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RESEARCH ARTICLE

Variation in Dietary Breadth Among Groups of Black Howler Monkeys is Not Associated With the Vegetation Attributes of Forest fragments

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Habitat disturbance alters vegetation structure and composition. For example, in forest fragments, the rate of secondary plant species recruitment and mortality in species typical of old-growth forests are higher. For many arboreal primates, movement between fragments is infrequent and difficult, thus the dietary breadth of herbivorous primates that live in fragments is expected to change. It is likely that the ability of howler monkeys (*Alouatta* spp.) to live in a large array of habitat types is related to their ability to exploit a broad set of both difficult to digest and high energy resources. However, if small fragments have fewer trees and plant species, food selection by howler monkeys could be limited, which would undermine their persistence. To address this question, we compared the dietary breadth of 14 groups of Yucatán black howler monkeys (*Alouatta pigra*) living in different fragments, and hypothesized that dietary breadth should be associated to the vegetation attributes of the habitat. We characterized the vegetation structure and composition in each fragment and collected a total of 3,747 focal hr on the feeding behavior of 60 adult individuals. Dietary diversity, both in terms of the rate of plant species used as food sources (plant species used per unit of time) and percentage of ingested food from the top five plant species with overall highest ingestion rate, was not related to vegetation attributes but rather associated with the degree of folivory, such that higher folivory led to more diverse diets. Groups living in fragments with higher tree density used a larger number of trees as food sources. Therefore, black howler monkeys living in small fragments with disturbed vegetation continued to preserve diet diversity, confirming that dietary diversification is an important goal in the foraging strategy of howler monkeys. Am. J. Primatol. © 2014 Wiley Periodicals, Inc.

Key words: ***Alouatta*; diet; folivory; habitat disturbance; forest fragment size; Mexico**

INTRODUCTION

The availability and quality of plants vary with environmental conditions. Therefore, foods selected at any particular time and place represent a subset of those that a forager can potentially digest and metabolize [Freeland & Janzen, 1974]. As a consequence, in addition to the physiological or morphological adaptations for consuming a certain array of foods, the ability of foragers to exploit a particular plant assemblage to meet their nutritional requirements is affected, among other things, by climate [e.g., Minder, 2012], plant phenology [e.g., van Schaik et al., 1993], and intra- and inter-specific competition [e.g., Ganzhorn, 1993; Hoeck, 1989]. Barriers to movement into neighboring microhabitats also may restrict the number and type of plant species and plant parts selected by foragers, affecting their dietary breadth [e.g., Erftemeier et al., 1993], which we define as the set of plants consumed over some extended period of time.

Many tree species disappear from small forest fragments (hereafter fragments), particularly those that are emergent and/or shade tolerant [Laurance et al., 2006; Tabarelli et al., 2012], resulting in lower plant species richness in fragments compared to more extensive forests [Hill & Curran, 2003; Laurance et al., 2002]. In the Neotropics, many of these species represent top food species for primates (e.g., *Brosimum alicastrum*, *Poulsenia armata*, *Pouteria* spp., *Dialium guianense*, *Nectandra* spp.), and hence, food

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availability can be lower in smaller fragments. For instance, *B. alicastrum*, which is a key food species for mantled howler monkeys [*Alouatta palliata*: Chapman, 1988] and Geoffroyi's spider monkeys [*Ateles geoffroyi*: Ramos-Fernández & Ayala-Orozco, 2003], is less common in forest fragments than in continuous forests [Arroyo-Rodríguez & Mandujano, 2006; Chaves et al., 2012]. In addition, moving among fragments separated by extensive open areas is infrequent and often dangerous for arboreal primates [e.g., Gilbert & Setz, 2001], which may further reduce access to food resources by individuals inhabiting isolated fragments. As a consequence, in fragments primates may be forced to change their dietary breadth.

Changes in dietary breadth in response to forest disturbance (i.e., loss, fragmentation, or modification: Cowlishaw & Dunbar, 2000) have been reported for a few primate species. For instance, in Sulawesi Tonkean macaques (*Macaca tonkeana*), diademed sifakas (*Propithecus diadema*) and Geoffroyi's spider monkeys, the diversity of plant species in the diet is higher among individuals that live in undisturbed or extensive forests than in individuals that live in smaller and fragmented forests [Chaves et al., 2012; Irwin, 2008; Riley, 2007]. Similarly, the number of plant species in the diet of several groups of black howler monkeys (*Alouatta pigra*) decreased dramatically after severe habitat disturbance caused by a hurricane [Behie & Pavelka, 2005]. Furthermore, in Sulawesi Tonkean macaques, diademed sifakas, and Geoffroyi's spider monkeys, there is evidence that habitat disturbance is associated with increased narrowing of dietary breadth [i.e., a higher proportion of the diet is concentrated in a reduced number of plant species; Chaves et al., 2012; Irwin, 2008; Riley, 2007]. In contrast, other studies have found no clear effects of changes in habitat, including fragment size and connectivity, on dietary breadth in terms of the number of consumed plant species [e.g., bearded saki monkeys, *Chiropotes chiropotes*: Boyle et al., 2012; golden-crowned sifakas, *Propithecus tattersalli*: Quéméré et al., 2013]. Still, even in these cases habitat disturbance affects the patterning of plant species use through the consumption of plants that are ignored in continuous forests, including the exploitation of cultivated plant species. This suggests that primates modify aspects of their diet and food selection in response to changes in their environment, which may enable them to exploit the set of plant species and food types available in fragments, improving their probablities of persisting in disturbed habitats.

Howler monkeys (*Alouatta* spp.) are arboreal quadrupeds that inhabit nearly all Neotropical forest types from southeastern Mexico to northern Argentina. Their ability to live in a large array of habitat types, including highly disturbed forests (e.g., small fragments, orchards), has been related to high dietary flexibility in terms of the plant parts used as food sources (e.g., leaves, fruits, flowers), the relative proportions of plant parts in the diet (i.e., from high folivory to high frugivory) and in the plant species that are consumed [e.g., Bicca-Marques, 2003; Cristóbal-Azkarate & Arroyo-Rodríguez, 2007; Crockett, 1998; Dias & Rangel-Negrín, in press]. Nevertheless, there is evidence suggesting that, as for other primate foragers [Freeland & Janzen, 1974; Westoby, 1978], a major goal of the foraging strategy of howler monkeys is nutrient balancing, which may serve to either obtain a complementary range of nutrients required for growth, maintenance, and reproduction or limit the amount of toxins ingested from any one plant species [e.g., Amato & Garber, in press; Behie & Pavelka, 2012a,b; Felton et al., 2009; Glander, 1978; Milton, 1980]. In this sense, if small fragments have fewer trees and plant species, limitations in the ability of howler monkeys to balance nutrients effectively could affect their survival.

