## Short Communication

# Nitrogen fertilization effects on forage production and nutritive value of 4 tropical grasses on alkaline soils in Argentina

*Efectos de la fertilización nitrogenada sobre la producción forrajera y el valor nutritivo de 4 pastos tropicales en suelos alcalinos en Argentina* 

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## Abstract

This study evaluated effects of nitrogen (N) fertilization and gypsum application and their interactions on total pasture forage production and nutritive values of *Chloris gayana* cultivars 'Santana' and 'Finecut' and *Panicum coloratum* cultivars 'Klein' and 'Bambatsi' growing in a moderately sodic soil (Typic Natracualf) in northcentral Santa Fe Province of Argentina over 3 years. Sown pasture forage production differed among cultivars. *C. gayana* cultivar 'Finecut' produced more forage biomass than the other 3 cultivars. *P. coloratum* cultivars were superior to *C. gayana* cultivars for nutritive value, showing lower NDF and ADF. N fertilization strongly increased forage production, total biomass and sown pasture biomass with improved crude protein content and improvement in plant N status. Addition of gypsum did not affect forage production and nutritive value. This research showed the feasibility to improve production and nutritive value of tropical pastures in subtropical areas by species/cultivar selection and N fertilization.

Keywords: C4 species, Chloris gayana, nitrogen nutrition index, Panicum coloratum.

## Resumen

Los objetivos de este trabajo fueron evaluar los efectos de la fertilización nitrogenada, el encalado y su interacción sobre la producción total de forraje y el valor nutritivo de cultivares de *Chloris gayana* 'Santana' y 'Finecut' y de cultivares de *Panicum coloratum* 'Klein' y 'Bambatsi' creciendo en un suelo sódico moderado (Natracualf típico) de la región centro-norte de la provincia de Santa Fe de Argentina durante tres años. Los cultivares difirieron en producción de forraje, siendo esta mayor en *C. gayana* 'Finecut' que en los otros tres cultivares. Los cultivares de *P. coloratum* fueron superiores en valor nutritivo a los de *C. gayana* por menores contenidos de FDN y de FDA. La fertilización nitrogenada aumentó fuertemente la producción total y de la pastura sembrada y mejoró el contenido de proteína y el estatus de nitrógeno en la planta. El encalado no afectó la producción forrajera ni el valor nutritivo. Esta investigación demostró la posibilidad de mejorar la producción y el valor nutritivo del forraje en áreas subtropicales mediante la selección de la especie/cultivar y la fertilización nitrogenada.

Palabras clave: Chloris gayana, índice nutrición nitrogenada, megatérmicas; Panicum coloratum.

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## Introduction

Extensive grazing on both natural grasslands and cultivated pastures is the predominant livestock production system in Argentina. Grazing in marginal areas with salinity, alkalinity and frequent flooding has become common in recent years as cropping expands (Manuel-Navarrete et al. 2009) in areas of central Argentina with mesothermal climate (Mosconi et al. 1981) and high temperatures. C4 tropical forage grasses adapted to salinity (Pittaro et al. 2015), waterlogging (Imaz et al. 2015) and their combined stress (Lifschitz et al. 2022) have proved to be adapted to environmental conditions in the area (Romero and Mattera 2013; Mattera et al. 2015; Tomás et al. 2018).

Nitrogen (N) fertilization increased forage production and nutritive value in Rhodes grass (Chloris gayana Kunth.) in a sodic soil in a subtropical region (Bruno et al. 1982) and was very responsive to N application associated with adequate rainfall (Boschma et al. 2014). Increases in forage production due to N application have been reported in C. gayana and Panicum coloratum L. (Pesqueira et al. 2016). Gypsum provides soluble calcium to the soil solution and replaces sodium, resulting in improved soil conditions (Qadir et al. 2007). The relationship between nitrogen and gypsum to improve forage production in these species is unknown. The aims of this study were to evaluate effects of N fertilization and gypsum application and their interactions on total pasture forage production and nutritive values of C. gayana cultivars 'Santana' and 'Finecut' and P. coloratum cultivars 'Klein' and 'Bambatsi' in a moderately sodic soil (Typic Natracualf) in northcentral Santa Fe Province of Argentina over 3 years.

#### **Materials and Methods**

The experiment was carried out at La Palmira Ranch, San Cristóbal Department, Santa Fe Province, Argentina (29°46'36'' S, 61°14'43'' W; 66 masl) from 2014–2016. The soil was Natracualf with soil capability  $VI_{ws}$  (Klingebiel and Montgomery 1961). Soil analysis of the 0–20 cm horizon before sowing showed the following parameters: Organic matter 3.6%, extractable phosphorus 22.7 ppm, pH 7.6, electrical conductivity 1.1 mmhos/cm, exchangeable sodium (ESP) 22%, and sodium to calcium ratio 0.66. Average annual precipitation at the site in the last 50 years was 1,103 mm and average temperature was 21 °C with a frost-free period from October to March. Precipitation during the experiment was above the historical average in the 3 years of the experiment (Table 1).

