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ORIGINAL PAPER

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Comparative analysis of the digestive efficiency and nitrogen and energy requirements of the phyllostomid fruit-bat (*Artibeus jamaicensis*) and the pteropodid fruit-bat (*Rousettus aegyptiacus*)

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Abstract Nitrogen (N) and energy (E) requirements of the phyllostomid fruit bat, *Artibeus jamaicensis*, and the pteropodid fruit bat *Rousettus aegyptiacus*, were measured in adults that were fed on four experimental diets. Mean daily food intake by *A. jamaicensis* and *R. aegyptiacus* ranged from 1.1–1.6 times body mass and 0.8–1.0 times body mass, respectively. Dry matter digestibility and metabolizable E coefficient were high (81.1% and 82.4%, respectively) for *A. jamaicensis* and (77.5% and 78.0%, respectively) for *R. aegyptiacus*. Across the four diets, bats maintained constant body mass with mean metabolizable E intakes ranging from 1357.3 kJ · $kg^{-0.75}$ · day^{-1} to 1767.3 kJ · $kg^{-0.75}$ · day^{-1} for *A. jamaicensis* and 1282.6–1545.2 kJ · $kg^{-0.75}$ · day^{-1} for *R. aegyptiacus*. Maintenance E costs were high, in the order of 3.6–5.4 times the basal metabolic rate (BMR). It is unlikely that the E intakes that we observed represent a true measure of maintenance E requirements. All evidence seems to indicate that fruit bats are E maximizers, ingesting more E than required and regulating storage by adjusting metabolic output. We suggest that true maintenance E requirements are substantially lower than what we observed. If it follows the eutherian norm of two times the BMR, fruit bats must necessarily over-ingest E on low-N fruit diet. Dietary E content did affect N metabolism of *A. jamaicensis*. On respective low- and high-E diets, metabolic fecal N were 0.492 mg N · g^{-1} and 0.756 mg N · g^{-1} dry matter intake and endogenous urinary N losses were 163.31 mg N · $kg^{-0.75}$ · day^{-1} and 71.54 mg N · $kg^{-0.75}$ · day^{-1}. *A. jamaicensis* required 332.3 mg · $kg^{-0.75}$ · day^{-1} and 885.3 mg · $kg^{-0.75}$ · day^{-1} of total N on high- and low-E diets, respectively, and 213.7 mg · $kg^{-0.75}$ · day^{-1} of truly digestible N to achieve N balance. True N digestibilities were low (29% and 49%) for low- and high-E diets, respectively. For *R. aegyptiacus*, metabolic fecal N and endogenous urinary N losses were 1.27 mg N · g^{-1} dry matter intake and 96.0 mg N · $kg^{-0.75}$ · day^{-1}, respectively, and bats required 529.8 mg · $kg^{-0.75}$ · day^{-1} (total N) or 284.0 mg · $kg^{-0.75}$ · day^{-1} (truly digestible N). True N digestibility was relatively low (50%). Based on direct comparison, we found no evidence that *R. aegyptiacus* exhibits a greater degree of specialization in digestive function and N retention than *A. jamaicensis*. When combined with results from previous studies, our results indicate that all fruit bats appear to be specialized in their ability to retain N when faced with low N diet.

Abbreviations *BMR* basal metabolic rate · *DMD* dry matter digestibility · *DLW* doubly labeled water · *E* energy · *EUN* endogenous urinary nitrogen · *FMR* field metabolic rate · *MFN* metabolic fecal nitrogen · *N* nitrogen · *SDA* specific dynamic action

Introduction

The sub-order Megachiroptera, comprising the Old World fruit bats of the family Pteropodidae, contains approximately 150 species which span the range from more or less obligate frugivores to specialized nectarivores, with intermediate mixed fruit and nectar feeders being common. Whether they radiated early from a chiropteran stock or arose as a convergence from an ancestral primate stock (Anderson and Jones 1967; Pettigrew 1995), it remains that the Pteropodidae represent the oldest group of "chiropteran" frugivores. This long association with a frugivorous diet is reflected in

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their simplified dentition, highly developed vision, and lack of echolocation (with the exception of the genus *Rousettus*, see Fenton et al. 1995). Although nitrogen (N) rich food such as insects and liquid fractions of leaves have been reported in their diets (Roberts and Seabrook 1989; Parry-Jones and Augee 1992; Kunz and Ingalls 1994; Kunz and Diaz 1995), there is little doubt that pteropodid bats are primarily fruit and nectar specialists.

The neotropical family Phyllostomidae includes a diverse array of approximately 137 species of insectivores, frugivores, nectarivores and vampires. The comparatively recent evolution of frugivory within the Phyllostomidae is indicated by the retention of many anatomical features associated with insectivory, including complex teeth, small eyes and echolocation. Even the most frugivorous of phyllostomids, such as members of the genera *Carollia* and *Artibeus*, continue to include both insects and the liquid fraction of protein-rich leaves in their diet, at least seasonally if not on a regular basis (Fleming et al.1972; Kunz and Diaz 1995).

The existence of two groups of frugivores within the same taxonomic unit (order Chiroptera), yet having distinct evolutionary histories, allows us to examine whether the evolution of frugivory as a dietary strategy necessarily entails adjustments in nutritional requirements and/or digestive physiology. Fruits are commonly considered to be nutritionally poor with most succulent fruits offering <1% N or <6% crude protein by dry mass (Howe and Estabrook 1977; Foster 1978). When one considers that most mammalian herbivores cannot achieve a positive N balance and hence sustain body condition and growth, on diets containing <6% crude protein (Holter et al. 1979; Milton 1979; Mattson 1980), there is reason to believe that frugivory imposes specific nutritional constraints, particularly with regards to nitrogen (N) extraction, retention, and metabolism.

To survive, grow, and reproduce on low-N fruit diets, frugivores may exhibit one or more of a number of physiological and behavioral adjustments. They may potentially limit N turnover. This would be observed as a reduction in the basal rate of urinary N output (endogenous urinary N; EUN) which is considered to represent N requirement for the maintenance and repair of tissue protein. The low maintenance N requirement of frugivorous oilbirds (*Steatornis caripensis*) has been attributed to such a reduction in N turnover (Bosque and De Parra 1992). Frugivores may also limit endogenous N losses in the feces resulting from the passage of dry matter through the digestive tract (metabolic fecal N; MFN). The comparatively low MFN values found for sugar gliders (Smith and Green 1987) and the phyllostomid fruit bat, *Carollia perspicillata*, (Delorme and Thomas 1996) may represent physiological adjustments in digestive function. Frugivores may increase the efficiency of protein digestion and absorption in the gut, resulting in an increased N retention. Finally, they may entirely escape the constraints imposed by the low N content of fruits by supplementing their diets with alternate protein-rich sources such as insects, pollen, and leaves. In this latter case, frugivores would be less subject to natural selection and one would not expect to see as strongly developed digestive or physiological adaptations to frugivory.

