Physiognomic changes in response to herbivory increase carbon allocation to roots in a temperate grassland of central Argentina

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Abstract   In most temperate grasslands, cattle grazing can promote physiognomic changes on plant communities, as well as changes in species growth patterns. Through these changes in vegetation, cattle grazing can generate changes in allocation to total, fine and coarse root biomass, which can further affect ecosystem processes. The objective of this work was to evaluate differences in root biomass, in the proportions of fine and coarse root biomass and in root C:N ratio across physiognomies from 0 to 30 cm. We selected plots of grazing lawns (associated to relatively high stocking rates), and plots of thin and thick tussock grasslands (associated to relatively low stocking rates) within the Pampa de Achala system in central Argentina. We found that total, coarse, and fine root biomass were highest in grazing lawns, intermediate in thin tussock grasslands, and lowest in thick tussock grasslands. Thick tussock grasslands showed the highest relative proportion of fine roots while root C:N ratio did not vary among physiognomies. Higher root biomass in lawns was probably caused by the dominance of species that allocate more biomass to roots than to shoots, as well as by the complementarity of soil resource extraction and by compensatory root growth in response to herbivory. In addition, and contrary to what was expected based on their above-

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ground attributes, thick tussock grasslands presented higher proportion of fine roots, which suggests a more acquisitive resource use strategy. However, root C:N from thick tussock grasslands did not differ from other physiognomies which might indicate there are not clear differences in below-ground resource use strategies. Our results show that grassland root biomass in this study system can increase in patches associated to higher cattle grazing due to changes in plant composition and physiognomy, while changes in root quality seem to be uncoupled from the above-ground traits.

Introduction

Grassland ecosystems worldwide are a major component of the global C cycle due to their extension (Gang et al. 2014) and due to the high amounts of organic carbon present in their soils (SOC; Hungate et al. 1997; King et al. 1997; Gill et al. 1999; Amundson 2001; Derner et al. 2006). SOC is a particularly stable C pool (Cotrufo et al. 2015) and its formation is highly linked to the decomposition of root tissues and exudates (Rasse et al. 2005; Mambelli et al. 2011; Freschet et al. 2013; Hu et al. 2016; Shahbaz et al. 2016). In this line, below-ground productivity can account for around 67% of total productivity in grasslands (Chapin et al. 2011), resulting in a below-ground biomass from two up to 30 times higher than above-ground biomass (Moretto et al. 2001; Gao et al. 2008). In spite of its importance, below-ground biomass in grassland ecosystems is still under-studied (Wilson 2014).

Currently, the increase in the use of lowlands for agriculture has increased the livestock pressure on most mountain systems, in particular on grassland physiognomies within them (Asner et al. 2004; FAO 2016). Grazing produces both short- and long-term changes in grasslands. The most immediate effect of grazing on vegetation is defoliation and, as a result of this, an increase in the below-ground biomass proportion relative to above-ground biomass. Also as a consequence of defoliation, consumed species can increase their absolute biomass allocation to below-

ground tissues, further differentiating above- and below-ground biomass compartments (McNaughton 1983; Shariff et al. 1994; Holland et al. 1996; Frank et al. 2002; Hamilton et al. 2008; Garcia-Pausas et al. 2011; Wilson et al. 2018). In the long term, grazing can lead to floristic changes (Bardgett and Wardle 2003; Díaz et al. 2007; Cingolani et al. 2008). Herbivory often favors the dominance of rosette or prostrate growth forms, which have higher root biomass relative to leaf biomass (Pucheta et al. 2004; Piñeiro et al. 2010; López-Mársico et al. 2015, 2016). This in turn increases even more the proportion of below-ground biomass. In systems without serious nutrient and water limitations, floristic changes may also involve the replacement of grazing intolerant, conservative species by grazing tolerant, acquisitive species (plants with higher Specific Leaf Area and N content, lower C:N ratio and total fiber content; Bardgett and Wardle 2003; Cingolani et al. 2007; Lavorel et al. 2007; Vaieretti et al. 2013; von Müller et al. 2017). Assuming that above- and below-ground plant functional strategies are coordinated (Freschet et al. 2013), it should be expected that grazing tolerant species have also acquisitive root attributes such as higher proportion of fine roots, lower concentrations of C and higher concentrations of N, compared to non-acquisitive and grazing intolerant plants (Lavorel et al. 2007; Bardgett et al. 2014; Prieto et al. 2015; Roumet et al. 2016). As such, changes in plant tissue quality could further affect nutrient and C cycling and its effects on decomposers activity (Bardgett and Wardle 2003).

Pampa de Achala, located on the highlands of Córdoba hills in Argentina, represents a grazed grassland without strong water or nutrient limitations (for water availability: Cingolani et al. 2015; Colladon et al. 2013; for nutrient availability: Vaieretti et al. 2010). In this system, high grazing pressure maintains a landscape dominated by lawn patches with low above-ground biomass covered by short species with acquisitive characteristics. Under grazing exclusion or very low stocking rates, the landscape is dominated by tall tussock grasses with more conservative characteristics (Pucheta et al. 1998; Cingolani et al. 2007; Vaieretti et al. 2013; von Müller et al. 2017).