To date, the study of the effects of habitat disturbance on the feeding ecology of howler monkeys has followed two main approaches: (1) case studies comparing the diet of a small number of groups (two to five groups) that live under varying habitat conditions (e.g., fragment size, plant species composition) [e.g., *A. palliata*: Asensio et al., 2007; *A. pigra*: Rivera & Calmé, 2006; *A. seniculus*: López et al., 2005], and (2) reviews of published studies that report both feeding behavior and habitat attributes that are related to disturbance, such as fragment size [*Alouatta* spp.: Bicca-Marques, 2003; Dias & Rangel-Negrín, in press; *A. guariba clamitans*: Chaves & Bicca-Marques, 2013; *A. palliata*: Cristóbal-Azkarate & Arroyo-Rodríguez, 2007]. All review studies have found that some features of the feeding behavior of howler monkeys do not vary as a function of fragment size, such as the relative proportion of leaves and fruits in the diet, whereas other features are consistently affected by attributes of fragmented populations (e.g., fragment size or population density), such as the number of plant species in the diet, which increases with increasing fragment size [e.g., Dias & Rangel-Negrín, in press; Cristóbal-Azkarate & Arroyo-Rodríguez, 2007]. Evidence from case studies is inconsistent, with some cases supporting positive effects of fragment size on time spent eating fruits or the richness of plant species composing the diet [*A. macconnelli*: López et al., 2005; *A. palliata*: Dunn et al., 2010; Juan et al., 2000], whereas others do not [*A. palliata*: Asensio et al., 2007; *A. pigra*: Rivera & Calmé, 2006].

Inconsistencies among case studies, as well as between case studies and review studies, could be related to multiple factors, including inter-specific differences in behavior or variation in methodology and sample size. Furthermore, in these studies, the effects of habitat disturbance on the feeding behavior of howler monkeys were assessed independently from variation in vegetation attributes (e.g., plant species composition, forest structure). As fragment size does not always correlate positively with food availability [e.g., Gómez-Espinosa et al., 2014], the consequences of habitat disturbance on the feeding behavior of howler monkeys should be evaluated through methods that allow greater discrimination of the effects of habitat spatial patterns (i.e., fragment size) from effects of variation in vegetation per se [e.g., plant species diversity: Arroyo-Rodríguez & Dias, 2010].

To address this limitation and improve our understanding of the dietary responses of howler monkeys to habitat disturbance, we investigated the dietary breadth of 14 groups of Yucatan black howler monkeys (*A. pigra*; hereafter, black howler monkeys) living in different fragments. We hypothesized that dietary breadth should be associated with the vegetation attributes of the habitat. We started by addressing the relationship between fragment size and vegetation attributes. Specifically, we predicted that fragment size would be positively associated with tree size and density, plant species diversity, and the presence of plant species typical of old-growth forests. We subsequently predicted that dietary breadth should be positively affected by tree size and density, plant species diversity, and the presence of primary plant species, so when living under these circumstances black howler monkeys should use more plant species and trees as food sources, and concentrate less of their feeding time on top food species (i.e., lower specialization). Because the number of plant species in the diet of howler monkeys increases with the degree of folivory [Dias & Rangel-Negrín, in press], the amount of leaves in the diet was considered an additional predictor of dietary breadth. Finally, as a consequence of changes in vegetation composition that occur in smaller fragments, we predicted an increase in the use of non-primary plant species in small fragments.

METHODS

This study complied with the legal requirements of the Estados Unidos Mexicanos (SEMARNAT SGPA/DGVS/01273/06 and 04949/07). The research also adhered to the American Society of Primatologists (ASP) Principles for the Ethical Treatment of Non-Human Primates.

Study Sites and Subjects

To reduce the potential confounding effects of variation in latitude and altitude on vegetation structure and composition, and as a consequence, on the feeding behavior of black howler monkeys living in different fragments, our study concentrated on populations that live in a ca. 20,000 km^2 area in the Campeche state, located in the Yucatan Peninsula, Mexico (Fig. 1). In Campeche, the climate is hot and humid [Vidal-Zepeda, 2005], and mean annual rainfall is 1,300 mm, with a dry season from November to May (43.7 ± 25.8 mm), and a rainy period between June and October (218.9 ± 14.1 mm). Mean annual temperature is 26°C.

From April 2005 to November 2008, we studied 14 groups of black howler monkeys that lived in

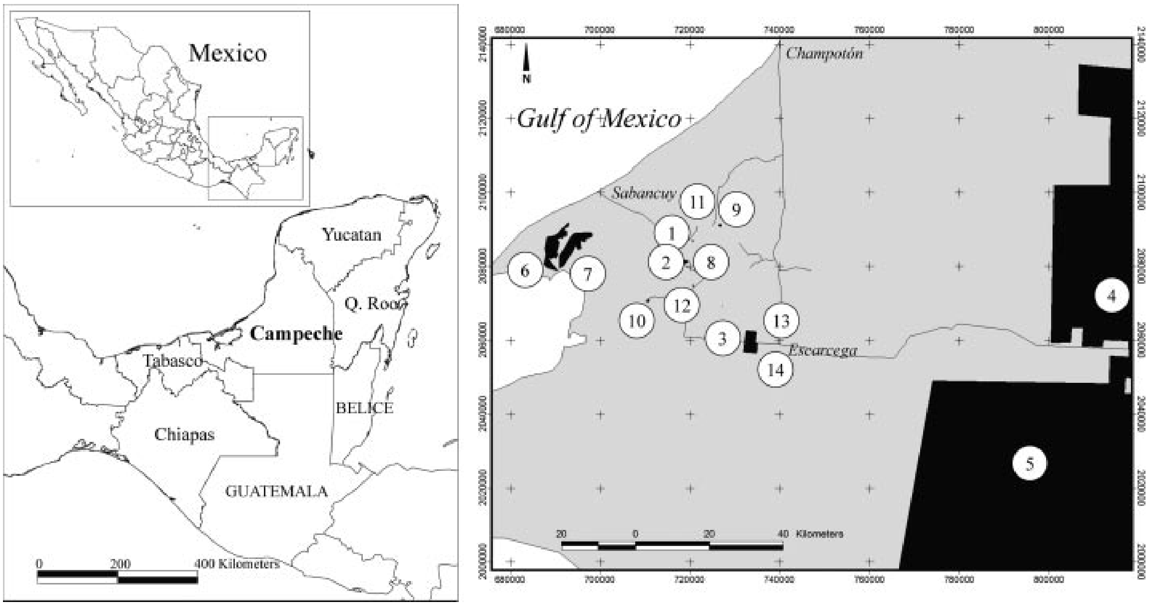

Fig. 1. Location of the 14 groups of black howler monkeys studied in the state of Campeche, Mexico. Groups: 1 = AA Álamo; 2 = Atascadero; 3 = Calakmul S; 4 = Calakmul N; 5 = Calaxchil; 6 = Chicbul; 7 = Chilar; 8 = Manantiales; 9 = Oxcabal; 10 = R Álamo; 11 = Subestación; 12 = T61 Calax; 13 = Tormento N; 14 = Tormento S.

different fragments (Table I). The selection of the study groups aimed to maximize differences in fragment size.

