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

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Abstract

Nothofaus antarctica forest extends from 46 to 56º SL been used as silvopastoral systems (SSP). Several studies have characterized understory of *N. antarctica* SSP. However, other practices like pasture sawn, irrigation and fertilization, to improve understory yield, remains undocumented. The aim of this work was to compare dry matter (DM) production and quality of natural understory vs. implanted pastures under different water and nutrient conditions. An experiment with fifteen plots of 6x 6 m where randomly established in a split-split plot design with specie as the main factor, water as the sub-plot factor and fertilizer application level as sub-sub-plot factor. 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 one and two years of experiment establishment. Significant differences were found in DM according to specie, water and fertilization treatment. The highest DM production was for *D. glomerata* with irrigation and 100 kg N ha^{-1} (6347 kg DM ha^{-1}), followed by natural understory with irrigation and 200 kg N ha^{-1} and *T. pratense* with irrigation after the first year. In contrast, after two years, significant differences were found only for specie and fertilizer treatments where the highest yield corresponded to *T. pratense* pasture. For quality *T. repens* had the highest digestibility values. This study showed that pure pastures of *D. glomerata* accompanied by N fertilization or *T. pratense*, are viable options to increase forage yield and quality in *N. antarctica* SSP.

Introduction

Native *Nothofaus antarctica* (ñire) forest extends from 46 to 56º S Latitude in Patagonia region of Argentina and Chile. Most of these forests have been used as silvopastoral systems (SSP), 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 SSP (Peri et al., 2016). However, other topics such as fertilization, irrigation or pasture sawn 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 it 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). Also, Gargaglione et al. (2014) reported that in good site qualities, understory DM production of natural grasses is lower under tree canopy than in 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 from 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, introduction of shade-tolerant grass and legume species in the appropriate mixture increases the herbage production and the nutritive value (Muñoz and Weaver, 1999) because of the ability of the legumes to fix atmospheric nitrogen and to benefit companion grasses (Ledgard, 1991).

In the context of climate change, there is a tendency to increase systems intensification to obtain higher yields per unit area by implementing different strategies such as the use of fertilization and irrigation, to increase productivity. In this sense, there is need to improve understory production in ñire SSP, either through the implementation of species adapted to shade, or other options as the use of fertilization or irrigation. Among the species commonly known as shade tolerant 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). Besides, although some native legumes grow in ñire understory (like *Vicia sativa*), their participation in the vegetation cover is very low, representing around 5% (Gargaglione and Peri, 2015). Incorporation of legumes in the understory of SSP may improve site fertilityby fixing atmospheric nitrogen (N) providing fertility and increase digestible dry matter for livestock consumption. In ñire forests, as in many other natural systems, N is the most limiting nutrient source (Diehl et al., 2008) mainly during spring, when low temperatures limits N mineralization (Bahamonde et al., 2012b). In this sense, another way to improve herbage DM production and quality could be adding fertilizer, although there is a lack of studies that evaluatedthe magnitude of DM response to fertilizers in ñire SSP. The aim of this work was to compare DM production and quality of natural understory vs. implanted pastures of grasses and legumes in young *N. antarctica* silvopastoral systems under different water and nutrient conditions.

Material and Methods

Study site

The site is located in Cancha Carrera ranch (51º 13' 21" S, 72º 15' 34" O) in SW of Santa Cruz province, Southern Patagonia, Argentina. This study was carried out in a naturally regenerated *N. antarctica* young forest (41 ± 6 years old) with an original density of 5820 tree ha^{-1}, 9 cm of diameter at breast height (DBH) and a total mean height of 8 m. The natural understory layer had 80–100% vegetation cover, being grasses the predominant group. The climate is cold temperate, with 6ºC of mean annual temperature and 379 mm of mean annual rainfall. Precipitation was recorded by automatic agro-meteorological station (Davis MB5 LR Vantage Pro 2 wireless), located at 51º 31' 58,77" SL; 72º 15' 45,35" WL. In addition, air and soil temperatures were measured continuously every 2 h with a data logging system (HOBOH8 Family, Onset Computer Corporation, USA). Sensors of air temperature were placed at 0.6 m height from ground level. Soil temperature was measured at 3 cm depth using soil thermometers (HOBO, Model TMC50-HA, USA). Soils are classified as Molisols, with 60 cm of depth, pH near of 4.7, 5.6% of organic carbon and nutrient concentrations of 0.6% of nitrogen and 23.5 ppm phosphorous.

Natural Understory evaluation

The botanical composition of natural understory was evaluated in two transects of 6 m length located in each plots of 6 x6 m (n = 3). In each transect, species cover was measured by the interception method (Levy and Madden, 1933), recording every 10 cm all plant species, litter or bare soil. With these data, the percentage of vegetation cover, specific richness and Shannon's diversity index were estimated.

