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The intake responses of three species of leaf-nosed Neotropical bats

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Abstract Flower-visiting bats encounter nectars that vary in both sugar composition and concentration. Because in the new world, the nectars of bat-pollinated flowers tend to be dominated by hexoses, we predicted that at equicaloric concentrations, bats would ingest higher volumes of hexoses than sucrose-containing nectars. We investigated the intake response of three species of Neotropical bats, *Leptonycteris curasoae*, *Glossophaga soricina* and *Artibeus jamaicensis*, to sugar solutions of varying concentrations (292, 438, 584, 730, 876, and 1,022 mmol L^{-1}) consisting of either sucrose or 1:1 mixtures of glucose and fructose solutions. Bats did not show differences in their intake response to sucrose and 1:1 glucose–fructose solutions, indicating that digestion and absorption in bat intestines are designed under the principle of symmorphosis, in which no step is more limiting than the other. Our results also suggest that, on the basis of energy intake, bats should not prefer hexoses over sucrose. We used a mathematical model that uses the rate of sucrose hydrolysis measured in vitro and the small intestinal volume of bats to predict the rate of nectar intake as a function of sugar concentration. The model was a good predictor of the intake responses of *L. curasoae* and *G. soricina*, but not of *A. jamaicensis*.

Abbreviations

ΔM	Change in mass
C_{sf}	Final concentration
V_0	Food intake
C_{s0}	Initial concentration
K_m	Michaelis–Menten constant
S_{max}	Rate of hydrolysis
$-r_s$	Sucrose hydrolysis
SI	Sugar intake
τ	Transit time
G	Volume of small intestine

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Introduction

The behavioural response of animals to foods that vary in energy concentration has been described in a variety of animals, ranging from insects (Slansky and Wheeler 1992; Josens et al. 1998) to large mammals (Spiegel 1973; Hansen et al. 1981). Typically, animals decrease intake as the concentration of assimilable energy in food increases (Montgomery and Bumgardt 1965; Slansky and Wheeler 1992). Castle and Wunder (1995) named the reciprocal relationship between intake and energy content "intake response".

The intake response has been well studied in nectar-feeding birds (Collins [1981]; López-Calleja et al. [1997]; McWhorter and Martínez del Río [2000]; Schondube and Martínez del Río [2003]). In these animals, intake decreases as a power function of sugar concentration [intake = a(concentration)[b]; where b < 0 (McWhorter and Martínez del Río [2000])]. Although less well known in bats, three recent studies, one on an old-world bat (*Rousettus aegyptiacus*; Korine et al. [2004]), and two on the New World bat *Glossophaga soricina* (Helversen and Winter [2003]; Ramírez et al. [2005]) suggest that the responses of nectar-feeding bats to sugar concentration in food are similar to those of nectar-feeding birds.

The power function relating intake with sugar concentration, has two non-exclusive explanations (Martínez del Río et al. [2001]). One is that nectar-feeding animals simply vary ingestion to maintain a constant sugar intake. In this compensatory feeding scenario, the exponent of the power function equals −1 (Montgomery and Bumgardt [1965]; Slansky and Wheeler [1992]), implying that sugar intake remains constant as concentration varies. Helversen and Winter ([2003]) reported that the bat *G. soricina* was able to achieve compensatory feeding, maintaining a constant energy intake of 48 kJ day^{-1}, by increasing food intake when its food concentration decreased from 876 to 292 mmol L^{-1} (sucrose equivalents). The alternative explanation to compensatory feeding is that a physiological process constrains food-intake (Levey and Martínez del Río [1999]; Martínez del Río et al. [2001]). If nectar-feeding animals are physiologically limited to achieve compensatory feeding, the exponent of the power function will be smaller than −1, implying that sugar intake increases with concentration.

McWhorter and Martínez del Rio ([2000]) constructed a mathematical model that predicts nectar intake from data on the kinetics of sucrose hydrolysis and the morphological characteristics of the small intestine. This model accurately predicts the relationship between intake and concentration in the broad-tailed hummingbird, *Selasphorus platycercus*. In contrast with the compensatory feeding explanation, the power function resulting from McWorther and Martínez del Rio's model has an exponent of −0.7. An exponent with such a value leads to a positive relationship between total sucrose intake and sugar concentration in food. Ramírez et al. ([2005]) applied the model to the nectar-feeding bat, *G. soricina*, under laboratory conditions and found that it describes the observed intake response of this species adequately. The exponent of the intake response power function in these bats was −0.52, indicating that the bats were physiologically limited to ingest food while feeding on sucrose solutions. Ramírez et al. ([2005]) hypothesized that sucrose

digestion may be the limiting step shaping the intake response of this species.

In this paper, we explored how bats modulate their nightly food intake in response to food of various concentrations and sugar compositions. Specifically, we investigated the role that sucrose digestion has over the intake responses of three Neotropical bat species in the family Phyllostomidae: *Leptonycteris curasoae*, *G. soricina*, and *Artibeus jamaicensis*. We measured the intake responses of bats to food containing various concentrations of sucrose or 1:1 mixtures of glucose–fructose. We expected bats to be like some passerine birds in which sucrose hydrolysis is more limiting than hexose uptake (Nicolson [2001]; Nicolson and Fleming [2003]; Schondube and Martínez del Rio [2003]), and hence that these animals would ingest more hexoses than sucrose if food contains the same caloric concentration. We use published data on the rate of sucrose hydrolysis and the small intestinal volume of bats (Hernández and Martínez del Rio [1992]; Schondube et al. [2001]) to examine whether the model predicts the bats' intake response accurately.