Habitat Characterization

To study the structure and composition of the vegetation in each fragment we used a modification of the Gentry [1982] protocol. Briefly, in each fragment we randomly located ten 50 × 2 m linear transects inside each group's home range. In each transect, we measured the diameter at breast height (DBH; ca. 1.3 m) of all trees with DBH $\geq$10 cm, which we identified at the species level based on morphology and classified according to its ecological group. Ecological groups refer to the light requirements of seeds for germination: primary species (shade tolerant), secondary species (shade intolerant), and non-secondary light demanding species (NSLD) [Arroyo-Rodríguez & Mandujano, 2006]. Secondary species grow only in clearings and at the forest edge. NSLD species can survive in primary or secondary forest, but they need intense light during the first stage of growth. This classification was based on information published in Etnoflora Yucatanense, Flora of Veracruz, and Neotropical Flora, as well as several species lists and consults with botanists [e.g., Ibarra-Manríquez et al., 1997; Popma et al., 1988]. Plants that could not be identified in the field were collected for identification at the "Alfredo Barrera Marín" (UADY, Yucatan, Mexico) and UCAM (Centro de Investigaciones Históricas y Sociales, UAC, Campeche, Mexico) herbaria.

We assessed the completeness of our vegetation samplings by calculating the proportion of observed plant species in each fragment with respect to the expected asymptotes of rarefaction curves. The expected number of plant species was calculated with the Clench equation [Soberón & Llorente, 1993]. We pooled the transect data for each fragment and treated each fragment as a unit for all vegetation analyses.

From transect data, we calculated species richness, density, and basal area for all sampled trees (Table II). We also calculated the proportion of primary tree species according to its ecological group. Total basal area and tree density are measures of vegetation structure, whereas the proportion of primary plant species and species richness are measures of vegetation composition. These measures have been frequently used to describe of the vegetation attributes of howler monkey habitats [e.g., Arroyo-Rodríguez & Mandujano, 2006; Dunn et al., 2009]. In the context of our prediction, smaller fragments should have lower species richness, tree density, basal area, and proportion of primary plant species.

Plant Species Used as food Sources

Each marked plant that was used as a feeding source was identified at the species level. During field identification we also noted the life growth form of each plant as epiphyte, shrub, tree or vine (both woody and non-woody vines), as well as the ecological group of each plant, as described above.

Behavioral Observations

We used focal animal sampling with continuous recording [1-hr samples; Altmann, 1974] to study feeding behavior. Samples in which focal animals were out of sight for more than 5 min were not used in our analyses. When focal animals fed, we noted the duration of the feeding episode and the plant part

TABLE I. Group Size and Composition, Fragment Size, and Sampling Effort for the 14 Groups of Black Howler Monkeys That Were Studied in Campeche

Group no.	Site	Group size[a]	Males	Females	Immatures[b]	Fragment size (ha)	Sampling effort (hr)
1	AA Álamo	4 (3)	2 (1)	2	1	35.3	270
2	Atascadero	4	2	1	1	1.15	250
3	Calakmul N	5	2	2	1	51,503	270
4	Calakmul S	9 (5)	3 (1)	2 (1)	4 (3)	140,000	257
5	Calaxchil	6	1	2	3	3,000	280
6	Chicbul	4 (5)	1	2	1 (2)	5	280
7	Chilar	8	2	3	3	2.1	270
8	Manantiales	3 (5)	1	1 (2)	1 (2)	50	270
9	Oxcabal	5 (7)	2 (3)	2	1 (2)	7	270
10	R Álamo	8	1	4	3	96	250
11	Subestación	5	2	2	1	6	250
12	T61Calax	9 (10)	2	3	4 (5)	300	270
13	Tormento N	10 (9)	3	4	3 (2)	600	280
14	Tormento S	6 (7)	2	2	2 (3)	800	280

Note: [a]Numbers in parenthesis represent changes in group composition between sampling periods.
[b]<30 months of age.

TABLE II. Variables Analyzed in This Study

Variable	Definition
Habitat variables	
Fragment size	Area (in ha) of each forest fragment (min = 1.2; max = 140,000; mean (±SD) = 14,029.0 ± 38,737.1 ha)
Total basal area of trees	Sum of basal areas of all trees with DBH >10 cm sampled in the Gentry transects (min = 2.8; max = 47.1; mean (±SD) = 9.6 ± 11.5 m^2)
Tree density	Number of trees sampled in the Gentry transects multiplied by 0.1 ha (min = 140; max = 1,120; mean (±SD) = 737.9 ± 267.2 trees/ha)
Plant species diversity	Species richness, as the total number of plant species sampled in the Gentry transects (min = 5; max = 33; mean (±SD) = 20.7 ± 7.8)
% Primary plant species	Percentage of plant species sampled in the Gentry transects classified as primary according to light requirements during seed germination (min = 14.3; max = 76; mean (±SD) = 46.3 ± 19.5%)
Dietary variables	
Folivory	Percentage of consumed food weight dedicated to eat leaves (min = 7.9; max = 86.9; mean (±SD) = 39.5 ± 24.1%)
Rate of plant species use	Total number of plant species used as food sources divided by total observation time per group (min = 0.13; max = 0.83; mean (±SD) = 0.49 ± 0.18 species/hr)
% Ingested food from top food species	Percentage of ingested food from the five food plant species with overall higher ingestion (min = 47.8; max = 98.9; mean (±SD) = 71.3 ± 14.9%)
Use of feeding trees (rate)	Number of trees used as food sources divided by total observation time per group (min = 0.07; max = 1.1; mean (±SD) = 0.69 ± 0.30 trees/hr)

that was consumed as young leaves, mature leaves, ripe fruits, unripe fruits, or flowers. Time spent eating each plant part was then multiplied by feeding rates reported for black howler monkey foods [Amato & Garber, in press] to obtain an estimation of the amount of ingested food (in grams) per plant part. From these data, we calculated percentages of ingested food weight per plant part. As mean percentage of mature leaf ingestion was only 3.1 ± 2.6% and four groups were not observed to ingest this plant item at all, we combined both young and mature leaves into one variable (folivory) for further dietary breadth analyses.

Observations were performed during complete days (i.e., 6:00 to 17:00 or 7:00 to 18:00, depending on daylight across the year), and all individuals were identified based on their natural anatomical and physiognomic characteristics, such as body size and proportions, scars, broken fingers, and genital morphology and pigmentation. Focal animals were selected on a pseudorandom basis, such that no individual was sampled twice until all were sampled once, and focal samples of each animal were evenly distributed throughout the day. We sampled only adult individuals ($N = 60$; 33 females and 27 males) and collected a total of 3,747 focal hr of behavioral data, with a mean observation time of 62.5 ± 19.8 hr per individual and 267.6 ± 11.4 hr per group (Table I). Each group was sampled during approximately 30 days in each season. Inter-annual variation in rainfall among sampling months during the 4 years that spanned our study was low for both the dry (coefficient of variation = 5%) and the wet seasons (coefficient of variation = 9%) (data from CONAGUA-SMN, Mexican National Water Committee-National Weather Service). Therefore, little annual variation in plant part production due to differences in climatic variables is expected to have occurred during the study.

