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Morpho-physiological responses of Italian ryegrass to volcanic ashfalls from Calbuco

Respuestas morfo-fisiológicas de ballica italiana a la caída de cenizas volcánicas del Calbuco

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† In memoriam of Lorena Riedel Stolzenbach (June 2, 1962 – November 19, 2018).

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A B S T R A C T

There are nearly 90 actives volcanoes in Chile that helped to shape the landscape of the country and created fertile soils. Productive pastures established on these soils are always at risk of being buried after fresh volcanic ashfalls. Therefore, our investigation aimed to characterise the physicochemical properties of the volcanic ashfalls (from the Calbuco volcano eruption) and their effect on the biomass response and morpho-physiological parameters of *Lolium multiflorum* Lam. The experiment considers: one control (100% soil), a mixture soil:ashfall (75:25 and 50:50) and two ashfall sizes, coarse and fine, respectively. Available nutrients, undisturbed height, number of leaves, herbage mass, and crude protein (CP) were determined. The ashfalls are low in macronutrients concentrations Ca^{2+} = 0.5 $cmol_+$ kg^{-1}, Olsen-P = 1.4 mg kg^{-1} and K^+ = 5.0 mg kg^{-1}, however, when mixed with soil the N concentration increases from 11.6 mg kg^{-1} to 18.4 mg kg^{-1}. The plants growing in mixtures with 50% fine ashfall were 2 cm higher and presented 10% less leaves than the control. No differences in herbage mass were observed. However, CP increased with mineral-N content and was higher when the ashfalls made up the 25% weight in the soil-ashfall mixture. Ashfalls incorporation into the soil as a result of cultivating and sowing new species such as *L. multiflorum* are a viable alternative towards the recovery of the pastureland after volcanic ashfalls.

RESUMEN

Cerca de 90 volcanes activos se encuentran en Chile, moldeando el paisaje y creando suelos fértiles. Praderas productivas establecidas sobre estos suelos, pueden ser enterradas después de la caída de cenizas volcánicas frescas. Considerando este proceso, el objetivo de nuestra investigación fue caracterizar fisicoquímicamente una ceniza volcánica (erupción volcán Calbuco) y su efecto en la biomasa y parámetros morfo-fisiológicos de *Lolium multiflorum* Lam. El experimento consideró: un control (100% suelo) una mezcla suelo:ceniza (75:25 y 50:50) y dos tamaños de ceniza, gruesa y fina, respectivamente. Se determinaron nutrientes disponibles, altura sin disturbar, número de hojas, masa herbácea y proteína cruda (CP). Las cenizas son bajas en macronutrientes Ca^{+2} = 0,5 $cmol_+$ kg^{-1}, P-Olsen = 1,4 mg kg^{-1} and K^+ = 5,0 mg kg^{-1}, sin embargo, al mezclarse con el suelo, la concentración de N incrementa de 11,6 mg kg^{-1} a 18,4 mg kg^{-1}. Las plantas que crecieron en mezclas con 50% de ceniza fina fueron 2 cm más altas que y presentaron 10% menos de hojas que el control. No se observaron diferencias en la masa herbácea. Sin embargo, CP incremento con el contenido de N-mineral y fue mayor cuando la cenizas constituían el 25% del peso de la mezcla suelo:ceniza. La incorporación de las cenizas al suelo mediante el cultivo y la siembra de nuevas especies como *L. multiflorum* son una alternativa viable para la recuperación de las praderas, después de la caída de cenizas volcánicas.

Palabras claves: mezcla suelo-ceniza, nitrógeno, proteína cruda, pradera.

INTRODUCTION

The "Circum-Pacific Ring of Fire" (RoF) is a direct consequence of the movement of tectonic plates, a theory developed by Alfred Wegener at the beginning of the 20th century that is known as "continental drift" (Gregory, 1925). RoF is the most volcanically active zone in the world (USGS, 2018), including volcanoes in New Zealand, Japan, the Philippines, Aleutian, Alaska, North America and South America (Shoji *et al*., 1993). The volcanism is intimately linked with the development of humankind, *e.g.* the Vesuvius eruption in AD 79 when Pompeii was buried under volcanic ashfalls, with the heat of pyroclastic surges and flows being the main cause of death of the population (Mastrolorenzo *et al*., 2010). Another good example is the Krakatau eruption and the effects of a tsunami in Sumatra in 1883 (Self, 1992).