Materials and methods

Study site

The study was conducted in the Chamela Biological Station (19°22′–19°35′N, 104°56′–105°03′W) located in the coast of Jalisco, Mexico. Vegetation is dominated by lowland deciduous forest with small patches of riparian forest Bullock ([1995]). The climate is tropical subhumid with a marked dry season. The warmest part of the year is from June to September and the coldest months are from January to March (García-Oliva et al. [2002]). Nectar concentration found in the species visited by bats at the study site varied from 3 to 33% (Rodriguez-Peña et al. [2007]).

Bat care and housing

Adult, non-reproductive individuals of the three species were captured using mist nets and transferred to a room temperature laboratory where they were maintained in colonies in cages (0.6 × 0.6 × 0.6 m). Bats were fed on the maintenance diet described by Mirón ([2005]) that offered 22.2% sucrose and 4.4% of protein. This diet was supplemented with NEKTON-Plus®. Individuals were marked on the forearm with a numbered band and weighed daily at the beginning and end of each feeding trial using an electronic balance (0.01 g precision). Captive bats maintained mass for the duration of the experiments and

were released at their capture site when experiments were finished.

Intake responses

We measured nightly food intake (g 10 h^{-1}) of individual bats in outdoor enclosures ($2 \times 4 \times 1.6$ m) placed within the forest where bats could forage freely. During each feeding trial, we offered a solution of sucrose or a 1:1 mixture of glucose–fructose from 2000 hours to 0600 hours. Six individuals were used for each of the three bat species for a total of 18 bats. Each bat was offered seven solutions of sucrose and seven solutions of glucose–fructose. In our experiments, each bat of the three species was confronted to the seven concentrations of sucrose and hexoses, one per day, for a total of 84 trials for each species. The sugar concentrations of these solutions were 146, 292, 438, 584, 730, 876, and 1,022 mmol L^{-1}. We defined our concentrations based on the natural range of concentrations found in the nectar of bat-pollinated flowers at our study site (Rodriguez-Peña et al. 2007) and previous work conducted with *G. soricina* (Helversen and Winter 2003; Ramírez et al. 2005). Because the sucrose molecule mass (1 mole = 342.3 g) is not exactly the double of the molecular weight of glucose and fructose (for each 1 mole = 180.2 g), solutions of these sugars made on a weight/weight basis are not equicaloric. Sucrose solutions contain 5% more energy than a glucose–fructose solution. To avoid this problem, we made our solutions in sucrose equivalents. To do so, we used the molecular weight for both, sucrose and hexoses, and balanced the solutions following Fleming et al. (2004). We offered individual bats each of the concentrations in a random order. We weighed solutions at the beginning and end of each feeding trial to quantify food intake (electronic balance 0.01 g precision). Because experimental solutions lacked nitrogen sources, our experiments consisted of three nights of experiments per day of resting. During the "resting day" bats received the maintenance diet. Each night we placed a feeder full of each concentration outside the flight cages to control for evaporation and changes in concentration. These feeders were covered with a mosquito mesh, to avoid insects and other nocturnal animals from drinking from them. Control feeders were weighed at the beginning and end of each trial, and the concentration of the solution was measured using a hand-held refractometer (Reichert 10431 0–50°compensated Brix temperature, Leica, Buffalo NY, USA) to account for changes in concentration. No changes in volume or concentration were observed in our control feeders.

Model and data analysis

We estimated the slopes and intercepts of the relationships between food intake and concentration with least squares regression analysis on the log-transformed data of each individual bat. We compared the slopes and intercepts of the intake responses of the sucrose, and the glucose–fructose diets with a paired *t* test. We compared the value of the intake responses' exponents to the −1 value expected from the compensatory feeding expectation using a one-sample *t* test. In addition, we calculated the change in mass (ΔM, in g) experienced by the bats by weighing them at the beginning and the end of each trial. We expected ΔM to correlate positively with their sugar intake (SI, in g 10 h^{-1}). To test this conjecture, we correlated ΔM against the SI of each bat with Spearman's rank correlation (r_s) and tested whether the average r_s was significantly greater than 0 using a *t* test. This procedure is appropriate because it avoids the pseudoreplication that one would incur when estimating r_s for pooled data. The average r_s values for a sample of bats satisfy the central limit theorem and hence one can make inferences about whether they are positive or negative (Stuart and Odd 1994). Additionally, we compared our results with intake predictions from McWhorter and Martínez del Rio's (2000) model. This model assumes that the intestine is analogous to a chemical reactor, in which sucrose hydrolysis ($-r_s$) follows Michaelis–Menten kinetics:

$$-r_s = \frac{S_{max} C_s}{K_m + C_s}, \tag{1}$$

where S_{max} is the rate of hydrolysis along the intestine (μmol min^{-1} μL^{-1}), K_m is sucrase's Michaelis–Menten constant (μmol μL^{-1}), and C_s is the concentration of sucrose (μmol μL^{-1}) along the intestine. The time (τ) required to reduce the initial concentration of sucrose (C_{s0}) to a given final value (C_{sf}) can then be integrated from Eq. 1 to:

$$\tau = \frac{K_m \ln(C_{s0}/C_{sf}) + (C_{s0} - C_{sf})}{S_{max}}, \tag{2}$$

Intake rate (V_0 in μL min^{-1}), can then be estimated using the volume of the small intestine G (in μL) as:

$$V_0 = \frac{G}{t}. \tag{3}$$

The parameters that we used to fit the model and the sources from which we obtained them are presented in Table 1. To compare observed intake of sucrose solutions with those predicted from the model, we used the coefficient of determination as a descriptor of goodness of fit (Anderson-Spechel 1994). We compared this coefficient of determination with that of a power function fitted to the