The experiment was carried out in an area of natural vegetation with the grasses C. gayana, Chloris halophila Parodi and Distichlis scoparia (Kunth) Arechav. without previous grazing and fertilization. The experiment included combinations of 3 factors: Cultivar (C. gavana cultivars 'Santana' and 'Finecut'; P. coloratum var. coloratum cultivar 'Klein' and P. coloratum var. makarikariense cultivar 'Bambatsi'), N fertilization (unfertilised and N applied as urea as a single dose of 100 kg N/ha/year) and gypsum amendment (unamended and 0.5 t gypsum/ha/year in the first year and 1 t gypsum/ha/year in following years). A randomized complete block design was used with split plot arrangement and 3 replicates. The main plot was the cultivar (40 m<sup>2</sup>/plot) and subplots were the factorial combination of N fertilization and gypsum amendment (10  $m^2/plot$ ).

Thousand seed weight of C. gayana was 0.86±0.02 g and  $1.00\pm0.02$  g with a germination percentage of 44±6% and 51±11% for cultivars 'Santana' and 'Finecut', respectively. P. coloratum 1,000 seed weight was 1.36±0.08 g and 2.63±0.17 g with a germination percentage of  $22 \pm 13\%$  and  $36 \pm 6\%$  for cultivars 'Klein' and 'Bambatsi', respectively. Seeds of cultivar 'Bambatsi' were coated with calcium carbonate to increase seed size while the remainder were uncoated and no treatments or inoculations were given. The seedbed was prepared by disc and tine harrows and the experiment seeded in rows at a sowing depth of 1 cm with row spacing of 16 cm on 22 January 2014. Seeding densities were high with the aim to obtain 250 seedlings/m<sup>2</sup>, ranging from 7 kg/ha (C. gayana cultivars), 23 kg/ha (P. coloratum cultivar 'Klein') and 26 kg/ha (P. coloratum cultivar 'Bambatsi'). N fertilization and gypsum amendment were performed

 Table 1. Rainfall (mm) at La Palmira Ranch during 2014–2016.

		( )			0								
	JAN	FEB	MAR	APR	MAY	JUN	JUL	AUG	SEP	OCT	NOV	DEC	TOTAL
2014	193	186	112	42	62	6	38	1	62	97	113	278	1,190
2015	222	117	158	40	8	18	0	89	5	142	224	112	1,135
2016	68	179	46	510	5	25	0	20	20	126	136	198	1,333

on the same day at sowing in the first growing season (22 January 2014) and in spring in the second and third growing seasons (14 October 2014 and 20 November 2015, respectively).

## Data collection

For forage evaluation, both total forage production in the plot from sown and unsown grass and other species (total pasture) and biomass production of the sown species only (sown pasture) were measured. Evaluation was carried out at several regrowth periods of different lengths according to phenological stage, with harvesting at beginning of reproductive development during 3 growing seasons (Table 2). The first evaluation, corresponding to the establishment period, was the time between sowing and first harvest. The second and third growing seasons covered 3 evaluations (regrowths after cleaning cut) each from spring to autumn. At each harvest an area of 5 m<sup>2</sup>, avoiding the borders of each plot, was harvested at 5 cm cutting height above ground level. Fresh material was weighed after each regrowth period in the field and 2 forage sub-samples were taken from each plot (200 g); the first sample was used to estimate dry matter content and the second to estimate the proportion of sown pasture compared to the total after it was separated from natural vegetation. The harvested forage was dried to constant weight in an air-forced oven at 60 °C.

Forage samples of sown pasture were taken to the laboratory from the first regrowth period for 2 growing seasons (2014 and 2014–15) for nutritional analysis. Analyses were performed at INTA Rafaela Forage Laboratory assessing crude protein (CP) (AOAC 1995), neutral detergent fiber (NDF) (Ankom 2005) and acid detergent fiber (ADF) (Ankom 2005). Additionally, nitrogen nutrition index (NNI) was estimated as an indicator of N plant status, calculated as:

NNI=Observed nitrogen content/Expected critical nitrogen content

Expected critical nitrogen content was estimated according to the reference curve for C4 grasses:

Critical nitrogen (%)= $3.6 \times \text{Biomass}^{-3.4}$  (<u>Duru et al.</u> 1997)

Biomass was considered as the forage production of the sown pasture only for the estimation. In the second growing season (2014–15), *P. coloratum* cultivar 'Bambatsi' was excluded from analysis because forage biomass was very limited in plots under natural conditions and it was impossible to obtain a representative sample.