Data Organization and Analyses

We used three measures of dietary breadth: (1) number of plant species used as food sources; (2) proportion of ingested food from top food species; (3) number of trees used as feeding trees (Table II). To account for variation among groups in sampling time, the number of plant species and trees used as food sources were converted to rates. Top food species was defined as the five plant species with overall highest ingestion for each group. In the context of our predictions, dietary breadth should decrease in fragments with lower species richness, tree density, basal area, and proportion of primary plant species. Therefore, the rates of plant species and tree use should decrease, whereas the proportion of food ingested from top food species should increase under those circumstances.

All analyses were performed at the fragment (fragment size and vegetation variables) and group (behavioral data) level ($N = 14$). We analyzed the relationship between fragment size and each vegetation variable with simple regression analyses, and explored the relationship between fragment size and folivory with a Pearson correlation. We used a simple regression analysis to test for the effect of the proportion of primary plant species in the habitat on the consumption of primary plant species.

Because vegetation variables measured in transects were correlated, we ran a principal components analysis (PCA) to reduce collinearity among predictor variables in models of dietary breadth. To model variation in dietary breadth we used regression analyses with Akaike's information criterion [AIC_c; Burnham & Anderson, 2010] to determine which predictive variables best explained variation among groups in the rate of plant species use, percentage of food ingested from top food species, and the rate of food tree use (Table II). Proportional data were normalized using the arcsine square root transformation, whereas fragment size was normalized using a logarithmic transformation. All tests were two-tailed and were performed with SPSS 20.0 (SPSS, Inc., Chicago, IL). Significance was set at $P < 0.05$.

RESULTS

Completeness of Vegetation Samplings

The mean proportion of plant species recorded in vegetation transects with respect to the estimated total number of species in each fragment was $73.80 \pm 0.03\%$, indicating that we sampled approximately two-thirds of all plant species present in fragments (Fig. 2). Variation among fragments in the proportion of plant species recorded was very low (coefficient of variation = 4%), suggesting that our method had a similar performance across forest fragments.

Effects of Fragment Size on Vegetation Attributes

Larger fragments had a higher basal area of trees ($R^2 = 0.78$, $F_{1,12} = 42.5$, $P < 0.001$; Fig. 3a), a more diverse plant species assemblage ($R^2 = 0.67$, $F_{1,12} = 24.4$, $P < 0.001$; Fig. 3b) and a higher proportion of primary plant species ($R^2 = 0.71$, $F_{1,12} = 29.2$, $P < 0.001$; Fig. 3c), whereas tree density was not related to fragment size ($R^2 = 0.007$, $F_{1,12} = 0.09$, $P = 0.763$; Fig. 3d). The proportion of consumed leaves was negatively related to habitat size ($r = -0.72$, $P = 0.004$).

A PCA of vegetation variables resulted in two components with eigenvalues ≥1, which explained 89% of the total variance. Component 1 (eigenvalue = 2.7, explained 67.5% of the variance) had strong, positive loadings for plant species richness (0.91), basal area of trees (0.87) and the proportion of primary plant species (0.92). Component 2 (eigenvalue = 1, explained 21.5% of the variance) had a strong, positive loading for tree density (0.84). These components were used as predictive variables (in addition to folivory) in models of dietary breadth.

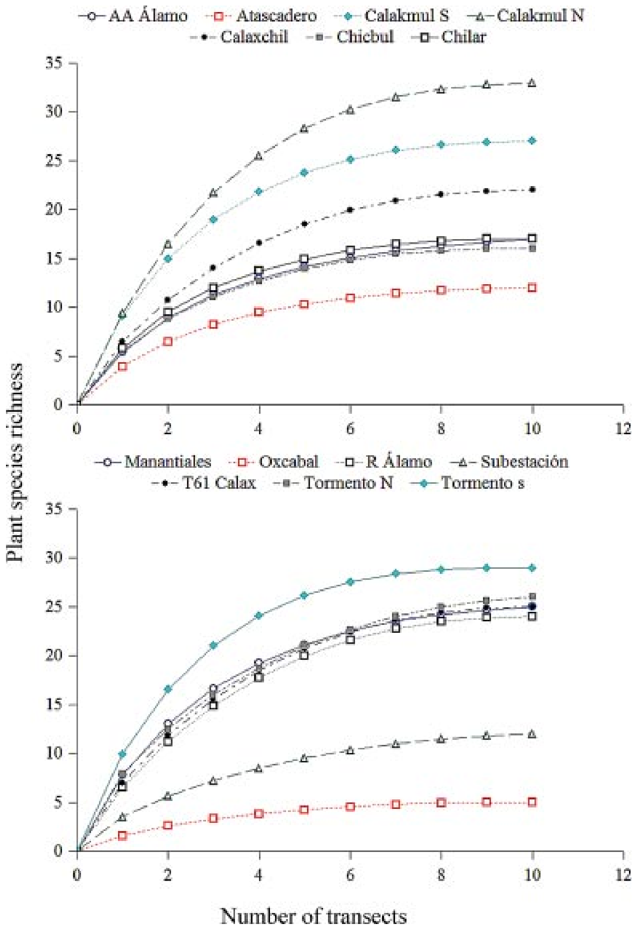

Fig. 2. Transect-based plant species accumulation curves for 14 forest fragments occupied by black howler monkeys in Campeche, Mexico. Species are represented as the mean number of new species observed for each transect accumulated up to the total sample size (i.e., 10 transects).

Dietary Breadth of Black Howlers in Fragments

Black howler monkeys were observed to consume 136 plant species and 28 morphospecies (Table SI). The majority of plants used were trees (67.1%), followed by vines (14.0%), lianas (7.3%), shrubs (6.7%), epiphytes (3.0%), and hemiparasites (1.8%). Of plants that could be identified at the species level, 38.2% were NSLD, 37.5% were primary, and 24.3% were secondary species. While no species was consumed by all groups, one species (*Bursera simaruba* (L.) Sarg.) was used by 12 groups, whereas 15 species were used by a single group. Thirty-one taxa (29 species and 2 morphospecies) were classified as top food species across the 14 groups (Table III), the majority of which (90.3%) were tree species. According to their ecological group, 44.8% were primary, 31.0% were NSLD, and 24.1% were secondary species. Two taxa (*B. alicastrum* Sw., *Manilkara*