Below-ground biomass in grazing lawns at the site have been found to be either equal (but with higher fine root proportion; Pucheta et al. 2004) or higher (Jaras Zamar 2016) than below-ground biomass at tussock

physiognomies, while information on below-ground traits is limited (e.g.,: C:N ratio; Jaras Zamar 2016). In this context, our first objective was to evaluate differences in root biomass between the dominant physiognomy found under higher grazing pressure (grazing lawns) and the dominant physiognomies found under lower grazing pressure (thin and thick tussock grasslands) in a temperate grassland system of central Argentina along the first 30 cm of soil and at three separate depths (from 0–5, 5–15, and 15–30 cm). Our second objective was to determine the differences in absolute and proportional amounts of fine and coarse root biomass between physiognomies at the same depths. Our third objective was to analyze the differences in total root biomass C:N ratio between physiognomies for the whole 30 cm of soil. Our first hypothesis states that in grazing lawns, the vegetation allocates a great amount of biomass to roots (Piñeiro et al. 2010), while null or low herbivory in tussock grasslands promotes the development of species with a smaller amount of biomass allocated to roots, and differences fade with depth because root biomass generally decreases rapidly at deeper soil layers (Prieto et al. 2015). Our second hypothesis states that, as grazing lawns are dominated by species with leaf attributes linked to a more acquisitive syndrome (Vaieretti et al. 2013; Poca et al. 2015), the absolute and relative abundance of fine roots (diameter < 1 mm; Pucheta et al. 2004) will be stimulated and roots will present a lower C:N ratio, in comparison to tussock-dominated patches (Pucheta et al. 2004; Lavorel et al. 2007). We predict that (1) total root biomass will be greater in lawns and these differences will be greater at a 0–5 cm depth, (2) absolute and relative fine root biomass will be greater in lawns, and (3) root C:N ratio will be lower in lawns, all compared to both types of tussock grasslands.

Methods

Study site

The study was developed in paddocks located inside the Quebrada del Condorito National Park (31°37′S, 64°48′W; at 2000 m a.s.l.) and in neighboring private properties, during February and March 2017 (i.e., plant maximum growth season; Pucheta et al. 2004). The mean temperatures of the coldest and warmest

months are 5.08 °C and 11.48 °C, respectively, and there is no frost-free period. Mean annual precipitation is ∼ 900 mm, with most rainfall concentrated in the warmest months (Colladon et al. 2013; Cingolani et al. 2015). Climate conditions during the year prior to the study (2016) were measured at a climatic station installed at one of the study paddocks. Mean temperatures of the coldest and warmest months were 2.04 and 15.86 °C, respectively, and precipitations were 732 mm. Soils in the area are derived from the weathering of the granitic substrate and fine-textured eolian deposits (Cabido et al. 1987). About 400 years ago, domestic European livestock were introduced in these mountains and their presence continues till date (von Müller et al. 2017). At present, cattle grazing is used inside the National Park as a management tool with conservation aims. Outside the Park, private-owned paddocks present comparatively higher stocking rates (see "Sampling design" section below). The landscape within the study area is an undulating plain with different vegetation patches, where more intensively grazed paddocks are predominantly covered by lawn patches dominated by species like *Lachemilla pinnata* (Ruiz & Pav.) Rothm., *Muhlenbergia peruviana* (P. Beauv.) Steud., *Carex fuscula* d'Urv., *Eryngium* spp, among others (hereafter *grazing lawns*). Within the same area, less intensively grazed or ungrazed paddocks are predominantly covered by tussock grassland patches dominated by species like *Poa stuckertii* (Hack.) Parodi (hereafter *thick tussock grasslands*), and *Festuca lilloi* E.B. Alexeev and *Deyeuxia hieronymi* (Hack.) Türpe (hereafter *thin tussock grasslands*; Cingolani et al. 2004; Vaieretti et al. 2013; von Müller et al. 2017).

Sampling design

We selected three pairs of paddocks, each pair constituted by two paddocks with contrasting plant physiognomy, separated by a wire-fence. By working with paired paddocks (in the form of wire-fence contrasts), and selecting plots relatively close to the wire-fence (plot distance to the fence ranged from 5 to 100 m), we maximized environmental and edaphic similarities, but avoided borderline effects. In this way, changes in root biomass and in root characteristics were attributed with more confidence to changes in vegetation physiognomy. Location of paddocks and plots within them is presented in Fig. 1. Paddocks
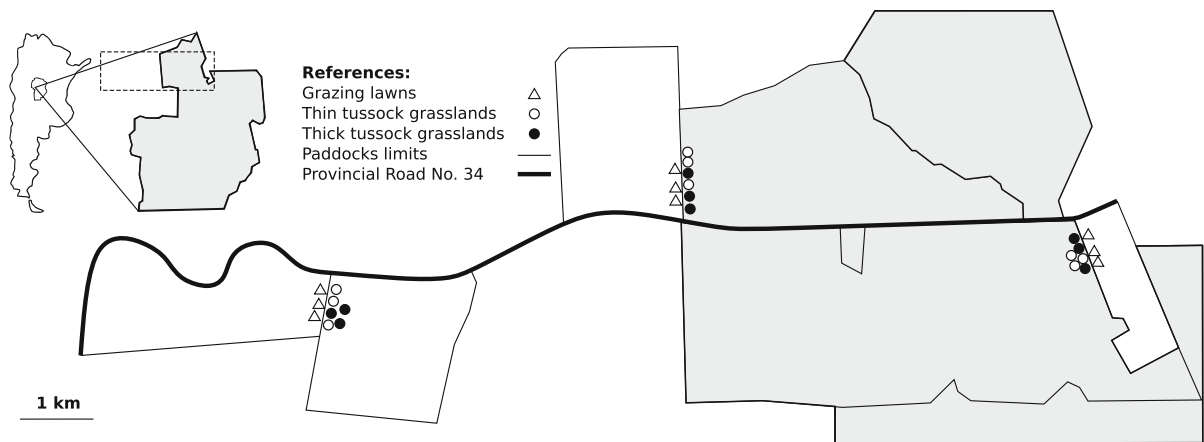
Fig. 1 Geographical disposition of the six study paddocks and grassland plots within them. Map of Argentina (top left) with the Córdoba province at the center and an expanded view of the Quebrada del Condorito National Park (gray areas) where the study area (expanded below) is marked with a dotted line

under higher grazing pressure are under annual effective stocking rates of above 0.23 animal units per ha of non-rocky surface (Vaieretti et al. 2018) while paddocks under lower grazing pressure are under stocking rates from 0.02 to 0.18 animal units per ha of non-rocky surface (Cingolani et al. 2014; von Müller et al. 2017). As a result of these differences in stocking rates, grazing lawn patches dominate the landscape at more intensively grazed paddocks, while tussock grassland patches dominate the landscape at less intensely grazed or ungrazed paddocks. Within each pair of paddocks, we selected three 25 m² grazing lawn plots on the side where this physiognomy was more abundant, and six tussock grassland plots (three thin and thick tussock grassland plots) on the other side, making a total of 27 plots (9 of each physiognomy; average distance to the nearest plot of 110 m). Total plant cover at all plots was nearly 100%.

