



Microhistological analysis of ancient camelid dung from the southern Argentinean Puna: Past vegetation composition and diet

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ABSTRACT

Identification of plant micro-remains in ancient dung is a tool used infrequently in archaeological studies to infer past vegetation diversity and herbivore diet. Through the microhistological analysis of dung of South American camelids from two micro-sectors of a rockshelter archaeological site in the Puna of Catamarca (Argentina), we identified plant micro-remains to illuminate our knowledge of the environment and the use of camelids during the late Holocene (540 ± 40 years BP). Grasses were the main plant category found in dung samples of both micro-sectors, followed by sedges (Cyperaceae), shrubs and annual herbs. However, relative abundance of several species and vegetation type varied between samples. Differences were also detected in the abundance of species from *vegas* (high Andean wetlands) and open areas. Our results could be explained by several factors, such as the season of the year in which dung was produced, differences in foraging habitats and foraging behavior of camelid species. Our work underscores the importance of microhistology as a useful tool for future paleoenvironmental studies. This method, combined with other reconstruction techniques (pollen, stable isotopes and macro-vegetation remains), has the potential to provide insights of paleovegetation histories and the use of wildlife by hunter-gatherers of the Southern Puna during the entire Holocene.

1. Introduction

Plant remains preserved in extant animal dung are considered an important source of information to infer animal diet, vegetation structure, habitat characteristics, and site ecology where the dung is found. Such remains may also contribute to improving our knowledge of past human populations. Among the most usual remains are vegetation macro-remains such as seeds, pollen and spherulites (the latter are microscopic structures of calcium carbonate produced in animal guts; Brochier et al., 1992; Canti, 1997). However, an alternative source of information, which has so far been infrequently used in archaeological studies is the identification of plant micro-remains, including the epidermis of leaves, stems, roots and, sometimes, flowers (Holechek et al., 1982). If the dung containing the micro-material is well preserved, it can give substantial information about past environmental conditions and cultural contexts (Velázquez et al., 2015). Moreover, the analysis of plant micro-remains can provide important complementary evidence to the other methodologies such as pollen analysis. Whereas with the former it is straightforward to perform taxonomic identifications to the species level, in the latter the level of taxonomic resolution usually does

not reach the level of genus or species (Caruso Fermé et al., 2018).

During the majority of the Holocene, the humans who have inhabited the Southern Argentinean Puna (Catamarca, Argentina) used several species of animals, mainly camelids such as guanacos (*Lama guanicoe*), vicuñas (*Vicugna vicugna*), and llamas (*Lama glama*) (Aschero and Martínez, 2001; Elkin, 1996; Mengoni Goñalons and Yacobaccio, 2006; Olivera, 1997; Panarello and Mondini, 2015; Urquiza and Aschero, 2014). Guanacos and vicuñas are the wild species that have inhabited the central Andean Puna since about 12,000–9000 years BP (Wheeler, 1995). The llama is the domestic variety of the guanaco and appeared around 4400 years BP in the Argentinean Puna (Aschero et al., 2012; Mengoni Goñalons and Yacobaccio, 2006; Olivera and Elkin, 1994; Urquiza and Aschero, 2014).

Among the three camelid species, only the remains of vicuñas and llamas dated to the late Holocene have been found in the majority of archaeological sites of Antofagasta de la Sierra, a locality in the Southern Puna (Aschero and Hocsman, 2011; Izeta, 2008; López Campeny et al., 2005; Olivera, 1997; Olivera et al., 2015; Ortiz and Urquiza, 2012; Urquiza et al., 2013; Urquiza and Aschero, 2014). In particular at the site of Punta de la Peña 4 (henceforth, PP4), humans