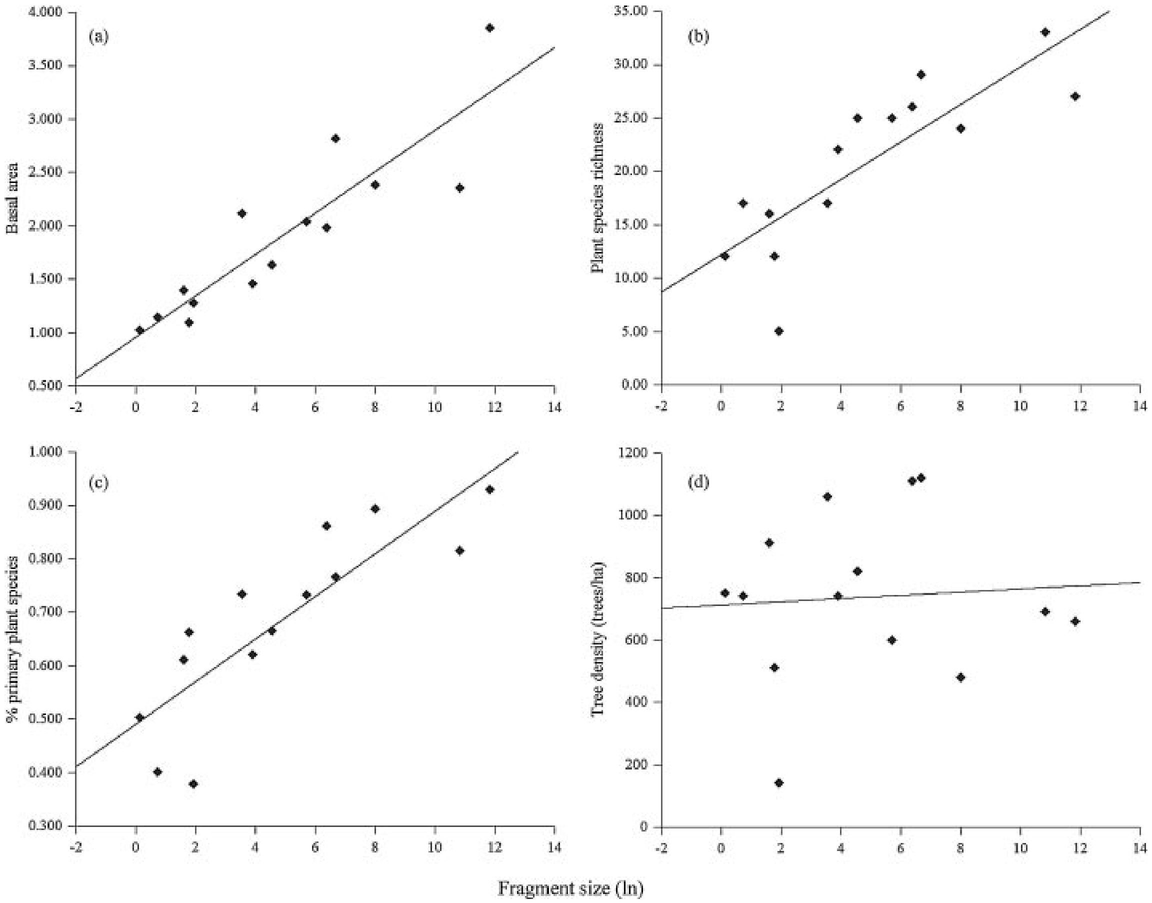

Fig. 3. Variation in vegetation composition and structure as a function of fragment size: (**a**) total basal area of trees; (**b**) plant species richness; (**c**) proportion of primary plant species; (**d**) tree density.

zapota (L.) P. Royen) were top food species for six groups, whereas 48.4% were top food species for a single group.

Effects of Vegetation Attributes on Dietary Breadth

The rate of plant species use was positively predicted by folivory (Table IV; Fig. 4a), whereas groups that consumed fewer leaves ingested more food from top food species (Fig. 4b). Groups that lived in habitats with a higher density of trees used more feeding trees (Fig. 4c). The proportion of primary plant species in the habitat positively predicted the consumption of primary plant species ($R^2_{1,12} = 0.37$, $P = 0.022$).

DISCUSSION

Our study demonstrates that, although in Campeche plant species richness, basal area and the proportion of primary plant species vary as a function of fragment size, the dietary breadth of black howler monkeys is little affected by the size of the fragment occupied. Dietary breadth, both in terms of the rate of plant species used as food sources and consumption of food from top food species, was not related to vegetation attributes of the habitat, but rather associated with feeding behavior, such that higher folivory led to more diverse diets. The only analyzed dietary variable that related to the vegetation attributes of the habitat was the number of used feeding trees: groups living in fragments with higher tree density used more trees as food sources. These results indicate that black howler monkeys that live in small fragments with disturbed vegetation preserve diet diversity, confirming that dietary diversification is an important goal in the foraging strategy of howler monkeys. Further, howler monkeys diversify their diet not only through the use of different plant species but also by feeding from more trees of each species, as suggested by the positive relationship between tree density and the number of feeding trees.

TABLE III. Proportion of the Diet Dedicated to the Consumption of Top Food Species for 14 Groups of Black Howler Monkeys in Campeche

				Group^b													
Family	Species	Growth form	Ecological group^a	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Apocynaceae	*Aspidosperma megalocarpon* Müll. Arg.	Tree	Primary				4.3										
Anacardiaceae	*Astronium graveolens* Jacq.	Tree	Primary								3.2						
Moraceae	*Brosimum alicastrum* Sw.	Tree	Primary		18.5	36.3	40.1	7.9							3.1		13.5
Burseraceae	*Bursera simaruba* (L.) Sarg.	Tree	NSLD			2.1			8.1		7.5						
Fabaceae	*Enterolobium cyclocarpum* (Jacq.) Griseb.	Tree	NSLD							2.6							
Myrtaceae	*Eugenia capuli* (Schltdl. & Cham.) Hook. & Arn.	Shrub	NSLD		5.3												
Moraceae	*Ficus cotinifolia* Kunth	Tree	Primary			41.1	17.0	17.2							23.1		
Moraceae	*Ficus lundellii* Standl.	Tree	NSLD	5.8						86.1							
Moraceae	*Ficus maxima* Mill.	Tree	Secondary									39.1					
Moraceae	*Ficus ovalis* (Liebm.) Miq.	Tree	NSLD			7.9						10.0					
Moraceae	*Ficus* sp.	Tree	ND							6.0				11.3	6.2		
Sterculiaceae	*Guazuma ulmifolia* Lam.	Tree	Secondary								10.8	10.5					
Fabaceae	*Leucaena leucocephala* (Lam.) de Wit	Tree	Secondary	4.6													
Fabaceae	*Lonchocarpus castilloi* Standl.	Tree	Secondary		18.7								20.5			13.7	
Fabaceae	*Lonchocarpus yucatanensis* Pittier	Tree	NSLD						24.8								
Fabaceae	*Lysiloma acapulcense* (Kunth) Benth.	Tree	Secondary							3.6							
Fabaceae	*Lysiloma latisiliquum* (L.) Benth.	Tree	Primary	6.0					10.0		37.9			9.2		9.8	
Fabaceae	*Lysiloma* sp.	Tree	ND					6.6									
Sapotaceae	*Manilkara zapota* (L.) P. Royen	Tree	Primary		7.1	2.2	27.6	27.1							33.4		13.6
Sapindaceae	*Melicoccus oliviformis* Kunth	Tree	Primary				2.4										
Anacardiaceae	*Metopium brownei* (Jacq.) Urb.	Tree	NSLD														36.7
Euphorbiaceae	*Phyllanthus liebmannianus* Mull. Arg	Tree	Secondary						10.0								
Myrtaceae	*Pimenta dioica* (L.) Merr.	Tree	Primary													9.1	
Fabaceae	*Piscidia piscipula* (L.) Sarg.	Tree	Primary							0.7		8.5	8.9	27.1			
Fabaceae	*Platymiscium yucatanum* Standl.	Tree	Primary					12.8			3.4				4.5		
Sapotaceae	*Pouteria campechiana* (Kunth) Baehni	Tree	Primary													7.0	
Simaroubaceae	*Simarouba glauca* DC.	Tree	Primary						6.2					14.8			6.4
Fam. 4	Sp. 4	Vine	ND									8.9					
Anacardiaceae	*Spondias radlkoferi* Donn. Sm.	Tree	NSLD										11.8				
Fabaceae	*Swartzia cubensis* (Britton & P. Wilson) Standl.	Tree	Primary	5.5													
Combretaceae	*Terminalia buceras* L.	Tree	Primary													7.0	
Moraceae	*Trophis racemosa* (L.) Urb.	Tree	NSLD	25.9									14.1				
Verbenaceae	*Vitex gaumeri* Greenm.	Tree	Secondary		25.7									9.2			7.4
Flacourtiaceae	*Zuelania guidonia* (Sw.) Britton & Millsp.	Tree	NSLD										3.3				