Measurements

In each plot, we took compound soil samples at three depths using a 4-cm-diameter soil corer (8, 4, and 4 soil cores, for each 0–5 cm, 5–15 cm, and 15–30 cm depth, respectively). Samples were taken systematically at each plot corner. We selected a maximum 30-cm soil depth considering rooting depth in the system has been found to be relatively shallow, with 86% of root biomass found in the first 20 cm of soil (Pucheta et al. 2004). Due to the shape of the soil corer used, we were unable to directly calculate the volume

of each soil sample to later estimate root biomass per unit area. In order to correct for this issue, we took a smaller soil sample of a known volume using a 1.8-cm-diameter soil corer in the center of the plot at each depth. This way, we assessed soil bulk density values which were later incorporated in the calculations. We kept soil samples in well-sealed plastic bags to avoid loss of humidity, during their transportation to the laboratory and their storage in fridges (at 6 °C), until samples were processed.

We hand-mixed compound soil samples in the laboratory and separated them in two homogeneous mounds: one for total root biomass extraction ($n = 27 = 3$ plots $\times$ 3 physiognomies $\times$ 3 blocks) and the other for fine and coarse root biomass extraction. This last measurement was done only in one plot of each physiognomy per pair of paddocks ($n = 9 = 1$ plot $\times$ 3 physiognomies $\times$ 3 blocks). We separated small root-free soil mounds to assess soil moisture by the gravimetric method and incorporated these values in the calculations.

To assess total root biomass, we oven dried soil samples (60 °C, 48 h), extracted roots and other below-ground organs with forceps, and washed them to remove adhered soil. We further oven dried (60 °C, 48 h) and weighed roots to measure dry weight (g). This root extraction technique allows to use a bigger sample size as it extends the period of time during which samples can be stored until processed, but it can produce a slight under-estimation of fine root biomass.

We expressed values as root biomass per land surface area (g/m^2) using Eq. 1:

$$\text{Root biomass} = D[B/(S/BD)], \qquad (1)$$

where D is soil depth (cm), B is root biomass (g), S is soil dry weight (g), and BD is soil bulk density (g/cm^3). To assess fine and coarse root biomass, we processed soil samples immediately after being brought back to the laboratory by wrapping them in voile fabric and washing fine soil particles under running water. This technique avoids problems related to fine root extraction such as decomposition and changes in root diameter, but it requires for immediate processing which in turn reduces the amount of samples that can be obtained on each field trip. We separated roots with forceps using a criterion of ≥ 1 mm diameter for coarse roots and < 1 mm for fine roots (Pucheta et al. 2004). We oven dried fine and coarse root samples (60 °C, 48 h) and expressed fine and coarse root biomass values by land surface area (g/m^2) using Eq. 1 as well, but S was calculated subtracting water content to fresh soil. We then used biomass values to calculate coarse:fine roots ratios. We ground root samples (ball mill Pulverisette 0, Fritsch) and we measured their C and N percentages with an elemental analyzer (Perkin Elmer 2400, II series). Thick tussock grassland coarse root samples from 15 to 30 cm depth did not reach the minimum weight necessary for this procedure (3 mg), so we estimated their C and N concentration as the means for thick tussock grassland coarse roots. We finally calculated total root biomass C:N ratios for the whole soil profile (0–30 cm).

Statistical analyses

We used Linear Mixed Models (LMM) to analyze total, coarse and fine root biomass and coarse:fine roots ratio for the whole soil profile (30 cm) and at each depth separately (0–5, 5–15, and 15–30 cm). We also analyzed total (coarse + fine) root C:N ratio for the whole soil profile. Physiognomy was considered as fixed effect and as grouping factor for the variance function (varIdent; Pinheiro and Bates 2000). We used wire-fence contrasts as random effects for total root biomass analysis considered at each depth and at the whole 30 cm, and also for coarse and fine root biomass at the whole 30 cm. We did not use any grouping factor for the variance function of the C:N ratio

analysis. We always chose best fit models considering the Akaike Information Criterion. For every LMM, we made Fisher's LSD tests to compare means. We carried out all tests with a significance level of 0.05 on Infostat (Di Rienzo et al. 2017) using an R interface (3.4.0 version 2017).

Results

Total root biomass

Along the whole soil profile analyzed (0–30 cm; $n = 27$ plots; $p \leq 0.0001$), lawns had twice as much root biomass (682.59 ± 88.82 g/m^2; mean ± SE) than thin tussock grasslands (343.88 ± 60.96 g/m^2) and 5 times as much biomass than thick tussock grasslands (135.12 ± 37.50 g/m^2; Fig. 2a). When each depth was analyzed separately, lawns had higher root biomass from 0 to 5 cm, followed by thin tussock grasslands and thick tussock grasslands, in that order ($p = 0.0001$). From 5 to 15 ($p \leq 0.0001$) and from 15 to 30 cm ($p = 0.0029$), lawns and thin tussock grasslands did not differ from each other, while thick tussock grasslands showed the least amount of root biomass (Table 1).

