

## BIPHASIC MASS GAIN IN MIGRANT HUMMINGBIRDS: BODY COMPOSITION CHANGES, TORPOR, AND ECOLOGICAL SIGNIFICANCE<sup>1</sup>

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**Abstract.** Body mass of migrant Rufous Hummingbirds (*Selasphorus rufus*) on refueling stopovers increased on average from 3.2 to 4.6 g over a period ranging from several days to 3 wk. In birds arriving with body masses below  $\approx 3.5$  g, the initial period of mass gain was very slow. This slow gain was not explained by energy costs associated with territory establishment or learning to secure food, since it occurred even in years when nectar resources were superabundant and territoriality was nearly nonexistent. Data on body composition indicate that mass gain up to  $\approx 3.5$  g was due to deposition of nonlipid body components, which we hypothesize to be proteins involved in rebuilding muscle catabolized during the last stage of the recent migratory flight. Following the initial phase of slow mass gain, an accelerating rise in body mass consisted entirely of lipid gain. On average, overnight mass loss decreased prior to migration, suggesting that nocturnal torpor facilitated lipid deposition. The slow phase of mass gain is a potentially important constraint on migrating hummingbirds, because if they deplete their fat stores and allow their body mass to fall below 3.5 g, they incur a substantial cost in terms of greatly increased time spent on the subsequent stopover.

**Key words:** California Sierra Nevada; fat deposition; fuel reserves; hummingbirds; mass gain; migration; migratory stopover; *Selasphorus rufus*; torpor.

### INTRODUCTION

Adaptive seasonal variation in body mass is perhaps most striking in migratory birds, some of which may nearly double their mass before initiating migration (e.g., Norris et al. 1957, Odum et al. 1961, Dowsett and Fry 1971, Fry et al. 1972, Mercier 1985, Jehl 1988). This variation in body mass is due at least in part to deposition and catabolism of fat, which serves as the principal source of energy for powering migratory flight (King and Farner 1965, Blem 1980, Dawson et al. 1983, Ramenofsky 1990). Fat is the best fuel for supporting long-distance migration: oxidation of fat yields more energy per unit mass than either carbohydrate or protein, and also yields a substantial amount of metabolic water, thereby helping to maintain water balance dur-

ing extended flight (Odum et al. 1964, Schmidt-Nielsen 1990).

Early studies suggested that nonfat body components of migrating birds remain in homeostasis down to some lower limit of body mass (Connell et al. 1960, Odum et al. 1964, Hicks 1967) and led to the claim that migratory birds are similar to airplanes in that “‘high octane’ fuel (that is, fat) is added to and used from pre-existing ‘tanks’ (tissue spaces) without appreciable change in the tissue structure of the body as a whole” (Odum et al. 1964:1037). However, more recent studies have demonstrated that lean dry body components vary in concert with lipid stores in some migrant species (Child 1969, Evans 1969, Fry et al. 1970, 1972, Evans and Smith 1975, Ward and Jones 1977, McLandress and Raveling 1981, Marsh 1984, Davidson and Evans 1988, Johnson et al. 1989, Gaunt et al. 1990, Piersma 1990, Ellis and Jehl 1991, Jenni-Eiermann

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and Jenni 1991). As a result of these discrepancies, the roles of different body components in migratory mass changes remain unclear and controversial (e.g., Baggott 1975, Piersma 1990).

Detailed analyses of energy acquisition and utilization, and of the resultant variation in body composition, are necessary for evaluating the different behavioral and physiological strategies of migration (Blem 1980, Walsberg 1990). Of the three major aspects of a migrant's energy budget, premigratory fuel storage, flight costs, and replenishment of fuel reserves en route, only the first two have received much attention. Unfortunately, data are rarely detailed enough to understand both the ecological and physiological processes taking place during en route stopovers (Lovei 1989). Many previous studies of mass gain by migrants have focused on changes in mean body mass in a population over time. Data from such studies may be confounded by seasonal variation in population composition and other sampling biases (e.g., Jehl 1988), and they generally provide little information about the ecophysiology of individual migrants. Other studies based on capture and recapture of individual birds during stopover periods provide more information on individual variation (e.g., Moore and Kerlinger 1987), but they are still constrained by uncertainty about when individuals arrive and depart.

We studied migrant Rufous Hummingbirds (*Selasphorus rufus*) for 11 summers (1976–1986) at a montane stopover site in the Sierra Nevada of California. Here, we present data that suggest that hummingbirds utilize only fat during migration unless body mass falls below  $\approx 3.5$  g. Nonfat substrate then is used. In replenishing these stores, there is a strong biphasic pattern of body mass gain, with the lean dry mass recovering very slowly, followed by a rapid rise in lipid. This pattern is most consistent with the earlier studies, but it also indicates that migrants do not always maintain homeostasis of nonfat body components during en route stopovers and the flights between them. We were able to detect and explain these patterns because our data set is uniquely powerful: we recorded body masses of unencumbered individuals throughout the duration of their stopovers, we know the departure dates of individuals, and we have body composition data over the range of premigratory masses.

