Supplementary Information for the article "**From structure to function in mutualistic interaction networks: topologically important frugivores have greater potential as seed dispersers**"

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Appendix S1. Description of sampling methodologies

Study area

Guacamayas. The Civilian Society Reserve Hacienda Betancí-Guacamayas (8° 11' 72''N, 75° 32' 78'' W) is located in the Buenavista municipality in the department of Córdoba, Colombia (Figure S1). The average altitude is 70 m a.s.l., and average temperature is 28° C. The reserve covers 460 ha and harbors fragments of tropical dry forest structurally connected by riparian corridors, all of which is embedded in a pasture matrix devoted to extensive livestock farming. This use of the land is a sustainable alternative that reduces the environmental impact of traditional production systems.

El Pino. The El Pino farm (8° 25' 11.33" N, 76° 3' 24.43" W) is located in the small town of Las Palomas, in Monteria, Colombia, on the Sinú River basin (Figure S1). El Pino has an area of 2,350 ha and is located at an average altitude of 35 m a.s.l. with an average annual temperature of 29° C. The farm, which has an artificial water channel that runs through it, is devoted to extensive livestock farming. The study area was limited to 150 ha that have been devoted to the conservation of a native forest remnant which is surrounded by a pasture matrix and is highly isolated from other forest fragments.

Figure S1. Geographical context of the study area and location of sampling sites (walking transects, bird census stations, and camera trapping stations).
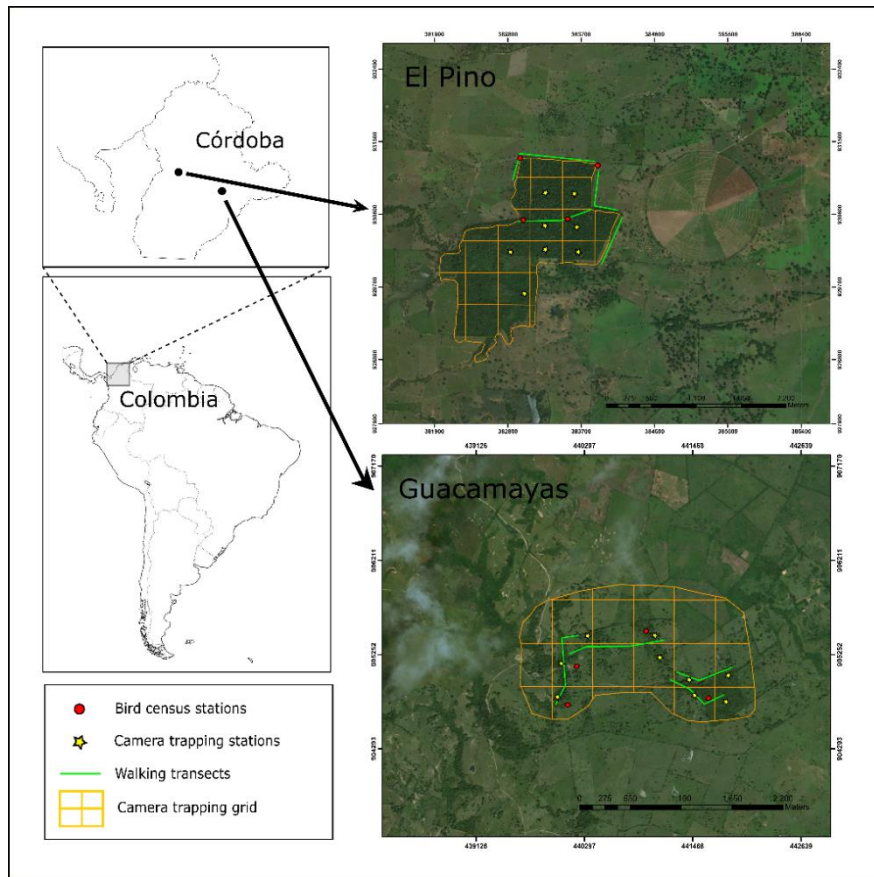
Behavioral characteristics of frugivores

For each frugivore species observed in a frugivory event, we collected the following behavioral information suitable for representing the species' potential to act as seed disperser:

Foraging strata

During each frugivory event we recorded the vegetation stratum in which the foraging took place, using the following level categories:

*Ground***:** The fruit had fallen from the source plant and the animal picked it up directly from the ground.

