

# Fertilization, irrigation and pasture sown to improve herbage production and quality in *Nothofagus antarctica* silvopastoral systems in southern Patagonia

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**Abstract** *Nothofagus antarctica* forest extends from 46 to 56° south latitude and its main use has been as silvopastoral systems (SPS). Several studies have characterized understory of *N. antarctica* SPS. However, other practices like pasture sowing, irrigation and fertilization, to improve understory yield, remains little documented. The aim of this work was to compare dry matter (DM) production and quality of natural understory vs. implanted pastures under different

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R. Christiansen INTA EEA Santa Cruz, Río Turbio, Santa Cruz, Argentina water and nutrient conditions. An experiment with fifteen plots of  $6 \times 6$  m was established in a strip-split plot design, with species as the main factor, water as the sub-plot factor (rainfed vs. irrigated) and fertilizer application level as sub-sub-plot factor (low, medium and high). The species evaluated were: Dactylis glomerata, Bromus catharticus, Trifolium pratense, T. repens and natural understory. Total dry matter (DM) production, leaf N concentration and digestibility were measured after 1 and 2 years of experiment establishment. Significant differences were found in DM according to species and fertilization treatments. The highest DM production was for T. pratense with medium fertilization level (5194 kg DM ha<sup>-1</sup>), followed by D. glomerata with 200 kg N ha<sup>-1</sup> (4984 kg DM ha<sup>-1</sup>). Natural understory increased DM with nitrogen fertilization from 1427 to 3980 kg DM  $ha^{-1}$ . In terms of quality, T. repens had the highest digestibility values. This study showed that pure pastures of T. pratense or D. glomerata accompanied by N fertilization are viable options to increase forage yield and quality in N. antarctica SPS.

**Keywords** Agroforestry · *Dactylis glomerata* · Native forest · ñire

### Introduction

Native *Nothofagus antarctica* (ñire) forest extends from 46 to 56° SL in Patagonia region of Argentina

and Chile. Most of these forests have been used as silvopastoral systems (SPS), where natural grasses growing under the tree canopy are grazed by cattle and sheep (Collado 2009; Peri and Ormaechea 2013). There are several studies that reported about quality and production of the understory stratum, animal performance and regeneration and production of N. antarctica trees under SPS (Peri et al. 2016). However, other topics such as fertilization, irrigation or pasture sowing to improve understory herbage production, remains little documented. It is known that understory dry matter (DM) production decreases if light intensity decreases (Devkota and Kemp 1999) and have been reported that in N. antarctica silvopastoral systems understory production largely depends on the interaction between soil water availability and light intensity (Peri 2005; Peri et al 2016). Additionally, Gargaglione et al. (2014) reported that in good site qualities, understory DM production of natural grasses is lower under tree canopy than under open condition (Gargaglione et al. 2014). This is consistent with other studies that showed a negative exponential relationship between herbage DM production and biomass, crown cover or basal area of trees (Scholes and Archer 1997; Peri et al. 2009). This aspect could be improved by increasing the proportion of shade tolerant species. Pang et al. (2019) evaluated 22 species of temperate forages and reported that most of them maintained yield and crude protein content under moderate and dense shade, (Pang et al. 2019). Devkota and Kemp (1999) concluded that successful and productive silvopastoral systems depended on well-adapted, shade-tolerant forage species. Thus, the introduction of shade-tolerant grass and legume species in the appropriate mixture increases herbage production and nutritive value (Muñoz and Weaver 1999) because of the ability of the legumes to fix atmospheric nitrogen (Ledgard 1991).