#### NATURAL HISTORY AND METHODS

Each summer, Rufous Hummingbirds migrate through mountains of the western United States from their breeding grounds in the Pacific Northwest to their wintering grounds in Mexico (Grant and Grant 1967, Calder 1987). En route, individuals stop in meadows and in most years establish feeding territories based on flowers (Gass 1979, Hixon et al. 1983). Over a period averaging  $\approx 8$  d, they gain  $\approx 1.5$  g and resume southward migration (Carpenter et al. 1983).

Our study site was a 2-ha, stream-watered meadow

at the base of the Sierra Nevada (1700 m elevation), 27 km northwest of Bishop, California (37°30' N, 118°30' W). A large population of the hemiparasitic flowering plant *Castilleja linariaefolia* (Scrophulariaceae) is associated with the willows, grass, and sagebrush near the stream. *C. linariaefolia* begins flowering from early to late July, and the first Rufous Hummingbirds arrive from mid-July to mid-August. As they refatten and resume migration, they are continually replaced by new arrivals into September.

During the summers from 1980 to 1986, we mist-netted hummingbirds at  $\approx 15$  sites throughout the meadow. Nets were opened from 0530 until mid-morning and again during the last 2 h of daylight. Each captured bird was weighed to the nearest 0.1 g with a Pesola balance and color-marked individually with three small strips of surveyor's flagging superglued to the skin of the back. These strips did not seem to bother the birds, nor did they affect body mass measurements or behavior (Carpenter et al. 1983). Every day we walked the length of the meadow with binoculars, identifying all individuals, marking their approximate territory boundaries on a map, and determining which individuals had resumed migration. Resumption of migration was conspicuous by several criteria: birds became obviously fat one to several days before disappearing from the site; birds often relaxed territorial defense the day before resuming migration; birds fed unusually heavily the evening before disappearing from the site; and some birds returned to their territories for 1–2 h of heavy feeding in the morning and then were observed to fly southward out of the meadow.

We recaptured and reweighed from 3 to 22% (depending on year) of our marked individuals. Each year, several individuals were selected for intensive behavioral observation and experimentation. These birds were weighed by means of an electronic artificial perch-balance (Carpenter et al. 1983; precision 0.01 g) throughout each of five 1-h observation periods per day over several days until they resumed migration. This technique provided detailed data on individuals without the potential problem of handling trauma affecting masses, and it also allowed us to measure nocturnal mass loss. As part of our standard protocol, we took continual measurements of body mass and behavior from 1830 to 1930 Pacific Daylight Time (PDT). Some individuals took unusually large meals just before going to roost, but before the end of the observation period (usually during the last 10 min). In these cases, we selected the final mass measurement prior to the onset of large meals as the final evening mass. Otherwise, if mass measurements were stable during the last hour, then the measurement closest to 1920 was selected as the final evening mass. When mass measurements fluctuated, as during breezy weather, we averaged measurements over a 5–10 min period. In the morning, body mass gain was often rapid before 0640 (possibly due to filling the crop to high levels to

facilitate rehydration), so we selected the first stable measurement after 0640. Therefore, we estimated overnight mass loss as the difference between the 0640 dawn mass and the 1920 mass of the previous dusk.

Selected individuals were time budgeted during 1-h observation periods beginning at 0630, 0930, 1230, 1530, and 1830. Full details of the procedures are given in Hixon and Carpenter (1988). Three timed variables contributed to foraging time: (1) nectar-feeding on the territory or in undefended areas; (2) thieving nectar from neighbors' territories; and (3) hawking insects (generally <30 s/h). Foraging time (i.e., the percentage of time spent foraging) was calculated for each observation period as the total foraging time divided by the total time during which the bird was in view. Average daily foraging time was calculated as the mean of the values obtained during the five observation periods. Incomplete budgets (<five observation periods) were obtained for some birds on some days, but because of diel cycles in the birds' behavior, the analyses reported here only consider days for which budgets were available during all five periods.

In 1976, we collected 18 Rufous Hummingbirds by shotgun from other meadows within a 64 km (40 mile) radius of the study area. Each carcass was immediately weighed to give fresh wet mass, and livers were dissected out and weighed. Entire carcasses were dried at 60°C until no further mass loss occurred and were reweighed, and the difference was taken as water content. Finally, lipid content was determined using Soxhlet ether extraction (Morton et al. 1974), and fat-free dry mass was obtained by subtraction. These birds ranged in fresh wet mass from 2.9 to 4.5 g. The initial masses of birds arriving at our meadow were as low as 2.8 g, and mean masses the night before migration ranged over the years from 4.5 to 4.8 g. Therefore, the collected birds spanned virtually the entire range of premigratory masses.