Coarse and fine root biomass

From 0 to 30 cm, coarse root biomass was higher in grazing lawns than in thick tussock grasslands ($p = 0.0432$), with thin tussock grasslands being intermediate (Fig. 1). When comparing values from 0 to 5 cm depth, both tussock grasslands had lower coarse root biomass than lawns ($p = 0.0140$). For the 5–15 cm depth, lawns showed higher amounts of coarse root biomass than thick tussock grasslands, with thin tussock grasslands being intermediate ($p = 0.0338$). At 15–30 cm soil depth, coarse root biomass in lawns decreased markedly, and did not differ from that of tussock physiognomies (Table 1).

From 0 to 30 cm, fine root biomass was higher in grazing lawns than in both types of tussock grasslands ($p = 0.0207$; Fig. 2b). When analyzed at each depth separately, differences in fine root biomass between physiognomies were only detected at a 5–15 cm depth ($p = 0.0144$), where lawns had higher fine root biomass than thick tussock grasslands, with thin tussock grasslands being intermediate (Table 1).
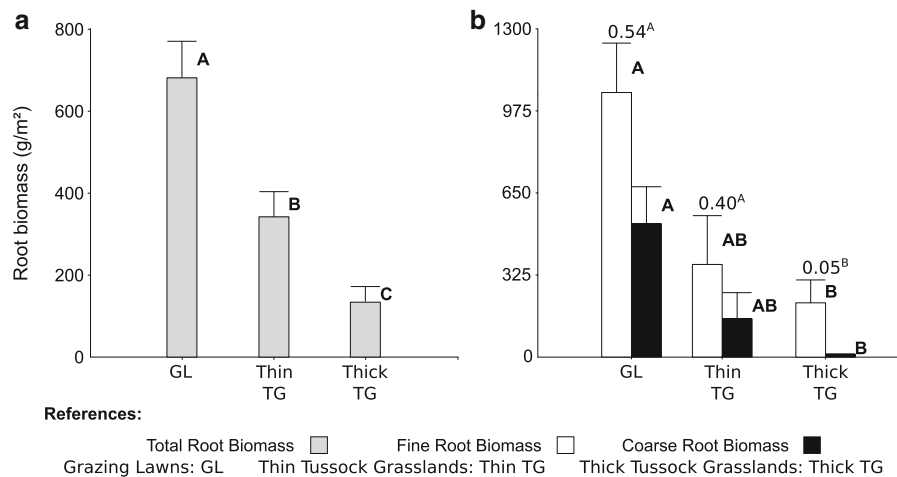
Fig. 2 Root biomass by physiognomy from 0 to 30 cm: **a** total root biomass, **b** coarse and fine root biomass. Different letters represent significant differences between physiognomies for total, coarse, and fine roots separately. Bars represent standard error. Values on top of bars are average coarse:fine roots ratios, and superscript letters indicate significant differences between physiognomies. Total root biomass data were obtained from roots retrieved from previously oven-dried soil samples which allow to maximize sample size ($n = 27$). Fine and coarse root biomass data were obtained from roots retrieved by washing fresh soil samples which minimize fine root loss ($n = 9$)

Table 1 Total, coarse, and fine root biomass and coarse:fine roots ratio for each depth and C:N ratio for the whole soil profile (mean $\pm$ SE)

Physiognomy	Depth (cm)	grazing lawns	Thin tussock grasslands	Thick tussock grasslands
Total root biomass (g/m²)	0–5	305.60 $\pm$ 56.24^A	130.75 $\pm$ 38.30^B	60.15 $\pm$ 35.45^C
	5–15	240.27 $\pm$ 43.74^A	144.84 $\pm$ 26.46^A	44.18 $\pm$ 9.71^B
	15–30	136.72 $\pm$ 35.54^A	68.29 $\pm$ 13.34^A	30.78 $\pm$ 4.14^B
Coarse root biomass (g/m²)	0–5	254.45 $\pm$ 60.35^A	29.98 $\pm$ 19.01^B	⁻4.69 $\pm$ 1.15^B
	5–15	239.88 $\pm$ 87.62^A	40.37 $\pm$ 15.00^{AB}	5.10 $\pm$ 2.06^B
	15–30	34.45 $\pm$ 6.78^A	80.42 $\pm$ 70.62^A	0.45 $\pm$ 0.06^A
Fine root biomass (g/m²)	0–5	436.21 $\pm$ 203.81^A	110.23 $\pm$ 79.16^A	109.54 $\pm$ 61.58^A
	5–15	482.00 $\pm$ 217.48^A	132.90 $\pm$ 83.94^A	74.01 $\pm$ 33.27^A
	5–30	126.59 $\pm$ 23.68^A	120.87 $\pm$ 99.23^{AB}	26.09 $\pm$ 2.76^B
Coarse:fine roots ratio	0–5	0.85 $\pm$ 0.32^A	0.32 $\pm$ 0.04^A	0.06 $\pm$ 0.02^B
	5–15	0.69 $\pm$ 0.39^A	0.54 $\pm$ 0.33^A	0.08 $\pm$ 0.04^A
	15–30	0.30 $\pm$ 0.08^A	0.53 $\pm$ 0.11^A	0.02 $\pm$ 0.0023^B
C:N ratio	0–30	38.80 $\pm$ 4.63^A	36.76 $\pm$ 4.63^A	45.84 $\pm$ 4.63^A

Upper-case superscript letters denote significant differences between physiognomies for each soil depth ($\alpha = 0.05$). Total root biomass data were obtained from roots retrieved from previously oven-dried soil samples which allow to maximize sample size ($n = 27$). The rest of the data was obtained from roots retrieved by washing fresh soil samples which minimize fine root loss ($n = 9$)

Results from supplementary measurements of C in total, coarse, and fine root biomass yielded very similar patterns to those presented for biomass alone (Figure S1 and Table S1).

