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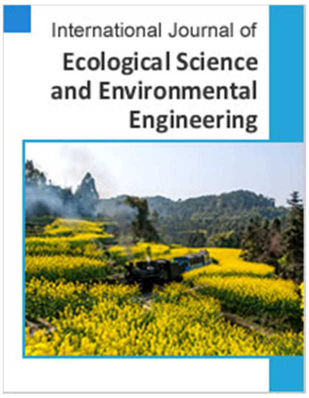

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Methodological Proposal for the Analysis of Spatio-Temporal Variability of Forage Value on *Paspalum quadrifarium*-Dominated Grasslands

Ilda Entraigas[1, 2, 3, *], Sergio Chiramberro[2], Horacio de Dominicis[2], Natalia Vercelli[1, 2, 4], Fabricio Bongiorno[2], Juan Carlos Ramaglio[2]

[1]Large Plains Hydrology Institute, National University of the Center of Buenos Aires, Azul, Argentina
[2]Faculty of Agronomy, National University of the Center of Buenos Aires, Azul, Argentina
[3]Commission of Scientific Research, La Plata, Argentina
[4]National Council of Scientific and Technical Research, Buenos Aires, Argentina

Email address

ilda@faa.unicen.edu.ar (I. Entraigas), sergio.chiramberro@hotmail.com (S. Chiramberro), hdedominicis@gmail.com (H. de Dominicis), natalia.vercelli@yahoo.com.ar (N. Vercelli), fbongiorno@faa.unicen.edu.ar (F. Bongiorno), ramaglio@faa.unicen.edu.ar (J. C. Ramaglio)
[*]Corresponding author

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Abstract

The Flooding Pampa is a subregion of Argentina where there is a plant community whose physiognomy is defined by tall-tussock grasses dominated by *Paspalum quadrifarium*. The hypothesis is that their floristic diversity allows a continuous forage supply, and this happens in a different way in each type of vegetation patch. The aim is to analyze the temporary and spatial variability of the forage value of these tallgrass prairies by the development of an index. Samplings were carried out throughout the year, for two consecutive years in three different sites. The index is made up of four components: Indicators of Cover/Abundance, Digestibility, Accessibility, and Phenological Stage of the vegetation. The site indexes ranged between 0.05 and 0.54, with minimum values in winter and maximum values in spring. Overall, the lax *pajonal* showed higher values than the dense *pajonal*, and within the latter one, the values of the short grass matrix were usually higher than those of the sector with dense patches dominated by *P. quadrifarium*. This index is useful to visualize the environmental conditions, seasons, vegetation patches and main species that influence the forage condition of a grassland ecosystem.

1. Introduction

In livestock production systems under grazing it is essential to know the seasonal availability of forage resources to take rational decisions about carrying capacity, stocking method and grazing management. The forage mass estimation is very important because it is the basis of animal production, and it can be estimate through different methods: harvest [28], height and volume [41], visual estimation [27], radiometry [29], photography [14] [30] and remotely sensed data [3], among others.

Methods for performing sampling depend on objectives persued and conditions of measurement, such as availability of human and material resources, precision required, scale of work and characteristics of vegetation. Likewise, the sampling procedure must be adapted to homogeneity, density, and botanical composition of vegetation [42].

On the other hand, the forage mass spatial and temporal variability is one of the factors that most affects the efficiency of livestock farms. This variability can often lead to underutilization of fodder, as well as overexploitation with the consequent deterioration of forage and edaphic resources [15].

The Flooding Pampa is a subregion in the central portion of Argentina that occupies about 90000 km^2. As it is a vast plain with soil and drainage conditions that severely limit the development of agricultural activity, almost 80% of its surface is not cultivated, therefore it keeps its natural or semi-natural vegetation [33]. These rangelands are currently the main source of nutrients for cattle, although they are at different levels of degradation mainly due to an improper resource management. This has led to both a loss of forage species in the grasslands and a gradual emergence of weeds. As a result, productivity decreases also due to the seasonality of production, which determines an important forage deficit in winter.

