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RESEARCH ARTICLE

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Increased physical activity is not related to markers of cardiometabolic health in two lemur species

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Abstract

Insufficient physical activity is a major risk factor for cardiometabolic disease (i.e., unhealthy weight gain, heart disease, and diabetes) in humans and may also negatively affect health of primates in human care. Effects of physical activity on energy expenditure and cardiometabolic health are virtually unstudied in nonhuman primates. We investigated physical activity and metabolic markers in 15 adult ring-tailed lemurs (*Lemur catta*) and 11 Coquerel's sifakas (*Propithecus coquereli*) at the Duke Lemur Center during a period of low activity in winter when the animals were housed in buildings (with outdoor access) and a period of high activity when individuals were free-ranging in large, outdoor, forested enclosures. We compared body mass, blood glucose, triglycerides, HDL- and LDL-cholesterol, physical activity via accelerometry, and total energy expenditure (TEE) via the doubly labeled water method (in ring-tailed lemurs only) between both conditions. Both species were more active and had a lower body mass in summer. Ring-tailed lemurs had a higher TEE and lower triglyceride levels in summer, whereas sifaka had higher triglyceride levels in summer. Individuals that increased their activity more, also lost more body mass. Individuals that lost more body mass, also had a positive change in HDL-cholesterol (i.e., higher values in summer). Changes in activity were not associated with changes in markers of metabolic health, body fat percentage and TEE (both unadjusted and adjusted for body composition). Older age was associated with lower activity in both species, and decreased glucose in ring-tailed lemurs, but was otherwise unrelated to metabolic markers and, for ring-tailed lemurs, adjusted TEE. Overall, body mass was lower during summer but the increase in physical activity did not strongly influence metabolic health or TEE in these populations.

KEYWORDS
cardiometabolic health, energetics, energy budget, metabolism, obesity

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Abbreviations: DLW, doubly labeled water; FFM, fat-free mass; FM, fat mass; FQ, food quotient; kD, depletion rate for ^{2}H (deuterium); kO, depletion rate for ^{18}O; ND, dilution space for ^{2}H (deuterium); NO, dilution space for ^{18}O; TBW, total body water; TEE, total energy expenditure.

1 | INTRODUCTION

Physical activity is central to both the evolved ecology and cardiometabolic health of humans and nonhuman primates (Aune et al., 2015; Lee et al., 2012; Schuler et al., 2013). Species' anatomy and physiology evolve to anticipate a particular level of physical activity. Consequently, species vary considerably in their activity level and the energy cost of daily physical activity. When the physical activity level differs from the species-typical range, health can be negatively affected. To date, this phenomenon has been studied most thoroughly in humans. Humans evolved as hunter-gatherers, with physically demanding foraging strategies that involved hours of moderate and vigorous physical activity each day (Pontzer et al., 2018; Wood et al., 2021). The lack of regular and vigorous physical activity in industrialized populations today is known to be a major risk factor for cardiometabolic disease (i.e., unhealthy weight gain, heart disease, and diabetes) and decreased longevity (Abdelaal et al., 2017; Aune et al., 2015; Lee et al., 2012; Schuler et al., 2013).

The relationship between daily physical activity and cardiometabolic health is not well studied in most other species, but it appears to be important, at least in species with habitually active lifestyles. Ad libitum fed laboratory rats with regular access to a running wheel have a lower body mass, no kidney lesions and more favorable health biomarkers compared to rats with little to no access to exercise (Alessio et al., 2005). In zoo elephants (*Loxodonta africana* and *Elephas maximus*), greater daily walking distance is associated with positive health outcomes including a healthier body condition and improved blood profiles (Meehan et al., 2016; Morfeld et al., 2016). Female baboons (*Papio cynocephalus*) that forage on natural food sources and travel long distances per day (8–10 km/day) have a lower body fat percentage (2%) compared to females (23%) that forage on a garbage dump and travel short distances (<4 km/day) (Altmann et al., 1993). Male baboons from the social group that uses the garbage dump have a reduced physical activity level, elevated cholesterol levels, and some are considered obese and have developed insulin resistance (Banks et al., 2003).

The findings of these previous studies are consistent with changes associated with the metabolic syndrome that increase the risk of heart disease in humans (McMillen & Robinson, 2005). Symptoms of the metabolic syndrome in humans include high blood pressure, high blood glucose, high plasma triglycerides, low HDL-cholesterol, and high waist circumference (The expert panel, 2001). Most of the studies conducted on animals to date are cross-sectional, but studies that assess within-individual changes in activity, cardiometabolic health and total energy expenditure (TEE; kcal/day) are required to better understand how changes in activity affect health and energy expenditure, and which factors explain interindividual variation in the response to an altered level of physical activity.

Activity level, TEE and body composition can change throughout an individual's lifespan. An age-related decline in physical activity has been reported for humans, nonhuman primates and other mammals (Chusyd et al., 2021; Holdgate et al., 2016; Ingram, 2000; Jänicke et al., 1986; Pontzer et al., 2021; Sallis, 2000). Age-related changes in TEE and body composition are best studied in humans, where TEE and fat-free mass (FFM) decrease in adults older than 60 years (Elia et al., 2000; Manini, 2010; Pontzer et al., 2021; Speakman & Westerterp, 2010). Fat mass (FM) has been found to increase (Coin et al., 2008; Kyle et al., 2001) and to decrease (Elia et al., 2000) with age in humans. Age-related changes in TEE and body composition have rarely been studied in nonhuman animals and thus, are not well-understood. Dog breeds differ in age-related changes in body composition, FM increases with age in a large breed, but is unrelated to age in medium and small-sized breeds (Speakman et al., 2003). Common bottlenose dolphins (*Tursiops truncatus*) show an age-related decline in TEE and an increase in body fat percentage (Rimbach et al., 2021), similar to patterns reported in humans. Age-related changes in TEE and body composition may thus be species-specific and data from additional species are needed to shed light onto these patterns.

Obesity and cardiometabolic health are common concerns for primates in human care (Clay et al., 2022; Cole et al., 2020; Mellor et al., 2020; Pontzer, 2023; Terranova & Coffman, 1997). Due to their close relatedness with humans, nonhuman primates may present a relevant biological model to study the relationship between cardiometabolic health, obesity and physical activity. Most research on primate cardiometabolic health and obesity to date has focused on a restricted number of genera, with bias towards macaques (*Macaca* spp.) and baboons (*Papio* spp.) (Pontzer, 2023). Other primate taxa, such as some lemur species, are commonly kept in zoos and research facilities (Association of Zoos and Aquariums, 2023; Reimes et al., 2021), and vary in their susceptibility to gain body mass in captivity. For example, ring-tailed lemurs (*Lemur catta*) are prone to elevated body masses in captivity, while other species, such as Coquerel's sifaka (*Propithecus coquereli*), are not (Mellor et al., 2020). To assess whether increases in physical activity are associated with changes in body mass and cardiometabolic health, we studied two lemur species that differ in their susceptibility to body mass gains in human care, the ring-tailed lemur (*L. catta*) and Coquerel's sifaka (*P. coquereli*). We measured physical activity, body mass, serum glucose and lipids of 15 adult ring-tailed lemurs and 11 adult Coquerel's sifakas in two different conditions, once during winter (low daily physical activity) and once during summer (high daily physical activity). For 11 ring-tailed lemurs, we also determined TEE and body composition (body fat percentage and FFM) in both conditions. The aims of this study were to (1) test whether increases in physical activity are associated with changes in body mass, cardiometabolic health, including serum glucose and lipids (HDL- and LDL-cholesterol, triglycerides), TEE and body fat percentage, and (2) to assess age-related changes in activity and metabolic phenotype in both lemur species. We predicted that blood glucose, LDL-cholesterol and triglyceride levels would be lower and that HDL-cholesterol would be higher in the high activity condition compared to the low activity condition. Further, we predicted body mass and body fat percentage to be lower in the high activity condition, and we did not expect changes in mass-adjusted TEE. We expected that an increase in activity would be associated with increased HDL-cholesterol, and decreased LDL-cholesterol and triglyceride levels.

2 | METHODS AND MATERIALS

2.1 | Study species and site

All subjects were housed at the Duke Lemur Center (DLC) in Durham, North Carolina (N 35° 59′ 35″, W78° 57′ 38″). All animal use and methods were approved by the Duke University Institutional Animal Care and Use Committee (Protocol #: A244-19-11) and the DLC Research Committee. The research also adhered to legal requirements and to the American Society of Primatologists Principles for the Ethical Treatment of nonhuman Primates.

We collected data on 15 adult ring-tailed lemurs (*L. catta*), seven females and eight males (age range: 5.2–27.3 years) and 11 adult Coquerel's sifakas (*P. coquereli*), five females and six males (age range: 3.1–17.0 years). Sample sizes were restricted by the number of animals available at DLC, and their health and reproductive status. Individuals were measured in two different conditions: a low activity (winter: *L. catta*: February to March 2020; P. *coquereli*: January to February 2021) and a high activity condition (summer: *L. catta*: July to August 2020; P. *coquereli*: April to July 2021). In winter, animals were kept indoors (room size: approximately 2.13 m length × 2.29 m width × 3.05 m height; one room per adult in the group), and were given access to attached outdoor enclosures (4.27 m length × 2.29 m width × 3.05 m height). In summer, a subset individuals acted as controls (nonfree-rangers; *L. catta*: $N = 5$; *P. coquereli*: $N = 3$) and were housed in the same set-up or outdoor-only chain-link silos (approximately 4.57 m diameter × 5.49 m height for the entire group) and the other individuals (free-rangers; *L. catta*: $N = 11$; *P. coquereli*: $N = 8$) were free-ranging in large forest enclosures that vary in size from 0.6 to 5.7 ha. Housing conditions were Continuous Full Contact (pair or group), meaning that two or more animals are housed in one space, enabling complete tactile interaction. Measurements of the control individuals were collected at the same time of year as for the free-ranging individuals to control for the potential effect of seasonal changes in metabolic phenotype that may obscure effects of physical activity. For all measurements, individuals were housed in indoor or outdoor enclosures for a minimum of 2 weeks before measurement.