Because phyllostomids have evolved a frugivorous habit more recently than pteropodids, as is testified by the different degrees of morphological specialization found in the two groups, one might expect that phyllostomids would exhibit a lower degree of specialization in digestive functions favoring efficient N extraction and retention. This would be particularly true if phyllostomid frugivores supplement their diets with insects on a regular basis and so escape the selective pressure imposed by an N-poor diet. Although several studies have addressed the problem of N balance in different members of the Phyllostomidae and Pteropodidae (e.g., Herbst 1986; Steller 1986; Law 1992; Delorme and Thomas 1996; Korine et al. 1996), there has been no direct comparison of digestive efficiency and N metabolism between two species of fruit-eating bats representing two distinct suborders and families (Microchiroptera: Phyllostomidae and Megachiroptera: Pteropodidae). Between-species comparisons are also complicated by the fact that, with one exception (Delorme and Thomas 1996), studies have not partitioned N losses between urinary and fecal routes, rendering it impossible to examine N metabolism and digestive function in detail.

In this paper we compare the digestive efficiency and both N and energy (E) requirements of two species of fruit-eating bats representing two suborders and families to test the prediction that one member of the Pteropodidae (*Rousettus aegyptiacus*) exhibits a greater degree of specialization in digestive function and N retention than a single member of the Phyllostomidae (*Artibeus jamaicensis*). We assume that if any fundamental physiological differences exist between the two sub-orders, Mega- and Microchiroptera, then these would be reflected in digestive function and/or N requirements of these two widely distributed representatives.

Materials and methods

We conducted this study at the Biodôme de Montréal using *A. jamaicensis* and *R. aegyptiacus* that had been born and raised in captivity. Prior to feeding trials bats were able to fly freely in a large exhibit where the ambient temperature was 23–26 °C and the light cycle was 12L:12D.

In this study, housing conditions, diets and feeding trials follow those described in Delorme and Thomas (1996). Briefly, for each feeding trial, we confined 5 *A. jamaicensis* and 5 *R. aegyptiacus* (2–3 males and 2–3 females per trial) separately in inverted 5-l or 18-l plastic bottles, respectively. Bats were fed a given diet (for composition see Table 1, Delorme and Thomas 1996) for 3 pre-trial days (acclimation) and 4 trial days (measurement). Feces and urine were collected daily at 0800 hours and stored frozen until subsequent analyses. Analyses for urinary, fecal and total N and E followed those described in Delorme and Thomas (1996).

We calculated dry matter digestibility (DMD), metabolizable E coefficient, and apparent and true N digestibilities following Robbins (1993). We estimated maintenance N requirement by regress-

Table 1 Food intake, total energy (*E*) intake, dry matter digestibility (DMD), metabolizable E coefficients and metabolizable E intake for *Rousettus aegyptiacus* fed four experimental diets. *P* is the probability from Kruskal-Wallis comparisons among groups. Analyses are based on $n = 20$ per diet, representing five bats held for four days. (*N* nitrogen)

	Low N/Low E ($n = 20$)	Low N/high E ($n = 20$)	High N/low E ($n = 20$)	High N/high E ($n = 20$)	*P*
Food intake ($g \cdot kg^{-0.75} \cdot day^{-1}$)	637.9 ± 41.6[a]	593.1 ± 33.4[a]	553.3 ± 24.9[a]	528.6 ± 28.4[a]	0.24
Total E intake ($kJ \cdot kg^{-0.75} \cdot day^{-1}$)	1715.9 ± 112.1[a]	1957.2 ± 110.1[a]	1731.8 ± 78.1[a]	1955.9 ± 105.0[a]	0.26
DMD (%)	82.3 ± 0.7[a]	80.4 ± 1.9[a]	73.1 ± 0.5[b]	74.4 ± 1.1[b]	0.0001
Metabolizable E coefficient (%)	82.4 ± 1.0[a]	80.2 ± 2.3[a]	74.3 ± 0.8[b]	74.8 ± 1.2[b]	0.0001
Metabolizable E intake ($kJ \cdot kg^{-0.75} \cdot day^{-1}$)	1407.7 ± 89.1[a]	1545.2 ± 82.8[a]	1282.6 ± 54.1[a]	1469.9 ± 87.4[a]	0.16

[a,b] Diets with the same letter are not significantly different based on a Tukey grouping

ing N balance against dietary N intake and calculating the N intake required to achieve N balance. We estimated MFN by regressing apparent N digestibility against N content of the diet, where MFN is the Y intercept. We estimated EUN by regressing urinary N content against dietary N intake, where EUN is the Y intercept. For comparison with bats of this study, we used data from Delorme and Thomas (1996) for the phyllostomid *Carollia perspicillata*.

Separate groups of five bats were subjected to the four different diets (total sample size = 20 bats/species) to eliminate the possible carry-over effect and cumulative stress that could potentially be induced by the individual confinement of colonial bats in bottles (see Widmaier and Kunz 1993). Statistical analysis was performed using repeated measures ANOVA. Because it was impossible to obtain normality of residuals under any transformation, and because of the small sample sizes, we used a Kruskal-Wallis nonparametric test. Multiple comparisons were done according to Tukey's method, using ranks. In regressions we present all measures, including measures for the same bat over successive days, as independent data points. Although this procedure may artificially inflate the degrees of freedom and hence significance levels, all regressions remained highly significant when the degrees of freedom were reduced to the number of individuals rather than the number of data points. We conclude that this degree of pseudoreplication did not affect our conclusions. Data are presented as mean ± SE.

Results

Food and E intake

Depending on day and diet, *R. aegyptiacus* ingested 71.7–205.0 g of food daily, representing intakes of 0.5–1.6 times body mass on a daily basis. Table 1 presents mass-specific food and E intakes and digestibilities. Food intake on a given diet was variable and coefficients of variation ranged from 20% on the high N/low E diet to 29% on the low N/low E diet. This substantial variation in food intake, which results in low statistical power, did not permit the detection of any significant difference between the four diets for the mass-specific food and gross E intake (food intake: Kruskall-Wallis $H = 4.2$, $df = 3$, $P = 0.24$; gross E intake: $H = 3.9$, $df = 3$, $P = 0.26$). However, on low-E diets, bats tended to have higher food but lower gross E intakes than on high-E diets. Although DMD and metabolizable E coefficient were both significantly lower on the high-N diets ($H = 35.6$, $df = 3$, $P < 0.0001$, and $H = 28.2$, $df = 3$, $P < 0.0001$, respectively), this did not result in any clear pattern on metabolizable E intake. As for the gross E intake, there were no significant differences in metabolizable E intake between diets ($H = 5.06$, $df = 3$, $P = 0.16$), probably due to the combined effects of low statistical power and substantial variation, which ranged from 19.0–28.3%, across diets. However, metabolizable E intake tended to be significantly lower on low-E than on high-E diets (Table 1).