Among the different expressions that natural grasslands in the region adopt, there is a plant community whose physiognomy is defined by dense and tall-tussock grasses, locally known as ʽpajonal' or ʽpajonales', according to its singular or plural form (ʽ*Paspaletum*' *sensu* [47]). It is a bilayer community, where the upper layer is formed by *Paspalum quadrifarium*, and the lower one is a matrix comprising species with trailing stems, rosette-like leaves or small tussocks. *P. quadrifarium* is a South American C-4 tussock grass that has great plasticity and adaptation to different environments as it tolerates high temperatures, drought, soil compaction, heavy traffic and flooding conditions.

Some authors [46] refer to these *P. quadrifarium*-dominated grasslands (hereafter *pajonal* or *pajonales*) and their evolution over time. The Flooding Pampa area was occupied mainly by these *pajonales* [47], and although floods/droughts alternance has determined that much of the area retains relatively natural features [38], these grasslands have suffered a setback in their extension by the application of herbicides, burning and tillage farming purposes [24].

These tallgrass prairies have been analyzed from several perspectives; for example, effects of different management strategies [7] [9], the role that this tussock grass plays in structuring species diversity patterns [32], the consequences of prescribed fire [36] [25] [48] [35], its fragmentation status [17] [18] [19] [22], and the botany characterization at a landscape scale [43] [45].

These grasslands distributed around the Flooding Pampa are Valuable Grassland Areas because they are considered physiognomic and floristic relicts of the landscape that dominated this region long time ago [4]. In addition, these remnants may provide different ecosystem services [11], while providing habitat for a wide variety of vertebrates that depend almost exclusively on the existence of a mature *pajonal* [10] [23] [20] [4].

The hypothesis that organizes this work is that the floristic diversity of these *pajonales* allows -along the whole year- a continuous forage supply, and that this happens in a different way in each vegetation patch type that form the internal mosaic of the community. Hence, the objective is to analyze the temporary and spatial variability of the forage value of these tallgrass prairies by the development of an index. Specifically, an index that captures the floristic diversity of these *pajonales* by considering the intrinsic characteristics (cover/abundance, digestibility, accessibility and phonological stage) of each species that forms this community at a given time. An index for measuring this variability would provide a metric to enable farmers and agricultural planners to make a sustainable use of their forage resources.

2. Methods

Study area

The study area is located in the central area of the Flooding Pampa, in the Azul county, Province of Buenos Aires (Figure 1). It is a plain area, extremely flat (slopes about 0.1%), with alkali and/or hydromorphic soils that show a hard carbonate crust of variable depth and continuity, between 0.5 and 1 m depth, locally known as ʽtosca'. According to data recorded in the Azul Aero Meteorological Office of the Servicio Meteorológico Nacional, the mean annual precipitation in Azul is 914 mm (1901-2014); the maximum monthly mean precipitation occurs in March (137 mm), while the minimum occurs in June (43 mm). The mean annual temperature is 14.5°C, with a maximum monthly mean in January of 21.4°C and the lowest monthly mean in July of 7.7 °C (1966-2009) [44]. According to the climate classification of Thornthwaite and Mather [40], the climate of the region is classified as subhumid-humid, mesothermal, with little or no water deficiency.

Sampling method

The study was carried out in a parcel of approximately 0.9 square kilometers that belongs to a livestock property (36° 38' 33.6" S – 59° 42' 17.0" W). The selected plot presents a typical *pajonal*, that is, with a wide internal heterogeneity in relation with the distribution of *P. quadrifarium* (Figure 1). Some sectors present tall, dense, and big patches dominated by *P. quadrifarium* (Site A) with some internal areas dominated by short grasses (Site B), and other sectors where tussocks are distributed in a lax way, so the matrix of short grasses has a remarkable development (Site C).