Coarse:fine roots ratio and root C:N ratio

Fine roots were always more abundant than coarse roots (78% of total root biomass, as it is also shown by the coarse:fine roots ratio values below 1 in all

physiognomies, Fig. 2b). Thick tussock grasslands had an even higher proportion of fine roots than the other two physiognomies ($p \leq 0.0001$). These differences were related to a significantly higher proportion of fine roots in thick tussock grasslands in the first 5 cm ($p = 0.0018$) and in the last 15 cm ($p = 0.0030$), while in between these depths the three physiognomies did not show significant differences (Table 1). Total root C:N ratio (i.e., coarse + fine) for the whole soil profile was not significantly different between physiognomies (Table 1).

Discussion

As it was expected according to previous studies in other grasslands, and supporting our first hypothesis, total root biomass was highest in lawns (physiognomy under more intense local grazing), intermediate in thin tussock grasslands, and lowest in thick tussock grasslands (the last two under less intense or null grazing). Supporting our second hypothesis, absolute fine root biomass was highest in lawns, medium in thin tussock grasslands, and lowest in thick tussock grasslands. However, as opposed to what was expected based on the above-ground attributes of dominant species at each physiognomy, root C:N ratio did not differ between physiognomies and fine root proportion was higher in thick tussock grasslands than in the other vegetation types, for which we partially reject our second hypothesis.

Root biomass

In coincidence with previous work from temperate grasslands of Southamerica (Piñeiro et al. 2010; López-Mársico et al. 2015, 2016; Jaras Zamar 2016), total root biomass was highest in grazing lawns, which are the dominant physiognomy under high disturbance intensity. Also, both the generally higher relative contribution of fine roots compared to coarse roots, and the higher absolute fine root biomass of grazing lawns were coincident with other results in our study region (Pucheta et al. 2004). The root biomass pattern found in our study is opposed to the above-ground biomass and litter patterns found in previous research within the same area (Table 2; Vaieretti et al. 2013). These opposite patterns are also in accordance with what is proposed for grasslands in general (Chapin

et al. 2011; Moretto et al. 2001; Gao et al. 2008). In particular, higher root biomass in lawns could be mainly related to species replacement associated to grazing (Cingolani et al. 2003, 2014), since many lawn species typically present below-ground reserve and reproductive organs like bulbs, rhizomes, and tap roots (Pucheta et al. 2004), which account for a substantial contribution of biomass to below-ground tissues. Additionally, we should have in mind another mechanism that could be operating in grazing lawns, namely the complementarity of soil resource extraction (Kell 2012; Mueller et al. 2013). The higher diversity that characterizes lawns could result in the presence of species with different soil exploration and nutrient uptake strategies, which make use of different soil resource patches. This could be generating the higher root density of lawns, even at 30 cm depth. Lastly, stimulated root growth as a response to above-ground herbivory could be contributing to the formation of this pattern (McNaughton 1983; Shariff et al. 1994; Holland et al. 1996; Frank et al. 2002; Garcia-Pausas et al. 2011; Wilson et al. 2018). This study did not test which one of these mechanisms is operating at a higher degree, but it is possible that all of them are acting simultaneously. Complementary designs and manipulative experiments could contribute to elucidate the importance of these processes in this study system.

Thin tussock grasslands presented intermediate values between the other two physiognomies. It is possible that, even when thin and thick tussock grasslands are both dominated by the same growth form (tussocks sensu Pérez Harguindeguy et al. 2013), species intrinsic differences in attributes could be the cause of their intermediate root biomass values. Thin tussock grasslands are typically dominated by *F. lilloi* and *D. hieronymi* (instead of the much highly dominant *P. stuckertii* from thick tussock grasslands; Cingolani et al. 2003) which are relatively more palatable than *P. stuckertii* and are thus sometimes consumed by herbivores (von Müller et al. 2017), and this may stimulate biomass allocation to roots. Moreover, although at our study sites both thin and thick tussock patches co-occurred, it has been pointed out that thin tussock grasslands do not develop under high soil moisture conditions as thick tussock grasslands do (Cingolani et al. 2014). The more developed root systems in thin tussocks in comparison to thick

Table 2 Above-ground biomass traits from the literature: 1 from Vaieretti et al. ([2013](#)) where thin and thick tussock grasslands are mentioned as open and closed tussock grasslands, respectively (mean ± SE); 2 from Poca et al. ([2014](#)) and 3 from Whitworth-Hulse et al. ([2016](#))

	Grazing lawns	Thin tussock grasslands	Thick tussock grasslands
Annual litterfall (g m^{-2} year^{-1})[1]	62.35 ± 11.34	527.13 ± 55.26	1075.51 ± 111.94
N (%)[1]	1.3 ± 0.07	0.9 ± 0.05	1 ± 0.06
Lignin (%)[1]	9.2 ± 0.43	6.9 ± 0.34	8.2 ± 0.45
Cellulose (%)[1]	33.8 ± 0.69	37.5 ± 0.67	39.1 ± 0.48
Hemicellulose (%)[1]	22.8 ± 1.17	27.7 ± 0.71	27.3 ± 1
C:N[1]	35.7 ± 2.07	50.8 ± 2.75	49.5 ± 0.35
Lignin: N[1]	7.1 ± 0.41	7.5 ± 0.24	8.5 ± 0.35
SLA (mm^2/mg) [2]	15.3	8	6
Ψ_{leaf} (Mpa)[3]	− 4.1	− 3.4	− 2.8

Values from 2 and 3 are means for the dominant species typical of each physiognomy: *M. peruviana*, *L. pinnata*, and *C. fuscula* for grazing lawns and *F. lilloi* and *D. hieronymi* for thin tussock grasslands. Only *P. stuckertii* values were used for thick tussock grasslands

tussocks could then be a useful adaptation for water exploitation in well-drained soils.