 Table 2. Forage evaluation harvesting dates over 3 growing seasons (2014–16).

Carriera concor	Start date	Hamvasting data			
Growing season	Start date	Harvesting date			
2014	Sowing: 22 Jan 2014	15 Apr 2014			
2014-15	Cleaning cut: 14 Oct	5 Dec 2014			
	2014	27 Jan 2015			
		17 Mar 2015			
2016	Cleaning cut: 20 Nov	5 Jan 2016			
	2015	3 Mar 2016			
		18 May 2016			

## Statistical analysis

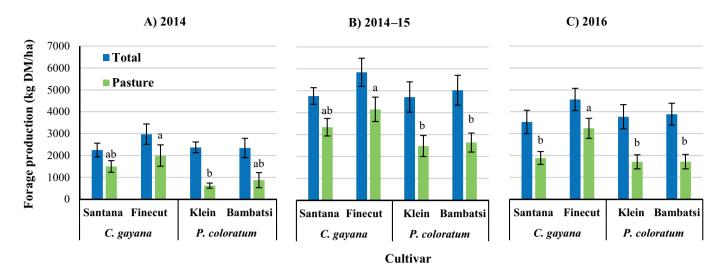
Total and sown pasture forage production were analyzed as a split plot design and factorial arrangement of fertilization and gypsum amendment. Repeated measures with R Core Team (2021) lme4 package (Bates et al. 2015) were used to evaluate the 3 growing seasons together. Additionally, independent ANOVAs were performed for each individual growing season. Normality and variance heteroscedasticity were checked and means compared with Fisher's least significant difference (LSD) to detect differences between treatments (P<0.05) using INFOSTAT software (Di Rienzo et al. 2017). For nutritive value variables and NNI, the same R package was used for the unbalance due to missing values for *P. coloratum* cultivar 'Bambatsi' in the second growing season.

#### Results

#### Forage production

No differences in total forage production (P>0.05; Figure 1, blue columns) were detected among cultivars. However, sown pasture forage production differed among cultivars with *C. gayana* cultivar 'Finecut' (Figure 1, green columns) showing significantly higher yields in all 3 growing seasons.

N fertilization had a significant positive effect on both total and sown pasture forage production (Figure 2). Increments were higher for the third growing season, followed by the second growing season (nitrogen × growing season interaction; P<0.05). *P. coloratum* showed a higher responsiveness to N fertilization for sown pasture forage production (cultivar × nitrogen interaction; P<0.05). Gypsum amendment did not affect forage production for any growing seasons and did not interact with the other factors (P>0.05).



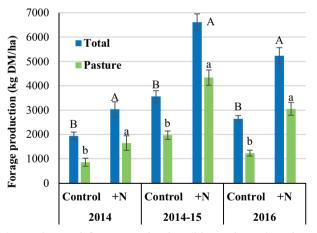
**Figure 1.** Total forage production (blue columns) and sown pasture forage production (green columns) of tropical grass cultivars (*C. gayana* cultivars 'Santana' and 'Finecut', *P. coloratum* cultivars 'Klein' and 'Bambatsi') in 3 growing seasons. Different lower-case letters indicate significant differences for total and sown pasture forage production, respectively (P<0.05). Bars represent standard errors.

#### Nitrogen plant status

Although NNI was below the reference curve, N fertilization improved NNI in both growing seasons (P<0.05) (Table 3), with a more pronounced increment in the second growing season (nitrogen × growing season interaction; P<0.05). *P. coloratum* had lower NNI than *C. gayana* in the first growing season (P<0.05).

#### Nutritive value

ADF differed between cultivars (P<0.05) with *P. coloratum* cultivar 'Bambatsi' having the lowest ADF (Table 4). In the second growing season (2014–15), *P. coloratum* cultivar Klein had a significantly lower ADF and NDF than *C. gayana* cultivars. N fertilization increased forage CP (P<0.05) whereas gypsum amendment did not affect nutritive value in any growing season (P>0.05). No significant interactions were detected between factors for forage nutritive value.



**Figure 2.** Total forage production (blue columns) and sown pasture forage production (green columns) of tropical grass cultivars (*C. gayana* cultivars 'Santana' and 'Finecut', *P. coloratum* cultivars 'Klein' and 'Bambatsi') in 3 growing seasons for different nitrogen fertilization. Different upper case and lower case letters indicate significant differences for total and sown pasture forage production, respectively (P<0.05). Bars represent standard errors.