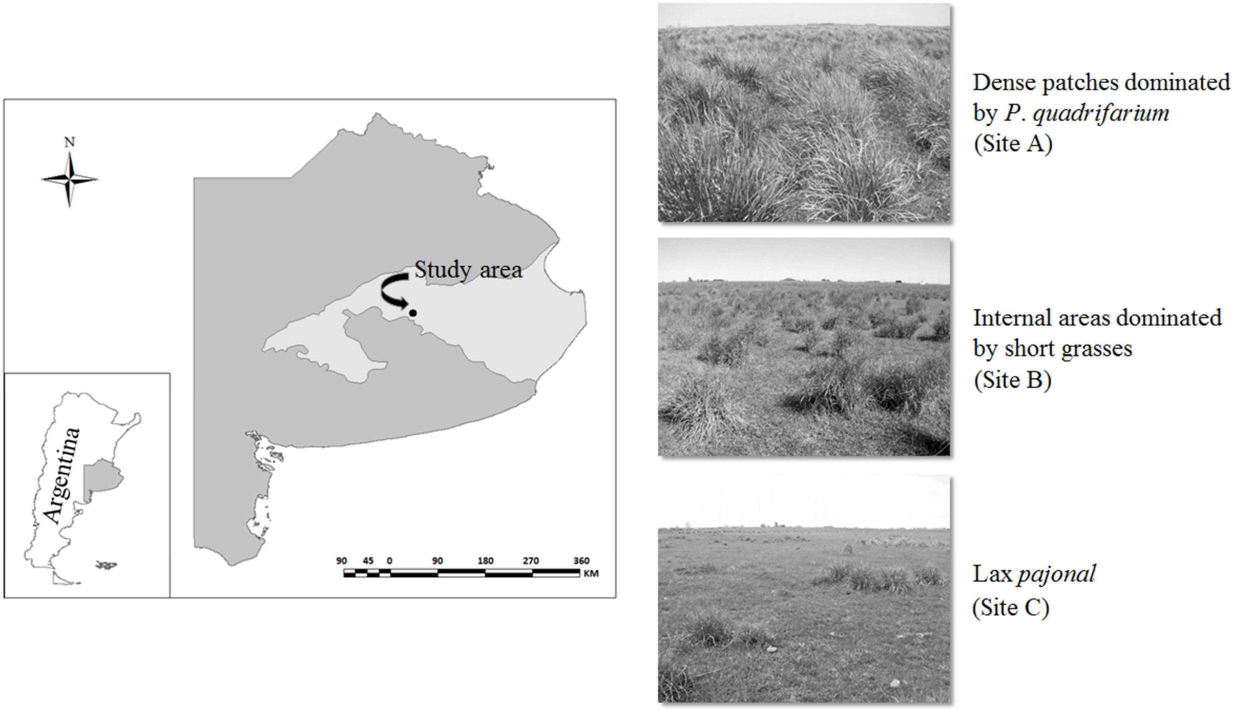

Figure 1. *Left: The study area (black dot) in the central area of the Flooding Pampa (light grey area) in the Province of Buenos Aires (dark grey area). Right: Physiognomy of the selected plots; tall, dense, and big patches dominated by P. quadrifarium (Site A); internal areas of the dense pajonal dominated by short grasses (Site B), and tussocks distributed in a lax way (Site C).*

Three samplings of vegetation were carried out throughout the year, for two consecutive years. They were held during the months of April (spring/summer species in reproductive status, and fall/winter species in vegetative status), August (fall/winter species in vegetative or early-reproductive status) and December (spring/summer species in vegetative status, and fall/winter species in reproductive status). In each site (A, B and C) the observations were made on a quadrant of 4 m^2, according to the concept of minimum area of the community [26], placing it at random to avoid bias.

Details about the Forage Value Index

The index proposed (Forage Value Index) for the analysis of spatio-temporal variability of forage value is made up of four components: Indicators of Cover/Abundance, Digestibility, Accessibility, and Phenological Stage of the vegetation.

Cover-abundance was estimated according to a visual assessment of the relative area covered by the different species in the quadrant, based on a modified Braun-Blanquet scale: 1% for a solitary individual with very low cover, 3% for few individuals with a cover less than 5%; and then, 5% - 100% as in a continuous scale.

Later, the Cover/Abundance indicator was developed by transforming the percentage of cover/abundance for each species into a decimal expression. So, 100% was expressed as 1; 50% as 0.5; 3% as 0.03, and so on.

Digestibility is the proportion of the dry matter, organic matter or nutrients absorbed during passage through the digestive tract, and is generally expressed on a percentage basis by the digestibility coefficient. We obtained percentage values for some species from literature [1] but for others we found authors that used different criteria to value digestibility in a vegetative stage [34] [8] [13]. For this reason, we developed our Digestibility Indicator from a comparative evaluation among them (Table 1).