Recent studies indicate that volcanic eruptions release stratospheric sulphated sprays, which change the quality and quantity of solar light, affecting negatively the performance of crops in C3 (rice, wheat, soybean) and C4 species (maize) (Proctor *et al*., 2018). However, volcanic eruptions have contributed to the resurgence of human activities, shaping the landscapes and creating new and fertile soils. After the eruption of Mount St. Helens, Cook *et al*. (1981) reported that the crop production was normal or above normal in wheat, potatoes, and apples trees. However, the same authors indicated that the ashfalls reduced the water infiltration and caused damage to insects due to their abrasive effect. The responses of soil fertility and pore space distribution should be considered in the short and long-term, depending on the chemical composition of the ashfalls (*e.g.* SiO_2), weather conditions, time, and tephra thickness and properties of the soil beneath, among others (Besoaín *et al*., 1995; González *et al*., 2015; Craig *et al*., 2016).

In Chile, nearly 90 volcanoes are active (SERNAGEOMIN, 2015) and after the eruption of Quizapú volcano in 1932[1] (Rovere *et al*., 2012), the most important eruptions in the last 30 years (Lonquimay: 1989; Hudson: 1991; Chaitén, 2008; Puyehue-Cordón Caulle, 2011; Calbuco; 2015) have concentrated between the Araucanía Region and the Aysén Region, where 51% of total volcanoes in Chile are located (Figure 1A). The consequences of volcanic ashfalls after the eruptions of volcanoes for the agriculture in this area are reported for Lonquimay (Besoaín *et al*., 1992); Hudson, (Besoaín *et al*., 1995; Craig *et al*., 2016); Chaitén (Lara, 2009), Puyehue-Cordón Caulle (Wilson *et al*., 2013) and Calbuco (Romero *et al*., 2016) volcanoes, however, there are not many studies related to the recovery of pastures under volcanic ashfalls deposits.

[1] One of the highest eruptions in the 20th century. The eruptive column reached 15-30 km height and released 30 km³ of tephra. The ashes reached Buenos Aires, Montevideo and the south of Brazil (Rovere *et al*., 2012).

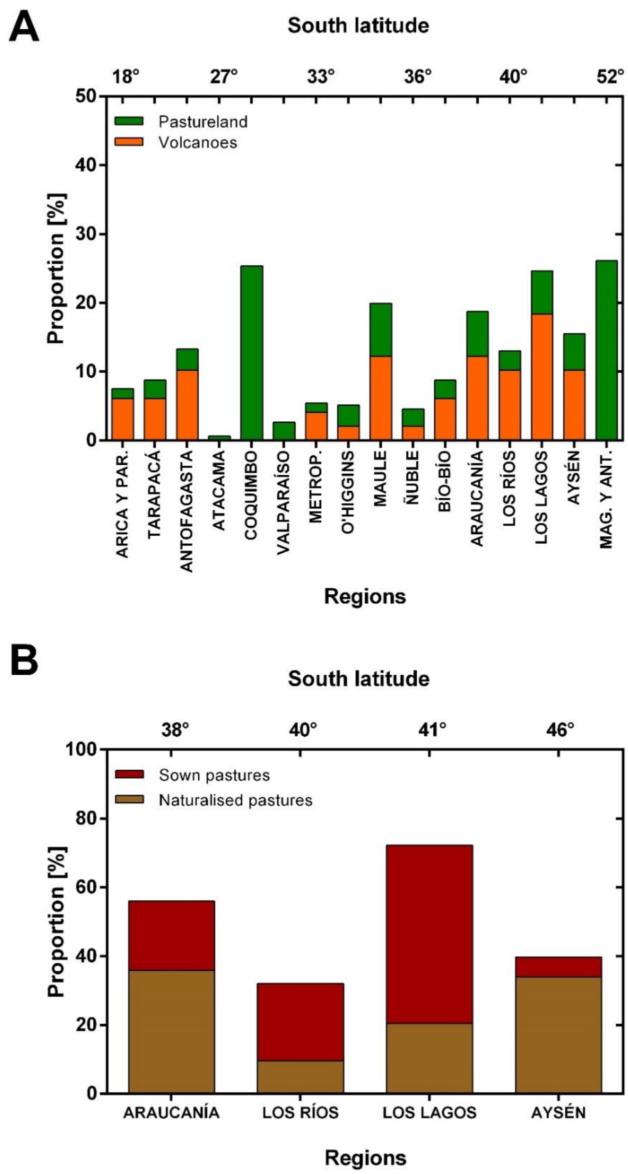

Figure 1. Volcanoes and pastureland distribution by administrative regions in Chile (1A). Naturalised and sown pastures between Araucanía and Aysén regions (1B)[2].

