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Seasonal changes in the diets of migrant and non-migrant nectarivorous bats as revealed by carbon stable isotope analysis

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Abstract. Three species of nectar-feeding bats migrate from tropical and subtropical Mexico into the Sonoran and Chihuahuan deserts during the spring and summer months. We examined geographic and seasonal changes in the diet of one migrant species, *Leptonycteris curasoae*, using carbon stable isotope techniques to determine the relative importance of C3 and CAM (Cactaceae, Agavaceae) plants in its diet. We also examined the diet of a non-migratory nectar-feeding bat, *Glossophaga soricina*, from southern Mexico using the same techniques. We found that *L. curasoae* feeds extensively or exclusively on CAM plants during migration and in the northern part of its range and feeds mostly on C3 plants in southern Mexico. This bat is a year-round resident on Baja California where it is a CAM specialist. The non-migrant *G. soricina* feeds mostly on C3 plants year-round. Phenological data suggest that certain species of columnar cacti and at least one group of paniculate *Agaves* on the Mexican mainland provide a spatio-temporally predictable nectar corridor along which nectarivorous bats may migrate in the spring and fall, respectively. Different flowering schedules of *Agaves* in Baja California appear to promote year-round dietary specialization and perhaps non-migratory behavior in nectar-feeding bats living there.

Key words: Nectarivory – Stable isotopes – Bats – Migration

Migrant flower-visiting birds and bats need nectar (and pollen) to fuel their migrations. We therefore expect the timing of migration to coincide with the blooming periods of suitable food plants. We also might expect nectar "corridors" – latitudinally broad paths of blooming plants pollinated primarily by migrant nectar-feeders – to exist between the northern and southern destinations of migrants. Such corridors are likely to exist, for example, along the migration routes of hummingbirds in the Sierra Nevada mountains of western North America (Grant and Grant 1966; Brown and Kodric-Brown 1979) but have not been explicitly demonstrated. Likewise, nectar corridors along the migration routes of other kinds of pollinators have not been documented.

Leptonycteris curasoae (formerly *L. sanborni*) (Phyllostomidae, Glossophaginae) is one of three species of North American nectarivorous bats (the others being *Leptonycteris nivalis* and *Choeronycteris mexicana*) that migrate from the tropics and subtropics of Mexico into the Sonoran and parts of the Chihuahuan deserts during the spring and summer and return south in the fall (Barbour and Davis 1969; Cockrum 1991). Winter populations of *L. curasoae* in southern Mexico feed on flowers from a variety of plants, including species in the Bombacaceae, Convolvulaceae, Leguminosae, as well as species in the Agavaceae and Cactaceae (Alvarez and Gonzalez 1970; Gardner 1977; Quiroz et al. 1986). Migrant populations in the north visit flowers in the Cactaceae and Agavaceae (Howell 1979; Howell and Roth 1981), but the degree to which they depend on these plants during migration is currently unknown. The diets of *L. nivalis* and *C. mexicana* are less well-known but also include many of the same kinds of plants visited by *L. curasoae* (Gardner 1977).

Because plants in the Agavaceae and Cactaceae use the CAM (Crassulacean acid metabolism) photosynthetic pathway and hence differ from C_3 plants (i.e. all other plants visited by *Leptonycteris*) in the ratio of $^{13}C/^{12}C$ in their tissues (Bender 1971; Osmond et al. 1973), it should be possible to determine the nutritional dependence of these bats on CAM plants by analyzing the carbon isotopic composition of their tissues (Tieszen et al. 1983). Here we examine seasonal changes in the degree of dietary specialization in *L. curasoae* using stable isotope techniques and relate this specialization to the blooming periods of plants along its potential migratory pathway. Our analysis reveals that at certain times of the year, this bat feeds heavily on flowers (and fruits in the case of cacti) of species of Cactaceae and Agavaceae whose

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blooming periods appear to form a latitudinally broad nectar corridor along the west coast of Mexico.

Materials and methods

We used muscle and skin tissue from a single toe from 110 museum specimens and 25 wild-caught individuals to determine the carbon isotopic composition of *L. curasoae* captured at locations ranging from northern Guatemala to southern Arizona (Appendix). For comparison, we also determined the carbon isotopic composition of two toes from 32 museum specimens of the smaller and more omnivorous non-migratory glossophagine bat *Glossophaga soricina* from tropical sites in southern Mexico.