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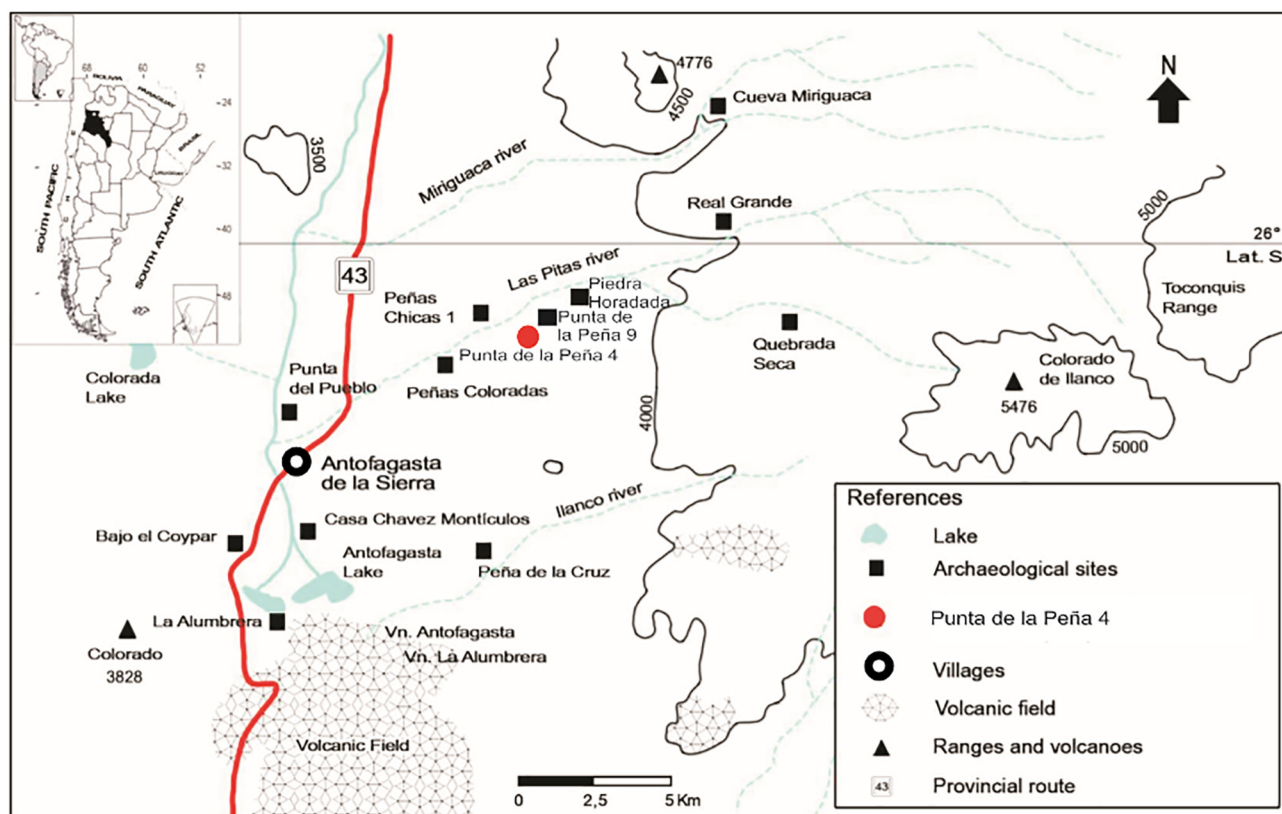


Fig. 1. Location and distribution map of archaeological sites in Antofagasta de la Sierra (Catamarca, Argentina). The location of the site Punta de la Peña 4 (PP4) is highlighted with a red circle. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

made wide use of camelids, valuing them as a source of meat, but also using their skin (leather), their coats (fibers) and their bones (Urquiza and Aschero, 2014). The composition of zooarchaeological assemblages attest to the varied use of the camelid remains and implied that these animals arrived virtually complete at the sites. Therefore, it is reasonable to assume that the slaughter and cleaning of intestinal tracts were carried out in situ (Urquiza and Aschero, 2014), preserving the waste and dung deposits within the associated stratigraphic records.

Vicuñas and llamas are still the most representative ungulates of the Puna. They are herbivorous mammals that forage on a wide variety of plants, so they can be considered a significant indicator of plant remains in the archaeological record (Browman, 1989; Hastorf and Wright, 1998). Vicuñas and llamas consume high proportions of grasses from a variety of sources from high-elevation grasslands to *vegas* (high Andean wetlands); this diet is supplemented with shrubs, sedges (Cyperaceae and Juncaceae species present exclusively in *vegas*), and annual herbs, although the proportions will vary between camelid species and depending upon seasons. Thus, during the wet season, the proportion of annual herbs increases while that of shrubs decreases, whereas in the dry season these proportions are reversed and consumption of *vegas* plants increases (Castellaro et al., 2004; Mosca Torres and Puig, 2010).

In Antofagasta de la Sierra the environmental conditions are currently extremely arid, vegetation productivity is low and pastures are poor. However, during the Holocene there were significant climatic changes associated with extreme wet and dry events (Olivera et al., 2004). Sedimentological, geomorphological and isotopic analyses indicate that from 8500 years BP there was a gradual process of increasing aridity, becoming markedly arid and warm around 6500–6300 years BP; however, this was followed by a period of high humidity starting around 4500–4000 years BP and ending around 1600 years BP (Olivera and Tchilinguirian, 2006; Tchilinguirian and Olivera, 2005). This end point marked a new phase of aridization,

which has underpinned the overall current environmental setting (Tchilinguirian and Olivera, 2014; Tchilinguirian et al., 2005). However, during the last 600 years, three periods of climatic instability have been identified (Grana, 2014): a wet phase (591–503 years BP); a wet and cold phase (490–450 years BP) and; a dry and possibly cold phase (449–40 years BP). Interestingly, despite these environmental changes during the Holocene, the vegetation has only varied in quantity and extent, but not in composition (Rodríguez, 2004).

The current study provides an analysis of micro-remains from camelid dung dated to around 540 ± 40 years BP (Urquiza and Aschero, 2014), thus corresponding to the wet phase described by Grana (2014). The goals of this study are twofold: to illuminate our knowledge of past environments during the late Holocene, and; to explore further the historical and contemporary accounts of the use of camelid resources by hunters and shepherds.

