Research Article

Native seed dispersers may promote the spread of the invasive Japanese raisin tree (*Hovenia dulcis* Thunb.) in seasonal deciduous forest in southern Brazil

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Abstract

Hovenia dulcis (the Japanese raisin tree) is an invasive tree in seasonal deciduous forest in Southern Brazil. The species has an uncommon infructescence morphology, with dry fruits attached to fleshy peduncles that attract animals. This study assesses the interaction between the raisin tree and frugivore animals in seasonal deciduous forest and identifies potential dispersal relationships. Local fauna consumers and dispersers of the raisin tree were recorded by camera traps and by focal observation. Fruit removal was assessed by experiments that excluded certain animal groups while allowing access by others. We assessed seed dispersal by endozoochory by collecting the feces of birds and mammals found in the study area. The seeds found in the animal feces were then subjected to germination tests. A sampling effort of 2,305 traps/day was taken during the fructification period of the raisin tree. As a result, 28 species (16 birds and 12 mammals) were recorded from pictures taken by camera traps, and two more bird species were recorded via focal observation, for a total of 30 recorded species. Intact seeds of the raisin tree were found in the feces of *Cerdocyon thous* (crab-eating fox). No difference was found in seed germination between seeds obtained from fecal samples and the control. Ants can also be secondary dispersers of the raisin tree based on two observations made during the research field trips. The results obtained from this study suggest the raisin tree has different dispersal mechanisms with which different native dispersal agents are associated. Mid-size mammals, such as the crab-eating fox, are secondary dispersers by endozoochory, while birds are primary dispersers by sinzoochory.

Key words: invasive alien plant; dispersion; mammals; birds; germination

Resumo

Hovenia dulcis (uva-do-japão) é uma árvore invasora em florestas estacionais deciduais no Sul do Brasil. Esta espécie apresenta uma infrutescência com uma morfologia incomum, com frutos secos presos a pedúnculos carnosos, os quais atraem a fauna. Este estudo tem como objetivo avaliar as interações entre a uva-do-japão e animais frugívoros na floresta estacional decidual, assim como as relações potenciais de dispersão. A fauna local consumidora e dispersora da uva-do-japão foi registrada por meio de armadilhas fotográficas e observações focais. A remoção de frutos foi avaliada experimentalmente através de gaiolas de exclusão de diferentes grupos animais em diferentes tratamentos. Amostras fecais de aves e mamíferos encontradas na área de estudo foram coletadas para uma avaliação da dispersão de sementes por endozoocoria. As sementes encontradas nas fezes foram submetidas a testes de germinação. Um esforço amostral total de 2305 armadilhas/dia foi feito durante o período de frutificação da uva-do-japão. Como resultado, 28 espécies (16 de aves e 12 de mamíferos) foram registradas pelas armadilhas fotográficas, e duas outras espécies de aves foram registradas por observações focais, totalizando 30 espécies registradas. Foram encontradas sementes intactas da uva-do-japão nas fezes de *Cerdocyon thous* (graxaim). Não foi encontrada diferença na germinação das sementes obtidas nas amostras fecais e no controle. Formigas podem ser consideradas dispersores secundários da uva-do-japão baseado em observações feitas durante as saídas de campo. Os resultados obtidos com este estudo sugerem que a uva-do-japão apresenta diferentes mecanismos de dispersão aos quais diferentes agentes dispersores nativos estão associados. Mamíferos de médio porte, como o graxaim, são dispersores secundários por endozoocoria, enquanto aves são dispersores primários por sinzoocoria.

Palavras-chave: planta invasora; dispersão; mamíferos; aves; germinação

Introduction

Once an introduced species establishes a local population, dispersal mechanisms may determine the arrival of propagules to new sites, the speed of species spread, and consequently, invasion success [1]. Mutualistic dispersal interactions facilitate and accelerate invasion by non-native plants [2]. Plant species with fruit or other structures attractive to vertebrate seed dispersers often have high invasive potential [3-5]. Dispersal by vertebrates may increase dispersal effectiveness at different spatial scales [6], in seed germination success [7, 8], and in seed deposition on sites favorable to seedling establishment [9].

The characteristics of the fruit determine its potential dispersers, which animal groups or species will be attracted to it, and dispersal distance [10, 11]. Fruit color and size, nutritional content, and pulp to seed ratio are determinants for frugivore attraction and are associated with animal-mediated invasions [11]. The species phenology may also influence fruit consumption according to the season and persistence of fructification, compared to other species whose fruit are consumed by local fauna. Other factors affecting animal attraction are plant crown size, fruit density, fruit accessibility, and distribution patterns [10].

