

Does avian gut passage favour seed germination of woody species of the Chaco Serrano Woodland in Argentina?

María Celeste Díaz Vélez, Ana Elisa Ferreras, Wesley Rodrigues Silva, and Leonardo Galetto

Abstract: Frugivorous birds are key dispersal agents of many plant species and also may facilitate seed germination after gut passage. However, the general effects of gut passage on seed germination are still not clear, with positive, negative, and neutral effects reported on seed germination. We evaluated seed germination of seven bird-dispersed plant species of the Chaco Serrano Woodland in Córdoba, Argentina: *Celtis ehrenbergiana* (Klotzsch) Liebm., *Condalia* spp. Cav., *Lantana camara* L., *Lithraea molleoides* (Vell.) Engl., *Lycium cestroides* Schltldl., *Schinus fasciculatus* (Griseb.) I.M. Johnst., and *Zanthoxylum coco* Gillies. We compared germination percentages and germination speed among seeds ingested by birds, manually extracted seeds, and seeds from intact fruits to understand which mechanisms are acting on bird gut-passed seeds. For six plant species, the action of frugivorous birds increased seed germination percentages and germination speed, through scarification, deinhibition, or combined mechanisms. Our results contribute to a better understanding of the mechanisms acting on seeds passed through bird gut. Also, we show the pivotal role that frugivorous birds exert on the seed germination of native woody plant species in the threatened ecosystem of the Chaco Serrano Woodland.

Key words: avian gut passage, deinhibition, germination, scarification, native woody species.

Résumé : Les oiseaux frugivores sont des agents clés de la dispersion de plusieurs espèces de végétaux et ils peuvent aussi faciliter la germination des graines après leur passage dans l'intestin. Cependant, les effets généraux du passage dans l'intestin sur la germination ne sont pas clairs, des effets positifs, négatifs et neutres sur la germination ayant été rapportés. Les auteurs ont évalué la germination de sept espèces de plantes dispersées par les oiseaux de la région forestière du Chaco Serrano à Córdoba, Argentine : *Celtis ehrenbergiana* (Klotzsch) Liebm., *Condalia* spp. Cav., *Lantana camara* L., *Lithraea molleoides* (Vell.) Engl., *Lycium cestroides* Schltldl., *Schinus fasciculatus* (Griseb.) I.M. Johnst. et *Zanthoxylum coco* Gillies. Ils ont comparé les pourcentages de germination et la vitesse de germination des graines ingérées par les oiseaux, des graines extraites manuellement et des graines de fruits intacts afin de comprendre quels sont les mécanismes qui agissent sur les graines ayant traversé l'intestin des oiseaux. Chez six espèces de plantes, l'action des oiseaux frugivores augmentait les pourcentages de germination et produisait une germination plus rapide, soit par scarification, désinhibition, ou par les deux mécanismes combinés. Les résultats des auteurs contribuent à une meilleure compréhension des mécanismes qui agissent sur les graines ayant traversé l'intestin. Ils montrent aussi le rôle essentiel que jouent les oiseaux frugivores dans la germination des graines d'espèces de plantes ligneuses indigènes dans l'écosystème menacé de la région forestière du Chaco Serrano. [Traduit par la Rédaction]

Mots-clés : passage dans l'intestin de l'oiseau, désinhibition, germination, scarification, espèces ligneuses indigènes.

Introduction

The consequences of avian fruit consumption on seed germination are important for assessing the effectiveness of frugivores in plant establishment and regeneration (Traveset 1998; Traveset et al. 2007; Schupp et al. 2010). The overall effects of gut passage on seed germination

are not consistent, with positive, negative, and neutral effects on seed germination all reported (Traveset 1998; Traveset et al. 2007). The results of the interaction seem to be highly dependent on the plant and bird species interacting, and on the methodology used in the testing (Robertson et al. 2006).

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In bird-dispersed species, facilitation of seed germination may occur through different mechanisms; among these, two of the most important are deoinhibition and scarification (Traveset 1998; Traveset and Verdú 2002; Robertson et al. 2006; Traveset et al. 2007). The deoinhibition effect implies that through avian fruit ingestion, germination is increased only by separating seeds from the fruit pulp. This effect could occur even if seeds are regurgitated. The fruit pulp may contain germination inhibitors such as lipids, glycoalkaloids, coumarin, and various light-blocking pigments (Cipollini and Levey 1997; Samuels and Levey 2005; Traveset et al. 2007). In addition, high sugar content in fruit pulp may provoke high osmotic pressure (Traveset 1998; Samuels and Levey 2005) or it can be a source of infections by fungi or other pathogens affecting seed viability and germination (Traveset 1998; Samuels and Levey 2005). The scarification effect (chemical or mechanical) on germination involves modifications of the seed coat (Traveset 1998; Traveset and Verdú 2002; Samuels and Levey 2005; Robertson et al. 2006; Traveset et al. 2007, 2008). The magnitude of the scarification effect depends on the characteristics of the frugivore species (length of the digestive tract, presence of a gizzard, and seed retention time) as well as fruit and seed traits of each plant species (pulp composition, seed age, seed size, and coat thickness and texture) (Traveset et al. 2007).