Our laboratory analyses followed standard techniques (DeNiro and Epstein 1978). Bat toes (ca. 3 mg) were declawed and soaked in 1N HCL solution for 4 h to dissolve bone carbonate. Samples were then washed to neutrality and freeze dried. Whole toes were combusted at 800° C for 3 h in Vycor ampules with 1 g of cupric oxide and copper and a small amount of silver foil (ca. 50 mg). Carbon dioxide was cryogenically purified from combustion products in a vacuum system. Purified carbon dioxide was analyzed in a PRISM (VC) mass spectrometer. Isotope ratios are expressed using the δ notation in parts per thousand (per mil) where

$$\delta\ ^{13}C\ (‰) = \left[\frac{(^{13}C/^{12}C)\ \text{sample}}{(^{13}C/^{12}C)\ \text{standard}} - 1\right] \times 10^3$$

The standard for $\delta\ ^{13}C$ values is the Peedee belemnite marine limestone (PDB). Precision of isotopic measurements for organic samples was ±0.4% (SD).

We analyzed muscle tissue because Tieszen et al. (1983) showed experimentally that its rate of carbon turnover (half-life = 27.6 days in gerbils *Meriones unguiculatus*) is relatively high, which gives us a relatively short (ca. 2–3 mos) time window into the diet of each specimen. Carbon turnover rates are likely to be higher in wild *L. curasoae* than in captive gerbils, whose mass is twice that of the bat.

To determine the carbon isotopic composition of known or potential bat-visited plants, we analyzed flower tissue from 10 species of Mexican or Central American C_3 plants and 21 species of CAM plants, including six species each in Gentry's (1982) Deserticolae, Ditepelae, and Hiemiflorae groups of the subgenus *Agave*. Migratory nectar-feeding bats are likely to encounter flowering individuals in these three groups of *Agaves* in the spring, summer and fall, and winter, respectively (Fig. 2). C_3 plants included *Crescentia cujete* (Bignoniaceae); *Ceiba acuminata, C. aescufolia, C. pentandra, Ochroma pyramidale, Pachira aquatica, Pseudobombax ellipticum* (Bombacaceae); *Bauhinia pauletia, Calliandra haematocephala,* and *Inga vera* (Leguminosae). CAM plants included *Carnegia gigantea, Pachycereus pringlei, Stenocereus thurberi* (Cactaceae); Agavaceae – Deserticolae group: *A. cerulata, A. deserti, A. gigantensis, A. mckelveyana, A. moranii, A. sobria;* Ditepelae group: *A. chrysantha, A. colorata, A. delamateri, A. murpheyi, A. palmeri, A. shrevei;* Hiemiflorae group: *A. atrovirens, A. congesta, A. hiemiflora, A. hurteri, A. pachycentra,* and *A. seemanniana.* The cactus tissue was collected at Bahia Kino, Sonora, Mexico. The other plant tissue came from herbarium specimens.

Comparison of the carbon composition of flower tissue and nectar in three species of cacti indicated that the $\delta\ ^{13}C$ values of these tissues differed by less than 1‰, which indicates that the carbon composition of flower tissue closely reflects the carbon that bats actually ingest. We assume that the same situation holds for the other flower tissue we analyzed.

Results and discussion

The carbon isotopic composition of tissues of 111 individuals of *L. curasoae* from Guatemala, mainland Mexico, and the southwestern United States varied seasonally and latitudinally. Tissues had lower $\delta\ ^{13}C$ values in the late fall and winter than in the spring, summer, and fall (Fig. 1). $\delta\ ^{13}C$ values in these samples were significantly correlated with latitude at which the bats were captured ($r = 0.61$, $F_{1,109} = 64.7$, $P < 0.001$); $Y = -22.11 + 0.31X$). Based on $\delta\ ^{13}C$ values, northern populations of *L. curasoae* appear to feed exclusively on CAM plants, whose $\delta\ ^{13}C$ averaged -12.62 ± 0.25 (SE) (Fig. 1), in the summer and probably also during their northward (February–April) and southward (August–October) migrations. Southern populations appear to feed on a mixture of C_3 and CAM plants during the winter. $\delta\ ^{13}C$ of the C3 plants averaged -26.41 ± 0.54 (Fig. 1). A few summer specimens south of the Sonoran desert (for example, two from Jalisco, Mexico, in July) had relatively low values of $\delta\ ^{13}C$ (-18.5 and -19.6), indicating that they were also feeding on C_3 plants.