Plants that attract birds usually produce small, brightly colored fruit, while mammal-attracting plants bear larger, aromatic fruit of delicate taste [12]. Birds are the most important dispersal agents for invasive trees and shrubs, as they are more efficient long-distance dispersal vectors [5, 13], while mammals can ingest larger quantities and a greater diversity of fruit [14]. Mammals are therefore important endozoochory agents, since they can defecate intact seeds throughout their home ranges [15]. For instance, ungulate mammals such as deer, moose, and cattle can disperse invasive alien plant seeds consumed in large quantities, which pass intact through the digestive tract [16].

The Japanese raisin tree, *Hovenia dulcis* Thunb. (Rhamnaceae) is a deciduous tree species native to east Asia [17] and invasive in forests in South America and Tanzania [17-20]. It is a fast-growing tree, reaching 25 m in height, and reproducing sexually by seed [21]. Fructification takes place between March and October in Brazil [21], with peak production between June and July [22]. Infructescences are composed of dark, dry, globose capsules measuring 6-7 mm in diameter, containing 2-4 seeds.

These are attached to a brown peduncle, rich in carbohydrates, which becomes thick and fleshy when ripe [21, 23]. It is the fleshy peduncle that is attractive to animals [23].

Based on fruit morphology and on field observation, various authors have proposed that birds and bats are potential dispersers of the raisin tree. According to these authors, birds and bats tend to be attracted by the fleshy peduncles and carry them to surrounding perches, where seeds would then be released [17, 24]. Zhou [23], however, stated that these suppositions were contradictory because birds are normally attracted by small, brightly colored dispersal units, traits which do not apply to the raisin tree peduncles. Both in its native range as well as in South American forests, intact seeds of the raisin tree are consumed [25] and dispersed long distances in the feces of large-sized mammals [26-30]. There is no evidence of bird endozoochorous seed dispersal in eastern Asia [23, 31], where birds feed on peduncles but do not swallow or carry seeds to great distances. In general, because birds grab infructescences by the peduncle and take them a short way to feed on, long-distance dispersal and seed dropping are incidental [23].

The raisin tree was introduced to the western part of Santa Catarina state in the second half of the 20th century [32]. It was intensely cultivated from the 1980s on, when the regional agroindustry promoted its usefulness for shading poultry and hog farms [33]. Since its introduction, the raisin tree has been planted extensively on rural properties as a source for lumber, fuel and, energy production, shade in agriculture and pasture applications, and windbreaks [21, 33]. The raisin tree spread in a fragmented landscape [34, 35]. Populations of this invasive species are currently present in the interior of forest fragments in different successional stages and along borders. We assessed the interaction between the raisin tree and frugivore animals in seasonal deciduous forest and identified potential dispersal relationships. The questions addressed by this study were: (1) which agents function as consumers and dispersers of seeds of the raisin tree? (2) how far can the seeds be dispersed by animals, and how does dispersal affect seed germination? and (3) does the abundance of animal species vary with vegetation structure and at different densities of the raisin tree?

Methods

Study area
This study was undertaken in remnants of seasonal deciduous forest (SDF) in the Fritz Plaumann State Park, Santa Catarina, Brazil (Fig. 1) (between coordinates 27°16'18" and 27°18'57" S, 52°04'15" and 52°10'20" W), in Cfa (subtropical) climate according to the Köppen-Geiger classification system. The park covers 740 hectares and is the only protected area conserving SDF remnants since the region was colonized by European immigrants in 1912 [32, 35]. The natural vegetation has been regenerating since 1998, when the park was established, after almost 50 years of land use for logging, agriculture, and grazing [35]. Data collection was carried out in different successional stages as previously described by Dechoum [22].

Data collection
Preliminary field observations showed that the fructification period of the raisin tree in the study area spans from May to September, therefore the field work was concentrated in these months in 2012 and 2013. Experiments and observations were undertaken in ten 10 x 10m² plots 100m apart. The term disperser was standardized in this study for animals capable of transporting seed or fruit (comprising seed and peduncle) to distances that vary from centimeters to kilometers. Species gathering fruit directly off tree crowns were primary dispersers, while those collecting fruits off the ground were secondary dispersers [36]. The term endozoochory was applied when the seeds were swallowed and ultimately dispersed via defecation, while sinzoochory was used when the seeds were actively transported by animals that fed on parts of the trees but did not ingest the seeds [37]. Our study was authorized by the Environmental Agency of Santa Catarina State (FATMA) (license number 028/2012 – GERUC/DPEC).

Fig. 1. Location of the Fritz Plaumann State Park, Santa Catarina state, southern Brazil [extracted from 25].