Table 1 Parameters used to fit the McWorther and Martínez del Rio's (2000) model

	L. curasoae	*G. soricina*	*A. jamaicensis*
[a]S_{max} (µmol min^{-1} µL^{-1})	0.049	0.115	0.055
[a]K_m (µmol µL^{-1})	0.052	0.044	0.059
[b]C_{sf}	0.009	0.009	0.009
[a]G (µL)	0.750	0.304	2.320

S_{max} rate of hydrolysis along the intestine, K_m sucrase Michaelis–Menten constant, C_{sf} final concentration of sucrose after digestion, *G* volume of the intestine

[a] From Schondube et al. (2001)

[b] We measured the digestion efficiency by quantifying the sugar content in the excreta of bats feeding exclusively on sugar solutions with a hand-held refractometer (Accuracy; Reichert 10431 0–50°Brix temperature compensated, Leica, Buffalo NY, USA; Schondube and Martínez del Rio 2003). Because solutes other than sugars bias refractometer readings (Hiebert and Calder 1983; Inouye et al. 1980), our measurements of sugar concentration in excreta were used only to generate a relative measurement of digestion efficiency

same data set using a non-linear regression routine (JMP 5.1® 2003).

Results

Does the intake response to sucrose solutions differ from that of hexose mixtures?

The three bat species decreased intake as sugar concentration increased and the relationship between food intake and sugar concentration was well described by power functions (Table 2, Fig. 1). The response of bats to sucrose solutions did not differ from their response to hexose mixtures. We found that the intercepts and slopes of these relationships did not differ (Table 2). The exponent of the functions relating intake with concentration was in all cases significantly different from −1 (Table 2). Sugar intake increased significantly with sugar concentration in food for the nectarivores *L. curasoae* and *G. soricina* (Fig. 2; r_s = 0.83 ± 0.04 SE and 0.67 ± 0.04, respectively, t_5 > 17.40, $P < 0.001$); however, in the frugivore *A. jamaicensis*, sugar intake and sugar concentration in food were not significantly correlated (average r_s = 0.386 ± 0.16 SE, t_5 = 2.41, P = 0.061). The average Spearman-rank coefficient of correlation between SI and ΔM was significantly different from 0 for *L. curasoae* (average r_s = 0.350 ± 0.08 SE, t_5 = 4.03, P = 0.020), *G. soricina* (average r_s = 0.210 ± 0.06 SE, t_5 = 3.35, P = 0.015), and *A. jamaicensis* (average r_s = 0.580 ± 0.12 SE, t_5 = 4.68, P = 0.005). However, the positive relationship between SI and ΔM in the three species were variable and weak (Fig. 3).

Table 2 Intake responses to sucrose and hexose mixtures with their respective exponents, intercepts, and coefficients of determination

	Exponent ± SE	[a]Intercept ± SE	r^2
G. soricina			
S	−0.76* ± 0.064	3.40 ± 0.180	0.94 ± 0.020
G:F	−0.75* ± 0.076	3.38 ± 0.090	0.92 ± 0.020
t test	0.18	0.08	0.55
P	0.86	0.93	0.60
L. curasoae			
S	−0.61* ± 0.040	3.24 ± 0.110	0.9 ± 0.030
G:F	−0.62* ± 0.026	3.22 ± 0.090	0.96 ± 0.020
t test	0.19	0.28	1.76
P	0.86	0.80	0.15
A. jamaicensis			
S	−0.70* ± 0.079	3.46 ± 0.234	0.85 ± 0.050
G:F	−0.77* ± 0.078	3.57 ± 0.217	0.85 ± 0.039
t test	0.65	0.38	0.05
P	0.54	0.71	0.96

S sucrose, *G:F* glucose–fructose

* Mean exponent was significantly higher than −1 ($t > 2.95$, $P < 0.005$)

[a] This intercept equals Log(a) in the equation log(intake) = Log(a) + bLog(concentration)

Does the model accurately predict the bat's intake response?

The model described the shape of the relationship between intake and sucrose concentration in *L. curasoae* (r^2 = 0.539) and *G. soricina* (r^2 = 0.673) well. However, the model underestimated intake in *L. curasoae* and overestimated it slightly in *G. soricina* (Fig. 4a, b). The power functions estimated by non-linear regression provided better fits to the data than the model (r^2 = 0.861 and r^2 = 0.837 for *L. curasoae* and *G. soricina*, respectively). In the case of *A. jamaicensis*, the model greatly overestimated intake (Fig. 4c). The residual sum of squares was much larger than the total sum of squares, yielding a negative value for r^2. This result implies a very poor fit of the model to the data. Indeed, it implies that the mean intake at all concentrations is a better descriptor of the relationship between intake and concentration than the model (Anderson-Sprechel 1994). A power function fitted using non-linear regression fitted the data well (r^2 = 0.69, Fig. 4c).