In the wild, the majority of ring-tailed lemurs disappear between 10 and 15 years of age and few individuals live beyond 15 years (Bennett et al., 2016; Cuozzo & Sauther, 2004). The age range of ring-tailed lemurs included in this study was 5.2–27.4 years, and that of Coquerel's sifaka was 3.1–17.3 years. At DLC, both species can live past 30 years (*L. catta*: 32.7 years; *P. coquereli*: 30.6 years), but ring-tailed lemurs have a higher median longevity of individuals that survived at least 30 days than Coquerel's sifaka (*L. catta*: 17.6 years; *P. coquereli*: 10.0 years) (Zehr et al., 2014).

2.2 | TEE

We calculated total energy expenditure (TEE, kcal/day) using the doubly labeled water (DLW) method (Lifson & McClintock, 1966; Speakman, 1997). This method uses water (H_2O) enriched in the safe, stable, nonradioactive isotopes (^2H, also called deuterium and ^{18}O) to track the production of CO_2, which yields a precise and reliable measure of energy expenditure. The dose of enriched water can be administered either by drinking it or through injection. In this study, subjects drank the dose. During the subsequent days, the hydrogen isotope, ^2H, is eliminated from the body via water (urine, sweat, insensible water loss), whereas the oxygen isotope ^{18}O is lost both via water and via expired CO_2. CO_2 production can be calculated by determining the difference between the elimination rates of the hydrogen and oxygen isotopes. The DLW method is the gold standard for measuring energy expenditure in free-living (i.e., outside of the lab) humans and nonhuman animals.

We determined TEE of 16 ring-tailed lemurs during winter 2020. Two ring-tailed lemurs did not drink the full dose of DLW and thus, their TEE could not be determined. Together with problems during sample collection (e.g., individuals had to be locked inside due to a hurricane warning or raccoons inside outdoor enclosures) and the death of one female, this resulted in a reduced sample size of 11 individuals (eight free-rangers (four females and four males) and three nonfree-rangers (one female and two males)) in summer (Table 1). We successfully determined TEE of eight Coquerel's sifakas (six free-rangers and two nonfree-rangers; four females and four males) during winter 2021, and TEE of two sifakas (one female and one male; both free-rangers) during summer 2021. During summer 2021, sample size of available sifaka for this study was reduced because the available population of sifaka was lower (e.g., individuals were transported to other facilities) and because we had access to fewer individuals due to DLC logistic constraints (such as breeding attempts and pregnancies). The largely reduced sample sizes for sifaka are mainly due to problems with administering the DLW dose to individual sifaka (i.e., they did not consume the DLW dose) and urine sample collection (i.e., sufficient amounts of urine could not be collected). In addition, one individual had to be taken into veterinary care and data collection on this individual was aborted. Thus, no repeat measures of TEE were successful for any sifaka.

Individuals ingested premeasured doses (*L. catta*: range: 6–9 g; *P. coquereli* range: 9–14 g) of doubly labeled water (6% ^2H_2O, 10% $H_2{}^{18}O$) tailored to body mass to provide sufficient initial isotopic enrichment (Speakman, 1997). Each individual provided one urine sample (0.5–4 mL) before dosing and another two samples postdose ingestion. The first postdose ingestion was collected 24.6 ± 5.7 h after dosing in ring-tailed lemurs and 20.6 ± 4.6 h after dosing in sifaka, and the last sample was collected 6–7 days post-dose ingestion. Samples were frozen (−20°C) and transported to Duke University for isotopic analysis.

2.3 | Isotope analysis

Samples were filtered using carbon black and a 30 kilodalton centrifuge concentrator (Vivaspin®). Enrichments of ^2H and ^{18}O were determined using integrated cavity off-axis spectroscopy (ABB®). We used the slope-intercept method to determine the

TABLE 1 Overview of sample sizes of measurements of body mass, physical activity, metabolic phenotype (glucose, HDL-cholesterol, LDL-cholesterol and triglycerides) and TEE in adult ring-tailed lemurs (*Lemur catta*) and adult Coquerel's sifakas (*Propithecus coquereli*) per study period.

Species	Period	Body mass	Activity	Blood parameters	TEE
Lemur catta	Winter	16	16	16	16
	Summer	15	15	15	12[a]
Propithecus coquereli	Winter	13	13	13	8
	Summer	14	14	14	2

Note: Sample size presented are total samples sizes obtained, including individuals for which we could not collect data in both study periods.

Abbreviation: TEE, total energy expenditure.

[a] TEE of 11 individuals was measured previously in winter, but one female died between both study periods and thus, we measured TEE of another female in the same ranging condition.

dilution spaces ND and NO and the depletion rates kD and kO for ^2H and ^{18}O, respectively (Berman et al., 2020; Pontzer, 2018; Speakman, 1997). We calculated the production rate of CO_2 from the single pool model as recommended for animals of this size (Speakman & Hambly, 2016; Speakman, 1997). Total body water (TBW) was determined from isotope dilution as:

$$\text{TBW} = (\text{NO}/1.007 + \text{ND}/1.041)/2$$

The average isotope dilution space ratio was 1.048 ± 0.011 (mean ± standard deviation [SD]) for ring-tailed lemurs and 1.076 ± 0.033 in sifaka. The rate of CO_2 production (mol day^{-1}) was calculated following equation 7.17 (Speakman, 1997):

$$rCO_2 = (N/2.078)*(kO - kD) - 0.0062*kD*N$$

CO_2 production was used to calculate total energy expenditure (TEE, kcal/day) using the Weir equation (Weir, 1949)

$$\text{TEE} = 22.4rCO_2(1.1 + 3.9/\text{FQ})$$

where FQ is the food quotient, which reflects the macronutrient content of the diet. We used a FQ = 0.9, and FFM was calculated from TBW assuming a hydration coefficient of 73.2% for FFM.

We calculated an adjusted TEE for each ring-tailed lemur based on a multiple regression model with TEE as the dependent variable and FFM and FM as independent variables. TEE, FFM, and FM were ln-transformed for this model, and we calculated FM by subtracting FFM from body mass. Using the predicted TEE and residual TEE for each measurement, we calculated adjusted TEE as

$$\text{adjusted TEE} = 100(1 + \text{Residual TEE}/\text{Predicted TEE})$$

Accordingly, an adjusted TEE of 110% indicates that the measured TEE that is 10% greater than predicted from body composition variables and an adjusted TEE of 90% is 10% less than predicted.

2.4 | Metabolic phenotype

DLC staff weighed each individual on the first and the last day of each measurement period, and we used the average of both measurements for all analyses. On the last day of each measurement period, 1 mL of blood was drawn from the femoral vein by DLC veterinary staff. The area of the blood draw was disinfected before the blood draw. Blood was allowed to clot for 15–20 min before samples were centrifuged for 10 min. Serum samples were stored at −80°C, and serum glucose, HDL- and LDL-cholesterol and triglycerides were measured at the Duke Metabolic Phenotyping Institute.

2.5 | Daily physical activity

All individuals were fitted with a MotionWatch8® (CamNtech) actigraphy data logger (weight: 7 g). Accelerometers were worn around the neck on a light nylon collar, 24 h per day for the entirety of each 1-week measurement period. The MotionWatch 8® sensor logs motion data using a built-in tri-axial accelerometer. The sensor samples data at 50 Hz and accumulates data over a user-defined epoch, and assigns an integer value on a ratio scale. We calculated an average activity level by dividing the sum of all activity values (of the entire 1-week period) by the number of sampling intervals.

2.6 | Statistical analyses

We used paired *t*-tests to assess changes in body mass, activity, blood glucose, HDL- and LDL-cholesterol and triglyceride levels between both study periods in both species, and to assess changes in (unadjusted and adjusted) TEE, FFM and body fat percentage in ring-tailed lemurs only. We used Welch Two Sample *t*-tests to assess whether free-rangers and nonfree-rangers differed in body mass, activity, blood parameters and (unadjusted and adjusted) TEE in summer (individuals were not free-ranging in winter). To meet assumptions, namely normally distributed residuals, we log-transformed glucose, HDL-cholesterol and FFM in ring-tailed lemurs, and in sifaka we log-transformed glucose and used a Box-Cox transformation for body mass.

2.7 | Changes in activity and metabolic phenotype

We used Welch Two Sample *t*-tests to assess whether individuals that were free-rangers in summer differed in the change of activity and metabolic phenotype from individuals that did not free-range in summer. We used linear models to assess the relationship between changes in physical activity and metabolic phenotype in both species together. We calculated the change in a parameter by subtracting the winter value from the summer value for the respective parameter. We used the change in activity, glucose, LDL- and HDL-cholesterol and triglyceride level as response variable in both species, and unadjusted TEE, adjusted TEE and body fat percentage as response variables in ring-tailed lemurs only. We included change in activity (not in the activity model), change in mass (in %; not in the model using adjusted TEE), species and sex as explanatory variables. Because activity and other parameters can decline with age, we also included age (at the first sampling period in winter) in the models. We tested for two-way interactions and removed nonsignificant interaction terms. We tested if model assumptions were met using the performance package (Lüdecke et al., 2021) and if required, transformed data to reach normally distributed residuals.