2. Materials and methods

2.1. Study area

Antofagasta de la Sierra (Catamarca, Argentina) is located in the southern Puna of Argentina and belongs to the Salt- or Desert-Puna (Troll, 1958). It is characterized by extensive areas of salt flats, formed as a result of low humidity and high evaporation (Olivera, 1988). The climate is cool and dry, with large daily and seasonal temperature ranges, intense frosts and scarce rainfall (0–200 mm/yr), the latter occurring almost entirely during summer. Phytogeographically, this area corresponds to the Puna Province of the Andean Domain (Cabrera, 1957), where the dominant vegetation is shrubby steppe, complemented by herbaceous, halophile and sammophile steppes. *Vegas* (wetlands with high plant cover) consist of short vegetation, where sedges are usually present in association with other small grasses (Cabrera and Willink, 1980).

2.2. Archaeological site

Punta de la Peña 4 (PP4) is an archaeological rockshelter site at 3650 m asl (26° 01' 40.52" S - 67° 20' 32.76" W), located in the middle reaches of the Las Pitás river (Fig. 1). It is interpreted as a residential base within an environment of shrub steppe called “*tolar*”, between the grassland steppe and the high *vegas*. The site provides good visibility of the valley offering a prime location for hunting, as well as annual water availability, firewood and opportunities for seasonal foraging (Urquiza and Aschero, 2014).

Urquiza and Aschero (2014) synthesized research conducted over more than 15 years at the site. This synthesis indicates continuity in the hunting practices of camelids and (water) birds during almost the entire Holocene, alternating with a mixed agropastoral economy. Vicuñas were used throughout the Holocene at the site, whereas llamas appeared during moments of transition (Middle-Late Holocene), confirming the presence of domesticated camelids in the southern Puna during this period.

PP4 offers an extensive archaeological record from the early to the late Holocene (ca. 8900–460 years BP), which can be divided into 8 stratigraphic levels (for detailed description of the characteristics of these levels, see Urquiza and Aschero, 2014; Rodríguez et al., 2003). Levels 3, 2 and 1 (ca. 760 to 460 BP) correspond to late occupation of the site, characterized by a layer of herbivore dung within the sediments which comprise a silty sand (Urquiza and Aschero, 2014). In this study, we investigated level 2, in particular two micro-sectors (G6b and H5a) within it from where the dung samples were obtained. This level has been dated to 540 ± 40 BP (UGA 9263) (Rodríguez, 2006; Urquiza and Aschero, 2014). This level is interpreted as belonging to a camelid herders settlement beneath the overhang (Urquiza and Aschero, 2014) and includes Camelidae remains (*V. vicugna* y *L. glama*), Rodentia, birds and Cervidae horns (*Hippocamelus antisensis*).

2.3. Analysis of dung samples

Samples of dung were collected from two micro-sectors in level 2 (G6b and H5a) and both were located within an area interpreted as “generalized area of activities” (Urquiza and Aschero, 2014), characterized by evidence of cleaning and abundant waste materials (e.g. dung, fleece, projectile points, bones). The micro-sector H5a was located in the discard area of level 2, at the limit between the drip line and the covered area of the overhang; whilst micro-sector G6b was located in the uncovered area (Fig. 2a and b).

Beneath the overhang of the site, preservation was good, and comprised cartilage tissue, tendons, hooves, leather, hair, feathers, wool, articulated bones, feces and textiles (Urquiza and Aschero, 2014; Urquiza and Echevarría, 2018). At this point it is opportune to highlight the excellent level of preservation of the dung samples, not only due to the dry climatic conditions of the area, but also because of the two layers of grasses (levels 1 to 3) that allowed the good preservation of other organic materials. The pH of the sediments is variable between neutral to alkaline (Brochier et al., 1992; Cabrera and Willink, 1980; Caruso Fermé et al., 2018) (Urquiza et al., 2009) and thus, it is possible to discard the impact which taphonomic processes could have affected the preservation of the dung.

In total, 31 organic-rich pellets were analyzed from sector G6b and 42 from H5b. To analyze the fragments present in the dung, we have used the microhistological technique described by Duci (1949) with some modifications to suit the vegetation of arid areas. This technique consists of the identification of plant epidermal tissues under a microscope (400× magnification), using diagnostic characteristics suitable for the differentiation of the species under study. These characteristics usually comprise epidermal cells, stomata, hairs and trichomes (Pelliza de Sbriller, 1993) and constitute reference standards that allow the taxonomic determination of plant fragments.

The technique was used to identify epidermis fragments of both

archaeological dung and modern plant reference material. The latter consisted of plant material collected in study area and herbarium specimens deposited in the Instituto Superior de Estudios Sociales (ISES), CCT CONICET Tucumán, Argentina. Reference microhistological slides and microphotographs were prepared for these plant species, and later used to identify the plant fragments from the dung slides. Reference slides were prepared by scraping (grasses and grass-like species), diaphanization (annual herbs, i.e., herbaceous dicotyledons), and maceration (shrubs). All the fragments were discolored in a solution of sodium hypochlorite (commercial bleach) and water. The epidermis samples were fixed on slides using glycerin as mounting medium. The histological slides of the epidermis were observed using a microscope (Olympus, CX21), equipped with a photographic-digital camera (Motic, MoticamX), connected to a computer. Photographs were taken of all the patterns and diagnostic traits observed.

