Altitude and temperature effects on the energetic cost of hover-feeding in migratory rufous hummingbirds, *Selasphorus rufus*

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Abstract: During migratory stopovers, rufous hummingbirds (*Selasphorus rufus* (Gmelin, 1788)) can achieve high daily rates of net energy intake and mass gain while foraging at a range of elevations and ambient temperatures, despite the high energetic costs of hovering flight and thermoregulation. To gain insights into the factors affecting the energetic costs incurred during foraging, we captured migratory hummingbirds and measured their oxygen consumption rates during hover-feeding. Measurements were performed in situ where rufous hummingbirds forage as they migrate at several locations along a gradient in elevation and over the range of ambient temperatures normally experienced. Oxygen consumption rates during hover-feeding varied between the sexes and between juveniles and adults. These differences appeared to reflect differences in the power requirements for hovering flight in relation to variation in wing morphology. Decreasing ambient temperature and increasing elevation both significantly increased oxygen consumption rate during hover-feeding. The effects of these two environmental variables were additive, suggesting that hummingbird thermoregulatory requirements were not met by the additional heat produced by the higher metabolic rate necessary to support hovering flight at higher elevation. These results provide insight into the ways different foraging strategies may allow hummingbirds to maximize net energy intake.

Résumé : Durant leurs arrêtes migratoires, les colibris roux (*Selasphorus rufus* (Gmelin, 1788)) peuvent réussir à atteindre des taux quotidiens nets d'accumulation d'énergie et des gains de masse élevés pendant leur recherche de nourriture à diverses altitudes, malgré les coûts énergétiques importants du vol stationnaire et de la thermorégulation. Afin d'essayer de comprendre les facteurs qui affectent les coûts énergétiques encourus durant la recherche de nourriture, nous avons capturé des colibris durant leur migration et mesuré leurs taux de consommation d'oxygène pendant l'alimentation en vol stationnaire. Les mesures ont été réalisées dans les sites mêmes dans lesquels les colibris roux s'alimentent durant leur migration à plusieurs stations dans le gradient d'altitude et la gamme de températures ambiantes qu'ils connaissent normalement. Les taux de consommation d'oxygène durant l'alimentation en vol stationnaire varient en fonction du sexe et différent chez les jeunes et les adultes. Ces différences semblent être le reflet de différences de besoins de puissance pour le vol stationnaire reliées aux variations de morphologie des ailes. Une baisse de la température ambiante et une augmentation de l'altitude accroissent significativement toutes deux le taux de consommation d'oxygène durant l'alimentation en vol stationnaire. Les effects de ces deux variables environnementales sont cumulatifs, ce qui laisse croire que les besoins de thermorégulation des colibris ne sont pas comblés par la production de chaleur additionnelle par le taux métabolique plus élevé requis pour permettre le vol stationnaire aux altitudes plus grandes. Ces résultats ouvrent des perspectives sur la manière dont les différentes stratégies de recerche de nourriture peuvent permettre aux colibris de maximiser leur apport net d'énergie.

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Introduction

Migratory birds face many environmental challenges as they cross latitudinal gradients and traverse mountain ranges, deserts, and open waters. Small birds, with their low thermal inertia, demonstrate large increases in energy expenditure with declining ambient temperature (T_a). In addition, their mode of flight is energetically expensive and highly reliant on aerobic energy metabolism. Aerodynamic power requirements are highly dependent on air density. Thus, variation in air density, as well as in the partial pressure of oxygen with elevation, can significantly increase the energetic cost of flight or even limit flight performance (Chai and Dudley 1995, 1996; Altshuler and Dudley 2003). T_a generally declines with increasing elevation, so the combined effects of increased power demands and limited oxygen availability on the energetic cost of flight, as well as the impact of these costs on the energetics of migratory flight, merit investigation.

Hummingbirds manage to stay in energy balance or make an energetic profit while foraging under a wide range of environmental conditions. Beginning in the late summer, rufous hummingbirds (*Selasphorus rufus* (Gmelin, 1788)) undertake an annual migration from breeding grounds in northwest North America to wintering grounds in central Mexico (Calder 1987). At various points along the migratory

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Table 1. Numbers and mean masses of rufous hummingbirds (*Selasphorus rufus*) included in study classified by age (AHY, after hatch year (>1 year old); HY, hatch year (<1 year old)) and sex (F, female; M, male).

Age class	Sex	n	Total number of trials*	Number of individual data points/trial (range)	Mean (±SE) mass during all experiments (g)
HY	F	1	2	34–74	3.86
AHY	F	5	17	20–67	3.52±0.06
HY	M	3	9	37–81	3.44±0.11
AHY	M	4	12	8–91	3.32±0.11

*A trial is defined as data from one individual at one elevation.

route including the Eastern Sierras, they establish and defend territories where they actively forage to rebuild fat stores. Such refuelling sites occur over a wide range of elevations. During early morning at higher elevations, rufous hummingbirds are often exposed to near-freezing T_a (Gass et al. 1999).