2.8 | Age-related changes

We used linear mixed models (LMMs) to assess age-related changes in activity, body mass and blood parameters. For these models, we made use of all available data (33 measurements of ring-tailed lemurs and 27 measurements of sifaka). In mammals, body mass increases with age until an asymptote is reached (Sebens, 1987; West et al., 2001), and in many species a quadratic relationship between age and body mass has been reported, where body mass decreases again at old age (Nussey et al., 2011; Pépin et al., 1996; Tafani et al., 2013). Because the relationships between age and body mass, markers of metabolic health, body fat percentage and adjusted TEE are likely nonlinear, we included age and its quadratic term, activity (not in the models for activity, body mass, adjusted TEE and body fat percentage), sex, season and species (not in models for adjusted TEE and body fat percentage) as explanatory variables and individual ID as random factor. We tested if model assumptions were met using the performance package (Lüdecke et al., 2021). To meet model assumptions, namely normally distributed residuals, we log-transformed activity, body mass, HDL-cholesterol, LDL-cholesterol, triglycerides and blood glucose before running models. We also removed one strong outlier of blood glucose (>three SDs above mean) before running the model. We tested for two-way interactions and removed nonsignificant interaction terms. LMMs for activity and triglycerides had a singular fit. When comparing a model with and without the random factor ID, there was no significant difference for triglycerides and activity, and for both parameters, the linear model had a smaller AIC (Akaike Information Criterion), and thus, we used a linear model for triglycerides and activity.

3 | RESULTS

Both ring-tailed lemurs and sifaka had a lower body mass and were more active in summer compared to winter (Table 2; Figure 1a,b and 2a,b). Ring-tailed lemurs had lower triglyceride levels in summer (Table 2 and Figure 1f), while sifaka had higher triglyceride levels in summer (Table 2 and Figure 2c). Blood glucose, HDL- and LDL-cholesterol levels did not differ between both periods in either species (Table 2; Figure 1 and Figure 2). Unadjusted TEE (Table 2; Figure 1d) and adjusted TEE (Paired *t* test: $t = 5.0205$, $df = 10$, $p = 0.0005$, Cohen's $d = 1.51$) of ring-tailed lemurs were higher in summer. FFM and body fat percentage of ring-tailed lemurs trended lower in the summer but did not reach the $p < 0.05$ threshold for statistical significance (Table 2 and Figure 1c,e).

In ring-tailed lemurs, summer activity did not differ between free-rangers ($N = 10$) and nonfree-rangers ($N = 5$), even when excluding the oldest individual that was also the only individual less active in summer (Welch Two Sample *t*-test, all $p > 0.22$). Free-rangers and nonfree-rangers also did not differ with regard to HDL- and LDL-cholesterol, glucose and triglyceride levels in summer (Welch Two Sample *t*-test, all $p > 0.26$). In summer, adjusted TEE was higher in nonfree-rangers ($N = 3$) compared to free-rangers ($N = 8$; Welch Two Sample *t*-test: $t = -3.517$, $df = 5.335$, $p = 0.015$, Cohen's $d = -2.19$), even when excluding the oldest and only individual less active in summer ($t = -3.185$, $df = 5.248$, $p = 0.023$, Cohen's $d = -2.07$). In Coquerel's sifakas, free-rangers ($N = 8$) and nonfree-rangers ($N = 3$) did not differ in activity, LDL-cholesterol, glucose and triglyceride levels in summer (Welch Two Sample t-test, all $p > 0.24$). HDL-cholesterol was higher in nonfree-rangers than in free-rangers ($t = -3.731$, $df = 8.997$, $p = 0.005$, Cohen's $d = -2.00$).

3.1 | Changes in activity and metabolic phenotype

There were no significant differences in the change of parameters between free-rangers and nonfree-rangers in ring-tailed lemurs (Welch Two Sample *t* tests: all $p > 0.22$) or Coquerel's sifakas (Welch Two Sample *t* tests: all $p > 0.37$). Individuals that increased their activity more from winter to summer also lost more body mass (LM: Estimate = −4.52, confidence interval [CI] = −8.69 to −0.35, $p = 0.035$; Adjusted $R^2 = 0.15$). Age at first measurement did not explain the extent to which individuals changed their activity ($p = 0.35$). Change in triglyceride level was not related to change in activity ($p = 0.79$), change in mass ($p = 0.42$) or age ($p = 0.13$). Change in triglyceride level was higher in male than in female sifaka, but there was no sex difference in ring-tailed lemurs (LM: interaction sex × species: Estimate = 99.87, CI = 23.98–175.76, $p = 0.013$; adjusted $R^2 = 0.59$). Change in HDL-cholesterol was higher in males than in females (Estimate = 3.69, CI = 2.17–5.21, $p < 0.001$) and individuals that lost more body mass, also had a positive change in HDL-cholesterol (i.e., higher values in summer; Estimate = 0.17, CI = −0.32 to −0.03, $p = 0.023$; adjusted $R^2 = 0.55$; Figure 3). Changes in body fat percentage, LDL-cholesterol, glucose, unadjusted TEE and adjusted TEE were not related to change in activity, sex, age or changes in body mass (all $p \geq 0.11$).

TABLE 2 Overview of average (±SD) activity, body mass, fat-free mass (FFM), body fat percentage, unadjusted TEE and blood parameters of adult ring-tailed lemurs (*Lemur catta*) and Coquerel's sifakas (*Propithecus coquereli*) measured both in winter (low activity condition) and summer (high activity condition).

Lemur catta	*N*	Winter	Summer	Paired *t* tests (*d* = Cohen's *d*)
Activity	15	232.5 ± 52.2	308.4 ± 52.5	$t = 5.243$, $p = 0.0001$, $d = 1.35$
Body mass (kg)	15	2.71 ± 0.31	2.47 ± 0.20	$t = -3.886$, $p = 0.0017$, $d = -1.00$
Blood glucose (mg/dL)	15	146.8 ± 66.4	129.8 ± 38.0	$t = -1.945$, $p = 0.3605$, $d = -0.24$
Triglycerides (mg/dL)	15	36.7 ± 14.2	27.5 ± 10.5	$t = -2.432$, $p = 0.0290$, $d = -0.63$
HDL-cholesterol (mg/dL)	15	59.0 ± 17.4	63.7 ± 16.5	$t = 1.370$, $p = 0.1921$, $d = 0.35$
LDL-cholesterol (mg/dL)	15	19.7 ± 6.6	17.8 ± 6.3	$t = -1.710$, $p = 0.1094$, $d = -0.44$
FFM (kg)	11	2.25 ± 0.19	2.14 ± 0.17	$t = -1.837$, $p = 0.0961$, $d = -0.55$
Body fat (%)	11	17.4 ± 6.6	12.0 ± 6.3	$t = -1.986$, $p = 0.075$, $d = -0.60$
Unadjusted TEE (kcal/d)	11	166.1 ± 15.7	176.7 ± 17.1	$t = 3.366$, $p = 0.0072$, $d = 1.01$
Propithecus coquereli				
Activity	11	195.3 ± 55.0	263.4 ± 41.6	$t = 3.675$, $p = 0.0043$, $d = 1.11$
Body mass (kg)	11	4.00 ± 0.68	3.84 ± 0.65	$t = -3.836$, $p = 0.0033$, $d = -1.16$
Blood glucose (mg/dL)	11	120.8 ± 15.1	118.5 ± 13.1	$t = -0.382$, $p = 0.7102$, $d = -0.12$
Triglycerides (mg/dL)	11	104.9 ± 45.6	177.5 ± 63.4	$t = 2.962$, $p = 0.0142$, $d = 0.89$
HDL-cholesterol (mg/dL)	11	129.8 ± 43.0	123.9± 40.1	$t = -0.905$, $p = 0.3869$, $d = -0.27$
LDL-cholesterol (mg/dL)	11	22.2 ± 11.7	22.3 ± 9.3	$t = 0.036$, $p = 0.9717$, $d = 0.01$

Note: Differences in sample sizes stem from logistic and health issues, especially from problems occurring during administering of the DLW dose in sifaka.

3.2 | Age-related changes

Activity decreased with age in both species (Table 3 and Figure 4a). Body mass of ring-tailed lemurs did not change significantly with age, while body mass of sifaka increased with age (Table 3 and Figure 4b). Blood glucose levels decreased with age in ring-tailed lemurs (age: Estimate = 0.04, CI = −0.00 to 0.08, $p = 0.066$; age^2: Estimate = −0.00, CI = −0.00 to −0.00, $p = 0.014$; conditional $R^2 = 0.39$; Figure 4c). Triglyceride level, HDL- and LDL-cholesterol did not show age-related changes (Figure 4). In ring-tailed lemurs, adjusted TEE (age: Estimate = 0.00, CI = −0.03 to 0.03, $p = 0.94$; age^2: Estimate = −0.00, CI = −0.00 to 0.00, $p = 0.66$; conditional $R^2 = 0.86$; Figure 4g) and body fat percentage (age: Estimate = −0.10, CI = −1.44 to 1.24, $p = 0.879$; age^2: Estimate = −0.01, CI = −0.05 to 0.03, $p = 0.642$; adjusted $R^2 = 0.23$; Figure 4h) did not decrease with age.