Note: [a]Not determined.
[b]Groups: 1 = AA Álamo; 2 = Atascadero; 3 = Calakmul S; 4 = Calakmul N; 5 = Calaxchil; 6 = Chicbul; 7 = Chilar; 8 = Manantiales; 9 = Oxcabal; 10 = R Álamo; 11 = Subestación; 12 = T61 Calax; 13 = Tormento N; 14 = Tomento S.

TABLE IV. Models of Dietary Breadth for Black Howler Monkeys

Dietary breadth	Model[a]	β	B	t	P
Rate of species use ($R^2 = 0.58$; $F_{2,11} = 7.7$; $P = 0.008$)	Leaf consumption	0.64	0.17	3.7	0.003
	Component 1	0.03	0.02	1.5	0.161
% Feeding from top food species ($R^2 = 0.78$; $F_{1,12} = 42.1$; $P < 0.001$)	Leaf consumption	−0.88	0.13	6.5	<0.001
Rate of tree use ($R^2 = 0.51$; $F_{2,11} = 5.8$; $P = 0.019$)	Component 2	0.24	0.07	3.3	0.007
	Leaf consumption	0.07	0.32	0.2	0.824

Note: β = regression coefficient; B = parameter.
[a] Variables included in models were selected by AIC_c.

Habitat Size, Vegetation Attributes, and the Dietary Breadth of Black Howler Monkeys

As a probable consequence of the significant effect of habitat size on vegetation composition, dietary composition differed greatly among groups (Table SI). However, although in small fragments there were fewer primary plant species, diversity in plant species use and specialization did not vary according to vegetation attributes. Howler monkeys living in fragments are probably maintaining dietary diversity through the exploitation of plant species that are either absent or are ignored in larger forests. For instance, in small fragments, individuals were observed to consume introduced plant species that were not observed in larger fragments (e.g., *Carica papaya* L.). These results confirm the dietary flexibility of black howler monkeys [Rivera & Calmé, 2006; Silver & Marsh, 2003], and converge with previous findings with other primates. For instance, bearded saki monkeys and golden-crowned sifakas do not consistently use more plant species when living in larger habitats, but show differences in the selection of foods that are dependent on plant availability [Boyle et al., 2012; Quéméré et al., 2013], while ursine colobus (*Colobus vellerosus*) are able to adjust species composition in its diet to accommodate the differences in plant composition in the forests they inhabit [Wong et al., 2006].

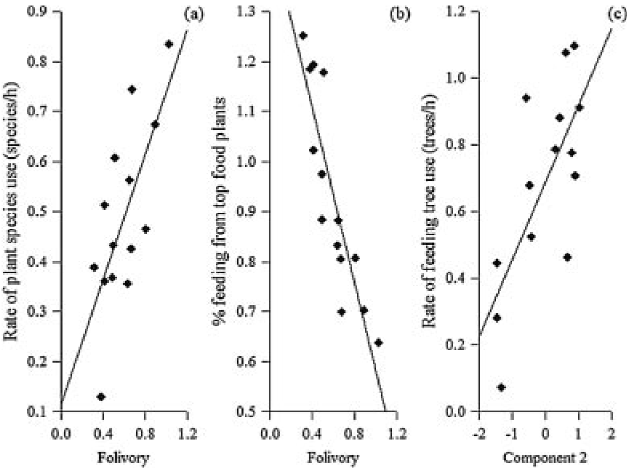

Fig. 4. Variation in dietary breadth of black howler monkeys: (**a**) rate of plant species use as a function of folivory; (**b**) food ingested from top food plant species as a function of folivory; (**c**) rate of feeding tree use as a function of component 2 (i.e., tree density).

The observed patterns of dietary diversification by black howler monkeys agree with previous case studies that compared feeding behavior among a few groups of howler monkeys. For instance, the number of plant species in the diet of three *A. palliata* groups living in contrasting habitats (e.g., fragment size was 1.3, 8.3, and 40 ha) differed by only 14% with respect to the total number of species used, and this difference did not parallel variation in fragment size [Asensio et al., 2007]. In contrast, literature reviews on the relationship between habitat disturbance and the feeding behavior of howler monkeys, which have analyzed dietary diversity, have consistently found negative associations between forest disturbance and the number of plant species used as food sources [Bicca-Marques, 2003; Chaves & Bicca-Marques, 2013; Cristóbal-Azkarate & Arroyo-Rodríguez, 2007]. It is possible that this difference is due to these studies not having standardized species diversity measures according to study duration in hours [Bicca-Marques, 2003; Chaves & Bicca-Marques, 2013; Cristóbal-Azkarate & Arroyo-Rodríguez, 2007]. We therefore recommend that future analyses of the dietary diversity of howler monkeys account for variation in observation effort, as performed in the present study.

Implications for the Survival of Black Howler Monkeys in Forest Fragments

Could the absence of differences among groups in dietary diversity represent good news for the conservation of howler monkeys in forest fragments? We do not believe so. As is the case for other primates that consume a variety of different plant tissues, howler monkeys have to balance the consumption of nutrients while avoiding the toxic effects of secondary metabolites coming from a single plant species [Felton et al., 2009; Freeland & Janzen, 1974; Pulliam, 1975]. This foraging goal can be attained through a mixed diet of different plant species, plant, and animal tissues, and different individual trees from each species [Milton, 1980]. We found that black howler monkeys living in smaller fragments maintain levels of dietary diversity comparable to those of individuals living in larger fragments by using more secondary and NSLD plant species. It has been speculated that non-primary vegetation (secondary and NSLD plant species) may have positive effects on the persistence of folivorous primates [Lovejoy et al., 1986], and particularly howler monkeys, in transformed habitats because: (1) leaves from secondary species may have lower levels of chemical defenses and fiber, and higher values of protein, digestible nutrients, and energy: Behie & Pavelka, 2012a; Chiarello, 2003; Lovejoy et al., 1986]; (2) there is lower seasonal variation in the production of young leaves (a preferred food item for howler monkeys) in secondary forests; and (3) secondary vegetation is on occasion positively associated with floristic diversity, facilitating dietary diversity [Cristóbal-Azkarate et al., 2005]. However, this prediction has not been formally tested as applied to howler monkeys' habitats and foods, and there is abundant data suggesting that howler monkey populations fare better (e.g., higher population size and occupancy of fragments) in mature or undisturbed forests than in disturbed secondary habitats [DeGamma-Blanchet & Fedigan, 2006; Estrada & Coates-Estrada, 1996; Rodríguez-Toledo et al., 2003; Sorensen & Fedigan, 2000].

