treatment.

Physiological traits

In sub-experiment 1 neither mulch nor mulch x soil conditioner interaction had a significant effect on LWP (Table 2). However, the addition of soil conditioner (New_SC40) significantly improved seedling water status compared to SC-. In contrast, we observed that SC dosage (sub-experiment 2) and SC formulation (sub-experiment 3) had no significant effect on LWP.

SPAD values were not affected by the different mulches. However, the use of New_SC40 resulted in a higher SPAD than SC-. Mulch x soil conditioner interaction was also significant. Exploring this interaction further we found that, in the presence of soil conditioner, all mulches enhanced the SPAD value compared to the Control, while in the absence of soil conditioner no mulch had any effect on SPAD.

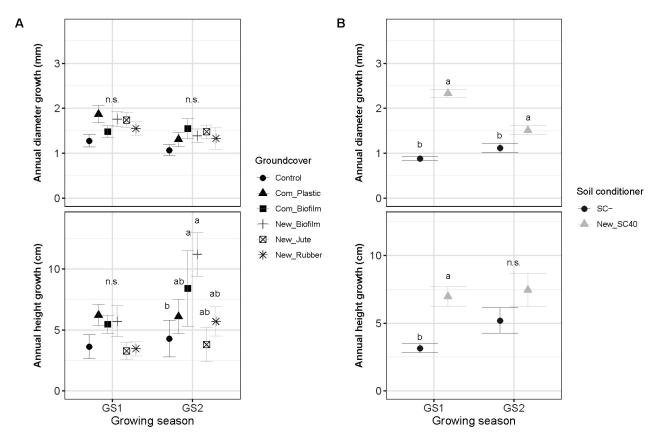


Figure 1. Annual diameter and height growth during the first two growing seasons (GS1-GS2) for sub-experiment 1. A: mulch treatments; B: presence of soil conditioner. For each variable and year, significant differences (p < 0.05) between treatments are indicated by different letters (Duncan test grouping), while "n.s." indicates not significant. Whiskers indicate standard error of the mean.

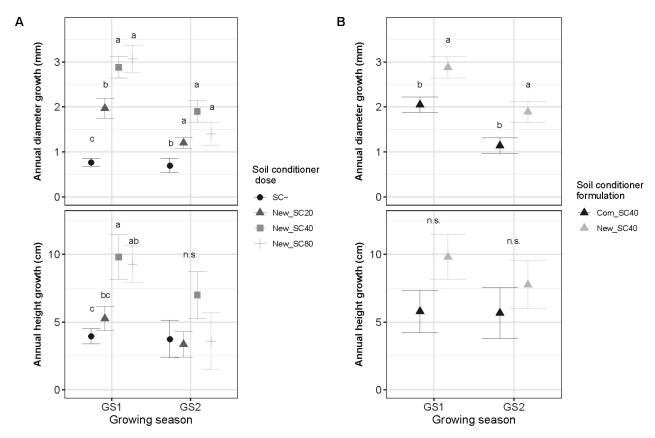


Figure 2. Annual diameter and height growth during the first two growing seasons (GS1-GS2) for (A) sub-experiment 2, and (B) sub-experiment 3. For each variable and year, significant differences (p < 0.05) between treatments are indicated by different letters (Duncan test grouping), while "n.s." indicates not significant. Whiskers indicate standard error of the mean.

Increasing the dose of New_SC (sub-experiment 2) to 40 or 80 g/seedling also led to enhanced SPAD compared to SC-. In sub-experiment 3 the new soil conditioner also resulted in significantly higher SPAD than the commercial one.

Table 3 shows the LWP and SPAD values of each treatment.

Mulch durability

The different mulches showed contrasting levels of damage after three growing seasons (Figure 3). The models with the highest integrity were New_Rubber (97% of units intact or with only slight damage) and Com_Plastic (91%). However, the biodegradable mulches had 20%, 8% and 14% (Com_Biofilm, New_Biofilm and New_Jute respectively) of the units severely damaged, while about half of the units were intact.

Discussion

Our study showed the importance that planting techniques can have on improving the early performance of broadleaf seedlings in south European mountain afforestation. The hypotheses of sub-experiment 1 were only partially corroborated: the addition of soil conditioner had a positive effect on all seedling performance parameters, but mulching and the interaction between mulch and soil conditioner (which was additive rather than synergistic) led to benefits below our expectations. In sub-experiment 2, the hypothesis was corroborated because the performance of the soil conditioner was better when the dose was increased from 20 to 40 g/seedling; however, there was a saturating effect at the 80 g/seedling dosage. Finally, the hypothesis of sub-experiment 3 was only partially corroborated, as we found similar height growth and seedling water statuses for the new soil conditioner and the commercial formulation, as initially foreseen; however, the new soil conditioner resulted in unexpected gains in diameter growth and seedling nutrient status.

