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Filling gaps in the seed dispersal effectiveness model for *Prosopis flexuosa*: quality of seed treatment in the digestive tract of native animals

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Abstract

For endozoochorous species, the quality component of seed dispersal effectiveness depends in part on the treatment seeds receive in the animal's gut. Covering a variety of taxa, diet, digestion system and body size of *Prosopis flexuosa* seed dispersers, we analysed differences among species in (1) mean retention time of ingested seeds, (2) recovery of viable seeds, (3) seed germination in comparison with seeds collected from trees and (4) germination of seeds after two different periods of retention in the gut. Feeding experiments were conducted with captive individuals of *Dolichotis patagonum*, *Lycalopex gymnocercus*, *Rhea americana*, *Chelonoidis chilensis* and *Lama guanicoe*. On the first day, we provided them with fruits containing controlled amounts of seed, and on the subsequent days, we collected faeces in order to recover seeds. We performed germination and viability tests on seeds coming from faeces and collected from trees. The results showed differences among species in the mean retention time of seeds. *Chelonoidis chilensis* had the longest mean retention time, but its effect on seed recovery and germination was similar to that of the other species, except for *L. guanicoe*, which showed the lowest seed recovery. When scarification and promotion of seed germination were considered, herbivorous mammals and tortoises (*L. guanicoe*, *D. patagonum* and *C. chilensis*) were the ones increasing germinability, whereas *R. americana* and *L. gymnocercus* did not significantly increase final seed germination percentage, which was similar to that for seeds collected from trees. *P. flexuosa* seeds receive a variety of treatments from endozoochorous dispersers, which might result in an overall fitness benefit for a plant living in unpredictable environments.

Introduction

Seed dispersal involves horizontal and vertical movements of seeds away from the parent plant (Chambers and MacMahon, 1994). It is a key process in the life history of plants that allows them to occupy new environments (Howe and Smallwood, 1982) and to escape the attack of pathogens, mortality by predation and seedling competition under the parent plant (Janzen, 1970). At a larger scale, long-distance dispersal may also be crucial for plants, driving species metapopulation structure and dynamics, gene flow, colonization opportunities and range expansion, and becomes particularly relevant in response to climate change and habitat fragmentation (Cain et al., 2000). Different abiotic (e.g. wind and water) and biotic agents (animals) are involved in fruit and seed dispersal until seeds reach the place where they will germinate and a new plant will establish (van der Pijl, 1982). Many plants have developed mutualistic relationships with animals to assure seed dispersal, in some cases offering attractive and nutritious fruits as rewards to endozoochorous dispersers (Jordano, 2000; Jordano et al., 2011).

In temperate communities, more than 60% of tree species are dispersed by vertebrates, and it has been estimated that at least 50% and often 75% or more of the tree species in tropical forests have fleshy fruits adapted for animal dispersal (Howe and Smallwood, 1982). Endozoochory involves fruit consumption by animals and seed dispersal through defecation; therefore, seed retention time in the digestive tract and distance travelled by animals during said time will affect seed dispersal distances (Nathan and Muller-Landau, 2000). Added to this, seeds can be scarified during the digestive process, increasing germination speed but with the risk of also increasing seed mortality if seed coats weaken too much. However, final seedling establishment depends on predation or subsequent movements of seeds from faeces, and on availability of suitable sites for plant establishment (Janzen et al., 1985).

Most seed dispersal studies over the last decades have taken the approach based on the seed dispersal effectiveness (SDE) framework (Schupp, 1993; Schupp et al., 2010). Disperser effectiveness was defined as the contribution a disperser makes to plant fitness, ideally measured as the number of reproductive adults recruited through the activity of a dispersal agent (Schupp, 1993). Later, it was proposed that SDE is a broader and more comprehensive concept, in that, it not only considers the effectiveness of dispersal that a single animal species provides but also the whole dispersal effectiveness that a plant receives from the assemblage of dispersers (Schupp et al., 2010). In addition, it is important not only to consider the effectiveness of seed disperser animals in plant fitness and population dynamics but also to highlight the benefits that animals obtain from seed dispersal mutualism, in the form of effective food resource provisioning (Schupp et al., 2017). According to the SDE model applied to endozoochorous seed dispersal systems, the effectiveness of animals as seed dispersers depend on two components, the quantity of dispersal, or number of seeds dispersed, and the quality of dispersal, which depends on the quality of the treatment in the digestive tract and the quality of seed deposition, which in turn determines the probability that a dispersed seed produces a new recruit (Schupp, 1993; Schupp et al., 2010).

The quality of seed treatment in the digestive tract depends on the animal's morphological and physiological characteristics, such as digestive system, body mass and diet (Traveset, 1998). During the passage, through the mechanical action of digestion, a seed's coat or endocarp can be altered by chewing. When the seed is consumed with other hard food items, including other seeds, it is likely to suffer mechanical breakdown (Samuels and Levey, 2005). Mechanical alteration of the seed coat or endocarp may facilitate imbibition at deposition sites, thus promoting germination. Alternatively, the gut's mechanical action may also destroy seeds (Samuels and Levey, 2005). Through the chemical action of the digestive tract, fluids may alter the seed coat or endocarp, thus affecting germination patterns. Digestive enzymes and stomach acids serve to break foods down, including seeds. This chemical treatment may also be affected by symbiotic bacteria and protozoa, which can digest structural cell wall polymers during fermentation (Traveset, 1998; Traveset et al., 2007).

Previous studies have shown that seed treatment in the digestive tract of animals has a more significant effect on seeds of trees from temperate than from tropical regions (Traveset, 1998; Traveset and Verdú, 2002). An example in the temperate Monte region could be the key tree species *Prosopis flexuosa* DC. (Fabaceae, Mimosoideae) (Álvarez and Villagra, 2009). The fruit of *P. flexuosa* is a modified indehiscent pod with a thin epicarp, a mesocarp that can be fleshy, sugary or fibrous and several endocarp segments (Burkart, 1976). Seed germination is hindered by physical dormancy and seed scarification by different agents increases germination (Catalán and Balzarini, 1992; Peláez et al., 1992; Peinetti et al., 1993; Campos and Ojeda, 1997; Ortega-Baes et al., 2002; Campos et al., 2008). Only a few days after reaching the soil, *P. flexuosa* fruits are removed by animals with different functional roles in the dispersal of the seeds. On the one hand, some opportunistic frugivores disperse seeds by endozoochory, such as medium-sized native mammals (e.g. *Dolichotis patagonum*, *Lagostomus maximus* and *Lycalopex gymnocercus*), large native herbivores (e.g. *Lama guanicoe* and *Rhea americana*) and non-native animals (e.g. *Lepus europaeus*, *Sus scrofa*, *Equus asinus* and *Bos taurus*; Campos and Ojeda, 1997; Campos et al., 2008, 2011, 2018; Campos and Velez, 2015). On the other hand, small rodents (e.g. *Graomys griseoflavus*, *Akodon dolores*, *Eligmodontia typus*, *Calomys musculinus* and *Microcavia maenas*) practice food hoarding, making both larderhoards and scatter-hoards with different effects on seed survival (Campos et al., 2007, 2017; Giannoni et al., 2013). In this relationship among *P. flexuosa* and its assemblage of dispersers, previous studies have shown that quantitative aspects of SDE directly related to animal visits and fruit removal are affected by changes in habitat heterogeneity under different land uses (Campos et al., 2016; Bessega et al., 2017; Tabeni et al., 2017; Miguel et al., 2017, 2018a,b). Although the development of the SDE model for *P. flexuosa* began years ago, using an approach that combines field and laboratory experiments, the aim of the present study is to provide data to fill in some gaps in the model. To do this, we focused on the experimental assessment of seed treatment quality in the digestive tract of some native animals. As was pointed out by Picard et al. (2015), studies of this kind remain rare because they rely on individual monitoring in controlled conditions, with heavy logistic constraints and associated with small sample sizes, mainly when wild species are considered. In particular, it remains important to establish whether variations occur among different species of animals consuming the same fruits and whether such variations could be attributed to differences among animal species.

