

Supporting Information (SI) text for

Pressure versus volume capabilities: A trade-off that has shaped the evolution of the human heart

Authors: Robert E. Shave^{1*†}, Daniel E. Lieberman^{2*†}, Aimee L. Drane³, Marcel G. Brown⁴, Alan M. Batterham⁵, Steven Worthington⁶, Rebeca Atencia^{2,7}, Yedra Feltrer², Jennifer Neary⁴, Rory B. Weiner⁴, Meagan M. Wafsy⁴, & Aaron L. Baggish^{4*†}

Correspondence to: rob.shave@ubc.ca, danlieb@fas.harvard.edu or abaggish@mgh.harvard.edu.

This PDF file includes:

Supporting Information
Figs. S1
Tables S1 to S11
Supporting Information References

Supporting Information

Research Subjects:

Chimpanzees and Gorillas. All chimpanzees included in this study were cared for at the Tchimpounga Rehabilitation Centre in Congo or at the Chimfunshi sanctuary in Zambia. On arrival at each of the sanctuaries, and every two to three years thereafter, each animal undergoes a complete health assessment, and in 2014 this included a full cardiac ultrasound examination conducted as part of the International Primate Heart Project (<http://primateheartproject.co.uk>). Healthy adult (>10 years of age) males (n=43) from these examinations were included in the initial comparison of cardiac structure and function between chimpanzees and humans. All chimpanzees included in the study were wild-born orphans confiscated by the wildlife authorities. The gorillas (n=5) included in the study were cared for in European zoological collections. Similar to the chimpanzees, all of the cardiac data from the gorillas were collected during routine veterinary procedures. The gorillas included were housed in zoological enclosures (Zoological Society of London UK, Paignton Zoo UK and Wilhelma Zoo, Stuttgart, Germany) that adhere to recognized husbandry guidelines (1). At the time of assessment all animals were asymptomatic and presumed healthy based on clinical observation and known history. Further detail regarding the animal sample and anesthetic protocols employed is provided below.

Justification of the Use of Semi-Natural Sanctuary Chimpanzees (Pan Troglodytes). It is not ethically appropriate to perform echocardiography or blood pressure assessment on wild chimpanzees (*Pan troglodytes*), necessitating the selection of semi-natural sanctuary chimpanzees undergoing regular health checks. Although semi-natural sanctuary chimpanzees are not wild there are several reasons to hypothesize that their activity levels do not differ

appreciably from those of wild chimpanzees. First, the semi-natural sanctuary chimpanzees studied have daily access to large enclosures of primary forest with dozens of edible plant species for *ad libitum* foraging and with a full range of climbing, nesting and trekking opportunities (1). The Chimfunishi sanctuary has 4 enclosures ranging from 19-77 hectares; the Tchimpounga sanctuary has 3 enclosures as large as 40 hectares. In semi-natural sanctuaries, mixed-sex groups of adult chimpanzees typically leave their dormitories in the morning to forage and explore in a rich social and physical environment prior to returning at night to feed and sleep (2). Chimpanzees in these semi-natural sanctuaries also exhibit species-typical fission-fusion behaviors, forming isolated subgroups whose composition changes throughout the day, and studies of their behavior indicate that these animals also exhibit species-typical behavior, physiology, and cognitive skills (2, 3). A second point is that wild chimpanzees are not highly physically active. In wild habitats such as the Kibale Forest, Uganda, chimpanzees walk less than 3 km/day (3). It is unlikely that semi-natural sanctuary chimpanzees are substantially less active than their wild counterparts, who spent most of the day resting, grooming and feeding quietly. Third, Pontzer *et al.*, (4) found no difference in total energy expenditure measurements between a wide range of wild, captive, and semi-natural sanctuary primates. Finally, and more speculatively, we note that differences in size-corrected resting cardiac output (ml/min/mass^{0.75}) are approximately 25% higher in humans than chimpanzees (Table 1), matching observed differences in size-corrected metabolic rate between the two species (5). These data raise the hypothesis that resting cardiac output correlates with metabolic rate in order to deliver appropriate levels of blood, hence energy.