Because of their small size, hummingbirds display high resting oxygen consumption rates ($\dot{V}_{O_2rest}$) that increase further as T_a declines below their thermoneutral zone and thermogenic mechanisms are activated (Lasiewski 1963; López-Calleja and Bozinovic 1995; Chai et al. 1998; Gass et al. 1999). The high energetic cost of flight among hummingbirds raises the question of whether the heat produced during exercise is sufficient to cover the energetic cost of thermoregulation when hover-feeding in cold environments. Previous attempts to quantify the energetic cost of hovering in hummingbirds at low T_a have led to somewhat contradictory results. Using indigo-capped (*Amazilia cyanifrons* (Bourcier, 1843); mean 4.8 g) and rufous-tailed (*Amazilia tzacatl* (De la Llave, 1833); mean 4.4 g) hummingbirds, Schuchmann (1979) found that mass-specific (M_b, body mass) oxygen consumption rate during hovering ($\dot{V}_{O_2hov}/M_b$) increased as T_a declined to 4 °C, with slopes similar to those at rest. This high added cost owing to thermoregulation during hovering flight contrasts with results obtained by Berger and Hart (1972) using glittering-throated emeralds (*Amazilia fimbriata* (Gmelin, 1788); mean 5.7 g) and Chai et al. (1998) using ruby-throated hummingbirds (*Archilochus colubris* (L., 1758); mean 4.5 g). Both groups found only modest increases in $\dot{V}_{O_2hov}/M_b$ during T_a hovering in response to declining T_a, indicating near-complete substitution of thermoregulatory heat production by the heat produced during exercise.

Since air density declines as a function of increasing elevation, theory predicts that mechanical power requirements should increase along with metabolic rates (Ellington 1984). Therefore, more heat should be generated as a by-product of hovering at higher elevation. In assessing the utility of this additional heat production, two possibilities can be imagined. First, the additional heat generated as a result of the additional energy expenditure during hovering at high elevation may be insufficient to meet the thermogenic requirements at low T_a. If so, the effect of T_a on $\dot{V}_{O_2hov}/M_b$ should be independent of altitude (i.e., the slope of the relationship between hovering oxygen consumption rate and T_a should be the same regardless of elevation). Alternatively, the additional heat generated as a result of the increased metabolism may substitute for the additional thermogenic requirements

associated with hovering at low T_a. In this case, the slope of the relationship between hovering oxygen consumption rate and temperature should decrease with increasing elevation.

Despite greatly increased metabolic costs incurred during hovering flight at low T_a and high elevation, refuelling rufous hummingbirds achieve remarkable rates of net energy gain, adding as much as 50% to their lean body mass in fat over a 1- to 2-week period at an elevation of ~1700 m (Carpenter et al. 1993a). One of the mechanisms employed to save energy and increase the rate of mass gain is nocturnal torpor (Carpenter et al. 1993a; Hiebert 1990; Hiebert 1993). Nevertheless, mass gain during daytime foraging can be achieved despite the high energetic costs of flight and thermoregulation. In short-term laboratory experiments at T_a of 5 °C, Gass et al. (1999) found that rufous hummingbirds are able to achieve an energetic profit and gain mass by foraging at high frequency on 30% (*m/v*) sucrose. Under similar conditions, broad-tailed hummingbirds (*Selasphorus platycercus* (Swainson, 1827)) are unable to maintain energy balance and lose mass (McWhorter and Martínez del Rio 2000). Although it has been possible to model hummingbird time and energy budgets to gain insights into how they might achieve net energy gain under various environmental conditions (Gass and Garrison 1999), the predictive power of such models can be improved through the incorporation of more realistic assumptions concerning the combined effects of T_a and altitude on energy expenditure. In this study, we captured rufous hummingbirds and measured their hovering oxygen consumption rates at several sites along a gradient in elevation and over a wide range of T_as. Our use of migratory hummingbirds to perform measurements in the actual habitats through which they migrate and forage differs from previous laboratory studies, some of which employed artificial mixtures of helium and oxygen to manipulate air density.

Materials and methods

Study organisms

Rufous hummingbirds of each sex and age class were captured in a modified Hall trap (Russell and Russell 2001) between 27 June and 8 August of 2004 and 2005 in Inyo (at 1539 m elevation) and Mono (at 2445 m elevation) counties, California. In every case, hummingbirds were held in captivity for at least 3 days before participating in the study, to allow for the rebuilding of any nonadipose tissue potentially catabolized during the previous period of migratory flight (Carpenter et al. 1993a). Table 1 shows the numbers of each age and sex class included in this study, as well as the

Table 2. Elevations and locations of research sites in California used to study the rufous hummingbirds (*Selasphorus rufus*).

Site	Elevation (m)	Location	County
1	12	University of California, Santa Barbara, aviary	Santa Barbara
2	1539	Well's Meadow, ~25 km NE of Bishop	Inyo
3	2445	Valentine Camp, Mammoth Lakes	Mono
4	3089	Mosquito Flats, Rock Creek	Mono

mean masses of hummingbirds in each group. Birds were fed ad libitum on a 13% (*m/v*) solution of Nektar®-Plus (Guenter Enderle, Tarpon Springs, Florida) supplemented with table sugar (5% *m/v*). This solution was also utilized during the experiments described below.