Experiment design and pasture installation

Previous this study, 0.4 ha was thinning by reducing crown cover to 50%. The site was fenced to avoid grazing in the SSP. In this thinned area, fifteen plots of 6x 6 m where randomly established in a split-split plot design (n = 3) with specie as the main factor, water as the sub-plot factor (6 x 3m) and fertilizer application level as sub-sub-plot factor. 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 sawn pastures, the natural sward (standing vegetation, rhizomes and roots) was removed and tilling the soil using a roto tiller and a rake. Plots were sown with *D. glomerata* at 12 kg ha^{-1}, *B. catharticus*and *T. pratense* at 15 kg ha^{-1} and *T. repens* at 10 kg ha^{-1}. All pasture seeds were tested for viability, and germinating were: 60, 80, 82 and 84% for DG, BR, TP and TR, respectively. Seed and fertilizer

were broadcast onto the plots by hand in September 2011 (early spring).

Water treatment consisted in a control and irrigation of90 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^{-1} for grass plots (DG, Br and NU) applied as urea, and 0, 50 and 100 kg Pha^{-1} for legume plots, applied as superphosphate. Fertilizer treatments were repeated once a year during spring (October) for two years. In addition, 3 plots with natural understory with the same water and fertilizer treatments were evaluated.

Measurements and Data analysis

Understory DM production and digestibility was measured after one (2013) and two years (2014) experiment establishment in the grow peak (late January) by clipping biomass within a 0.1 m^2quadrat. Material collected was sorted in green, senescent and flower components. Then, material was oven dried at 60º C to get dry weight. Subsamples of 20 g were sent to the Forage Laboratory of INTA Balcarce (Buenos Aires, Argentina) for *in vitro* digestibility analysis. In addition, forage quality was evaluated by studding foliar N concentrations monthly among the growing season, since the first season (2012) and the two years after. Subsample of 20 g was sent to LANAQUI laboratory (Universidad Nacional del Sur, Bahía Blanca, Buenos Aires) for total nitrogen analysis with Kjeldahl method. Data was analyzed using the INFOSTAT 2.0 software (Di Rienzo 2018) in a split plot design ANOVA with a significance level of 0.05%. Significant differences were tested using Tukey test.

Results

Environment conditions

Total rainfall in both years of study was higher than the historic mean, being 424 and 400 mm for 2013 and 2014, respectively. Mean air temperatures during the growing season were 10.2 and 10.7 °C for 2013 and 2014, respectively, meanwhile mean soil temperature was around 9.9 °C in both years (Figure 1). Instead of similar mean air temperature values between years, the first year (2013) presented more days with air temperatures above 16-18 °C (Figure 1 A) than the second year (Figure 1 B).

Figure 1. Mean daily air (blue lines) and soil (red lines) temperatures of the *N. antarctica* silvopastoral system studied in south Patagonia, after the first year 2013 (A) and the second year (B) of the experiment establishment.

Natural understory

Natural SSP 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 1). Mean species richness and Shannon diversity index were 16 and -1.7, respectively (Figure 2).

Table 1. Specie composition and their relative cover in natural understory of *N. antarctica* silvopastoral system in south Patagonia, Argentina.

Specie	Relative cover (%)
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

Water and fertilizer application had little effect in vegetation cover, although Shannon diversity index decreased from -1.7 (control) to -1.44 for the treatment 200 kg N ha^{-1} (Figure 2B). Likewise, relative cover of understory species had some variations according to water and fertilizer treatments. For example, under irrigation and 200 kg N ha^{-1} *P. pratensis* cover to 24% and *D. glomerata* increased from 16 to 43 %. Other species did not show changes in relative cover according to irrigation or fertilization treatments.

Figure 2. A) vegetation cover and B) Shannon index diversity of natural understory of *N.antartica* silvopastoral system in soutn Patagonia in response to water irrigation (90 mm added, grey bars) and fertilizer application (0, 100 and 200 kg N ha^{-1}).

Forage implantation and yield

With exception of *Bromus catharticus*, all species were established successfully with a vegetation cover of 80 -90%. Significant differences were found in DM production according to specie, water and fertilization treatment, with some significant interactions between treatments (Table 2). After one year of experiment establishment, the highest DM production was for *D. glomerata* with irrigation and 100 kg N ha^{-1} with 6347 kg DM ha^{-1} (Figure 3 B, Table 3) followed by natural understory with irrigation and 200 kg

N ha^{-1} (Figure 3 A, Table 3) and *T. pratense* with irrigation and no fertilization with 5207 kg DM ha^{-1} (Figure 3 C, Table 3). *T. pratense* and natural understory in the control presented the lowest biomass production with 419 and 852 kg DM ha^{-1}, respectively (Table 3). In contrast, after two years of pasture implantation, significant differences ($p < 0.05$) were found only for specie and fertilizer treatments (Table 2). The highest yield was for *T. pratense* with 50 kg P ha^{-1}(8489 kg DM ha^{-1}, Figure 3 G, Table 3), followed by *D. glomerata* with 100 kg N ha^{-1} (5985 kg DM ha^{-1}, Figure 3 F, Table 3), natural understory (3883 kg MS ha^{-1}, Figure 3 E) and *T. repens* with 3096 kg MS ha^{-1} (Figure 3 H). At this year, pasture with no fertilization presented the lowest DM production compared with medium and high fertilization levels (Figure 3, Table 3).