Table 1. *Digestibility Indicator proposed (Idig) and its correspondence with the different criteria used on the reviewed literature.*

Digestibility (%)	Rosengurtt (1979)	Cahuépé (1994)	Fernández Greco and Viviani Rossi (2011)	Digestibility Indicator (Idig)
>70	Fine	5	Very good	1
70-60	Soft	4	Good	0.83
59-50	Regular	3	Regular	0.53
49-45	Hard	2	Low	0.33
44-40	Very hard	1	Very bad	0.13
<40	Weeds	0	Negligible	0

In some cases, digestibility values could not be obtained (either because it was not possible to determine the species of a given genus or even the family of a collected specimen, or because no information was found in the literature about that parameter for an identified species). So a general rule was adopted for those situations: the digestibility coefficient average value of the family (for unknown genus) or of the genus (for unknown species), 10% being substracted in order to avoid overestimation in that parameter.

On the other hand, in many grazing situations, forage availability (kg DM / ha) is the main factor limiting consumption. In some forage species, availability is not related to consumption because the variables that make the structure of the forage mass (accessibility) are more important: height, spatial distribution, leaf/stem ratio, live/dead ratio, and so on [1].

In this research, the accessibility was included taking into account the height measured in centimeters from the ground to the highest point reached by a leaf. Some authors [16], meanwhile, associate the bite size with the height of the plant. From those statements, we developed the Accessibility Indicator (Table 2).

Table 2. *Accessibility Indicator proposed (Iacc) and its correspondence with the parameters taken into account by Galli et al. (1996) and the plant height measured on field.*

Galli et al. (1996)		Plant height (cm)	Accessibility Indicator (Iacc)
Accessibility	**Weight of the bite (g of dry matter)**		
Very high	1.38	>30	1.00
High	1.16	30-25	0.84
Medium	0.88	25-20	0.76
Low	0.59	20-15	0.67
Very low	0.34	15-5	0.58
Inaccessible	0	<5	0.00

Quality, both in legumes and grasses, has a close dependence on the phenological stage: in tillering (grass) or early stages of the development cycle (legumes), forage quality is high; then quality begins to decline from the time of internodes elongation in grasses or early flowering stage in legumes, and it reaches its lowest level when the plants are heading or late flowering [1]. In the same way some researches [21], based on the dynamism of the life cycle of certain species, analyzed the nutritional profile of many of them from a set of chemical parameters of nutritional importance (moisture, ash, crude fiber, lignin, etc.), and taking those data as reference, the Phenological State Indicator was developed (Table 3).

Table 3. *Phenological State Indicator proposed (Ips) for grasses and legumes, and their correspondence with the different criteria used on the reviewed literature.*

	Phenological state	Digestibility (%)		Phenological State Indicator (Ips)
Grasses		Aello and Di Marco (2004)	Jaurena et al. (1994)	
	Tillering	70-75	70-75	1
	Internodes elongation	60-65	62-65	0.87
	Heading	50-55	55-57	0.73
Legumes		Jaurena et al. (1994)		
	Early stages	>65		1
	Early flowering	61		0.94
	Late flowering	53		0.82

P. quadrifarium has a very particular tussock architecture that some authors [2] have described as consisting of two cones. An external one, composed of new leaves emerging radially, with high forage quality and available for livestock feed; and an inner one, with lower nutritional quality, and whose leaves are not available to livestock because they are under a compact mass of severed or dead leaves. Due to the situation described above, it is considered that cattle consume only about 20% of the total offered by the tussock. Therefore, for the particular case of *P. quadrifarium*, the index was calculated as for any other species but considering only 20% of its value.

The Forage Value Index is structured as follows:

First, in each site, the Primary Index for each recorded species is estimated (values for each Indicator are determined from their respective already detailed scales):

$$InPsp = Icob * Idig * Iacc * Ips$$

Where:

InPsp: Primary Index for each species

Icob: Cover/Abundance Indicator

Idig: Digestibility Indicator

Iacc: Accessibility Indicator

Ips: Phenological Stage Indicator

Then, the Total Primary Index is obtained for each site (InTPst), which is the result of the summation of the Primary Index for each recorded species in this site.