We tested differences among five disperser species covering a variety of taxa, diet, digestive system and body size: *Dolichotis patagonum* (Mammalia, Rodentia; herbivore; hindgut fermenter; ≈8 kg), *Lycalopex gymnocercus* (Mammalia, Carnivora; carnivore and opportunistic frugivore; monogastric hindgut fermenter; ≈6 kg), *Rhea americana* (Birds, Rheiform; herbivore; hindgut fermenter; ≈30 kg), *Chelonoides chilensis* (Reptilia, Testudines; herbivore; hindgut fermenter; ≈1.5 kg) and *Lama guanicoe* (Mammalia, Artiodactyla; herbivore; foregut fermenter; ≈90 kg). The animal species were assumed to have different retention times and digestive treatment effects because of their variation in taxa, digestive system and body mass (Illius and Gordon, 1993). Our objective was to analyse differences among species in the quality of seed treatment in the digestive tracts considering: (1) mean retention time of ingested seeds, (2) recovery of viable seeds, (3) seed germination in comparison with seeds collected from trees and (4) germination of seeds after two different periods of retention in the gut. We tested three predictions:

1. Because *L. guanicoe* is a foregut fermenter with a digestive process that includes rumination ('ruminant-like'; Hume, 1989) and tortoises, such as *C. chilensis*, have low metabolic rate and food intake (Bjorndal, 1997), the mean seed retention time will be longer in these species compared to the others.
2. Because of the long mean retention time, total recovery and germination will be lower for seeds ingested by *L. guanicoe* and *C. chilensis* than for those consumed by the other species.
3. Germination will be higher for seeds recovered during the first period of passage through the digestive tracts of all species.

Materials and methods

Feeding and seed recovery experiments

The fruits of *P. flexuosa* were collected from 20 adult trees randomly selected in Ischigualasto Provincial Park (29°55′S, 68°

Table 1. Animal species that ingested *P. flexuosa* seeds

Species	No. of offered seeds	Mean of ingested seeds	Duration of experiment (total days)	Mean retention time (days) mean (±SE)
Dolichotis patagonum	100	98.43	8	1.61 (±0.19) A
Lycalopex gymnocercus	100	54.67	14	2.88 (±0.38) AB
Lama guanicoe	300	213.20	12	3.78 (±0.29) AB
Rhea americana	300	255.40	14	2.93 (±0.53) AB
Chelonoidis chilensis	50	44	35	16.29 (±3.65) B

Experimental details are shown. Letters indicate differences in mean retention time (days) among species.

05′W, San Juan Province) during the fruiting seasons (January to March) of 2014–2017. Fruits were stored in paper bags in a refrigerator at 5°C until the start of experiments, following the methodology proposed by Cony (1993) for preserving seeds. The feeding experiments were conducted with captive animals from three institutions: the Ecological Reserve and Wildlife Rehabilitation Centre (Mendoza), the Mendoza Zoo and the Centre for Rehabilitation of Wild Fauna, Environmental Education and Recreation (San Juan). Animals were kept and handled according to the Animal Care and Use Committee of the American Society of Mammalogists (Sikes et al., 2011) and conforming to the legal requirements of the Agencies of Renewable Natural Resources of Mendoza and San Juan (No. 444/2016). Experiments were performed from November 2016 to December 2018 with captive adult individuals of *D. patagonum* ($N = 8$), *L. gymnocercus* ($N = 3$), *L. guanicoe* ($N = 6$), *R. americana* ($N = 5$) and *C. chilensis* ($N = 6$). Each animal was isolated in a clean enclosure and fed on its usual food and with drinking water provided *ad libitum*; therefore, no adaptation period was needed before the experiments. The selected individuals showed no signs of stress during the trials because they had been kept in captivity since an early age and were used to human presence. They were active and in good health conditions throughout the study. During the first day of the trial, and for once, we offered them fruits containing controlled amounts of seed (between 30 and 300 seeds depending on the species) mixed with their usual food to facilitate a good acclimation to the experimental condition. We selected apparently undamaged fruits, discarding by external observation the ones with holes in the epicarp produced by the emergence of adult insects (Velez et al., 2018). We quantified the number of seeds provided, assuming a single seed in each pod segment. We began the experiments in the morning, ensuring that each animal ingested as many seeds as possible (Table 1). We interrupted the first-day feeding session when it appeared that all seeds had been ingested or when the animal refused to eat, and the remaining seeds were removed. The days following ingestion of the seeds, each animal had free access to freshwater and received its usual food every day. The faeces of each individual were collected daily and stored in labelled paper bags until processing. To ensure that we covered retention time until reaching a plateau in the cumulative proportion of retrieved seeds when it was logistically possible, we collected all fresh faeces for 2 weeks, except for tortoise faeces that were collected for 35 d. To recover the seeds, each faecal sample was thoroughly washed under running water in a sieve (71 µm). Seeds were dried at room temperature and stored in the laboratory until the germination experiment.