Figura 1. Distribución de volcanes y praderas por regiones administrativas en Chile (1A). Praderas naturalizadas y sembradas entre las regiones de la Araucanía y Aysén (1B)[2].

In Chile, soils derived from volcanic materials (Andosols) are dominant between latitudes (38° to 46° S) (Luzio and Alcayaga, 1992). Pastures in that area represent 20% of the national surface, and sown pastures prevail in both regions, Los Ríos and Los Lagos (Figure 1B).

A wide range of cultivated pastures in the south of Chile are dominated by grasses such as *Lolium perenne* L. Demanet (2000), the most used species in dairy

[2] Figures elaborated with information obtained from INE (2007) and SERNAGEOMIN (2018).

and beef production which covers 65% of the pastures. Second to that is *Lolium multiflorum* Lam. (Ballica italiana or Italian ryegrass), a bi-annual grass used in short rotation and characterised by a fast growth with successful establishment in soils with medium to low fertility (Parga, 2008).

Volcanic ashfalls deposits are frequently found in the south of Chile, therefore the following questions arise: can we use volcanic ashfall? (*e.g.* in agriculture); which are the physicochemical properties of the volcanic material?; how relevant is their thickness?; is it possible to recover the agronomic attributes of pastures buried under volcanic ashfalls deposition?

This manuscript was developed in the framework of the "XV Congreso Regional Escolar de la Ciencia y la Tecnología Explora de CONICYT Región de Los Ríos 2018" and the results characterise the physicochemical characteristics of the ashfalls and analyse the effect of ashfall in the soil and the response related to both, biomass production and morpho-physiological parameters of *L. multiflorum*.

MATERIALS AND METHODS

Site description and sampling

The soils are in the geomorphological unit corresponding to the San José depression, where volcanic ashfalls were deposited over a cemented tuff known as sandstone ("cancagua" in Spanish). The classification of the soil corresponds to Petroduric Silandic Andosol (Valdivia soil series). These soils are deep, well drained and the topography is dominated by slopes between 5% to 8% (CIREN, 2003; Salazar *et al*., 2005). The sampling was performed at "Estación Experimental Agropecuaria Austral" of the Universidad Austral de Chile (39°47' S, 73°13' W). Sixty kg of disturbed soil were collected from the first 20 cm soil depth and stored in plastic bags.

The volcanic ashfall depositions over the soils were originated during the eruption of the Calbuco volcano (April 22nd, 2015). The material was collected around the Rupanco Lake (40° 53' S; 72° 15' W) and in Ensenada village (41° 12' S; 72° 32' W), 56 km and 15 km from Calbuco volcano, and the samples were identified as fine ashfall and coarse ashfall, respectively. The particle size of volcanic ashfall is shown in Figure 2.

Eighty seven percent of coarse ashfall ranged between 4000 μm to 600 μm being classified as gravel size, while 89% of fine ashfalls ranged between 212 μm to 125 μm in the fine sand size.

Soil analyses

The samples were homogenised, sieved (< 2 mm) and dried (~25 °C) for soil chemical analyses. Soil pH was determined by using a soil:solution in a proportion (1:2.5), soil organic carbon (SOC) was measured through wet combustion, mineral nitrogen (N-min) through Kjeldahl distillation and available phosphorus using Olsen-P. The exchangeable cations (Ca^{2+}, Mg^{2+}, Na^{+}, K^{+}) were determined using an ammonium acetate solution and expressed as effective cation exchange capacity (ECEC). Exchangeable aluminium (Al^{3+}) was determined using a KCl 1N and the micronutrients (Fe, Cu, Zn and Mn) were determined using DTPA solution at pH 7.3 (Sadzawka, 1990). The elements concentration was determined by atomic absorption spectroscopy, (SavantAA AAS-GBC, Australia).

Experimental design

The experiment design was completely randomised and considered four treatments and one control with four replicates each (Table 1) distributed in twenty plastic pots (l = 16 cm; a = 269 cm^2).