#### Statistical methods

Initial examination of data on mass gain and body composition suggested transitions in patterns at  $\approx 3.5$  g. We therefore explored several possible statistical descriptions of body mass as a function of time before migration, and body component masses as functions of body mass. The models we used were:

- 1) linear  $Y = c + bX$ ,
- 2) exponential  $Y = c + be^{ax}$ ,
- 3) piecewise linear  $Y = c_1 + b_1X$   
( $X \leq \text{threshold}$ )  
 $Y = c_2 + b_2X$   
( $X > \text{threshold}$ ),

We also examined the following combination of linear and exponential models as a possible description of mass gain:

- 4) piecewise hybrid  $Y = c_1 + b_1X$   
( $X \leq \text{threshold}$ )  
 $Y = c_2 + b_2e^{ax}$   
( $X > \text{threshold}$ ).

Linear models were fitted using ordinary least squares regression. Other models were fitted using the nonlinear estimation program in SYSTAT (Wilkinson 1990). The threshold point was included as one of the parameters to be estimated in piecewise models. In piecewise models of mass gain, where our sample size is very large, we forced continuity at the threshold point by including an appropriate term in the penalty function. We did not impose continuity restrictions at threshold points in piecewise models of body components, because sample sizes are relatively small, and we wished to avoid depleting degrees of freedom in statistical tests.

We used the adjusted  $R^2$  (denoted  $R^2_{\text{adj}}$ ) as a criterion for model selection (Glantz and Slinker 1990). The value of  $R^2_{\text{adj}}$  for a particular model indicates the expected proportion of variance in the dependent variable that would be accounted for by the predictor when applying the model to a new sample from the same population of data.  $R^2_{\text{adj}}$  is similar to the usual  $R^2$  in regression, but incurs a penalty in the form of lost degrees of freedom as more variables or parameters are included in the model. Approximate  $F$  tests (Glantz and Slinker 1990) were used to test whether the selected models significantly ( $P < .05$ ) reduced the unexplained variance.

Data on foraging time were examined for trends using linear regression, with time before migration (days) as the independent variable.

## RESULTS

### Mass gain

Gain of body mass after arrival in the meadow accelerated through time (Fig. 1). This pattern occurred despite the fact that, on average, the time spent feeding decreased through time (percent foraging time =  $17.79 + 0.65 \cdot [\text{days before migration}]$ ,  $N = 96$ ,  $r^2 = 0.09$ ,  $P = .002$ ). Birds gained mass slowly before reaching  $\approx 3.5$  g, but rapidly thereafter. The pattern held both for the entire sample (Fig. 1A) and for individual birds (Fig. 1B), and was similar in all years (Fig. 1C), despite the fact that ecological and behavioral conditions varied tremendously. In particular, food was so abundant in 1982 and part of 1983 that Rufous Hummingbirds arrived heavier and were effectively nonterritorial (Carpenter 1987), unlike in the other, highly competitive years. Nonetheless, the few individuals in those years that arrived below 3.5 g tended to show the same slow initial rate of mass gain (Fig. 1C), despite the absence of competition for food resources.

Based on the criterion of  $R^2_{\text{adj}}$ , the exponential and piecewise hybrid models provided identical fits (Table 1). Both models significantly reduced the unexplained