Discussion

The three species of bats we studied had intake responses similar to those that have been observed in bats (Korine
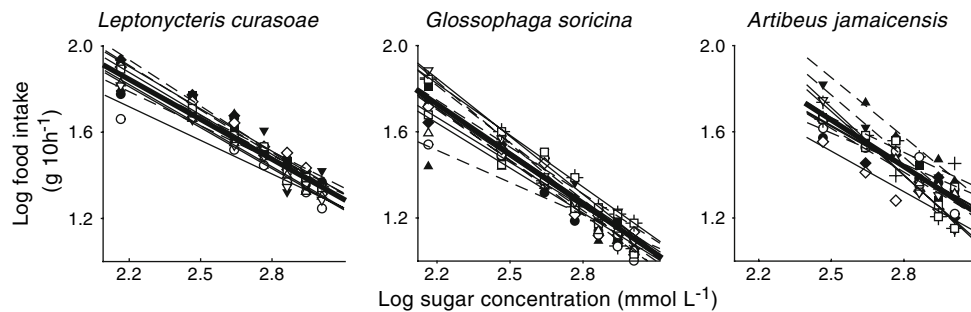
Fig. 1 Intake response of the three species of phyllostomid bats. The axes are logarithmic, and a different symbol and a different regression line represent each individual. The *heavy line* is the common regression line obtained from the average of all intercepts and slopes. *Closed symbols* represent sucrose whereas *open symbols* represent hexoses
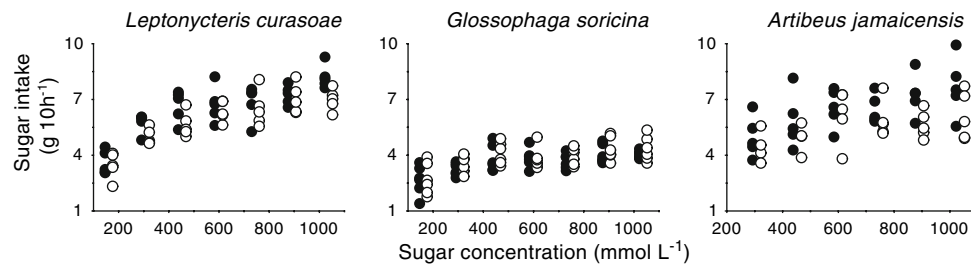
Fig. 2 Sugar intake increased significantly with sugar concentration in food for *L. curasoae* and *G. soricina* but not in *A. jamaicensis*. *Closed symbols* represent sucrose whereas *open symbols* represent hexoses. Both sucrose and hexose solutions had the same concentration; however, in order to avoid data point overlapping, we dislodge the *open symbols* to the *right* by 2 mm

et al. 2004; Ramírez et al. 2005) and birds (Martínez del Río et al. 2001; McWhorter and Lopez-Calleja 2000 among others). Furthermore there were no differences in the intake between sucrose and the 1:1 glucose–fructose solutions. The exponents of the intake responses for the three species of bats were different from −1, showing that sugar intake is positively correlated with sugar concentration (Levey and Martínez del Rio 1999; Martínez del Río et al. 2001). Here, we first discuss the potential prevalence of intake responses among nectar-feeding vertebrates and the consequences of this prevalence for osmoregulation in nectar and fruit-eating bats. Next, we consider the implications of no differences in the intake response of bats to sucrose and hexose solutions for the interaction between bats and the plants that they pollinate. Finally, we evaluate the role that digestive processes, like sucrose hydrolysis and hexose transport, have over the food intake of bats.

The osmoregulatory consequences of the prevalence of intake responses among nectar-feeding vertebrates

Our results demonstrate that three Neotropical bat species, with different levels of dietary specialization for nectar, show intake responses similar to those found in other nectar and fruit-eating animals. The intake responses of *L. curasoae*, *G. soricina*, and *A. jamaicensis* were similar to those reported for nectar and fruit-eating bats in the past (Thomas 1984; Helversen and Winter 2003; Korine et al. 2004; Ramírez et al. 2005). As in these studies, we found that bats respond to sugar concentration in food in the same fashion as birds do. We suggest that their response probably leads to the same consequences observed in birds. For example, Bakken et al. (2004) and McWhorter et al. (2004) found that intake responses in birds lead to very high water ingestion rates at low sugar concentrations. Similarly at low concentrations, *L. curasoae* and *G. soricina* ingested from 1.0 to 3.5 and 1 to 5 times, respectively, of their body mass in water (Fig. 1).

Bakken et al. (2004) identified the osmoregulatory quandary faced by hummingbirds. When feeding, these birds must dispose of ingested water rapidly. However, while they are fasting, hummingbirds must conserve water. This predicament is aggravated by the apparent inability of hummingbirds to concentrate urine (Lotz and Martínez del Río 2004). Our intake response results suggest that nectar-feeding bats probably share this osmoregulatory dilemma with hummingbirds. They ingest, and must dispose of large amounts of water when they are feeding. Like hummingbirds, the kidneys of nectar and fruit-eating bats seem better suited to dispose of water and to recover electrolytes than to concentrate urine (Studier and Wilson 1983; Herrera et al. 2001; Schondube et al. 2001). The rate at which bat kidneys can get rid of absorbed water when feeding at

Fig. 3 Mass change (ΔM, in g) increased as a function of sugar intake (SI, in g 10 h^{-1}). ΔM was calculated by weighting the bats at the beginning and the end of each feeding trial. In the three species, the average Spearman-rank correlation coefficient was significantly different from 0. Individuals are represented by *different symbols*. *Closed symbols* represent sucrose whereas *open symbols* represent hexoses

dilute nectar could limit their capacity to ingest additional water, possibly affecting food ingestion. The mechanisms used by bats to resolve the osmoregulatory predicament posed by intake responses are unknown and need to be explored.

The intake responses of bats did not differ between sugar types

Bats' intake responses to sucrose and hexoses were indistinguishable. This implies that for equicaloric concentrations, bats ingested the same amount of energy of glucose and fructose than of sucrose. Our results are supported by the findings of Voight and Speakman (2007) in the bat, *G. soricina*. They found that the rates of fractional incorporation of dietary sugars into the pool of metabolized substrates did not differ among sucrose, glucose, and fructose. The results we report, and the findings by Voight and Speakman (2007), are surprising on two accounts. First, the New World nectar-pollinated flowers secrete primarily hexose-dominated nectars (Pyke and Waser 1981; Baker and Baker 1983; Baker et al. 1998). Second, Herrera (1999) found that nectar-feeding bats preferred sucrose to equicaloric hexose solutions. Our results suggest that bats should be indifferent to sugar composition, because both the sucrose and hexose solutions have the same energetic value for them. One possible explanation is that bats are using cues other than energy intake, such as taste, when they make choices between different sugar solutions (Herrera 1999; Herrera et al. 2000). The preferences of bats for different sugars and the reasons that explain the prevalence of hexose-dominated nectars among New-world bat-pollinated plants remain unexplained.