Records of local fauna and potential consumers and dispersers of the raisin tree
Local fauna consumers and dispersers of the raisin tree were recorded in the study area by camera traps and focal observations. We placed ten digital Tigrinus® camera traps, one in each sampling plot. In eight of the plots the camera traps were directed at the base of trees of the raisin tree to document feeding on fruit on the ground [38], and in two plots the camera traps were installed on suspended tree platforms 2m off the ground to record arboreal animals. The camera traps were installed in three 40-60 hectare forest fragments, and were activated between April and September of 2012 and 2013.

Focal observations were undertaken between July and August 2012 and 2013, at peak fructification periods, to record birds feeding on the raisin tree. Bird visits were recorded only when the birds seized fruits or peduncles of the raisin tree, not when using trees only for perching (based on [39]). Eight trees bearing mature fruit and clear crown views were selected for observation, usually close to trails. The focal tree method is described by Galetti [38] and the observations were made using binoculars (Nikula® 8 x 30) and bird identification guides [40] at 10 m from each tree. Each focal tree was observed in the morning (from 7:00 to 11:00 am) and at the end of the afternoon (from 4:00 to 6:00 pm), totaling 48 hours of observations (16 hours in 2012 and 32 hours in 2013). Visiting species and fruit destination (dropped, consumed, or transported) were recorded during observations.

Fruit removal by different consumer groups
Fruit removal was assessed by experiments that promoted the exclusion of certain animal groups while allowing access by others. Two removal stations per plot were installed 10 m apart, each subdivided in four treatments 2 m apart in a square formation. Ten infructescences were used in each treatment and eight plots were used. The proportion of removed fruit was evaluated ten days after the beginning of the experiment, in July, 2013. Treatments were adapted from Galetti [38] as follows: a) open (control): infructescences were placed on 10 cm diameter plastic plates set on the forest floor, allowing access to all animals (mammals, birds, and invertebrates); b) access to large mammals: infructescences were placed on plastic plates mounted and nailed on top of 2l plastic bottles sticked 40cm above the ground, with Tangle-Trap® applied to the base of the bottles to prevent access by

terrestrial invertebrates [41] (only animals larger than 40 cm in height had access to infructescences); c) access to small mammals: infructescences were placed on plastic plates mounted on three 15 cm long wooden staves 5 cm above the ground inside metal cages measuring 26 x 10.5 x 14 cm with 14 x 10.5 cm openings (adapted Young type traps) - Tangle-Trap® was applied on the wooden staves to avoid access by ants (rodents and marsupials were considered small mammals) and; d) ant access: infructescences were placed on plastic plates inside enclosed metallic traps (Young type traps).

Dispersal distance and seed viability
String reels attached to infructescences were used to determine how far seeds of the raisin tree are transported by animals as well as dispersal destination (method adapted from [42, 43]). Five string reels attached to infructescences were placed in front of each of the ten camera traps directed at the base of trees. Reels were revised and infructescences replaced at every monitoring activity (n=11), totaling 550 repetitions. The experiment was checked every 30 days, and the infructescences were classified according to destination, in accordance with Peres and Baider [44]: (1) not displaced and intact; (2) consumed but not displaced (reel not unspooled, peduncle consumed); (3) displaced (infructescence can no longer be found at the end of unspooled string); (4) effectively dispersed (infructescence displaced, peduncle attached to string). Monitoring was maintained during the span of fructification of the raisin tree, between May and September, both in 2012 and in 2013. The string reels and infructescences were replaced every time their appearance or odor showed they were no longer fresh, as well as when they were consumed or taken away. Displacement was measured in straight lines from the experiment installation site to the location where infructescences were found.

The assessment of seed dispersal by endozoochory was implemented by collecting feces of birds and mammals found in the study area, analyzed for presence or absence of seeds of the raisin tree. Existing trails in forest remnants in different sucessional stages were covered. Bird and mammal feces were separated and, whenever possible, the species to which the fecal sample belonged was identified based on characteristics such as size, format and odor. Approximately 140 km were covered in a total of 20 field trips (10 per year).

Seeds found in animal feces were subjected to germination tests. The feces collected were washed in running water over a fine mesh sieve. After drying in open air, the seeds were placed to germinate on vermiculite substrate in trays in a greenhouse [38]. A control treatment was also undertaken using the same number of seeds that did not go through animal digestive tracts. These seeds were obtained from fruit found on the forest floor. All rotting fruit or fruit covered by fungi were discarded.

Relationship between fauna and vegetation structure
In the same study period, Dechoum [22] evaluated vegetation structure and diversity in the established plots. Part of the data obtained by Dechoum [22] was used to check whether the frequency of animal species was associated with vegetation structure and with the presence of the raisin tree. All woody plants larger than 5 cm in diameter at ground level were sampled, identified, and their heights were visually estimated. Total density, average height, average basal area, species richness of native woody plants, and density of the raisin tree were quantified in each sampling plot [22]. These data were combined with the animal records for each plot to determine whether the abundance of animal species was related to vegetation structural characteristics and to density of the raisin tree.