In the context of climate change, there is a tendency to intensify systems to achieve higher yields per unit area by implementing various strategies such as fertilization and irrigation to increase productivity. Consequently, there is a need to enhance understory production in ñire silvopastoral systems (SPS), either through the introduction of shade-adapted species or other methods such as fertilization or irrigation. Among the species commonly known for their shade tolerance are orchard grass (*Dactylis glomerata*), barley (Bromus catharticus), and red clover (Trifolium repens) (Maddaloni and Ferrari 2001; Devkota et al. 1997; Peri et al. 2007; Kyriazopoulos et al. 2013). These species have already been tested in the region with good results in implementation and production, but only in steppe areas (Christiansen et al. 2007). Additionally, although some native legumes grow in ñire understory (like Vicia sativa), their contribution to vegetation cover is very low, typically around 5% (Gargaglione and Peri 2015). Incorporating legumes into the understory of SPS may enhance site fertility by fixing atmospheric nitrogen (N), thereby increasing fertility and enhancing digestible dry matter for livestock consumption. In ñire forests, like many other natural systems, N is the most limiting nutrient source (Diehl et al. 2008) particularly during spring when low temperatures restrict N mineralization (Bahamonde et al. 2012). In this sense, another approach to enhance herbage DM production and quality could involve the application of fertilizer, although there is a scarcity of studies evaluating the extent of DM response to fertilizers in ñire SPS. Furthermore, it is well-known that N. antarctica can thrive under a wide range of environmental conditions, from temporarily flooded soils with high precipitation to drier sites, bordering the Patagonian steppe (Veblen et al. 1996). It is also well documented that understory DM production in these SPS is positively correlated with water availability (Peri 2005; Peri et al. 2016). Therefore, including treatments that enhance available water could provide insights into wetter sites compared to those presented in the current study. The aim of this study was to compare dry matter (DM) production and quality between natural understory and sown pastures of grasses and legumes in young N. antarctica silvopastoral systems under varying water availability and nutrient conditions.

### Material and methods

#### Study site

The site is located at Cancha Carrera ranch  $(51^{\circ} 13' 21'' \text{ S}, 72^{\circ} 15' 34'' \text{ W})$  in the southwest of Santa Cruz province, Southern Patagonia, Argentina. This study was carried out in a naturally regenerated *N. antarctica* young forest  $(41 \pm 6 \text{ years old})$  with an original density of 5820 tree ha<sup>-1</sup>, averaging 9 cm in diameter at breast height and a mean total height of

8 m. The natural understory layer exhibited 80-100% vegetation cover, dominated by grasses. The climate is cold temperate, with a mean annual temperature of 6 °C and mean annual rainfall of 379 mm.

# Environmental measurements

Precipitation was recorded by an automatic agrometeorological station (Davis MB5 LR Vantage Pro 2 wireless), located at 51° 31' 58, 77" S; 72° 15' 45,35" W. Additionally, air and soil temperatures were measured continuously every 2 h with a data logging system (HOBOH8 Family, Onset Computer Corporation, USA). Air temperature sensors were positioned at 0.6 m above ground level, while soil temperature was measured at a depth of 3 cm using soil thermometers (HOBO, Model TMC50-HA, USA). The site's soils are classified as Mollisols, with a depth of 60 cm and a pH about 4.7. Prior to establishing the study, six soil samples were randomly taken from plots (to a depth of 20 cm) to characterize the soil. The samples were air dried and transported to the laboratory for analysis: (i) organic carbon (C) was determined by spectrophotometry according to Kumies after wet oxidation in acid medium (Houba et al. 1988); (ii) total nitrogen content (N) was determined using the semi-micro Kjeldahl method (Sparks 1996); (iii) available phosphorus (P) content was assessed using the Olsen method; (iv) exchangeable potassium (K) was determined by saturation with sodium acetate, washed with ethylic alcohol, and displacement through pH 7 buffered ammonium acetate. Soil pH was obtained by potentiometric measurement in a water saturated paste.

### Natural understory evaluation

The botanical composition of the natural understory was evaluated in two 6 m long transects located in each plot of  $6 \times 6$  m (n=3). In each transect, species cover was measured by the interception method (Levy and Madden 1933), recording all plant species, litter or bare soil, every 10 cm interval. Using these data, the percentage of vegetation cover, species richness, and Shannon's diversity index were estimated.