Is food intake constrained in bats?

In all cases, the exponents of the intake responses were higher than −1 (Table 2). This exponent implies a positive correlation between sugar/energy intake and the sugar concentration of ingested food, and for this reason it has been interpreted as evidence of feeding constrained by one or many physiological processes (McWorther and Martínez
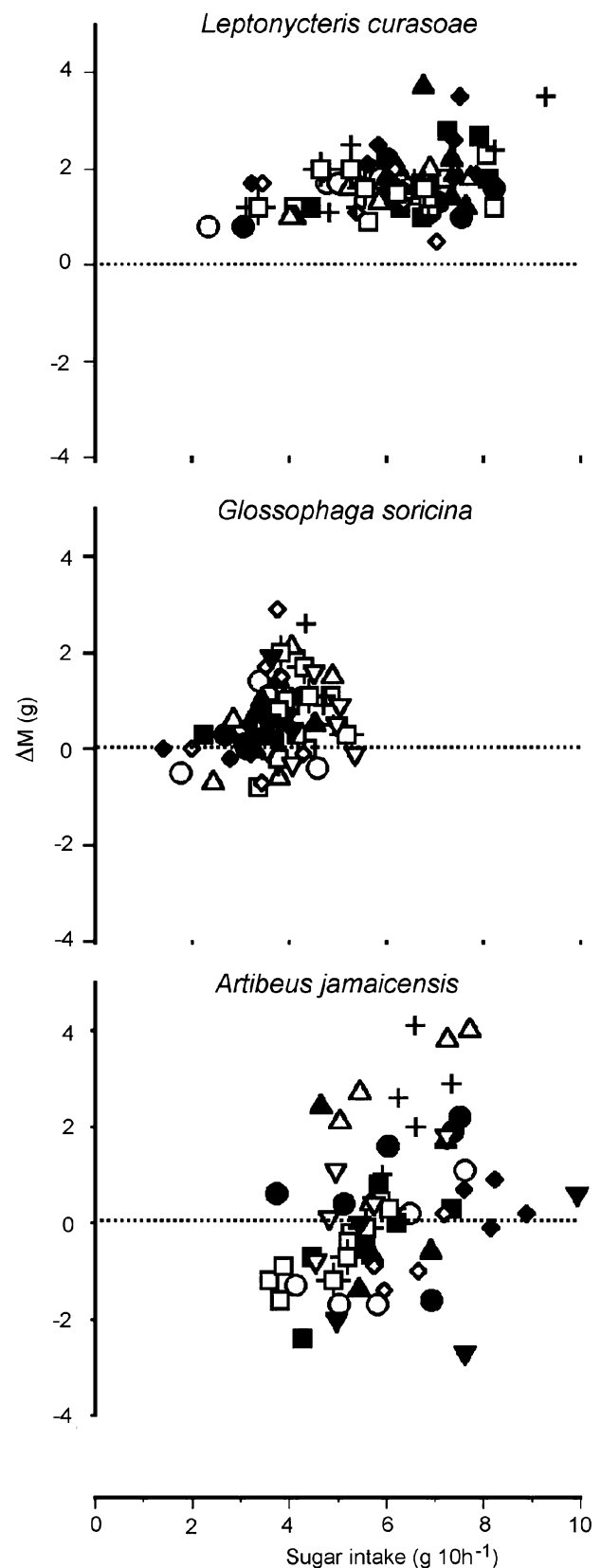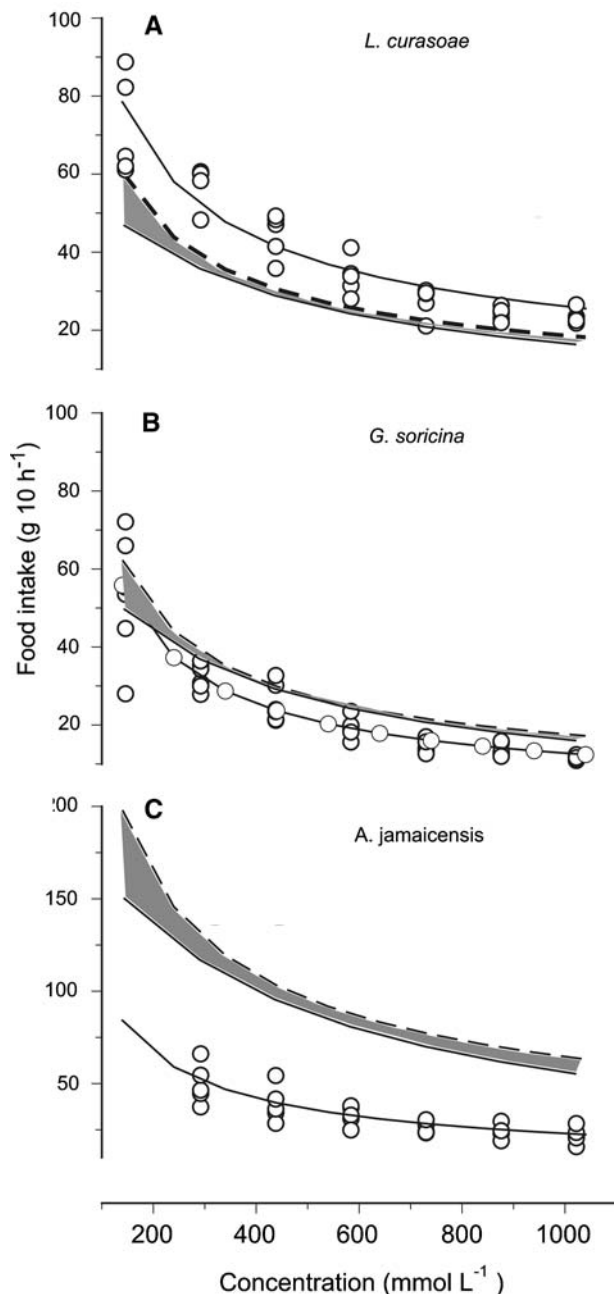
Fig. 4 Predictions generated by McWorther and Martínez del Rio's model (2000) are shown as a *dashed line*. A power function fitted with a non-linear square routine is represented with the *solid line*. The *grey area* represents the range of values predicted by the model when sugar assimilation is varied from from 0.998 (from Winter 1998) to 0.991 (data obtained from bats' excreta). Data are only for the intake response of bats to sucrose solutions