Anesthetic Protocols for Chimpanzees and Gorillas. Before immobilization, all animals were fasted overnight with water available *ad libitum*. Prior to each health check, body mass was estimated based on previous assessments. Chimpanzees were anesthetized using either: a combination of Medetomidine (0.05 mg/kg) and ketamine (5 mg/kg) (n=24); Tiletamine-Zolazepam (10 mg/kg) (n=4); a combination of Tiletamine-Zolazepam (2 mg/kg) and Ketamine (5 mg/kg) (n=5); a combination of Medetomidine (0.03 mg/kg) and Tiletamine-Zolazepam (2 mg/kg) (n=9); or, a combination of Medetomidine (0.03 mg/kg) and Ketamine (3 mg/kg) delivered via remote dart injection before being intubated and maintained with isoflurane using a semi-closed rebreathing anesthetic circuit (n=1). Anaesthetic agents were administered intramuscularly by hand injection, or remote dart injection. After completion of all procedures, animals anesthetized with Medetomidine were reversed using Atipamezole (0.25 mg/kg). Once the chimpanzees were anaesthetized and unresponsive to external stimuli they were transported to a field-based assessment center. The gorillas were anesthetized using either a combination of Medetomidine (0.3mg/kg) and Ketamine (1mg/kg) (n=2), or Tiletamine-Zolazepam (1mg/kg) and Medetomidine (0.02mg/kg) (n=3). Irrespective of protocol the anesthetic agents were delivered by remote dart injection and anesthesia was maintained using Isoflurane as outlined above. Similar to chimpanzees, when gorillas had been anesthetized with Medetomidine this was reversed using Atipamezole (0.25 mg/kg) following completion of the health check. All of the protocols adopted in this study have been approved by the Pan African Sanctuary Alliance Advisory Council, adhere to the legal requirements of the Republic of Congo and Zambia, and have been endorsed by the British and Irish Aquarium and Zoo Association. Furthermore, all procedures involving animals received approval from the Cardiff Metropolitan University Ethics Committee.

Tarahumara. We collected echocardiographic and blood pressure data from 42 Tarahumara (who call themselves Rarámuri) men between the ages of 17 and 46 (mean age, 33 ± 8 yrs; body mass, 62 ± 8 kg; height, 164 ± 5 cm). We also collected blood pressure data on an additional 61 Tarahumara men between the ages of 14 and 93 (mean age, 49 ± 20 yrs; body mass, 61 ± 10 kg; height, 163 ± 6 cm). All participants were recruited from the Municipality of Urique in the western part of the state of Chihuahua, Mexico. Details regarding participant recruitment and physical activity patterns of the Tarahumara are outlined below and in *Table S2*. Unfortunately, Tarahumara women were reluctant to participate and so our comparisons are limited to a male population.

Recruitment of Tarahumara. Tarahumara Participants were recruited by word of mouth with the help of a resident (M. Mahaffey) who is well known to many Tarahumara, and a local Tarahumara farmer (S. Cubesare) who speaks Rarámuri (the native language of the Tarahumara). Only men were studied because women were generally reluctant to participate. All research was conducted at the government medical clinic in the town of Cerocahui. Some participants walked to the clinic, but transportation was provided to the majority of participants, who lived as far as 40 km from the clinic. Although the Tarahumara reside in a mountainous region they typically spend the majority of their time in the valley floors and are only exposed to modest altitude (6000-7000 feet) occasionally. Exclusion criteria included the presence of any medical condition that would be reasonably expected to interfere with normal daily activities. All participants provided verbal informed consent, which was administered by translators who spoke Spanish

and Rarámuri. All aspects of recruitment and enrollment were approved by the Partners Human Research Committee (Somerville, MA, USA).