Low: The fruit was picked from plant's branches from zero to two meters off the ground.

Medium The fruit was taken from a height of between two and eight meters off the ground.

High: The fruit was taken from heights above eight meters off the ground.

Fruit handling type

For each frugivory event, we characterized the type of fruit/seed handling behavior of the animal using the following classification:

Predation: The frugivore opened the fruit to consume the seed by cracking it. This behavior is common among Psittacidae family.

Pulp consumption: The frugivore consumed the fleshy pulp, without swallowing or cracking seeds. This behavior is common in small vertebrates consuming large fleshy fruits.

Fruit transport: The frugivore transported the fruit (in its mouth or beak) from the encounter site to another location. This behavior may be followed by other handling types when, after displacement, it was still possible to watch how the animal handled the fruit.

Endozoochory: The frugivore swallowed the entire fruit, later releasing the seed(s) intact via regurgitation or defecation.

Appendix S2. *Sampling completeness and interaction accumulation curves*

In order to evaluate the representativeness of frugivore-plant interaction sampling we built interaction accumulation curves, using the number of observed interactions in relation to sampling effort, the latter being measured as the number of interaction events recorded (Chacoff et al., 2012; Jordano, 2016). In this procedure, the expected number of interactions was calculated using the Chao1 estimator, and it was compared to the number of interactions observed as a percentage of occurrence of interactions (Chacoff et al., 2012; Chao et al., 2009). Calculations were made using EstimateS 9.1.0 software (Colwell, 2013) and showed that the sampling effort applied led to the detection of 80% of expected interactions in both localities (Figure S2), suggesting the high representativeness of our interaction matrices.

Figure S2. Interaction accumulation curves representing the Chao1-estimated expected interactions for Guacamayas (A) and El Pino (B).
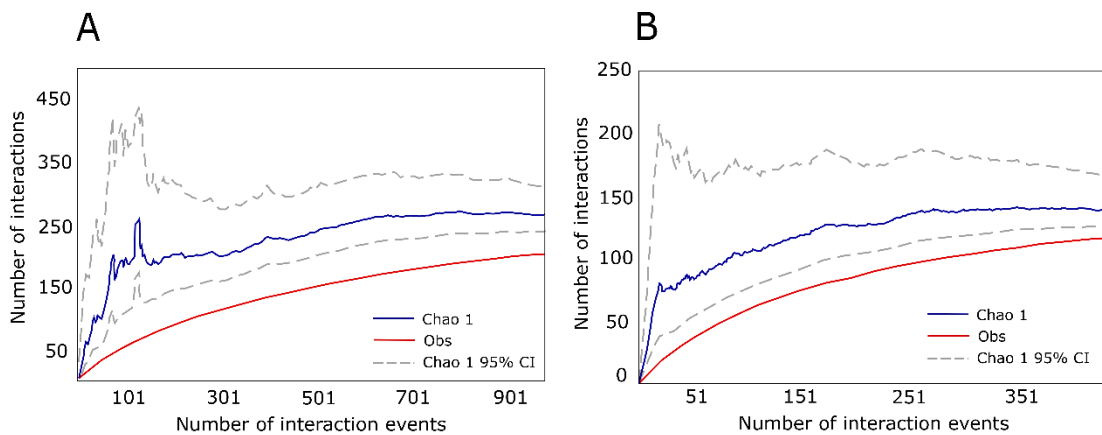
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Appendix S3. Morphological and behavioral characteristics of frugivores

In order to represent the functional effect that each frugivore species may provide, through seed dispersal, to community-level plant recruitment, we took into account a set of morphological and behavioral characteristics of frugivores, that are considered to affect plant recruitment, by modifying pre- and post-dispersal seed fate as well as seedling establishment (Table S3). These frugivore characteristics were quantitative parameters whose values were assumed to be positively correlated with the potential of frugivores to perform as seed dispersers.