Each dung sample was ground with a coffee grinder, then placed in a container with 70 ml of water and 30 ml of sodium hypochlorite for 20 min to discolor plant fragments. The sample was then washed and sieved through a 71 µm sieve. With a small spatula, part of the sample was placed on each slide with a drop of glycerin as a mounting medium. Fifteen microscope slides were made for each sample, and 70 fields per slide were systematically examined using a 400× magnification. Plant species were grouped into five categories according to their life forms: grasses; sedges; shrubs; annual herbs; and succulents, although the succulent category was not considered in the statistical analyses because it was rare (< 0.5%). Plant identifications were made to the genus level, and to the species level whenever possible.

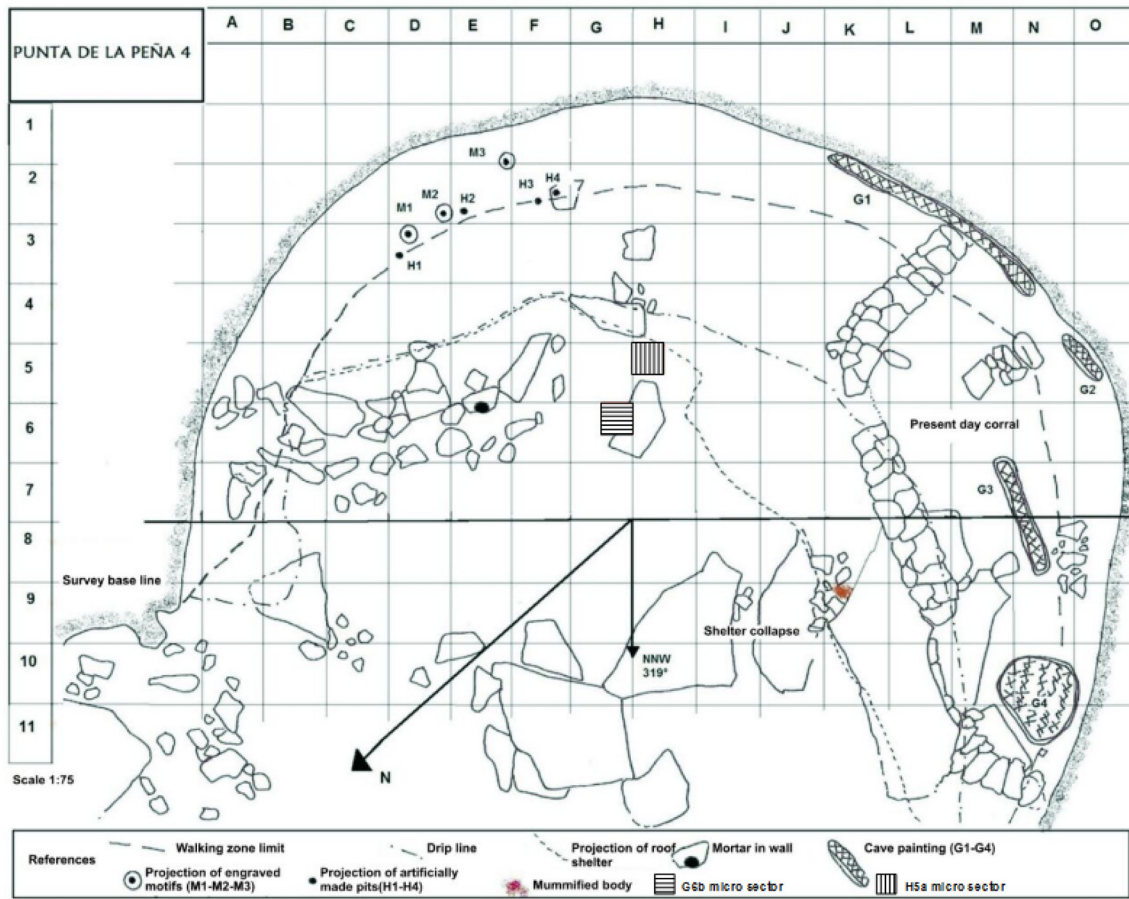
Currently, there are several procedures to help improve fragment recognition in the sample and the accuracy of quantification during the analyses (Catán et al., 2007; Dacar and Giannoni, 2001; Williams, 1969). Due to the differences in digestibility between plant remains, some species can be under- or overestimated in the quantification (Vavra et al., 1978). Therefore, several authors have proposed the use of quantification criteria or correction factors (Catán et al., 2007; David et al., 1983; Johnson and Wofford, 1983). We used the quantification criterion suggested by Catán et al. (2007) to count epidermal fragments of annual herbs. These authors suggest that the presence of three or more loose hairs per microscopic field of an identified fragment can help to avoid an underestimation of the species.