The treatments were designed to evaluate the addition of volcanic ashfalls in (25%) and (50%) in the soil

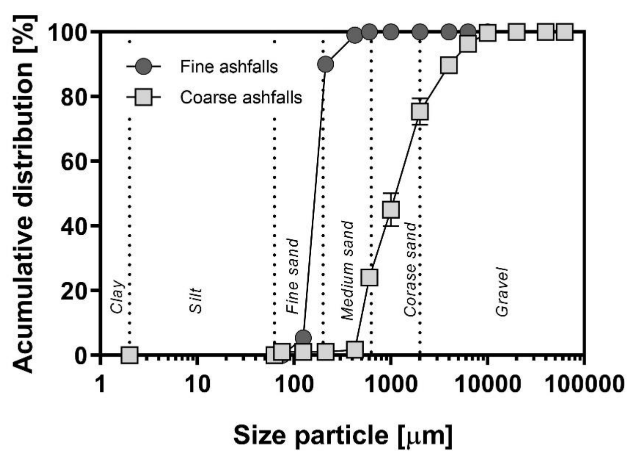

Figure 2. Size particle distribution of coarse and fine volcanic ashfalls.

Figura 2. Distribución del tamaño de partículas de cenizas gruesas y finas.

Table 1. Treatment descriptions and proportion of volcanic ash soil (VAS) and ashfalls according to size.

Cuadro 1. Descripciones de los tratamientos y proporción de suelo de cenizas volcánicas (VAS) y cenizas según tamaño.

Control	100% Volcanic ash soil (VAS = 3 kg)
T1	75% VAS (2.25 kg) + 25% fine ashfalls (0.75 kg)
T2	75% VAS (2.25 kg) + 25% coarse ashfalls (0.75 kg)
T3	50% VAS (1.50 kg) + 50% fine ashfalls (1.50 kg)
T4	50% VAS (1.50 kg) + 50% coarse ashfalls (1.50 kg)

profile after a volcanic eruption. The calculations of the proportion soil:ashfall considered a bulk density (0.7 g cm^{-3}) representative of the soil series (Dec *et al*., 2011). In each pot, eight seeds of *L. multiflorum* cv. Tama were sowed considering a commercial dose of 25 kg ha^{-1}. To ensure the germination and growth of the seeds, the pots were located inside a glasshouse with temperature (20 °C), light (16 h d^{-1}) and irrigation control.

Plants height and herbage mass

For each plant, morphological measurements such as undisturbed length of the plant (cm) and live leaf numbers were performed every 7 days, but some measurements were extended to 10 days. A leaf blade was considered "live" when the leaf was over 50% green and the number of leaves considered the sum of leaf totally expanded and leaves in process of expansion (Canseco *et al*., 2007). These measurements were conducted from June 26th to August 10th, 2018. At that time, the plants were defoliated (just once) at 4 cm, placed inside paper bags and dried in an oven at 60 °C for 48 h for dry matter determination (Canseco *et al*., 2007). At the same time, a soil sample was taken to evaluate the effect of soil:ashfall mixture over the ryegrass.

Crude protein

After defoliation (August 10th), the nitrogen concentration was determined in the leaves by LECO FP528 based in Dumas method (AOAC, 1995) and the crude protein (CP) was calculated as follows:

$$CP = N*6.25 \tag{1}$$

Where: CP = Crude protein (%); N = Nitrogen content (%).

Statistical analyses

A Shapiro-Wilks test ($p \leq 0.05$) was performed to evaluate the normality of dataset and ANOVA analyses conducted to compare the soil:ashfall mixes and the response of the plant growth and herbage mass treatments. A Multiple comparison test was performed using a Tukey test. Linear regressions were conducted to evaluate the relationship between soil N and crude protein content. The analyses were conducted using STATISTICA 7.0 and GraphPad Prism version 6.01 for Windows.

RESULTS AND DISCUSSION

Soil and ashfalls properties

The analyses conducted to know the initial nutritional state of the studied materials and their chemical characteristics are presented in Table 2.

Olsen-P was higher ($p \leq 0.001$) in the control (5.4 mg kg^{-1}) compared to the fine and coarse ashfalls (0.8 mg kg^{-1} and 1.5 mg kg^{-1}, respectively). Exchangeable potassium reached 133 mg kg^{-1} in the soil, while reaching a very low value (5 mg kg^{-1}; $p \leq 0.001$) in ashfalls. Ashfalls had a low Al saturation (5.6%) compared to the control (22%). Iron was higher in the soil (29 mg kg^{-1}), while fine ashfalls presented 39% lower in iron concentration compared to coarse ashfalls ($p \leq 0.01$).

In general, these properties remained stable in the soil-ashfall mixtures investigated in this research (Table 3).

