Feral horses dung piles as potential invasion windows for alien plant species in natural grasslands

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Abstract Small scale disturbances could act as patches that provide sites for the colonization of competitively inferior species, promoting the establishment of non-native species in some cases. We analyzed the vegetation associated with feral horse dung piles in montane pampas grasslands in Mid-East Argentina and described the changes following their abandonment, evaluating whether dung piles act as invasion windows, allowing the entrance of alien plant species. We estimated the portion of the study area directly covered by horse manure and dung height was used to estimate the time elapsed after the abandonment of each pile. Vegetation replacement on dung piles of different ages was assessed and compared with grassland controls using discriminant analysis. We used regression analysis to look for changes in vegetation cover, species richness, species diversity and evenness in response to height (age) of the dung piles, and principal component analyses (PCA) to identify groups of plants associated with different successional stages. We compared cover of alien plant species on dung piles with grassland controls using one-way ANOVA. On average, 2.5% of the study area was covered by horse dung. Total vegetation cover, species richness, diversity and evenness increased after the piles were abandoned. Characteristic plant groups were associated with initial, middle and last phases of the studied succession. Vegetation on the dung piles significantly differed from that in grassland controls and two species were consistently associated with dung piles: the invasive Red Star Thistle, *Centaurea calcitrapa*, and a native grazing-intolerant grass, *Nassella clarazii*. Non-native species cover was also higher in dung piles than in control plots. Dung piles cover a significant portion of grassland area in our study site, produce significant changes in the vegetation and are associated with some invasive alien plants that could eventually colonize more pristine areas in the vicinity. On the other hand, they might represent refuges for palatable species, since horses seem to avoid them for grazing.

Keywords Conservation · Invasive alien species · Natural grasslands · Refuges · Stud pile · Manure

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Introduction

Invasive alien species usually modify the disturbance regime of invaded ecosystems changing patterns of succession and promoting colonization by pioneer or

ruderal species, which results in the modification of natural communities (Vitousek et al. 1996; Mack et al. 2000). The presence of a non-native species quite often favours the introduction of others through a process called "invasion meltdown" (Parker et al. 1999; Grosholz 2005; Facon et al. 2006), as has been cited in several cases when the arrival of plant species was associated with the presence of large alien herbivores (Parker et al. 2006). The introduction of domestic and feral livestock results in degradation of the native flora affecting the whole landscape, decreasing the cover of the herbaceous layer and favouring the dominance of plants that are less susceptible to grazing, such as unpalatable (Huntly 1991; Vázquez 2002; Tood 2006), spiny or rosette species (Fernández-Alès et al. 1993), and of medium to low height species (Lavorel et al. 1999), increasing the richness and diversity of non-native plants in many cases (Chaneton et al. 2002; Vázquez 2002; Parker et al. 2006).

The introduction of herbivores results in higher impacts when introduced animals are functionally different from those already present, or thrive in greater densities in respect to those found naturally in the native community (Hobbs and Huenneke 1992). Large alien ungulates have produced smaller changes in the savannas of Africa and North America, possibly because large, hoofed, congregating grazing mammals already existed in the evolutionary history of these habitats, resulting in the evolution of grasses tolerant to heavy grazing—including both tussock and rhizomatous or turf grasses. The situation is different on the large South American plains where herbivores of this kind were not previously present and so dominant tussock grasses are intolerant to heavy grazing (Mack 1989). Similarly, the grazing intensity by domestic herbivores in southern South America is thought to be about ten times more than it was before their introduction (Oesterheld et al. 1992).

Grazing and trampling are the best known agents of change associated with the presence of introduced herbivores (Trimble and Mendel 1995; Vázquez 2002; Rook et al. 2004), whereas, in comparison, much less is known, about the impact of the accumulation of manure, which is a conspicuous effect of ungulate activity. Feral horses and other ungulates, e.g. vicuñas, guanacos, llamas and Przewalski horses (Bouman 1986; Vilá 1994; Dalmasso et al. 1999; Beever and Brussard 2000; King and Gurnell 2007), behave in a