Experiment design and pasture installation

For the establishment of the SPS, a homogeneous forest area of 0.4 ha was thinned by reducing tree density from 2300 to 1000 tree ha<sup>-1</sup>, which represented a crown cover of around 50%. The site was fenced to prevent grazing in the SPS. In this thinned area, fifteen plots of  $6 \times 6$  m were established in a strip-split plot design (n=3), with species as the main factor (randomly assigned), water as the sub-plot factor ( $6 \times 3$  m) and fertilizer application level as sub-subplot factor (Fig. 1). The pasture species evaluated were: *Dactylis glomerata* var. Porto (DG), *Bromus catharticus* var. Fierro Plus INTA (BR), *Trifolium pratense* var. Quiñequeli (TP), *Trifolium repens* var. Nimbus (TR), and natural understory (NU).

Before sowing the pastures, the natural sward (standing vegetation, rhizomes and roots) was removed by tilling the soil using a roto tiller and a rake. Plots were sown with *D. glomerata* at 12 kg ha<sup>-1</sup>, *B. catharticus* and *T. pratense* at 15 kg ha<sup>-1</sup> and *T. repens* at 10 kg ha<sup>-1</sup>. Forage seeds of each species were tested for viability through a germination test. This test involved placing 100 seeds in trays with moistened cotton using distilled water. The trays were placed in a germination chamber (INGELAB, model I-502 PF) at 25 °C and 16 h of light. The number of germinated seeds was counted



Fig. 1 Schematic diagram of the  $6 \times 6$  m plots established for the experiment. Species (main factor) was assigned randomly. Water treatments (irrigated and rainfed) and fertilization levels (low, medium and high) were applied in a strip plot design within each plot. Initials correspond to: DG: *Dactylis glomerata*; Br: *Bromus catharticus*; Tr: *Trifolium repens*; TP: *Trifolium pratensis* and NU: *Natural understory*. Numbers indicate replicates

daily for a period of 15 days. The germination percentages were: 60, 80, 82 and 84% for DG, BR, TP and TR, respectively.

Seed and fertilizer were broadcast by hand onto the plots in September 2011 (early spring). The water treatment consisted in a control and irrigation of 90 mm during the growing season (December to March), applied manually every 12 days. Fertilizer levels corresponded to 0, 100 and 200 kg de N ha<sup>-1</sup> for grass plots (DG, Br and NU) applied as urea, and 0, 50 and 100 kg Pha<sup>-1</sup> for legume plots, applied as superphosphate. Fertilizer treatments were repeated once a year during spring (October) for 2 years. Additionally, three plots with natural understory with the same water and fertilizer treatments were evaluated.

# Measurements and data analysis

Understory DM production and digestibility were measured once a year at the growth peak (late January) by clipping biomass within a  $0.1 \text{ m}^2$ quadrant after one (2013) and two years (2014) of experiment establishment. Collected material was sorted into green, senescent and flower components. Then, the material was oven dried at 60 °C to obtain dry weight. DM production was calculated as green leaves plus florescence. Subsamples of 20 g of green leaves were sent to the Forage Laboratory of INTA Balcarce (Buenos Aires, Argentina) for in vitro digestibility analysis. In addition, forage quality was evaluated by studying foliar N concentrations in a 20 g subsample sent to LANAQUI laboratory (Universidad Nacional del Sur, Bahía Blanca, Buenos Aires) for total nitrogen analysis with Kjeldahl method.

Aboveground dry matter production, Ν concentration and digestibility were analyzed with mixed effects models with normal error distribution (lme in nlme package in R; Pinheiro et al. 2022). Mixed-effects models considered repeated measurements (2 years) nested in subplots and subplots nested in plots. Water level, fertilization level, the species and the interactions were the predictors. The homogeneity of the variances was evaluated by residual plots as a function of adjusted values and predictors. When necessary, variance functions for groups (verIdent) were applied. The normal distribution of the residuals was evaluated using quantile theoretical distribution plots (Q–Q plot). Inferences about each model were made using the Anova function (car package, Fox and Weisberg 2011). When main predictors or the interactions were significant, least squares mean adjusted by "sidak" and *p*-value <0.05 were used to compare means between levels of factors (emmeans function in emmeans package; Lenth 2017). The conditional and marginal coefficients of determination (R2c and R2m, respectively) were obtained from r.squared GLMM function (MuMIn package Barton 2016. These analyses were made in the R-cran environment, version 3.6.1 (R Core Team, 2022) and with RStudio; version 1.2.1335 (RStudio Team 2020).