Table 2. Analysis of variance of dry matter production comparing three forage species implanted (*Trifoliumrepens,* T.*pratense* and *Dactylis glomerata*) and natural understory with two watertreatments (irrigated and control) and three fertilizer levels (low, medium, high)after one and two years of experiment establishment in a *N. antarctica*silvopastoral system in southern Patagonia, Argentina. Fertilizer levels corresponded to three levels of nitrogen applied to grasses (01 100 and 200 k N ha^{-1}) and three levels of phosphorus applied for legumes (0, 50 y 100 kg de P ha^{-1}).

	First year			Second year		
Factor de Variación	**gdl**	**Fvalue**	**p value**	**gdl**	**Fvalue**	**p value**
Model	39	3,52	0,0002	39	5,41	<0,0001
Specie	3	8,89	0,0063	3	9,23	0,0056
Water	1	34,39	0,0004	1	0,76	0,4086
Fertilizer level	2	9,15	0,0007	2	13,34	0,0001
Watter* specie	3	4,48	0,0399	3	0,34	0,7985
Water * Fertilizer	2	4,6	0,0175	2	1,22	0,3077
Specie* Fertilizer	6	6,17	0,0002	6	2,53	0,0404
water* specie *Fertilizer	6	1,86	0,1196	6	0,4	0,875

Figure 3. Mean annual biomass production (kg DM ha^{-1} yr^{-1}) of pure pastures implanted (*Dactylis glomerata, Trifolium pratense* y *T. repens*) and natural understory in a *N. antartica* silvopastoral system (southern Patagonia, Argentina) after one (A, B, C, D) and two years (E, F, G y H) of pasture sawn, under different water regime (control, white bars, vs. irrigated, grey bars) and three fertilization levels: low (control =0), medium and high. For grasses, fertilization treatments were: 0, 100 y 200 kg de N ha^{-1} and for legumes pastures fertilizer levels corresponded to 0, 50 y 100 kg de P ha^{-1}.Vertical bars indicate standard deviation of the means.

Table 3. Mean values of dry matter production of three pure pastures implanted (*Dactylis glomerata, Trifolium pretense* and *T. repens*) and natural understory in a *N. antarctica* silvopastoral system (southern Patagonia, Argentina) after one (2013) and two (2014) years of experiment implantation. Different letters indicate significant differences between treatments with a p value < 0.05.

	Irrigated			Control		
	Fertilizer level			Fertilizer level		
January 2013	**Low**	**Medium**	**High**	**Low**	**Medium**	**High**
Natural understory	1705 abcde	3936 abcdef	5729 ef	852 a	2720 abcdef	3388 abcdef
Dactylis glomerata	1093 *abc*	6347 f	3525 abcdef	969 ab	2635 abcdef	5054 bcdef
Trifolium repens	1113 *abcd*	2305 abcdef	2306 abcdef	923,7 ab	1159 abcd	1783 abcde
Trifolium pratense	5217 cdef	5256 def	2932 abcdef	2758 abcdef	419 a	1555 abcd
January 2014						
Natural understory	1378 ab	3049 abcde	3762 abcdef	1773 abc	3883 abcdef	3041 abcde
Dactylis glomerata	2227 abcd	4033 abcdef	5985 cdefg	1255 a	5475 abcdefg	5370 abcdefg
Trifolium repens	2346 abcde	2660 abcde	2503 abcde	2804abcde	2769abcde	2801 abcde
Trifolium pratense	5566 bcdefg	6611 efg	6535 efg	6147 defg	8489 g	7413 fg

For legume pastures, after one year, irrigation had significant effects on *T. Pretense* DM production by increasing yield by from 419 to 5256 kg DM ha^{-1} (Figure 1 G). In contrast, after two years, irrigation did not present significant differences in dry matter production. Similarly, P fertilization did not show significant effects in the second year. By another way, *T. repens* produced 923-2300 kg DM ha^{-1} in the first year, and 2300-2800kgDM ha^{-1} in the second year, without significant differences in biomass production according to water or fertilizer treatments.

Forage quality

Significant differences were found (p <0.05) in leaf N concentration according to species, water and fertilization, and some significant interactions between treatments (Table 4). Differences among species occurred mainly in spring (December 2012), months later of experiment establishment, when *T. pratense* had significantly (p <0.05) higher N concentrations than other species (Figure 4). In the spring of the second year, *T. repens* had the highest N concentration, followed by *T. repens* and grasses (Figure 4). Fertilizer treatment influenced in N concentrations depending on date and significant differences were found in February 2012, January and March 2013 (Table 4), where *D. glomerata* and natural understory whit no fertilization had lower values than those growing in medium and high fertilization levels (Figure 4).