4 | DISCUSSION

As expected, both lemur species showed a higher level of activity in summer compared to winter. Individuals that showed larger increases in activity from winter to summer also lost more body mass. Individuals that lost more body mass, also had a positive change in HDL-cholesterol (i.e., higher values in summer). But changes in activity were not associated with changes in markers of metabolic health, suggesting that increases in physical activity are associated with only few improvements in markers of cardiometabolic health in both lemur species.

Both lemur species were more active in summer than in winter, and this pattern was independent of ranging status. Individuals that did not free-range also showed an increased activity level in summer, indicating that there might be a seasonal shift in activity level due to external cues such as photoperiod or ambient temperature that trigger changes in activity level independent of space available. Photoperiod and temperature have previously been identified as cues for adjustments in activity patterns in birds, mammals and primates (Daan & Aschoff, 1975; Kavanau & Peters, 1976).

In contrast to our predictions, we found little evidence that increases in physical activity were associated with improvements in markers of cardiometabolic health in either lemur species. Individuals that lost more body mass, also had a positive change in HDL-cholesterol (i.e., higher values in summer). However, change in activity was not related to changes in triglyceride levels, LDL- and HDL-cholesterol or glucose of either species, and also not to changes in body fat percentage, unadjusted TEE or adjusted TEE of ring-tailed lemurs. Similarly, in zoo elephants, distance walked was not correlated with the amount of FM relative to body mass or blood glucose (*E. maximus*) (Chusyd et al., 2021), or with body condition, health or behavioral outcomes (*E. maximus* and *L. africana*) (Holdgate et al., 2016). In contrast, another study found that increased walking exercise (≥14 h/week) was associated with a decreased risk of obesity in zoo (*L. africana* and *E. maximus*) elephants (Morfeld et al., 2016).

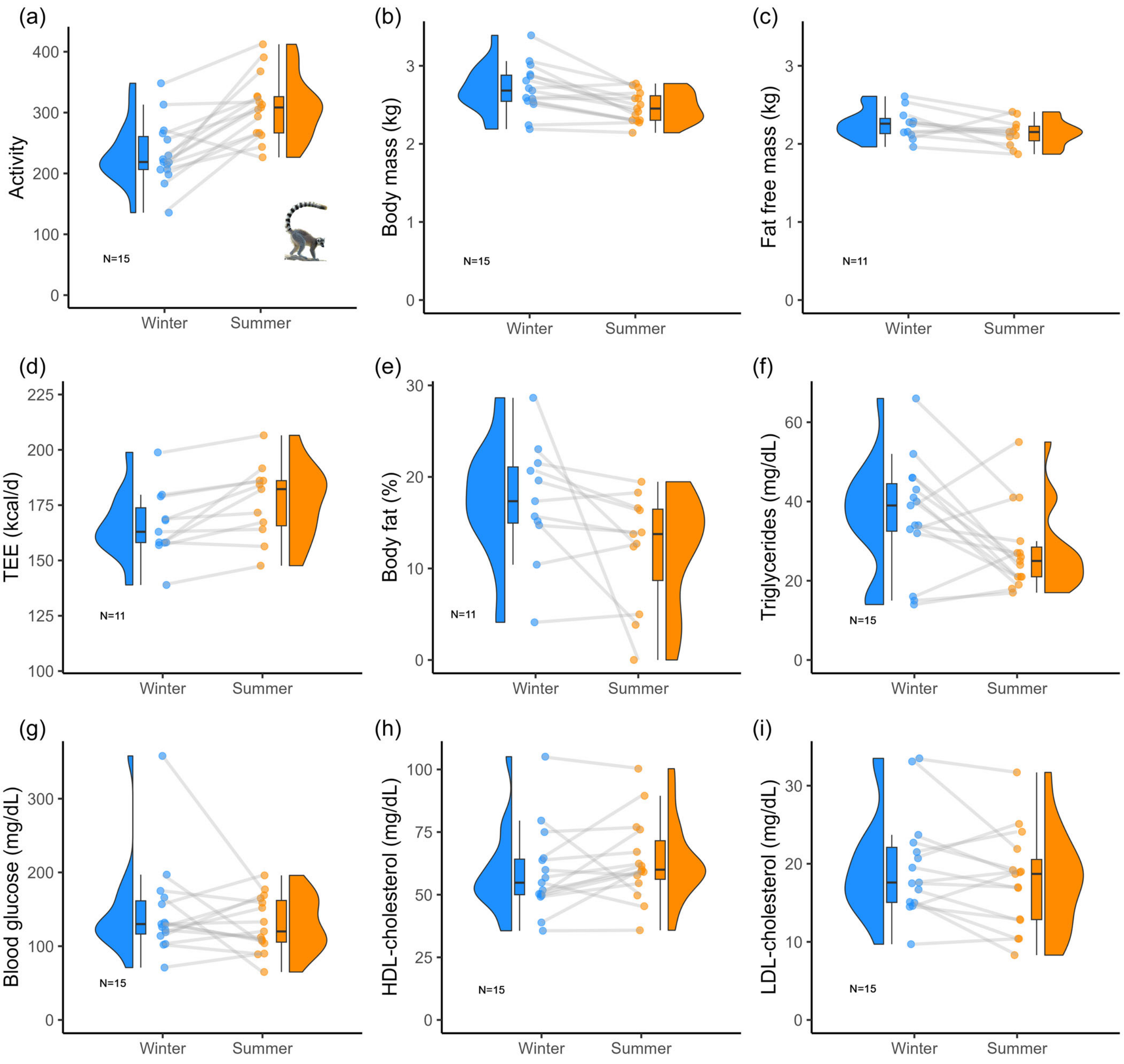

FIGURE 1 Intraindividual changes in activity, body mass, body composition, total energy expenditure and metabolic health parameters from winter (low activity condition; blue) to summer (high activity condition, orange) in ring-tailed lemurs (*Lemur catta*). Individuals were more active (a), had a lower body mass (b), higher unadjusted TEE (d) and lower triglyceride levels (f) in summer compared to winter. FFM (c) and body fat percentage (e) showed a trend towards higher measurements in winter. Differences in blood glucose (g), HDL-cholesterol (h) and LDL-cholesterol (i) between low activity and high activity conditions were not significant. Note the truncated y-axis in (d). The half-violin plots represent kernel density plots, boxplots show median values as well as 1st and 3rd quartiles, points represent individual values and repeated data points from the same individual are connected by gray lines (number of repeatedly measured individuals is presented). The species picture was taken from Pixabay (https://pixabay.com/images/).

There are several possible explanations for the lack of correspondence between physical activity and metabolic markers in this study. First, it is possible that the methods used to measure activity did not capture its most salient aspects. We used accelerometry to calculate an average activity level for a week-long period. It could be that activity intensity, not average activity level, more strongly influences blood profiles and cardiometabolic health. Total or average daily activity is generally considered a useful and reliable measure in human studies (Sjöros et al., 2020), but the effect of total activity time vs the effect of activity intensity on metabolic health is not well studied in animals. Second, although ring-tailed lemurs in this study had a larger body mass than individuals in the wild, blood glucose and blood lipid values fall within reference values for the species (Table 4).