Data collection

All experiments were conducted while the birds were inside a 0.6 m × 0.6 m × 0.6 m cage composed of window screen and acrylic. Data were collected from each bird at 1–4 separate sites (3 ± 1 sites) along a gradient in elevation spanning >3000 m (12–3089 m; see Table 2). The elevation at each site was determined by GPS. Data collection took place between 0530 and 1850 on any given day, although no period of data collection on any individual bird on any day exceeded 6 h in duration. Data collection was conducted continuously utilizing only one bird at any time and lasted between 2 and 6 h. Ambient temperature during the period of data collection changed as the natural result of heating following the rising of the sun in the morning hours and cooling during late afternoon hours. This natural temperature variation was exploited during the period of data collection to explore the effect of temperature on oxygen consumption rate during hovering. All hummingbirds were allowed to feed for a minimum of 30 min prior to each period of data collection to ensure that hummingbirds were oxidizing predominantly carbohydrate during the entire period of data collection (Suarez et al. 1990; Welch et al. 2006, 2007; Welch and Suarez 2007). All data collection was conducted while the experimental cage was placed outdoors, shaded from direct sunlight. Data collection conducted at higher elevation sites (1539–3089 m in elevation) within Inyo and Mono counties, California, took place between 29 June and 14 August of 2004 and 16 June and 30 August of 2005 (Table 2). Throughout this period, birds were housed in similar cages on or nearby the grounds of the University of California Valentine Eastern Sierra Reserve – Valentine Camp. Hummingbirds were transported in cages, with constant access to food, in the early morning to the 1539 and 3089 m data collection sites by automobile. Data collection at 12 (see below) and 2445 m took place in areas immediately adjacent to the holding facilities. Data collection conducted at the lower elevation site (12 m in elevation) on the campus of the University of California, Santa Barbara, (UCSB) took place between 9 September and 14 October of 2004 and 6 September and 26 October of 2005 (Table 2). During these periods, birds were housed at the UCSB aviary in individual outdoor, wire-mesh enclosures measuring 1.8 m tall × 0.6 m wide × 2.4 m long, or larger. Capture, housing, and experimental protocols adhered to the recommendations of the Guide for the Care and Use of Laboratory Animals (Institute of Laboratory Animal Resources Commission on Life Scien-

ces 1996) and were approved by the UCSB Institutional Animal Care and Use Committee (protocol No. 672).

Respirometry

Oxygen consumption rates during hovering were obtained by inducing hummingbirds to voluntarily place their heads inside a respirometry mask to access the feeding solution following the technique described by Bartholomew and Lighton (1986). A 50 mL conical tube, capped by a rubber stopper, served as the reservoir for the Nektar®-Plus and table sugar solution. The rubber stopper was pierced by an 8 mm (inner diameter) glass tube of ~20 cm in length. This glass tube was bent at a ~30° angle approximately halfway along its length and fire polished so that the external opening had closed to roughly 2–3 mm in diameter. A plastic cylinder (16 mm inner diameter), which served as a mask whenever the hummingbird accessed the feeder, was attached to the front of a red plastic flower (Perky-Pet® brand, Denver, Colorado) and this was fastened to the glass feeding tube with a rubber grommet secured to the back end of the plastic flower. The cylinder was punctured by 0.25 inches (1 inch = 25.4 mm) inner diameter polyethylene tubing approximately halfway along its length, serving as the conduit through which air could be drawn through the mask and delivered on to the respirometric equipment. A photoresistor was attached to the bottom of the plastic mask adjacent to the open end. When a hummingbird's head was inside the mask (during a feeding event), the photoresistor was occluded. By determining the length of time the resistance signal from the photoresistor was elevated (indicating occlusion), the duration of any given feeding event could be calculated. All feeding events <1 s in duration were excluded from analysis. Overall, feeding events averaged 6.4 ± 3.8 s (range 1.0–36.0 s) in duration.

Air was drawn through the feeder mask and then passed through a column of Drierite–Ascarite–Drierite (Drierite: W.A. Hammond Drierite Co., Ltd., Xenia, Ohio; Ascarite II: Arthur H. Thomas Company, Philadelphia, Pennsylvania) to remove water vapour and carbon dioxide from the airstream. Subsequently, the air passed into the oxygen analyzer. During 2004, the oxygen analyzer was the FOX® model (Sable Systems International Inc., Las Vegas, Nevada). During 2005, the oxygen analyzer was a FOXBOX® model (Sable Systems International Inc.). In each case, flow rate corrected to standard temperature and pressure (STP) was sensed by an internal mass-flow meter and was maintained between 1000 and 1100 mL·min^{-1} by control of pumping intensity via a mechanism internal to each analyzer. STP-corrected oxygen data associated with each feeding event were first corrected by subtracting baseline values (determined as the linear extrapolation of points directly before and after the feeding event in question). These baseline-

corrected data were then converted to millilitres of gas using eq. 3b of Withers (1977). Rates of oxygen consumption were determined by dividing the calculated volume of oxygen consumed by the duration of the feeding event. Prior to each period of data collection, the oxygen analyzer was calibrated with well-mixed ambient air drawn through the mask in the absence of the hummingbird.

The only perch provided to the hummingbird inside the experimental chamber was placed on top of a balance. Thus, whenever the bird perched, its mass was measured. A thermoresistor was placed adjacent to the feeder mask, allowing for recording of T_a. Analog voltage outputs from the thermoresistor, the balance, the photoresistor, and the oxygen analyzer were conveyed to a UI2 Universal Interface (Sable Systems International Inc.) that converted all data to digital format and transmitted it to a notebook computer where it was recorded.