Significant changes were found in mineral-N concentration ($p \leq 0.05$) and exchangeable potassium ($p \leq 0.0001$). N concentration was higher in the treatments (14.0 mg kg^{-1} to 18.4 mg kg^{-1}) compared to N concentration in the control (11.6 mg kg^{-1}). Potassium concentration decreased in the following order: Control > T2 > T1 > T4 > T3, with values between 165 mg kg^{-1} to 58 mg kg^{-1}.

Romero *et al*. (2016) reported that the ashfalls from the Calbuco eruption is basaltic andesite composed mainly by plagioclase, volcanic glass, pyroxene and lithics, describing irregular shards and blocky particles with inclusions of Fe and Ti oxides that could be related with the high Fe concentrations found in this study. In general, research performed to determine chemical properties of volcanic ashfalls has been focused on determining the oxide weight composition (*e.g.* SiO_2) (Vogel *et al*., 2017), however, the results that address the link of agronomic value of nutrients with Calbuco volcanic ashfalls are scarce.

Beutell (1893) quantified calcium, magnesium and potassium oxides and phosphoric acid among others, gave values of 6.38%, 1.50%, 0.55% and 0.20% respectively. Thus, ashfalls themselves are not a valid source of nutrients because they are unable to pass through fast enough towards the soil solution and be made available for absorption by plant roots. These results of ashfall nutrients availability are opposite to the findings reported by Besoaín *et al*. (1995), indicating values of 1,378 mg kg^{-1}, 70 mg kg^{-1} and 18 mg kg^{-1} of calcium, potassium and phosphorus, respectively, in volcanic ashfalls from the Hudson volcano distributed between Coyhaique and Puerto Aysén during the eruption of 1991.

Morpho-physiological responses to soil:ashfall mixes

The responses in undisturbed plant height showed significant differences ($p \leq 0.05$) among the control and T3 and T1 *vs*. T3 (Figure 3A), while the leaf number showed differences two (July 13th) and eight weeks after sowing (August 10th) (Figure 3B).

Plant height was similar at the beginning of the experiment (June 26th), however, after one week (July 5th) the plants of treatment 3 tended to be 2 cm taller, difference that significantly increased by July 24th and July 31st

Table 2. Chemical properties in control (Andosol), fine and coarse ashfalls before the experiment. Values correspond to available fractions and are shown as mean values ± standard error.

Cuadro 2. Propiedades químicas en control (Andosol), cenizas finas y gruesas antes del experimento. Los valores corresponden a las fracciones disponibles y se muestran como valores medios ± error estándar.

Soil property		Control	Fine ashfalls	Coarse ashfalls	Significance
pH	[log H^+]	5.60 ± 0.00	6.35 ± 0.05	6.15 ± 0.05	ns
SOC	[%]	6.50 ± 0.20	n.d.	n.d.	-
P-Olsen	[mg kg^{-1}]	5.35 ± 0.15	0.80 ± 0.00	1.45 ± 0.85	***
K^+	[mg kg^{-1}]	132.50 ± 2.50	5.00 ± 0.00	5.00 ± 0.00	***
Na^+	[$cmol_+$ kg^{-1}]	0.05 ± 0.00	0.12 ± 0.00	0.06 ± 0.00	ns
Ca^{2+}	[$cmol_+$ kg^{-1}]	1.01 ± 0.21	0.52 ± 0.03	0.29 ± 0.00	ns
Mg^{2+}	[$cmol_+$ kg^{-1}]	0.44 ± 0.03	0.09 ± 0.02	0.06 ± 0.01	ns
Al^{3+}	[$cmol_+$ kg^{-1}]	0.52 ± 0.02	0.04 ± 0.00	0.03 ± 0.01	ns
ECEC	[$cmol_+$ kg^{-1}]	2.36 ± 0.21	0.78 ± 0.04	0.45 ± 0.02	ns
Al saturation	[%]	22.15 ± 2.85	5.65 ± 0.15	5.65 ± 0.25	****
Zn	[mg kg^{-1}]	0.21 ± 0.00	0.19 ± 0.02	0.16 ± 0.00	ns
Fe	[mg kg^{-1}]	29.10 ± 0.20	5.50 ± 0.00	8.95 ± 1.05	****; **
Cu	[mg kg^{-1}]	0.81 ± 0.00	0.40 ± 0.00	0.54 ± 0.03	ns
Mn	[mg kg^{-1}]	1.28 ± 0.03	0.80 ± 0.00	0.78 ± 0.06	ns

ns: $p > 0.05$; *: $p \leq 0.05$; **: $p \leq 0.01$; ***: $p \leq 0.001$; ****: $p \leq 0.0001$; n.d.: not detected.