particular way: herds or family groups deposit their dung at special sites, forming conspicuous mounds, called *dung piles* or *stud piles* (King and Gurnell 2007). It is thought that these accumulations mark the territory occupied by a male and they also serve as a signal for reproduction, letting a male know when females in his territory are on heat (Bouman 1986; Vilá 1994). Manure in dung piles increases the availability of soil nutrients, especially nitrogen and phosphorus (Nielsen 1997; Dai 2000; Aarons et al. 2004) and favours the conservation of humidity. This might contribute to the development of special patches of vegetation growing directly on manure and in its surroundings (Shiyomi et al. 1998; Dai 2000). In many ecosystems, the invasion of non-native plant species is dependent on this type of disturbance that creates appropriate microhabitats for their establishment, especially if the disturbance results in an increase in water and nutrients (Lake and Leishman 2004). These microhabitats can act as "invasion windows" fostering the establishment and potential dispersal of invasive alien species. Moreover, many species are transported in manure, which also contributes to their establishment (Dobbie et al. 1993; Malo and Suárez 1995a, b; Campbell and Gibson 2001; Wells and Lauenroth, 2007). Previous studies are concerned with analysis of the effects of manure of domestic livestock (Shiyomi et al. 1998; Dai 2000; Loucougaray et al. 2004), but no evaluation of the impact of wild herbivore dung piles has been undertaken, especially feral horses on natural ecosystems.

Horses have been present in the Argentine plains since the beginning of European colonization (Vázquez 2002) and they form feral populations in several natural and semi-natural grasslands and shrublands across the country (INBIAR, Database on Biological Invasions in Argentina 2007). A population of feral horses inhabits the Ernesto Tornquist Provincial Park (ETPP), one of the last few relicts in a relatively good state of conservation in the Pampas Phytogeographic Province (Scorolli 1999). The ETPP has provided a favourable habitat for the growth of this population which started with a nucleus of a stallion and four mares that were introduced in 1942 (Scorolli 1999), reaching a current density of 25 horses per km^2 (A. Scorolli, Departamento de Biología, Bioquímica y Farmacia, Universidad Nacional del Sur, pers. comm.). The presence and activity of feral horses in the area results in a highly altered matrix of low

grassland from which small areas of taller tussock grasses (*Paspalum quadrifarium*) emerge, in association with low, wet sites. It is known that grazing in the reserve results in a reduction of vegetation cover and height and increases the abundance of shrub species and unpalatable grasses (A.E. de Villalobos and S.M. Zalba pers. comm.), as well as the cover of alien species (Frangi and Bottino 1995; Long and Grassini 1997). However, the impact of dung piles on vegetation communities in the reserve had not been previously analyzed.

In this study, we assess the area directly affected by feral horse manure in the ETPP. We evaluate differences in plant communities growing on dung piles compared with control plots without manure, and describe the successional changes in dung pile vegetation over time and also discuss the possible role of dung piles in the introduction and dispersal of invasive plants in the reserve.

Methods

Study area

The study was carried out in the Ernesto Tornquist Provincial Park (ETPP), which has an area of approximately 6,700 ha in the Ventania mountains, between 38°00′–38°10′ S and 61°45′–62°08′ W, and includes mountains of around 1,000 m a.s.l. Climate is temperate, with an average annual temperature of 14°C (Burgos 1968). The average precipitation is 800 mm, with occasional snow in the winter.

The study area in the reserve consists of 2,000 ha of piedmont valleys with 5 –11% slopes, shallow soils and occasional rocky outcrops. In the absence of intensive grazing the grass canopy is 50–60 cm tall, dominated by *Piptochaetium hackelii*, *P. napostaense*, *Nassella melanosperma* and *Chascolytrum subaristatum* among others (Frangi and Bottino 1995). Tussock paspalum (*Paspalum quadrifarium*) is dominant on stream edges and in areas where the soil is saturated with water during rainy periods (Frangi and Bottino 1995).

Sampling design and statistical analysis

The sampling was carried out in January and February 2004 over an area where horses were active

(Scorolli 1999). Fifteen linear transects 50-m long were placed at random, in order to determine the percentage of the study area was directly covered by manure.