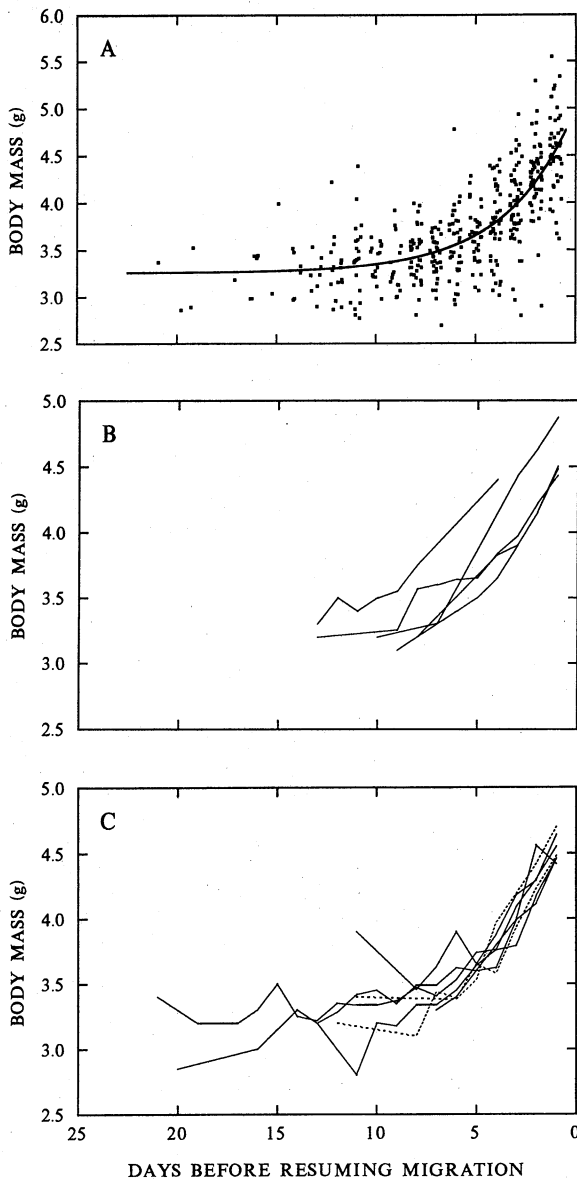


FIG. 1. Changes in body mass over the days preceding resumption of migration. (A) Data for all individuals and all years combined. Only body masses measured during the afternoon and evening are included. When more than one measurement was made on an individual in a given day, the last "good" measurement was used (details in *Natural history and methods*). A small amount of uniform random error has been added to all data points to avoid overlap in the figure. Reflecting the fact that most individuals spent <10 d on the stopover, sample sizes for days 1 through 21 (right to left on abscissa) are as follows: 46, 51, 53, 42, 33, 36, 35, 35, 15, 15, 21, 16, 8, 6, 2, 5, 1, 0, 2, 1, 1. The curve superimposed on the data is the best fitting exponential model (body mass =  $3.26 + 1.75 \cdot e^{(-0.297 \cdot [\text{days before migration}] )}$ ,  $R^2_{\text{adj}} = 0.57$ ). (B) Five typical individuals. (C) Seven years (1980–1986) plotted separately. The lines connect the mean body masses of the measured population on each day prior to migration. The 2 yr when food resources were most abundant (1982, 1983) are indicated by  $\cdots$ ; other years are indicated by  $\text{—}$ .

TABLE 1. Values of  $R^2_{\text{adj}}$  for the best fitting models of changes in body mass, body composition, and liver mass in Rufous Hummingbirds. Piecewise hybrid models were not applicable (NA) to body composition data.

Model*	Dependent variable				
	Body mass†	Water mass‡	Lean dry mass‡	Fat mass‡	Liver mass‡
Simple linear	0.44	0.57	0.27	0.83	0.69
Exponential	0.57	0.65	0.22	0.85	0.65
Piecewise linear	0.56	0.65	0.42	0.89	0.72
Piecewise hybrid	0.57	NA	NA	NA	NA

\* Equations are shown in *Statistical methods*.

† Independent variable is number of days before migration is resumed, during a refueling stopover.

‡ Independent variable is body mass.

variance in the body mass data (approximate  $F$  tests,  $P < .001$ ). The estimated threshold point in the best fitting piecewise hybrid model was 8.5 d before migration, corresponding to a body mass of 3.39 g. The fitted exponential model is shown in Fig. 1A. Both models indicated a relatively slow rate of increase at low body masses, with mass gain accelerating rapidly thereafter.

#### Body composition

Piecewise linear models provided best fits for two of the body components and for liver mass, and tied with the exponential model for water mass (Table 1). In all four cases, approximate  $F$  tests indicated that piecewise models significantly reduced the unexplained variance ( $P < .05$ ). The fact that neither linear nor exponential models outperformed the piecewise model for any component strongly indicates a biphasic pattern in the data. Estimated thresholds for the three body components (water, nonfat tissue, fat) and liver mass were 3.70, 3.50, 3.50, and 3.34 g, respectively (Table 2). Because of the close proximity of these values, and because the mean threshold for the three body components (3.57 g) is close to the point of transition identified in the pattern of overall body mass gain (3.39 g), it seems likely that the shifts in the patterns of mass gain and body composition reflect a common "critical mass" where an important transition in substrate metabolism occurs. The slight differences among the statistically identified break points probably reflect sampling and estimation errors, so we will assume that the threshold point is  $\approx 3.5$  g.

Based on these results, we apply the terms "Phase 1" to the relatively flat part of the curves in Fig. 1 up to  $\approx 3.5$  g, and "Phase 2" to the steeply rising part after 3.5 g. During Phase 1, both water and fat-free dry mass increased as body mass increased, but no lipid accumulated (Fig. 2A–C, Table 2). In contrast, during Phase 2 water and fat-free dry mass showed no consistent changes, but lipid accumulated (Fig. 2A–C, Table 2). That is, lipid alone accounted for the rapid rise in body mass in the accelerating phase of the curves in Fig. 1.

TABLE 2. Piecewise regressions of body components and liver mass on fresh wet body mass for migrating Rufous Hummingbirds during a refueling stopover. The "thresholds" given are the values that maximize  $R^2_{adj}$  (Table 1). Sample sizes are as in Fig. 2. Symbols: \*\*\*  $P < .001$ ; \*  $P < .05$ ; NS = not significant.