Nevertheless, our results should be verified with further experiments involving a wider variety of tree species, a longer time span, and whenever possible a thorough study of the changes in the soil water status

Table 3. Midday Leaf Water Potential (LWP) and SPAD of the measurements taken in the summers of 2014 and 2015. Values are expressed as mean \pm standard error. Significant differences (p < 0.05) between treatments are indicated by different letters, according to Duncan's posthoc test grouping.

	LWP (bar)	SPAD					
Sub-experiment 1 (Mulch and soil conditioner)							
Mulch							
Control	-22.0 ± 0.3 a	$29.1 \pm 0.7 \; a$					
Com_Plastic	$-21.8 \pm 0.5 \text{ a}$	$30.5\pm0.7~a$					
Com_Biofilm	-21.2 ± 0.4 a	$31.1\pm0.7~a$					
New_Biofilm	$-22.2 \pm 0.4 a$	$29.6 \pm 0.7 \; a$					
New_Jute	$-21.5 \pm 0.5 \text{ a}$	$31.1\pm0.7\;a$					
New_Rubber	-21.0 ± 0.4 a	$30.7 \pm 0.7 \; a$					
Soil conditioner							
SC-	$-22.1 \pm 0.3 \text{ b}$	$29.8 \pm 0.4 \; b$					
New_SC40	-21.2 ± 0.3 a	$30.9 \pm 0.4 \; a$					
Sub-experiment 2 (soil conditioner dose)							
SC-	-21.9 ± 0.7 a	$28.8 \pm 0.9 \text{ b}$					
New_SC20	$-22.0 \pm 0.5 \text{ a}$	$30.7 \pm 0.9 \; ab$					
New_SC40	-21.7 ± 0.6 a	$32.3 \pm 0.8 \; a$					
New_SC80	$-21.8 \pm 0.5 \text{ a}$	$33.1\pm1.2\;a$					
Sub-experiment 3 (soil conditioner formulation)							
Com_SC40	$-20.9 \pm 0.5 \text{ a}$	$28.9 \pm 0.8 \ b$					
New_SC40	-21.7 ± 0.6 a	$32.3\pm0.8\;a$					

throughout the seasonal cycle, and how it is affected by the presence of the mulches and/or soil conditioners. Our results may also have been affected by the exceptionally wet summer (50% higher than the reference value) during the first growing season, which could have contributed to the very high survival rate (99%). If the first summer had been drier, the effect of the soil conditioners and especially the mulches may have been

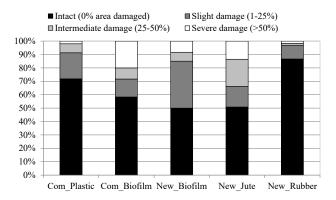


Figure 3. Mulch durability: proportion of the different mulch models in each level of physical damage, after 30 months in the field.

more evident because the seedlings would have been exposed to a more intense water stress that would have been alleviated by the two planting techniques.

Overall mulch performance

In general, mulching had no significant effect on seedling performance with the only exception of New Biofilm, which resulted in larger height growth compared to the Control in the second growing season. Although the other variables did not result in significant differences between mulches, the Control was the treatment with the lowest diameter growth and SPAD, and the second lowest height growth and LWP. This seems to imply that mulching has a slightly positive effect. Based on over 110 reports, Van Sambeek (2010) analysed the relative productivity response of different weeding techniques on various tree species, finding a mulch:control response ratio for *Fraxinus* spp. of 222, meaning that mulched seedlings yielded on average 122% more than those without mulch. In our study, however, the mean mulch:control response ratio was 132 for diameter growth and 148 for height growth. Dostálek et al. (2007) also found a negligible height growth response in Fraxinus excelsior subject to 40 cm wide textile mulching compared to the control, after 5 growing seasons. However, the same authors found a positive effect of fresh bark mulch with similar dimensions as it increased (2-fold) the overall height growth rate compared to the control. The mulches tested here were similar to those evaluated in a previous study (Coello et al., 2018), in which we found no significant differences between them with regard to seedling water status. In contrast, Paris et al. (1998) found that 2 m wide plastic mulching had a positive effect on walnut LWP. However, similarly to our study, Cregg et al. (2009) and Coello et al. (2017) observed that mulching had no effect on seedling early nutrient status. These results question the interest of mulching in the study conditions. That the mulch performance was below the initial expectations could be due to three factors:

- (i) the competitiveness of spontaneous vegetation during the first two growing seasons was poor because (a) the soil preparation left 40 x 40 cm of bare soil prior to planting, and (b) the site quality was low due to low temperatures and a stony, coarse-textured soil. Indeed, most of the unmulched planting pits did not show relevant proliferation of extant vegetation, and therefore the expected gains induced by the weeding effect of mulching could have been masked.
- (ii) the mulches in our study were rather small (40 x 40 cm) compared to most previous works. Previous studies with small mulches (60 x 60 cm or less) suggest that there are lower gains in seedling

performance (Navarro *et al.*, 2005; Dostálek *et al.*, 2007; Valdecantos *et al.*, 2009, 2014; Coello *et al.*, 2018) compared to the more evident benefits of mulches sized 80 x 80 cm or larger (Jiménez *et al.*, 2014, 2016; Vitone *et al.*, 2016; Coello *et al.*, 2017). Davies (1988) found that the mulches with sides sized 120 cm or larger were more effective than those with sides sized 60 cm or less.

(iii) the summer rainfall during the first growing season was 50% higher than the reference value, which may have masked the positive effect of mulching on soil water retention (Barajas-Guzmán & Barradas 2011, McConkey *et al.*, 2012).

Mulch models

There were only minor differences between the mulch models, in line with Maggard et al. (2012) and Coello et al. (2017, 2018). Only New_Rubber resulted in a much lower survival rate (82%) than the other mulches (95% in average). Overall, and despite the general lack of significant differences between them, the best performance could be attributed to New_Biofilm, which ranked first in overall seedling growth, and to Com_Biofilm, which ranked second in both LWP and SPAD. The sound performance of these models, together with their degradability and renewable origin, make them, along with New_Jute, suitable alternatives to plastic mulching with added benefits from the technical (do not need to be removed) and environmental points of view.

The durability of the mulches was assessed after 30 months, a time span long enough to provide an initial idea of their service life. The plastic mulch in the study performed in line with the literature: Haywood (2000) and Coello et al. (2017) found respectively 70% and 66% of plastic mulches with 20% or less damaged area after five years, similarly in our case 91% of units had only 20% of damage or less. The biodegradable mulches also performed in line with previous studies: a biofilm similar to New Biofilm tested by Coello et al. (2017) showed 33% of units severely damaged after 40 months of study, while in our case the values ranged between 8% (New Biofilm) and 20% (Com Biofilm). We can therefore initially conclude that the biodegradable mulches are expected to meet the desirable service life of 4-5 years during which more than half of the units should remain intact or be only slightly damaged (Coello & Piqué, 2016).

Overall performance of soil conditioners

The use of SCwSAPs was positive for early seedling performance, with an overall improvement in all assessed parameters, in line with a previous study with the same products evaluated at an afforestation site with *Pinus halepensis* in a coarse-textured soil in semiarid conditions (Coello *et al.*, 2018). However, these results contrast with most previous afforestation studies using SCwSAPs, which found negligible or even negative effects when the SCwSAPs were applied inadequately (1.5 years after planting – Bulíř, 2006; superficial application – Hueso-González *et al.*, 2016), in a low or inaccurate dosage (15 g/seedling – Navarro *et al.*, 2005; unknown dose – Frigola & Nadal, 2013) or in fine-textured, clayish soils (Bulíř, 2005; Navarro *et al.*, 2005; Del Campo *et al.*, 2011; Frigola & Nadal, 2013).

After two growing seasons the average survival for seedlings including any type or dose of soil conditioner (New_SC20, New_SC40, New_SC80 and Com_SC40) was 95%, slightly higher than the 90% of SC- seedlings. Hüttermann *et al.* (2009) also reported a positive effect of soil conditioners on the survivorship of various species and Chirino *et al.* (2011) found a positive effect for *Quercus ilex* L.

With regard to annual diameter and height growth, New_SC40 seedlings grew 92% and 72% more than those of the control treatment, similarly to our previous study (Coello *et al.*, 2018). The growth gains induced by pure SAP (not including fertilizers and other components present in the SCwSAPs tested) in previous afforestation studies with broadleaved species are much less evident than in the present study (Rowe *et al.*, 2005; with *Alnus cordata* (Loisel.) Dub., *Alnus glutinosa* (L.) Gaertn and *Salix x reichardtii*) or even negligible (Clemente *et al.*, 2004 with *Ceratonia siliqua* L., *Olea europaea* L. and *Pistacia lentiscus* L.; Chirino *et al.*, 2011 with *Quercus suber* L.).