Most studies that evaluated the effects of avian fruit consumption on seed germination tested the combined effects of scarification and deoinhibition without separating the two (Traveset and Verdú 2002; Samuels and Levey 2005). Samuels and Levey (2005) reviewed the literature and found that only 18% of the studies evaluating the effect of gut passage on germination included comparisons that allowed the distinction between scarification and deoinhibition effects to be made (this requires a comparison of seeds from intact fruits, manually extracted seeds, and gut-passed seeds). The recognition of the two components of germination stimulation has greatly clarified what is required to improve our knowledge on the role of frugivorous birds as seed dispersers in natural environments. Several of the previous studies suggested that pulp removal alone by frugivorous birds would be sufficient to increase seed germination, without providing evidence of a scarification effect (Izhaki and Safrieli 1990; Barnea et al. 1991; Reid and Armesto 2011; Lehouck et al. 2011). On the other hand, a review by Traveset and Verdú (2002) showed that avian-gut-passed seeds have increased germination percentages compared with manually extracted seeds, suggesting that the scarification effect is important. Moreover, Robertson et al. (2006) showed that deoinhibition studies are prone to artefacts of the testing environment, where whole fruits are tested in Petri dishes that prevent the washing away of inhibitors that would happen naturally in the soil under field conditions. Thus, the importance of the deoinhi-

tion effect may have been overestimated as a result of using an inappropriate test method.

The Chaco subtropical forest of Argentina became a priority for biodiversity conservation because it is a threatened ecosystem that has suffered a strong increase in the rate of deforestation and habitat fragmentation due to the expansion of agriculture and urbanization (Gavier and Bucher 2004; Zak et al. 2004; Galetto et al. 2007; Aide et al. 2012), and has also suffered from the proliferation of invasive exotic plant species (Giorgis et al. 2011). There are many species of woody plants, shrubs, grasses, and vines in this ecosystem that are consumed and dispersed by frugivorous birds (Ponce et al. 2012; Díaz Vélez 2013). Birds are the only volant frugivores (no frugivorous bats exist in the area), and they may play an important role in maintaining bird-dispersed plant populations, not only by promoting seed movement, but also by increasing the likelihood or speed of seed germination. However, only a few studies have examined seed germination of bird-dispersed species in Chaco (Funes et al. 2009; Grilli and Galetto 2009; Grilli et al. 2009; Ashworth and Martí 2011), and even fewer studies have evaluated the different mechanisms that could facilitate seed germination (Renison et al. 2010). In this work, address two main questions: (i) does avian gut passage have a positive, negative, or neutral effect on the seed germination of native woody plant species?; and (ii) if it has a positive effect, is this facilitation occurring through deoinhibition, scarification, or both?

Materials and methods

Study site

The study was performed in the Chaco Serrano Woodland of Córdoba, Argentina. The mean annual temperature is 17.5 °C. Most precipitation falls from October to March (on average 750 mm per season) (Luti et al. 1979; Moglia and Gimenez 1998), coinciding with the fruiting peak of bird-dispersed plants. The dry season extends from April to September (on average 116 mm per season), which coincides with the period of low temperatures (Capitanelli 1979).

The Chaco vegetation at the study site is highly fragmented because of recent agricultural and urban expansion (Zak et al. 2004). Fruits were collected from eight of the fragments (31°09'S to 31°13'S, and 64°13'W to 64°17'W) that are composed of secondary forest with a canopy that usually reaches 7–9 m high. Native vegetation includes numerous bird-dispersed species, the majority of which are trees and shrubs, with some climbers (such as *Passiflora morifolia* Mast. and *Passiflora Suberosa* L.) and many herbs (such as *Salpichroa organifolia* (Lam.) Thell., *Solanum chenopodioides* Lam., *Rivina humilis* L., and *Lantana grisebachii* Stuck. ex Seckt.) (Cagnolo et al. 2006; Ponce et al. 2012; Díaz Vélez 2013).

Table 1. List of species studied in the Chaco Serrano Woodland in Córdoba, Argentina.