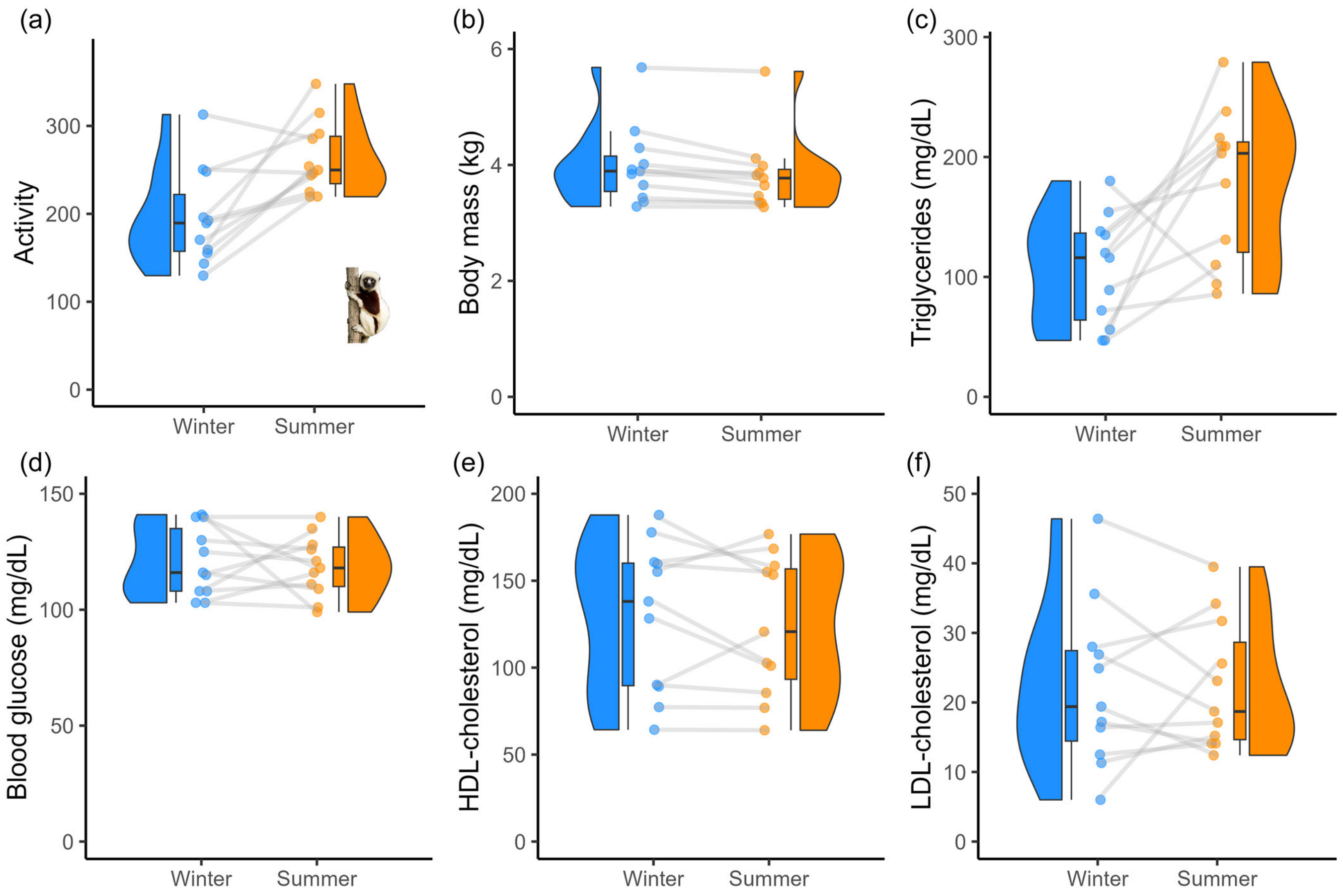

FIGURE 2 Intraindividual changes in activity, body mass and metabolic health parameters from winter (low activity condition; blue) to summer (high activity condition; orange) in 11 adult Coquerel's sifakas (*Propithecus coquereli*). Individuals were more active (a), had a lower body mass (b) and higher triglyceride levels (c) in summer compared to winter. Differences in blood glucose (d), HDL-cholesterol (e) and LDL-cholesterol (f) between low activity and high activity condition were not significant. The half-violin plots represent kernel density plots, boxplots show median values as well as 1st and 3rd quartiles, points represent individual values and repeated data points from the same individual are connected by gray lines. The species picture was taken from Pixabay (https://pixabay.com/images/).

Reference values from wild Coquerel's sifaka or other individuals in human care are not available for comparison. Body mass of sifaka included in this study (3.91 ± 0.61 kg) is well within the range reported for species (3.7–4.3 kg) (DLC, 2023; Mittermeier et al., 2013; Pontzer, 2023). None of study individuals in the low activity condition were largely overweight or obese. Thus, it may be that—at least in ring-tailed lemurs—increases in activity level from winter to summer did not lead to improvements of an already "healthy" blood profile. But irrespective of why individuals are active, the simple act of being active may be beneficial, as physical activity is known to improve health, independent of loss of body or FM or changes in blood lipids (Di Blasio et al., 2014).

Third, we found large between-individual variation regarding absolute changes and the direction of changes in metabolic phenotype (Figure 1 and 2). In humans, cardiometabolic responses to exercise can vary greatly between individuals. Intervention studies have found that most individuals show improved blood profiles (e.g., glucose and cholesterol), heart rate, blood pressure, among other metabolic variables, in response to weeks- or even months-long exercise interventions (Bonafiglia et al., 2016; Bouchard & Rankinen, 2001), but the range of responses is varied and some individuals show no improvement while others show declines (Bonafiglia et al., 2016; Bouchard & Rankinen, 2001). The causes of this heterogeneity in metabolic response to changes in exercise are an area of active research (Bonafiglia et al., 2016; Marsh et al., 2020), and genetic predisposition and genetic relatedness seem to be one factor for the observed variation, at least in humans (Claude Bouchard, 2019; Tremblay et al., 1997). The large between-individual variation in changes in metabolic phenotype together with the restricted sample size of 26 individuals in this study may account for the lack of relationships between increases in activity and improved cardiometabolic health in the study populations. Additional studies with larger sample sizes, potentially with individuals living in different housing conditions (e.g., research facilities, zoos) will show how generalizable the results of this study are.

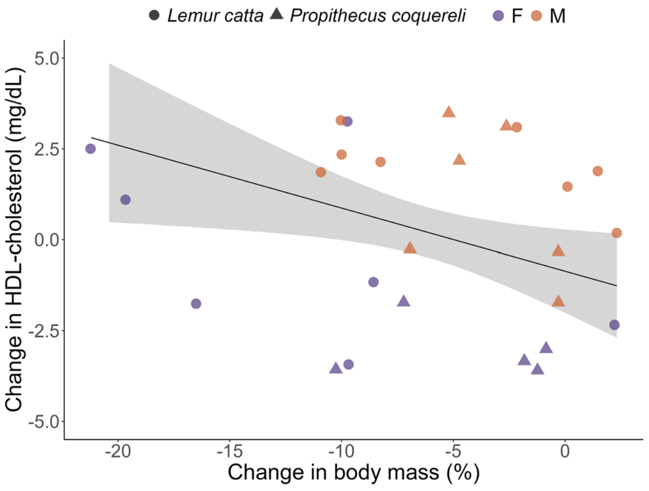

FIGURE 3 Changes in HDL-cholesterol values in 15 adult ring-tailed lemurs (*Lemur catta*, indicated by points) and 11 adult Coquerel's sifakas (*Propithecus coquereli*, indicated by triangles). Larger decreases in body mass were associated with a higher HDL-cholesterol values in summer compared to winter (i.e., positive change in HDL-cholesterol). Model estimates (lines) and confidence bands (shaded areas) for the fitted values based on standard errors computed from the covariance matrix of the fitted regression coefficients are shown (purple indicates female and orange indicates males).

4.1 | Age-related changes

We found an age-related decline in activity in both lemur species. Similar findings have been reported in a long-lived bird, the wandering Albatross (*Diomedea exulans*), Asian and African elephants (*E. maximus* and *L. africana*) and humans (Chusyd et al., 2021; Holdgate et al., 2016; Lecomte et al., 2010; Wolff-Hughes et al., 2015). Body mass and body condition decline with age in some wild mammals (Altmann et al., 2010; Hoffman et al., 2010; Nussey et al., 2011; Tafani et al., 2013), whereas other species, such as naked-mole rats (*Heterocephalus glaber*), show no age-related change in body mass (O'Connor et al., 2002). In gray mouse lemurs (*Microcebus murinus*), an age-related decline in body mass occurs in captivity, but not in the wild (Hämäläinen et al., 2014). We found that adult body mass did not decline with age in ring-tailed lemurs, similar to results from wild and captive ring-tailed lemurs (Clauss et al., 2021; Koyama et al., 2008). Body fat percentage and adjusted TEE of ring-tailed lemurs did not change with age. Blood glucose levels decreased with age in ring-tailed lemurs in this study. In contrast, blood glucose levels increase with age in rhesus macaques (*Macaca mulatta*) (Mattison et al., 2012). We found no other age-related changes in LDL- and HDL-cholesterol or triglycerides. Body mass and triglycerides increased during ageing in laboratory

TABLE 3 Results of a linear model to test for age-related changes in activity level (left panel) and a linear mixed model to test for age-related changes in body mass (right panel) of adult ring-tailed lemurs (*Lemur catta*; *N* = 33 measurements) and adult Coquerel's sifaka (*Propithecus coquereli*; *N* = 27 measurements).

log (activity)				**log (body mass)**			
	Estimate	**CI**	***p***		**Estimate**	**CI**	***p***
(Intercept)	6.10	5.83–6.38	<0.001	(Intercept)	1.04	0.81–1.26	<0.001
Age	−0.05	−0.09 to −0.01	0.008	Age	−0.00	−0.04 to 0.03	0.885
Age^2	0.00	−0.00 to 0.00	0.073	Age^2	−0.00	−0.00 to 0.00	0.683
Sex [M]	0.00	−0.12 to 0.13	0.940	Sex [M]	−0.05	−0.14 to 0.04	0.294
Season [Winter]	−0.17	−0.29 to −0.06	0.003	Season [Winter]	0.07	0.04–0.10	<0.001
Species [*P. coquereli*]	−0.30	−0.43 to −0.18	<0.001	Species [*P. coquereli*]	0.18	0.07–0.29	0.002
				Age^2 × species	0.00	0.00–0.00	0.001
				Random Effects			
				σ2	0.003		
				ICC	0.789		
$N_{\text{Individuals}}$	31			$N_{\text{Individuals}}$	31		
Observations	60			Observations	60		
R^2	0.420			Marginal R^2	0.721		
Adjusted R^2	0.366			Conditional R^2	0.941		

Note: The marginal R^2 considers only the variance of the fixed effects, while the conditional R^2 takes both the fixed and random effects into account. Abbreviations: σ2, variance of the random effect "individuals ID"; ICC, Intraclass coefficient of variation.