A. jamaicensis ingested 28.1–80.8 g food daily which represented intakes of 0.8–2.2 times body mass. Mass-specific food intake ($g \cdot kg^{-0.75} \cdot day^{-1}$) of *A. jamaicensis* differed significantly between the four diets (Kruskall-Wallis, $H = 26.2$, $df = 3$, $P < 0.0001$) (Table 2). *A. jamaicensis* showed a clear pattern of a reduction in food intake on high-E diets. Bats ingested 18% less food on low N/high E diet than on low N/low E (499.9 ± 15.1 $g \cdot kg^{-0.75} \cdot day^{-1}$ vs 606.3 ± 30.2 $g \cdot kg^{-0.75} \cdot day^{-1}$, respectively) and 23% less on the high N/high E diet than on high N/low E diet (556.6 ± 21.4 $g \cdot kg^{-0.75} \cdot day^{-1}$ vs 720.1 ± 36.2 $g \cdot kg^{-0.75} \cdot day^{-1}$,

Table 2 Food intake, total E intake, DMD, metabolizable E coefficients and metabolizable E intake for *Artibeus jamaicensis* fed four experimental diets. *P* is the probability from Kruskal-Wallis comparisons among groups. Analyses are based on $n = 20$ per diet, representing five bats held for four days

	Low N/Low E ($n = 20$)	Low N/high E ($n = 20$)	High N/low E ($n = 20$)	High N/high E ($n = 20$)	*P*
Food intake ($g \cdot kg^{-0.75} \cdot day^{-1}$)	606.3 ± 30.2[ab]	499.9 ± 15.1[c]	720.1 ± 36.2[a]	556.6 ± 21.4[bc]	0.0001
Total E intake ($kJ \cdot kg^{-0.75} \cdot day^{-1}$)	1628.6 ± 81.1[b]	1647.0 ± 49.8[b]	2253.5 ± 113.2[a]	2058.5 ± 79.3[a]	0.0001
DMD (%)	82.6 ± 0.4[a]	85.6 ± 0.3[b]	76.7 ± 0.4[c]	79.4 ± 0.5[d]	0.0001
Metabolizable E coefficient (%)	83.3 ± 0.8[ab]	87.2 ± 0.8[a]	78.8 ± 2.3[b]	80.2 ± 2.2[b]	0.0009
Metabolizable E intake ($kJ \cdot kg^{-0.75} \cdot day^{-1}$)	1357.3 ± 68.0[b]	1435.0 ± 43.0[b]	1767.3 ± 97.0[a]	1649.8 ± 78.9[ab]	0.005

[a,b,c,d] Diets with the same letter are not significantly different based on a Tukey grouping

respectively). Because of the interaction between food intake and dietary E content, gross E intake varied significantly between the diets ($H = 26.2, df = 3, P < 0.0001$). Bats ingested 38.4% more E on high N/low E than on low N/low E diet (2253.5 ± 113.2 kJ · $kg^{-0.75}$ · day^{-1} vs 1628.6 ± 81.1 kJ · $kg^{-0.75}$ · day^{-1}) and 25% more on the high N/high E than on low N/high E diet (2058.5 ± 79.3 kJ · $kg^{-0.75}$ · day^{-1} vs 1647.0 ± 49.8 kJ · $kg^{-0.75}$ · day^{-1}). DMD and metabolizable E coefficient varied significantly between diets ($H = 62.1$, $df = 3$, $P < 0.0001$, and $H = 16.4$, $df = 3$, $P < 0.0009$, respectively). As with *R. aegyptiacus*, DMD and metabolizable E coefficient were significantly lower on high-N than on low-N diets, but DMD and metabolizable E coefficient were also significantly higher on high-E than on low-E diets (Table 2). Changes in DMD and metabolizable E coefficient did not act to stabilize metabolizable E intake, which varied significantly between diets ($H = 12.8$, $df = 3$, $P < 0.005$). Compared with the mean intake on low N diets (1396.1 kJ · $kg^{-0.75}$ · day^{-1}), bats assimilated a mean of 26.6% more E on the high N/low E diet (1767.3 kJ · $kg^{-0.75}$ · day^{-1}).

Depending on day and diet, daily metabolizable E intake varied from 757.5 kJ · $kg^{-0.75}$ · day^{-1} to 2873.5 kJ · $kg^{-0.75}$ · day^{-1} for *A. jamaicensis* and from 670.8 kj · $kg^{-0.75}$ · day^{-1} to 2299.4 kJ · $kg^{-0.75}$ · day^{-1} for *R. aegyptiacus*. Despite this more than three-fold variation in E intake, daily changes in body mass were not correlated with metabolizable E intake (*A. jamaicensis*: $r^2 = 0.0$, $df = 1,78$, $P > 0.05$; *R. aegyptiacus*: $r^2 = 0.0$, $df = 1,78$, $P > 0.05$; Fig. 1).