Plant species		<i>Condalia</i> spp. ^a	<i>Lantana camara</i>	<i>Lithraea molleoides</i>	<i>Lycium cestroides</i>	<i>Schinus molle</i>	<i>Zanthoxylum coco</i>
Family	Celtidaceae	Rhamnaceae	Verbenaceae	Anacardiaceae	Solanaceae	Anacardiaceae	Rutaceae
Common name	Tala	Piquilín	Bandera Española	Molle de Beber	Tumiñico	Moradillo	Coco
Growth form	Tree	Shrub-small tree	Shrub	Tree	Shrub-small tree	Shrub	Tree
Fruiting period ^b	Jan.–Aug.	Jan.–Feb.	Jan.–May	Jan.–Oct.	Jan.–May	Jan.–Aug.	Jan.–Aug.
Fruit type ^c	Drupe	Drupe	Drupe	Drupe	Berry	Drupe	Dehiscent follicle
Fruit diameter (mm)	6–9	8	4–7	6–8	5–6	4–5	5–7
Fruit colour	Yellow–orange	Dark purple–dark red	Blue–black	Translucent with black pulp	Dark red–purple	Violet–purple	Shiny black
Bird species ^d	Elasp, Pitsul, Salaur, Thrbon, Turama	Elasp, Pitsul, Salaur, Thrbon, Turama	Elasp, Turama	Colmel	Elasp, Pitsul, Poonig, Salaur, Thrbon, Turama	Elasp, Salaur, Thrbon, Turama	Elasp, Pitsul, Salaur

^aThis includes three species (*Condalia montana*, *Condalia buxifolia*, and *Condalia microphylla*); they are difficult to tell apart because their seeds are very similar.

^bData obtained from seed traps located in the forest fragments (Díaz Vélez 2013).

^cOne seed per fruit in all, except for *L. cestroides* (29 seeds per fruit on average).

^dData obtained from personal observations in the study area and literature (De la Peña 2011; Ponce et al. 2012; Díaz Vélez 2013). Colmel, *Colaptes melanochloros*; Elasp, *Elaenia parvirostris* and *Elaenia albicep*; Pitsul, *Pitangus sulphuratus*; Poonig, *Pooispiza nigrorufa*; Salaur, *Saltator aurantirostris*; Thrbon, *Thraupis bonariensis*; and Turama, *Turdus amaurochalinus*.

Fig. 1. Experimental identification of mechanisms related to the increase of seed germination and acting (or not) on seeds passed through bird gut, following the scheme proposed by Samuels and Levey (2005). G, gut-passed seeds; F, intact fruit; and M, manually extracted seeds.

G > M > F	Scarification + Deinhibition effect
G = M > F	Deinhibition effect
G > M = F	Scarification effect
G = M = F	Neutral effect

Species studied

We collected fruits of seven native bird-dispersed woody species that are commonly eaten by birds and that were the most abundant in a bird-dispersed seed rain experiment performed within the study area (Díaz Vélez 2013). The characteristics of the plant and frugivorous bird species that consume their fruits are listed in Table 1.

Seed treatments and germination experiments

To understand which mechanisms are acting on bird gut-passed seeds, the scheme proposed by Samuels and Levey (2005) was used to compare germination percentages among seeds ingested by birds, manually extracted seeds, and seeds from intact fruits (Fig. 1). The three seed treatments were as follows:

(a) Gut-passed seeds (G): seeds were defecated and (or) regurgitated by birds. If this treatment results in an increase of the percentage of seed germination, the mechanisms inferred are deinhibition and scarification.

(b) Intact fruits (F): mature fruits were collected from at least 10 individual plants of each species and stored in paper bags until the beginning of the germination experiment. The dehydration process that fruits experience inside the paper bags simulates what would happen with nondispersed fruits, i.e., fruits that fall on the ground. Since *Lycium cestroides* Schltld. is the only species with more than one seed per fruit, the percentage of seed germination was calculated by adding the germinated seeds plus the number of ungerminated seeds within intact fruits opened at the end of the experiment to get a total seed count per fruit.

(c) Manually extracted seeds (M): seeds were manually removed from mature fruit, and any pulp remaining on the seed coat was washed off. This condition was used to determine whether the seeds are subject to an inhibitory mechanism produced by the pulp. For *Zanthoxylum coco* Gillies, the seeds were left in water for one day to facilitate the extraction of the oily blackish mesocarp that is firmly attached to the seed. Most of the oily mesocarp was extracted using tweezers.

Each replicate, for each treatment, consisted of a group of seeds or fruits from only one species placed in a

Table 2. Sources of bird-dispersed seeds, number of seeds per replicate, and number of replicates per treatment.