Germination and viability of seeds

Germination and viability were measured for seeds ingested by animals. In July 2018 and May 2019, germination tests were performed in incubators (Precision GCA Corporation, Scientific Model 818, Chicago, Illinois, USA) in the dark, and at a constant temperature of 30°C (Cony and Trione, 1996). Seeds were placed in sterile, plastic Petri dishes (9 cm diameter) containing filter paper disks resting on a thin layer of cotton, all materials having previously been sterilized. Ingested seeds were previously immersed in a solution of sodium hypochlorite (2%) for 10 min to remove fungi and superficial bacteria (Sauer and Burroughs, 1986). Dishes were initially moistened with a suspension of Captan fungicide at 2 g l^{-1}, being thereafter moistened as needed with sterile water. Apparently, healthy seeds recovered from animal faeces were cultivated, with sample sizes determined by seed availability. Seeds recovered from each individual by deposition day were kept in separate Petri dishes containing a maximum of 25 seeds. Dishes were randomly repositioned every week to avoid a chamber position effect. Germination, identified as visible radicle protrusion, was recorded every day for 30 d. In order to know the viability of the seeds offered to animals considering the condition and provenance of seeds, we carried out germination tests with seeds collected from 20 trees. Collected seeds were mixed in a pool. We used visually healthy seeds with endocarps and made 25 replicates of 20 seeds. It should be noted that there is a 32% pre-dispersal loss of *P. flexuosa* seeds due to abortion and insect predation. In many cases, insects die during development and do not emerge, then endocarps remain intact and seed loss cannot be externally determined (Velez et al., 2018).

To establish whether seeds that had failed to germinate in the previous experiments were viable, non-germinated seeds from trees and ingested by animals were subjected to the standard treatment with a 2,3,5-triphenyltetrazolium chloride test (Pili-Sevilla, 1987), which detects seed viability by staining the embryo tissue pink/red. Then, those seeds that germinated and those that were positive in the viability test were considered viable seeds.

Data analysis

Seed recovery was calculated as the proportion of viable seeds daily removed from faeces to the viable seeds consumed by individuals. Because animals were fed on whole fruits containing viable and non-viable seeds, the pre-ingestion loss was estimated using the seeds collected from trees.

Mean retention time is the time that the average digested particle remains in the digestive tract (Warner, 1981) or the time for

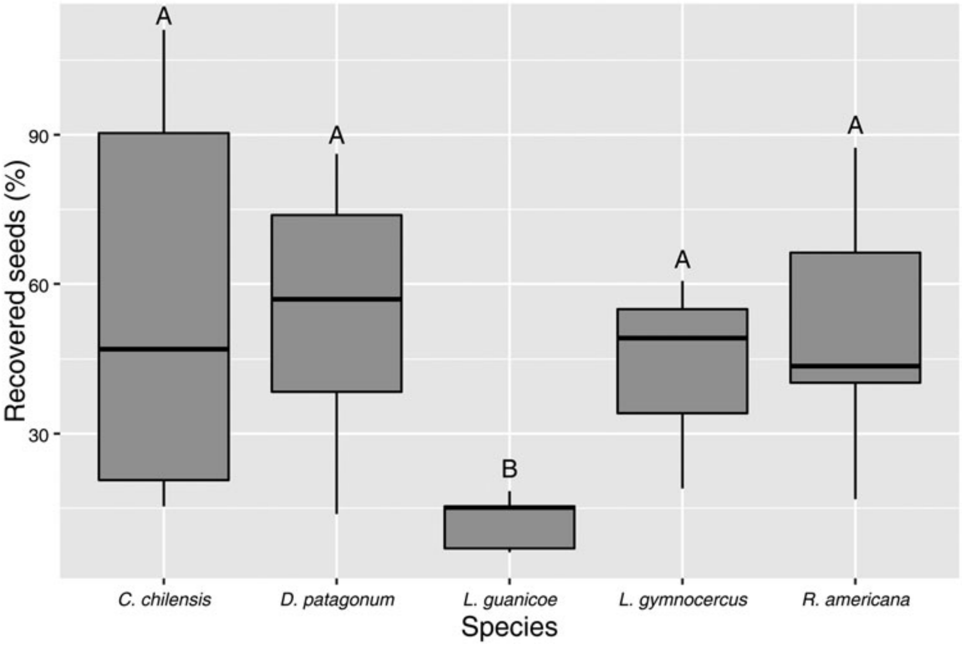

Fig. 1. Total percentages of recovered seeds of *P. flexuosa* after passing the digestive tracts of animal species. Letters show significant differences among species. The line in the box indicates the median value of the data. The upper and lower hinges of the box indicate the 75th and 25th percentiles of the dataset, respectively. The ends of the vertical lines indicate the minimum and maximum data values.

a 50% seed recovery. Expressing the results in terms of mean retention time is a convenient method for comparing the curves of cumulative seed recovery over time along their entire lengths (Castle, 1956). The mean retention time of seeds ingested by each individual was computed as the weighted sum of seeds recovered per day related to the total number of seeds recovered (Gardener et al., 1993; Razanamandranto et al., 2004). Differences among species in mean retention time were analysed using a Kruskal–Wallis rank-sum test with an *a posteriori* pairwise Wilcox test.

In order to analyse differences in the recovery of seeds ingested by species, we used a generalized linear mixed model (GLMM) with a binomial error distribution and a logit link function because there was no overdispersion. The total number of seeds recovered from faeces was used as a numerator in the response variable, and the number of seeds ingested by individuals as a denominator. Individuals were considered a random effect nested within species.

Differences in germination were analysed between seeds recovered from faeces and seeds collected from trees using a GLMM with a binomial error distribution and a logit link function. The total number of germinated seeds was used as a numerator in the response variable. The number of seeds recovered from faeces of individuals or the number of viable seeds in the replicates of seed collected from trees was used as denominators.

In order to evaluate the effect of retention time in the digestive tract on seed germination, we estimated for every species the time needed for an 80% seed recovery; using this information, we grouped the data into two periods: the first considering the days required to reach 80% seed recovery (P1) and the latter including the days until the end of the experiments (P2). We chose this percentage of recovery because values showed low variations among individuals of each species. We used a GLMM with a binomial error distribution and a logit link function to analyse the effect of periods (P1 and P2) on the percentages of germinated seeds. For this model, the number of germinated seeds during every period was used as a numerator in the response variable and the total number of germinated seeds as a denominator. Individuals nested within species were considered as a random effect. All analyses and graphs were performed in R version 3.6.1 (Team RC, 2016).

Results

A mean of 65% (SE = 4.5) of the seeds obtained from trees were viable, and it represents the viability of seeds ingested by animals in our experiment. Similar results were found by Velez et al. (2018). Cumulative curves of seed recovery over time for each species showed the same general sigmoid shape. After an initial rise, the curves rose sharply and gradually flattened in the final stages of seed recovery. It is important to note that the seed recovery for *C. chilensis* lasted 30 d (Supplementary Fig. S1). The mean retention time of ingested seeds varied among the studied species. *Chelonoidis chilensis* showed the longest mean retention time, followed by *L. guanicoe*, *L. gymnocercus*, *R. americana* and *D. patagonum* in the decreasing order. Only between *C. chilensis* and *D. patagonum*, there was a statistically significant difference (Kruskal–Wallis chi-squared = 20.50, df = 4, *P*-value < 0.001; Table 1).

Total recovery of seeds after passage through digestive tracts was around 50% of the viable ingested seeds for most animal species, except for *L. guanicoe* (Fig. 1 and Table 2). The model fitted to analyse the percentages of seeds recovered from faeces of different animals showed that the seeds ingested by *L. guanicoe* were recovered to a lower percentage than those ingested by the other species (Table 2).