Fifteen dung piles >1 m in diameter were selected in order to avoid biases due to very small dung piles or isolated dung (i.e. dung not grouped in piles). The area of dung piles was calculated assigning each pile to either a circular or oval shape. The height of each pile was calculated as the average from five measurements taken in the centre of the pile and in the middle of their largest and smallest radiuses. Mean height was used to estimate how much time had passed since abandonment by the animals (age of the dung pile). Loss of humidity, decomposition of organic material and physical factors such as wind and rain, result in hardening and then disintegration and fragmentation of the manure over time and also in the loss of the boundaries of the lumps (Dai 2000). These changes in turn cause a decrease in the height of the pile and a concomitant increase in its area. Therefore, it can be assumed that dung height shows a negative correlation with time after abandonment of the pile and so it can be used as an indicator of its age. Alternatively, differences in dung height might respond to an original variation in the size of the pile; if dung height reduces with time, we would expect to find a negative correlation between both variables. Actually, the area of dung piles (2.80 ± 0.324 m^2, mean $\pm$ SE) and their height were negatively correlated ($R = -0.679$, $P < 0.01$, $n = 15$), supporting the idea of reductions in dung height and increases in size over time.

We determined the percentages of plant species cover and of bare manure by visual estimation for each dung pile. Plant species were collected and identified in the laboratory according to Zuloaga and Morrone (2007).

Plant species richness and Shannon's diversity index and evenness (Begon et al. 1996) were calculated for the vegetation on the dung piles. A linear regression analysis was undertaken between the height of the dung (as an indicator of the time passed since abandonment of the pile) and the total percentage of vegetation cover, species richness, Shannon's diversity index as well as the evenness in the whole dung pile.

A principal components analysis (PCA) was carried out using the correlation matrix between the percentage cover for each species in order to search for patterns in the floristic composition of the dung

piles. Occasional species (i.e. species present on only one dung pile with a percentage cover <1%) were excluded. As the first principal component explained a meaningful part of the total variation according to Kaiser–Guttman criterion (Legendre and Legendre 1998), we correlated it with dung pile height in order to describe plant species replacement with time.

Twenty 1-m^2 grassland controls placed at least at 10 m from the nearest dung pile were randomly selected. Species composition on these plots was assessed and compared with dung piles using step-wise discriminant analysis. A canonical variate analysis was used to determine which variables were more important in separating dung piles from grass-land controls. We also compared non-native species cover per square meter between control plots and dung piles by Kruskal–Wallis non-parametric analysis of variance.

Results

Manure cover at the study site was 254 ± 52 m^2 ha^{-1}, representing 2.5% of the total surface area. About 78 species were found growing directly on the dung piles, among which 35 (45%) were considered occasional. About 61 species were detected in control plots of which nine (15%) were considered occasional. The percentage of uncovered substrate (i.e. non-vegetated substrate covered only by manure) increased with the height of the dung pile, i.e. greater percentages of bare manure were associated with more recent dung piles
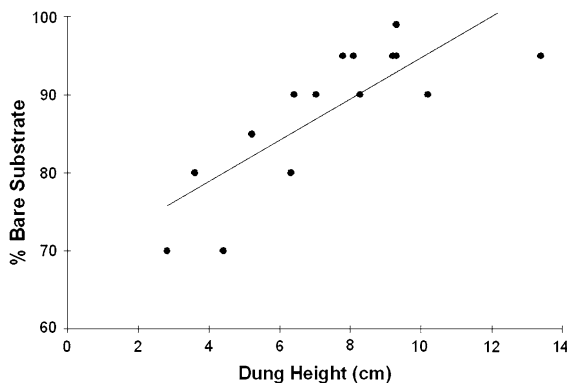

Fig. 1 Linear regression between the percentage of bare substrate and dung height. The height of the dung is used as a proxy inversely related to the time passed since the abandonment of the dung pile. $R^2 = 0.64$, $n = 15$, $P < 0.01$
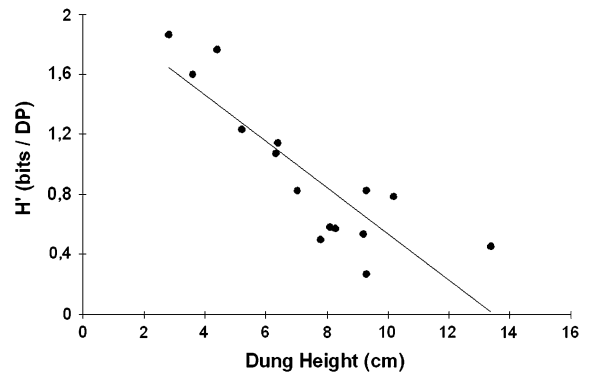