Physical activity in the Tarahumara. The Tarahumara who live in the Sierra Madre Occidental, often called the Sierra Tarahumara, are included as a comparison sample in this study because they are among the last populations in North America that still engages in subsistence farming without leading westernized lifestyles and who participate minimally in the market economy (6). We note that although our sample of older Tarahumara from the Cerocahui area were not connected much to the market economy, the Tarahumara population in some other areas is undergoing rapid change. Although the Tarahumara are famous for long distance running, most Tarahumara men are farmers who report that they rarely if ever take part in long distance running races and never train. In the past, most running occurred in the context of persistence hunting in which small groups of hunters would run down deer, or in ball game races known as *rarajipari* that would occur several times a year (7, 8). Our participants report that persistence hunting is no longer regularly practiced, that *rarajipari* races occur only a few times a year, and that most members of the community run only a portion of *rarajipari* races (9). Instead, most Tarahumara physical activity consists of walking, often up and down steep paths in the highly mountainous Sierra Tarahumara, as well as agricultural tasks such as plowing, harvesting, and carrying things. All the Tarahumara men in our sample reported that they rarely, if ever, smoke tobacco (a contrast to Hadza and Bushmen hunter-gatherers, who smoke frequently).

To assess physical activity levels quantitatively, a subsample (n=21, age = 35±12 years) of the Tarahumara men whose hearts were measured using ultrasound were given wrist-mounted accelerometers (Actigraph GT3X). All men were recruited from the communities of Cerocahui and Huisuchi. We selected a 14-day accelerometer measurement period spanning the 3rd and 4th

weeks of December 2015 as this time period did not include any traditional running ceremonies (Rarajipari) and was not a time of active crop harvest as both factors may have over-estimated routine physical activity patterns. At the start of the study period, accelerometers were placed on each participant's preferred wrist by a study investigator using single-use, non-removable plastic wristband and were then activated using ActiLife 6 Data Analysis Software (version 6.13.3). Accelerometers were subsequently worn continuously for the entire 14-day study period during which participants were instructed not to deviate from normal physical activity patterns. Accelerometer data were collected in 60-second epochs at a rate of 30Hz. At the conclusion of the study period, accelerometers were removed and returned to the study team for analysis. Accelerometer data were downloaded and processed in ActiLife 6 Data Analysis Software (version 6.13.3). Data analysis was limited to the waking hours of 6:00am to 9:00pm. In order to quantify physical activity, cut points were established utilizing per epoch counts per minute (CPM). CPMs were then stratified as follows using the Freedson Adult VM3 (2011) algorithm (10): sedentary (0-99 CPM), light physical activity (100-2689 CPM), moderate physical activity (2690-6166 CPM), vigorous physical activity (6167-9642 CPM) and very vigorous physical activity level (≥ 9643 CPM) (*Table S2*). Vector Magnitude was accounted for in both the sedentary analysis and in establishing physical activity cut points. The calculated time spent in sedentary level activity was extracted from the calculated time spent in light physical activity to prevent double counting of sedentary minutes.

Sedentary controls. 40 minimally active or sedentary (≤ 2 hours exercise/week), normotensive, healthy men between the ages of 21 and 35 (mean age, 27 ± 4 yrs; body mass, 75 ± 9 kg; height, 176 ± 7 cm) were recruited from the Greater Boston (Massachusetts, USA) area using an

institutional research study volunteer program (www.RVSPforhealth.org) at the Massachusetts General Hospital. Advertisements were posted on line that recruited participants to serve as disease free, normally active, controls. Inclusion criteria were: 1) Male gender, 18-35 years of age; 2) Less than 2 hours of routine physical activity per week; 3) Body Mass Index (BMI) of 18 to 28 kg/m²; 4) Lifelong non-smoker or tobacco user; 5) No prior history of clinically significant hematological, renal, endocrine, pulmonary, gastrointestinal, cardiovascular, hepatic, psychiatric, neurologic, or allergic disease. Exclusion criteria were: 1) Regular alcohol consumption exceeding 14 drinks/week; 2) Use of prescription or non-prescription drugs and dietary supplements during the 1-year prior to study enrollment; 3) Prior participation in organized high school or collegiate interscholastic athletics. Participants meeting enrollment criteria provided written informed consent at the time of study enrollment. All aspects of recruitment, enrollment, and data collection were approved by the Partners Human Research Committee (Somerville, MA, USA) and written informed consent was obtained from all participants before enrollment.