The model fitted to test whether the percentages of germinated seeds differ among sources (recovered from faeces and collected

Table 2. Percentages of total recovered seeds after ingestion by animals, and total germination of ingested seeds and seeds collected from trees

Sources	Total recovered seeds	Total germinated seeds	Germinated seeds Period 1	Germinated seeds Period 2
Dolichotis patagonum	54.50 (±10.43) A	55.46 (±5.08) A	93.80 (±2.72)	6.20 (±2.72)[a]
Lycalopex gymnocercus	42.94 (±12.42) A	8.00 (±1.08) B	100 (±0.00)	0.00 (±0.00)[a]
Lama guanicoe	12.41 (±2.47) B	62.12 (±9.29) A	73.33 (±8.10)	26.67 (±8.10)[a]
Rhea americana	50.87 (±12.03) A	21.18 (±3.68) B	82.02 (±5.07)	17.98 (±5.10)[a]
Chelonoidis chilensis	56.19 (±17.38) A	52.17 (±13.00) A	63.89 (±20.37)	19.44 (±16.33)[a]
Trees		14.83 (±3.43) B		

Percentages of germinated seeds after the two periods of retention in the digestive tracts of animal species. Mean percentages (±SE) are shown. Values of recovered seeds were recalculated considering the viability obtained for ingested seeds (65%). Letters indicate significant differences among seed sources.
[a] Significant differences between periods.

Table 3. Results of generalized linear mixed models with a binomial error structure testing whether the proportion of *P. flexuosa* seeds recovered from faeces differs among species, the proportion of germinated seeds differs among sources (faeces and trees), and between periods of retention (P1 and P2) in the digestive systems of animals

Adjusted model	Effects	Estimate	Std. error	*z*-value	*P*-value
Proportion of recovered seeds ~ species + (1|species/individual)	Intercept (*L. guanicoe*)	−2.16	0.27	−7.97	1.64×10^{-15}
	C. chilensis	1.38	0.38	3.65	0.000264
	L. gymnocercus	1.20	0.45	2.64	0.008221
	D. patagonum	1.42	0.35	4.05	5.01×10^{-5}
	R. americana	1.36	0.37	3.67	0.000239
Total proportion of germinated seeds ~ sources + (1|species/individual)	Intercept (*L. guanicoe*)	−0.405	0.18	−2.55	0.010792
	C. chilensis	−0.02	0.25	−0.07	0.942307
	L. gymnocercus	−2.14	0.63	−3.41	0.000659
	D. patagonum	−0.08	0.21	−0.40	0.687014
	R. americana	−1.01	0.22	−4.61	4.01×10^{-6}
	trees	−1.50	0.26	−5.76	8.39×10^{-9}
Proportion of germinated seeds ~ period + (1|species/individual)	Intercept (P1)	−0.13	0.08	−1.65	0.09
	P2	−1.99	0.18	−11.17	2×10^{-16}

from trees) showed that germination was significantly low for seeds ingested by *R. americana* and *L. gymnocercus*, and for seeds collected from trees (Tables 2 and 3). Total seed germination was over 50% for seeds consumed by *L. guanicoe*, *D. patagonum* and *C. chilensis* (Fig. 2).

The germination analysis of seeds recovered after two periods of retention in the digestive tract showed a lower germination for seeds recovered during P2, that is, the seeds that remained in the digestive tracts after 80% of the ingested seeds had been recovered (Tables 2 and 3).

Discussion

The different ways in which seeds are treated in the digestive systems of endozoochorous animals is one of the components of dispersal quality (Schupp, 1993; Schupp et al., 2010). This study demonstrated some important differences in the quality of the treatment that *P. flexuosa* seeds receive when they pass through the digestive tract of native animals. The mean retention time of seeds does not seem to be the most important factor in total seed recovery and germination because the species with the longest retention time (*C. chilensis*) was not the one causing the greatest loss of seeds. In general, after a mean retention time of 5.5 d (SE = 2.72, range 1–16), it was possible to recover nearly half the viable seeds ingested, except for seeds consumed by *L. guanicoe*. Of the recovered seeds, more than 50% germinated during a month after recovery, except seeds ingested by *R. americana* and *L. gymnocercus*, which germinated in low proportions similarly as seeds collected from trees. For all animal species, germination was lower for seeds excreted during P2.

Some limitations are recognized due to working with animals in captivity because of alterations in the animals' activity patterns imposed by confined spaces, and shifts in their diets relative to those of animals in the wild (Picard et al., 2015). We observed that retrieved seeds were sometimes consumed again by individuals, a behaviour often seen in captive animals such as *R. americana* (Renison et al., 2010), and common in some cecum fermenters such as *D. patagonum* (Hume and Warner, 1980). Beyond its limitations, this type of study allows us to estimate the effect of different digestive tract treatments on a known number of seeds consumed in a controlled moment.

Since the digestive systems differ greatly by taxon, both morphologically and physiologically, we can expect differences in gut retention times with effects on recovery and germination of

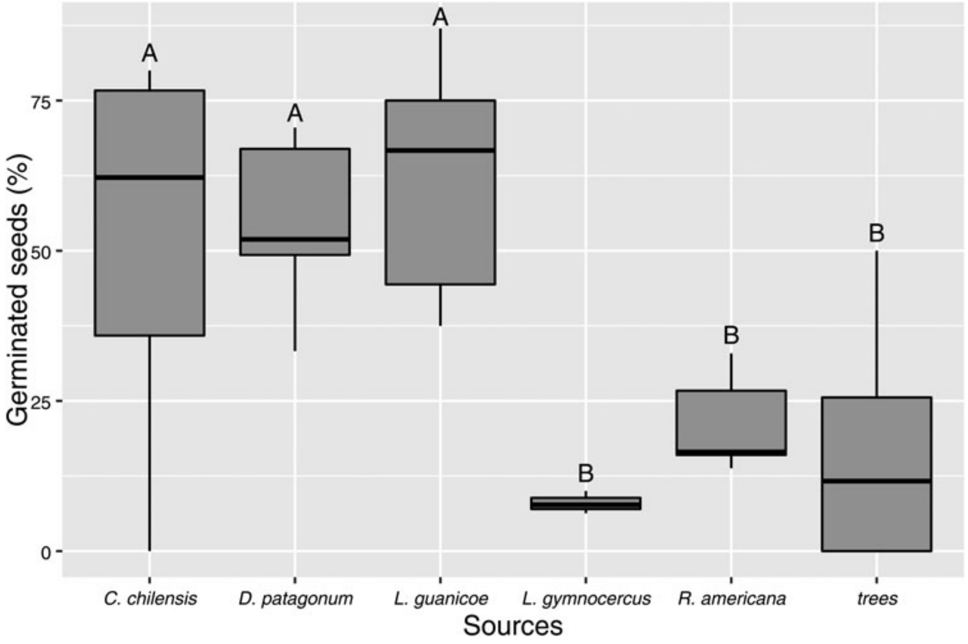

Fig. 2. Total percentages of germination of *P. flexuosa* seeds recovered after passing the digestive tracts of animal species. Letters show significant differences among species. The line in the box indicates the median value of the data. The upper and lower hinges of the box indicate the 75th and 25th percentiles of the dataset, respectively. The ends of the vertical lines indicate the minimum and maximum data values.