Fig. 2 Linear regression between Shannon's diversity (H′) and height of the dung piles. $R^2 = 0.725$, $n = 15$, $P < 0.01$. DP: Dung pile
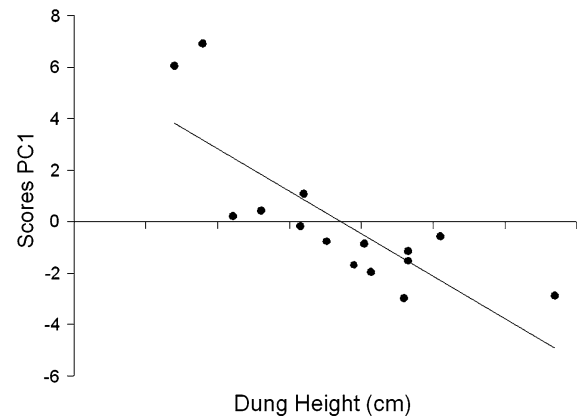

Fig. 3 Linear regression between dung piles scores on the first principal component (PC1) and the height of each pile

($R^2 = 0.640$) (Fig. 1). Shannon's diversity index increased significantly with reductions in height of the dung pile ($R^2 = 0.725$) (Fig. 2). The same occurred with species richness and evenness ($R^2 = 0.461$ and $R^2 = 0.701$, respectively). The first princi-pal component of the PCA obtained from the data of species cover in the dung piles explained 18% of the total variance, while the second component explained 13%. No clearly defined groups of dung piles were detected by their ordination on these first two axes.

The height of the dung pile was negatively corre-lated with the position of the dung piles on the first principal component (correlation coefficient = −0.798; $P < 0.01$) (Fig. 3). The ordering of dung piles on the first principal component reveals changes in the abundance of different species in association with the age of the dung pile, according to species loads in

Table 1 Correlation coefficients (R) between species cover and the coordinates of dung piles on the first principal component (PC1). Only species with correlation coefficients, equal to or greater than half the highest value obtained are shown

Species	R PC1
Nassella clarazii	0.9350
Evolvulus sericeus	0.8444
Chaptalia exscapa	0.7924
Cyperus aggregatus	0.6770
Paronychia spp.	0.6622
Pfaffia gnaphaloides	0.6468
Centaurea calcitrapa	0.5720
Conyza monorchis	0.5642
Piptochaetium medium	0.5275
Acmella decumbens	0.5181
Helenium radiatum	0.4879
Margyricarpus pinnatus	−0.2537
Hedeoma medium	−0.3438
Berroa gnaphalioides	−0.3499
Baccharis rufescens	−0.4058
Chevreulia sarmentosa	−0.4551

the first PCA axis (Table 1). Some species, e.g. *Eragrostis lugens, Eryngium nudicaule, Margyricarpus pinnatus* and *Pavonia cymbalaria,* were present in most of the dung piles and some of them, e.g. *Aristida spegazzinii, Pfaffia gnaphaloides* and *Piptochaetium stipoides,* showed greater cover in the oldest dung piles. Another group of plants including *Baccharis rufescens, Chevreulia sarmentosa, Hedeoma medium, Piptochaetium montevidense* and *Plantago myosuros* appeared in recent and middle aged dung piles. Other plants associated with dung piles of middle age continued to be present in the oldest ones, e.g. *Centaurea calcitrapa, Chaptalia exscapa, Cyperus aggregatus, Eleusine tristachya, Evolvulus sericeus, Nassella clarazii, Piptochaetium medium* and *Sporobolus indicus* (Table 2).
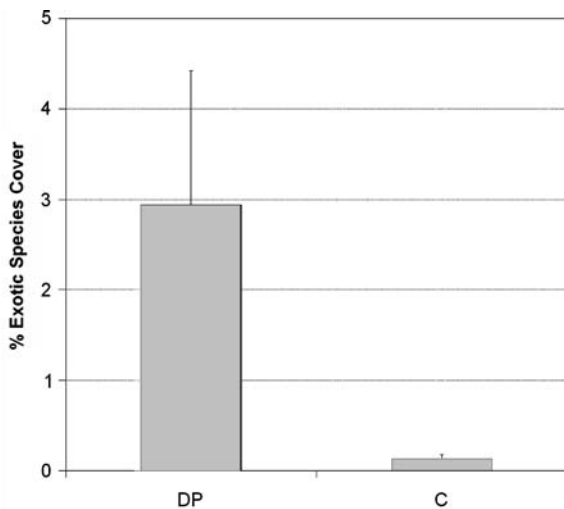
The best stepwise discriminating model obtained when comparing vegetation on the dung piles and in the control selected thirteen species from a total of 67 (Table 3). *Centaurea calcitrapa* and *Nassella clarazii* only appeared growing on the dung piles, while *Baccharis rufescens, Helenium radianum* and *Pfaffia gnaphaloides* showed higher cover on them. Alternatively, *Aristida spegazzinii, Baccharis stenophylla, Chaptalia exscapa, Eragrostis lugens, Juncus tenuis, Mimosa rocae* and *Piptochaetium stipoides* were consistently associated with the control plots. The discriminant function discriminates significantly between both vegetation groups (Wilk's-Lambda = 0.008, df = 13, $P < 0.001$) with a 100% of the samples correctly allocated.