In small fragments, howler monkeys may be metabolically constrained, because energy intake is supposedly lower due to a reduction in the ingestion of preferred foods, such as fruit, which black howler monkeys consume according to its availability [e.g., Behie & Pavelka, in press; Pavelka & Knopff, 2004; Silver et al., 1998], and folivory increases, leading to higher foraging effort [e.g., higher travel time: Dunn et al., 2009]. In the current study, folivory and the number of used feeding trees increased in smaller habitats, supporting the existence of a link among feeding behavior, dietary breadth, and habitat disturbance. Therefore, future research should aim at determining the fitness consequences of variation in the dietary breadth of howler monkeys, including the assessment of the quality (digestible nutrients) and anti-quality (non-digestible nutrients and toxic components) of the foods that are consumed in relation to landscape structure [*sensu* Turner, 1989]. When such data become available, we will be able to better predict the long-term survival probabilities of black howler monkey populations living in disturbed habitats.

CONCLUSION

We conclude that the dietary breadth of black howler monkeys is little affected by the vegetation attributes of the habitat that were measured in the present study, and is mainly affected by folivory, such that more folivorous diets lead to more diverse diets. Finally, it is noteworthy that 95% of the 136 plants consumed by black howler monkeys that we identified at the species level have not been evaluated in terms of conservation status [IUCN, 2013]. To better understand how black howler monkeys, and primates in general, adjust to the transformation of their habitats, we will have to improve our knowledge on the patterns of occurrence and abundance of the plant species that they use as food sources.

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REFERENCES

Altmann J. 1974. Observational study of behavior: sampling methods. Behaviour 49:227–267.

Amato K, Garber PA. In press. Nutrition and foraging strategies of the black howler monkey (*Alouatta pigra*) in Palenque National Park, Mexico. Am J Primatol doi: 10.1002/ajp.22268

Arroyo-Rodríguez V, Dias PAD. 2010. Effects of habitat fragmentation and disturbance on howler monkeys: a review. Am J Primatol 71:1–16.

Arroyo-Rodríguez V, Mandujano S. 2006. Forest fragmentation modifies habitat quality for *Alouatta palliata*. Int J Primatol 27:1079–1096.

Asensio N, Cristóbal-Azkarate J, Dias PAD, Veà JJ, Rodríguez-Luna E. 2007. Foraging habits of *Alouatta palliata mexicana* in three forest fragments. Folia Primatol 78:141–153.

Behie AM, Pavelka MSM. 2005. The short-term effect of Hurricane Iris on the diet and activity budget of black howlers (*Alouatta pigra*) in Monkey River, Belize. Folia Primatol 76:1–9.

Behie A, Pavelka MSM. 2012a. Food selection in the black howler monkey following habitat disturbance: implications for the importance of mature leaves. J Trop Ecol 28:153–160.

Behie A, Pavelka MSM. 2012b. The role of minerals in food selection in a black howler monkey (*Alouatta pigra*) population in Belize following a major hurricane. Am J Primatol 74:1054–1063.

Behie AM, Pavelka MSM. In press. Fruit as a key factor in howler monkey population density: conservation implications. In: Kowalewski M, Garber PA, Cortés-Ortiz L, Urbani B, Youlatos D, editors. Howler monkeys: examining the evolution, physiology, behavior, ecology and conservation of the most widely distributed Neotropical primate. New York: Springer.

Bicca-Marques JC. 2003. How do howler monkeys cope with habitat fragmentation. In: Marsh LK, editor. Primates in fragments: ecology and conservation. New York: Kluwer. p 283–303.

Boyle SA, Zartman CE, Spironello WR, Smith AT. 2012. Implications of habitat fragmentation on the diet of bearded saki monkeys in central Amazonian forest. J Mammal 93:959–976.

Burnham KP, Anderson DR. 2010. Model selection and multimodel inference: a practical information-theoretic approach. New York: Springer.

Chapman CA. 1988. Patterns of foraging and range use by three species of Neotropical primates. Primates 29:177–194.

Chaves OM, Bicca-Marques JC. 2013. Dietary flexibility of the brown howler monkey throughout its geographic distribution. Am J Primatol 75:16–29.

Chaves OM, Stoner KE, Arroyo-Rodríguez V. 2012. Differences in diet between spider monkey groups living in forest fragments and continuous forest in Mexico. Biotropica 44:105–113.

Chiarello AG. 2003. Primates of the Brazilian Atlantic forest: the influence of forest fragmentation on survival. In: Marsh LK, editor. Primates in fragments: ecology and conservation. New York: Kluwer. p 99–121.

Cowlishaw G, Dunbar RIM. 2000. Primate Conservation Biology. Chicago: The University of Chicago Press.

Cristóbal-Azkarate J, Arroyo-Rodríguez V. 2007. Diet and activity pattern of howler monkeys (*Alouatta palliata*) in Los Tuxtlas, Mexico: effects of habitat fragmentation and implications for conservation. Am J Primatol 69:1013–1029.

Cristóbal-Azkarate J, Veà JJ, Asensio N, Rodríguez-Luna E. 2005. Biogeographical and floristic predictors of the presence and abundance of mantled howler monkeys (*Alouatta palliata mexicana*) in rain forest fragments at Los Tuxtlas, Mexico. Am J Primatol 67:209–222.

Crockett CM. 1998. Conservation biology of the genus *Alouatta*. Int J Primatol 19:549–578.

DeGamma-Blanchet H, Fedigan LM. 2006. The effects of forest fragment age, isolation, size, habitat type, and water availability on monkey density in a tropical dry forest. In: Estrada A, Garber P, Pavelka M, Luecke L, editors. New perspectives in the study of Mesoamerican primates: distribution, ecology, behavior and conservation. New York: Kluwer. p 165–188.

Dias PAD, Rangel-Negrín A. In press. Diets of howler monkeys. In: Kowalewski M, Garber PA, Cortés-Ortiz L, Urbani B, Youlatos D, editors. Howler monkeys: examining the evolution, physiology, behavior, ecology and conservation of the most widely distributed Neotropical primate. New York: Springer.

Dunn JC, Cristóbal-Azkarate J, Veà J. 2009. Differences in diet and activity pattern between two groups of *Alouatta palliata* associated with the availability of big trees and fruit of top food taxa. Am J Primatol 71:654–662.

Dunn JC, Cristóbal-Azkarate J, Veà J. 2010. Seasonal variations in the diet and feeding effort of two groups of howler monkeys in different sized forest fragments. Int J Primatol 31:887–903.

Erftemeier PL, Djunarin A, Moka W. 1993. Stomach content analysis of a dugong (*Dugong dugong*) from Sulawesi, Indonesia. Aust J Mar Freshwater Res 44:229–233.

Estrada A, Coates-Estrada R. 1996. Tropical rain forest fragmentation and wild populations of primates at Los Tuxtlas, Mexico. Int J Primatol 17:759–783.