	Plant species						
	<i>Celtis ehrenbergiana</i>	<i>Condalia</i> spp.	<i>Lantana camara</i>	<i>Lithraea molleoides</i>	<i>Lycium cestroides</i>	<i>Schinus fasciculatus</i>	<i>Zanthoxylum coco</i>
Source of bird-dispersed seeds	Traps ^a	Mist nets ^b	Traps	Traps	Mist nets	Traps	Traps
No. of seeds per replicate	16	7	20	10	28	20	16
No. of replicates per treatment	13	4	5	6	17	7	5

^aSeed traps: a total of two hundred and forty 50 cm × 50 cm nylon traps were placed below tree species in eight forest fragments of the Chaco Serrano Woodland during 6 months (January to June 2011).

^bMist nets: nets (12 m × 2.6 m, 36 mm mesh size) were placed in different locations in eight forest fragments (total 1023 net hours) during the fruit peak of bird-dispersed plants (January to April 2009). We placed a plastic sheet beneath each deployed net to collect the faecal samples as birds upon being caught might drop them. Also, we held captured birds in cloth bags for no more than 20 min to collect faecal samples.

transparent plastic container that was 5 cm high and 8 cm in diameter (250 cm³) (Table 2). For the “intact fruit” and “manually extracted seeds” treatments, we used mature fruit collected from the same individual plant or seed trap. The number of seeds per replicate and replicates per treatment varied among plant species and were determined according to the average number of seeds found in the faecal or regurgitated samples (Table 2). Seed sources and replicates for the three germination treatments are shown in Table 2.

Germination experiments were carried out in a germination chamber. Each replicate was placed in a transparent plastic container, covered with holes for ventilation, containing 1 cm of sterilized soil mixed with sand. The plastic containers were kept in a germination chamber at 25 °C with 14 h of light. Seeds or fruit were sown in the substrate and watered regularly. The groups of seeds or fruit were examined for germination (radicle emergence) twice a week. Germinated seeds were counted and removed to reduce interaction with ungerminated seeds and to avoid recounting. Germination was monitored for one year in *L. cestroides* and *Condalia* spp. Cav. (from August 2009 to August 2010) and for 8 months in the remaining plant species (from June 2011 to February 2012). We reduced the time of the second germination experiment because we did not record seed germination after 8 months in the first experiment.

Statistical analyses

We calculated the number and percentage of germinated seeds for each treatment at the end of the experiment for the seven plant species selected. The percentage of seed germination was analyzed with a generalized linear model with a binomial error structure and logit link function, using the seed treatments as the fixed factor (three levels: gut-passed seeds, manually extracted seeds, and intact fruits). A quasi-binomial error structure was used because of over-dispersion in the response variables in many species (see Table 3). When differences among treatments were significant, the Di Rienzo, Guzmán, and Casanoves a-posteriori test was performed. The generalized linear models were performed in Infostat (Di Rienzo et al. 2013) and its interface to R (R 2.15.0, R Development Core Team 2012). As an estimate of germination speed,

Table 3. Degrees of freedom (df) and *P* values for the generalized linear model performed for each plant species comparing intact fruit, manually extracted seeds, and gut-passed seeds.

Plant species	df	<i>P</i> value
<i>Celtis ehrenbergiana</i> ^a	2	0.05
<i>Condalia</i> spp. ^a	2	0.03
<i>Lantana camara</i> ^a	2	<0.0001
<i>Lithraea molleoides</i>	2	<0.0001
<i>Lycium cestroides</i> ^a	2	<0.0001
<i>Schinus fasciculatus</i> ^a	2	<0.0001
<i>Zanthoxylum coco</i>	2	<0.0001

^aSpecies that were analyzed with a quasi-binomial error structure because of overdispersion.

we used the survival analysis module of Kaplan–Meier available in SPSS 10.0 to compare cumulative germination percentages among treatments. We used log-rank tests (Mantel–Cox), a nonparametric test recommended for evaluating the effect of categorical predictive variables. In all cases, values of *P* ≤ 0.05 were used to assess statistically significant differences among treatments (Traveset et al. 2008).