Table 2 Species cover on dung piles and mean cover in control plots. Cover: **1**. 0–1%; **2**. 1–5%; **3**. 5–25%, **4**. 25–50%, **5**. 50–75%, **6**. 75–100%. Species were selected considering changes in abundance in association with piles of different height

Species	Dung Pile height (cm)															Control Plots
	13.4	10.2	9.3	9.3	9.2	8.3	8.1	7.8	7.0	6.4	6.3	5.2	4.4	3.6	2.8	
Piptochaetium stipoides	1	0	1	1	1	0	1	1	2	1	1	2	1	1	2	5
Margirucarpus pinnatus	1	1	1	0	1	1	0	1	1	1	0	1	1	0	1	2
Eragrostis lugens	1	1	1	0	1	0	0	0	1	1	1	0	1	1	1	3
Aristida spegazzini	1	1	1	0	1	0	1	1	0	1	0	0	2	1	0	2
Pavonia cymbalaria	1	1	1	0	1	0	0	0	1	2	0	1	1	1	0	2
Pfaffia gnaphalioides	1	1	1	0	0	0	0	0	1	1	1	0	1	1	2	1
Eryngium nudicaule	1	1	0	0	1	1	0	1	1	0	1	0	1	1	0	1
Baccharis rufescens	1	0	0	0	1	0	1	0	0	0	0	0	0	0	0	1
Hedeoma medium	1	0	1	0	1	0	0	0	0	1	0	0	0	0	0	1
Chevreulia sarmentosa	1	0	0	0	1	0	0	1	0	0	0	0	0	0	0	1
Piptochaetium haeckelii	0	2	0	0	0	0	0	0	1	0	0	0	0	0	0	0
Plantago myosurus	0	0	1	0	0	0	0	0	1	1	0	0	0	0	0	1
Piptochaetium medium	0	0	0	0	0	1	1	0	0	0	1	2	1	1	2	2
Eleusine tristachya	0	0	0	0	0	0	1	0	0	1	0	0	1	1	0	1
Cyperus aggregatus	0	0	0	0	0	0	0	1	0	0	0	0	0	1	1	1
Sporobolus indicus	0	0	0	0	0	0	0	0	1	1	0	1	0	1	0	0
Nassella clarazii	0	0	0	0	0	0	0	0	0	1	0	0	0	2	2	0
Evolvulus sericeus	0	0	0	0	0	0	0	0	0	1	0	0	0	1	1	1
Centaurea calcitrapa	0	0	0	0	0	0	0	0	0	0	2	2	3	0	3	0
Chaptalia exscapa	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	1

Table 3 Standardized canonical discriminant function coefficients (C) from canonical variate analysis on plant species cover between dung piles and grassland controls

Species	C
Centaurea calcitrapa	−2.021
Nassella clarazii	−1.360
Helenium radianum	−1.244
Juncus tenuis	−1.202
Baccharis crispa	−0.958
Baccharis rufescens	−0.624
Pfaffia gnaphalioides	0.870
Aristida spegazzinii	1.025
Eragrostis lugens	1.912
Mimosa rocae	1.199
Baccharis stenophylla	2.404
Chaptalia exscapa	3.047
Piptochaetium stipoides	3.435
Functions at group centroids	
Dung piles	−12.273
Grassland controls	9.205

Fig. 4 Alien species cover per square meter in dung piles (DP) and control plots (C). The bars represent standard deviation

Non-native species cover was higher in dung piles than in grassland control plots ($H = 4.69$, $P < 0.05$) (Fig. 4). Only one alien species, *Geranium dissectum*, was found growing in control plots with a mean cover of <0.15%. However, nine alien species were associated with dung piles with *Centaurea calcitrapa* being the most abundant species in some of the piles (Table 4).