Data analysis
The number of species occurrences in each plot was obtained from camera trap records. A trapping success index (TSI) was calculated for each species using the formula TSI = $T_{species}/S_{plot}$, where $T_{species}$ = number of photographs of each species. $plot^{-1}$, and S_{plot} = number of camera traps.$plot^{-1}$.day^{-1}(i.e. the sampling effort undertaken in the same plot) [45]. Same species records in the same plot obtained on

the same day at intervals shorter than one hour were considered as one record to avoid recording the same individual [46]. The photographic records that show animals feeding on fruit were used to indicate consumer species of the raisin tree. Bird observations denoted which species were consuming fruits or peduncles of the raisin tree. These observations were used to determine whether the species' foraging behavior may help to disperse the seeds of the studied invasive tree.

A nested analysis of variance by permutation (5,000 replicates) was run in order to determine whether one of the groups was more representative than the others in removing raisin tree fruit from cages in the exclusion experiment. We used the permutation test because the data obtained did not present either a normal distribution nor homoscedasticity. The analysis was run in the R software version 3.2.1 [47] using "coin" package [48] and "sciplot" package [49] for the graphic representation of the number of fruits (mean ± se) removed by group of animals. Descriptive statistics (percentages of fruit removal, mean ± sd) were used to synthesize the data from the string reel removal distance experiment.

The searches of existing trails resulted in the collection of 27 fecal samples, 10 of which contained seeds of the raisin tree. Because the number of seeds varied among feces samples, one seed in each fecal sample and one in the control lot were selected at random, and the sum of germinated and non-germinated seeds from each condition were used in a contingency table. The Pearson's chi-squared test with the Yates' continuity correction was run to test whether the germination of seeds from animal feces and from the control lot were different.

The vegetation variables – density of the raisin tree, total tree richness in sample plots, average tree height and average tree basal area including the raisin tree - were transformed by standardization. A canonical correspondence analysis (CCA) was carried out [50] to relate vegetation variables to the frequency of animal species recorded in each plot. The CCA was first performed with all vegetation descriptive variables. When collinearity among variables was verified using the Variance Inflation Factor (VIF), those presenting lower correlation values were selected. The selected variables were: total density, density of the raisin tree, and species richness in each plot. Then a new CCA was performed using the R software [47] and the package "vegan", and a Monte Carlo permutation test was carried out to determine which variable is the most significant in the canonical correspondence analysis [51].

To check for spatial autocorrelation between experimental sites, we ran a Mantel's test between a Bray-Curtis dissimilarity matrix (based on the frequency of the sampled species in each plot) and an Euclidian distance matrix of each sampled site (based on their geographical coordinates), using the R software version 3.2.1 [47] "ecodist" package [52]. No spatial autocorrelations were found (r=0.07, p=0.16).

Results
Animal species feeding on the raisin tree
A sampling effort of 2,305 traps/day was made during the fructification period of the raisin tree in 2012 and in 2013. As a result, 28 species were recorded: 16 birds and 12 mammals (Tables 1 and 2). A wild rodent not identified at the species level belongs to the Cricetidae family (Fig. 2). Apart from species identified from pictures taken by camera traps, two additional bird species were recorded by focal observations, for a total of 30 species.

Table 1. Mammals recorded by camera traps installed in seasonal deciduous forest fragments in the Fritz Plaumann State Park (Brazil) in 2012 and in 2013. TSI = trapping success index (TSI = $T_{species}/S_{plot}$).

Order	Family	Latin name	Common name	Number of records	TSI	*H. dulcis* consumption
Artiodactyla	Cervidae	*Mazama nana*	Southern bororó	39	0.17	No
Carnivora	Felidae	*Leopardus tigrinus*	Oncilla	2	0.009	*
		Leopardus wiedii	Margay	4	0.02	*
		Puma yagouarondi	Jaguarundi	1	0.004	*
	Canidae	*Cerdocyon thous*	Crab-eating fox	20	0.09	Yes
	Procyonidae	*Nasua nasua*	South American coati	20	0.09	Yes
		Procyon cancrivorus	Crab-eating raccoon	3	0.01	No
Primates	Cebidae	*Sapajus nigritus*	Black-horned tufted capuchin	13	0.06	No
Rodentia	Cuniculidae	*Cuniculus paca*	Spotted paca	6	0.03	No
	Cricetidae	(not identified)	-	1	0.004	No
	Sciuridae	*Guerlinguetus ingrami*	Southeastern squirrel	3	0.01	Yes
Xenarthra	Cingulata	*Dasypus novemcinctus*	Nine-banded armadillo	60	0.26	Yes
			Total	**172**		

*carnivorous species.