Table S3. Morphological and behavioral characteristics of frugivores and their relationship with the species' potential as a seed disperser.

Frugivore characteristic	Relationship to seed dispersal effectiveness
Body mass	From a whole community, coarse grain perspective, larger frugivores show longer gut retention times (Karasov & Levey, 1990; Levey & Grajal, 1991) which, consequently, leads to longer dispersal distances and concomitant recruitment advantages (due to a variety of factors: escape from specialist predators and pathogens, avoidance of conspecific competition, genetic advantages, etc. Wotton & Kelly, 2011; Jenkins et al., 2007; Pérez-Méndez, Jordano, & Valido, 2015)
Foraging strata	Vegetation vertical stratification in tropical forests affects frugivore-plant interactions (Schleuning et al., 2011). Species able to forage in different strata can reach more plants species (Malmborg & Willson, 1988), and disperse seeds across a wider range of forest microsites (Schleuning et al., 2011). Thus, from the plant community perspective, they would offer a better seed dispersal service to a wider variety of species.

Forest habit	This represents seed dispersers role at a landscape scale, bearing in mind that forest habitat represents different microhabitats suitable for recruitment of different forest species (areas under tree canopies and small forest gaps; e.g. Wenny & Levey, 1998), but that deforested land (here pastures devoted to cattle grazing and crops) represents an unsuitable habitat for forest species in general (Holl, Loik, Lin, & Samuels, 2000). Thus, a frugivore dependent on forest cover would disperse most seeds under forest canopies and forest gaps, both within forest remnants, but also in forest islands or plantations (very relevant for forest recovery; Zahawi, Holl, Cole, & Reid, 2013).
Handling type	Seed germination and the establishment of a new seedling are conditioned by the embryo surviving fruit/seed handling by the frugivore (Jordano, 2014). Some behaviors, such as pulp picking, decrease dispersal success whereas others, such as endozoochory, largely favor dispersal success (Simmons et al., 2018).
Range of fruit size	Fruit and seed size is an important trait in the reproductive success of plants (Leishman, Wright, Moles, & Westoby, 2000). Different seed sizes can be advantageous depending on species' reproductive strategies and environmental conditions (Dalling & Hubbell, 2002). Frugivores able to consume a larger diversity of fruit sizes would therefore provide seed dispersal service to plants with a greater diversity in life histories, distributing the advantages of dispersal among more functional types of plants.

Estimation of frugivore characteristics

Body mass. The parameter used was the logarithm of the median of the body mass for each frugivore species (Wilman et al., 2014).

Foraging strata. For each frugivore species, we calculated the diversity of foraging strata used by means of a Simpson index applied to the frequency of use of each strata category. The following equation was used:

$$Foraging\ strata = 1 - \frac{\sum_{i=1}^{s} n_i(n_i - 1)}{N(N-1)}$$

Where,
S: number of strata
N: total observations
n: number of observations per strata

Forest habit. In order to determine the frequency of use of forest habitat by each frugivore species, we considered the abundance of that particular frugivore at sampling stations (census points and camera trapping stations) representing varied values of forest cover. Forest cover was estimated in a 100-meter radius area around sampling stations (Figure S3), by using the Normalized Difference Vegetation Index (NDVI). This index quantifies the amount and type of vegetation based on the measurement of radiation intensity in the different bands of electromagnetic spectrum emitted or reflected by the vegetation (Purevdorj, Tateishi, Ishiyama, & Honda, 1998). For this we used LandSat 8 images of 30-meter resolution (https://landsat.gsfc.nasa.gov/landsat-8/landsat-8-bands/). Image pixels were classified into two categories (forest and non-forest), where forest corresponded to those of NDVI >0.4 (Figure S3). We verified visually whether areas with pixels of lower values corresponded to open non-forest vegetation types or areas with few and scattered trees.