# Results

# Environment conditions

Total rainfall in both years of the study exceeded the historical average of 379 mm, with 424 and 400 mm per year for 2013 and 2014, respectively. Mean air temperatures during the growing season were 10.2 and 10.7 °C for 2013 and 2014, respectively, while the mean soil temperature averaged around 9.9 °C in both years (Fig. 2). Despite similar mean air temperature values between years, 2013 experienced more days with air temperatures above 16–18 °C (Fig. 2A) compared with 2014 (Fig. 2B).

Soils were slightly acidic without salinity problems and with intermediate values of N, C and P (Table 1).

# Natural understory

Natural SPS understory had a vegetation cover of 95–98%, being the most abundant species: *Poa pratensis* (41%) *Dactylis glomerata* (17%), *Taraxacum officinale* (14%), *Osmorhiza chilensis* (13%) and *Gallium aparine* (Table 2). Mean species richness and Shannon diversity index were 16 and -1.7, respectively (Fig. 3).

Water and fertilizer application had little effect on vegetation cover (Fig. 3A), although Shannon diversity index decreased from -1.7 (control) to -1.44 with the 200 kg N ha<sup>-1</sup> treatment (Fig. 3B). Similarly, the relative cover of understory species showed some variations according to water and fertilizer treatments. For example, *P. pratensis* decreased in vegetation



Fig. 2 Mean daily air (solid lines) and soil (doted lines) temperatures of the *N. antarctica* silvopastoral system studied in south Patagonia, after the first year 2013 (A) and the second year 2014 (B) of the experiment establishment

Table 1 Mean soil characteristics of the study site previous to the experiment installation, in south Patagonia, Argentina

C (%)	N (%)	C:N	P (ppm)	K (meq/100 g)	pH	Salinity (mmhos/cm)
7.13 (2.65)	0.53 (0.16)	13.93 (4.73)	23.96 (0.16)	0.25 (0.001)	5.7 (0.28)	0.54 (0.16)

In parenthesis standard deviation of the mean

**Table 2** Species composition and relative cover in natural understory of *N. antarctica* silvopastoral system in south Patagonia, Argentina

Specie	Relative
	(%)
Poa pratensis	41.3
Dactylis glomerata	16.8
Taraxacum Officinale	14
Osmorhiza chilensis	12.7
Trisetum spicatum	1.7
Gallium aparine	2.7
Deschampsia flexuosa	1.3
Gallium fuegianum	0.7
Cerastium Fontanum	0.5
Calciolaria biflora	0.3
Viola magallanica	0.6
Vicia sativa	0.8
Agrostis tenuis	1.1
Bromus unioloides	0.6
Rumex acetocella	0.1
Acaena magellanica	0.3

cover with increased N, from 41 to 24%, while *D. glomerata* increased from 16 to 43% with fertilization and irrigation. Other species did not show changes in relative cover related to irrigation or fertilization treatments.

#### Forage implantation and yield

Except for *Bromus catharticus*, all species were successfully established with a vegetation cover of 80–90%. Significant differences in DM production were found among species and fertilization, with a significant interaction between treatments (Table 3). In contrast, no significant differences were observed with respect to water treatments (Table 3). The highest DM production was for *T. pratense* at 50 kg P ha<sup>-1</sup> with a mean value of 5194 kg DM ha<sup>-1</sup> (Table 4), followed by *D. glomerata* at 200 kg N ha<sup>-1</sup> with 4983 kg DM ha<sup>-1</sup> (Fig. 4). Plots without fertilization in *D. glomerata* and natural understory showed the lowest DM accumulation, with 1386 and 1427 kg DM ha<sup>-1</sup>,