Dependent variable	Threshold (g)	Regression equations and statistics	
		Body mass $\leq$ threshold	Body mass $>$ threshold
Water mass	3.70	$Y = 0.52 + 0.52 \cdot X$ (***)	NS
Lean dry mass	3.50	$Y = -0.24 + 0.31 \cdot X$ (*)	NS
Fat mass	3.50	NS	$Y = -2.36 + 0.79 \cdot X$ (***)
Liver mass	3.34	NS	$Y = -0.07 + 0.06 \cdot X$ (*)

Overnight mass loss

On average, overnight loss of body mass decreased over time during Phase 2 of the refueling stopover. This was true both for individual birds, and for pooled data on many individuals (Fig. 3).

DISCUSSION

Fat deposition and the use of torpor

In contrast to the pattern observed in Phase 1, a steeply accelerating accumulation of lipid was evident during Phase 2 of mass gain. Fig. 2D suggests that

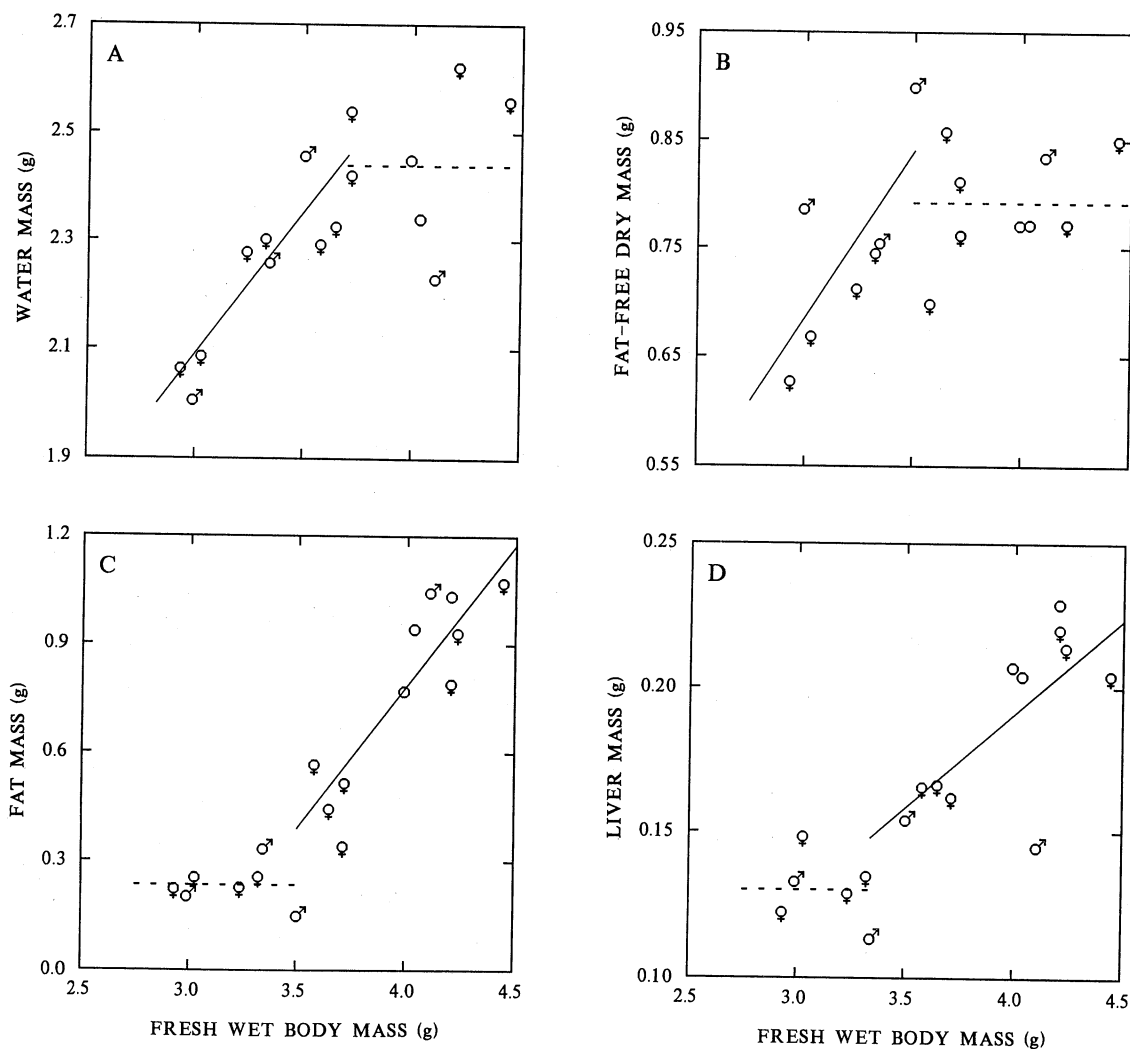


FIG. 2. Variation in (A) water mass, (B) fat-free dry mass, (C) fat mass, and (D) fresh liver mass, as a function of variation in fresh wet body mass. Each point within one figure represents a different bird. The sex of each bird is represented by standard symbols; ○ indicate birds of unknown sex. Significant regressions (Table 2) are indicated by —.