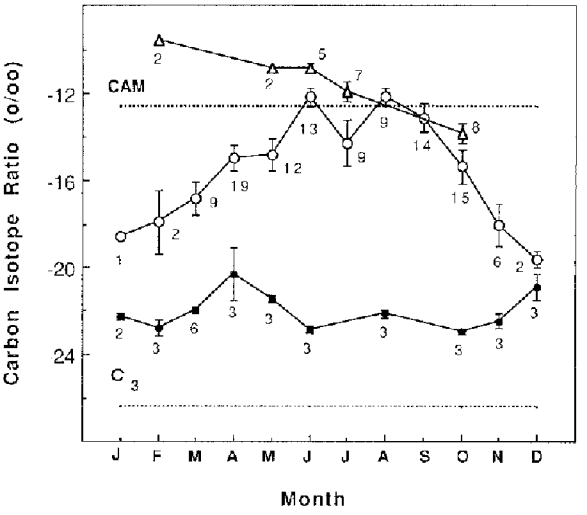

Fig. 1. Monthly values (mean ± 1 SE, sample size) of $\delta\ ^{13}C$ in muscle tissue from the bats *Leptonycteris curasoae* (*open circles* = mainland, open triangles = Baja California) and *Glossophaga soricina* (*closed circles*). The dashed lines labeled CAM and C_3 indicate mean values of plant tissues analysed in this study. Standard errors around these means are 0.21 for 21 CAM species and 0.54 for 10 C_3 species

Examination of individuals of *L. curasoae* captured in April and June 1989 and 1990 at Bahia Kino, Sonora, Mexico, supports our spring dietary scenario. Bats captured in April shortly after they had arrived from the south contained less CAM carbon and had a mean $\delta\ ^{13}C$ of -14.45 ± 0.55 SE ($n = 16$, range = -19.47 to -11.31) compared with bats captured in June, in which $\delta\ ^{13}C$ averaged -11.46 ± 0.41 ($n = 9$, range = -14.19 to -10.11). These means differ significantly ($t = 3.73$, d.f. = 23, P (one-tailed) = 0.0005).

In contrast to the mainland situation, strong seasonal changes in carbon composition were absent in 24 samples of *L. curasoae* from Baja California (Fig. 1). All of these samples had high $\delta\ ^{13}C$ values, although monthly means differed significantly (Kruskal-Wallis Anova: $X^2 = 14.9$, $P = 0.005$). These data suggest that the population there feeds exclusively on CAM plants throughout the year and may not be migratory, as evidenced by their year-