Table 3. Chemical properties in control and soil:ashfall mixture after the experiment. Values correspond to available fractions and are shown as mean values ± standard error. Letters indicate differences among treatments.

Cuadro 3. Propiedades químicas en control y las mezclas suelo:ceniza después del experimento. Los valores corresponden a las fracciones disponibles y se muestran como valores medios ± error estándar. Letras indican diferencias entre los tratamientos.

Soil property		Control	T1	T2	T3	T4	Significance
pH	[log H^+]	5.53 ± 0.06	5.68 ± 0.08	5.60 ± 0.07	5.63 ± 0.05	5.53 ± 0.02	ns
Mineral-N	[mg kg^{-1}]	11.55 ± 0.73b	18.38 ± 3.07a	15.75 ± 1.05a	14.00 ± 2.73a	14.47 ± 1.23 a	*
P-Olsen	[mg kg^{-1}]	3.90 ± 0.16	3.63 ± 0.14	4.00 ± 0.39	3.68 ± 0.05	4.08 ± 0.13	ns
K^+	[mg kg^{-1}]	164.75 ± 4.55a	107.25 ± 4.61bc	124.75 ± 5.00b	58.00 ± 1.87d	100.50 ± 3.93 c	****
Na^+	[$cmol_+$ kg^{-1}]	0.10 ± 0.00	0.14 ± 0.00	0.15 ± 0.01	0.17 ± 0.03	0.20 ± 0.02	ns
Ca^{2+}	[$cmol_+$ kg^{-1}]	0.96 ± 0.19	0.86 ± 0.02	1.00 ± 0.03	0.77 ± 0.03	1.07 ± 0.05	ns
Mg^{2+}	[$cmol_+$ kg^{-1}]	0.45 ± 0.05	0.36 ± 0.01	0.43 ± 0.01	0.28 ± 0.01	0.39 ± 0.02	ns
Al^{3+}	[$cmol_+$ kg^{-1}]	0.53 ± 0.00	0.43 ± 0.01	0.45 ± 0.01	0.31 ± 0.00	0.38 ± 0.00	ns
ECEC	[$cmol_+$ kg^{-1}]	2.45 ± 0.25	2.05 ± 0.05	2.35 ± 0.04	1.68 ± 0.06	2.29 ± 0.01	ns
Al saturation	[%]	21.85 ± 1.71	20.73 ± 0.35	19.15 ± 0.48	18.70 ± 0.56	16.65 ± 0.53	ns

ns: $p > 0.05$; *: $p \leq 0.05$; ****: $p \leq 0.0001$.

($p \leq 0.05$). At these dates the plant height growth rate was of 0.23 cm d^{-1} for the control, while in T3 was 0.29 cm d^{-1} (Figure 3A). The average leaf number was 10% and 11% lower in the control compared to T3 ($p \leq 0.05$) and T4 ($p \leq 0.01$), respectively, in July 13th. At the end of the experiment, these differences were also observed between the control and T4 ($p \leq 0.001$; 50% coarse ashfalls), and between T2 and T4 ($p \leq 0.01$; 25% coarse ashfalls).

Responses in biomass and leaf protein content

The biomass responses to treatments and the relationship between mineral-N and crude protein (CP) are shown in the Figures 4A and 4B, respectively.

The herbage mass did not show significant differences ($p = 0.33$) among the treatments (Figure 4A) and ranged between 10 Mg DM ha^{-1} to 16 Mg DM ha^{-1}. However, as the soil mineral nitrogen increased, the concentration of CP in the leaves also increased ($R^2 = 0.95$). CP varied between 2.3% to 2.9%, while the mineral-N between 14 mg kg^{-1} to 18 mg kg^{-1}. This increase was greater in plants that grew in the mixture with 25% ashfalls, compared to treatments that contained 50% ashfalls, however, at the lowest mineral-N concentration (control = 11.5 mg kg^{-1}), *L. multiflorum* plants had a high protein content in their leaves (2.8%) (Figure 4B).