Data analysis

Data from any individual at a given elevation were excluded from analysis if the temperature range over which data were obtained spanned <10 °C. This was done because scatter in the data potentially obscured actual correlations between oxygen consumption rate and temperature when the temperature range was not sufficiently large. Data were analyzed using multiple linear regression on each individual hummingbird independently. The effects of temperature, elevation, and their interaction were examined in each case, except for one hummingbird for which data were only available from only one elevation. All data are reported as means ± SE.

Results

Colder T_a and higher elevation independently increased metabolic rates during hovering in migratory rufous hummingbirds. Multiple linear regression modelling on each individual revealed that $\dot{V}_{O_2hov}/M_b$ increased significantly as temperature decreased ($P \leq 0.0029$). Additionally, $\dot{V}_{O_2hov}/M_b$ increased significantly as elevation increased for each individual ($P < 0.0001$). The effect of the interaction of elevation and temperature on $\dot{V}_{O_2hov}/M_b$ was significant ($P < 0.05$) for 6 out of 12 hummingbirds (Fig. 1).

The calculated slopes of the regression of $\dot{V}_{O_2hov}/M_b$ (mL $O_2 \cdot g^{-1} \cdot h^{-1}$) vs. T_a (i.e., the thermal conductance) at each elevation at which an individual was studied were significantly less than zero ($P \leq 0.0104$) and ranged from –0.87 to –0.25 mL $O_2 \cdot g^{-1} \cdot h^{-1} \cdot °C^{-1}$. Table 3 lists the calculated values of thermal conductance at each site along the gradient in elevation averaged by age class and sex. The mean thermal conductance across individuals was –0.58 ± 0.04 mL $O_2 \cdot g^{-1} \cdot h^{-1} \cdot °C^{-1}$ ($n = 13$).

The slope of the relationship of $\dot{V}_{O_2hov}/M_b$ to elevation, averaged across individuals, was 4.3 ± 0.3 mL $O_2 \cdot g^{-1} \cdot h^{-1} \cdot km^{-1}$ ($n = 12$). Figure 2 shows mean values of $\dot{V}_{O_2hov}/M_b$ at 0 °C, calculated from estimates of thermal conductance for each bird, at each elevation, averaged for each age and sex class. These values of $\dot{V}_{O_2hov}/M_b$ permit comparison of $\dot{V}_{O_2hov}/M_b$ across elevations at a constant T_a (Fig. 2). For each age and sex class (except HY F, for which there were just two values available), the relationship between intercept values and elevation was significant (Fig. 2; $P \leq$

0.0047). While the values of thermal conductance (and the values of $\dot{V}_{O_2hov}/M_b$ at 0 °C calculated from thermal conductance, see below) calculated for each individual at each elevation are not strictly equivalently independent, linear regression was employed to ensure that the results are more accessible to all readers.

For 6 of 12 individual hummingbirds, there were significant differences in the estimates of thermal conductance among one or more sites along the gradient in elevation. However, there was no consistent pattern of increasing or decreasing thermal conductance with increasing elevation. This is apparent in Fig. 3, which shows mean thermal conductance at each elevation in relation to elevation itself grouped by age and sex class. Linear regression revealed no significant trend in thermal conductance as a function of elevation for each age and sex class (Fig. 3; $P \geq 0.2685$). While there is the suggestion of an inverted U-shaped relationship between thermal conductance and elevation, fitting of a second-degree polynomial curve to the data reveals a significant relationship only for AHY M ($F_{[3,13]} = 1.1320$, $P = 0.0434$). For all other groups, the fitted second-degree polynomial curves were not significant ($P \geq 0.3724$).

Figure 2 reveals that mean $\dot{V}_{O_2hov}/M_b$ varied consistently between age and sex classes across elevations when compared at a given T_a. Nonparametric statistical analysis revealed significant concordance between age and sex class and mean intercept values at the 1539 and 2445 m sites (Kendall's $\tau = 1.0$, $P = 0.0415$ for each), but not at the 12 and 3089 m sites (Kendall's $\tau = 1.0$, $P = 0.1172$ for each). In each case the correlation was complete, and it is likely that limitations in sample size led to a lack of statistical significance being detected at the 12 and 3089 m sites.

Discussion

When faced with declining T_a, hummingbird $\dot{V}_{O_2rest}$ dramatically increases as thermogenic mechanisms are activated. Gass et al. (1999) found that rufous hummingbirds respond to low T_a by increasing foraging effort and, when provided sufficiently high sucrose concentrations in their diet, are able to maintain energy balance or achieve net energy gain despite the high costs of hovering and thermoregulation. That laboratory study led to the present one which aimed to characterize the energetic costs incurred by migratory rufous hummingbirds as they experience the range of air densities, oxygen partial pressures, and T_a values in the actual habitats in which they forage. The natural variation in environmental conditions inherent in the field-based measurements described here precludes rigorous mechanistic conclusions. Rather than inferring details concerning physiological or biochemical mechanisms as in previous studies (Suarez et al. 1990; Welch et al. 2006, 2007; Welch and Suarez 2007), our intention was to estimate the energetic costs incurred by rufous hummingbirds as they hover during foraging under the field conditions they naturally encounter during migratory refuelling.