		NNI 2014	NNI 2014–15
Cultivar	Klein	0.25 b	0.42
	Bambatsi	0.27 b	-
	Finecut	0.41 a	0.47
	Santana	0.41 a	0.48
Nitrogen fertilization	Natural condition	0.30 b	0.31 b
	Nitrogen	0.37 a	0.60 a
Gypsum amendment	Natural condition	0.33	0.44
	Gypsum	0.34	0.46
CV (%)		19.61	20.54

**Table 3.** Nitrogen fertilization and gypsum amendment effects on nitrogen nutrition index (NNI) of tropical grass cultivars (*C. gayana* cultivars 'Santana' and 'Finecut', *P. coloratum* cultivars 'Klein' and 'Bambatsi') in a Natracualf soil according to the growing season.

\*Different letters between levels of each factor indicate significant differences (P<0.05)

Table 4. Nitrogen fertilization and gypsum amendment effects on nutritive value of tropical grass cultivars (C. gayana cultivars
'Santana' and 'Finecut', P. coloratum cultivars 'Klein' and 'Bambatsi') in a Natracualf soil according to the growing season.

					-		
			2014			2014-15	
		CP (%)*	NDF (%)	ADF (%)	CP (%)	NDF (%)	ADF (%)
Cultivar	Klein	7.09	67.85	34.96 ab**	10.77	64.74 b	28.72 b
	Bambatsi	7.42	65.25	32.39 c	s.d.***	s.d.	s.d.
	Finecut	7.90	66.26	35.58 a	9.07	67.49 a	33.60 a
	Santana	8.53	65.83	33.11 bc	9.75	66.86 a	33.33 a
Nitrogen fertilization	Natural condition	7.84	65.65	33.80	9.16 b	66.93	31.52
	Nitrogen	7.63	66.64	34.22	10.57 a	65.80	32.25
Gypsum amendment	Natural condition	7.71	66.69	34.20	9.86	66.13	31.63
	Gypsum	7.76	65.61	33.82	9.87	66.59	32.14
CV (%)		11.08	3.53	6.70	12.07	2.69	3.61

\*CP=crude protein; NDF=neutral detergent fiber; ADF=acid detergent fiber. \*\*Different letters between percentages of each factor indicate significant differences (P<0.05). \*\*\*No analysis due to scarce biomass of *P. coloratum* cultivar 'Bambatsi' in 2014–2015.

## Discussion

This study confirms previous reports that showed good adaptation of *C. gayana* in northcentral Santa Fe Province (Bruno et al. 1982; Romero and Mattera 2013; Mattera et al. 2015). Forage production of *P. coloratum* was lower than *C. gayana*, mainly due to a lower percentage of plant biomass in plots. This lower production of *P. coloratum* in sub-tropical environments, despite persistence and good tillering capacity, is in line with results reported by Pesqueira et al. (2016). *C. gayana* was recognized as a weed in *P. coloratum* plots, probably from the seed soil bank as *C. gayana* is naturalized in the area (Pisani et al. 2009).

High exchangeable sodium limits forage productivity and can be alleviated with application of gypsum due to replacement of sodium by calcium in the soil solution (Qadir et al. 2007). In this study, neither forage production nor nutritive value were significantly improved by addition of gypsum with low impact on soil pH and electrical conductivity (data not shown). Leaching of salt would be required to decrease exchangeable sodium from the soil profile (Qadir et al. 2007) but was probably prevented by poor drainage and a fluctuating water table (Hein and Hein 1986).

N fertilization influenced forage production, corroborating previous studies that showed N responsiveness of these grasses in subtropical and temperate environments (Bruno et al. 1982; Boschma et al. 2014; Pesqueira et al. 2016). A moderate single annual dose of N (100 kg N/ha) increased forage production between 70% and 147%. Although sown pasture forage production increased in all cultivars with N fertilization, the increment was particularly important in *P. coloratum* as an important strategy to improve pasture establishment and subsequent production. Improved plant status of N increased plant growth and forage production and was reflected in increase in NNI value, which is a good indicator of plant response to nitrogen (Lemaire et al. 2008). In this study, a higher relative increase in NNI was recorded in the second growing season, probably associated with well-established adult plants with well-developed root systems. NNI values were below the reference curve, indicating N-fertilized pastures were deficient in N, allowing a response in forage production. Future studies to assess nitrogen recovery and nitrogen use efficiency would add to the understanding of N dynamics in pastures because it was not possible to determine N absorption in this study.

Although forage nutritive value was higher in *P. coloratum* than *C. gayana*, impact on overall feed value may be low due to lower percentage of the species in total forage. As expected, crude protein content increased with N fertilization, coincident with increased forage production and NNI improvement.

#### Conclusions

*C. gayana* cultivar 'Finecut' was the most productive of the 4 cultivars of tropical grasses tested in the study in a subtropical area. *P. coloratum* cultivars had higher nutritive value but were few in the pasture. Nitrogen fertilization increased forage production and crude protein content through improving N status in fertilized plots. During the evaluation period, addition of gypsum had no effect on forage production nor forage nutritive value of the 4 cultivars tested.

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