The decrease in root biomass with soil depth is a common pattern associated to less favorable conditions in deeper soil layers (e.g., lower nutrient availability and higher soil compaction; Prieto et al. [2015](#)). Additionally, in the highly diverse lawns, interspecific competition could be further decreasing the relative abundance of deep roots by reducing plant vigor to such an extent that plants cannot produce roots deep enough to exploit resources at lower depths (Wardle and Peltzer [2003](#)). As for both thin and thick tussock grasslands, interspecific competition and its effect on plant vigor might be less intense under these physiognomies. This in turn should account for the less steep decrease in below-ground biomass with depth under these vegetation patches.

Coarse:fine roots ratio and root C:N ratio

Coarse:fine roots ratio and total root C:N ratio patterns did not correspond to what was expected in the context of leaf attributes of dominant species from each physiognomy (Table [2](#)). Leaves of species from grazing lawns are palatable and more on the acquisitive end of the spectrum (SLA and N content, lower C:N ratio, and total fiber content) while leaves of species in tussock physiognomies are unpalatable and present traits typical of a more conservative syndrome (Cingolani et al. [2007](#); Vaieretti et al. [2013](#); Poca et al.

[2014](#); Whitworth-Hulse et al. [2016](#)). However, thick tussock grasslands presented a higher proportion of fine roots, which is generally associated to an acquisitive strategy (Lavorel et al. [2007](#)). At the same time, root C:N ratio did not differ between the three physiognomies, suggesting that their root quality was similar, although measurements with a bigger sample size and spanning a wider time lapse are required to make a stronger assertion. This apparent absence of coordination between above and below-ground traits has been already found in other non-palatable grass species (Moretto et al. [2001](#)), and in other plant communities of the world (e.g., Freschet et al. [2015](#)). This outcome may result from diverse above- and below-ground "environmental and evolutionary constraints on plant species-average traits, as well as plastic responses to local environmental conditions" (Freschet et al. [2015](#)). In addition, other authors actually doubt of *P. stuckertii* being classified as a conservative species, considering its high growth rate and water uptake rate, typical of an acquisitive water use syndrome (Cingolani et al. [2015](#); Withworth-Hulse et al. 2016). Finally, in regard to coarse and fine roots characterization, it has been noted that the way these two groups are defined (i.e., according to diameter or functional criteria) can influence the results, especially when comparing different species (McCormack et al. [2015](#); Freschet and Roumet [2017](#)).

Connecting the dots: further considerations

The contrasting amounts of root biomass in each physiognomy, as well as the different proportions of fine root biomass, could have different implications on ecosystem processes at our study site. One aspect of ecosystem functioning that should be further studied is the annual dynamics in root biomass. Within the study area, Pucheta et al. (2004) found a decrease in root biomass values in winter compared to summer, especially at grazing lawns, but this was only measured in two plots. A better characterization of root biomass seasonal changes, as well as seasonal changes in root traits, is thus required to better understand below-ground biomass dynamics. Our findings may also have implications in relation to soil C dynamics, because above- and below-ground biomass at each physiognomy and their turnover make up contrasting amounts of C inputs to soils. Root derived C (both from biomass turnover and from rhizodeposition) would expectedly be higher in lawns than in tussock physiognomies (Figure S2), while the opposite is true for leaf derived C (Vaieretti et al. 2013). How this affects soil processes will depend mainly on the carbon use efficiency of the soil microbiota and the stabilization capacity of the soil matrix (Cotrufo et al. 2013; Oldfield et al. 2018), but it is yet to be tested. In relation to this, it has been proposed that the input of more labile litter may stimulate soil organic matter stabilization (Cotrufo et al. 2013, 2015). Grazing lawns seem to be receiving more decomposable leaf litter, but roots do not seem to be different in quality according to their C:N ratio (unless we consider their higher absolute amounts of fine root biomass compared to tussock grasslands). How this changes in leaf and root quality may affect C respiration and stabilization also remains to be explored.

References

Amundson R (2001) The carbon budget in soils. Annu Rev Earth Planet Sci 29:535–562

Asner GP, Elmore AJ, Olander LP, Martin RE, Harris AT (2004) Grazing systems, ecosystem responses, and global change. Annu Rev Environ Resour 29:261–299

Bardgett RD, Wardle DA (2003) Herbivore-mediated linkages between above-ground and below-ground communities. Ecology 84:2258–2268

Bardgett RD, Mommer L, De Vries FT (2014) Going underground: root traits as drivers of ecosystem processes. Trends Ecol Evol 29(12):692–699

Cabido M, Breimer R, Vega G (1987) Plant communities and associated soil types in a high plateau of the Córdoba mountains, central Argentina. Mt Res Dev 7:25–42

Chapin FS III, Matson PA, Vitousek PM (2011) Principles of terrestrial ecosystem ecology. Springer, New York

Cingolani AM, Cabido MR, Renison D, Solís Neffa V (2003) Combined effects of environment and grazing on vegetation structure in Argentine granite grasslands. J Veg Sci 14:223–232

Cingolani AM, Renison D, Zak MR, Cabido M (2004) Mapping vegetation in a heterogeneous mountain rangeland using landsat data: an alternative method to define and classify land-cover units. Remote Sens Environ 92:84–97

Cingolani AM, Cabido M, Gurvich DE, Renison D, Díaz S (2007) Filtering processes in the assembly of plant communities: are species presence and abundance driven by the same traits? J Veg Sci 18:911–920

Cingolani AM, Noy-Meir I, Renison D, Cabido M (2008) La ganadería extensiva, ¿es compatible con la conservación de la biodiversidad y de los suelos? Ecología Austral 18:253–271

Cingolani AM, Vaieretti MV, Giorgis MA, Poca M, Tecco PA, Gurvich DE (2014) Can livestock grazing maintain landscape diversity and stability in an ecosystem that evolved with wild herbivores? Perspect Plant Ecol 16:143–153

Cingolani AM, Poca M, Giorgis MA, Vaieretti MV, Gurvich DE, Whitworth-Hulse JI, Renison D (2015) Water provisioning services in a seasonally dry subtropical mountain: identifying priority landscapes for conservation. J Hydrol 525:178–187