Our data indicate that the heat generated by increased metabolism during hovering flight in hummingbirds is not sufficient to fully meet their thermogenic requirements at low T_a under field conditions. The values of thermal conductance (i.e., slopes of the relationship between $\dot{V}_{O_2hov}/M_b$ and

Fig. 1. Plots of the relationship between hovering mass-specific oxygen consumption rate and temperature by elevation for each rufous hummingbird (*Selasphorus rufus*). Multiple linear regression reveals a significant effect of temperature ($P \leq 0.0029$) and elevation ($P < 0.0001$) for most individuals. The effect of elevation on $\dot{V}_{O_2hov}/M_b$ could not be evaluated for individual N43934 because data were available at only one site along the gradient in elevation. AHY, after hatch year (>1 year old); HY, hatch year (<1 year old); F, female; M, male.
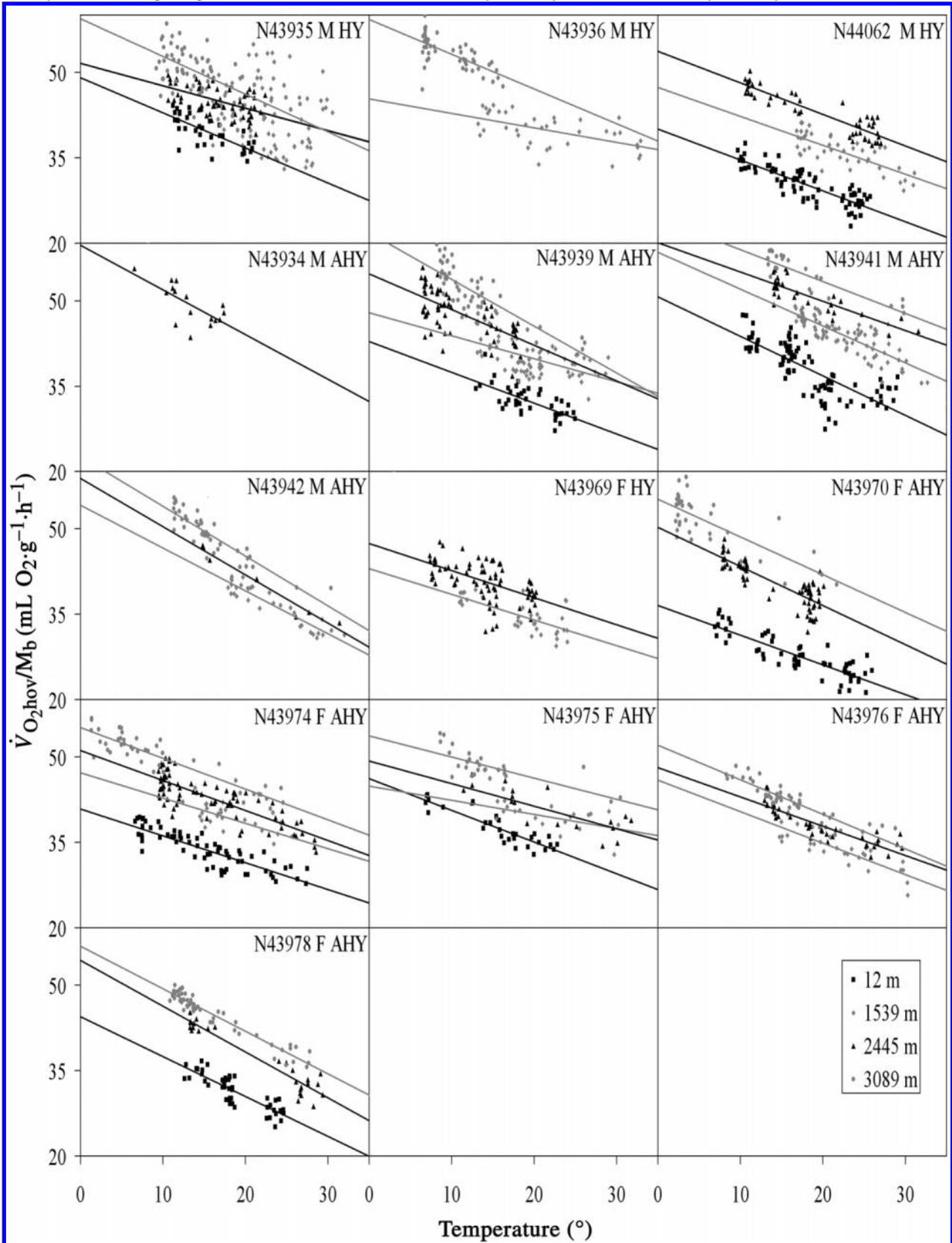
Table 3. Mean thermal conductance (i.e., slopes of the relationship between hovering mass-specific oxygen consumption rate and temperature) of rufous hummingbirds (*Selasphorus rufus*), grouped by age and sex class, at each elevation site.