Figure 4. Mean leaf N concentration values (%) during the growing season in *Dactylis glomerata*, *Trifolium repens*, *T. pretense* pastures and natural understory in *N. antarctica* silvopastoral system in Southern Patagonia, Argentina.

Table 4. Analysis of variance (p values) of leaves N concentration according to specie (*Dactylis glomerata, Trifolium repens, Trifolium pratense* and natural understory), water treatment (irrigation and control) and fertilizer level (0, 100 and 200 kg Nha^{-1} for grasses and 0, 50 y 100 kg Pha^{-1}, for legumes).

Date	specie	water	Fertilizer level	water*specie	water*fertilizer	specie*fertilizer
Feb 2012	0.2023	**0.0051**	**0.0017**	0.7341	0.2172	0.0697
Dic 2012	**0.0051**	0.4342	0.359	**0.0461**	0.8285	0.0759
Ene2013	0.0873	0.0818	**<0.0001**	0.1367	0.2043	**<0.0001**
Mar 2013	0.6866	**0.0247**	**0.0001**	0.4779	**0.0065**	0.134
Dic2013	**0.0021**	0.7614	0.8278	0.4659	0.355	0.0781
Ene2014	**<0.0001**	0.3183	0.6381	0.1857	0.7469	0.0628

Figure 5 shows means N concentrations values in peak of biomass. Natural understory and *D. glomerata* pastures presented similar N concentrations values and similar response to fertilizer application, whit values around 1.2 % and 1.5 % N in control for first and second year, respectively and increased to 2.9 and 3% in medium and high N applied levels in the first year (Figure 5 A). In contrast, during the second year, N concentration did not show differences according to N application, oscillating between 1.5 and 1.7 among all N levels. Legumes instead, fertilizer application did not produce significant differences in N concentrations (Figure 5).

Figure 5. Mean values of N concentration after A) one year and B) two-year implantation of *Dactylis glomerata*, *Trifolium repens*, T. pratense and the natural understory in *N. antarctica* silvopastoral system in response to three levels of fertilization (control =0, medium and high). Fertilization applied was 0, 100 and 200 kg Nha^{-1} for *D. glomerata* and natural and understory and (0, 50 y 100 kg Pha^{-1}) for legumes.

Digestibility

Significant differences ($p < 0.05$) were found in *in vitro* dry matter digestibility according to specie, water, fertilizer level and year, although there were no significant interactions between treatments (Table 5). In both years, *T. repens* was the species that accounted with the highest digestibility values with 81.6 and 80.0 % for first and second year, respectively (Table 5). *D. glomerata* had the lowest values with 72.0 and 66.0 % for first and second year, respectively, followed by natural understory with 74.0 and 67.0 %, respectively (Table 5). Watter and fertilizer treatments had influence on leaves digestibility depending on the year. For example, in the first-year, plants under irrigation presented higher digestibility values than control, without significant differencesduring the second year. In addition, fertilizer level presented significant differences only after two years of experiment establishment, where pastures with the higher fertilizer level showed highermean in vitro digestibility values (71.4 %) compared with control (69.3 %).

Table 5. Digestibility values in peak of biomass (late January) after one and two years of implanted plots of *Dactylis glomerata, Trifolium repens, T. pratense* and natural understory in a *N. antarctica* silvopastoral system in southern Patagonia (Argentina), under water treatment (irrigation vs control) and three fertilizer levels (control, medium and high). Fertilizer treatment correspond to three levels of N applied (0, 100 y 200 kg ha^{-1}) in *D. glomerata* and natural understory plots and three levels of P for legume plots (0, 50 and 100 kg Pha^{-1}).

	Digestibility (%)	
Factor	After one year	After two years
Specie		
Dactylis glomerata	72.5 a	66.52 a
Natural understory	73.8 a	67.46 a
Trifolium pratense	74.25 a	66.79 a
Trifolium repens	81.58 b	80.04 b
Water treatment		
Irrigation	76.08 a	69.91 a
control	75.00 b	70.49 a
Fertilizer level		
Low	75.25 a	69.33 a
Medium	75.6 a	69.91 ab
High	75.76 a	71.37 b

Discussion

Natural Understory

Natural understory of this *N. antarctica* silvopastoral system contains exotic grasses, like *Dactylis glomerata*, *Poa pratensis* and others dicotyledonous as *Taraxacum officinale* probably introduced in the past, by the ranchers. The native *Osmorhiza chilensis* represented 12% of vegetation cover with little variation between applied treatments. The botanical composition of the present study contrast with Peri et al. (2016) who indicated that dominant species at a regional level in ñire silvopastoral systems are dominated by *Carex andina* (20–30%), *Poa pratensis, Phleum commutatum, Dactylis glomerata, Bromus setifolius, Agrostis flavidula, Deschampsia flexuosa* and *Festuca pallescens.*

Water and fertilizer application decreased the Shannon index diversity of understory, mainly due to an increase in *D. glomerata* cover with irrigation and N application. Biomass production found in this study for natural understory in control plots (852 kgha^{-1}) was in the range of those informed by other authors for ñire forests in south Patagonia (Bahamonde et al., 2012; Peri et al., 2016). Understory dry matter production in ñire silvopastoral systems depends on the interaction of soil water availability and light intensity that reaches the soil, since under moderate water stress conditions DM production decrease from 2800 Kg DM ha^{-1} in open sites (100% light transmittance) to 500 kg kg DM ha^{-1} in severe shade (Peri et al., 2016).