L. multiflorum is widely used in pastures in southern Chile particularly on intensive grazing systems during

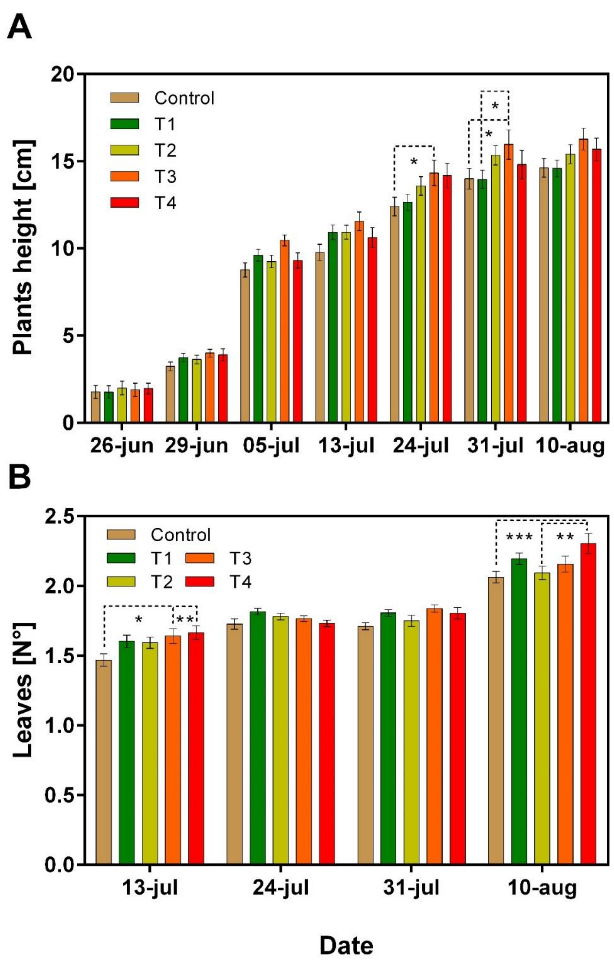

Figure 3. Plants height (A) and leave numbers (B) in treatments during the experimental period. Mean values ± standard error is shown. Asterisks denote significant differences among treatments. *: $p \leq 0.05$; **: $p \leq 0.01$; ***: $p \leq 0.001$.

Figura 3. Altura de plantas (A) y número de hojas (B) por tratamiento durante el periodo experimental. Se presentan valores medios ± error estándar. Asteriscos denotan diferencias significativas entre tratamientos. *: p ≤ 0,05; **: p ≤ 0,01; ***: p ≤ 0,001.

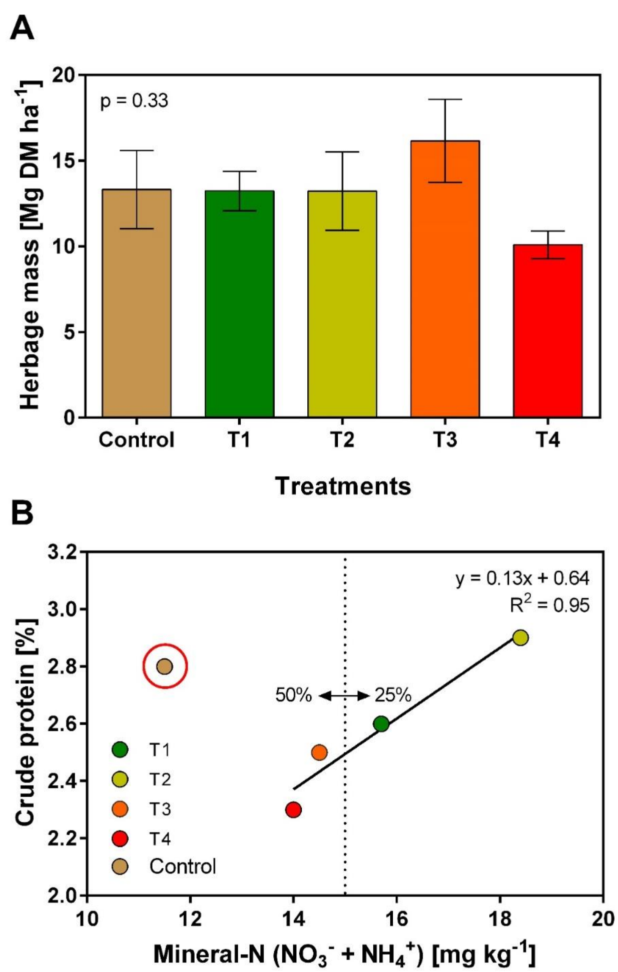

Figure 4. Mean values ± standard error for herbage mass (A) and the relationship between crude leaf protein and soil mineral nitrogen (B). Red circle indicate that control was excluded from regression analyses.