$$InTPst = InPsp_1 + InPsp_2 + InPsp_3 + InPsp_4 + \ldots\ldots + InPsp_n$$

Afterwards, based on the visual interpretation of satellite images, the areal representativeness of each vegetation patch type (in our case, sites A, B and C) is evaluated. In addition, to validate the different tussock densities recognized from the images, the Line Interception Method [5] [39] was used on field. In our work, 2 transects of 30 m (on the same starting point and at randomly selected directions) were located. Then, a 4 m measuring tape was stretched perpendicular to the baseline (2 m to each side of the baseline) spaced at 5 m, recording the number of centimeters in which there was either a *P. quadrifarium* tussock or the short grass matrix. This procedure was repeated 4 times in each site. Now, the Total Primary Index at a site level (InTPst) can then be weighted by the proportion of the parcel surface occupied by each vegetation patch type (sup) to obtain the corresponding Index of Forage Value at a parcel scale (InFVpar).

$$InFVpar = InTPstA * supA + InTPstB * supB + InTPstC * supC$$

The index can obtain values that range from 0 to 1, where 0 represents a parcel without any vegetation, either without forage species, or with plants that cannot be reached by the bite of livestock; and 1 represents a parcel with total coverage of species with a digestibility value higher than 70%, totally accessible for livestock, and in a stage of nutritional splendor.

Considering the years as pseudo-replications and sites and sampling event as factors, the analyses of variance (ANOVA model III) was performed. Means separation was based on Fisher's Least Significant Difference (LSD) test at a 5% significance level. Infostat [12] was the statistical package used.

3. Results and Discussion

On the analyzed *pajonal*, the site indexes ranged between 0.05 and 0.54, with minimum values in winter and maximum values in spring, reflecting the typical behavior of natural grasslands. The analysis of variance showed significant differences between sites (Table 4; $p<0.05$), and the Fisher's LSD test showed that Site A was significantly different from the others two (Table 5). Overall, the lax *pajonal* (Site C) showed higher values than the dense *pajonal*, and within the latter one, the InTPst of the short grass matrix (Site B) were usually higher than those of the sector with dense patches dominated by *P. quadrifarium* (Site A). Furthermore, taking into account only the three species that make the greatest contribution to the index, it is noticeable how, on average, they contribute about 71% of its value (standard deviation of 14.8). Although these species change in each site and occasion, it is clear that certain species have a key role based on their frequency. It is noteworthy that *Lotus tenuis* and *Lolium mutiflorum*, for example, are certainly the two species that appear in more than half of the sampling opportunities, contributing greatly to the calculation of InTPst (Table 6).

Table 4. *Analysis of variance table (SS: Sum of Squares, df: degrees of freedom, MS: Mean Squared).*

Source of variation	SS	dg	MS	F ratio	p_value
Site	0.13	2	0.07	7.76	0.0110
Sampling event	0.07	2	0.04	4.18	0.0519
Site x Sampling event	0.01	4	2.3 E-03	0.27	0.8929
Error	0.08	9	0.01		
Total	0.29	17			

Table 5. *Fisher's Least Significant Difference (LSD) test table (alfa = 0.05; Least Significant Difference = 0.12128; Error = 0.0086; dg = 9. Values followed by the same letter are no significantly different).*

Site	Mean	N	Standard Error
A	0.15 a	6	0.04
B	0.30 b	6	0.04
C	0.36 b	6	0.04

Table 6. *Total number of species, number and percentage of species with forage value, value of the Total Primary Index (InTPst), and species with the highest incidence in the index (expressed in percentage of the index value) on the dense patches dominated by P. quadrifarium (Site A), on internal areas dominated by short grasses (Site B), and on the lax pajonal (Site C), for each sampling event.*