**Fig. 3** A Vegetation cover and **B** Shannon index diversity of natural understory of *N.antartica* silvopastoral system in south Patagonia in response to water irrigation (90 mm added, grey bars) and fertilizer application (0, 100 and 200 kg N ha<sup>-1</sup>)



**Table 3** Mixed effects model analysis of dry matter production, digestibility, and total nitrogen comparing three forage species (*T. repens, T. pratense* and *D. glomerata*) and natural understory under two water levels (irrigated and control) and

three fertilizer levels (low, medium, high) in a *N. antarctica* silvopastoral system in southern Patagonia, Argentina

	df	Dry matter (kg/ha)		Digestibility (%)		Total nitrogen (%)	
		Chisq	p value	Chisq	p value	Chisq	p value
Species	3	39.01	< 0.0001	635.83	< 0.0001	236.45	< 0.0001
Water level	1	3.22	0.073	4.55	0.033	6.68	0.0097
Fertilizer level	2	54.50	< 0.0001	3.42	0.181	19.85	< 0.0001
Water* species	3	1.64	0.651	4.43	0.218	3.28	0.349
Water * fertilizer	2	0.80	0.669	3.94	0.139	2.46	0.291
Species* fertilizer	6	36.55	< 0.0001	7.39	0.286	42.35	< 0.0001
Water* species *fertilizer	6	3.66	0.723	9.63	0.141	4.34	0.630

Columns show the degree of freedom (df), the statistic Chi squared (Chisq) and the associated probability (*p*-value). Numbers in bold indicated significant effects

**Table 4** Mean values of dry matter production (Kg DM  $ha^{-1}$ ) of pastures implanted (*D. glomerata*, *T. pratense* and *T. repens*) and natural understory and three fertilization levels: low (con-

trol=0), medium and high, in a *N. antarctica* silvopastoral system in southern Patagonia, Argentina

Species	Fertilizer level					
	Low	Medium	High			
Natural understory	1427 (-15.9; 2870) a	3397 (1488.8; 5304.9) bc	3980 (2151.7; 5808.6) bc			
Dactylis glomerata	1386 (-248.8; 3056.5) a	4622 (2156.3; 7088.2) bc	4984 (2648.2; 7318.7) bc			
Trifolium repens	1797 (319.9; 3273.5) ab	2223 (696.9; 3750.0) ab	2348 (830.9; 3865.3) ab			
Trifolium pratense	4922 (2700.9; 7143.3) bc	5194 (806.1;9581.4) bc	4609 (544.3; 8673.3) bc			

Different letters indicate significant differences according to models presented in Table 3. Lower and upper confidence intervals of the GMM model are in parenthesis

respectively (Table 4). Senescent components were not included in the DM estimations, and they represented 11%; 17%; 20% and 23% of the total DM for *T. pratense, natural understory, D. glomerata* and *T. repens*, respectively.

#### **Forage quality**

Significant differences were found (p < 0.05) in leaf N concentration according to species, water and fertilization, with some significant interactions between treatments (Table 3). *D. glomerata* and

**Fig. 4** Mean annual biomass production (kg DM ha<sup>-1</sup> yr<sup>-1</sup>) of pastures implanted (*D. glomerata, T. pratense* and *T. repens*) and natural understory in a *N. antarctica* silvopastoral system (southern Patagonia, Argentina) and three fertilization levels: low (control=0), medium and high. Vertical bars indicate standard deviation of the means

Fig. 5 Mean values of leaves N concentration for D. glomerata, T. repens, T. pratense and the natural understory at the growth peak (January) in response to three levels of fertilization (control = 0, medium and high) in a N. antarctica silvopastoral system. Vertical bars indicate the standard deviation of the mean. Different letters indicate significant differences between species and fertilization



natural understory with no fertilization presented the lowest values of % N with 1.32 and 1.46%, respectively, while both legumes showed the highest values (Fig. 5). Likewise, natural understory and *D. glomerata* significantly increased their leaf N concentration as fertilizer levels increased to 2.33 and 2.40%, respectively (Fig. 5). In contrast, no differences were found in legumes with fertilization. Overall, irrigation treatments tended to result in higher N concentrations, which was more pronounced in legumes (Fig. 5).