ingested seeds (Traveset et al., 2007). A rather long retention time with a prolonged exposure to digestive fluids may result in the removal of protective coats with an increase in germination speed or damage of the embryo (Gardener et al., 1993; Traveset and Verdú, 2002). Because of that, we expected to find that seeds ingested by *L. guanicoe* and *C. chilensis* would have longer mean retention time, lower seed recovery and lower germination of recovered seeds due to embryo damage by mastication and long exposure to digestive fluids.

Our prediction was partially justified. Results showed that *C. chilensis* had the longest mean retention time of ingested seeds, and the rodent *D. patagonum* had the shortest, whereas the other species had intermediate values. For *C. chilensis*, the mean retention time for ingested seeds of *P. flexuosa* was 16 d, similar to the 5–15 d reported in experiments with *P. nigra* seeds (Varela and Bucher, 2002). For *P. nigra*, seed recovery was almost 90% (Varela and Bucher, 2002), whereas recovery was 56% for *P. flexuosa* seeds. Despite the long retention time of seeds in the digestive tract of tortoises, their keratinized beak, instead of teeth, allows swallowing whole fruits, with seeds appearing in faeces with little physical change (Varela and Bucher, 2002).

Although the mean retention time of *P. flexuosa* seeds ingested by *L. guanicoe* was not longer than in the other studied species (4 d), and falls within the range of mean retention time recorded for markers ingested by *L. lama* (2–10 d depending on particle size; Clemens and Stevens, 1980), the lowest percentages of viable seed were recovered after *L. guanicoe*'s digestion (12%). A previous study that analysed faeces of *L. guanicoe*, collected in the field, documented large percentages of damaged seeds of *P. torquata*, *P. chilensis* and *P. flexuosa* (20–45%; Campos et al., 2008). Despite the low seed recovery, the present study showed that 62% of the seeds were able to germinate, similar to the proportion of germinable seeds of *P. chilensis* (70%) and *P. flexuosa* (50%) found in faeces collected from the field (Campos et al., 2008). From captivity experiments and field studies, we can observe a low recovery of viable *P. flexuosa* seeds with approximately half of them germinating after passage through the digestive tract of *L. guanicoe*. This effect on seed survival can be explained by the digestive process of this species, which includes rumination and the action of a forestomach with three gastric compartments for microbial fermentation, all resulting in high digestibility of plant material (Esteban and Thompson, 1988).

Despite the marked difference in retention times between *C. chilensis* and *D. patagonum*, both the total percentage of seeds recovered (56 and 54%, respectively) and the germination of seeds (52 and 55%) were similar. Then, these similarities observed between both dispersers suggest that their difference in retention time is not the most important factor affecting seed recovery and germination. For many species, the time that seeds remain in the digestive tract of dispersers does not have an effect on germination, either positive or negative. Other factors, such as the type of food ingested along with fruits (with variable acidity, water content, etc.), may even be more relevant in determining the extent to which seeds are mechanically or chemically abraded (Traveset, 1998). *Dolichotis patagonum* is a small hindgut fermenter or a cecum fermenter, with a cecum as the primary site of microbial activity (Hume and Warner, 1980). In captivity, mean retention time of fluid and particles in the gastrointestinal tract of *D. patagonum* was 27 h (Sakaguchi et al., 1992), with high digestive efficiency (Kufner and Durañona, 1991). From faeces of *D. patagonum* collected in the wild, 65% of *P. flexuosa* seeds were viable and between 12 and 25% were able to germinate (Campos and Ojeda, 1997; Campos et al., 2008), although, from these studies, we do not know how long those seeds remained in the digestive tract or how many seeds were originally ingested.

For the remaining two species, *R. americana* and *L. gymnocercus*, our results showed intermediate values of retention time, a higher seed recovery after ingestion than for *L. guanicoe*, and the lowest percentages of germination among all studied species. *Rhea americana* is a large herbivorous bird that eats fruits and seeds of a wide range of sizes (Pratolongo et al., 2003; Renison et al., 2010). *Rhea* species are characterized by a short colon with particularly enormous paired caeca as the main site for microbial fermentation (Stevens and Hume, 1995). They have efficient digestibility, with a mean digesta retention time for small particles (8 mm) of 7–19 h (Frei et al., 2015), although our results showed a mean retention time of 3 d for *P. flexuosa* seeds (5 mm). We recovered 51% of the ingested seeds, of which 21% were able to germinate. A previous study performed with captive individuals showed that less than 10% of *P. nigra* and *P. alba* seeds were recovered within 36 h following ingestion, and germinability of the recovered seeds was 5% for *P. alba* and 20% for *P. nigra* (Renison et al., 2010). The effect of digestion on seed germination was similar to that found in our study, although the low recovery percentage reported by Renison et al. (2010) might suggest that in this work the observation time was not long enough. It has been proposed that an excessive mechanical grinding in a bird's gizzard may be detrimental for seeds (Traveset et al., 2007). Even though *R. americana* did not appear to scarify seeds and increase germination percentage as others species do. We found no strong effect on viability because seed retrieval after passage through its gut was similar to that obtained for the other species, except for *L. guanicoe*, a species quite similar to *R. americana* in body size, but different in being a foregut fermenter.