Colladon L, Felici GS, Pazos I (2010) Anuario pluviométrico 2005–2010. Cuenca del Río San Antonio. Sistema del Río Suquía - Provincia de Córdoba. Instituto Nacional del Agua y del Ambienten (INA) y Centro de Investigaciones de la Región Semiárida (CIRSA), Córdoba

Cotrufo MF, Wallenstein MD, Boot CM, Denef K, Paul E (2013) The Microbial Efficiency-Matrix Stabilization (MEMS) framework integrates plant litter decomposition with soil organic matter stabilization: do labile plant inputs form stable soil organic matter? Glob Change Biol 19(4):988–995

Cotrufo MF, Soong JL, Horton AJ, Campbell EE, Haddix ML, Wall DH, Parton WJ (2015) Formation of soil organic

matter via biochemical and physical pathways of litter mass loss. Nat Geosci 8:776–779

Derner JD, Boutton TW, Briske DD (2006) Grazing and ecosystem carbon storage in the North American Great Plains. Plant Soil 280:77–90

Di Rienzo JA, Casanoves F, Balzarini MG, Gonzalez L, Tablada M, Robledo CW (2017) InfoStat versión 2017. Grupo InfoStat, FCA, Universidad Nacional de Córdoba, Argentina. https://www.infostat.com.ar. Accessed April 2017

Díaz S, Lavorel S, McIntyre S, Falczuk V, Casanoves F, Milchunas DG, Skarpe C, Rusch G, Sternberg M, Noy-Meir I, Landsberg J, Zhang W, Clark H, Campbell BD (2007) Plant trait responses to grazing—a global synthesis. Glob Change Biol 12:1–29

FAO (2016) FAOSTAT Emissions Database. https://faostat3.fao.org/home/E12. Accessed June 2016

Frank DA, Kuns MM, Guido DR (2002) Consumer control of grassland plant production. Ecology 83(3):602–606

Freschet GT, Roumet C (2017) Sampling roots to capture plant and soil functions. Funct Ecol 31(8):1506–1518

Freschet GT, Cornwel WK, Wardle DA, Elumeeva TG, Liu W, Jackson BG, Onipchenko VG, Soudzilovskai NA, Tao J, Cornelissen JHC (2013) Linking litter decomposition of above- and below-ground organs to plant–soil feedbacks worldwide. J Ecol 101:943–952

Freschet GT, Kichenin E, Wardle DA (2015) Explaining within-community variation in plant biomass allocation: a balance between organ biomass and morphology above vs below-ground? J Veg Sci 26(3):431–440

Gang C, Zhou W, Chen Y, Wang Z, Sun Z, Li J, Qi J, Odehet I (2014) Quantitative assessment of the contributions of climate change and human activities on global grassland degradation. Environ Earth Sci 72:4273–4282

Gao YZ, Giese M, Lin S, Sattelmacher B, Zhao Y, Brueck H (2008) below-ground net primary productivity and biomass allocation of a grassland in Inner Mongolia is affected by grazing intensity. Plant Soil 307:41–50

Garcia-Pausas J, Casals P, Romanyà J, Vallecillo S, Sebastià MT (2011) Seasonal patterns of below-ground biomass and productivity in mountain grasslands in the Pyrenees. Plant Soil 340:315–326

Gill R, Burke IC, Milchunas DG, Lauenroth WK (1999) Relationship between root biomass and soil organic matter pools in the shortgrass steppe of eastern Colorado. Ecosystems 2:226–236

Hamilton EW III, Frank DA, Hinchey PM, Murray TR (2008) Defoliation induces root exudation and triggers positive rhizospheric feedbacks in a temperate grassland. Soil Biol Biochem 40:2865–2873

Holland JN, Cheng W, Crossley DA Jr (1996) Herbivore-induced changes in plant carbon allocation: assessment of below-ground carbon fluxes using carbon-14. Oecologia 107:87–94

Hu Y, Zeng D, Ma X, Chang S (2016) Root rather than leaf litter input drives soil carbon sequestration after afforestation on a marginal cropland. For Ecol Manage 362:38–45

Hungate BA, Holland EA, Jackson RB, Chapin FS III, Mooneyk HA, Field CB (1997) The fate of carbon in grasslands under carbon dioxide enrichment. Nature 388(7):576–579

Jaras Zamar DJ (2016) Efecto del pastoreo sobre los reservorios y flujos de carbono en pastizales de altura en las Sierras Grandes de Córdoba. Graduate thesis, University of Córdoba

Kell DB (2012) Large-scale sequestration of atmospheric carbon via plant roots in natural and agricultural ecosystems: why and how. Phil Trans R Soc B 367:1589–1597

King AW, Post WM, Wullschleger SD (1997) The potential response of terrestrial carbon storage to changes in climate and atmospheric CO_2. Clim Change 35:199–227

Lavorel S, Díaz S, Cornelissen JHC, Garnier E, Harrison SP, McIntyre S, Pausas JG, Pérez-Harguindeguy N, Roumet C, Urcelay C (2007) Plant functional types: are we getting any closer to the holy grail? In: Canadell JG, Pataki D, Pitelka L (eds) Terrestrial ecosystems in a changing world global change—The IGBP series. Springer, Berlin, pp 149–164

López-Mársico L, Altesor A, Oyarzabal M, Baldassini P, Paruelo JM (2015) Grazing increases below-ground biomass and net primary production in a temperate grassland. Plant Soil 392(1–2):155–162

López-Mársico L, Oyarzabal M, Altesor A, Paruelo JM (2016) Efecto del pastoreo sobre la productividad primaria neta subterránea de un pastizal templado. Ecología Austral 26:256–263

Mambelli S, Bird JA, Gleixner G, Dawson TE, Torn MS (2011) Relative contribution of foliar and fine root pine litter to the molecular composition of soil organic matter after in situ degradation. Org Geochem 42:1099–1108