In this study we found that fertilizer application produced significant changes in natural understory biomass accumulation in both water treatments, increasing from 852 to 3388 kg DM ha^{-1} in the first year and from 1772 to 3883 Kg DM ha^{-1}in the second year. This is concordant 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, in this study no significant differences were found in biomass accumulation between 100 and 200 Kg N ha^{-1}, thus, an important increase in natural forage yield is possible with intermediate levels of N applications. It is possible that the increase in biomass production with N application could be related with the increase in *D. glomerata* abundance as N and water increase.

Forage yield of introduced pastures.

Results found in this study show that implantation of pure pastures of *D. glomerata* accompanied by N fertilization or pure pastures of *T. pretense*, are viable options to increase forage yield in *N. antarctica* silvopastoral systems, by increasing biomass production compared with natural understory. In contrast, *Bromus catharcticus* establishment failed in these sites despite repeated sawing attempts, and *T. repens* did not exceed the productivity of the natural understory. Water treatments had different effects according to the year. While after one year, water addition had significant effects by increasing dry matter understory productions, this effect was not evident in the second year. These could be related to differences in environment conditions between years. Although mean annual rainfall and air temperatures were similar in both years, year 2013 presented more days with temperatures above 18°C, which could produce higher water stress in control plots and higher yield under irrigation. Also, it is possible that at the second year pastures were better established with a larger root system that facilitate the uptake of water and nutrients from deeper soil. In contrast, fertilizer had significant effects in both evaluated years, mainly owing to a greater response to N fertilization in natural understory and *D.*

glomerata pasture. There are few antecedents of introduce pastures in Patagonian *N. antarctica* silvopastoral systems, where predominate extensive practices with a low level of intensification, since many region land managers use ñire forest mainly during breeding season, or maintenance or without specific objectives (Per et al., 2016). Mayo et al. (2014) evaluated an implanted mixed pasture of *T. repens* and *D. glomerata* without water restrictions in ñire forest in Santa Cruz province(south Patagonia), and they reported a dry matter production of 6929 kg DM ha^{-1}, an increase of 84% compared with control treatments (3761 Kg DM ha^{-1}). These authors also reported no significant differences in DM production between 50% and 100% of light transmissivity, indicating that these species are able to implant in silvopastoral systems (Mayo et al., 2014). Hansen et al., (2013) working with *N. antarctica* silvopastoral systems in North Patagonia (Chubut province), reported that a mixture of *D. glomerata*, *Festuca sp*. and *T. pratense* increased the productivity of the herbaceous layer from 172 kg DM ha^{-1} to 2178 kg DM ha^{-1}. These lower values lower values compared with our study may be due to lack of nitrogen fertilization or to more severe water stress.

Forage quality: N concentrations and digestibility

N concentrations values in natural understory found in this study are concordant with those reported by Bahamonde et al (2012) for ñire understory in similar sites of southern Argentina, and with those reported by Peri et al (2016). Likewise, we found that N application increased in 250% in N concentrations in leaves of understory, indicating again, a strong N limiting in these systems and shows that a substantial improvement in the quality of the natural stratum of these areas can be obtained with the application of medium levels of N fertilization (100 kg Nha^{-1}). Natural understory and the implanted *D. glomerata* pasture had similar response to N addition, increasing N concentrations in leaf tissues. However, the response varied according to year. In contrast, fertilizer application did not affect forage quality in legume pastures, indicating that available P probably was not limiting for these kinds of pastures. We observed that legumes always had higher N concentrations compared with grasses. This is concordant with Kyriazopoulos et al. (2013) and Bakoglu et al. (1999) who reported that pastures of *T. subterraneum* presented higher N concentrations than *D. glomerata* pasture and with Bakoglu et al. (1999). Legume forages usually have higher % crude protein than grasses due to lower concentrations of fiber (Ball et al. 2001; Bonin and Tracy 2011, Pang et al., 2019). Likewise, Pang et al. (2019) postulated that high resilience in % of crude protein (or foliar N concentrations) for legumes may be due to their nitrogen fixing ability while grasses depend mainly on available soil N (Pang et al., 2019).