The quality of treatment of *L. gymnocercus*, a monogastric (or 'single-stomached') animal was similar. As most carnivores, it has a relatively short and simple gastrointestinal tract; the glandular stomach is a unilateral dilatation of the digestive tract and a distinct hindgut is absent (Stevens and Hume, 1998). The mean retention time of *P. flexuosa* seeds was almost 3 d, longer than the mean retention time found in previous studies for *Schinus molle* fruits ingested by *L. culpaeus* (19 h; Silva et al., 2005), and for *Vachellia (Acacia) aroma* seeds ingested by *L. gymnocercus* and *Cerdocyon thous* (14 h; Varela and Bucher, 2006). It is important to note that alteration of an animal's activity patterns imposed by space limitation is a good reason to be careful about gut retention times obtained in captive carnivores (González-Varo et al., 2013). In one of the studies, authors had to discard 20 individuals because they exhibited stress symptoms, which can affect gastrointestinal motility by accelerating seed evacuation (Varela and Bucher, 2006). Several studies reported seed dispersal of different plants by species of *Lycalopex* (e.g. *Pseudalopex* and *Dusicyon*), mainly comparing the viability and germination of seeds obtained from faeces collected in the field and seeds from trees (Bustamante et al., 1992; Castro et al., 1994; León-Lobos and Kalin-Arroyo, 1994; Campos and Ojeda, 1997; Varela and Bucher, 2006; Dellafiore, 2018; Maldonado et al., 2018). Only two studies carried out experiments in captivity, both of them with *L. gymnocercus*. Dellafiore (2018) reported a 100% seed recovery and a 42% germination of *Pyracantha atalantoides* seeds after 24 h of retention in the gut. Varela and Bucher (2006), after 36 h, recovered 52% of viable seeds of *Acacia aroma* ingested by animals and achieved 36% germination. After a 3-d mean retention time for *P. flexuosa* seeds, our results showed a 43% recovery of the viable ingested seeds, of which only 8% were able to germinate, similar to the results for germination of *P. flexuosa* seeds obtained from faeces collected in the field (Campos and Ojeda, 1997). Other studies have also reported a low effect of digestion on germinability of seeds of *Lithrea caustica* and *P. alba*, and it has been proposed that retention time in the fox's digestive tract is too short to break the physical dormancy of seeds (León-Lobos and Kalin-Arroyo, 1994; Maldonado et al., 2018).

The variety of seed treatments offered by endozoochorous dispersers to *P. flexuosa* represents part of the qualitative component of SDE. Seeds contained in an indehiscent fruit need a mechanism to be released. Decomposition of the pericarp in dryland soil can take some years owing to fruit hardness and slowness of decomposition processes. During that time, seeds are exposed to predation by arthropods and small-sized rodents (Ortega-Baes et al., 2001; Giannoni et al., 2013; Velez et al., 2018). In this scenario, fruit ingestion by endozoochorous species represents for seeds a way by which they are released from fruit avoiding predators under the parent tree, with some costs to seed recovery after the passage through the animals' digestive systems. In the studied assemblage, the highest cost was for seeds ingested by *L. guanicoe*.

Seed germination does not seem to be significantly favoured when passing through digestive tracts, compared to the germination obtained in previous laboratory studies using healthy seeds that have been scarified by mechanical or chemical means, which is optimal for breaking dormancy and maximizing germination under laboratory conditions (e.g. Catalán and Macchiavelli, 1991; Catalán and Balzarini, 1992; Cony and Trione, 1996; Ortega-Baes et al., 2002). Nevertheless, in the assemblage of seed dispersers, the passage by the digestive system of some species partially released seeds from the physical dormancy imposed by hard impermeable seed coats and increased germination percentages (seeds ingested by *L. guanicoe*, *D. patagonum* and *C. chilnesis*), and other dispersers (*R. americana* and *L. gymnocercus*) kept germination similar to that of seeds from trees receiving no scarification treatment (i.e. seeds from trees not released from endocarps). In the last case, seeds must wait for a subsequent environmental scarification by soil abrasion or extreme temperature cycles. It has been proposed that the passage through the digestive tract of animals produces asynchrony because endozoochory changes the timing of germination. In habitats characterized by environmental unpredictability, germination asynchrony might result in an overall fitness benefit for the plant that allows waiting for good conditions for seedling survival (Fedriani and Delibes, 2009). Thus, the benefits of germinating faster depend on the context.

In a global study, it has been proposed that mammals have a positive effect as enhancers of seed germination, although some groups, such as rodents, cause low seed germination and high mortality after ingestion. Particularly for Fabaceae seeds, mammals do not have a significant effect on germination (except in the case of Primates; Torres et al., 2020). In contrast to this general study, we found that the quality of seed treatment by *D. patagonum*, the rodent of our study, was similar in its high seed recovery to *L. gymnocercus*, and similar in its high seed germination to *L. guanicoe*. Furthermore, we consider that mammals effectively contribute to dispersal of *P. flexuosa* seeds.

Because field studies of *P. flexuosa* report high quantities of viable seeds in faeces (e.g. Campos and Ojeda, 1997; Campos et al., 2008), the numbers obtained in captivity experiments imply that a large number of seeds must have been consumed to compensate for the recovery of approximately half the seeds, with half of them having germination success after gut passage. In order to complete the SDE model for *P. flexuosa*, future research would be needed to compare the faeces of different dispersers as germination substrate, and to determine the conditions for seed germination and seedling establishment in the microhabitats where seeds are dispersed. In addition, data from animal movement patterns (home range, habitat use) would provide information about the potential seed dispersal distance by animal species.

Supplementary material. To view supplementary material for this article, please visit: https://doi.org/10.1017/S096025852000032X.

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Author contributions.
C.M.C., M.I.C. and F.M.C. conceived the ideas and designed methodology; L.R., N.M., M.I.C., C.S., C.M.C., C. de los R. and F.M.C. collected and analysed the data; C.M.C. and F.M.C. acquired funding for the project; C.M.C. led the writing of the manuscript. All authors contributed critically to the drafts and gave final approval for publication.

References

Álvarez JA and Villagra PE (2009) *Prosopis flexuosa* DC. (Fabaceae, Mimosoideae). *Kurtziana* **35**, 47–61.

Bessega C, Pometti C, Campos C, Saidman BO and Vilardi JC (2017) Implications of mating system and pollen dispersal indices for management and conservation of the semi-arid species *Prosopis flexuosa* (Leguminosae). *Forest Ecology and Management* **400**, 218–227.

Bjorndal KA (1997) Fermentation in reptiles and amphibians, pp. 199–230 *in* Mackie RO and White BA (Eds) *Gastrointestinal microbiology*. Boston, MA, Chapman and Hall Microbiology Series, Springer.

Burkart A (1976) A monograph of the genus *Prosopis* (Leguminosae subfam. Mimosoidae). *Journal of the Arnold Arboretum* **57**, 219–525.

Bustamante RO, Simonetti JA and Mella JE (1992) Are foxes legitimate and efficient seed dispersers? A field test. *Acta Oecologica* **13**, 203–208.

Cain ML, Milligan BG and Strand AE (2000) Long-distance seed dispersal in plant populations. *American Journal of Botany* **87**, 1217–1227.

Campos C and Ojeda R (1997) Dispersal and germination of *Prosopis flexuosa* (Fabaceae) seeds by desert mammals in Argentina. *Journal of Arid Environments* **35**, 707–714.

Campos CM and Velez S (2015) Almacenadores y frugívoros oportunistas: el papel de los mamíferos en la dispersión del algarrobo (*Prosopis flexuosa* DC) en el desierto del Monte, Argentina. *Revista Ecosistemas* **24**, 28–34.

Campos C, Giannoni S, Taraborelli P and Borghi C (2007) Removal of mesquite seeds by small rodents in the Monte desert, Argentina. *Journal of Arid Environments* **69**, 228–236.