Significant differences (p < 0.05) were found in in vitro dry matter digestibility according to specie and water treatments, although no significant differences were observed based on fertilizer levels (Table 3). *Trifolium repens* accounted with the highest digestibility values (80.8%), meanwhile all other

**Table 5** Mean in vitro dry matter (DM) digestibility values in peak of biomass (late January) of *Dactylis glomerata, T. repens, T. pratense* and natural understory in a *N. antarctica* silvopastoral system in southern Patagonia (Argentina), under two water treatments

Digestibility (%)			
Specie	Mean		
Dactylis glomerata	69.5 a		
Natural understory	70.6 a		
Trifolium pratense	70.5 a		
Trifolium repens	80.8 b		
Water treatment			
Irrigation	73.5 a		
Control	72.3 ab		

Different letters indicate significant differences

species had digestibility values around 69.5 and 70.6% (Table 5).

# Discussion

#### Natural understory

The natural understory of this *N. antarctica* SPS includes exotic grasses such as *Dactylis glomerata*, *Poa pratensis* and others dicotyledonous species like *Taraxacum officinale*, probably introduced in the past by the ranchers. The native *Osmorhiza chilensis* accounted for 12% of vegetation cover with little variation between applied treatments. The botanical composition observed in this study contrasts with that reported by Peri et al. (2016), who identified dominant species at a regional level in ñire SPS represented predominantly by *Carex andina* (20–30%), along with *Poa pratensis, Phleum commutatum, Dactylis glomerata, Bromus setifolius, Agrostis flavidula, Deschampsia, flexuosa* and *Festuca pallescens*.

Water and fertilizer application decreased the Shannon diversity index of the understory, mainly due to an increase in *D. glomerata* cover. The biomass production observed in this study for natural understory in control plots  $(1427 \text{ kgha}^{-1})$  falls withing the range reported by other authors for ñire forests in south Patagonia (Bahamonde et al. 2012; Peri et al. 2016). Understory DM production in ñire SPS depends on the interaction between soil

water availability and light intensity reaching the soil. Under moderate water stress conditions, DM production decreases from 2800 kg ha<sup>-1</sup> in open sites (100% light transmittance) to 500 kg kg ha<sup>-1</sup> in severe shade (Peri et al. 2016).

In this study, we found that fertilizer application produced significant changes in natural understory biomass accumulation, increasing from 1427 to 3980 kg DM ha<sup>-1</sup>. This is consistent with Dhiel et al. (2008) and Gargaglione et al. (2014), who reported that N is the most limiting nutrient in these Patagonian forests. In addition, our study found no significant differences in biomass accumulation between the 100 and 200 kg N ha<sup>-1</sup> fertilization levels, suggesting that a substantial increase in natural forage yield is possible with intermediate levels of N application. Likewise, although water treatments led to some biomass accumulation; no significant differences were detected, indicating that water was not the most limiting factor for DM production during study years.

Forage yield of introduced pastures

The results of this study showed that the establishment of pure pastures of D. glomerata accompanied by N fertilization, or pure pastures of T. pratense, are viable options to increase forage yield in N. antarctica SPS, compared with natural understory. In contrast, the establishment of B. catharcticus failed in these sites despite several planting attempts, and T. repens did not exceed the productivity of the natural understory. Water treatments had little effect on forage DM accumulation, probably because total rainfall in both studied years was higher than the historical mean, making this factor less limiting than it might be in drier years. Conversely, fertilizer had significant effects on DM accumulation, mainly due to a greater response to N fertilization in natural understory and D. glomerata pasture. There are few precedents of introduced pastures in Patagonian N. antarctica SPS, where extensive practices with a low level of intensification predominate, as most farmers use ñire forest mainly during the breeding season or animal maintenance (Peri et al. 2016).

Our results are consistent with Mayo et al. (2014) who evaluated an established mixed pasture of *T. repens* and *D. glomerata* without water restrictions in ñire forest in south Patagonia (Santa Cruz province).