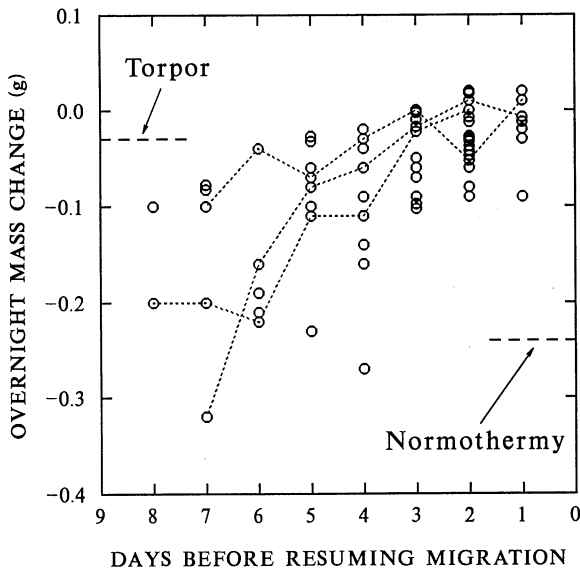


FIG. 3. Overnight mass loss in relation to time before migration. On average, Phase 2 begins 8–9 d before migration (Fig. 1, Table 2). Data from all years and all individuals are combined, representing 23 individuals with a mean of 2.8 measurements per individual. Data for three individuals with seven data points each are indicated by dotted lines. The horizontal dashed lines labeled “Normothermy” and “Torpor” indicate the theoretical expectations for overnight mass loss by normothermic and fully torpid birds in sheltered roosts, as calculated by Carpenter and Hixon (1988). A small amount of error ( $\pm 0.0025$  g) has been added to overlapping data values in the figure so that sample sizes are visually evident. Positive overnight mass changes represent experimental error.

$\approx 10\%$  of the gain could be accounted for by deposition of lipid in the liver. The rest is probably stored in adipose tissue (Johnston 1970): conspicuous fat depots develop in the birds’ furculum and subcutaneously over the entire body.

The positive correlation between time before migration and foraging time indicates that the pattern of mass gain in Phase 2 was not simply due to variation in foraging behavior. Instead, the acceleration in mass gain apparently resulted at least in part from an increasing use of torpor for fat conservation. Under the assumption that overnight mass loss is inversely proportional to the amount of time spent in torpor, we can use laboratory measurements of oxygen consumed overnight with and without torpor at different temperatures (Hiebert 1990) to calculate the mass birds should have lost overnight at our field temperatures. If a Rufous Hummingbird spent all night normothermic in a sheltered roost, it should have used  $\approx 0.24$  g of fat, whereas if it spent all night torpid in a sheltered roost, it should have used only 0.03 g (calculations in Carpenter and Hixon 1988).

The average bird began Phase 2 losing  $\approx 0.2$  g overnight, and therefore most individuals were probably using little or no torpor. Over the next several nights,

apparently more birds used torpor and spent successively greater proportions of the night in torpor. All birds appear to have spent at least some part of the night torpid during the three nights prior to the resumption of migration. On the final night before leaving, birds lost an average of only 0.02 g overnight, nearly identical to the value that we calculated they should have lost if the entire night were spent torpid.

A possible alternative explanation for the progressive reduction in overnight mass loss is that the increasingly thicker fat layer acts as an efficient insulator, reducing overnight heat loss. Gradually improving insulation might lead to a gradual reduction in overnight mass loss as a bird fattens, even if it remains normothermic at night. However, whole-body oxygen consumption of fat (4.5 g) and lean (3.3 g) birds differs by only 1% when the birds are normothermic, and oxygen consumption during torpor is 10% higher in fat birds than in lean ones (Hiebert 1989). Thus, reduction in mass-specific oxygen consumption in fat birds cannot account for reduction in overnight mass loss relative to lean birds. Furthermore, direct observations demonstrate that very fat birds in fact do go torpid part or all of the night (Carpenter and Hixon 1988, Hiebert 1990).

Thus, although our data for torpor are indirect, we hypothesize that Rufous Hummingbirds may wait until their internal conditions are minimally appropriate for migrating before using torpor as a means to conserve and increase fat. The determining internal condition may involve the completion of muscle rehabilitation, as discussed below.

#### *Source of the nonfat dry mass changes*

Water and some nonfat component are accumulated during Phase 1 of premigratory mass gain (Fig. 2A, B, Table 2). The anabolic process taking place during Phase 1 apparently is slow, requiring up to 2 wk in some individuals who arrived with a body mass much below 3.5 g (Fig. 1). Furthermore, this process apparently is completed before fat deposition begins.