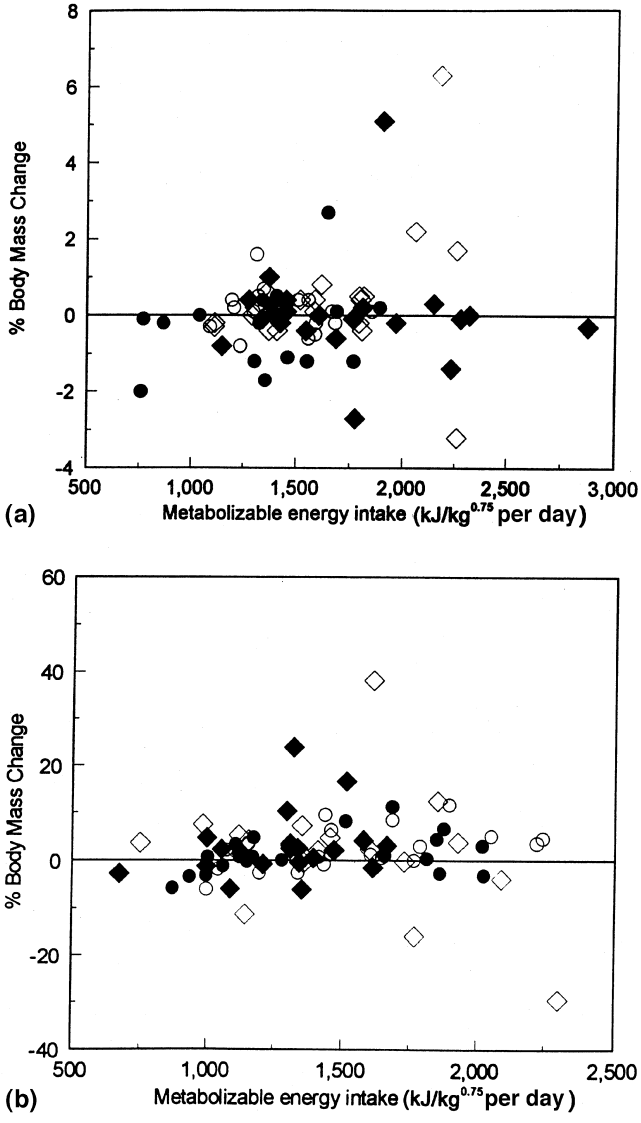


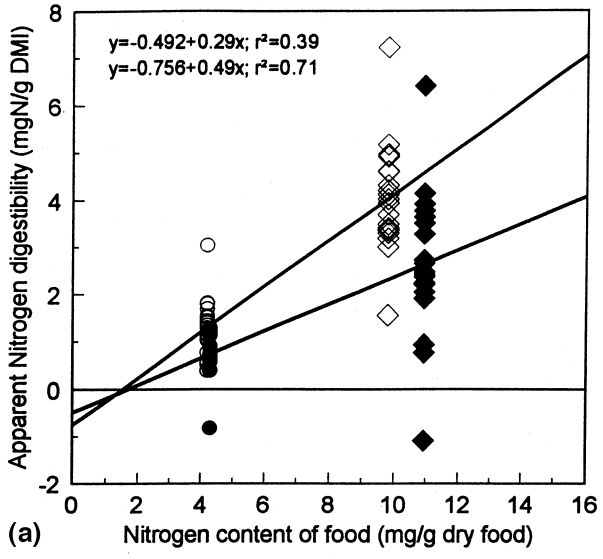
Fig. 1A,B Relationship between metabolizable energy (E) intake and percent change in body mass in **A** *Artibeus jamaicensis* ($n = 20$ per diet) and in **B** *Rousettus aegyptiacus*. *Open* and *filled circles* indicate low nitrogen (N)/high E and low N/low E diets, respectively. *Open* and *filled squares* indicate high N/high E and high N/low E diets, respectively

N digestibility and requirement

For *R. aegyptiacus*, apparent N digestibility was significantly correlated with dietary N intake ($r^2 = 0.58$, $df = 1,78$, $P < 0.001$; Fig. 2b) where: apparent N digestibility (mg N · g^{-1} dry matter intake) = −1.27 + 0.50 (N content). True N digestibility (the slope) was 0.50 and MFN (the Y-intercept) was 1.27 mg N · g^{-1} dry matter intake. True N digestibilities did not differ significantly between the four diets ($H = 7.2$, $df = 3$, $P > 0.05$). N balance was significantly related to both total dietary N intake ($r^2 = 0.49$, $df = 1,78$, $P < 0.001$; Fig. 3a) and truly digestible N intake ($r^2 = 0.87$, $df = 1,78$, $P < 0.001$; Fig. 3b). *R. aegyptiacus* achieved N balance with a minimum intake of 529.8 mg · $kg^{-0.75}$ · day^{-1} (132.0 mg · day^{-1}) of total N or 284.0 mg · $kg^{-0.75}$ · day^{-1} (70.7 mg · day^{-1}) of truly digestible N for an 156.7-g individual.

EUN was determined by regressing urinary N against dietary N intake and extrapolating to zero N intake (Fig. 4). For *R. aegyptiacus*, EUN was estimated at 96.0 mg N · $kg^{-0.75}$ · day^{-1} or 24 mg N · day^{-1} whereby: urinary N output (mg N · $kg^{-0.75}$ · day^{-1}) = 96.02 + 0.09 [Dietary N intake] ($r^2 = 0.28$, $df = 1,78$, $P < 0.0001$).

For *A. jamaicensis*, inspection of Figs. 2a and 5a seems to show that dietary E content affects apparent N digestibility and N balance. Low-E diets result in lower apparent N digestibility and less positive N balances at both low and high dietary N levels. We thus analyzed apparent N digestibility and N balance for low-E and high-E diets separately. For low-E diets, apparent N digestibility was significantly correlated with dietary N content where apparent N digestibility = −0.492 + 0.29 [N content], ($r^2 = 0.39$, $df = 1,73$, $P < 0.001$). For high-E diets, apparent N digestibility = −0.756 + 0.49 [N content], ($r^2 = 0.71$, $df = 1,73$, $P < 0.001$; Fig. 2a). The slopes of these regressions estimate true N digestibility as 0.29 and



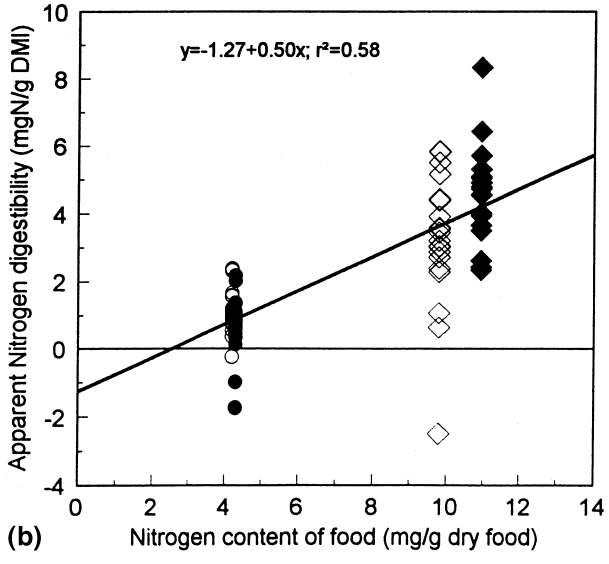
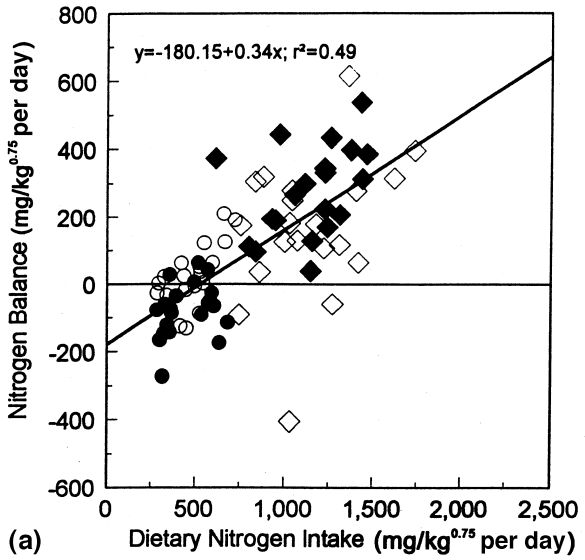


Fig. 2A,B Relationship between apparent N digestibility and N content of food in **A** *A. jamaicensis* (n = 20 per diet except for the low N/low E n=15) and in **B** *R. aegyptiacus* (n = 20 per diet). Symbols represent the four diets as in Fig. 1