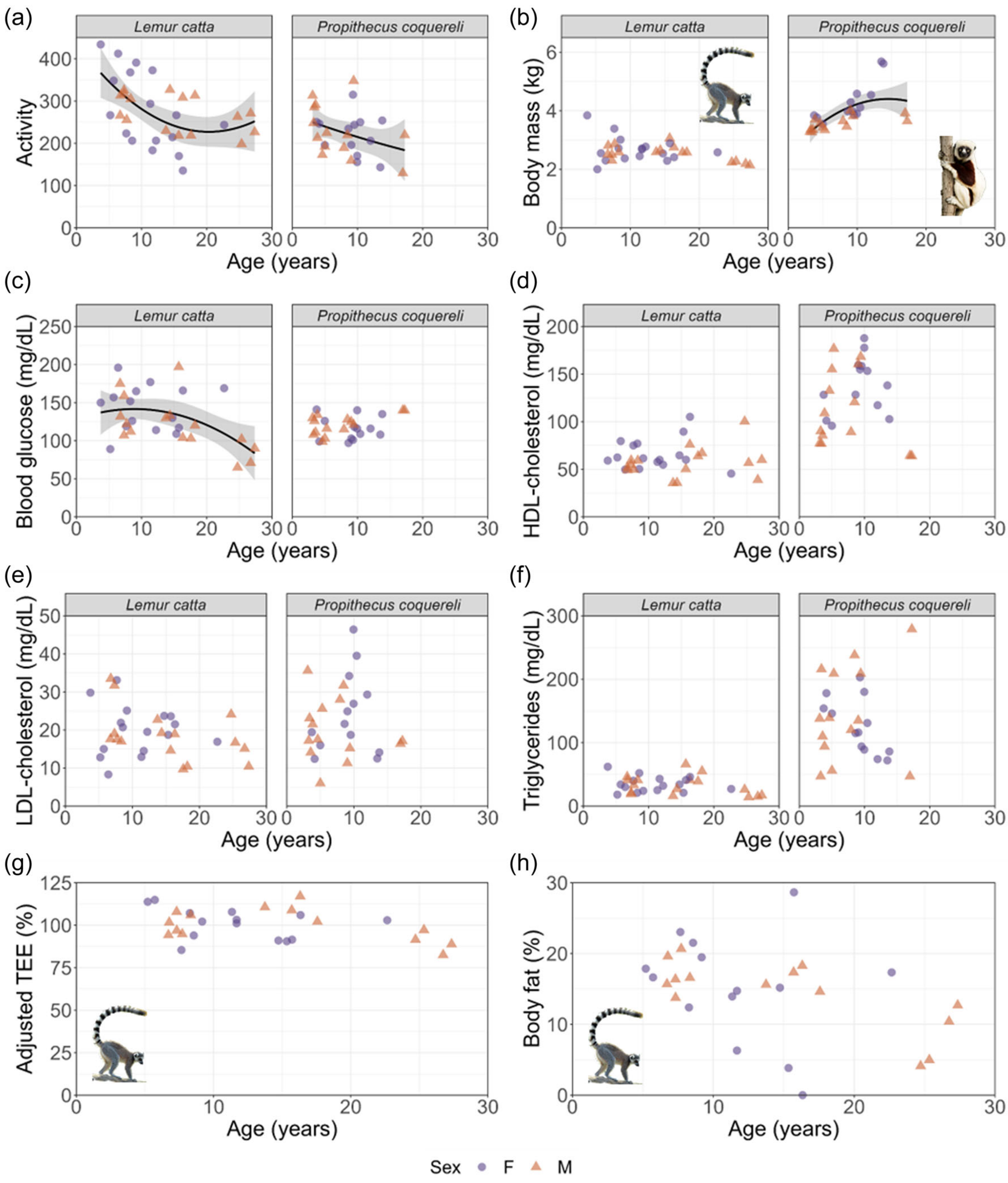

FIGURE 4 Age-related changes in adult ring-tailed lemurs (*Lemur catta*; *N* = 33 measurements) and adult Coquerel's sifakas (*Propithecus coquereli*; *N* = 27 measurements). Activity level decreased with age in both species (a), body mass increased with age in sifaka but not in ring-tailed lemurs (b), blood glucose decreased with age in ring-tailed lemurs, but not sifaka (c), and other markers of metabolic health did not show age-related changes (d–f). Adjusted TEE and body fat percentage of ring-tailed lemurs (*N* = 28 measurements) showed no age-related changes (g, h). Black lines show quadratic regression lines and shading represents 95% confidence intervals (purple points indicate females and orange triangles indicate males). Species pictures were taken from Pixabay (https://pixabay.com/images/).

populations of chimpanzees (*Pan troglodytes*), and these populations also showed age-related decreases of total cholesterol, HDL- and LDL-cholesterol (Cole et al., 2020). Body mass increased with age in sifaka, but they showed no other age-related change in markers of cardiometabolic health. These results may be an important consideration for the management of older sifaka in human care.

4.2 | Summary

Seasonal changes in the management and housing of captive primates offer a potentially powerful approach for studying the effects of physical activity. In the ring-tailed lemur and sifaka populations in this study daily physical activity increased during the summer months, but these changes were not strongly related to

TABLE 4 Body mass and blood lipid values of ring-tailed lemurs (*Lemur catta*) housed at Duke Lemur Center, Durham, USA, other captive colonies, and free-ranging individuals from Beza Mahafaly and Tsimanampetsotsa Strict Nature Reserve (TSNR), Madagascar.

	DLC winter[a]	DLC summer[a]		Beza Mahafaly[b]		TSNR[c]		captive[d]	
Parameter	Mean ± SD	Mean ± SD	*n*	Mean ± SD	*n*	Mean ± SD	*n*	Mean ± SD	*n*
Body mass (kg)	2.71 ± 0.31	2.47 ± 0.20	15	2.2 ± 0.2	70	1.99 ± 0.34	19	-	-
Blood glucose (mg/dL)	146.8 ± 66.4	129.8 ± 38.0	15	188.6 ± 80.1	27	136 ± 36.9	20	142 ± 75	901
Triglycerides (mg/dL)	36.7 ± 14.2	27.5 ± 10.5	15	43.0 ± 10.4	27	20.8 ± 7.03	20	69 ± 35	472
Cholesterol (mg/dL)	-	-	-	16.4 ± 15.3	27	44.3 ± 7.4	19	89 ± 26	768
HDL-cholesterol (mg/dL)	59.0 ± 17.3	63.6 ± 16.5	15	-	-	-	-	-	-
LDL-cholesterol (mg/dL)	19.6 ± 6.6	17.8 ± 6.3	15	-	-	-	-	-	-

[a] This study;

[b] (Miller et al., 2007);

[c] (Dutton et al., 2003);

[d] (Teare, 2002).

common biomarkers of metabolic health or total energy expenditure. Increased physical activity was apparent for individuals housed indoors as well as those in large outdoor enclosures, suggesting the change in activity may owe more to seasonal environmental cues than to enclosure size. Age-related changes in body mass and activity may be important considerations in the management of older primates in human care.

AUTHOR CONTRIBUTIONS

Rebecca Rimbach and Herman Pontzer designed the study; Rebecca Rimbach collected and analyzed the data; Rebecca Rimbach drafted the manuscript; Herman Pontzer revised the manuscript; all authors gave final approval for publication.

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CONFLICT OF INTEREST STATEMENT

The authors declare no conflict of interest.

DATA AVAILABILITY STATEMENT

The data and source code that support the findings of this study are openly available in the OSF repository at https://doi.org/10.17605/OSF.IO/TG5QP (https://osf.io/tg5qp/).

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REFERENCES

Abdelaal, M., le Roux, C. W., & Docherty, N. G. (2017). Morbidity and mortality associated with obesity. *Annals of Translational Medicine*, *5*(7), 161. https://doi.org/10.21037/atm.2017.03.107

Alessio, H. M., Hagerman, A. E., Nagy, S., Philip, B., Byrnes, R. N., Woodward, J. L., Callahan, P., & Wiley, R. L. (2005). Exercise improves biomarkers of health and stress in animals fed ad libitum. *Physiology & Behavior*, *84*(1), 65–72. https://doi.org/10.1016/j.physbeh.2004.10.010

Altmann, J., Gesquiere, L., Galbany, J., Onyango, P. O., & Alberts, S. C. (2010). Life history context of reproductive aging in a wild primate model. *Annals of the New York Academy of Sciences*, *1204*, 127–138. https://doi.org/10.1111/j.1749-6632.2010.05531.x

Altmann, J., Schoeller, D., Altmann, S. A., Muruthi, P., & Sapolsky, R. M. (1993). Body size and fatness of free-living baboons reflect food availability and activity levels. *American Journal of Primatology*, *30*(2), 149–161. https://doi.org/10.1002/ajp.1350300207

Association of Zoos and Aquariums. (2023). https://www.aza.org/

Aune, D., Norat, T., Leitzmann, M., Tonstad, S., & Vatten, L. J. (2015). Physical activity and the risk of type 2 diabetes: a systematic review and dose-response meta-analysis. *European Journal of Epidemiology*, *30*(7), 529–542. https://doi.org/10.1007/s10654-015-0056-z

Banks, W. A., Altmann, J., Sapolsky, R. M., Phillips-Conroy, J. E., & Morley, J. E. (2003). Serum leptin levels as a marker for a syndrome X-like condition in wild baboons. *The Journal of Clinical Endocrinology & Metabolism*, *88*(3), 1234–1240. https://doi.org/10.1210/jc.2002-021695

Bennett, G., Malone, M., Sauther, M. L., Cuozzo, F. P., White, B., Nelson, K. E., Stumpf, R. M., Knight, R., Leigh, S. R., & Amato, K. R. (2016). Host age, social group, and habitat type influence the gut microbiota of wild ring-tailed lemurs (*Lemur catta*). *American Journal of Primatology*, *78*(8), 883–892. https://doi.org/10.1002/ajp.22555

Berman, E. S. F., Swibas, T., Kohrt, W. M., Catenacci, V. A., Creasy, S. A., Melanson, E. L., & Speakman, J. R. (2020). Maximizing precision and accuracy of the doubly labeled water method via optimal sampling protocol, calculation choices, and incorporation of 17O measurements. *European Journal of Clinical Nutrition*, *74*, 454–464. https://doi.org/10.1038/s41430-019-0492-z