The calculation of NDVI is shown by the formula:

NDVI = (NIR — VIS) / (NIR + VIS)

Where,

VIS = Red visible (Band 4, LandSat 8)

NIR = Near infrared (Band 5, LandSat 8)

With this information, we determined the frequency of forest habitat use according to forest cover using the following formula:

$$Forest\ habit = \frac{\sum_{i=1}^{s} \left(\frac{f_i}{R_i}\right) * \left(\frac{FP_i}{P_i}\right)}{N}$$

Where,

S: number of sampling stations

f: frequency of occurrence of the frugivore species being observed at the point i

R: total repetitions of point i

FP: pixels corresponding to forest at the point i (100-meter radius)

P: total pixels at point i (100-meter radius)

N: number of sampling stations in which the frugivore species was registered

Higher values indicate higher frequency of occurrence of a frugivore species in sites with high forest cover.

Figure S3. Zoom examples of forest cover estimation within a 100-meter radius around sampling stations. Dark green areas indicate forest and light green areas non-forest, based on classification of pixels from NDVI values.
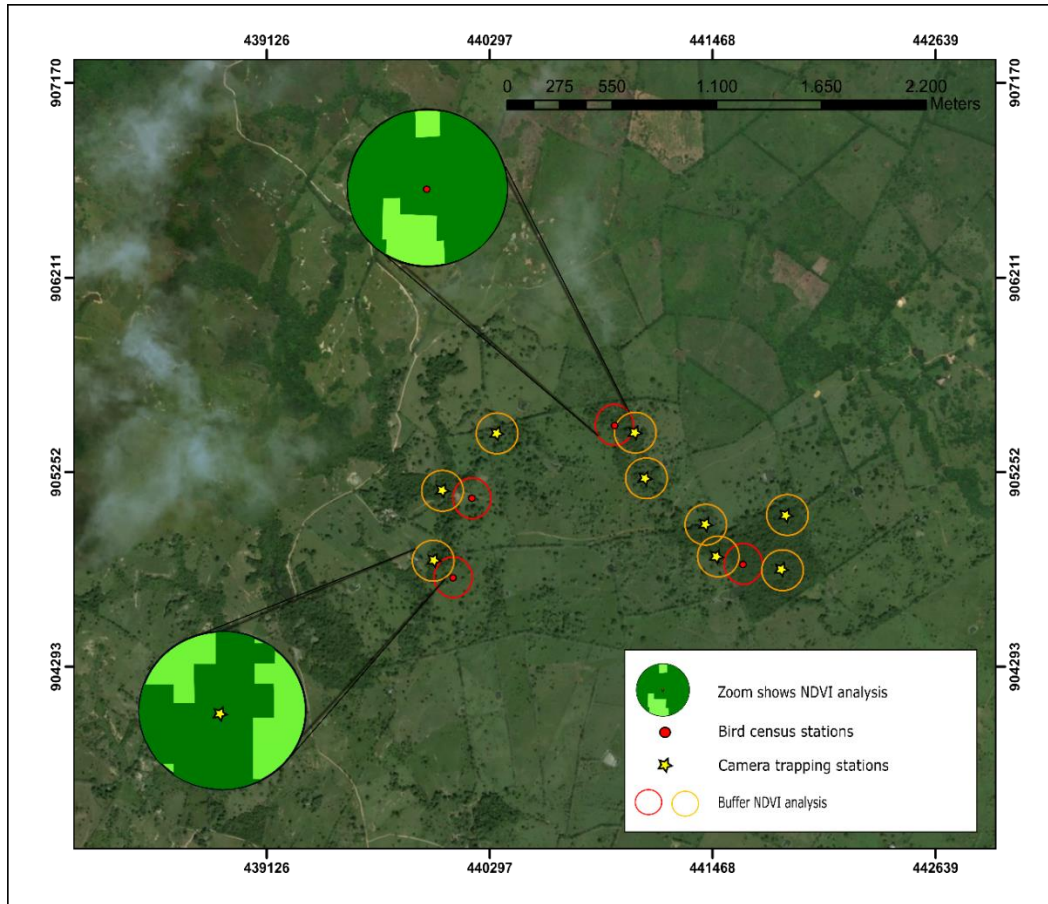
Handling type. We considered four different types of fruit/seed handling behavior observed in the field: predation, pulp consumption, fruit transport and endozoochory. For each handling type, we assigned an effect value, which reflected quantitatively the effect of handling in terms of seed survival, in a gradient from negative to positive and from seed predation to legitimate seed dispersal: predation, -1; pulp consumption, 0.1; fruit transport, 0.3; endozoochory, 1. For each frugivore species, the qualitative metric was estimated as the sum of the products of the frequency of occurrence and the effect value of the various handling types across frugivory events, as indicated in the following equation:

$$Handling\ Type = \sum_{i=1}^{S} (n_i * C_i)$$

Where,

S: number of frugivory events

n: frequency of handling type *i*

C: effect value of handling type *i* according to the gradient C=-1 (predation), C=0.1 (pulp consumption), C=0.3 (transport), C=1 (endozoochory)

Range of fruit size. We estimated the average size (fruit diameter) of fruits of the plant species consumed by frugivores, based on a sample of ten fruits from each of ten individuals from each species. When it was not possible to sample fruits in the field, fruit size data were obtained from literature, after which we estimated the range of fruit size by means of the coefficient of variation (CV) of average fruit size among the plant species consumed by each frugivore species.

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Appendix S4. Principal Component Analysis on species-level network metrics.

The Principal Component Analysis (PCA) on the four species-level network metrics for frugivores (degree, c and z, contribution to nestedness) showed that the first axis (PC1) accounted for 70% of variance in Guacamayas, and 56% in El Pino (Table S4.1). The loads of each species-level network metrics in PC1 in both networks varied between 0.40 and 0.62 (Table S4.2).

Table S4.1. Percentage of variance explained by the first three PCA axes in each locality.

Locality	PCA	PC1	PC2	PC3
Guacamayas	Standard Deviation	1.67	0.89	0.52
	Prop. Variance	0.70	0.20	0.06
	Cum. Proportion	0.70	0.90	0.97
El Pino	Standard Deviation	1.47	0.97	0.77
	Prop. Variance	0.56	0.24	0.15
	Cum. Proportion	0.56	0.80	0.95

Table S4.2. Loads of species-level network metrics (degree, c and z, contribution to nestedness) on PCA axes for each locality

Locality	PCA	PC1	PC2	PC3
Guacamayas	Degree	0.574	0.101	0.179
	c	0.421	-0.745	0.419
	z	0.454	0.655	-0.491
	Contribution to nestedness	0.536	-	-0.818
El Pino	Degree	0.628	-	0.127
	c	0.402	0.733	0.387
	z	0.430	-0.681	0.415

Contribution to nestedness	0.508	-	-0.814

Appendix S5. Detection and analytical management of outliers

In the network of El Pino, the species *Dasyprocta punctata* had a high SDP despite having a very low CNS, diverging greatly from the general trend of SDP being directly proportional to CNS. After analyzing the linear relationship between SDP and CNS for this locality, the data for *D. punctata* was considered to be an analytical outlier according to the *Bonferroni Outlier Test* implemented in the *car* package in R (Fox & Weisberg, 2011) (*D. punctata* rstudent = 6.54, Bonferonni p <0.001; Figure S5.1).