Endurance and Resistance Athletes. Athletic participants were recruited from the Harvard Athlete Initiative (HAI), a research collaboration between the Massachusetts General Hospital and the Harvard University Department of Athletics, that was designed as a platform to conduct prospective, longitudinal studies focused on exercise physiology and cardiac remodeling in competitive athletes (11-15). For the purpose of this study, we enrolled 82 male athletes \geq 18 years of age who were free of cardiovascular disease. Written informed consent was obtained from all participants before enrollment. The Partners Human Research Committee and Harvard University institutional review board approved all aspects of this protocol before initiation. All participants were first-year university students who were recruited to be members of varsity

athletic programs. The endurance athlete cohort ($n=42$, 20 ± 2 yrs; body mass, 67 ± 5 kg; height, 179 ± 6 cm) was comprised of long distance runners who were members of the men's track and field program while the resistance athlete cohort ($n=40$, 19 ± 1 yrs; body mass, 102 ± 14 kg; height, 185 ± 9 cm) was comprised of American-style football players who played at a lineman field position (i.e. tackle, guard, center, or defensive end).

Blood pressure data from the National Health and Nutrition Examination Survey

(NHANES): The National Health and Nutrition Examination Survey (NHANES) is an ongoing survey coordinated by the Centers for Disease Control and Prevention. The study uses a stratified, multistage, clustered probability sample design to acquire a representative sample of the US population. A complete description of the NHANES design/methodology can be found at (<http://www.cdc.gov/nchs/nhanes.htm>). As outlined in the NHANES methodology, all procedures were approved by the National Center for Health Statistics review board and respondents gave informed consent prior to participation. The precise procedures and instrumentation for blood pressure assessment in the NHANES study have been described previously (16). Briefly, systolic and diastolic blood pressures were assessed following 5 minutes of seated rest, with an average of three measures reported. We extracted blood pressure data from the 2011-2012 and 2013-2014 NHANES cohorts. All male participants aged between 8 and 80 years of age were initially selected from the database, participants were then excluded from further analyses if they had: eaten, consumed coffee or alcohol, or smoked in the 30 minutes prior to the BP measurement being made, or if an incomplete BP assessment was noted, data were also excluded if the pulse was defined as irregular, or if the participant reported to be routinely using medication. Following exclusions, a sample of 3495 males remained with a mean

age of 29 ± 17 years. These data were then compared with the data collected from the Tarahumara population using LOWESS regression analysis.

Echocardiographic Protocol. All measurements of cardiac structure and function were performed by a single highly trained sonographer (A.L.D.). Left ventricular (LV) structure and function were assessed using 2D, Doppler and speckle tracking ultrasound technology (Vivid Q Ultrasound, GE, Horten, Norway). Images were acquired from the parasternal long and short-axis (basal, mid- and apex levels) views as well as from the apical 4- and 2-chamber views. Assessment of LV structure and geometry included: 1) end-diastolic wall thickness (septal wall thickness + posterior wall thickness/2); 2) LV end-diastolic internal cavity diameter at the basal and mid-level; and 3) relative wall thickness (RWT; posterior wall thickness*2 / diastolic internal cavity dimension) at both the base and mid-level. LV end diastolic and systolic volumes (EDV and ESV respectively) were obtained using the Simpson's methodology (37). Left ventricular sphericity index was calculated using the ratio of LV length to mid cavity diameter with both measured at end diastole from the apical 4-chamber view. The LV outflow tract diameter was measured from the parasternal long axis view during systole. Systolic function was assessed by calculating stroke volume (EDV-ESV), and from the systolic tissue velocities averaged from the septum and lateral walls (S'). Diastolic function was assessed using trans-mitral Doppler filling velocities (Early – E and atrial A) and tissue Doppler velocities during the early (E') and late (A') diastolic period. In a subset of the chimpanzees (n=36), Tarahumara (n=33), sedentary controls (n=10), long distance runners (n=9) and American football linemen (n=9) short axis images were analyzed using speckle-tracking imaging (Echopac, GE Medical, Horten, Norway, version 110.1.8) to assess LV basal and apical rotation and rotation velocities, and to calculate LV twist and LV untwisting velocity as described previously (17). Appropriate