Di Blasio, A., Izzicupo, P., D'Angelo, E., Melanzi, S., Bucci, I., Gallina, S., Di Baldassarre, A., & Napolitano, G. (2014). Effects of patterns of walking training on metabolic health of untrained postmenopausal women. *Journal of aging and physical activity*, *22*(4), 482–489. https://doi.org/10.1123/JAPA.2013-0043

Bonafiglia, J. T., Rotundo, M. P., Whittall, J. P., Scribbans, T. D., Graham, R. B., & Gurd, B. J. (2016). Inter-individual variability in the adaptive responses to endurance and sprint interval training: a randomized crossover study. *PLoS One*, *11*(12), e0167790. https://doi.org/10.1371/journal.pone.0167790

Bouchard, C. (2019). DNA sequence variations contribute to variability in fitness and trainability. *Medicine & Science in Sports & Exercise*, *51*(8), 1781–1785. https://doi.org/10.1249/MSS.0000000000001976

Bouchard, C. & Rankinen, T. (2001). Individual differences in response to regular physical activity. *Medicine and Science in Sports and Exercise*, *33*, S446–S451. https://doi.org/10.1097/00005768-200106001-00013

Chusyd, D. E., Nagy, T. R., Golzarri-arroyo, L., Dickinson, S. L., Speakman, J. R., Hambly, C., Johnson, M. S., Allison, D. B., & Brown, J. L. (2021). Adiposity, reproductive and metabolic health, and activity levels in zoo Asian elephant (*Elephas maximus*). *Journal of Experimental Biology*, *224*, jeb219543. https://doi.org/10.1242/jeb.219543

Clauss, M., Trümpler, J., Ackermans, N. L., Kitchener, A. C., Hantke, G., Stagegaard, J., Takano, T., Shintaku, Y., & Matsuda, I. (2021). Intraspecific macroscopic digestive anatomy of ring-tailed lemurs (*Lemur catta*), including a comparison of frozen and formalin-stored specimens. *Primates*, *62*(2), 431–441. https://doi.org/10.1007/s10329-020-00873-8

Clay, A. W., Crane, M. M., & Bloomsmith, M. A. (2022). Weight management towards physiological and behavioral wellbeing for chimpanzees living under human care. *Zoo Biology*, *41*(3), 200–217. https://doi.org/10.1002/zoo.21668

Coin, A., Sergi, G., Minicuci, N., Giannini, S., Barbiero, E., Manzato, E., Pedrazzoni, M., Minisola, S., Rossini, M., Del Puente, A., Zamboni, M., Inelmen, E. M., & Enzi, G. (2008). Fat-free mass and fat mass reference values by dual-energy x-ray absorptiometry (DEXA) in a 20-80 year-old Italian population. *Clinical Nutrition*, *27*(1), 87–94. https://doi.org/10.1016/j.clnu.2007.10.008

Cole, M. F., Cantwell, A., Rukundo, J., Ajarova, L., Fernandez-Navarro, S., Atencia, R., & Rosati, A. G. (2020). Healthy cardiovascular biomarkers across the lifespan in wild-born chimpanzees (*Pan troglodytes*). *Philosophical Transactions of the Royal Society, B: Biological Sciences*, *375*(1811), 20190609. https://doi.org/10.1098/rstb.2019.0609

Cuozzo, F. P. & Sauther, M. L. (2004). Tooth loss, survival, and resource use in wild ring-tailed lemurs (*Lemur catta*): implications for inferring conspecific care in fossil hominids. *Journal of Human Evolution*, *46*(5), 623–631. https://doi.org/10.1016/j.jhevol.2004.02.004

Daan, S. & Aschoff, J. (1975). Circadian rhythms of locomotor activity in captive birds and mammals: their variations with season and latitude. *Oecologia*, *18*(4), 269–316. https://doi.org/10.1007/BF00345851

Duke Lemur Center. (2023). *Coquerel's sifaka*. https://lemur.duke.edu/discover/meet-the-lemurs/co

Dutton, A., Christopher, J., & Randall, E. (2003). Biomedical evaluation of free-ranging ring-tailed lemurs (*Lemur catta*) in Tsimanampetsotsa Strict Nature Reserve, Madagascar. *Journal of Zoo and Wildlife Medicine*, *34*(1), 16–24. https://doi.org/10.1638/1042-7260(2003)34[0016:beofrl]2.0.co;2

Elia, M., Ritz, P., & Stubbs, R. (2000). Total energy expenditure in the elderly. *European Journal of Clinical Nutrition*, *54*, S92–S103. https://doi.org/10.1038/sj.ejcn.1601030

Hämäläinen, A., Dammhahn, M., Aujard, F., Eberle, M., Hardy, I., Kappeler, P. M., Perret, M., Schliehe-Diecks, S., & Kraus, C. (2014). Senescence or selective disappearance? Age trajectories of body mass in wild and captive populations of a small-bodied primate. *Proceedings of the Royal Society B: Biological Sciences*, *281*, 20140830. https://doi.org/10.1098/rspb.2014.0830

Hoffman, C. L., Higham, J. P., Mas-Rivera, A., Ayala, J. E., & Maestripieri, D. (2010). Terminal investment and senescence in rhesus macaques (*Macaca mulatta*) on Cayo Santiago. *Behavioral Ecology*, *21*(5), 972–978. https://doi.org/10.1093/beheco/arq098

Holdgate, M. R., Meehan, C. L., Hogan, J. N., Miller, L. J., Soltis, J., Andrews, J., & Shepherdson, D. J. (2016). Walking behavior of zoo elephants: associations between GPS-measured daily walking distances and environmental factors, social factors and welfare indicators. *PLoS One*, *11*, e0150331.

Ingram, D. K. (2000). Age-related decline in physical activity: generalization to nonhumans. *Medicine & Science in Sports & Exercise*, *32*(9), 1623–1629. https://doi.org/10.1097/00005768-200009000-00016

Jänicke, B., Coper, D., & Jänicke, U.-A. (1986). Motor activity of different-aged cercopithecidae: silvered-leaf monkey (*Presbytis cristatus* Esch.), lion-tailed monkey (*Macaca silenus* L.), moor macaque (*Macaca maura* Cuv.) as observed in the zoological garden, Berlin (West). *Gerontology*, *32*, 133–140.

Kavanau, J. L. & Peters, C. R. (1976). Activity of nocturnal primates: influences of twilight zeitgebers and weather. *Science*, *191*(4222), 83–86.

Koyama, N., Aimi, M., Kawamoto, Y., Hirai, H., Go, Y., Ichino, S., & Takahata, Y. (2008). Body mass of wild ring-tailed lemurs in Berenty Reserve, Madagascar, with reference to tick infestation: A preliminary analysis. *Primates*, *49*(1), 9–15. https://doi.org/10.1007/s10329-007-0051-4

Kyle, U. G., Genton, L., Hans, D., Karsegard, V. L., Michel, J. P., Slosman, D. O., & Pichard, C. (2001). Total body mass, fat mass, fat-free mass, and skeletal muscle in older people: cross-sectional differences in 60-year-old persons. *Journal of the American Geriatrics Society*, *49*, 1633–1640. https://doi.org/10.1111/j.1532-5415.2001.49272.x

Lecomte, V. J., Sorci, G., Cornet, S., Jaeger, A., Faivre, B., Arnoux, E., Gaillard, M., Trouvé, C., Besson, D., Chastel, O., & Weimerskirch, H. (2010). Patterns of aging in the long-lived wandering albatross. *Proceedings of the National Academy of Sciences*, *107*(14), 6370–6375. https://doi.org/10.1073/pnas.0911181107

Lee, I.-M., Shiroma, E. J., Lobelo, F., Puska, P., Blair, S. N., & Katzmarzyk, P. T. (2012). Effect of physical inactivity on major non-communicable diseases worldwide: an analysis of burden of disease and life expectancy. *The Lancet*, *380*(9838), 219–229. https://doi.org/10.1016/S0140-6736(12)61031-9

Lifson, N. & McClintock, R. (1966). Theory of use of the turnover rates of body water for measuring energy and material balance. *Journal of Theoretical Biology*, *12*(1), 46–74. https://doi.org/10.1016/0022-5193(66)90185-8

Lüdecke, D., Ben-Shachar, M., Patil, I., Waggoner, P., & Makowski, D. (2021). performance: an R package for assessment, comparison and testing of statistical models. *Journal of Open Source Software*, *6*(6), 3139. https://doi.org/10.21105/joss.03139

Manini, T. M. (2010). Energy expenditure and aging. *Ageing Research Reviews*, *9*, 1–11. https://doi.org/10.1016/j.biotechadv.2011.08.021.Secreted

Marsh, C. E., Thomas, H. J., Naylor, L. H., Scurrah, K. J., & Green, D. J. (2020). Fitness and strength responses to distinct exercise modes in twins: studies of twin responses to understand exercise as a THerapy (STRUETH) study. *The Journal of Physiology*, *598*(18), 3845–3858. https://doi.org/10.1113/JP280048

Mattison, J. A., Roth, G. S., Beasley, T. M., Tilmont, E. M., Handy, A. M., Herbert, R. L., Longo, D. L., Allison, D. B., Young, J. E., Bryant, M., Barnard, D., Ward, W. F., Qi, W., Ingram, D. K., & De Cabo, R. (2012). Impact of caloric restriction on health and survival in rhesus monkeys from the NIA study. *Nature*, *489*(7415), 318–321. https://doi.org/10.1038/nature11432

McMillen, I. C., & Robinson, J. S. (2005). Developmental origins of the metabolic syndrome: prediction, plasticity, and programming. *Physiological Reviews*, *85*, 571–633. https://doi.org/10.1152/physrev.00053.2003

Meehan, C. L., Mench, J. A., Carlstead, K., & Hogan, J. N. (2016). Determining connections between the daily lives of zoo elephants and their welfare: an epidemiological approach. *PLoS One*, *11*, e0158124.