They reported a DM production of 6929 kg DM ha<sup>-1</sup>, an increase of 84% compared with control treatments (3761 kg DM ha<sup>-1</sup>). These authors also noted no significant differences in DM production between 50 and 100% light transmissivity, indicating that these species can be established in SPS (Mayo et al. 2014). Hansen et al. (2013), working with *N. antarctica* SPS in northern Patagonia (Chubut province), reported that a mixture of *D. glomerata*, *Festuca sp.* and *T. pratense* increased the productivity of the herbaceous layer from 172 to 2178 kg DM ha<sup>-1</sup>. These lower values compared with our study may be due to a lack of nitrogen fertilization or to water stress at their sites.

# Forage quality: leaves N concentrations and digestibility

N concentrations values in the natural understory found in this study align with those reported by Bahamonde et al. (2012) and Peri et al. (2016) for ñire understory in similar sites of southern Argentina. Furthermore, we observed a 250% increase in N concentrations in understory leaf tissues, highlighting N as a significant limiting factor in these systems. This suggests that substantial improvements in the quality of the natural stratum in these areas can be achieved with moderated levels of N fertilization (100 kg N ha<sup>-1</sup>). Both natural understory and the implanted D. glomerata pasture showed similar response to N addition, increasing N concentrations in their leaf tissues. In contrast, fertilizer application did not significantly affect forage quality in legume pastures, indicating that available P was likely not limiting in the evaluated pastures. We observed consistently higher N concentrations in legumes compared to grasses. This finding is consistent with studies by Kyriazopoulos et al. (2013) and Bakoglu et al. (1999), who reported that pastures of T. subterraneum presented higher N concentrations than D. glomerata pastures. Legume forages typically contain a higher percentage of crude protein than grasses due to lower fiber concentrations (Ball et al. 2001; Bonin and Tracy 2011; Pang et al. 2019). Pang et al. (2019) have suggested that the higher resilience in crude protein percentage (or foliar N concentrations) observed in legumes may be attributed to their ability to fix nitrogen, whereas grasses primarily depend on available soil N (Pang et al. 2019).

For in vitro DM digestibility, we found that T. repens presented the highest values. This is consistent with Peri et al. (2016), who reported that the quality of the understory in ñire SPS improved considerably when this species was incorporated into forage consumption. It is important to note that T. repens has been established in many sites of this region, but generally with a low proportion in vegetation cover (Peri et al. 2016). Furthermore, digestibility values found in this study for grasses (D. glomerata and natural understory) were higher than those reported by Peri and Bahamonde (2012) for natural understory in *N. antarctica* SPS in southern Patagonia (50–63%). These differences may be owing to variations in environmental conditions, as these authors reported negative associations between digestibility and increasing soil or air temperature and positive correlations with an increment in soil moisture (Peri and Bahamonde 2012).

#### Conclusions

In the context of climate change, the intensification of productive systems is needed to obtain greater yield per unit of area. In this study, we showed that D. glomerata with medium levels of N fertilization produced 30% more biomass than natural understory in southern Patagonian N. antarctica SPS, while T. pratense produced 70% more biomass than natural understory, with no fertilizer addition. Regarding forage quality, both legumes always provided higher values in foliar N concentrations and digestibility than natural understory. Finally, it is important to note that natural understory of the studied site could increase its productivity up to two times by adding intermediate levels of N fertilization, making this a viable alternative to increase DM production in these systems.

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Author contribution Gargaglione Verónica wrote the manuscript text, Peri Pablo reviewed the manuscript and contributed to the discussion section. Juan Pablo Mayo and Christensen Rodolfo contributed with meteorological and environmental data and field work. Cecilia Casas contributed adding new statistical analyses, using general mixed models, as suggested by one of the reviewers. She also reviewed the manuscript incorporating improvements in data presentation.

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**Data availability** No datasets were generated or analysed during the current study.

#### Declarations

**Conflict of interest** The authors declare that there are not conflicts of interest to publish this manuscript and no practices were carried out with animals or related to human health.

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