Table 4 Alien plant species frequency (Freq.) and mean cover (M.C.) on dung piles and control plots

Species	Dung Pile ($n = 15$)		Control plots ($n = 20$)	
	Freq.	M.C. (%)	Freq.	M.C. (%)
Bromus hordeaceus	1	0.03	0	0
Centaurea calcitrapa	4	2.40	0	0
Echium plantagineum	1	0.20	0	0
Geranium dissectum	3	0.10	5	0.13
Petrorhagia nanteulli	2	0.07	0	0
Plantago lanceolata	1	0.03	0	0
Polycarpon tetraphyllum	1	0.03	0	0
Scleranthus annuus	1	0.03	0	0
Taraxacum officinale	1	0.03	0	0

Discussion

Despite the widespread presence of feral ungulates in grassland ecosystems, this is the first effort to assess the effect of feral horse dung piles on plant communities. The area covered by manure in the ETPP (ca. 2.5%) is greater than the results obtained for cattle dung by other authors in other ecosystems of the world. Dai (2000) found around 1% cover of cattle manure in a field on the island of Öland, in the south of Sweden. Omaliko (1981) reports that the percentage of surface covered by cattle manure reached 0.38% in a savanna area in Nigeria. Malo and Suárez (1995c), working in a Mediterranean pasture in Spain, found that the cover of cattle manure reached 0.5% of the area, varying during the year, and locally reaching 2%.

In our study, the percentage of bare substrate decreases with the age of the dung piles whereas species richness, diversity and evenness increase. It is possible that the initial impact of the accumulation of manure restricts the colonization of the site to a reduced group of species capable of tolerating the high concentration of salts that comes from the dung and urine of the horses. With the passage of time, environmental conditions may become less extreme, allowing the establishment of other species. The coexistence of both groups could explain the increase in total vegetation cover, richness, diversity and evenness that follows. This conclusion has to be taken with caution, as there is a risk of confounding

the effect of time with the concomitant increase in area that occurs as dung piles disintegrate. However, plant cover (shown as the complement of bare substrate in Fig. 1) increases more rapidly than dung piles area: since the area of the dung piles augments in a factor of 5 times (ca. 1 m² to ca. 5 m²), the percentage plant cover changes from ca. 1% to ca. 30% in the studied dung piles. On the other hand, the recorded increases in plant species richness and diversity greatly exceed those expected only by an increase in area, considering that minimal sampling area in this grasslands is about 1 m² (Zalba 1994).

A process of successional replacement of the vegetation becomes evident when the variation in the specific composition of dung piles over time is analyzed. The presence of a particular species would depend on its relative capacity to arrive at the site, germinate and become established, as well as to resist competitive replacement by new species. A group of plants (e.g. *Aristida spegazzinii, Piptochaetium stipoides, Pfaffia gnaphalioides*) that commonly appear in the plots sampled in the surrounding grassland matrix was also present on dung piles of different ages, showing greater cover in the older ones. It is possible that at least some of these species were present before the site was covered by dung and were capable of resisting the impact of manure, resprouting from underground structures, such as rhizomes, stolons, root suckers or bulbs, after the abandonment of the dung piles. Another group of species might initially respond in the same way, but then not be able to tolerate the environmental conditions in the dung piles in successive stages, or the competition from opportunistic species that take advantage of this habitat, and eventually disappearing. This group of plants appeared in young dung piles, but was not found in the oldest ones (e.g. *Hedeoma medium, Baccharis rufescens*). Finally, a third group of species was only associated with the lowest dung piles, indicating that they appeared after a certain time had passed after the abandonment of the dung pile (e.g. *Centaurea calcitrapa, Chaptalia exscapa, Nassella clarazii*).