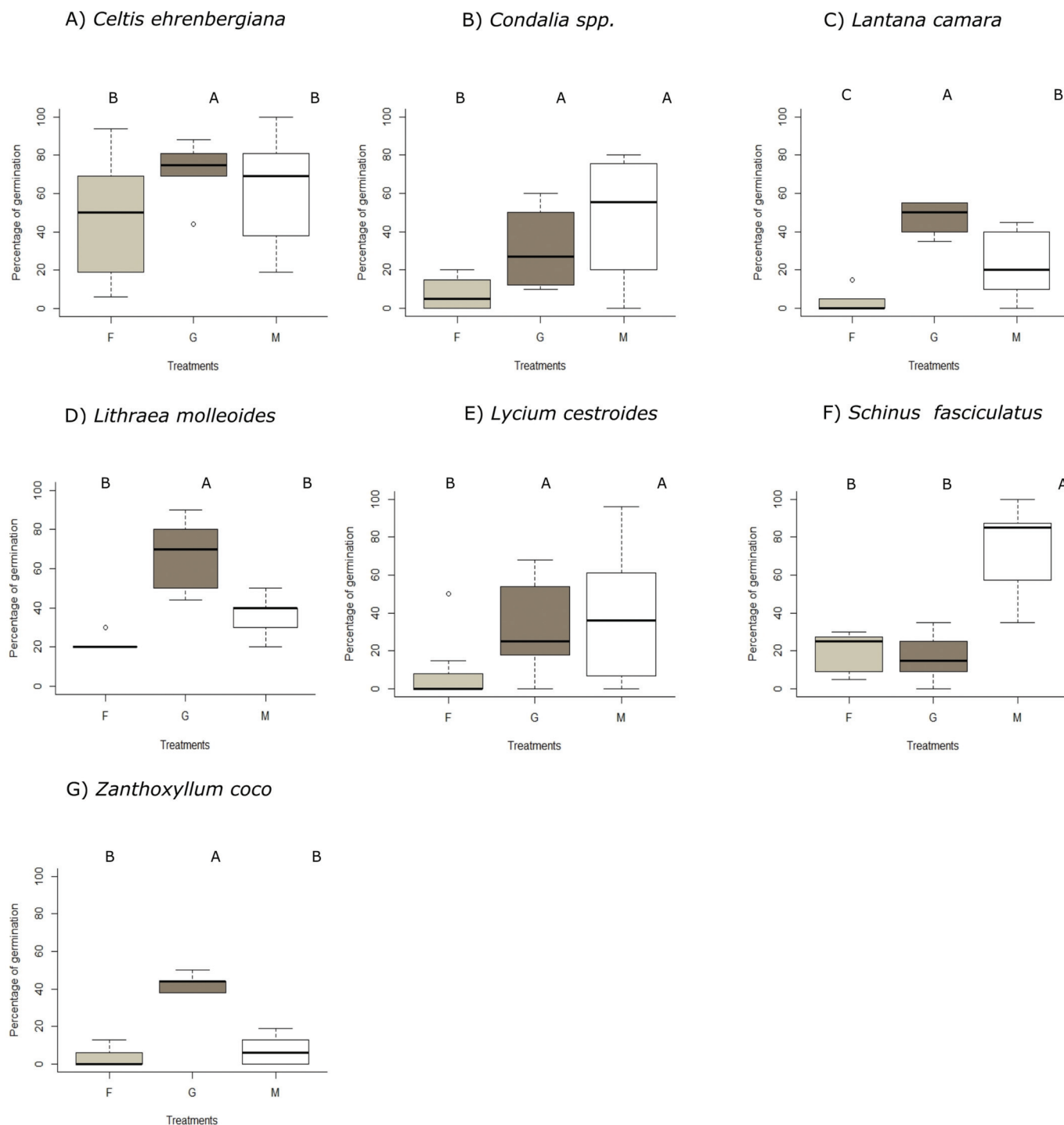
Results

Germinability

There were significant treatment effects on germination percentage for all species (Table 3; Fig. 2). In general, there was a reduced likelihood of seed germination in intact fruits, ranging from 4% to 22%; however, *Celtis ehrenbergiana* (Klotzsch) Liebm. presented an average germination percentage >50%. Germination varied from 17% to 74% for gut-passed seeds, and from 8% to 73% for manually extracted seeds.

Gut-passed seeds showed significantly higher germination percentages than intact fruits in all species, with the exception of *Schinus fasciculatus* (Griseb.) I.M.Johnst. (Fig. 2F). In *S. fasciculatus*, the germination of manually extracted seeds was significantly higher than gut-passed seeds and intact fruit. For *C. ehrenbergiana*, *Lantana camara* L., *Lithraea molleoides* (Vell.) Engl., and *Z. coco*, gut-passed seeds showed significantly higher values than manually

Fig. 2. Germination percentages for each seed treatment (G, gut-passed seeds; F, intact fruits; and M, manually extracted seeds) for (A) *Celtis ehrenbergiana*, (B) *Condalia* spp., (C) *Lantana camara*, (D) *Lithraea molleoides*, (E) *Lycium cestroides*, (F) *Schinus fasciculatus*, and (G) *Zanthoxylum coco*. Different letters above the columns indicate significant differences, $P \leq 0.05$. [Colour online.]



extracted seeds (Figs. 2A, 2C, 2D, and 2G). Of these four species, manually extracted seeds also differed from intact fruit in *L. camara* only (Fig. 2C). In *L. cestroides* and *Condalia* spp., manually extracted seeds differed from intact fruit, but not from gut-passed seeds (Figs. 2B and 2E).