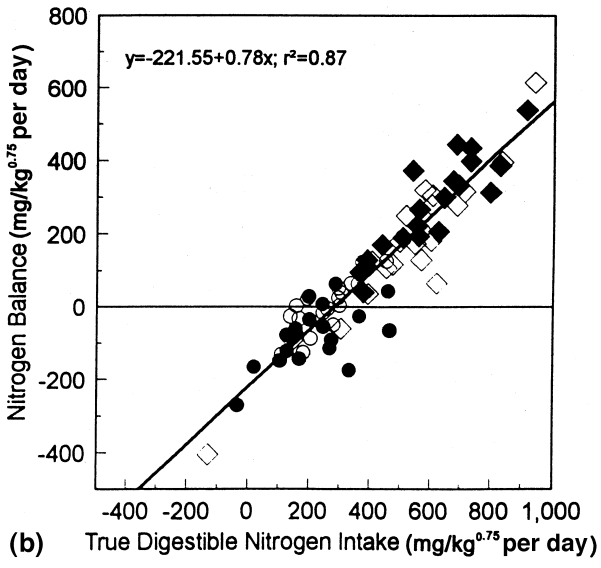


Fig. 3A,B Relationship between N balance and total dietary N intake **A** or truly digestible N intake **B** in *R. aegyptiacus* (n = 20 per diet). Symbols represent the four diets as in Fig. 1

0.49 for low- and high-E diets, respectively. The intercepts estimate MFN at 0.492 mg N · g^{-1} and 0.756 mg N · g^{-1} dry matter intake for low- and high-E diets, respectively. True N digestibilities did differ significantly between the four diets (H = 26.6, df = 3, P < 0.0001). Because apparent N digestibility was affected by dietary E content, bats achieved N balance with intakes of either 332.3 mg · $kg^{-0.75}$ · day^{-1} of total N or 885.3 mg · $kg^{-0.75}$ · day^{-1}. This represents intakes of 27.9 mg · day^{-1} or 74.4 mg · day^{-1} of total N for a 36.8-g individual maintained on high- and low-E diets, respectively. Once the effect of dietary E content on true N digestibility is accounted for, N balance was highly correlated with truly digestible N intake (r^2 = 0.92, df = 1,73, P < 0.001; Fig. 5b). *A. jamaicensis* requires a minimum intake of 213.7 mg · $kg^{-0.75}$ · day^{-1} of truly digestible N to achieve N balance or 18.0 mg · day^{-1} for a 36.8-g individual.

For *A. jamaicensis,* the effect of dietary E content on N metabolism seems again reflected in urinary N output. Inspection of Fig. 4a shows that bats fed low-E diets lost more urinary N than those fed high-E diets. On low-E diets, urinary N output (mg N · $kg^{-0.75}$ · day^{-1}) = 163.31 + 0.04 [Dietary N intake] (r^2 = 0.14, df = 1,73, P < 0.0001) whereas for high-E diets, urinary N output (mg N · $kg^{-0.75}$ · day^{-1}) = 71.54 + 0.05 [Dietary N intake] (r^2 = 0.21, df = 1,73, P < 0.0001). The intercepts of these two graphs estimate EUN at 163.31 mg N · $kg^{-0.75}$ · day^{-1} or 13.7 mg N · day^{-1} (low-E diets) or 71.54 mg N · $kg^{-0.75}$ · day^{-1} or 6.0 mg N · day^{-1} on high-E diets.

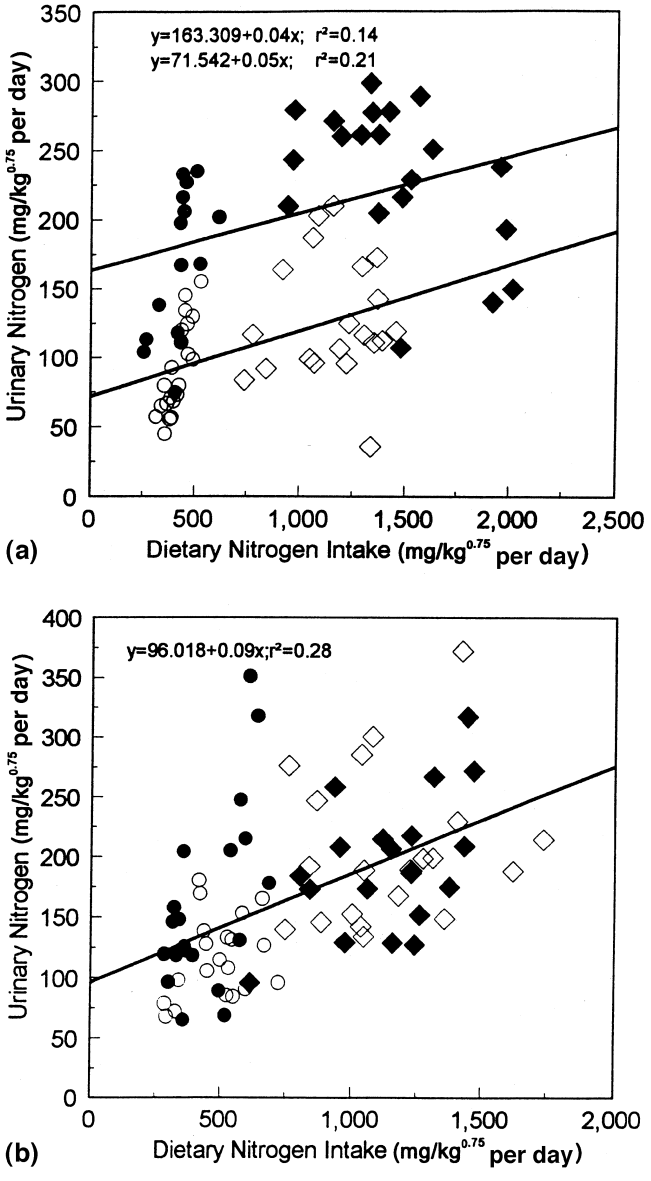


Fig. 4A,B Relationship between urinary N and dietary N intake in **A** *A. jamaicensis* ($n = 20$ per diet except for the low N/low E, $n = 15$) and in **B** *R. aegyptiacus* ($n = 20$ per diet). Symbols represent the four diets as in Fig. 1