The identity of the lean dry mass is unknown, but glycogen and protein are obvious candidates. In contrast to migratory flight, the very high ATP requirements of hovering, which is used extensively for feeding during stopovers, are supported largely by the oxidation of carbohydrate (Suarez et al. 1990). Therefore, it seems plausible that the first stage in a migratory stopover could be devoted to replenishing glycogen stores. Anabolism of glycogen is thought to require hydration at rates varying from 2.7 to 4 g water to 1 g glycogen (McBride et al. 1941, Olsson and Saltin 1970, but see Sherman et al. 1982). The observation that our leanest birds gained 400 mg in water (Fig. 2A) suggests that all of the variation in lean dry mass (up to 180 mg; Fig. 2B) might be accounted for by glycogen. However, glycogen is unlikely to account for the mass

gain during Phase 1, because liver mass did not change over the 2.9–3.5 g range (Fig. 2D), and because only limited amounts of glycogen have been found in the muscle of free-living migratory birds and other vertebrates (<2% of wet muscle mass; e.g., Farner et al. 1961, King et al. 1963, Vallyathan and George 1964, John and George 1965, George and Chandra-Bose 1967, Hultman 1967, Gollnick et al. 1972, Kerem et al. 1973, Marsh 1983). For example, Suarez et al. (1990) estimated that  $\approx 5$  mg of glycogen occurs in 1 g of hummingbird flight muscle. It therefore seems more likely that the fat-free dry mass gained by lean birds consisted primarily of protein. This conclusion is also supported by a trend for birds to spend more time hawking insects (their principal source of amino acids for protein building) when lean than when fat (F. L. Carpenter, *unpublished data*).

Several hypotheses have been proposed to account for mass changes in migrants involving protein. First, the flight muscles of some species may undergo an adaptive hypertrophy to accommodate the increased power demands of flight caused by the elevation of body mass (Evans 1969, Fry et al. 1972, Marsh 1984). If such muscle hypertrophy occurs, it is presumably regulated by a "feedback mechanism involving the dynamic load placed on the muscles during flight" (Dawson et al. 1983:R759). A second possibility involves the requirement for a supply of amino acids for general structural repair and maintenance. Protein reserves within the body may have to be utilized in order to meet dietary requirements during the obligatory period of fasting associated with long-distance flight (Evans and Smith 1975, Ward and Jones 1977, Piersma 1990, Piersma and Jukema 1990). The protein source may be hypertrophied flight musculature (Ward and Jones 1977) or other, more labile, protein reserves (Kendall et al. 1973). Third, regulation of substrate metabolism may result in flight muscle catabolism by migrants. Dawson et al. (1983) suggested that there may be a trade-off between carbohydrate balance and protein balance during long-distance flights. To balance tissue uptake of blood glucose (and avoid hypoglycemia or fatigue), an input of carbohydrate from elsewhere is required. Since the liver only contains small stores of glycogen, gluconeogenesis from glycerol or amino acids is probably required. Uptake of glucose at higher rates than can be supplied from glycerol may therefore necessitate the catabolism of muscle proteins to supply amino acids (Daniel et al. 1977, Dawson et al. 1983).

The hypotheses outlined above all predict that changes in fat and protein stores should be concurrent rather than sequential during migration. A more parsimonious explanation for the patterns we observed in Rufous Hummingbirds is that these birds frequently deplete their energy stores while en route. Although fat is the primary source of energy for powering migratory flight, if storage fat is depleted, then nonfat tissue must be burned as an emergency measure (Odum et al. 1964).

This nonfat tissue might involve flight muscle, which has high levels of proteolytic enzymes (Swain 1992). Degraded muscle tissue would presumably have to be rebuilt or repaired during the early stage of stopover before fat redeposition could begin. Depletion of fat reserves is therefore consistent with our observation of sequential patterns of variation in fat and nonfat dry body components.

Studies of body composition changes in sedentary animals undergoing starvation frequently indicate that fat and protein are utilized concurrently (e.g., Entenman et al. 1958, Cherel et al. 1988, Groscolas et al. 1991). However, protein degradation during endurance exercise is poorly known (Goldspink 1991), and there is no reason to assume that the physiological changes occurring during fasting in sedentary and exercising animals must be identical (cf. Shephard 1982).

As far as we are aware, use of nonfat tissue for migratory fuel following the depletion of fat stores en route has not previously been documented in detail. However, examination of migrant birds following their completion of long-distance flights across major ecological barriers often suggests that breast muscle has been catabolized (e.g., Miller 1963, Rogers and Odum 1966, Kuenzi et al. 1991). For example, in their study of North American migrants following trans-Gulf migration, Rogers and Odum (1966:418) noted that in some of the very light birds, "the breast muscles seemed reduced so that the keels of the sternums protruded noticeably." Our data together with these anecdotal reports suggest that, during migration, muscle has a homeostatic role involving the provisioning of substrate for combustion during energy emergencies. This conclusion is in accord with recent studies of brood-rearing passerines, which demonstrated a role of flight muscle protein in fuel homeostasis during fasting (Swain 1992).