Germination speed

There were large treatment effects on the speed at which seeds germinated (Table 4, Fig. 3). Overall, the cumulative germination of gut-passed seeds differed significantly from manually extracted ones, being faster in *C. ehrenbergiana*, *L. camara*, *L. molleoides*, and *Z. coco*

Table 4. Chi-squared values (χ^2) and *P* values for the log-rank tests (Mantel–Cox) performed for each plant species comparing intact fruit (F), manually extracted seeds (M), and gut-passed seeds (G).

Plant species	Pairwise comparisons					
	F vs. G		F vs. M		G vs. M	
	χ^2	<i>P</i> value	χ^2	<i>P</i> value	χ^2	<i>P</i> value
<i>Celtis ehrenbergiana</i>	56.243	<0.0001	5.106	0.024	33.703	<0.0001
<i>Condalia</i> spp.	3.451	0.063	12.07	0.001	4.003	0.045
<i>Lantana camara</i>	50.469	<0.0001	16.950	<0.0001	11.955	0.001
<i>Lithraea molleoides</i>	24.007	<0.0001	4.27	0.039	5.757	0.016
<i>Lycium cestroides</i>	130.102	<0.0001	168.646	<0.0001	3.944	0.047
<i>Schinus fasciculatus</i>	0.181	0.67	85.855	<0.0001	76.59	<0.0001
<i>Zanthoxylum coco</i>	34.956	<0.0001	1.684	0.194	26.341	<0.0001

(Figs. 3A, 3C, 3D, and 3G), and slower in *Condalia* spp., *L. cestroides*, and *S. fasciculatus* (Figs. 3B, 3E, and 3F). Gut-passed seeds also germinated significantly faster than those of intact fruit, but the differences were not significant in *Condalia* spp. and *S. fasciculatus* (Figs. 3B and 3F). Manually extracted seeds germinated significantly faster than those of intact fruit for all species except *Z. coco*, for which the difference was not statistically significant (Fig. 3G).

Discussion

Frugivorous birds are key agents for the maintenance and expansion of many plant species, because they disperse the plant's seeds and may facilitate their germination. In this study we analyzed germination percentage and germination speed in seven species of the Chaco Serrano Forest of Córdoba, Argentina, to disentangle whether birds are having a positive, negative, or neutral effect on seed germination. In addition, as proposed by Samuels and Levey (2005), we aimed to understand the mechanisms through which this interaction is occurring. For the majority of plant species in this study (six out of seven), the action of frugivorous birds facilitated seed germination through scarification (*C. ehrenbergiana*, *L. molleoides*, and *Z. coco*), deoinhibition (*Condalia* spp. and *L. cestroides*), or through combined mechanisms (*L. camara*).