Mellor, E. L., Cuthill, I. C., Schwitzer, C., Mason, G. J., & Mendl, M. (2020). Large lemurs: ecological, demographic and environmental risk factors for weight gain in captivity. *Animals: An Open Access Journal from MDPI*, *10*(8), 1443. https://doi.org/10.3390/ani10081443

Miller, D. S., Sauther, M. L., Hunter-Ishikawa, M., Fish, K., Culbertson, H., Cuozzo, F. P., Campbell, T. W., Andrews, G. A., Chavey, P. S., Nachreiner, R., Rumbeiha, W., Stacewicz-Sapuntzakis, M., & Lappin, M. R. (2007). Biomedical evaluation of free-ranging ring-tailed lemurs (*Lemur catta*) in three habitats at the beza mahafaly special reserve, Madagascar. *Journal of Zoo and Wildlife Medicine*, *38*(2), 201–216. https://doi.org/10.1638/1042-7260(2007)038[0201:beofrl]2.0.co;2

Mittermeier, R. A., Schwitzer, C., Louis, J., Edward, E., & Richardson, M. C. (2013). Family Indriidae (Woolly Lemurs, Sifakas and Indri). In R. A. Mittermeier, A. B. Rylands, & D. E. Wilson (Eds.), *Handbook of the Mammals of the World—Primates*.

Morfeld, K. A., Meehan, C. L., Hogan, J. N., & Brown, J. L. (2016). Assessment of body condition in African (*Loxodonta africana*) and Asian (*Elephas maximus*) elephants in North American zoos and management practices associated with high body condition scores. *PLoS One*, *11*(7), e0155146. https://doi.org/10.1371/journal.pone.0155146

Nussey, D. H., Coulson, T., Delorme, D., Clutton-Brock, T. H., Pemberton, J. M., Festa-Bianchet, M., & Gaillard, J. M. (2011). Patterns of body mass senescence and selective disappearance differ among three species of free-living ungulates. *Ecology*, *92*(10), 1936–1947. https://doi.org/10.1890/11-0308.1

O'Connor, T. P., Lee, A., Jarvis, J. U. M., & Buffenstein, R. (2002). Prolonged longevity in naked mole-rats: age-related changes in metabolism, body composition and gastrointestinal function. *Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology*, *133*(3), 835–842. https://doi.org/10.1016/S1095-6433(02)00198-8

Pepin, D., Faivre, R., & Menaut, P. (1996). Factors affecting the relationship between body mass and age in the izard. *Journal of Mammalogy*, *77*(2), 351–358. https://doi.org/10.2307/1382807

Pontzer, H. (2018). Method and rationale for recalculating dilution spaces to a single, common time point in doubly labeled water studies. *European Journal of Clinical Nutrition*, *72*(12), 1620–1624. https://doi.org/10.1038/s41430-018-0361-1

Pontzer, H. (2023). The provisioned primate: patterns of obesity across lemurs, monkeys, apes and humans. *Philosophical Transactions of the Royal Society, B: Biological Sciences*, *378*, 20220218.

Pontzer, H., Wood, B. M., & Raichlen, D. A. (2018). Hunter-gatherers as models in public health. *Obesity Reviews*, *19*, 24–35. https://doi.org/10.1111/obr.12785

Pontzer, H., Yamada, Y., Sagayama, H., Ainslie, P. N., Andersen, L. F., Anderson, L. J., Arab, L., Baddou, I., Bedu-addo, K., Blaak, E. E., Blanc, S., Bonomi, A. G., Bouten, C. V. C., Bovet, P., Buchowski, M. S., Butte, N. F., Camps, S. G., Close, G. L., Cooper, J. A., … Speakman, J. R. (2021). Daily energy expenditure through the human life course. *Science*, *373*, 808–812.

Reimes, T., Nijssen, T., & Valente, L. (2021). Captive populations of lemurs in European zoos: mismatch between current species representation and ex-situ conservation needs. *Lemur News*, *23*, 61–67.

Rimbach, R., Amireh, A., Allen, A., Hare, B., Guarino, E., Kaufman, C., Salomons, H., & Pontzer, H. (2021). Total energy expenditure of bottlenose dolphins (*Tursiops truncatus*) of different ages. *Journal of Experimental Biology*, *224*, jeb242218. https://doi.org/10.1242/jeb.242218

Sallis, J. F. (2000). Age-related decline in physical activity: A synthesis of human and animal studies. *Medicine & Science in Sports & Exercise*, *32*(9), 1598–1600. https://doi.org/10.1097/00005768-200009000-00012

Schuler, G., Adams, V., & Goto, Y. (2013). Role of exercise in the prevention of cardiovascular disease: Results, mechanisms, and new perspectives. *European Heart Journal*, *34*(24), 1790–1799. https://doi.org/10.1093/eurheartj/eht111

Sebens, K. P. (1987). The ecology of indeterminate growth in animals. *Annual Review of Ecology and Systematics*, *18*(1), 371–407.

Sjöros, T., Vähä-Ypyä, H., Laine, S., Garthwaite, T., Lahesmaa, M., Laurila, S. M., Latva-Rasku, A., Savolainen, A., Miikkulainen, A., Löyttyniemi, E., Sievänen, H., Kalliokoski, K. K., Knuuti, J., Vasankari, T., & Heinonen, I. H. A. (2020). Both sedentary time and physical activity are associated with cardiometabolic health in overweight adults in a 1 month accelerometer measurement. *Scientific Reports*, *10*(1), 20578. https://doi.org/10.1038/s41598-020-77637-3

Speakman, J. R. (1997). *Doubly-labelled water: Theory and practice*. Chapman and Hall.

Speakman, J. R., Van Acker, A., & Harper, E. J. (2003). Age-related changes in the metabolism and body composition of three dog breeds and their relationship to life expectancy. *Aging cell*, *2*(5), 265–275. https://doi.org/10.1046/j.1474-9728.2003.00061.x

Speakman, J. R. & Hambly, C. (2016). Using doubly-labelled water to measure free-living energy expenditure: some old things to remember and some new things to consider. *Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology*, *202*, 3–9. https://doi.org/10.1016/j.cbpa.2016.03.017

Speakman, J. R., & Westerterp, K. R. (2010). Associations between energy demands, physical activity, and body composition in adult humans between 18 and 96 y of age. *The American Journal of Clinical Nutrition*, *92*, 826–834. https://doi.org/10.3945/ajcn.2009.28540.826

Tafani, M., Cohas, A., Bonenfant, C., Gaillard, J. M., Lardy, S., & Allainé, D. (2013). Sex-specific senescence in body mass of a monogamous and monomorphic mammal: the case of Alpine marmots. *Oecologia*, *172*(2), 427–436. https://doi.org/10.1007/s00442-012-2499-1

Teare, A. (2002). *Reference ranges for physiological values in captive wildlife*. International Species Information System.

Terranova, C. J. & Coffman, B. S. (1997). Body weights of wild and captive lemurs. *Zoo Biology*, *16*(1), 17–30. https://doi.org/10.1002/(sici)1098-2361(1997)16:1<17::aid-zoo4>3.0.co;2-e

The expert panel. (2001). Executive summary of the third report of The National cholesterol education program (NCEP) expert panel on detection, evaluation, and treatment of high blood cholesterol in adults (Adult Treatment Panel III). *JAMA: The Journal of the American Medical Association*, *285*, 2486–2497.

Tremblay, A., Poehlman, E. T., Després, J. P., Thériault, G., Danforth, E., & Bouchard, C. (1997). Endurance training with constant energy intake in identical twins: changes over time in energy expenditure and related hormones. *Metabolism: Clinical and Experimental*, *46*(5), 499–503. https://doi.org/10.1016/S0026-0495(97)90184-0

Weir, J. B. V. (1949). New methods for calculating metabolic rate with special reference to protein metabolism. *The Journal of Physiology*, *109*, 1–9.

West, G. B., Brown, J. H., & Enquist, B. J. (2001). A general model for ontogenetic growth. *Nature*, *413*, 628–631.

Wolff-Hughes, D. L., Fitzhugh, E. C., Bassett, D. R., & Churilla, J. R. (2015). Waist-worn actigraphy: population-referenced percentiles for total activity counts in U.S. adults. *Journal of Physical Activity and Health*, *12*(4), 447–453. https://doi.org/10.1123/jpah.2013-0464

Wood, B. M., Harris, J. A., Raichlen, D. A., Pontzer, H., Sayre, K., Sancilio, A., Berbesque, C., Crittenden, A. N., Mabulla, A., McElreath, R., Cashdan, E., & Jones, J. H. (2021). Gendered movement ecology and landscape use in Hadza hunter-gatherers. *Nature Human Behaviour*, *5*, 436–446. https://doi.org/10.1038/s41562-020-01002-7

Zehr, S. M., Roach, R. G., Haring, D., Taylor, J., Cameron, F. H., & Yoder, A. D. (2014). Life history profiles for 27 strepsirrhine primate taxa generated using captive data from the Duke Lemur Center. *Scientific Data*, *1*, 140019. https://doi.org/10.1038/sdata.2014.19

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