speckle-tracking optimization was not possible in all participants, hence the reduced n for these variables. All cardiac variables were measured and averaged over three cardiac cycles.

As cardiac structure is known to vary as a function of body size (18) all data were scaled to body mass raised to an allometric exponent based on the model of quarter-power scaling (19). To confirm the appropriateness of the exponents used we also calculated sample-specific allometric exponents and compared differences between the Tarahumara and chimpanzees using the two different exponents. Differences between these theoretical and empirical comparisons were not substantial (*Table 1, Table S2*).

Blood Pressure Assessment. During the chimpanzee health checks, systolic and diastolic blood pressures were recorded using manual auscultation. Once animals were positioned supine a manual blood pressure cuff (Welch Allyn, Skaneateles Falls, NY 13153, USA) was placed around the animal's right arm in the brachial position. As animals varied in size a range of cuffs (sizes 8-11) was used to ensure a standardized position. Recordings were made at least three times over the course of the health check procedure and the average of these measures was used for analysis. Blood pressure recordings were not included within the 20-minute period immediately following anesthetic delivery so as to minimize the effects of the stress associated with anesthetic induction and the initial hemodynamic response to the anesthetic agents (20). Obtaining blood pressure measurements in adult male gorillas was logistically challenging using standard techniques, as such we were unable to report reliable data for the five gorillas included in this study.

Blood pressures in the sedentary controls, long distance runners and American football linemen were measured at the point of study enrollment using a clinical grade, carefully calibrated aneroid sphygmomanometer and an appropriately sized arm cuff. Measurements were made while participants were in the supine position after at least 10 minutes of quiet rest. Reported values represent the average of triplicate measurements. Similarly, blood pressure measurements in the Tarahumara were assessed following 10 minutes of supine rest using an automated device (Mobil-O-Graph. I.E.M GmbH, Stolberg, Germany) according to the manufacturer's guidelines. Three consecutive measures were taken with at least two minutes rest between recordings. Previously, the Mobil-O-Graph has been validated against standard auscultation and has met the criteria set by the Association for the Advancement of Medical

Instrumentation (AAMI), the European Society of Hypertension and the British Hypertension Society (21-23).

Acute Pressure versus Volume Challenges. Randomly selected participants from each of the long distance running and American football linemen cohorts described above were chosen to participate in the acute physiologic provocation protocols to measure the cardiac response to acute pressure (n=20) and volume challenges (n=16).

Isometric Handgrip Testing (IHGT). To measure responses to an acute pressure challenge, IHGT was performed using a commercially available hand dynamometer (North Coast Medical, Gilroy, CA) with subjects in the supine position to facilitate simultaneous echocardiography as previously described. The protocol began with the determination of maximum force generation, during which each participant performed three maximum effort handgrip tests with the hand-held dynamometer in the dominant hand. The average value of the three trials was computed. Each participant then performed a prolonged (3 minutes) handgrip test at 40% of their maximal value. Systemic arterial blood pressure was assessed using an automated cuff applied to the non-dominant arm at 1-minute intervals during IHGT and for 3 minutes after completion of IHGT. Echocardiographic assessments of left ventricular stroke volume were performed at baseline and during the final minute of IHGT (i.e., peak afterload).

Normal Saline Infusion Protocol. To measure responses to an acute volume challenge, isotonic saline (0.9% NaCl) was infused via an 18-gauge intravenous cannula inserted into a superficial antecubital arm vein as previously described (24). A standardized volume (1200 mL x body surface area) of saline was infused at a rate of 10 mL per unit of body-surface area (BSA) per minute over 120 minutes. Participants remained in a supine position during the infusion.