Seven species consumed fruits of the raisin tree: *Cerdocyon thous* (crab-eating fox), *Dasypus novemcinctus* (nine-banded armadillo), *Guerlinguetus ingrami* (southeastern squirrel), *Nasua nasua* (south american coati) (Fig. 3), *Penelope obscura* (dusky-legged guan), *Pyroderus scutatus* (red-ruffed fruitcrow), and *Trogon surrucura* (southern surucua trogon). The first five consumer species may play the role of secondary disperser, since they feed on peduncles and fruits off the ground, while the last two species may be primary dispersers, gathering fruits directly off tree crowns. Therefore, 44.4% of the mammals and 20% of the birds that have fruit as part of their diet consumed fruits of the raisin tree and are potential seed dispersal agents of the species.

One specimen of red-ruffed fruit crow and another one of southern surucua trogon were recorded feeding on peduncle of the raisin tree during informal field observations. The southern surucua trogon specimen pulled a tiny branch off the infructescence and perched on a neighboring tree. It shook the branch in its beak, trying to get rid of the globose fruit, and consumed only the fleshy peduncle. Then it repeated the same procedure and moved to a more distant tree for foraging.

Fruit removal, dispersal distance and effective dispersal agents
The exclusion and control treatments did not differ in the number of removed fruits (Z=1.84, p=0.21), i.e, no animal group was a more efficient remover of fruits of the raisin tree than any other in the study site (Fig. 4).

Table 2. Birds recorded in seasonal deciduous forest fragments in the Fritz Plaumann State Park (Brazil) in 2012 and in 2013. TSI = trapping success index (TSI = $T_{species}/S_{plot}$).

Order	Family	Latin name	Common name	Number of records	TSI	*H. dulcis* consumption
Columbiformes	Columbidae	*Leptotila rufaxilla*	Gray-fronted dove	23	0.09	No
		Leptotila verreauxi	White-tipped dove	3	0.01	No
Galliformes	Cracidae	*Penelope obscura*	Dusky-legged guan	17	0.07	Yes
Gruiformes	Rallidae	*Aramides saracura*	Slaty-breasted wood-rail	8	0.03	No
Passeriformes	Conopophagidae	*Conopophaga lineata*	Rufous gnateater	10	0.04	**
	Corvidae	*Cyanocorax chrysops*	Plush-crested jay	2	0.009	No
	Cotingidae	*Pyroderus scutatus**	Red-ruffed fruitcrow	1	-	Yes
	Dendrocolaptidae	*Dendrocolaptes platyrostris*	Planalto woodcreeper	1	0.004	**
	Thraupidae	*Lanio melanops*	Black-goggled tanager	1	0.004	No
		Saltator similis	Green-winged saltator	2	0.009	No
		Tachyphonus coronatus	Ruby-crowned tanager	1	0.004	No
	Turdidae	*Turdus leucomelas*	Pale-breasted thrush	49	0.21	No
		Turdus albicollis	White-necked thrush	3	0.01	No
		Turdus rufiventris	Rufous-bellied thrush	60	0.26	No
Strigiformes	Strigidae	*Megascops* sp.	-	1	0.004	***
Tinamiformes	Tinamidae	*Crypturellus tataupa*	Tataupa tinamou	1	0.004	No
		Crypturellus obsoletus	Brown tinamou	1	0.004	No
Trogoniformes	Trogonidae	*Trogon surrucura**	Southern surucua trogon	1	-	Yes
			Total	**185**		

*species recorded in focal observations.
**insectivorous species.
***carnivorous species.

Fig. 2. Unidentified Cricetidae specimen.

The destinations of peduncles tied to string reels in the dispersal distance experiment were: 91.6% (n=504) were not displaced, remaining intact; 4.7% (n=26) were consumed but not displaced; 2.7% (n= 15) were consumed and displaced, but the fruits were not found at the ends of the strings; and 0.9% (n= 5) were effectively dispersed, i.e., displaced and found still tied to the strings. The average displacement distance of fruit effectively dispersed and consumed was 1.04m, varying from 8.7m to 0.35m from the initial position.

The total number of seeds found in feces was 73, and the largest fecal sample with the highest number of seeds (n=53) belonged to a crab-eating fox that consumed both fruit and peduncle. No difference was found in seed germination between seeds obtained from fecal samples and those in the control group (χ^2=0.23, p=0.63, df=1). This suggests that the passage of seeds through the digestive tracts of animals does not influence seed germination of the raisin tree. Because the seeds in the feces were both intact and germinated, this species may be considered a seed disperser of the raisin tree.