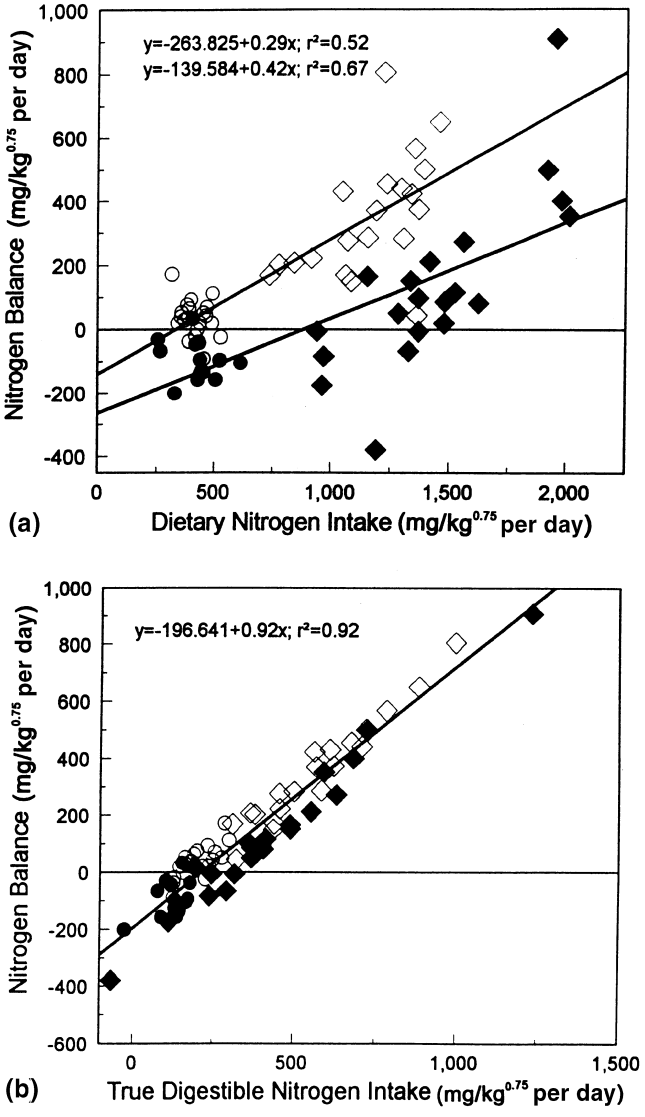


Fig. 5A,B Relationship between N balance and total dietary N intake **A** or truly digestible N intake **B** in *A. jamaicensis* ($n = 20$ per diet except for the low N/low E, $n = 15$). Symbols represent the four diets as in Fig. 1

Discussion

Food and E intake and digestibility

The striking feature of fruit bat feeding, whether we are referring to *A. jamaicensis* or *R. aegyptiacus*, is the large volume of food ingested and the high intake of metabolizable E. In this study, *R. aegyptiacus* ingested a mean of 0.8–1.0 times body mass and 1282.6–1545.2 kJ · $kg^{-0.75}$ · day^{-1} of metabolizable E across the four diets while *A. jamaicensis* ingested a mean of 1.1–1.6 times body mass and 1357.3–1767.3 kJ · $kg^{-0.75}$ · day^{-1} of metabolizable E across the four diets. These values for food intake are more or less comparable to those for other fruit and nectar-feeding phyllostomids and pteropodids (Table 3). Even though the bats were housed in small cages in this study and so could not fly, they still ingested quantities of fruit that were comparable to those reported for bats allowed to fly freely either in the wild or in a large flight cage (Morrison 1978; von Helverson and Reyer 1984; Thomas 1984; Table 3). Food intake thus appears to be independent of the level of locomotory activity, suggesting that intake is regulated by factors other than the energy expenditure associated with locomotion. The consequence of ingesting such a large food mass is an extraordinarily high E intake. Indeed, even though bats could not fly under the conditions imposed by laboratory ad lib feeding experiments (this study; Steller 1986; Delorme and Thomas 1996;

Table 3 Summary of food and E intake [gross E intake (*GEI*) and metabolizable E intake (*MEI*)], measured with different methods, in fruit and nectar feeding phyllostomid and pteropodid bats. Methods: (**1**) ad lib feeding in lab, (**2**) doubly labeled water, (**3**) estimated from field as extrapoled from lab estimates of diet composition

Species	Mass (g)	Food intake $g \cdot day^{-1}$ ($g \cdot kg^{-0.75} \cdot day^{-1}$)	GEI $kJ \cdot day^{-1}$ ($kJ \cdot kg^{-0.75} \cdot day^{-1}$)	MEI $kJ \cdot day^{-1}$ ($kJ \cdot kg^{-0.75} \cdot day^{-1}$)	Method	Source
C. perspicillata	18.5	26.8–30.1 (534.5–599.6)	67.2–97.4 (1339.5–1941.4)	58.6–79.1 (1168.7–1577.3)	1	Delorme and Thomas (1996)
A. jamaicensis	36.8	42.0–60.5 (499.9–720.1)	136.8–189.3 (1628.6–2253.5)	114.0–148.5 (1357.3–1767.3)	1	This study
A. jamaicensis	50.0	–	61.1 (578.3)	–	3	Morrison (1978)
A. jamaicensis	50.0	46.3 (437.7)	59.7 (564.5)	–	1	Morrison (1980)
A. caudifer	11.5	–	–	51.9 (1477.3)	2	Helverson and Reyer (1984)
M. pusillus	26.0	46.1–67.3 (712.8–1040.0)	64.9–92.5 (1002.2–1428.7)	–	1	Thomas (1984)
R. aegyptiacus[a]	144.0	88.9–96.2 (380.3–411.5)	185.9–222.5 (795.4–952.0)	164.9–209.0 (705.6–894.2)	1	Korine et al. (1996)
R. aegyptiacus	156.7	131.6–158.9 (528.6–637.9)	427.3–487.4 (1715.9–1957.2)	319.4–384.8 (1282.6–1545.2)	1	This study
E. buettikoferi	175.3	170.5–256.9 (629.3–948.2)	252.6–386.8 (932.6–1427.9)	–	1	Thomas (1984)
P. poliocephalus[b]	850.0	188.0 (212.5)	–	590.4 (667.0)	1	Steller (1986)

[a] Values are only for fruits with high water content

[b] Values are based on native fig diet

Korine et al. 1996), their E intake was still on the order of 2.3–4.9 times the basal metabolic rate (BMR). These E intakes are 20–164% greater than the mean eutherian maintenance E requirement, which is roughly two times the proposed Kleiber (1961) mean eutherian BMR of 293 $kJ \cdot kg^{-0.75} \cdot day^{-1}$.

Why do fruit bats ingest such large amounts of food and consequently of E ? Thomas (1984) suggested that obligate frugivorous bats faced a dietary constraint imposed by the low N content of their fruits and were forced to over-ingest E in order to ensure an adequate intake of N to meet their minimum requirements for maintenance. On the other hand, Delorme and Thomas (1996) concluded that *Carollia perspicillata* was not forced to over-ingest E to meet their N requirements. Deciding which of these two opposing analyses applies (i.e., deciding whether or not fruit bats over-ingest E) necessarily requires that we know what the minimum E requirements are. Maintenance E requirements are typically determined by regressing change in body mass against E intake to determine the E intake required for stable body mass. However, our studies of *Carollia perspicillata*, *A. jamaicensis*, and *R. aegyptiacus* (as well as Thomas' work on *Epomops buettikoferi* and *Micropteropus pusillus*) indicate that fruit bats regulate body mass independently of the mean daily E intake. Although it could be argued that the time-frame over which bats regulate body mass and E intake exceeds the 24-h measurement period imposed by most laboratory feeding studies, we found no evidence that bats either gained or lost body mass over the 7 days of confinement (3 pre-trial days and 4 feeding trial days). This necessarily means that the average daily E intake and expenditure were balanced.