Age class	Sex	Mean (±SE) thermal conductance (mL $O_2 \cdot g^{-1} \cdot h^{-1} \cdot °C^{-1}$) by elevation			
		12 m	1539 m	2445 m	3089 m
HY	F		–0.45 (1)	–0.47 (1)	
AHY	F	–0.56±0.05 (4)	–0.41±0.09 (3)	–0.58±0.07 (5)	–0.59±0.06 (5)
HY	M	–0.58±0.04 (2)	–0.38±0.07 (3)	–0.47±0.08 (2)	–0.63±0.03 (2)
AHY	M	–0.62±0.08 (2)	–0.60±0.10 (3)	–0.69±0.08 (4)	–0.75±0.10 (3)

Note: AHY, after hatch year (>1 year old); HY, hatch year (<1 year old); F, female; M, male. Values in parentheses are the number of birds studied at each elevation. The regressions are significantly different from zero for each individual hummingbird at each site ($P \le 0.0104$).

Fig. 2. Mean (±SE) mass-specific oxygen consumption rate of rufous hummingbirds (*Selasphorus rufus*) at 0 °C (based on calculated thermal conductance) as a function of elevation. There is a significant positive relationship between the intercept value and elevation ($P \le 0.0047$) for each age and sex class, except for HY F, which only had two values available. AHY, after hatch year (>1 years old); HY, hatch year (<1 year old); F, female; M, male.
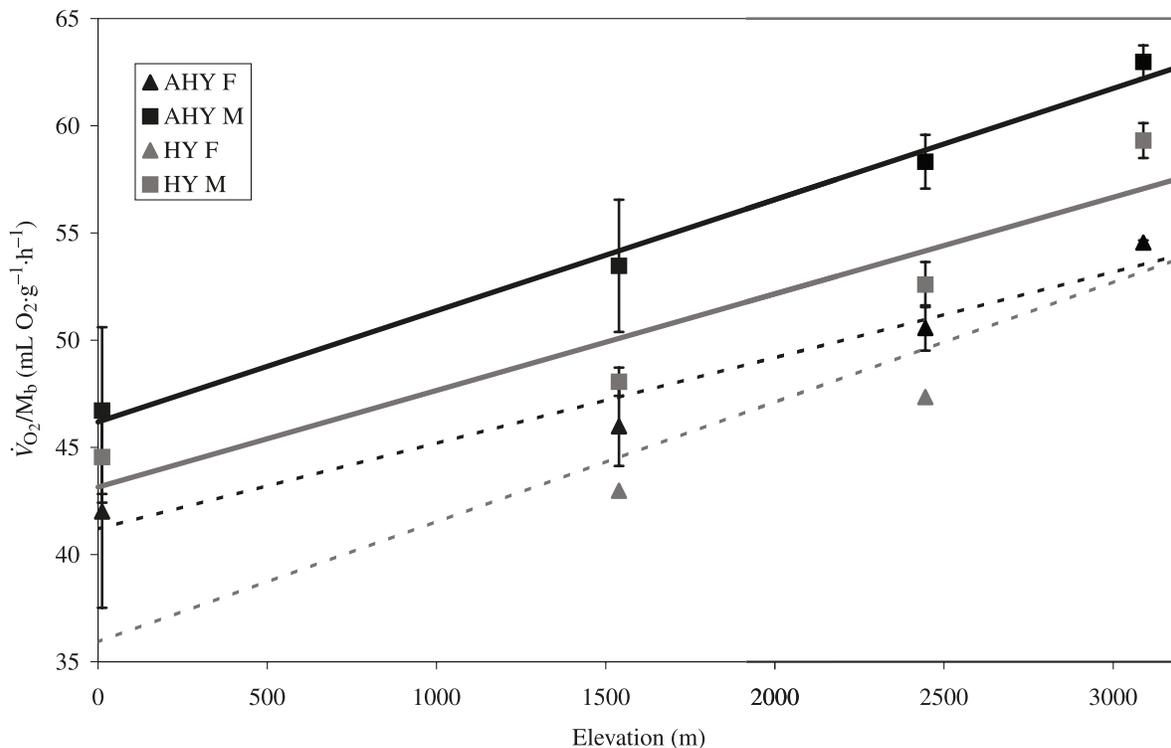

temperature) reported here (–0.58 ± 0.04 mL $O_2 \cdot g^{-1} \cdot h^{-1} \cdot °C^{-1}$) are similar to those reported by Hiebert (1990) and Lasiewski (1963) for perched rufous hummingbirds and Schuchmann (1979) for rufous-tailed hummingbirds but substantially greater than those reported for glittering-throated emeralds and ruby-throated hummingbirds (Berger and Hart 1972; Chai et al. 1998). The mean body mass of the migratory rufous hummingbirds used in our study, 3.47 ± 0.06 g, is significantly lower than those of the other species used in these laboratory studies. The differences in results across studies are likely due to a combination of interspecific variation and differences in experimental conditions (i.e., natural vs. controlled).