Fig. 3. *Cerdocyon thous* **(crab-eating fox) (A),** *Nasua nasua* **(South American coati) (B) and** *Guerlinguetus ingrami* **(southeastern squirrel) (C), consumers of** *Hovenia dulcis* **recorded by camera traps in seasonal deciduous forest fragments in the Fritz Plaumann State Park (Brazil). In "C", suspended tree platforms were used to record arboreal animals.**
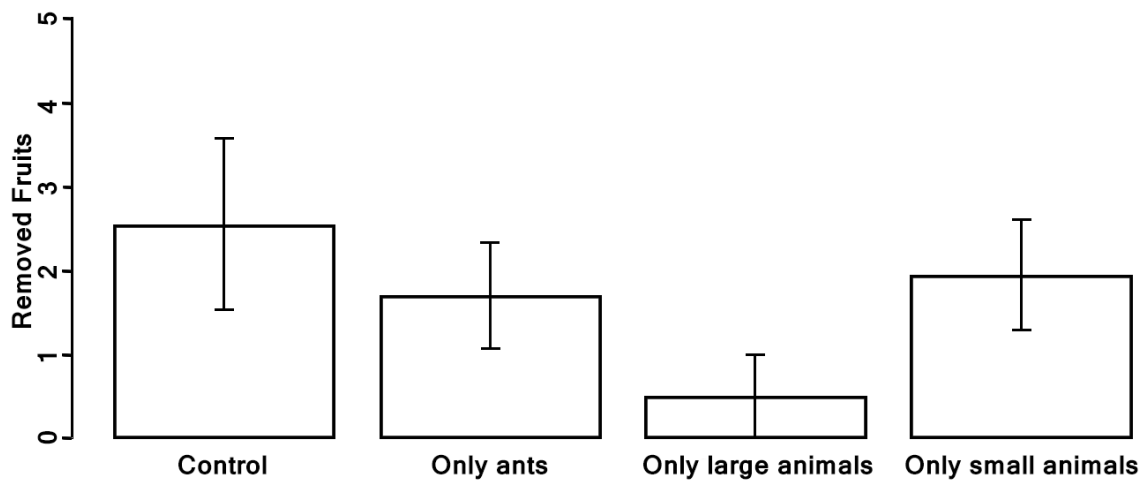
Fig. 4. *Hovenia dulcis* removed fruit numbers (mean ± standard error) per treatment in experiments undertaken in fragments of seasonal deciduous forest in the Fritz Plaumann State Park (Brazil). Treatments were compared using a nested analysis of variance by permutation. Treatments (x axis): control - all animals had access to infructescences; only ants - only ants had access to infructescences; only large animals - only animals larger than 40 cm in height had access to infructescences; only small animals – only mammals smaller than 14 cm had access to infructescences.

Relationship between fauna and vegetation structure

The CCA results showed that the three vegetation variables used (total tree density, total tree richness, and density of the raisin tree) explained 66.5% of animal species variance (total variance = 1.39, constrained variance = 0.93), leaving 33.5% of data variance unexplained (Fig. 5). The variance proportions explained by the two first axes are CCA1: 51.8% (F=6.19, p<0.05), and CCA2: 29.2% (F=3.49, p<0.01); i.e., the sum of both axes represent 81% of the relation between vegetation variables and animal species. The Monte Carlo permutation test (F=3.98, p<0.05) also indicated a significant correlation between species presence in plots and vegetation variables. The first ordination axis, which covers most of the variance, was mainly correlated with total tree richness (score = -0.977) and density (score = -0.884). On the second axis, the abundance of the raisin tree (score = 0.752) best explained data distribution. According to the Monte Carlo permutation test, the most significant variable for the analysis was total tree density (F=5.37, p<0.05).