2.4. Statistical analysis

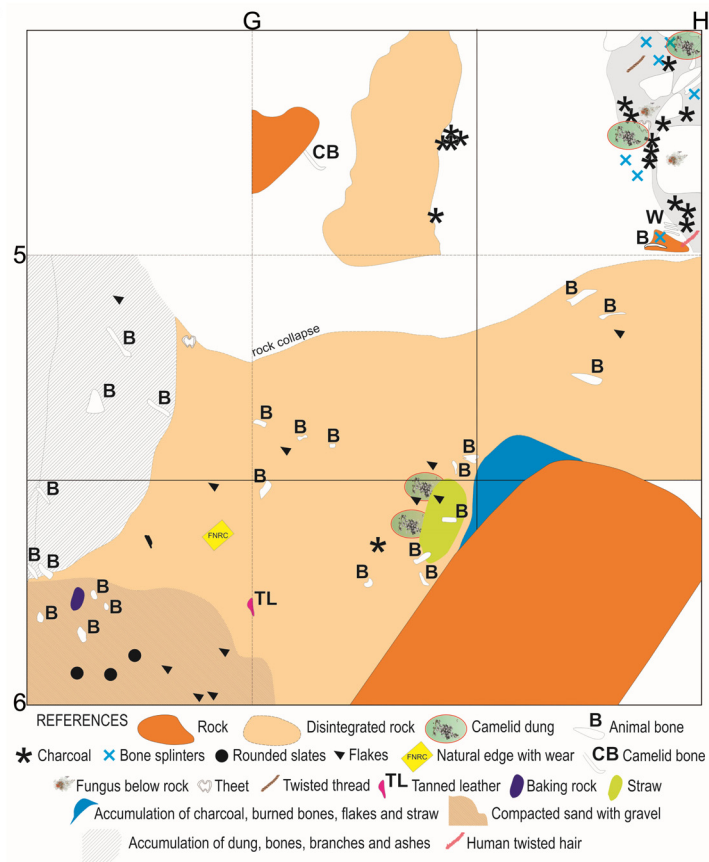
Shannon Index (H' ; Colwell and Futuyma, 1971) was used to estimate diversity of species remains in the dung samples, whereas Simpson Index was used to estimate species richness. Pielou's Evenness Index (J') was used to show how variable the number of fragments of each species was between dung samples. The latter index is the quotient between H' and H'_{\max} , where H'_{\max} is the maximum possible value of H' if every species is equally likely. J' is constrained between 0 and 1. The lower the variation in samples between the species (and the presence of a dominant species), the lower the value of J' . We used Jaccard's Similarity Index (Jaccard, 1908) to assess similarities in composition among micro-sectors. This index summarizes how much the composition of two sets of species overlaps, and it is the number of species shared between both sectors divided by the total number of species in the two samples.

To conduct a comparative analysis of the proportion of plant categories and species (or genus) between the G6b and H5b micro-sectors, we fitted generalized linear models (GLM) with a binomial error distribution (McCullagh and Nelder, 1989). In each model, the response variables were the proportions mentioned above and the micro-sector was the predictor factor. Only differences in plant species with relative frequencies above 5% were considered for comparisons between micro-sectors. All analyses were performed using the packages “MASS” (Venables and Ripley, 2002), “simba” (Juraskinski and Retzer, 2012) and “vegan” (Oksanen et al., 2018) of R statistical software, version 3.4.2 (R

a



b



(caption on next page)

Fig. 2. a. Plan view of PP4 archaeological site. Adapted from Rodríguez et al. (2006) by Urquiza and Aschero (2014). Scale 1:75.

b. Context plans of site PP4, level 2 (540 ± 40 yBP). Draft showing the location of the dung camelids remains in micro-sectors G5b and H5a. Each square is 1 × 1 m.

Table 1

Values of richness, diversity, evenness and similarity indexes of dung samples of camelids from G6b and H5b micro-sectors of the level 2 in PP4 archaeological site.

Index	G6b	H5b
Diversity (H')	2.21	2.01
Richness (S)	18	18
Pielou (J')	0.76	0.70
Jaccard	0.80	0.80

Core Team, 2017).

3. Results

In total, 21 species were identified in the dung samples (Table 1), with a maximum of 18 species per sector. Diversity (H') and evenness (J') were similar in both sectors, and the similarity index indicated that both dung assemblages shared 80% of the species (Table 1).

Grasses were the main plant category found in the samples of both micro-sectors, followed by sedges, shrubs and annual herbs (Table 2). The most important species were the grasses *Distichlis humilis*, *Deyeuxia eminens* and *D. chrysophylla*, the sedges *E. albibracteata* and *Carex* sp., and the shrub *Adesmia* sp. (Table 2). Among the annual herbs, it was not possible to distinguish between species or genera and only the family was identified, which in this case was Malvaceae. Fragments of these plant species were found almost exclusively in dung samples of G6b (Table 2).

In spite of their overall similarity in plant diversity, evenness and composition, these plant community attributes differed sharply among micro-sectors. For example, species fragments from the vega represented 49% from the total in G6b and 79% in H5b. The two micro-sectors differed also in the relative abundance of several plant species and, therefore, vegetation categories (Fig. 3). Grasses were significantly less abundant in samples of G6b than in H5b (48% vs. 73%), whereas

Table 2

Relative abundances of plant species founded in dung samples of camelids from G6b and H5a micro-sectors of the level 2 in PP4 archaeological site. (*): species found only in one micro-sector.