del Río 2000). The existence of a constraint is further supported by the positive correlation between sugar intake and sugar concentration. Similar results were reported for *G. soricina* by Roces et al. (1993). In compensatory feeders, energy intake remains relatively constant across concentrations (Lopez-Calleja et al. 1997; Martínez del Río

et al. 2001; Helversen and Winter 2003). Ramírez et al. (2005) suggested that sucrose hydrolysis might be the process that constrains intake in *G. soricina*. However, we found that there were no differences in the bats' intake response to glucose and fructose. This result is similar to that reported by Schondube and Martínez del Río (2003) in magnificent hummingbirds (*Eugenes fulgens*). The absence of a difference in the response of bats to sucrose and hexose solutions suggests that neither sucrose hydrolysis nor the uptake of hexoses is limiting. Sugar digestion and absorption in bat intestines seem to be designed according to the principle of symmorphosis, which postulates that physiological processes in series are designed so that no step is more limiting than others (Weibel 2000). Similar results have been found for several species of nectar-feeding birds, including hummingbirds (López Calleja et al. 1997; McWhorter and López-Calleja 2000; McWhorter and Martínez del Rio 2000; Martínez del Rio et al. 2001; Schondube and Martínez del Rio 2003; Fleming et al. 2004) and sunbirds (Fleming et al. 2004).

Although, the intake responses of the three species were qualitatively similar, there were some interesting differences. These differences are best highlighted by considering how well McWhorter and Martínez del Rio's (2000) model described the intake responses of bats. The model performed very poorly for the frugivore *A. jamaicensis*. These bats appear to ingest much less food than their intestines are capable of processing. Indeed, when all trials are considered, *A. jamaicensis* was the only species to maintain neutral mass ($\Delta M = 0.48$ g $\pm$ 0.75 SD, $t = 1.80$, $P > 0.05$). Both of the nectarivores, *G. soricina* ($\Delta M = 0.59$ g $\pm$ 0.75 SD, $t = 6.7$, $P < 0.001$) and *L. curasoae* ($\Delta M = 1.7$ g $\pm$ 0.62 SD, $t = 22.8$, $P < 0.001$) gained mass in these trials. Intake response studies in nectar-feeding birds emphasize potential post-ingestional constraints on feeding. Since *A. jamaicensis* is a frugivore that only ingests nectar occasionally, its oral morphology is better suited to chew fruit than to lap nectar (Ortega and Castro-Arellano 2001). We hypothesize that the intake response of *A. jamaicensis* was constrained more by pre-ingestional than by post-ingestional factors (Paton and Collins 1989; Mitchell and Paton 1990.

Both Helversen and Winter (2003) and Ramírez et al. (2005) measured the intake response of *G. soricina*. While Helversen and Winter found that bats were able to maintain a constant daily energy intake under laboratory conditions, Ramírez et al. (2005) reported that *G. soricina* was unable to perform compensatory feeding. Our experiments support the last result. Even if we exclude the intake values from the lowest concentration (146 mmol L^{-1}), at which bats ingested large amounts of diluted nectar, our results indicate that bats were unable to maintain compensatory feeding (mean exponent $\pm$ SE $= -0.85 \pm 0.03$; -0.76 ± 0.03,

and -0.70 ± 0.07, for *G. soricina*, *L. curasoae*, and *A. jamaicensis*, respectively; all values statistically different from -1, $P < 0.03$ for all cases). Ramírez et al. (2005) found that *G. soricina* had an exponent of -0.52, higher than the one we found (-0.76), and that bats ingested significantly less food per hour than in our experiments. One possible explanation for differences among the three studies is the use of different experimental designs. Helversen and Winter (2003) conducted their experiments in large flying cages, controlling temperature, relative humidity, and availability of food per visit. Ramírez et al. (2005) also conducted their experiments under controlled temperature and humidity, but used small cages ($0.2 \times 0.18 \times 0.18$ m) that did not allow flying, and offered the bats an unlimited amount of food. Our experiments were conducted under natural conditions (in the forest) within medium size cages ($2 \times 2 \times 1.6$ m) that allowed flying and ad libitum food. We suspect that the differences between the environmental and space conditions among the three studies lead to the different results. The constant temperature and relative humidity in Helversen and Winter's (2003) experiment could have reduced the variation in energy expenditures and the evapotranspiration of the bats. While in the Ramírez et al. experiment, the reduction of exercise due to the use of small cages could have lessened the metabolic expenditures causing a decrease in intake with respect to our data. Our experiments confronted bats with large natural changes in temperature. Nighttime temperatures during the experiments at our study site in February varied from 25°C at sunset to a minimum of 15°C at 0200 hours (Estación de Biología Chamela, unpublished data). These values are below the thermo neutral zone of some nectar-feeding phyllostomid bats (30–35°C; Cruz-Neto and Abe 1997). A 10°C reduction in ambient temperature, which is similar to the nightly temperature change experienced by our bats, could double their oxygen consumption (Cruz-Neto and Abe 1997). Under these conditions bats could become physiologically limited to maintain a constant influx of energy when feeding at a large range of sugar concentrations.