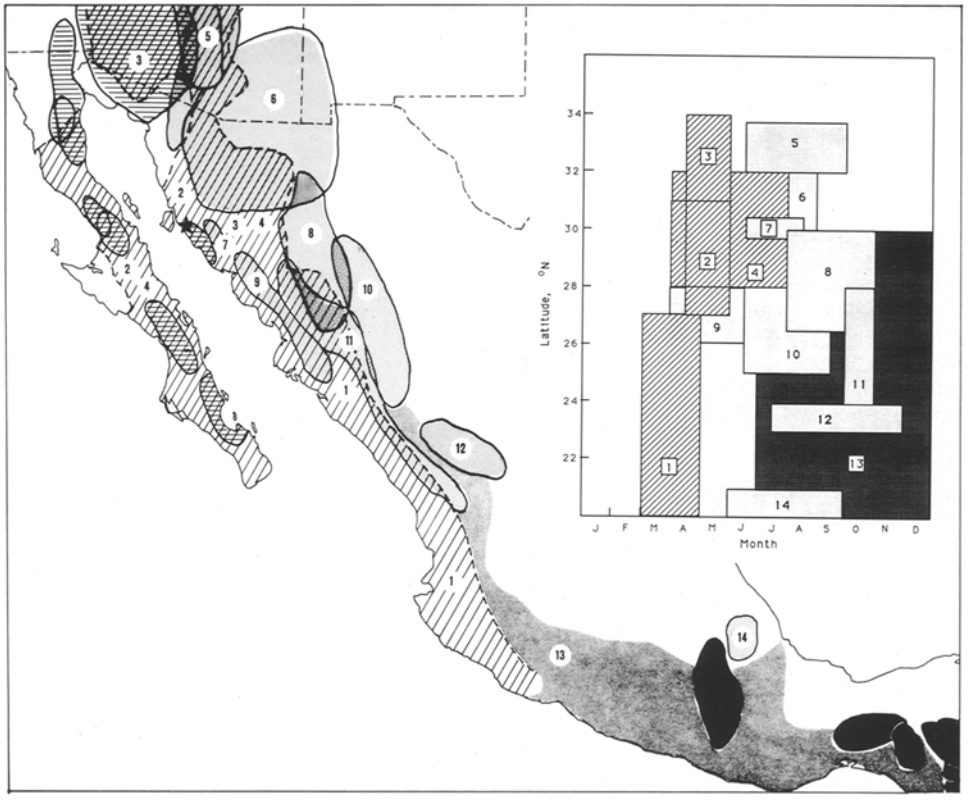

Fig. 2. The blooming periods and geographic distributions of four species of columnar cacti *(diagonal hatching)* and nine species of *Agave* in the Ditepelae group *(light stippling)* and one species in the Rigidae group *(dark stippling)* of Gentry (1982). Also shown are the ranges of species in the Deserticolae (horizontal hatching) and Hiemiflorae (black figures) groups of *Agaves*. Species are: 1. *Pachycereus pecten-aboriginum*, 2. *P. pringlei*, 3. *Carnegia gigantea*, 4. *Stenocereus thurberi*, 5. *Agave chrysantha*, 6. *A. palmeri*, 7. *A. fortidens*, 8. *A. shrevei*, 9. *A. colorata*, 10. *A. flexispina*, 11. *A. wocomahi*, 12. *A. durangensis*, 13. *A. angustifolia*, and 14. *A. applanata*. The star indicates the location of Bahia Kino, Sonora, Mexico. The inset is modified from Fleming (1992)

round presence in southern Baja California (Woloszyn and Woloszyn 1982).

In contrast to *L. curasoae*, tissues of the non-migratory *G. soricina* from southern Mexico were basically C_3 in their carbon isotopic composition year-round (Fig. 1). Although monthly values of $\delta\ ^{13}C$ varied significantly (Kruskal-Wallis Anova: $X^2 = 23.7$, $P = 0.005$), $\delta\ ^{13}C$ was not correlated with latitude in this species ($r = 0.084$, $F_{1,30} = 0.21$, $P = 0.65$). Pollen analysis reveals that in southern Mexico this bat visits flowers in the Bignoniaceae, Bombacaceae, and Leguminosae more frequently and flowers in the Agavaceae and Cactaceae less frequently than *L. curasoae* (Alvarez and Gonzalez 1970; Quiroz et al. 1986). It also eats insects and fruits of many C_3 plants during the tropical wet season (Heithaus et al. 1975; Quiroz et al. 1986).

The carbon composition of tissues of one migratory nectar- and pollen-feeding bat indicates that this species feeds extensively or exclusively on CAM plants (i.e. Cactaceae and Agavaceae) during migration and in the northern parts of its geographic range. Our preliminary data on *L. nivalis* ($n = 4$ specimens) and *Choeronycteris mexicana* ($n = 14$) show a similar pattern. These results suggest that there should exist a nectar corridor primarily composed of CAM plants that feed these bats as they move into and away from the Sonoran and Chihuahuan deserts. Data on the spatio-temporal distributions of flowering times of bat-visited cactus and *Agave* species (Fig. 2) are consistent with this expectation. In the spring, the corridor is comprised of at least four species of night-blooming columnar cacti plus *Agave colorata* which flower in northward progression along Mexico's dry west coast. Although the three northern cactus species have broadly overlapping flowering periods, detailed analysis at Bahia Kino in Sonora (Fig. 2) indicates that their flowering peaks are displaced; *Pachycereus pringlei* is the earliest blooming species and *Stenocereus thurberi* is the latest. Flowering peaks of these two species coincide with peak numbers of migrant bats (THF, personal observations).