Figura 4. Valores promedio ± error estándar para masa herbácea (A) y la relación entre la proteína cruda de la hoja y el nitrógeno mineral del suelo (B). El círculo rojo indica que el control fue excluido del análisis de regresión.

winter (Demanet, 2000) due to competitive strategies and higher growing rate. Kobayashi *et al*. (2008) evaluated the nutritive value of Italian ryegrass in Japan during the winter period and reported differences in plant height of *L. multiflorum* after 30 days from sowing in the late autumn, similarly to the result of this research after 28 and 35 days after sowing.

The interaction between soil:ashfall and the ecophysiological responses in *Embothrium coccineum* J.R. Forst. & G. Forst were reported by Delgado *et al*. (2015). In extremely disturbed environments (*e.g.* after volcanic ashfalls), *E. coccineum* can colonise the volcanic substrate through the cluster roots mechanism and solubilise phosphorus from this volcanic material, so it is considered as pioneer plant in harsh environments. On the other hand, España *et al*. (2016) studied the effect of slurry pig mixtures application as amendment in tailings, using *L. perenne* as bioindicator plant and reported that the increase of slurry concentration in *L. perenne* inhibits both the germination and the root growth compared with tailing, even when the slurry increased the macronutrient concentration the sandy texture of mine tailings seems to be the first factor in the establishment of *L. perenne*. The effects of volcanic ashes over particle size distribution were reported by González *et al*. (2015), using different types of soils (Andosols and Ultisols) and soil:ashfall concentration doses. An increase in volumetric water content at -6 kPa and a decrease at -1543 kPa of matric potential as the dose increased, is reflected in an increase of plant available water. The pore interconnection, reflected on the air conductivity, increased in Andosols and diminished in Ultisol. This has been related to soil shrinkage at initial matric potentials (near to saturation), causing a diminishing of the soil macropore volume and the air convective fluxes in soils (Dörner *et al*., 2013b).

A direct relationship between mineral-N and CP has been measured. Pavinato *et al*. (2014) reported increasing levels of CP of 146 g kg^{-1} to 163 g kg^{-1} and 130 g kg^{-1} to 156 g kg^{-1} in seasons 2009 and 2010, respectively, as the N rate increased (0; 40; 80 and 120 kg N ha^{-1}). However, in this study, the lowest value of mineral-N (11.5 mg kg^{-1} in the control) reached one of the highest values of CP (2.8%). This could be have been related to the hypothesis proposed by Clunes and Pinochet (2019) about the physical storage of inorganic nitrogen in the soil. The predominance of coarse particles in the ashfalls (similar as sand particles) causes a pore size redistribution and changes in the air/water fluxes (González *et al*., 2015) and, consequently, changes in the nitrogen availability. Regardless of the size of the ashfalls (coarse or fine), soils mixed with ashfalls in 25% by weight had more mineral-N and CP compared to the treatments with mixed 50% soil and 50% ashfalls. The ashfalls deposited and mixed in Andosols favours the formation of macropores (> 50 µm) increasing the infiltration but also the N availability due to their capacity to store plant available water at low water tensions (Dörner *et al*., 2013a) and, consequently, available N. The higher CP found in the control could have been caused by an organic mineralisation of N. An increase of plant available water in the soil can promote "pulses" of N, increasing the nitrogen diffusion from soil matrix and favouring an accumulation of nitrates in the soil solution. If the roots and the nutrients match in the same space, a synchrony between the nitrate and absorbing hair could have occurred; in this way, the plant can absorb the nitrate and transform it into protein molecules, regardless of the low mineral-N in the soil (Clunes and Pinochet, 2019).

CONCLUSIONS

The available concentrations of macronutrients found in the ashfalls were low in calcium, available phosphorus and exchangeable potassium compared with the control. However, the mineral-N increased in soil:ashfall mixture.

The ashfalls mixed with soils in different concentration caused changes in the morpho-physiological parameters in *L. multiflorum*. Plants grown in T3 were 2 cm higher than those in the other treatments, while the leaf number was 10% and 11% lower in the control compare to T3 and T4, respectively. No differences in herbage mass between treatments were found.

The CP increased as mineral-N increased in the substrate. CP was higher when the ashfalls made up to 25% of the soil:ashfall mixture, regardless to the size of the ashfalls.

After the volcanic ashfall, their incorporation into the soil, for example by mixing them via soil cultivation and then sowing grasses as *L. multiflorum,* is a realistic alternative to recover pastureland. However, this will depend of properties such as ashfall thickness, nutrient concentration of the ashfalls, weather conditions and farmer technological level.

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