We found that McWhorter and Martínez del Rio's model (2000) explained 53 and 67% of the variation in intake response for *L. curasoae* and *G. soricina*, respectively. Although, power functions described the data better than this model (Fig. 4a, b), the adequate fit of the model is remarkable. It is more remarkable given that the enzyme and morphology data that we used to fit the model was obtained from the literature (Hernandez and Martínez del Río 1992; Schondube et al. 2001), and represents a different set of individuals. In addition, we used Winter's assimilation efficiency (1998) data to test the model sensitivity to variation in this parameter. A relatively small decrease in sugar assimilation efficiency from 0.998

(Winter 1998) to 0.991 (data obtained from bats' excreta), leads to a large change (17%) in intake at the lowest concentration (Fig. 4). We suggest that the model could describe accurately the intake responses of bats, but ideally its parameters must be drawn from the same population of bats in which the intake response is measured.

Both our experimental results and the predictions from the model indicate that both sucrose digestion and the uptake of hexoses can limit the food intake of nectar-feeding bats. These bats have high sucrose digestion rates that seem to be coupled with high transport rates of glucose and fructose. We speculate that food intake is not limited by a single factor but potentially by many steps along the digestion to catabolism pathway. The high water intakes of bats at the lowest sugar concentrations also lead us to speculate that getting rid of excess absorbed water may represent a problem to these animals. Limitations to food intake, while feeding on nectar, in fruit- and nectar-feeding bats, seem to be caused by both pre- and post-ingestional factors, such as mouth and tongue morphology in the fruit-eating bat *A. jamaicensis*, and digestive, osmoregulatory, and metabolic processes in *L. curasoae* and *G. soricina*. The existence of physiological limitations that prevent leaf-nosed bats from achieving compensatory feeding could have fitness effects on them, when they face dilute-diets and/or high energetic demands, like cold weather, lactation, or migratory demands.

References

Anderson-Sprecher R (1994) Model comparisons and R^2. Am Stat 48:113–117

Bakken BH, McWorther TJ, Tsahar E, Martínez del Río C (2004). Hummingbirds arrest their kidneys at night: diel variation in glomerular filtration rate in *Selasphorus platycercus*. J Exp Biol 207:4383–4391

Baker HG, Baker IS (1983) Floral nectar constituents in relation to pollinator type. In: Jones CE, Little RJ (eds) Handbook of pollinators. New York Scientific and Academic, pp 117–141

Baker HG, Baker IS, Hodges A (1998) Sugar composition of nectars and fruits consumed by birds and bats in the tropics and subtropics. Biotropica 30:559–586

Bullock SH (1995) Plant reproduction in neotropical dry forest. In: Bullock SH, Mooney HA, Medina E (eds) Seasonally dry tropical forest. Cambridge University Press, Cambridge, pp 277–203

Castle KT, Wunder BA (1995) Limits to food intake and fiber utilization in the prairie vole *Microtus orchrogaster*: effects of food quality and energy need. J Comp Physiol A 164:609–617

Collins BG (1981) Nectar intake and water balance for two species of Australian honeyeater, *Lichmera indistincta* and *Acanthorynchus superciliosis*. Physiol Zool 54:1–13

Cruz-Neto AC, Abe AS (1997) Metabolic rate and thermoregulation in the nectarivorous bat *Glossophaga soricina* (Chiroptera, Phillostomidae). Rev Brasil Biol 57:203–209

Fleming PA, Bakken BH, Lotz CN, Nicolson SW (2004) Concentration and temperature effects on sugar intake and preferences in a sunbird and Hummingbird. Funct Ecol 18:223–232

García-Oliva F, Camou A, Mass JM (2002) El clima de la región central de la costa del pacífico mexicano. In: Noguera F, Vega SH, García Aldrete AN, Quesada M (eds) Historia natural de chamela. Universidad Nacional Autónoma de México, México, pp 3–10

Hansen CB, Jen KC, Kribs P (1981) Regulation of food intake in monkeys. Physiol Behav 26:479–486

Helversen O, Winter Y (2003) Glossaphagine bats and their flowers: costs and benefits for plants and pollinators. In: Kunz TH, Fenton MB (eds) Bat ecology. University of Chicago Press, Chicago, pp 346–397

Hernandez A, Martínez del Río C (1992) Intestinal disaccharidases in five species of phyllostomid bats. J Comp Physiol B 103:105–111

Herrera LG (1999) Preferences for different sugars in Neotropical nectarivorous and frugivorous bats. J Mammal 80:683–688

Herrera LG, Leblanc D, Nassar J (2000) Sugar discrimination and gustatory thresholds in captive-born frugivorous Old World bats. Mammalia 64:135–143

Herrera LG, Martínez del Rio C, Braun E, Hobson K (2001) Renal structure in phyllostomid bats: using stable isotopes to explore relationships between diet and morphology. Isotopes Environ Health Stud 37:1–11