Felton AM, Felton A, Lindenmayer DB, Foley WJ. 2009. Nutritional goals of wild primates. Funct Ecol 23:70–78.

Freeland WJ, Janzen DH. 1974. Strategies in herbivory by mammals: the role of plant secondary compounds. Am Nat 108:269–289.

Ganzhorn JU. 1993. Flexibility and constraints of *Lepilemur* ecology. In: Kappeler PM, Ganzhorn JU, editors. Lemur social systems and their ecological basis. New York: Plenum Press. p 153–165.

Gentry AH. 1982. Patterns of Neotropical plant species diversity. Evol Biol 15:1–85.

Gilbert KA, Setz EZ. 2001. Primates in a fragmented landscape. Six species in Central Amazonia. In: Bierregaard RO Jr, Gascon C, Lovejoy TE, Mesquita R, editors. Lessons from Amazonia: the ecology and conservation of a fragmented forest. New Haven: Yale University Press. p 262–270.

Glander KE. 1978. Howling monkey feeding behaviour and plant secondary compounds: a study of strategies. In: Montgomery GG, editor. Ecology of arboreal folivores. Washington: Smithsonian Institution Press. p 561–574.

Gómez-Espinosa E, Rangel-Negrín A, Chavira R, Canales-Espinosa D, Dias PAD. 2014. The effect of energetic and psychosocial stressors on glucocorticoids in mantled howlers (*Alouatta palliata*). Am J Primatol. doi: 10.1002/ajp.22240

Hill JL, Curran PJ. 2003. Area, shape and isolation of tropical forest fragments: effects on tree species diversity and implications for conservation. J Biogeogr 30:1391–1403.

Hoeck HN. 1989. Demography and competition in hyrax. A 17 years study. Oecologia 79:353–360.

Ibarra-Manríquez G, Martínez-Ramos M, Dirzo R, Núñez-Farfán J. 1997. La vegetación. In: González-Soriano E, Dirzo R, Vogt RC, editors. Historia Natural de Los Tuxtlas. México, DF: UNAM. p 61–85.

Irwin M. 2008. Feeding ecology of diademed sifakas (*Propithecus diadema*) in forest fragments and continuous forest. Int J Primatol 29:95–115.

IUCN. 2013. Red list of threatened species. Available online at: www.iucnredlist.org (consulted on September 15, 2013).

Juan S, Estrada A, Coates-Estrada R. 2000. Contrastes y similitudes en el uso de recursos y patrón general de actividades en tropas de monos aulladores (*Alouatta palliata*) en fragmentos de selva en Los Tuxtlas, México. Neotrop Primates 8:131–135.

Laurance WF, Lovejoy TE, Vasconcelos HL, et al. 2002. Ecosystem decay of Amazonian forest fragments: a 22-year investigation. Conserv Biol 16:605–618.

Laurance WF, Nascimento HEM, Laurance SG, et al. 2006. Rain forest fragmentation and the proliferation of successional trees. Ecology 87:469–482.

López GO, Terborgh J, Ceballos N. 2005. Food selection by a hyperdense population of red howler monkeys (*Alouatta seniculus*). J Trop Ecol 21:445–450.

Lovejoy TE, Bierregaard RO Jr, Rylands AB, et al. 1986. Edge and other effects of isolation on Amazon forest fragments. In: Soulé ME, editor. Conservation biology: the science of scarcity and diversity. Sunderland: Sinaur. p 257–285.

Milton K. 1980. The foraging strategy of howler monkeys: a study in primate economics. New York: Columbia University Press.

Minder I. 2012. Local and seasonal variations of roe deer diet in relation to food resource availability in a Mediterranean environment. Eur J Wildl Res 58:215–225.

Pavelka MSM, Knopff K. 2004. Diet and activity in black howler monkeys (*Alouatta pigra*) in southern Belize: does degree of frugivory influence activity level? Primates 45:105–111.

Popma J, Bongers F, Martínez-Ramos M, Veneklaas E. 1988. Pioneer species distribution in treefall gaps in Neotropical rain forest; a gap definition and its consequences. J Trop Ecol 4:77–88.

Pulliam HR. 1975. Diet optimization with nutrient constraints. Am Nat 109:765–768.

Quéméré E, Hibert F, Miquel C, et al. 2013. A DNA metabarcoding study of a primate dietary diversity and plasticity across its entire fragmented range. PLoS ONE 8: e58971.

Ramos-Fernández G, Ayala-Orozco B. 2003. Population size and habitat use of spider monkeys in Punta Laguna, Mexico. In: Marsh L, editor. Primates in fragments: ecology and conservation. New York: Kluwer. p 191–209.

Riley EP. 2007. Flexibility in diet and activity patterns of *Macaca tonkeana* in response to anthropogenic habitat alteration. Int J Primatol 28:107–133.

Rivera A, Calmé S. 2006. Forest fragmentation and its effects on the feeding ecology of black howlers (*Alouatta pigra*) from the Calakmul area in Mexico. In: Estrada A, Garber PA, Pavelka MSM, Luecke L, editors. New perspectives in the study of Mesoamerican primates: distribution, ecology, behavior, and conservation. New York: Springer. p 189–213.

Rodríguez-Toledo EM, Mandujano S, García-Orduña F. 2003. Relationship between forest fragments and howler monkeys (*Alouatta palliata mexicana*). In: Marsh LK, editor. Primates in fragments: ecology and conservation. New York: Kluwer. p 79–97.

Silver SC, Marsh LK. 2003. Dietary flexibility, behavioral plasticity, and survival in fragments: lessons from translocated howlers. In: Marsh LK, editor. Primates in fragments: ecology and conservation. New York: Kluwer. p 251–265.

Silver SC, Ostro LET, Yeager CP, Horwich RH. 1998. Feeding ecology of the black howler monkey (*Alouatta pigra*) in northern Belize. Am J Primatol 45:263–279.

Soberón JM, Llorente JB. 1993. The use of species accumulation functions for the prediction of species richness. Conserv Biol 7:480–488.

Sorensen TC, Fedigan LM. 2000. Distribution of three monkey species along a gradient of regenerating tropical dry forest. Biol Conserv 92:227–240.

Tabarelli M, Peres CA, Melo FPL. 2012. The “few winners and many losers” paradigm revisited: emerging prospects for tropical forest biodiversity. Biol Conserv 155:136–140.

Turner MG. 1989. Landscape ecology: the effect of pattern on process. Annu Rev Ecol Evol 20:171–197.

van Schaik CP, Terborgh J, Wright SJ. 1993. The phenology of tropical forests: adaptive significance and consequences for primary consumers. Annu Rev Ecol Evol 23:353–377.

Vidal-Zepeda R. 2005. Las regiones climáticas de México, I. 2.2. México, DF: Instituto de Geografía, UNAM.

Westoby M. 1978. What are the biological bases of varied diets? Am Nat 112:627–631.

Wong SNP, Saj TL, Sicotte P. 2006. Comparison of habitat quality and diet of *Colobus vellerosus* in forest fragments in Ghana. Primates 47:365–373.

Supporting Information

Additional Supporting Information may be found in the online version of this article.