Hovering flight at higher altitude is associated with significantly greater oxygen consumption rates in rufous hummingbirds compared with lower altitudes. This is predicted by aerodynamic theory for hovering animals (Ellington 1984), which says that the power requirements for hovering should increase as a function of decreasing air density. In laboratory studies, Berger (1974) observed an increase in oxygen consumption rate of 6%–8% when simulating a change in elevation between sea level and 4000 m in sparkling violetears (*Colibri coruscans* (Gould, 1846)) and glittering-throated hummingbirds. In the present study, we estimated a substantially greater increase in $\dot{V}_{O_2 hov}/M_b$ as a function of elevation in rufous hummingbirds. At 20 °C, rufous hummingbirds increased their $\dot{V}_{O_2 hov}/M_b$ by ~30% as elevation increased from 12 to 3089 m. In laboratory experiments, Altshuler and Dudley (2003) report that rufous hummingbirds are capable of hovering flight under conditions of normobaric hypoxia when oxygen partial pressures approximated that found at an elevation of 6000 m. Given

Fig. 3. Mean (±SE) thermal conductance values (i.e., slopes of the linear regression of relationship between hovering mass-specific oxygen consumption rate of rufous hummingbirds (*Selasphorus rufus*) to ambient temperature as a function of elevation. There is no significant effect of elevation on the thermal conductance when a linear fit is applied ($P \geq 0.2685$). The fitted second-degree polynomial curve was significant for AHY M ($P = 0.0434$) but not for AHY F, HY M, or HY F ($P \geq 0.3724$). AHY, after hatch year (>1 year old); HY, hatch year (<1 year old); F, female; M, male.
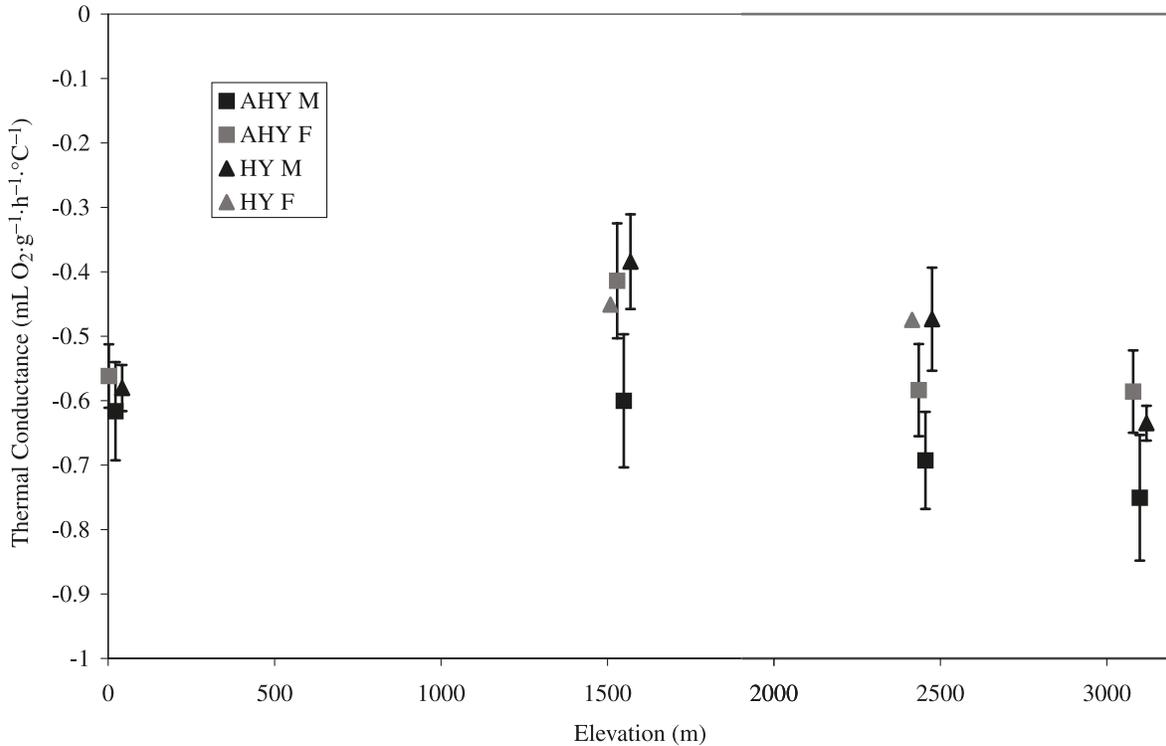
the dramatic increase in $\dot{V}_{O_2 hov}/M_b$ as a function of increasing elevation observed here, as well as the ability of rufous hummingbirds to sustain hovering flight when the oxygen content of the air is greatly decreased, it is clear that these birds possess large reserve capacities for the delivery of oxygen to exercising muscle tissues.

The additional power requirements necessary to support hovering at high elevation should lead to the production of additional heat as a by-product of increased metabolic rate. While there was statistically significant variation in the slopes of the relationship between $\dot{V}_{O_2 hov}/M_b$ and T_a across elevations for 6 of 12 individual hummingbirds, the variation in slopes appears not to follow any pattern and is essentially random. Our results suggest that the additional heat associated with increased metabolism during hovering at higher elevation does not provide additional thermoregulatory benefit. Thus, to maintain thermal homeostasis, rufous hummingbirds must increase their metabolic rate during hovering at low T_a at higher elevations to the same relative extent that they do at lower elevations.