Figure S5.1. Studentized residuals of a linear regression model between SDP (variable response) and CNS (predictor) in the El Pino locality. The red dot, indicating *D. punctata*, is considered an outlier value according to *Bonferroni* test. The red line represents the critical distance, set at 3 standard deviations.
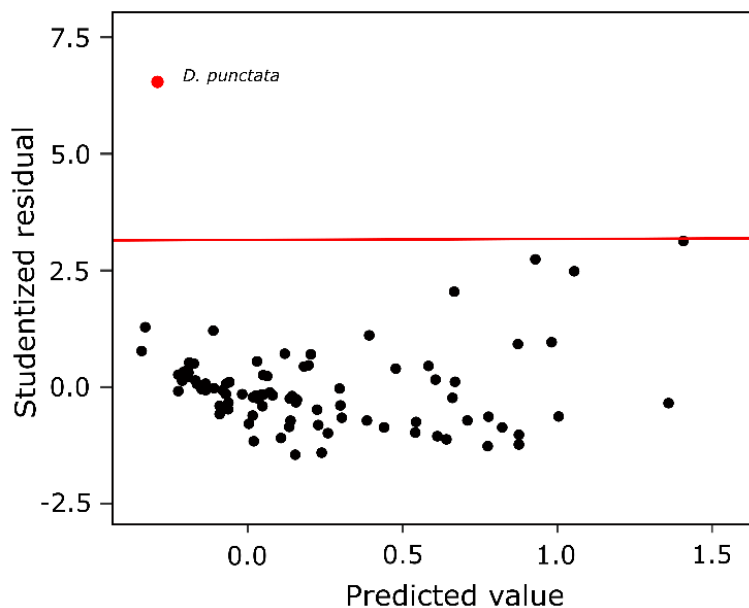
A generalized linear mixed model (GLMM) that considered the dataset of both localities, along with data for *D. punctata* in El Pino, showed a positive and significant effect of CNS on SDP (Table S5.2). GLMM also showed a significant interaction between CNS and locality (Table S5.1), which indicates a difference in the slope of SDP-CNS relationship between localities, mostly due to the inclusion of

D. punctata in El Pino. Independent GLMM models for each locality also indicated positive and significant effects of CNS on SDP in both localities (Table S5.2).

Table S5.1. Generalized linear mixed model verifying the effect of CNS and locality (fixed effects) on SDP, including *D. punctata*. The model included taxonomical identity (nested categories: family, genus, and species) as a random effect. Marginal and conditional (in parentheses) R^2 values are shown.

SDP $R^2 = 0.244$ (0.645)				
Fixed effects	**Estimate**	**SE**	**t-Value**	**p-Value**
CNS	0.288	0.049	5.812	0.000
Locality	-0.013	0.103	-0.127	0.899
CNS: Locality	-0.166	0.071	-2.343	0.026
Random effects	**Variance**	**SD**		
Species[Genus[Family]]	0.334	0.451		

Table S5.2. Generalized linear mixed models verifying the effect of CNS on SDP for each locality, including *D. punctata*. The model included taxonomical identity (nested categories: family, genus) as a random effect. Marginal and conditional (in parentheses) R^2 values are shown.

| SDP				
Guacamayas $R^2 = 0.475$ (0.602)				
Fixed effects	**Estimate**	**SE**	**t-Value**	**p-Value**
CNS	0.319	0.045	6.944	0.000
Random effects				
Genus[Family]	2.251e-5	0.495		
El Pino $R^2 = 0.112$ (0.830)				
Fixed effects				

CNS	0.217	0.062	3.496	0.008
Random effects	**Variance**	**SD**		
Genus[Family]	0.312	0.393		

References

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Appendix S6. Relationship between frugivore characteristics and their contribution to network structure