In the fall, the nectar corridor likely consists of a series of species of *Agave*, primarily of the Ditepelae group which Gentry (1982) has postulated as being coevolved with bats. A comparison of the blooming times of Ditepelae *Agaves* with those of all species of the subgenus *Agave* based on herbarium data presented in Gentry (1982) indicates that the Ditepelae bloom significantly later in the year (with a peak in August compared with June and July for the entire subgenus) (Kolmogorov-Smirnoff two-sample test: $D_{max} = 0.271$, $P < 0.05$). This result is consistent with Gentry's coevolutionary hypothesis, but field studies are needed to determine whether *Agave* flowering peaks in Mexico correspond with peak numbers of migrant bats.

Our data on the carbon composition of *L. curasoae* from Baja California suggest that nectar-feeding bats feed on CAM flowers or fruit nearly year-round, including the winter months when *Agave* and cactus flowers are unavailable in northern parts of the mainland Sonoran desert. Available phenological data (Wiggins 1980) indicate that more species of *Agave* (four or five species) flower in December through March in Baja California than at other times of the year (zero to two species with a second peak occurring in June and July). Winter- and

summer-blooming *Agaves* plus spring-blooming and summer-fruiting cacti (*Pachycereus* and *Stenocereus*) thus appear to provide *Leptonycteris* bats with CAM resources during most of the year and may promote non-migratory behavior.

In conclusion, we propose that the flowering times of certain species of columnar cacti and of at least one group of *Agaves* on the Mexican mainland provide a spatio-temporally predictable nectar corridor and apparently promote seasonal dietary specialization in migratory glossophagine bats. This specialization has important conservation implications for the federally-endangered *L. curasoae* because elimination of plant populations along either leg of the corridor will negatively affect the bats and possibly the reproductive success of other plant populations. Our results also suggest that because they share the same specialized pollinators, certain species of Cactaceae and Agavaceae are effective mutualists (Waser and Real 1979) on a broad geographic scale. Finally, the flowering schedules of *Agaves* and columnar cacti on Baja California appear to promote year-round dietary specialization and perhaps nonmigratory behavior in nectar-feeding bats living there.

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Appendix

Month of capture and localities of specimens of bats analyzed in this study. Sample sizes are in parentheses

A. *Leptonycteris curasoae from Guatemala, mainland Mexico, and the southwestern United States*

January: Nayarit (1), Mexico; February: Colima (2), Mexico; March: Guatemala (3); Chiapas (2), Colima (2), Queretaro (2), Nayarit (1), Mexico; April: Guatemala (1); Chiapas (1), Oaxaca (1), Sonora (16), Mexico; May: Chiapas (4), Oaxaca (7), Tamaulipas (1), Mexico; June: Queretaro (4), Sonora (9), Mexico; July: Queretaro (3), Jalisco (3), Mexico; New Mexico (3), USA; August: Michoacan (2), Mexico; New Mexico (7), USA; September: Morelos (1), Jalisco (1), Nayarit (2), Sinaloa (2), Mexico; Arizona (4), New Mexico (4), USA; October: Guatemala (1); Chiapas (1), Colima (2), Nayarit (9), Sonora (2), Mexico; November: Guerrero (3), Nayarit (3), Mexico; December: Guerrero (2), Mexico.

B. *L. curasoae from Baja California*

All specimens came from the Cabo region of Baja California Sur except for three specimens taken in May on Isla Angel de la Guardia in Baja California Norte.

C. *Glossophaga soricina from southern Mexico*

January: Pueblo (2); February: Chiapas (3); March: Chiapas (3), Oaxaca (3); April: Chiapas (2), Oaxaca (1); May: San Luis Potosi (3); June: Veracruz (3); August: Chiapas (3); October: Chiapas (3); November: Chiapas (3); December: Chiapas (3).

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