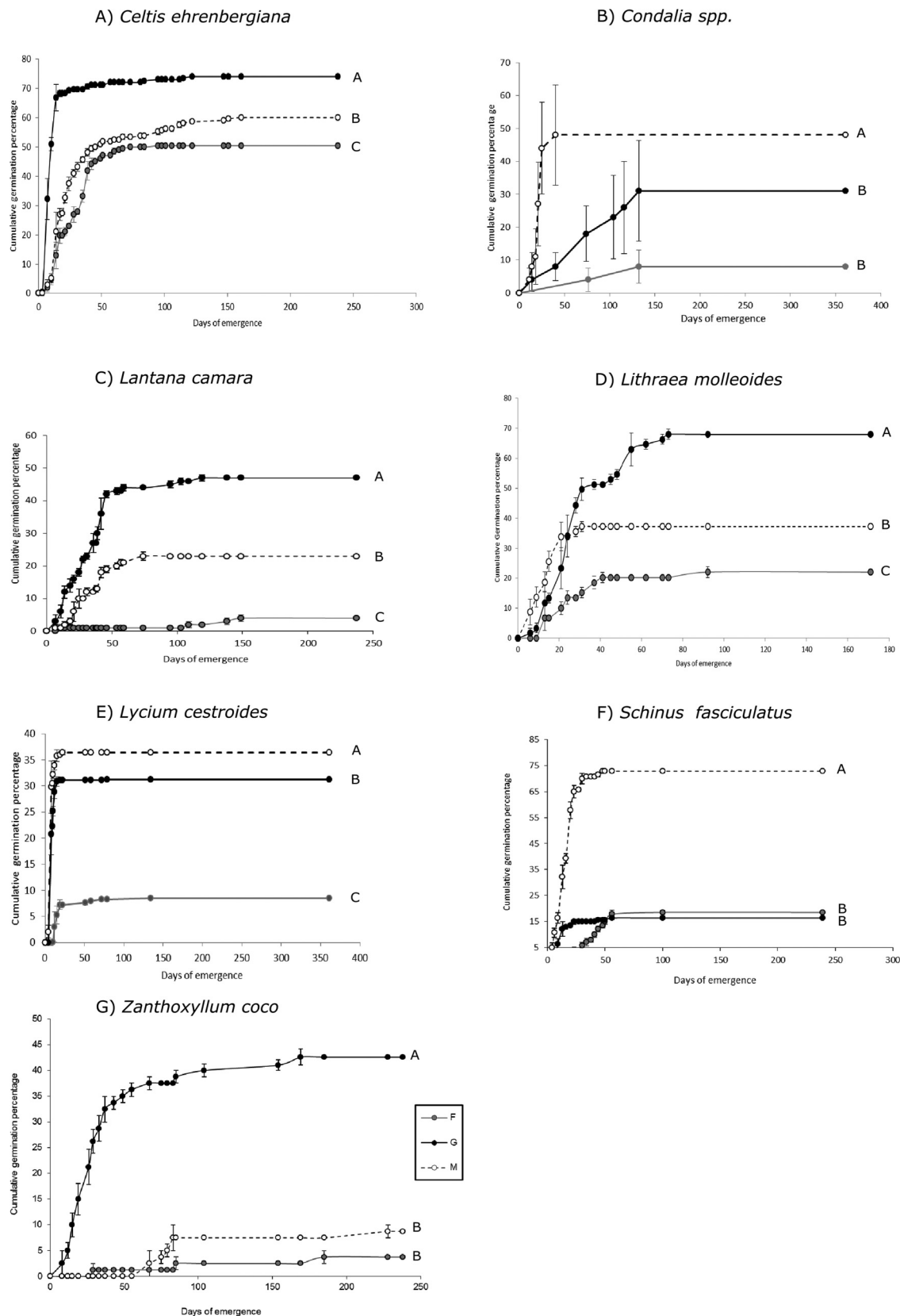
In *C. ehrenbergiana*, *L. molleoides*, and *Z. coco*, seed germination percentage and germination speed were significantly higher in gut-passed seeds compared with manually extracted seeds, which is indicative of a scarification effect. Therefore, these species must have a double dependency on birds, i.e., for dispersal and germination. It is possible that in *C. ehrenbergiana* and *L. molleoides* a deoinhibition effect also exists, which was only detected through their germination speed. It is important to highlight that the seeds in more than 50% of even the intact fruits of *C. ehrenbergiana* germinated successfully. In addition, Renison et al. (2010) found that seeds of this species when ingested by *Rhea americana* had lower germination than those of intact fruit, and suggested that *C. ehrenbergiana* seeds could be adapted to dispersal by small bird species with shorter gut retention

times, which is supported by our results. In *Z. coco*, because pulp removal of this species is difficult (probably not fully removed in our manually extracted treatment), we cannot discard a deoinhibition effect. However, in another congeneric species, *Zanthoxylum dissitum*, Ying-Zi et al. (2009) observed higher germination after seeds were scarified with sulfuric acid. Therefore, our results and those in *Z. dissitum* suggest that scarification is the main mechanism in this genus.

In *Condalia* spp. and *L. cestroides*, higher germination percentages in manually extracted seeds and gut-passed seeds than those from intact fruits were observed, suggesting that the seeds increased their germinability through a deoinhibition effect (Traveset 1998; Traveset and Verdú 2002; Robertson et al. 2006; Traveset et al. 2007). The speed of germination also suggested this pattern, even though it does not fully support it. In both species, the speed of seed germination for the extracted seeds was significantly faster than that for seeds residing in intact fruit, but the germination speed of extracted seeds was also faster than that for gut-passed seeds.