Echocardiographic assessments of left ventricular stroke volume were performed at baseline and during the final 10 minutes of saline infusion.

Comparison of left ventricular apical trabeculations in chimpanzees and humans. To examine whether the apex of the chimpanzee's left ventricle is more trabeculated than in humans, we assessed the ratio of non-compacted to compacted myocardial tissue in chimpanzees and Tarahumara subsistence farmers, which are representative of other human groups in this respect (Table 1). Using the previously described Jenni criteria (25) we show that chimpanzees have a statistically significant greater degree of trabeculation ($P < 0.0001$) in the left ventricular apex when compared to the subsistence farmers (*Figure S1*). This statistical significance likely explains the biologically significant lack of apical rotation in the chimpanzees.

Echocardiogram Data from Gorillas (*Gorilla gorillas*). To test if the chimpanzee left ventricle is representative of the primitive phenotype we also assessed ventricular structure and function in an outgroup sample of five adult male gorillas (*Gorilla gorillas*). Unlike chimpanzees these animals were cared for in zoological collections and were not wild-born. These data, showing thick, trabeculated, ventricular walls and low sphericity index (*Table S3*) support our contention that the primitive cardiac phenotype is pressure adapted.

Principal components analysis of chimpanzee and human left ventricles. A comparison of left ventricular (LV) morphology associated with adaptations to either pressure or volume was conducted among the five young adult male populations (chimpanzees, Tarahumara, football players, endurance runners, and sedentary controls from the Boston area). The comparison involved a two-stage analysis. In the first stage of analysis, two derived variables were created to represent LV morphology associated with pressure and volume. This was achieved by selecting structural and functional LV variables determined a priori to be associated with either pressure (n=7; systolic ventricular wall velocity (S'), left ventricular wall thickness (WT), relative wall thickness (RWT), mid RWT, systolic blood pressure (SBP) and diastolic blood pressure (DBP)), or volume (n=9; early trans-mitral filling velocity (MVE), early diastolic ventricular wall velocity (E'), sphericity index (SI), stroke volume (SV), left ventricular length (LVL), end diastolic ventricular volume (EDV), end systolic ventricular volume (ESV), and left ventricular outflow tract (LVOT)), and then including these variables in two separate principal components analyses (PCA). All structural data entered into the PCAs were scaled as per the methods noted above. These PCAs generated new variables, called principal component (PC) scores, which are linear combinations (weighted-sums) of the original raw variables. Components are ordered decreasingly by the amount of variance they explain, so that the first component explains the greatest variance. The scores for first PC are therefore the best one-dimensional representation of the higher-dimensional original variables, with regard to explained variance. Since the PCAs were conducted using the correlation matrix of raw variables, the resulting PC scores are standardized (i.e., z-scores).

In the second stage of analysis, differences in LV adaptations to pressure and volume were inferred among the five populations. The first PC scores from each PCA were extracted and

used as representations of pressure and volume related LV adaptations. To perform inference, two general linear models were conducted with these derived pressure and volume variables regressed separately on four binary indicator population variables representing population identities with respect to a reference group. After the models were fitted, predicted means and 95% confidence intervals for the mean were estimated along with pair-wise contrasts between means for each population (*Tables S4-5*) p -values were adjusted for family-wise error using the sequential Bonferroni method.