Genera/species	G6b (%)	H5b (%)
<i>Distichlis humilis</i>	31.3	8.4
<i>Deyeuxia eminens</i>	5.8	38.5
<i>Deyeuxia chrysophylla</i>	4.1	12.4
<i>Deyeuxia rigescens</i>	0.2	0.8
<i>Deyeuxia</i> sp.	1.9	4.9
<i>Hordeum halophilum</i>	0.9	0.3
<i>Stipa</i> sp. (*)	0.0	0.3
<i>Festuca</i> sp.	0.5	1.3
<i>Festuca chrysophylla</i>	3.2	5.9
Total grasses	48.0	72.7
<i>Lycium chananar</i> (*)	0.1	0.0
<i>Senecio</i> spp.	3.4	3.1
<i>Junellia</i> sp.	2.4	2.2
<i>Doniophyton</i> sp.	0.4	0.3
<i>Adesmia</i> spp.	6.1	4.0
<i>Artemisia copa</i> (*)	2.6	0.0
<i>Parastrephia</i> sp.	1.4	2.2
<i>Hofmanseggia</i> sp. (*)	0.0	0.2
Total shrubs	16.4	12.0
<i>Eleocharis albibracteata</i>	18.0	0.3
<i>Carex</i> sp.	11.8	14.9
Total grass-like	29.8	15.2
Malvaceae*	5.8	0.1
Total annual herbs	5.8	0.1

sedges and annual herbs were more abundant in G6b than in H5b (30% vs. 15% and, 6% vs. 0.1%, respectively; Fig. 3a. and Appendix A). Relative abundances of shrubs were similar between sites and represented less than the 20% of the fragments found in both dung samples.

Among grass species, *Distichlis humilis* was significantly more abundant in samples of G6b than in samples of H5b, whereas the abundance of *Deyeuxia eminens*, *D. chrysophylla* and *Festuca chrysophylla* were significantly lower in G6b than in H5b (Fig. 3b. and Appendix A). Finally, the relative abundance of two species of sedges, *E. albibracteata* and *Carex* sp. and of Malvaceae species, were significantly higher in samples of G6b than in H5b (Fig. 3b. and Appendix A).

4. Discussion

The 21 species found in the PP4 dung samples correspond to 35.2% of a total of 55 current species recorded in the study area by Rodríguez (2013). Although this percentage might be considered low, 9 of the 21 species have not been recorded previously across several archaeological sites in the Antofagasta de la Sierra, including PP4 (Rodríguez, 2008, 2013). The species (or genera) among grasses are: *Distichlis humilis*, *Deyeuxia chrysophylla* and *Hordeum halophilum*, the shrubs *Artemisia copa*, *Doniophyton* sp., *Junellia seriphoides* and *Lycium chananar*, the sedges *Carex* sp. and *Eleocharis* aff. *albibracteata*, and finally, species of the family Malvaceae. Therefore, we can consider that the micro-histological analysis of ancient dung can be a useful tool that complements our knowledge of past vegetation history constructed using other environmental techniques. The importance of this technique has also been mentioned by Caruso Fermé et al. (2018), who performed a multiproxy study of macro and micro-botanical remains recovered from Cerro Casa de Piedra 7, an archaeological site in Santa Cruz Province (Argentina), inhabited during the early and middle Holocene.

It is interesting to note that the preservation of annual herbs such as species of Malvaceae, is quite remarkable since preservation of ephemeral species is challenging under the natural climatic conditions. Therefore, its presence in dung samples provides an excellent opportunity to investigate seasonality and/or interfaces between dry and wet periods.

The high abundance of grasses compared to shrubs, sedges and annual herbs, matches that reported in other studies about seasonal use of vegetation by vicuñas and llamas (Borgnia et al., 2010; Castellaro et al., 2004; Mosca Torres and Puig, 2010; Posse and Livraghi, 1997). During the short wet season (summer), the vegetation of the Puna is characterized by the presence of annual herbs and by the higher nutritional quality of the plants foraged (Borgnia et al., 2010). Therefore, the presence of these species in samples of G6b and their absence in samples of H5b indicates that the dung was produced during the wet season. It is interesting to note that although the dung analyzed in this study was produced during a wet period according to Grana (2014), the absence and/or scarcity of some plant species demonstrate the presence of seasonality. In this regard, more guano from the same period (i.e. the last 600 years) should be analyzed from the same and from different archaeological sites to support, with greater certainty, what has been observed by the current authors.