The positive effect of SCwSAP or pure SAP on seedling water status has been reported in nursery conditions for other broadleaved species: Citrus sp (Arbona et al., 2005); Populus euphratica Oliv. (Luo et al., 2009); Quercus suber (Chirino et al., 2011) and Fagus sylvatica L. (Beniwal et al., 2011; Jamnicka et al., 2013). The improved water status induced by the SCwSAP suggests that the seedlings use the water stored by the polymer (Del Campo et al., 2011; Jamnicka et al., 2013). The improvement in nutrient status induced by New SC40 was also described by Machado et al. (2016), who reported an increase in the level of macro and micronutrients in soils when a SCwSAP was used. In contrast, the use of pure SAP by Clemente et al. (2004) did not increase seedling chlorophyll or nitrogen content. This suggests that some of the ingredients of New SC other than SAP (probably the fertilizers, humic acids, and/or the root growth precursors) help improve tree nutrient status, which is a relevant advantage in soils with a poor nutrient retention capacity, i.e. coarse textured and/or stony soils.

The apparent reduction over time in the soil conditioner effect, which was detected in this study in the second growing season, was also reported by Chirino *et al.* (2011), Oliveira *et al.* (2011) and Coello *et al.* (2018), and could be because (i) the nutrients added via the soil conditioner are progressively exhausted, and/or (ii) the seedling develops new roots out of the planting pit, thus accessing unconditioned soil volume.

Soil conditioner dosage and formulation

Increasing soil conditioner dosage (sub-experiment 2) was generally positive, in line with Al-Humaid & Moftah (2007) and Hüttermann et al. (2009). The most cost-effective dose was 40 g/seedling, coinciding with the manufacturer's recommendation for 30 x 30 x 30 cm soil mixing volume. This finding is also in line with our previous study (Coello et al., 2018), in which we found that the 20 g/seedling dose led to better results than the control less often than the 40 g/seedling dose, which provided outcomes that were similar to the highest dose (80 g/seedling) for all variables. Future costeffectiveness studies considering various dosages, as well as the application of this product at an operational scale, could be used to fine tune the viability and the most recommendable dosage for different site conditions.

In sub-experiment 3 New SC40 led to better results than Com SC40 for diameter growth (both growing seasons) and SPAD. These findings contrast with our previous study in semiarid conditions (Coello et al., 2018), in which we generally found similar results for both products. This suggests that the ingredients contained in the new formulation and not in the commercial one (humic acids and the type of root growth precursors and polymers) may be particularly beneficial in cold, montane conditions or for nutrient-demanding species such as Fraxinus excelsior. Another strength of the new formulation is that it is expected to have higher social acceptability than most commercially available SAPs or SCwSAPs that contain polyacrylamide (DRI, 2008), like the commercial model in our study. Future experiments with these products will make it possible to ascertain the site conditions and species for which each formulation is particularly efficient.

Interactions between mulch and soil conditioner

As mulching had predominantly no or only minor effects, applying this technique combined with the soil conditioner seems to be of little interest in these conditions. The interaction between the two techniques was only significant in the case of SPAD, where the soil conditioner combined with any mulch type resulted in

better seedling nutrient status than when applied alone (unmulched seedlings), while this did not occur in the absence of soil conditioner. This suggests that, for this particular variable (SPAD), the positive effects of mulching may be enhanced when fertilizers are added to the plantation pit, in line with Vincent & Davies (2003).

Implications for management

The soil conditioner with water super-absorbent polymers in a synergistic mixture with other ingredients (fertilizers, humic acids and root growth precursors) is a cheap and easily applied technique that does not require maintenance, and which had a positive effect on all seedling performance indicators in our study. The tested polyacrylamide-free prototype increased both seedling water status, probably due to the polymer; and nutrient status, probably due to the fertilizers and humic acids. This technique ultimately increased seedling survival and growth. The most cost-effective soil conditioner dose was the one recommended by the manufacturer, 40 g/seedling, which often led to better results than the lower dose (20 g/seedling) but never worse than the higher one (80 g/seedling). The new prototype tested seems a suitable alternative to the commercially available version, with technical and social advantages: higher growth rates and better seedling nutrient status, while also being polyacrylamide-free.

On the other hand, the use of small mulches (40 x 40 cm) had a slightly positive effect compared to untreated seedlings, which was usually not statistically significant. Therefore, this technique does not seem a first priority option for enhancing the early establishment of seedlings at poor-quality, montane afforestation sites that have low competitiveness of extant vegetation. Among the different mulch models, the biodegradable ones (prototypes based on biofilm and woven jute, as well as the commercial biofilm) performed similarly to the plastic mulch, and therefore show added value from the technical (do not require removal) and social (come from renewable sources) perspectives.

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