The slow rate of mass gain by Rufous Hummingbirds during Phase 1 may be due to a constraint on the dietary availability of amino acids to these nectar-feeding birds, to a higher energy cost of synthesizing protein than fat, or to other, as yet unknown factors. Dietary availability of amino acids is probably not the explanation since insects were abundant at our study site. Furthermore, the few data we have on birds hawking for insects before and after reaching 3.5 g suggest that birds spent more time flycatching when they weighed <3.5 g (F. L. Carpenter, *unpublished data*).

On the other hand, we can discount the possibility that lean birds simply devoted less time or effort to foraging. We found that foraging time tended to decrease as birds became fatter, and other studies have suggested that lean migrants may have a greater impulse to acquire energy than fat birds (Mascher 1966, Loria and Moore 1990, Kuenzi et al. 1991). Although probably physiological in nature, the precise reason for the slow rate of mass gain in lean birds remains unexplained, and should be the subject of future study.

*Ecological significance*

Our results have general implications for understanding the migratory strategies used by birds. First, we found that virtually all mass gain above 3.5 g was due to lipid deposition. This pattern of body composition change is consistent with Odum et al.'s (1964) historically influential hypothesis that nonfat body components of migrating birds remain in homeostasis, down to some lower limit of body mass. Those workers suggested that depletion of storage fats occurs at a fat index (ratio of fat mass to nonfat dry mass) of 0.2 to 0.3; below this level, nondepot fat and/or nonfat tissue must be catabolized. Consistent with this hypothesis, Fig. 2 indicates that the critical mass of  $\approx 3.5$  g in Rufous Hummingbirds is associated with a fat index of  $\approx 0.35$ .

Migration often occurs during times of unfavorable or unpredictable weather (Gass and Lertzman 1980) and intense competition for food resources (Rappole and Warner 1976, Kodric-Brown and Brown 1978, Gass 1979, Moore and Yong 1991), and it is generally a time of high mortality in birds (Spendelow 1985, Lindstrom 1989, Alerstam 1990). Therefore, natural selection should favor the ability to deposit fat efficiently and regain body mass rapidly during refueling stopovers (Alerstam and Lindstrom 1990). While some studies have presented data suggesting that migrants gained mass rapidly over the duration of their stopover (Cherry 1982, Bairlein 1985, Moore and Kerlinger 1987), other studies have documented an initial period of loss or maintenance of body mass by migrants for one to several days following their first capture (and presumed arrival) at the stopover site (Davis 1962, Gladwin 1963, Nisbet et al. 1963, Mascher 1966, Mueller and Berger 1966, Page and Middleton 1972, Langslow 1976, Rappole and Warner 1976, Mehlum 1983a, Biebach et al. 1986). Previous workers have hypothesized that low rates of mass gain during the early stages of migratory refueling stopovers are caused by a delay in the onset of hyperphagia in newly arrived migrants (Mascher 1966, Langslow 1976), costs associated with territory establishment (Davis 1962, Rappole and Warner 1976, Mehlum 1983a, b), or handling trauma (Mueller and Berger 1966). Our results suggest an alternative explanation for variation in mass gain by Rufous Hummingbirds, namely, that migrants suffer a period of slow mass gain upon stopping only if they need to restore nonfat body components degraded during the preceding migratory flight.

Some workers have argued that catabolism of surplus flight muscle for fuel may be adaptive in that it reduces the power output for flight and extends the maximum range of a migratory flight (Pennycuik 1975, 1978). Our data do suggest that some individuals may catabolize muscle toward the end of a long-distance migratory flight. However, such a strategy imposes a potentially severe constraint on the subsequent rate of

refattening, which entails costs in terms of a delay in the resumption of migration, and therefore probably represents an emergency measure resorted to only when all nonessential fat stores have been depleted.

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NOTE ADDED IN PROOF—While this paper was in press, Å. Lindstrom and T. Piersma ("Mass changes in migrating birds: the evidence for fat and protein storage re-examined," *Ibis* **135**:70–78), showed that individual variation in structural size, reserve levels, or timing of storage can cause body composition data for a population to exhibit a purely artifactual increase in fat-free mass. Their model predicts apparent deposition of fat-free tissue along with fat throughout the range of body mass gain, even if only fat is being deposited in reality. Their model has merit, but we think it does not apply to our data because our analyses suggest that mass gain in Rufous Hummingbirds above 3.5 g consists entirely of fat, and all gain below 3.5 g is non-fat.