For digestibility, in this study *T. repens* presented the highest values in both evaluated years. This is concordant with Peri et al. (2016) who reported that understory quality of ñire silvopastoral systems improved considerably when this species was incorporated into forage consumption. It is important to note that *T. repens* had been established in many sites of this region, but in general 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* silvopastoral systems in south Patagonia (50–63%). These differences may be owing to differences in environment conditions, since 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 et al., 2012). This was demonstrated in the present study, where in the first year, irrigation increased digestibility values although in the second year, was the fertilizer level that increased digestibility.

Conclusions

In the context of climate change, 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 N fertilization produced 60 and 40% more biomass than natural understory in southern Patagonian *N. antarctica* silvopastoral systems, after one and two years of implantation, respectively. *T. pratense* also is a good option by producing 200% more biomass than natural understory, after two years on established. In contrast, *B. catharticus* could not be implemented and *T. repens* had lower yield compared with natural understory. With respect to forage quality, *T. repens* and *D. glomerata* had higher values in foliar N concentrations and digestibility than natural understory.

Finally, is important to note that natural understory of the site studied could increase its productivity up to three times by adding intermediate levels of N fertilization, under no limiting water.

Declarations

Author Contribution

Gargaglione Verónica wrote the main manuscript text, Peri Pablo reviewed manuscript and contribute to the discussion, Juan Pablo Mayo and Christensen Rodolfo contributed with meteorological and environmental data and field work.

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Statements and Declarations

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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Figures

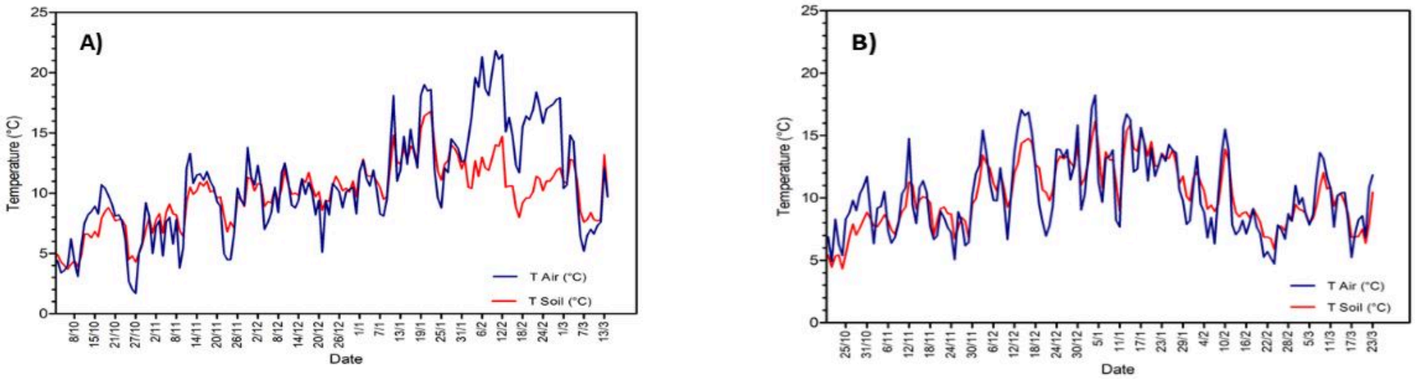

Figure 1

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

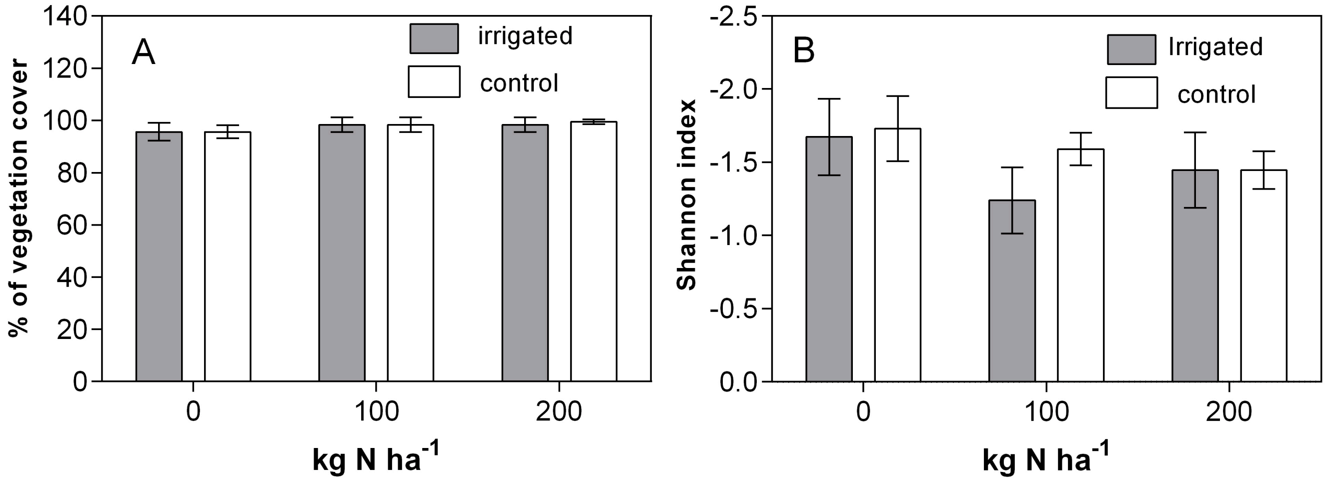

Figure 2

A) vegetation cover and B) Shannon index diversity of natural understory of *N.antartica* silvopastoral system in soutn Patagonia in response to water irrigation (90 mm added, grey bars) and fertilizer application (0, 100 and 200 kg N ha^{-1}).