The species most strongly correlated to the abundance of the raisin tree are either omnivore or frugivore [dusky-legged guan, *Sapajus nigritus* (black-horned tufted capuchin), crab-eating fox, *Lanio melanops* (black-goggled tanager) and *Turdus albicollis* (white-necked thrush)]. The species *Megascops* sp., the crab-eating fox (score = 1.131) and the dusky-legged guan (score = 0.765), which are some of the species that were best correlated to the second axis (Fig. 4). As abundance of the raisin tree best explained the data on the second axis, the analysis corroborates the association between the raisin tree and these animal species. Some mammal species were more abundant in plots where abundance of the raisin tree was higher, such as the crab-eating fox and the black-horned tufted capuchin, both strongly related to the abundance of the raisin tree in the CCA.
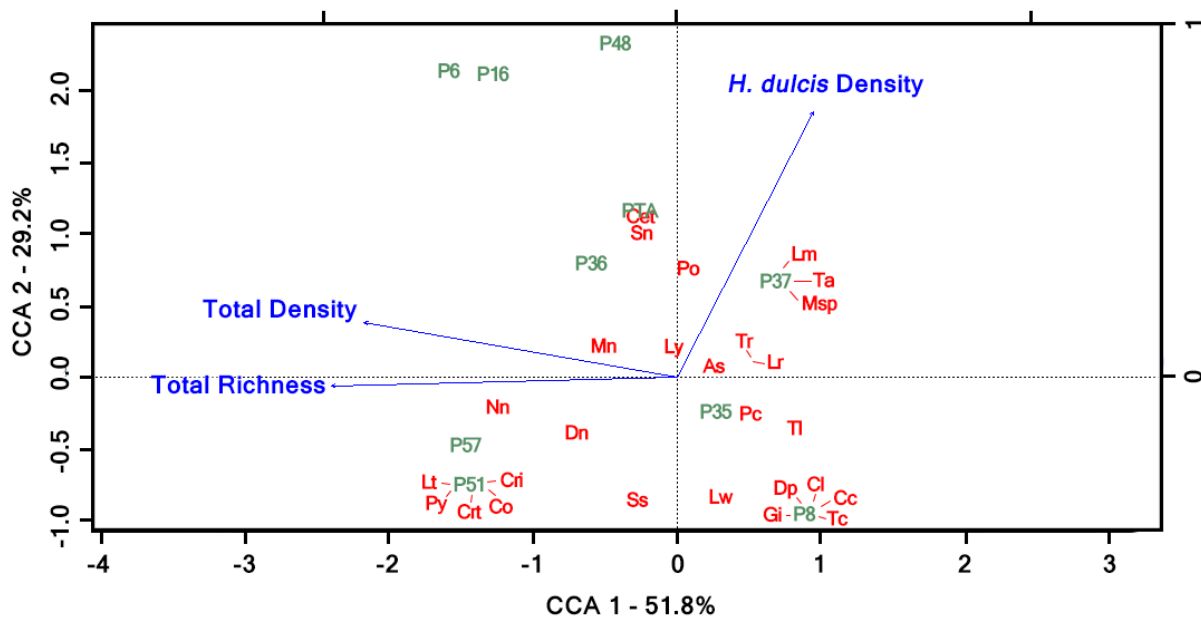
Fig. 5. Canonical correspondence analysis (CCA) of total tree density, total tree richness, and *Hovenia dulcis* density, including frequency of animal records in plots installed in fragments of seasonal deciduous forest in the Fritz Plaumann State Park (Brazil) in 2012 and in 2013. The three vegetation variables explained 66.5% of animal species variance. The first ordination axis was mainly correlated with total tree richness (score = -0.977) and density (score = -0.884), while on the second axis, the *H. dulcis* abundance variable (score = 0.752) best explained data distribution. Legend: mammals – Cet: *Cerdocyon thous*; Cri: Cricetidae; Cp: *Cunuculus paca*; Dn: *Dasypus novemcinctus*; Gi: *Guerlinguetus ingrami*; Lt: *Leopardus tigrinus*; Lw: *Leopardus wiedii*; Mn: *Mazama nana*; Nn: *Nasua nasua*; Pc: *Procyon crancrivorous*; Py: *Puma yagouarondi*; Sn: *Sapajus nigritus*. Birds – As: *Aramides saracura*; Cl: *Conophaga lineata*; Co: *Crypturellus obsoletus*; Crt: *Crypturellus tataupa*; Cc: *Cyanocorax chrysopsl*; Dp: *Dendrocolaptes platyrostrisI*; Lm: *Lanio melanops*; Lr: *Leptotila rufaxilla*; Lv: *Leptotila verreauxi*; Msp: *Megascops* sp.; Po: *Penelope obscura*; Ss: *Saltator similis*; Tc: *Tachyphonus coronatus*; Ta: *Turdus albicollis*; Tl: *Turdus leucomelas*; Tr: *Turdus rufiventris.*

Discussion

The results obtained from this research suggest the raisin tree has different dispersal mechanisms with which different native dispersal agents are associated. Mid-size mammals are secondary dispersers by endozoochory, such as the crab-eating fox, while birds are primary dispersers by sinzoochory. Similar results had been observed in the species native range both for birds and for medium and large-size mammals [23, 31]. Birds are considered the most important long-distance dispersal agents among animal seed vectors [5]. However, the raisin tree seems to be an exception since birds did not displace seeds to greater distances by endozoochory, but only to short distances from reproductive trees by sinzoochory.

Ants may also contribute to the establishment success of invasive alien plants [2] and can be considered secondary dispersal agents of the raisin tree, as they were found to displace seeds off the forest floor in the exclusion experiment using cages. Secondary dispersal may take place when an animal removes and carries seeds that have already been displaced, dropped, or defecated by another frugivore animal in repeated short-distance dispersal events, increasing the chance of seeds escaping the influence of the mother-plant [53]. Two observations corroborating the evidence of seed dispersal by ants were made during the research field trips. On one occasion, *Atta sexdens* ants were seen carrying seeds of the raisin tree across the forest floor. On another occasion an ant nest from which

seedlings of the raisin tree were growing was found (Fig. 6); however, determining the ant species to which this nest belonged was not feasible, as it had been abandoned [54].