Campos CM, Peco B, Campos VE, Malo JE, Giannoni SM and Suárez F (2008) Endozoochory by native and exotic herbivores in dry areas: consequences for germination and survival of *Prosopis* seeds. *Seed Science Research* **18**, 91–100.

Campos CM, Campos V, Mongeaud A, Borghi C, de los Ríos C and Giannoni S (2011) Relationships between *Prosopis flexuosa* (Fabaceae) and cattle in the Monte desert: seeds, seedlings and saplings on cattle-use site classes. *Revista Chilena de Historia Natural* **84**, 289–299.

Campos CM, Campos VE, Miguel F and Cona MI (2016) Management of protected areas and its effect on an ecosystem function: removal of *Prosopis flexuosa* seeds by mammals in Argentinian drylands. *PLoS ONE* **11**, 9.

Campos CM, Campos VE, Giannoni SM, Rodríguez D, Albanese S and Cona MI (2017) Role of small rodents in the seed dispersal process: *Microcavia australis* consuming *Prosopis flexuosa* fruits. *Austral Ecology* **42**, 113–119.

Campos CM, Velez S, Miguel MF, Papú S and Cona MI (2018) Studying the quantity component of seed dispersal effectiveness from exclosure treatments and camera trapping. *Ecology and Evolution* **8**, 5470–5479.

Castle EJ (1956) The rate of passage of foodstuffs through the alimentary tract of the goat. 1. Studies on adult animals fed on hay and concentrates. *British Journal of Nutrition* **10**, 15–23.

Castro SA, Silva SI, Meserve PL, Gutiérrez JL, Contreras LC and Jaksic FB (1994) Frugivoría y dispersión de semillas de pimiento (*Schinus molle*) por el zorro culpeo (*Pseudalopex culpaeus*) en el Parque Nacional Fray Jorge (IV Región, Chile). *Revista Chilena de Historia Natural* **67**, 169–176.

Catalán LA and Balzarini M (1992) Improved laboratory germination conditions for several arboreal *Prosopis* species: *P. chilensis*, *P. flexuosa*, *P. nigra*, *P. alba*, *P. caldenia* and *P. affinis*. *Seed Science and Technology* **20**, 293–298.

Catalán LA and Macchiavelli RE (1991) Improving germination in *Prosopis flexuosa* D.C. and *P. alba* Griseb. with hot water treatments and scarification. *Seed Science and Technology* **19**, 253–262.

Chambers J and MacMahon J (1994) A day in the life of a seed: movements and fates of seeds and their implications for natural and managed systems. *Annual Review of Ecology and Systematics* **25**, 263–292.

Clemens ET and Stevens CE (1980) A comparison of gastrointestinal transit time in ten species of mammal. *Journal of Agricultural Science, Cambridge* **94**, 735–737.

Cony M (1993) Programa de Conservación y Mejoramiento de Especies del Género Prosopis en la Provincia Fitogeográfica del Monte, Argentina. Convenio CIID-IADIZA, pp. 37–72 *in* IADIZA (Ed.) Contribuciones Mendocinas a la Quinta Reunión de Regional para América Latina y el Caribe de la Red de Forestación del CIID. Conservación y Mejoramiento de Especies del Género Prosopis. Mendoza, IADIZA-CRICYT-CIID.

Cony MA and Trione SO (1996) Germination with respect to temperature of two Argentinian *Prosopis* species. *Journal of Arid Environments* **33**, 225–236.

Dellafiore CM (2018) ¿Afecta el zorro (*Lycalopex gymnocercus*) la germinación de piracanta (*Pyracantha atalantoides*) Rosaceae? *Mastozoología Neotropical* **25**, 53–58.

Esteban LR and Thompson JR (1988) The digestive system of New World camelids - common digestive diseases of llamas. *Iowa State University Veterinarian* **50**, 117–121.

Fedriani JM and Delibes M (2009) Functional diversity in fruit-frugivore interactions: a field experiment with Mediterranean mammals. *Ecography* **32**, 983–992.

Frei S, Ortmann S, Reutlinger C, Kreuzer M, Hatt JM and Clauss M (2015) Comparative digesta retention patterns in ratites. *The Auk* **132**, 119–131.

Gardener CJ, McIvor JG and Jansen A (1993) Passage of legume and grass seeds through the digestive tract of cattle and their survival in faeces. *Journal of Applied Ecology* **30**, 63–74.

Giannoni SM, Campos VE, Andino N, Ramos-Castilla M, Orofino A, Borghi CE, de los Ríos C and Campos CM (2013) Hoarding patterns of sigmodontine rodent species in the Central Monte desert (Argentina). *Austral Ecology* **38**, 485–492.

González-Varo JP, López-Bao JV and Guitián J (2013) Functional diversity among seed dispersal kernels generated by carnivorous mammals. *Journal of Animal Ecology* **82**, 562–571.

Howe HF and Smallwood J (1982) Ecology of seed dispersal. *Annual Review of Ecology and Systematics* **13**, 201–228.

Hume ID (1989) Optimal digestive strategies in mammalian herbivores. *Physiological Zoology* **62**, 1145–1163.

Hume ID and Warner ACI (1980) Evolution of microbial digestion in mammals, pp. 665–684 *in* Ruckebusch Y and Thivend P (Eds) *Digestive physiology and metabolism in ruminants*. Dordrecht, Netherlands, Springer.

Illius AW and Gordon IJ (1993) Diet selection in mammalian herbivores: constraints and tactics, pp. 157–181 *in* Highes RN (Ed.) *An interdisciplinary approach to foraging behaviour*. Boston, Blackwell Scientific Publishers.

Janzen DH (1970) Herbivores and the number of tree species in tropical forests. *American Naturalist* **104**, 501–528.

Janzen DH, Demment MW and Robertson JB (1985) How fast and why do germinating Guanacaste seeds (*Enterolobium cyclocarpum*) die inside cows and horses? *Biotropica* **17**, 322–325.

Jordano P (2000) Fruits and frugivory, pp. 125–166 *in* Fenner M (Ed.) *Seeds: the ecology of regeneration in plant communities*. Wallingford, UK, CABI Publishing.

Jordano P, Forget PM, Lambert JE, Böhning-Gaese K, Traveset A and Wright SJ (2011) Frugivores and seed dispersal: mechanisms and consequences for biodiversity of a key ecological interaction. *Biology Letters* **7**, 321–323.

Kufner MB and Durañona G (1991) Consumo y eficiencia digestiva del mara, *Dolichotis patagonum* (Rodentia: Caviidae). *Ecologla Austral* **1**, 50–55.

León-Lobos PM and Kalin-Arroyo MT (1994) Germinación de semillaje *Lifhrea caustica* (Mol.) H. et A. (Anacardiacea) dispersadas por *Pseudalopex* sp. (Canidae) en el bosque esclerófilo de Chile central. *Revista Chilena de Historia Natural* **67**, 59–64.