Among the 9 species of grasses consumed, 4 were recorded above 5%, and two of them showed the highest relative abundance, *Distichlis humilis*, in G6b, and *Deyeuxia eminens*, in H5b. The first one is common in moist and saline soils, while the latter grows exclusively in damp soils, near rivers and in vegas (Cuello, 2006). The highest abundance of vega grasses, *D. eminens* and *D. chrysophylla*, were recorded in H5b compared to G6b; in addition to the considerable high abundance of *Carex* sp., this explains the high abundance of fragments of vega species

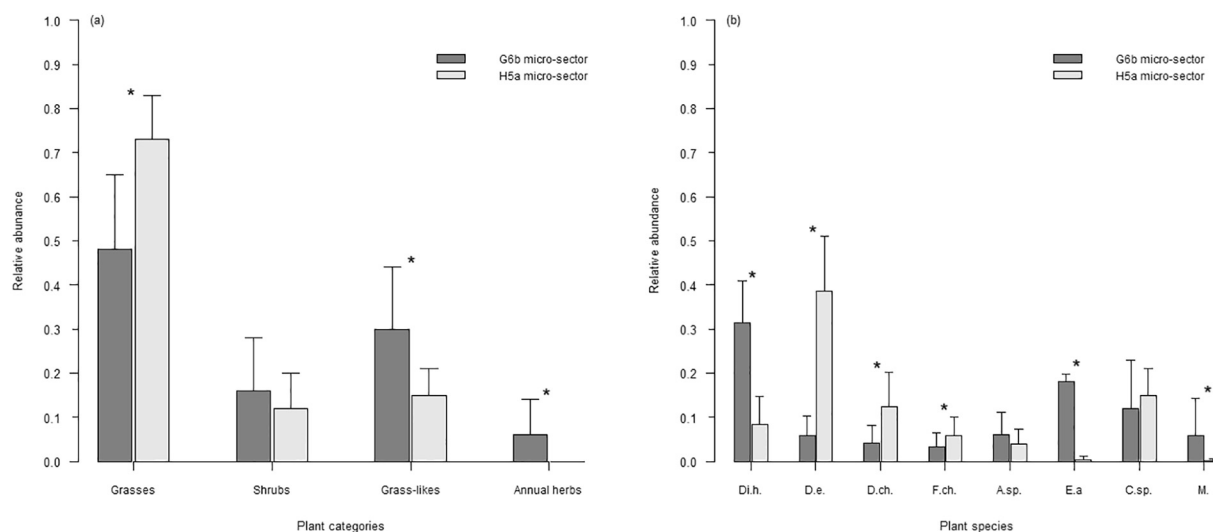


Fig. 3. Relative abundances of plant categories (a), and species (b) in dung samples of G5b and H5a micro-sectors in Punta de la Peña 4 archaeological site. *: Significant differences ($p \leq 0.05$) between sectors. Only those species detected in more than 5% were included in the graph. Di.h. *Distichlis humilis*; D.e.: *Deyeuxia eminens*; D.ch: *Deyeuxia chrysophylla*; F.ch: *Festuca chrysophylla*; A.sp: *Adesmia* sp.; E.a.: *Eleocharis albibracteata*; C.sp: *Carex* sp.; M: species of Family Malvaceae.

in H5b. In contrast, in the G6b sample, the fragments representing the vega species were mostly those of the sedges *Carex* sp. and *E. albibracteata*. The latter is practically absent in the H5b samples, possibly due to a decrease in its availability or absence in the vegas.

It is noteworthy that among the species of shrubs, those of the genus *Adesmia* were the most common in samples of both micro-sectors, although the overall relative abundances were low. Even though we were not always able to discriminate among the species, as they share some epidermal characteristics, most of the fragments identified correspond to *Adesmia horrida*. This species is also very frequent in the archaeological record of PP4 and other sites within our study area (Rodríguez, 2000, 2004, 2006, 2008), as well as at present (Cuello, 2006; Rodríguez, 2013). Throughout the Holocene, *A. horrida* has been mainly used as fuel because its wood has a high heat value (Rodríguez, 2008). On the other hand, this species is important in the diet of vicuñas in Laguna Blanca, particularly during the dry season (Benítez et al., 2006; Borgnia et al., 2010). Therefore, it is possible to consider that the genus *Adesmia* was a key resource not only for the settlers of Antofagasta de la Sierra, but also for camelids, and it is still important nowadays.

Due to the differences found in the relative abundances between micro-sectors, we could infer that the camelid individuals that produced the dung of G6b used vegas and open areas to forage, whereas those producing the dung of H5b fed mainly in vegas or rivers. In both cases, and in spite of the differences between sectors, the use of plant species from the vega is substantial. Studies on the use of vegas by llamas and vicuñas argue that during the dry season or in times of drought, these habitats are preferentially used by camelids due to their constant supply of forage and water (Borgnia et al., 2010; Castellaro et al., 2004; Mosca Torres and Puig, 2012).

Regarding the camelids that produced the dung, it was not possible to differentiate between species according to the size and shape of the dung. However, if we consider the origin of the plant species found, we can infer that dung of G6b could be produced by vicuñas, whereas dung samples of H5b could belong to llamas. The high percentage of vega species found in the H5b dung, agrees with the observation of Rodríguez (2004) on the different hunting strategies of the hunter-gatherers that inhabited the Puna. This author considers that llamas were raised in the *tolar* and the *vega* near the site, while the vicuñas were hunted in the grasslands (5 km from PP4; Rodríguez et al., 2006). However, there is also the possibility that the vicuñas were hunted in the *vega* while they were drinking and foraging. Several authors have

observed that vicuñas leave their territory once or twice a day to move to water sources to meet their daily survival needs (Mosca Torres and Puig, 2012; Vilá and Roig, 1992). Therefore, hunters could have taken advantage of this behavior to secure their prey without moving great distances. This idea is corroborated by the research of Urquiza and Aschero (2014) who determined for level 2 a similar percentage of llama bones (21,2%) to those of vicuñas (22,34%); primary processing of both species would have been undertaken inside the overhang with the dung removed from the abdominal cavity during evisceration.