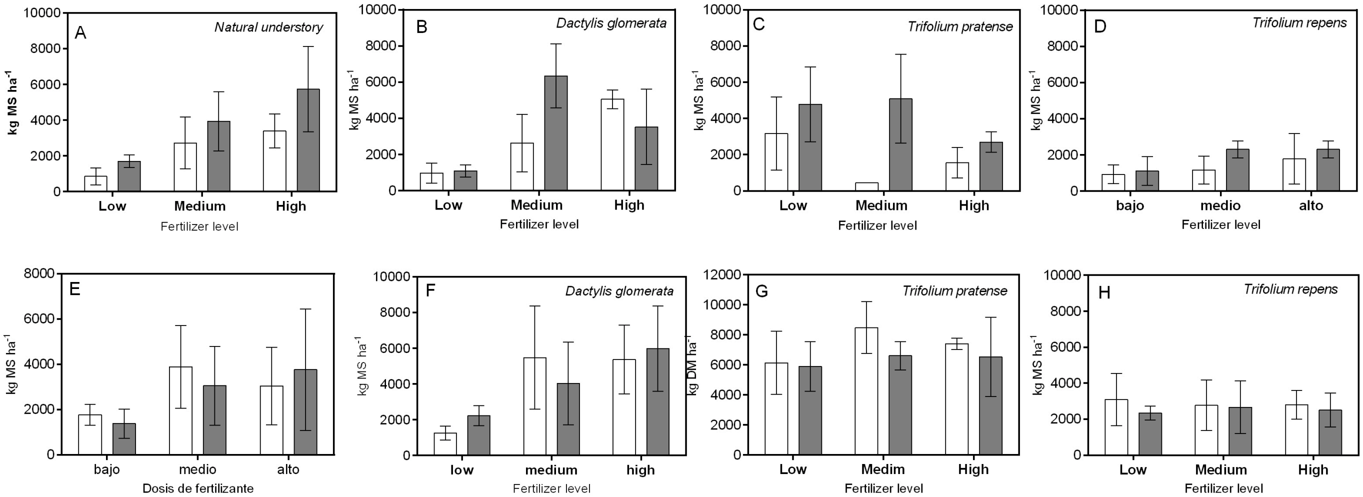

Figure 3

Mean annual biomass production (kg DM ha^{-1} yr^{-1}) of pure pastures implanted (*Dactylis glomerata, Trifolium pratense* y *T. repens*) and natural understory in a *N. antartica* silvopastoral system (southern Patagonia, Argentina) after one (A, B, C, D) and two years (E, F, G y H) of pasture sawn, under different water regime (control, white bars, vs. irrigated, grey bars) and three fertilization levels: low (control =0), medium and high. For grasses, fertilization treatments were: 0, 100 y 200 kg de N ha^{-1} and for legumes pastures fertilizer levels corresponded to 0, 50 y 100 kg de P ha^{-1}.Vertical bars indicate standard deviation of the means.