Maldonado DE, Loayza AP, Garcia E and Pacheco LF (2018) Qualitative aspects of the effectiveness of Culpeo foxes (*Lycalopex culpaeus*) as dispersers of *Prosopis alba* (Fabaceae) in a Bolivian dry valley. *Acta Oecologica* **87**, 29–33.

Miguel F, Cona MI and Campos CM (2017) Seed removal by terrestrial mammals in Argentinian drylands under different land management practices. *Seed Science Research* **27**, 174–182.

Miguel MF, Tabeni S, Cona MI and Campos CM (2018a) Secondary seed dispersal by mammals between protected and grazed semiarid woodland. *Forest Ecology and Management* **422**, 41–48.

Miguel MF, Jordano P, Tabeni S and Campos CM (2018b) Context-dependency and anthropogenic effects on individual plant-frugivore networks. *Oikos* **127**, 1045–1059.

Nathan R and Muller-Landau HC (2000) Spatial patterns of seed dispersal, their determinants and consequences for recruitment. *Trends in Ecology and Evolution* **15**, 278–285.

Ortega-Baes P, de Viana M and Saravia M (2001) The fate of *Prosopis ferox* seeds from unremoved pods at National Park Los Cardones. *Journal of Arid Environments* **48**, 185–190.

Ortega-Baes P, de Viana M and Suhring S (2002) Germination in *Prosopis ferox*: effects of mechanical, chemical and biological scarificators. *Journal of Arid Environments* **50**, 185–189.

Peinetti R, Pereyra M, Kin A and Sosa A (1993) Effects of cattle ingestion on viability and germination rate of caldén (*Prosopis caldenia*) seeds. *Journal of Range Management* **46**, 483–486.

Peláez DV, Bóo RM and Elia OR (1992) Emergence and seedling survival of caldén in the semiarid region of Argentina. *Journal of Range Management* **45**, 564–566.

Picard M, Papaix J, Gosselin F, Picot D, Bideau E and Baltzinger C (2015) Temporal dynamics of seed excretion by wild ungulates: implications for plant dispersal. *Ecology and Evolution* **5**, 2321–2632.

Pili-Sevilla E (1987) Germination and tetrazolium testing. *Seed Science and Technology* **15**, 691–698.

Pratolongo P, Quintana R, Malvárez I and Cagnoni M (2003) Comparative analysis of variables associated with germination and seedling establishment for *Prosopis nigra* (Griseb.) Hieron and *Acacia caven* (Mol.) Mol. *Forest Ecology and Management* **179**, 15–25.

Razanamandranto S, Tigabu M, Neya S and Odén PC (2004) Effects of gut treatment on recovery and germinability of bovine and ovine ingested seeds of four woody species from the Sudanian savanna in West Africa. *Flora* **199**, 389–397.

Renison D, Valladares G and Martella MB (2010) The effect of passage through the gut of the Greater Rhea (*Rhea americana*) on germination of tree seeds: implications for forest restoration. *Emu* **110**, 125–131.

Sakaguchi E, Nippashi K and Endoh G (1992) Digesta retention and fibre digestion in maras (*Dolichotis patagonum*) and guinea-pigs. *Comparative Biochemistry and Physiology* **101**, 867–870.

Samuels IA and Levey DJ (2005) Effects of gut passage on seed germination: do experiments answer the questions they ask? *Functional Ecology* **19**, 365–368.

Sauer DB and Burroughs R (1986) Disinfection of seed surfaces with sodium hypochlorite. *Phytopathology* **76**, 745–749.

Schupp EW (1993) Quantity, quality and the effectiveness of seed dispersal by animals. *Vegetatio* **107/108**, 15–29.

Schupp EW, Jordano P and Gómez JM (2010) Seed dispersal effectiveness revisited: a conceptual review. *New Phytologist* **188**, 333–353.

Schupp EW, Jordano P and Gómez JM (2017) A general framework for effectiveness concepts in mutualisms. *Ecological Letters* **20**, 577–590.

Sikes R, Gannon W and The Animal Care and Use Committee of the American Society of Mammalogists (2011) Guidelines of the American Society of Mammalogists for the use of wild mammals in research. *Journal of Mammalogy* **92**, 235–253.

Silva SI, Jacksic FM and Bozinovic F (2005) Nutritional ecology and digestive response to dietary shift in the large South American fox, *Pseudalopex culpaeus*. *Revista Chilena de Historia Natural* **78**, 239–246.

Stevens CE and Hume ID (1995) *Comparative physiology of the vertebrate digestive system*. New York, Cambridge University Press.

Stevens CE and Hume ID (1998) Contributions of microbes in vertebrate gastrointestinal tract to production and conservation of nutrients. *Physiological Reviews* **78**, 393–427.

Tabeni S, Miguel MF, Campos CM and Cona M (2017) Small mammal abundance and seed predation across boundaries in a restored-grazed woodland interface. *Restoration Ecology* **26**, 787–795.

Team RC (2016) *R: a language and environment for statistical computing*. Vienna, Austria, R Foundation for Statistical Computing.

Torres D, Castaño JH and Carranza-Quiceno JA (2020) Global patterns in seed germination after ingestion by mammals. *Mammal Reviews*. doi: 10.1111/mam.12195.

Traveset A (1998) Effect of seed passage through vertebrate frugivores' guts on germination: a review. *Perspectives in Plant Ecology, Evolution and Systematics* **1**, 151–190.

Traveset A and Verdú M (2002) A meta-analysis of the effect of gut treatment on seed germination, pp. 339–350 *in* Levey DJ; Silva WR and Galetti M (Eds) *Seed dispersal and frugivory: ecology, evolution and conservation*. Wallingford, CABI Publishing.

Traveset A, Robertson AW and Rodríguez-Pérez J (2007) A review on the role of endozoochory in seed germination, pp. 78–103 *in* Dennis A; Green R; Schupp E and Westcott D (Eds) *Seed dispersal: theory and its application in a changing world*. Norfolk, UK, CABI Publishing.

van der Pijl L (1982) *Principles of dispersal in higher plants* (3rd edn). Berlin, Springer-Verlag.

Varela RO and Bucher EH (2002) Seed dispersal by *Chelonoidis chilensis* in the Chaco Dry woodland of Argentina. *Journal of Herpetology* **36**, 137–140.

Varela O and Bucher EH (2006) Passage time, viability, and germination of seeds ingested by foxes. *Journal of Arid Environments* **67**, 566–578.

Velez S, Chacoff NP and Campos CM (2018) Pre-dispersal seed loss in two *Proposis* species (Fabacea: Mimosoidea) from the Monte Desert, Argentina. *Ecología Austral* **28**, 361–373.

Warner ACI (1981) The mean retention times of digesta markers in the gut of the tammar, *Macropus eugenii*. *Australian Journal of Zoology* **29**, 759–771.