Hiebert SM, Calder WA III (1983) Sodium, potassium, and chloride in floral nectars: energy-free contributions to refractive index and salt balance. Ecology 64:399–402

Inouye DW, Favre ND, Lanum JA, Levine DM, Meyers JB, Roberts MS, Tsao FC, Wang Y-Y (1980) The effects of non-sugar nectar constituents on estimates of nectar energy content. Ecology 61:992–996

JMP 5.1® (2003) SAS institute, Cary, NC, USA

Josens RB, Farina WM, Roces F (1998) Nectar feeding by the ant *Camponotus mus*: intake rate and crop filling as a function of sucrose concentration. J Insect Physiol 44:579–585

Korine C, Speakman J, Arad Z (2004) Reproductive energetics of captive free-ranging Egyptian fruit bats *Rossetus aegymtiacus*. Ecology 85:220–230

Levey DJ, Martínez del Rio C (1999) Test, rejection, and reformulation of a chemical reactor-based model of gut function in a fruit-eating bird. Physiol Biochem Zool 72:369–383

López-Calleja MV, Bozinovic F, Martínez del Rio C (1997) Effects of sugar concentration on hummingbird feeding and energy use. Comp Biochem Physiol A 118:1291–1299

Lotz C, Martínez del Rio C (2004) The ability of rufous hummingbirds (*Selasphorus rufus*) to dilute and concentrate urine. J Avian Biol 35:54–62

Martínez del Rio C, Schondube JE, McWhorter TJ, Herrera LG (2001) Intake responses in nectar feeding birds: digestive and metabolic causes, osmoregulatory consequences, and coevolutionary effects. Am Zool 41:902–915

McWorther TJ, López-Calleja MV (2000) The integration of diet, physiology and ecology of nectar-feeding birds. Rev Chil Hist Nat 73:415–460

McWorther TJ, Martínez del Rio C (2000) Does gut function limit hummingbird food intake? Physiol Biochem Zool 73:313–324

McWorther TJ, Martínez del Rio C, Pinshow B, Roxburg L (2004) Renal function in Palestine Sunbirds: elimination of excess water does not constrain energy intake. J Exp Biol 23:3391–3398

Mirón ML (2005) Efecto de la dieta en el fraccionamiento isotópico de 13C y 15N en el murciélago *Glossophaga soricina* (Chiroptera: Phillostomidae). Tesis de maestría (Maestría en ciencias biológicas), Facultad de Ciencias, UNAM

Mitchell RJ, Paton DC (1990) Effects of nectar volume and concentration on sugar intake rates of Australian honeyeaters (Meliphagidae). Oecologia 83:238–246

Montgomery MJ, Baumgardt BR (1965) Regulation of food intake in ruminants. 2. Pelleted rations varying in energy concentration. J Dairy Sci 48:569–577

Nicolson SW (2001) Pollination by passerine birds: why are nectars so dilute? Comp Biochem Physiol 4:1–8

Nicolson SW, Fleming PA (2003) Nectar as food for birds: the physiological consequences of drinking dilute sugar solutions. Plant Syst Evol 238:139–153

Ortega J, Castro-Arellano I (2001) *Artibeus jamaicensis*. Mammal Species 662:1–9

Paton DC, Collins BG (1989) Bills and tongues of nectar-feeding birds: a review of morphology, function and performance, with intercontinental comparisons. Aust J Ecol 14:473–506

Pyke GH, Waser NM (1981) The production of dilute nectars by hummingbird and honeyeater flowers. Biotropica 13:260–270

Ramírez NP, Herrera GM, Mirón L (2005) Physiological constraint to food ingestion in a New World nectarivorous bat. Physiol Biochem Zool 78:1032–1038

Roces F, Winter Y, Helversen O (1993) Nectar concentration preference and water balance in a flower visiting bat, *Glossophaga soricina antillarum*. In: Barthlott W (eds) Animal–plant interactions in tropical environments. Museum Koening, Bonn, pp 159–165

Rodriguez-Peña N, Stoner K, Schondube J, Ayala-Berdón J, Martínez del Río C (2007) Effects of sugar composition and concentration on food selection by *Leptonycteris curasoae* and *Glossophaga soricina* (Chiroptera: Phyllostomidae: Glossophaginae). J Mammal (in press)

Schondube JE, Martínez del Río C (2003) Concentration-dependent sugar preferences in nectar-feeding birds: mechanisms and consequences. Funct Ecol 17:445–453

Schondube JE, Herrera LG, Martínez del Río C (2001) Diet and the evolution of digestion and renal function in phyllostomid bats. Zoology 104:59–73

Slansky F, Wheeler GS (1992) Caterpillars compensatory feeding response to diluted nutrients leads to toxic allelochemical dose. Entomol Exp Appl 65:171–186

Spiegel TA (1973) Caloric regulation of food intake in man. J Comp Physiol Psychol 84:24–37

Stuart A, Ord JK (1994) Kendall's advanced theory of statistics, vol 1. Edward Arnold, New York

Studier EH, Wilson DE (1983) Natural urine concentrations and composition in Neotropical bats. Comp Biochem Physiol 75:509–515

Thomas DW (1984) Fruit intake and energy budgets of frugivorous bats. Physiol Zool 57:457–467

Voight CC, Speakman JR (2007) Nectar-feeding bats fuel their high metabolism directly with exogenous carbohydrates. Funct Ecol 21:913–921

Weibel ER (2000) Symmorphosis: on form and function in shaping life. Harvard University Press, Cambridge

Winter Y (1998) In vivo measurement of near maximal rates of nutrient absorption in a mammal. Comp Biochem Physiol 119:853–859