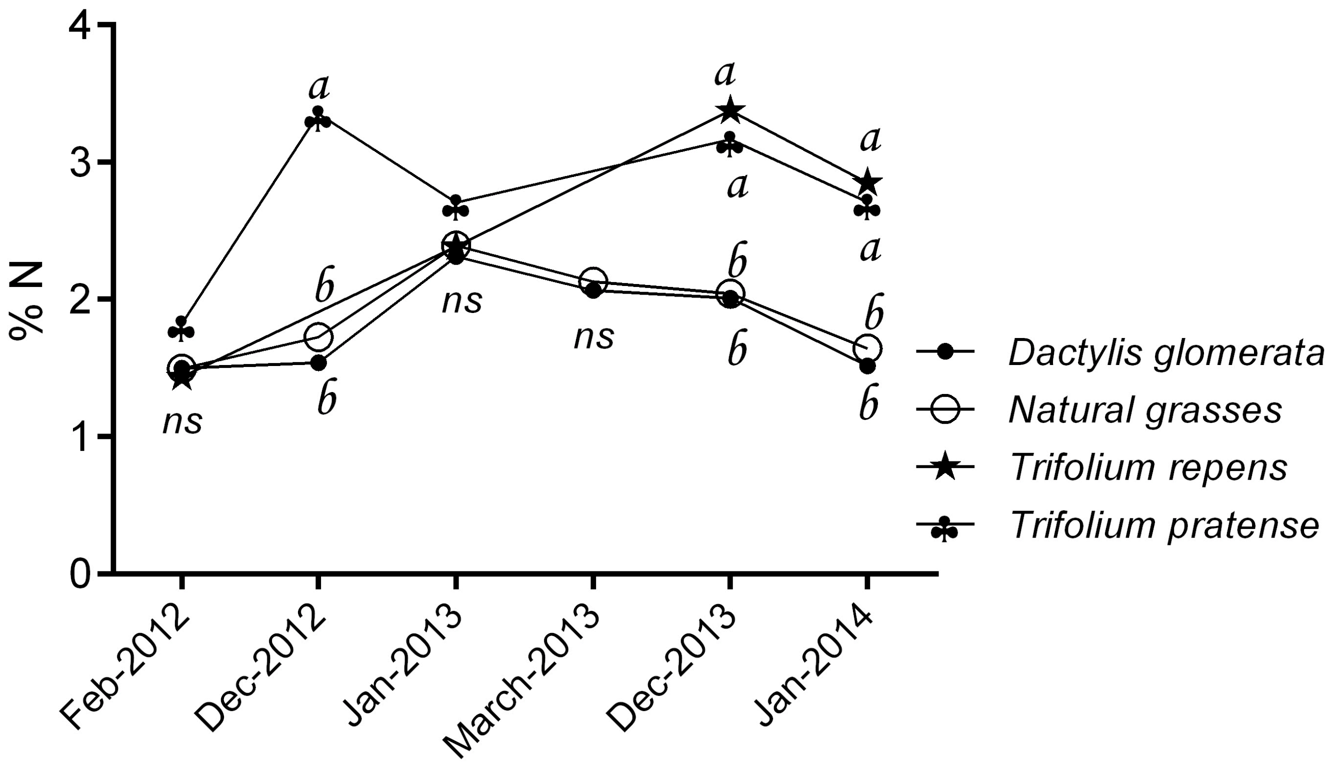

Figure 4

Mean leaf N concentration values (%) during the growing season in *Dactylis glomerata*, *Trifolium repens*, *T. pretense* pastures and natural understory in *N. antarctica*silvopastoral system in Southern Patagonia, Argentina.

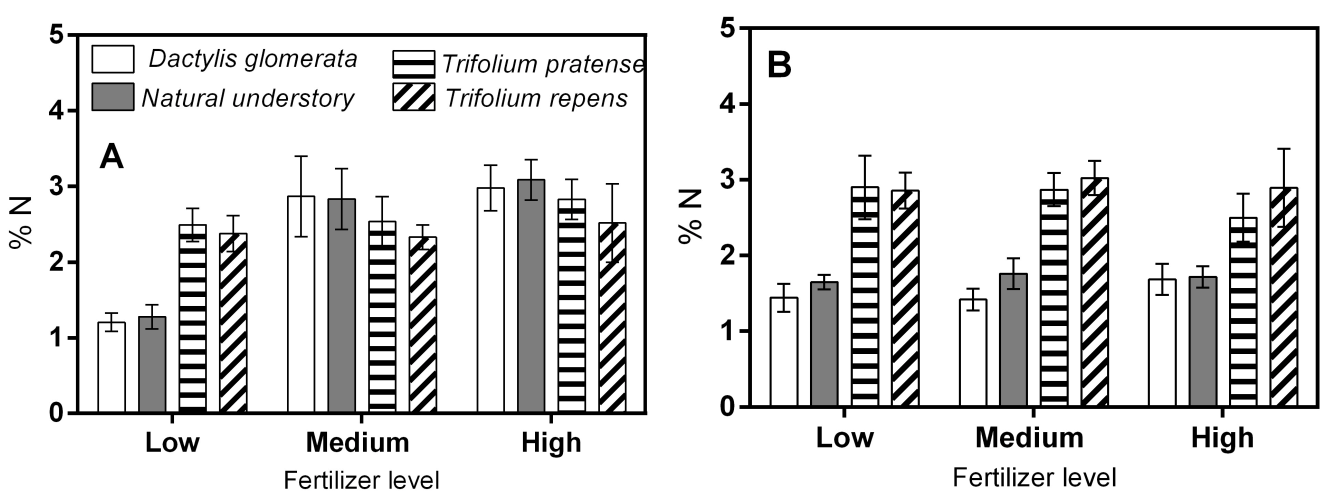

Figure 5

Mean values of N concentration after A) one year and B) two-year implantation of *Dactylis glomerata*, *Trifolium repens*, T. pratense and the natural understory in *N. antarctica* silvopastoral system in response to three levels of fertilization (control =0, medium and high). Fertilization applied was 0, 100 and 200 kg Nha^{-1} for *D. glomerata* and natural and understory and (0, 50 y 100 kg Pha-^{1}) for legumes.