Our data also show that $\dot{V}_{O_2 hov}/M_b$ varies in a consistent way between each age and sex class across all elevations regardless of T_a. $\dot{V}_{O_2 hov}/M_b$ values at a given elevation and T_a for each age and sex class are related in the following way: AHY M > HY M > AHY F > HY F. This is an intriguing result given that relative competitive dominance in territorial interactions between members of differing age and sex classes of rufous hummingbirds follows an identical hierarchy (Carpenter et al. 1993*b*, 1993*c*; Kodric-Brown and

Brown 1978). Table 4 lists published average wing length values, as well as wing disc loading (ratio of body mass to area swept out by the wings) values, calculated using mean hummingbird mass in this study for each rufous hummingbird age and sex class. Among AHY M, HY M, and AHY F rufous hummingbirds, increases in mean wing disc loading correspond with greater territorial dominance (Table 4). Thus, relative territorial dominance correlates with higher wing disc loading (Feinsinger and Chaplin 1975; Feinsinger and Colwell 1978; Feinsinger et al. 1979). Altshuler et al. (2004) contend that wing length is a superior predictor of territorial dominance compared with wing disc loading. Wing lengths from published data support this suggestion across all four rufous hummingbird age and sex classes (Table 4). In these studies, it is assumed that general aerial maneuverability should increase as wing length decreases (and by extension wing disc loading increases), thus enabling greater fighting prowess in more dominant territorial hummingbirds. Carpenter et al. (1993*c*) noted that despite their subordinate territorial status, AHY and HY F rufous hummingbirds are able to gain mass approximately as quickly as HY M individuals. They hypothesized that lower wing disc loading affords these age and sex classes lower energetic costs during hovering flight, which allows them to achieve high net energy intake rates despite exclusion from the highest quality resources. Our data show that the more subordinate age and sex classes of rufous hummingbirds display lower mass-specific metabolic rates during hovering when compared at a given temperature and

Table 4. Mean published wing lengths (Kodric-Brown and Brown 1978; Carpenter et al. 1993c) and calculated mean wing disc loading (WDL) for each age and sex class of rufous hummingbirds (*Selasphorus rufus*).

Age class	Sex	n	Mean (±SE) wing length (mm)*	Mean (±SE) mass during all experiments (g)	Mean WDL (g/cm^2)[†]
HY	F	1	45.1±0.1	3.86	0.0405
AHY	F	5	44.6±0.2	3.52±0.06	0.0377
HY	M	3	43.0±0.1	3.44±0.11	0.0394
AHY	M	4	40.7	3.32±0.11	0.0421

Note: AHY, after hatch year (>1 year old); HY, hatch year (<1 year old); F, female; M, male.

*Data from Carpenter et al. (1993c) and Kodric-Brown and Brown (1978).

[†]WDL is calculated as $M_b/(2 \cdot \text{mean} + 0.809 \cdot \text{mean}^{0.6})$ based on the equation in Greenewalt (1975).

elevation and support the hypothesis of Carpenter et al. (1993c). Hiebert (1993) reported that captive rufous hummingbirds, like wild birds, underwent seasonal changes in daily mass that appeared to reflect their predisposition to increase net energy intake during migratory periods. Because rufous hummingbirds used in this study were captured on migratory refuelling grounds, presumably during the beginning or during a period of mass gain (Carpenter et al. 1983, 1993a), it is likely that they would gain some mass during the course of the study. As this mass gain would affect wing disc loading, thus confounding any effects of temperature or elevation on hovering metabolic rate, care was taken to test hummingbirds at each elevation in a random order. However, travel between the three higher elevation sites in Mono and Inyo counties and the lowest elevation site in Santa Barbara county on a daily basis was not possible, and thus all studies at this low-elevation site took place last. While hummingbirds were, on average, slightly heavier when tested at this lowest elevation, the difference in mass was not statistically significant ($F_{[3,36]}$ = 2.3336, P = 0.0903), and thus any changes in wing disc loading are likely minimal.

Gass et al. (1999) show that rufous hummingbirds typically were unable to maintain or gain mass at 5 °C when feeding on 15%–20% sucrose solutions, approximating the low end of the range of sucrose concentrations in flowers hummingbirds typically visit in the wild (18%–26%, Bolten et al. 1979). This mass loss occurs despite observed increases in foraging frequency at lower T_a. However, rufous hummingbirds can clearly gain mass when foraging on dilute nectar at higher T_a (Carpenter et al. 1993a). We have shown here that the energetic cost of hovering (foraging) flight is significantly increased with increased elevation and lower T_a. Such a combination would be expected to adversely impact the ability of hummingbirds to make an energetic profit. On this basis, we hypothesize that rufous hummingbirds should prefer floral resources located at lower elevation and restrict foraging at high elevation to warmer times of the day when lower elevation resources are readily available. A possible strategy would involve beginning foraging in the morning at lower elevations, and moving upslope to forage on nectar at higher elevation as temperatures climb. Consistent with the idea put forward by Carpenter et al. (1993c), we propose that these high-elevation floral resources are more likely to be exploited by HY and AHY F rufous hummingbirds owing to competitive exclusion from lower elevation sites, as well as their

ability to profit more from these resources owing to their relatively lower energetic costs of hovering.

In summary, we have shown that environmental factors including temperature and elevation can have profound effects on the energetic costs of foraging in migratory rufous hummingbirds, and that strategies for achieving maximal rates of net energy gain might differ among age and sex classes. These data add further support for the suggestion by the late William Calder (Calder 1979) that physiological data can provide useful insights in studies of feeding strategies in hummingbirds.

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