Fig. 6. Abandoned ant nest with germinating *Hovenia dulcis* seedlings.

The raisin tree seems to benefit from a small number of animal species compared to the total number of species observed: 44.44% of recorded omnivorous mammals and 20% of omnivorous birds were observed consuming fruit of the raisin tree. These species, however, are also the most frequent ones, suggesting that the raisin tree may benefit mainly from omnivore or generalist animals. It is a well-known pattern that invasive plants often attract generalist vertebrate dispersal agents [55]. Considering the mammals recorded in this study, for example, with the exception of carnivores represented by three wild cat species, the other nine species are omnivores, frugivores, or occasionally include fruit in their diets [56, 57]. It is important to stress a possible limitation of our study in that the fruit removal experiment was conducted at the time of raisin tree fructification when there is a large supply of fruit on the ground, which may have satiated the fauna and interfered with the results of the experiment.

The crab-eating fox, one of the mammals confirmed as dispersal agent of the raisin tree in the study area, is an opportunistic species whose diet changes according to the availability of food resources [58]. The species is also an efficient seed dispersal agent since it feeds on large quantities of seed and is capable of covering long distances [59]. Besides aiding long-distance dispersal, it generates new invasion foci. It often defecates in open areas, forest borders or trails, sites which favor seed germination and seedling establishment of the raisin tree [21].

Three bird species dusky-legged guan, red-ruffed fruitcrow, and southern surucua trogon, were recorded consuming fruits of the raisin tree. Among the bird species observed, 13 are fruit consumers and only three are not [*Conopophaga lineata* (rufous gnateater), *Dendrocolaptes platyrostris* (planalto woodcreeper), and *Megascops* sp.] [60]. The species *Leptotila rufaxilla* (gray-fronted dove), dusky-legged guan, *Aramides saracura* (slaty-breasted wood-rail), *Cyanocorax chrysops* (plush-crested jay), black-goggled tanager, *Tachyphonus coronatus* (ruby-crowned tanager), *Crypturellus obsoletus* (brown tinamou), and southern surucua trogon live in forest habitats; *Leptotila verreauxi* (white-tipped dove), red-ruffed fruitcrow, *Saltator similis* (green-winged saltator), and white-necked thrush live in forest borders or clearings; and *Turdus leucomelas* (pale-breasted thrush), *Turdus rufiventris* (rufous-bellied thrush), and *Crypturellus tataupa* (tataupa tinamou) are adapted to anthropogenic

habitats [61]. Considering the current vegetation in the study area, it would be logical to conclude that these thirteen fruit consumer species can use or live in fragments in different successional stages as well as use the raisin tree in their foraging, becoming potential dispersal agents of the invasive species.

Implications for Conservation

The introduction of invasive species to forest ecosystems can have important impacts on interaction webs, disrupting mutualisms [62], and/or playing the role of the main source of fruits and seeds. In this study, the raisin tree seems to have been successfully integrated into the dispersion web as a generalist, benefitting from the dispersal services of both vertebrates and invertebrates.

As most animal species recorded were frugivore or omnivore, their presence in the study area may depend on fruit availability [30] produced by both by native and non-native plants. Some invasive plants may become essential food items for native animals, particularly in areas where most of the native vegetation has been destroyed or during food shortage periods [63-65]. In the Galapagos Islands, for example, asynchronous fruit production by the introduced species *Rubus niveus* (snowpeaks raspberry) and *Psidium guajava* (guava), compared to most native species, possibly represents a competitive advantage for these invasive species [65]. In areas where invasive plants are part of native frugivore animal diets there may be a conservation conflict between controlling the invasive species and maintaining the status of frugivore populations, especially when other threats such as habitat destruction have reduced populations of native species [66].

In the study area, at least 15 zoochorous plant species totally or partially synchronize with the raisin tree in fruit production in seasonal deciduous forest [54]. The raisin tree is therefore a potential competitor for seed dispersal agents, so the removal of the invasive species could promote the reestablishment of frugivore animals and native plant species [67]. Fruit characteristics that may affect frugivore choice, such as size, morphology, color, peel edibility, chemical defenses, crown size, and phenology must be considered when selecting species for restoration or enrichment [63, 66, 68]. The offer of alternative food resources to animals is one strategy to mitigate impacts from the control of invasive alien species bearing fleshy fruit [10, 69]. The raisin tree control programs must therefore include complementary management practices, especially planting native species to replace the food resources provided by the target species to help restore previously functioning interaction networks.

Acknowledgements

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