Therefore, in *L. cestroides* the results for germination speed suggest that even though birds provide a deoinhibition effect, they are not capable of promoting the greatest germination velocity possible for the species. In *Condalia* spp., gut-passed seeds showed similar speeds to intact fruits, thus birds do not seem to exert any benefit in the speed of germination, and deoinhibition may be the only factor affecting the germinability of this species. For *Condalia* spp., Renison et al. (2010) also found that seed germination was higher in manually extracted seeds of *Condalia microphylla* compared with intact fruits, but seeds ingested by *R. americana* showed lower germination percentages. Altogether, the comparison of our results and the previous work of Renison et al. (2010) for *C. ehrenbergiana* and *Condalia* spp. suggests that different bird species may exert different effects on seed germination, and that the different plant species also respond differently to gut passage. In addition, performing experiments that address the different *Condalia* species co-existing in the study area (*Condalia buxifolia*, *Condalia*

Fig. 3. Cumulative seed germination percentages for each seed treatment (G, gut-passed seeds; F, intact fruit; and M, manually extracted seeds) for (A) *Celtis ehrenbergiana*, (B) *Condalia* spp., (C) *Lantana camara*, (D) *Lithraea molleoides*, (E) *Lycium cestroides*, (F) *Schinus fasciculatus*, and (G) *Zanthoxylum coco*. Different letters beside the curves indicate significant differences, $P \leq 0.05$.



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microphylla, and *Condalia montana*) would be important for assessing the array of interactions between these plant species and their seed dispersers.

Only in *L. camara* do birds seem to exert both germination-promoting mechanisms, because statistically significant differences were observed between gut-passed seeds and manually extracted seeds, and between the manually extracted seeds and seeds from intact fruit in both of the variables analyzed. Thus, birds are highly important for seed germination of this species, which is considered a native invasive species because it is originally from other phytogeographic regions of the country (Giorgis and Tecco 2014). Therefore, these results are important if they are intended to develop a management strategy for this species.

Finally, in *S. fasciculatus*, the fruit pulp is firmly attached to the seed, making pulp removal by abrasion in bird guts possibly difficult, which might explain the higher germination and speed in manually extracted seeds compared with gut-passed ones. The low values obtained in gut-passed seeds suggested that in this species birds might be exerting a negative effect on seed germination. It is possible that other bird species, or other terrestrial dispersers, may be involved in pulp removal of *S. fasciculatus* fruit, producing different results. A deinhibition mechanism was observed in other congeneric species (*Schinus terebinthifolius*) from Argentina, Paraguay, and Brazil (Panetta and Mckee 1997; D'Avila et al. 2010).

Our detailed sampling effort to obtain defecated seeds contributes to a better understanding of the role that the community of frugivorous birds exerts on the seed germination of native plant species in the Chaco Serrano Woodland. In addition, except for *S. fasciculatus*, most species of the Chaco Serrano region depend on birds for their dispersal and germination, and therefore, birds facilitated the maintenance of their populations in this region. However, it would be ideal to repeat these experiments in field conditions, because it is possible that even though we did not use Petri dishes, the relatively small volume of soil used in each container within the experimental chambers may have been insufficient to allow inhibitors to leach away and may have altered the microorganisms available for decomposition, modifying the pulp breakdown as it would occur naturally in the soil (Robertson et al. 2006). These alterations may have exacerbated the inhibitor's effects observed here in intact fruits.

Finally, it is important to clarify that a faster germination speed could have either negative or positive consequences for plant populations, depending on climatic conditions of the area at the time when seeds germinate. The environmental conditions in the Chaco Serrano Woodland are very different in the wet and dry seasons (Torres and Galetto 2011). The species reported here are dispersed during the wet season (Díaz Vélez 2013). There-

fore, a faster germination speed may be advantageous for these species, because it would ensure seedling establishment during the wet period, as also occurs in other regions with pronounced seasonality, such as some temperate and desert areas (Traveset 1998). However, the exact consequences of different germination speeds for Chaco Serrano Woodland species requires further study. Germination speed is a complex variable and the advantages or disadvantages for each plant species may be influenced by the action of granivores and herbivores, as well as fluctuations in resource availability during seedling establishment (Daws et al. 2002; Yirdaw and Leinonen 2002). Also, it has been suggested that variability in germination speed adds temporal heterogeneity to seed germination responses, which may favour species establishment and regeneration (Izhaki and Safrieli 1990; Reid and Armesto 2011).

Concluding remarks

Despite the effects of frugivorous birds on seed germination, it is essential to establish the regeneration possibilities for the bird-dispersed plant species (Baskin and Baskin 1998), which remain unknown for many ecosystems in Argentina and South America (Reid and Armesto 2011). Because our study considers the seed dispersal of common plant species by the most abundant bird dispersers of the area (Ponce et al. 2012; Díaz Vélez 2013), we provide a broad view of the impact of bird species on these woody species. Frugivorous birds not only move seeds to different microsites, but also provide different treatments to the seeds, enhancing the chances of seed germination and plant establishment in the Chaco Serrano Woodland (Díaz Vélez 2013).

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