Dung piles represent a sudden and significant disturbance due to the accumulation of a large amount of manure, with the consequent decrease in the cover of the underlying vegetation and the enrichment of nutrients and humidity. According to the Grime′s triangular model for representing plant strategies (CSR theory) (Grime 1977, 2001) plant competition is not important in recently disturbed areas with great availability of resources and rapid colonization. So, the initial stages of plant establishment on a dung pile would be determined by the survival of tolerant species that are already present in the site, plus pioneer species present in the soil seed bank, in seed rain or transported in the dung. Seeds of species of some genera recorded on dung piles in ETPP, e.g. *Carex, Conyza, Cyperus, Daucus, Eleusine, Erodium, Geranium, Juncus, Oxalis* and *Plantago*, among others, are able to survive the passage through the digestive tract of horses and other ungulates and are effectively dispersed by them (Malo and Suárez 1995b; Campbell and Gibson 2001; Cosyns and Hoffmann 2005; Mouissie et al. 2005). As time passes, the conditions in the dung piles would become more moderate, allowing the arrival of other species. Competition might then become the factor that structures the communities.

Changes in vegetation growing on dung piles respect to grassland controls should be taken with care considering that manure covers an important portion of the area and it is possible that control plots could have been covered by dung in the past. To have a real knowledge of the effect of manure, plots historically free of dung should be used, what is actually not possible due to the extensive use of the area for cattle ranching. Nevertheless, an effort has been made to select controls that were free of dung evidences and we think that the described restriction does not preclude drawing conclusions about the effect of dung piles.

The association of alien species with dung piles is of particular interest. In particular *Centaurea calcitrapa,* the most abundant non-native species found in the dung piles, is relatively uncommon in natural grasslands at the study area, but very frequent on road edges and other disturbed places (Long and Grassini 1997). During this study, *C. calcitrapa* was not recorded in grassland habitats away from the dung piles, but it was very frequent in old dung piles, exceeding percentage covers of 5%. The species was seen flowering on the piles, supporting the idea that it can complete its life cycle before the complete disintegration of the pile and so producing seeds that can disperse from there. Dung piles in which the species is present might therefore function as a source from where the species could colonize grassland

habitats in the future, provided that certain ecological, genetic and/or demographic changes occur, constituting a potential centre of dispersion of propagules over the area.

The case of *Nassella clarazii* is also interesting as it was only found in the oldest dung piles and the specimens found were more than 25-cm tall (A. Loydi, pers. observ.). *N. clarazii* is very palatable and sensitive to grazing (Rúgolo de Agrasar et al. 2005), and so it is not common in this intensively grazed ecosystem and in fact it was not found in the grassland control plots. Abundance of sensitive species to grazing seems to depend on refuge availability in both terrestrial and aquatic ecosystems (Quenga Kerr and Paul 1995; Lavorel et al. 1999; Callaway et al. 2000; Milchunas and Noy-Meir 2002; Oesterheld and Oyarzábal 2004; Callaway et al. 2005; Rebollo et al. 2005; Baraza et al. 2006; Graff et al. 2007). These refuges allow threatened organisms to escape from the effect of herbivores, avoiding local extinction in areas that are heavily grazed (Callaway et al. 2000). The presence of ungrazed specimens of *N. clarazii* associated with dung piles might be because of feral horses avoid grazing there and therefore these areas might act as refuges that favour palatable species. The tendency of domestic horses to avoid grazing in latrine areas has been reported by numerous authors (Nielsen 1997; Shiyomi et al. 1998; Loucougaray et al. 2004), however, Beever and Brussard (2000) and Lamoot et al. (2004) did not record this behaviour when analyzing populations of feral and free-ranging horses. Alternatively, the presence of the species might be related to the particular ecological conditions at dung piles and this could be further tested by experimental manipulation with exclosures.

It can be concluded that dung piles cause local changes in vegetation communities of Pampas grassland that may scale up both their spatial and temporal boundaries. These might still affect plant communities after the disintegration of the manure, the virtual disappearance of the piles and even after the eventual removal of the population of the invasive herbivore, mainly due to their role as facilitators of invasion processes. Dung piles in some cases may act as invasion windows and sources of propagules for invasive alien species, leading to potentially irreversible changes in the grassland. On the other hand, the presence of grazing-sensitive species on the dung piles is indicative of the effect of grazing outside their boundaries and also of the potential for recuperation of grassland by managing the stocking rate of non-native herbivores.

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