To determine maintenance E requirements, it is essential to identify the threshold E intake below which bats are unable to maintain constant body mass. For at least two reasons, however, it appears unlikely that the E intakes that have been reported to date represent a true measure of maintenance E requirement. First, the maintenance E requirement represents the sum of BMR, digestive and absorptive costs (often refered to as specific dynamic action; SDA), and includes only limited locomotory costs. Although flight is energetically expensive, bats do not normally fly during the feeding trials. As a result, there is no reason to expect that maintenance E costs for bats should differ markedly from the mammalian mean of roughly $2 \times BMR$ (Maynard and Looslie 1962). BMR's have been reported for *Carollia perspicillata* (Audet and Thomas 1997), *A. jamaicensis* (Mc Nab 1969), and *R. aegyptiacus* (Korine and Arad 1993) as 1.9, 1.7, 0.95 ml $O_2 \cdot g^{-1} \cdot h^{-1}$, respectively, representing E expenditure of 338.0, 358.8, 288.1 $kJ \cdot kg^{-0.75} \cdot day^{-1}$, respectively. Depending on food intake for a given diet, mean daily metabolizable E intakes ranged from 1168.7 $kJ \cdot kg^{-0.75} \cdot day^{-1}$ to 1577.3 $kJ \cdot kg^{-0.75} \cdot day^{-1}$ for *Carollia perspicillata* ($3.4–4.7 \times BMR$), 1357.3 $kJ \cdot kg^{-0.75} \cdot day^{-1}$ to 1767.3 $kJ \cdot kg^{-0.75} \cdot day^{-1}$ for *A. jamaicensis* ($3.6–4.3 \times BMR$), and 1282.6 $kJ \cdot kg^{-0.75} \cdot day^{-1}$ to 1545.2 $kJ \cdot kg^{-0.75} \cdot day^{-1}$ for *R. aegyptiacus* ($4.4–5.4 \times BMR$). Although the large volumes of food ingested daily by fruit bats may result in an elevated SDA, as was indicated by Noll (1979) for *R. aegyptiacus*, it is unlikely that this digestive cost could account for the extraordinarily high intakes of E that we have reported. We calculated that the SDA for frugivorous bats would have to be 12 times greater than for other mammals before it could begin to explain the high metabolizable E intake that we have observed for fruit bats, yet the SDA associated with tropical fruit is not abnormally high (see Peters 1983). A second line of evidence suggests that the

high metabolizable E intakes that we observed are not a true measure of maintenance E requirement. Using the doubly labeled water (DLW) method, Kunz et al. (1998), von Helverson and Reyer (1984), Thomas (in Fleming 1988) and Bell et al. (1986) measured field metabolic rate (FMR) for *Phyllostomus hastatus, Anoura caudifer*, *Carollia perspicillata* and *Macrotus californicus* as 876.7–1051.6, 1477.3, 1543.5 and 592.2 kJ · $kg^{-0.75}$ · day^{-1} respectively, and from time-E budgets Morrison (1978) estimated FMR for *A. jamaicensis* as 449.3 kJ · $kg^{-0.75}$ · day^{-1}. These measures of FMR represent approximately 1.5–4.7 × BMR and, with the exception of *A. jamaicensis*, correspond with DLW estimates of FMR for birds and mammals. Figure 6 presents these measurements of FMR through the regression lines proposed by Nagy (1987) for DLW estimates of FMR for birds and mammals along with our lab data for comparison. This figure shows that our data for ad lib intake are similar to FMR values measured with DLW for free-ranging birds and nectar and fruit bats. Although in our laboratory conditions *Carollia perspicillata*, *A. jamaicensis* and *R. aegyptiacus* were unable to fly and so experienced no major locomotory costs associated with foraging or social interactions, they exhibited E intakes roughly equivalent to those experienced in the field and they did so without gaining weight. It might be argued that the high E intake and expenditure that we observed are an artifact induced by the stress and by the individual confinement of normally colonial bats. Widmaier and Kunz (1993) showed that handling and the isolation of *Pteropus hypomelanus* in small containers results in a massive increase in the secretion of the major glucocorticoids, indicating substantial stress. This may be the case for our bats, but we do not believe that this has a major effect on metabolic rate. If stress or solitary confinement did in fact result in artificially high metabolic rates, then this would have been noted in studies of BMR (or RMR) and thermal conductance. However, the BMR's reported for *Carollia perspicillata* (1.9 ml $O_2 \cdot g^{-1} \cdot h^{-1}$, Audet and Thomas 1997), *A. jamaicensis* (1.7 ml $O_2 \cdot g^{-1} \cdot h^{-1}$, Mc Nab 1969), and *R. aegyptiacus* (0.95 ml $O_2 \cdot g^{-1} \cdot h^{-1}$, Korine and Arad 1993) closely follow the mammalian mean once mass is controlled for. There is thus no evidence that stress or solitary confinement has a marked effect on metabolic rate and hence E requirements.

On the basis of the preceeding arguments, we suggest that the true maintenance E requirements for *Carollia perspicillata*, *A. jamaicensis* and *R. aegyptiacus* are substantially lower than the metabolizable E intakes that we observed in our feeding trials. In this case, these bats must regulate body mass independently of food and E intake. As Thomas (1984) concluded, regulation does not occur at the pre-absorption level because all our analyses were based not on gross E intake but rather on metabolizable (assimilated) E intake. This leaves only one possible route for regulating body mass in the face of highly variable E intake, and that is by the adjustment of metabolic rate. The most likely candidates offering a regulatory mechanism are peptides in the central nervous system and circulating hormones which are believed to play a major role in E consumption and expenditure in mammals. Rapidly accumulating evidence suggests that OB protein (also known as leptin), which is secreted from adipose tissue, plays an important role in the control of body fat stores not only through regulation of ingestion (e.g., Campfield et al. 1995; Halaas et al. 1995; Pelleymounter et al. 1995), but also through adjustments in E