Figure S6. Relationships between abundance, and morphological and behavioral characteristics of frugivores, and CNS, for Guacamayas (A) and El Pino (B) localities. Each graph shows the value of Spearman correlation coefficient and its significance level (*: p ≤ 0.05; **: p ≤0.01; *** p ≤ 0.001). For body mass, a quadratic model was fitted to the data (global significance is quoted).
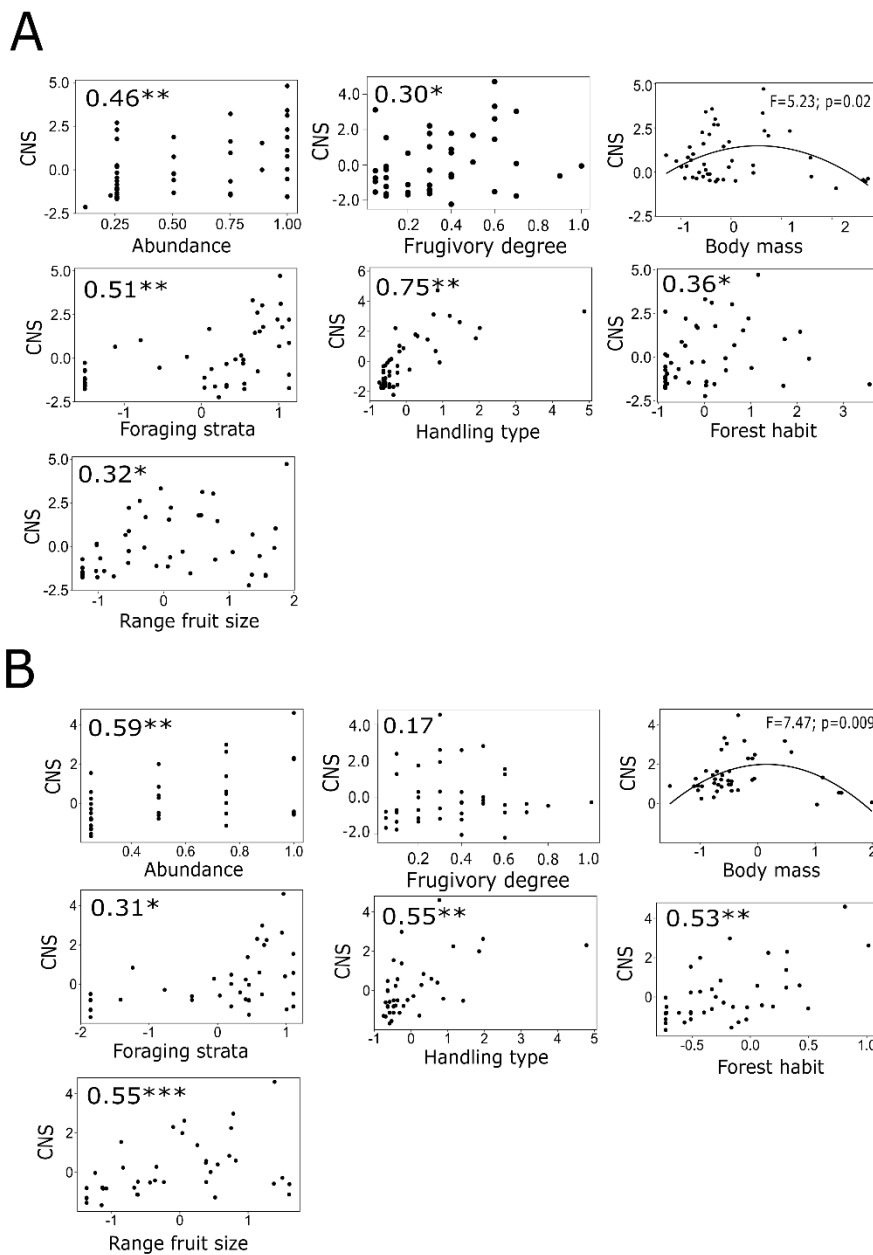
Appendix S7. Correlations between frugivore characteristics

Figure S7. Correlation matrix (Spearman correlation coefficient) among frugivore characteristics for Guacamayas (A) and El Pino (B) localities. Each graph shows the correlation coefficient and its significance (*: p ≤ 0.05; **: p ≤0.01; *** p ≤ 0.001).