Microhistological analyses of contemporary dung samples of vicuñas and llamas are necessary to further explore the origins of dung collected from archaeological sites. Furthermore, dung samples of both species should be collected in different seasons of the year and across a range of different habitats (*grassland*, *tolar* and *vega*) to compare with ancient samples.

5. Conclusions

Results of microhistological analysis of camelid dung presented here show differences in plant species relative abundances between micro-sectors dated to the same period of the late Holocene. The main differences detected were the variations in the abundance of species from the *vega* vs. species from more open and dry areas (*tolar* and *grassland*). These differences reinforce the evidence that the site was occupied during different seasons of the year (Rodríguez, 2004; Urquiza and Aschero, 2014). Moreover, the results show that the camelids that produced the dung were probably hunted in different habitats within the area of the Puna; however, we could not differentiate between vicuñas and llamas.

While complementary analyses are needed to improve the interpretations and inferences of the results reported here, we consider that they provide a significant starting point to demonstrate the potential of microhistological techniques as a very useful and simple tool for future paleoenvironmental reconstruction studies. One of the major advantages of this technique is that in many cases plant fragments can be taxonomically identified to the level of genus or species, providing a wide range of information for vegetation reconstructions. Therefore, this technique, combined with other reconstruction proxies such as pollen, stable isotopes and macro-vegetation remains, would help to improve our knowledge, not only of the paleovegetation record during the entire Holocene, but also of the use of faunal resources by the former settlers of Antofagasta de la Sierra.

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Appendix A. Appendix

Generalized linear models (GLM) with a Binomial distribution comparing relative abundances of plant categories and species between dung samples of G6b and H5a micro-sectors in PP4 archaeological site. The response variables are the relative abundances of plant categories and species, and the explanatory factors are the two micro-sectors, G6b and H5a, being G6b the reference level. Di.h: *Distichlis humilis*; D.e.: *Deyeuxia eminens*; D.ch: *Deyeuxia chrysophylla*; F.ch: *Festuca chrysophylla*; A.sp: *Adesmia* sp.; E.a.: *Eleocharis alibracteata*; C.sp: *Carex* sp.; M.: species of Family *Malvaceae*.

Model	Variables	Estimate	Std. error	Z value	p Value
Plant categories					
Grasses ~ micro-sector, family = binomial (link = "logit")	Intercept	-0.734	0.066	-11.13	< 0.001
	Micro-sector	0.415	0.083	4.99	< 0.001
Shrubs ~ micro-sector, family = binomial (link = "logit")	Intercept	-1.806	0.169	-10.69	< 0.001
	Micro-sector	-0.318	0.239	-1.33	0.180
Grass-likes ~ micro-sector, family = binomial (link = "logit")	Intercept	-1.212	0.104	-11.66	< 0.001
	Micro-sector	-0.664	0.158	-4.19	< 0.001
Annual herbs ~ micro-sector, family = binomial (link = "logit")	Intercept	-2.849	0.266	-10.70	< 0.001
	Micro-sector	-4.165	1.831	-2.27	0.023
Plant species					
Di.h. ~ micro-sector, family = binomial (link = "logit")	Intercept	-1.161	0.096	-12.04	< 0.001
	Micro-sector	-1.321	0.177	-7.48	< 0.001
D.e. ~ micro-sector, family = binomial (link = "logit")	Intercept	-2.849	0.192	-14.82	< 0.001
	Micro-sector	1.894	0.206	9.19	< 0.001
D.ch ~ micro-sector, family = binomial (link = "logit")	Intercept	-3.185	0.255	-12.47	< 0.001
	Micro-sector	1.099	0.288	3.81	< 0.001
F.ch ~ micro-sector, family = binomial (link = "logit")	Intercept	-3.445	0.248	-13.88	< 0.001
	Micro-sector	0.605	0.296	2.04	0.041
A.sp ~ micro-sector, family = binomial (link = "logit")	Intercept	-2.789	0.181	-15.38	< 0.001
	Micro-sector	-0.440	0.266	-1.65	0.098
E.a. ~ micro-sector, family = binomial (link = "logit")	Intercept	-1.717	0.191	-8.97	< 0.001
	Micro-sector	-4.199	1.270	-3.31	< 0.001
C.sp ~ micro-sector, family = binomial (link = "logit")	Intercept	-2.135	0.180	-11.89	< 0.001
	Micro-sector	0.239	0.228	1.05	0.290
M. ~ micro-sector, family = binomial (link = "logit")	Intercept	-2.849	0.266	-10.70	< 0.001
	Micro-sector	-4.165	1.831	-2.27	0.023

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