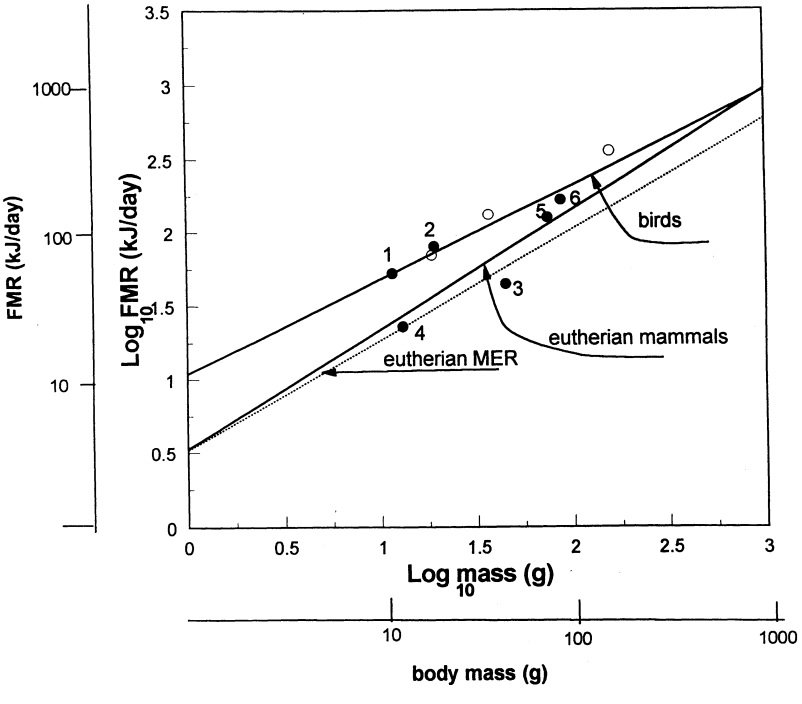


Fig. 6 Field metabolic rate (*FMR*) as a function of body mass in birds, eutherian mammals and bats. Data on the FMR of birds and eutherian mammals are derived from Nagy (1987). Eutherian maintenance E requirement (*MER*) (*dotted line*) represents 2 × BMR of Kleiber (1961). Species identification numbers are: *1 A. caudifer*, von Helverson and Reyer 1984; *2 Carollia perspicillata*, Thomas (in Fleming 1988); *3 A. jamaicensis*, Morrison 1978; *4 Macrotus californicus*, Bell et al. 1986; *5* and *6 Phyllostomus hastatus*, Kunz et al. 1998. Our lab data for *Carollia perspicillata*, *A. jamaicensis*, and *R. aegyptiacus* are indicated by *open circles*

metabolism (e.g., Halaas et al. 1995; Hwa et al. 1996). Recent physiological studies have also shown that leptin interacts with several neuropeptides such as neuropeptide Y and corticotrophin releasing hormone which have been implicated in food regulation of mice, rats and many other species (Kaiyala et al. 1995; Stephens et al. 1995; Schwartz et al. 1996). Brown fat has frequently been implicated in the regulation of fat stores and body composition, offering at least one of the metabolic pathways for unloading excess E through heat production (e.g., Rothwell and Stock 1979). Okon (1980) showed that the pteropodid bat, *Eidolon helvum*, had large deposits of brown adipose tissue and the lipid content underwent a sizeable circadian cycle indicating high metabolic activity. McNab (1976) also found seasonal variation in fat reserves of bats in tropical environments. While we have no direct evidence for high metabolic activity in brown adipose tissue of *Carollia perspicillata*, *A. jamaicensis* and *R. aegyptiacus*, the tight regulation of body mass in these bats despite considerable variation in E intake strongly suggests that variation in metabolic output (possibly implicating brown adipose tissue and/or leptin) is the basis for E management. All evidence seems to indicate that fruit bats do not modulate fruit intake precisely but rather are E maximizers, ingesting more E than is required and regulating storage by adjusting metabolic output.

If, for the sake of argument, we assume that maintenance E requirements of fruit bats follow the eutherian norm at roughly $2 \times$ BMR, then we can question whether bats could potentially regulate fruit and E intake at a lower level (the eutherian norm) and still balance their N requirement. In this case, *Carollia perspicillata*, *A. jamaicensis* and *R. aegyptiacus* would require 677.8, 716.4 and 576.2 $kJ \cdot kg^{-0.75} \cdot day^{-1}$, respectively, or the equivalent of 34.0, 60.2 and 143.5 $kJ \cdot day^{-1}$ of metabolizable E intake. This would require an intake of 2.4–11.2 g on our experimental diets, 2.4–10.7 g on a natural fig (*Ficus ovalis*) diet and 2.5–11.1 (dry mass) on *Piper amalago* (fruit data from Herbst 1986). To meet their maintenance N requirements of 442.0–529.8 $mg \cdot kg^{-0.75} \cdot day^{-1}$ or the equivalent of 22.2–132.0 $mg \cdot day^{-1}$, non-breeding *Carollia perspicillata*, *A. jamaicensis* and *R. aegyptiacus* would require an intake of 5.2–30.7 g on our experimental diets, 3.9–23.1 g on a fig diet and 2.2–13.2 g (dry mass) on *P. amalago* (Herbst 1986), and breeding females would require substantially more. This analysis shows that non-breeding bats would not be able to face shortfalls of 2.8–19.5 g on our low N diets, and 1.5–12.4 g on a *Ficus* diet. However, on N-rich fruit like *P. amalago,* only *R. aegyptiacus* would face shortfall of 2.1 g (dry mass). Thus, on the basis of the assumption that maintenance E requirements follow the eutherian norm, we conclude that fruit bats must necessarily over-ingest E if they feed exclusively on low-N fruit.

The fact that the phyllostomids, *Carollia perspicillata* and *A. jamaicensis*, and the pteropodids, *Epomops buettikoferi*, *Micropteropus pusillus*, and *R. aegyptiacus*, are all able to maintain health and body condition as well as to regulate body mass under captive conditions proves that insect or leaf supplements are not a fundamental nutritional requirement. These bats are clearly able to simultaneously adjust fruit intake to achieve N balance and to regulate metabolic output to avoid excessive mass (fat) storage. This would make phyllostomid and pteropodid fruit bats ideal candidates for the study of the role of leptin or neuropeptide Y in the regulation of body mass and metabolic output. However, it remains to be seen whether fruit bats face a problem of E over-ingestion in the field. Both pteropodid and phyllostomid bats do either occasionally or regularly ingest insects, pollen, and the liquid fraction of leaves (Law 1992; Bhat 1994; Kunz and Diaz 1995). By supplementing their protein intake when specific types of leaves, flowers, or insects are available, these bats may be able to dissociate N and E intakes and so escape the problem of an over-ingestion of E. This is the "mixed" strategy proposed by Thomas (1984, Fig.3). At this point, we do not know the exact nutritional role of leaf-eating, although Kunz and Diaz (1995) proposed that it is an adaptation specifically aimed at avoiding N constraint. If this is the case, then fruit bats that resort to leaf-eating (or supplement their protein intake by other means) should be able to simultaneously reduce their daily fruit and E intake. Under these conditions, we would predict that DLW studies will reveal FMR that are substantially lower than the E intakes and expenditures reported from laboratory studies where N and E intakes are not uncoupled.

In conclusion, our comparative analysis of the digestive efficiency and N metabolism of *A. jamaicensis* and *R. aegyptiacus* suggest that both species showed a similar degree of specialization. Fruit bats appear to be particularly efficient in limiting endogenous N losses in the feces. As was pointed out by Delorme and Thomas (1996), it is the low MFN level that allows fruit bats to survive on low-N diets. The results from the present study support the conclusion that, although phyllostomids have likely evolved a frugivorous habit more recently than pteropodids, *A. jamaicensis* do not exhibit a lower degree of specialization in digestive functions favoring efficient N extraction and retention.

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