	Sampling date	Species (Nº)	Forage species		InTPst	Highest incidence in the index	
			(Nº)	(%)		Species	(%)
Site A	04/2012	19	8	42	0.23	*Trifolium repens* *Lolium multiflorum* *Lotus tenuis*	80
	08/2012	19	5	26	0.05	*P. quadrifarium* 'Unknown Poaceae 1' *Lotus tenuis*	80
	12/2012	19	5	26	0.29	*Lolium multiflorum* *Paspalum vaginatum* *Lotus tenuis*	72
	04/2013	25	10	40	0.16	*Cynodon dactylon* *Paspalum dilatatum* *Lotus tenuis*	64
	08/2013	12	3	25	0.07	*Lolium multiflorum* *P. quadrifarium* *Cynodon dactylon*	100
	12/2013	16	9	56	0.11	*P. quadrifarium* *Lolium multiflorum* *Hordeum pusillum*	63
Site B	04/2012	25	10	40	0.25	*Plantago lanceolata* *Paspalum dilatatum* *Lotus tenuis*	55
	08/2012	16	8	50	0.26	*Nassella* sp. *Lolium multiflorum* *Eleocharis* sp.	70
	12/2012	29	13	45	0.23	*Cynodon dactylon* *Nassella* sp. *Lolium multiflorum*	66

	Sampling date	Species (Nº)	Forage species		InTPst	Highest incidence in the index	
			(Nº)	(%)		Species	(%)
	04/2013	28	13	46	0.42	*Lotus tenuis* *Paspalum dilatatum* *Plantago lanceolata*	74
	08/2013	23	9	39	0.20	*Lolium multiflorum* *Nassella phillippi* *Bothriochloa laguroides*	91
	12/2013	29	18	62	0.43	*Nassella melanosperma* *Lolium multiflorum* *Lotus tenuis*	49
Site C	04/2012	23	12	52	0.29	*Setaria viridis* *Poa* sp. *Lotus tenuis*	59
	08/2012	24	9	38	0.28	*Lolium multiflorum* *Nassella* sp. *Lotus tenuis*	86
	12/2012	24	9	38	0.38	*Lolium multiflorum* *Cynodon dactylon* *Nassella* sp.	60
	04/2013	25	11	44	0.42	*Nassella* sp. 'Unknown Poaceae 1' *Lotus tenuis*	60
	08/2013	24	4	17	0.23	*Nassella formicarum* *P. quadrifarium* *Cynodon dactylon*	93
	12/2013	30	20	67	0.54	*Lotus tenuis* *Lolium multiflorum* *Nassella formicarum*	53

However, differences in the magnitude of the maximum and minimum values for each analyzed year were observed, and to understand the causes of this behavior, it is necessary to refer to environmental issues that affected the *pajonal* distinctively in those two periods. According to the records, rains were distributed very differently during the two sampling years. Total precipitated rain in 2012 was 1,434 mm (57% more than the historical average), while in 2013 it was 689 mm (25% less than the historical average).

As a result of these events, the *pajonal* suffered prolonged flooding during most of the 2012, which impacted on the overall behavior of the vegetation community. That is, the values of InTPst in sites B and C were lower compared to 2013; however, in Site A, in which *P. quadrifarium* is the almost exclusive protagonist, this situation seems not to affect it. Probably, being a native species strongly adapted to situations of prolonged flooding is what allows such behavior.

With reference to the values of Index of Forage Value at a scale parcel (InFVpar), they reflect the typical dynamics of pampean natural grasslands, ie vegetation with marked seasonality in their forage supply, but maintaining its productive activity throughout the entire year (Figure 2).

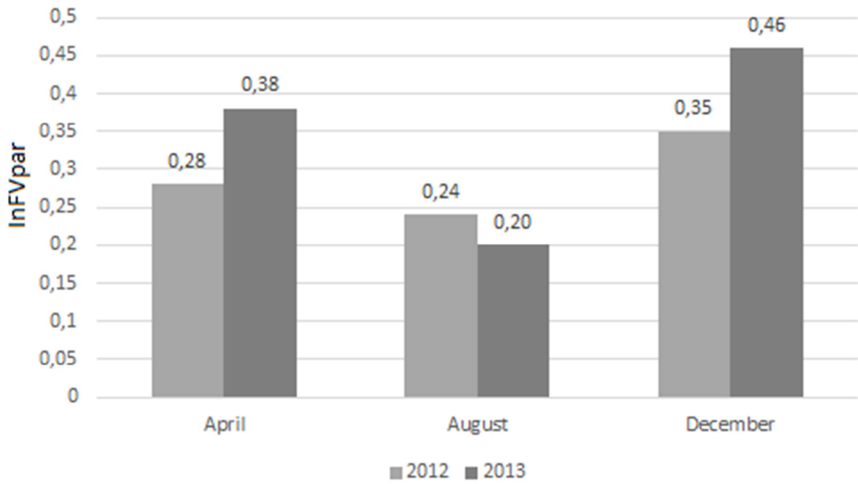

Figure 2. *Values of Index of Forage Value at a scale parcel (InFVpar) for each sampling event.*