McCormack ML, Dickie IA, Eissenstat DM, Fahey TJ, Fernandez CW, Guo D, Helmisaari H, Hobbie EA, Iversen CM, Jackson RB, Leppälammi-Kujansuu J, Norby RJ, Phillips RP, Pregitzer KS, Pritchard SG, Rewald B, Zadworny M (2015) Redefining fine roots improves understanding of below-ground contributions to terrestrial biosphere processes. New Phytol 207:505–518

McNaughton SJ (1983) Compensatory plant growth as a response to herbivory. Oikos 40:329–336

Moretto AS, Distel RA, Didoné NG (2001) Decomposition and nutrient dynamic of leaf litter and roots from palatable and unpalatable grasses in a semi-arid grassland. Appl Soil Ecol 18:31–37

Mueller KE, Tilman D, Fornara DA, Hobbie SE (2013) Root depth distribution and the diversity–productivity relationship in a long-term grassland experiment. Ecology 94:787–793

Oldfield EE, Crowther TW, Bradford MA (2018) Substrate identity and amount overwhelm temperature effects on soil carbon formation. Soil Biol Biochem 124:218–226

Pérez Harguindeguy N, Díaz S, Garnier E, Lavorel S, Poorter H, Jaureguiberry P, Bret-Harte MS, Cornwell WK, Craine JM, Gurvich DE, Urcelay C, Veneklaas EJ, Reich PB, Poorter L, Wright IJ, Ray P, Enrico L, Pausas JG, de Vos AC, Buchmann N, Funes G, Quétier F, Hodgson JG, Thompson K, Morgan HD, ter Steege H, van der Heijden MGA, Sack L, Blonder B, Poschlod P, Vaieretti MV, Conti G, Staver AC, Aquino S, Cornelissen JHC (2013) New handbook for standardised measurement of plant functional traits worldwide. Aust J Bot 61:167–234

Piñeiro G, Paruelo JM, Oesterheld M, Jobbágy EG (2010) Pathways of grazing effects on soil organic carbon and nitrogen. Rangel Ecol Manage 63:109–119

Pinheiro JC, Bates DM (2000) Mixed-effects models in S and S-PLUS. Springer, New York

Poca M, Pérez-Harguindeguy N, Vaieretti MV, Cingolani AM (2014) Descomposición y calidad físico-química foliar de 24 especies dominantes de los pastizales de altura de las sierras de Córdoba, Argentina. Ecología Austral 24:249–257

Poca M, Vaieretti MV, Cingolani AM, Pérez-Harguindeguy N (2015) Scaling-up from species to ecosystems: how close can we get to actual decomposition? Acta Oecol 64:1–9

Prieto I, Roumet C, Cardinael R, Dupraz C, Jourdan C, Kim JH, Maeght JL, Mao Z, Pierret A, Portillo N, Roupsard O, Thammahacksa C, Stokes A (2015) Root functional parameters along a land-use gradient: evidence of a community-level economics spectrum. J Ecol 10:361–373

Pucheta E, Vendramini F, Cabido M, Díaz S (1998) Estructura y funcionamiento de un pastizal de montaña bajo pastoreo y su respuesta luego de su exclusión. Revista de la Facultad de Agronomía 103(1):77–92

Pucheta E, Bonamici I, Cabido M, Díaz S (2004) Below-ground biomass and productivity of a grazed site and a neighbouring ungrazed exclosure in a grassland in central Argentina. Austral Ecol 29:201–208

Rasse DP, Rumpel C, Dignac MF (2005) Is soil carbon mostly root carbon? Mechanisms for a specific stabilisation. Plant Soil 269:341–356

Roumet C, Birouste M, Picon-Cochard C, Ghestem M, Osman N, Vrignon-Brenas S, Cao K, Stokes A (2016) Root structure–function relationships in 74 species: evidence of a root economics spectrum related to carbon economy. New Phytol 210(3):815–826

Shahbaz M, Kuzyakov Y, Heitkamp F (2016) Decrease of soil organic matter stabilization with increasing inputs: mechanisms and controls. Geoderma 304:76–82

Shariff AR, Biondini ME, Grygiel CE (1994) Grazing intensity effects on litter decomposition and soil nitrogen mineralization. J Range Manage 47:444–449

Vaieretti MV, Cingolani AM, Pérez-Harguindeguy N, Gurvich DE, Cabido M (2010) Does decomposition of standard materials differ among grassland patches maintained by livestock? Austral Ecol 35(8):935–943

Vaieretti MV, Cingolani AM, Pérez-Harguindeguy N, Cabido M (2013) Effects of differential grazing on decomposition rate and nitrogen availability in a productive mountain grassland. Plant Soil 371:675–691

Vaieretti MV, Iamamoto S, Pérez-Harguindeguy N, Cingolani AM (2018) Livestock grazing affects microclimate conditions for decomposition process through changes in vegetation structure in mountain grasslands. Acta Oecol 91:101–107

von Müller AR, Renison D, Cingolani AM (2017) Cattle landscape selectivity is influenced by ecological and management factors in a heterogeneous mountain rangeland. Rangel J 39:1–14

Wardle DA, Peltzer DA (2003) Interspecific interactions and biomass allocation among grassland plant species. Oikos 100:497–506

Whitworth-Hulse JI, Cingolani AM, Zeballos SR, Poca M, Gurvich DE (2016) Does grazing induce intraspecific trait variation in plants from a sub-humid mountain ecosystem? Austral Ecol 41(7):745–755

Wilson SD (2014) Below-ground opportunities in vegetation science. J Veg Sci 25:1117–1125

Wilson CH, Strickland MS, Hutchings JA, Bianchi TS, Flory SL (2018) Grazing enhances below-ground carbon allocation, microbial biomass, and soil carbon in a subtropical grassland. Glob Change Biol. https://doi.org/10.1111/gcb.14070

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