Due to the internal heterogeneity presented in these *P. quadrifarium*-dominated grasslands, considering the different patches of vegetation over time was useful to get an integral analysis of the spatio-temporal variability of forage value on these communities. In fact, the apparent structural homogeneity of these grasslands induced their early classification as a unique community [47] [6], but another authors found different species assemblages within it that can be considered as different communities [31] [32].

The lax *pajonal* generally recorded a higher number of forage species, but even in those cases where the number was similar to the other sites, the index value was higher because these species could express themselves better (greater coverage and accessibility) due to the absence of competition with *P. quadrifarium*. This species has intrinsic characteristics (only about 20% of its foliage is consumed by animals and also has a low digestibility) which make its contribution to the index value not so significant, despite having a very high coverage in Site A.

On the other hand, it seems that prolonged flooding suffered by the *pajonal* during 2012, mainly affected sites B and C as the indexes obtained were lower than those of 2013. However, Site A, where *P. quadrifarium* is almost the exclusive protagonist, showed higher values than those of the following year. We hypothesize that the intrinsic structural characteristics of *P. quadrifarium* are the key factor for this particular situation that occurs during periods of very abundant rainfall. The aerial architecture of this species allows a significant amount of water to be trapped in its canopy and, therefore, evaporates directly from there without reaching the ground. On the other hand, as regards the radical structure of *P. quadrifarium*, it is demonstrated that *pajonales* have a positive effect on the hydrological properties of the soils. In fact, recent studies in the region postulate that areas with patches formed by this species present soils with lower values of apparent density than those with short grasses [37]. The plant architecture formed by these aerial and underground characteristics may be responsible for the particular water dynamics in these sectors. In addition, tussocks are high enough not to be affected by flooding in a dramatic way. However, the same conditions become extremely adverse for the great majority of species that form the *pajonal* matrix, since they remain submerged during several days because of their characteristics of being short grasses. All these issues turn Site A in an area where, during prolonged flooding periods, the conditions become particularly favorable for companion species of *P. quadrifarium* (many of them forage species) to develop comparatively better than under normal conditions and, consequently, the index values increase. In this way, Site A is likely to become a nursery habitat defined by both the aerial and underground architecture of *P. quadrifarium* and the environmental conditions derived from these characteristics.

Lolium mutiflorum and *Lotus tenuis* are clearly the two species that appear in more than half of the sampling occasions, contributing greatly to InTPst calculation. At the same time, some species such as *Plantago lanceolata*, *Piptochaetium stipoides*, *Nassella phillipii*, *Jarava plumosa*, *Nassella longiglumis*, *Nassella melanosperma* and *Nassella neesiana*, which are not currently considered important forage species, are highlighted in this research by demonstrating their impact on the index.

The above mentioned ensures that the *pajonal* has a lot of species with a clear foraging behavior, so many monospecific promotion practices that often take place in the region would be damaging the natural grassland expression, in a clear position of underestimation of the capacity of these environments.

4. Conclusions

The Index of Forage Value presented here takes into account the specific richness of these grasslands and highlights their forage supply for cattle in the region (out of a total of 109 identified species, 61 of them were of forage value, of which 61% were native). This is an important question because they are generally underestimated and considered low productive environments, so the index would allow fair assessment of the *pajonales*.

This index may not represent an easy metric for farmers and agricultural planners to estimate taking into account the whole indicators involved for all the species in a community. But once the species that play a key role in the value of the forage index in a given grassland are identified, it is possible to focus on them the monitoring as a proxy of forage quality in a given grassland.

We consider that this application has been useful to visualize the environmental conditions, seasons, vegetation patches and main species that influence the forage condition of a *pajonal*. Surely these factors change in different grassland ecosystems, but its application is feasible if structure, function and dynamics of each particular community are considered. Future works may use it as functional trait in ecological analysis, especially in ecosystems that have internal heterogeneity, defined seasonality and alternation of environmental conditions that deserve to be highlighted and analyzed differentially.

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