Longitudinal Athlete Training Programs. Participants were studied before (initiation of pre-season training) and after a 90-day period of training and competition (completion of the autumn competitive sports season). As previously shown, this model reproducibly captures the impact of exercise dose intensification and sport-specific physiological stimulus on cardiac structure and function (11-15). Demographic and anthropomorphic information including age, self-reported ethnicity, medication and/or over the counter supplement use, personal and family medical history, height, weight, resting heart rate and blood pressure (*Table S6*) were obtained for each participant at both time points. Exercise training volume during an 8-week pre-study time period, and during the 90-day period of training intensification, were assessed for each participant as characterized by total number of training hours per week and the hours per week dedicated to either resistance or endurance activities as previously reported (14). Blood pressure at both study time points was measured using a clinical grade, carefully calibrated aneroid sphygmomanometer as outlined above. Comprehensive echocardiographic assessment of left ventricular structure and function were also performed for each participant at both time points (*Table S7 and S8*). Each participant was subject to testing for performance enhancing drugs as dictated by National Collegiate Athletic Association (NCAA) standards. Subjects were excluded from the final data analysis if they undertook any breaks in training of ≥ 3 days during the study time period due to injury or any other circumstances.

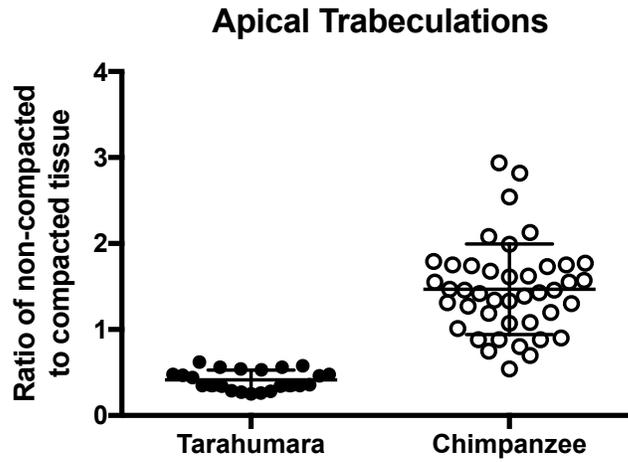


Figure S1: A comparison of left ventricular apical trabeculations (as quantified by the ratio of non-compacted to compacted myocardial tissue) between the Tarahumara (left) and chimpanzees (right). *P*-value is for test of equality of means between groups (chimpanzee vs. subsistence farmers) using a 2-tailed unpaired Welch's *t*-test for unequal variances.

Table S1. Demographic, hemodynamic and left ventricular structure and function in semi-wild sanctuary Chimpanzees, Tarahumara, sedentary humans, long distance runners and American football linemen including non-scaled variables omitted from Table 1 in the manuscript.

	Chimpanzees (n=43)	All Humans (n=164)	Tarahumara (n=42)	Sedentary Humans (n=40)	Long-Distance Runners (n=42)	American Football Linemen (n=40)	P-value [§]
Demographics							
Age (years)	21 ± 5	25 ± 8	33 ± 8*	27 ± 4*	20 ± 2	19 ± 1	0.015
Body Mass (kg)	55 ± 8	76 ± 18	62 ± 8*	75 ± 9*	67 ± 5*	102 ± 14*	<0.001
Height (cm) [^]	127 ± 9	176 ± 10	164 ± 5*	176 ± 7*	179 ± 6*	185 ± 9*	<0.001
Hemodynamics							
SBP (mmHg)	138 ± 21	116 ± 11	113 ± 11*	115 ± 8*	110 ± 8*	127 ± 9	<0.001
DBP (mmHg)	92 ± 17	67 ± 11	69 ± 10*	67 ± 8*	60 ± 8*	74 ± 13*	<0.001
Heart Rate (b/min)	58 ± 10	60 ± 9	59 ± 9	61 ± 8	56 ± 8	66 ± 6*	0.78
Heart Rate/mass ^{-0.25}	158 ± 28	178 ± 31	165 ± 26	179 ± 24*	159 ± 24	209 ± 24*	0.002
CO (l/min)	2.91 ± 0.94	5.33 ± 1.56	3.96 ± 0.74*	4.75 ± 0.83*	5.83 ± 1.11*	6.76 ± 1.68*	<0.001
CO (l/min)/mass ^{0.75}	0.147 ± 0.047	0.208 ± 0.049	0.181 ± 0.036*	0.188 ± 0.035*	0.250 ± 0.047*	0.210 ± 0.045*	<0.001
LV Structure							
LV WT (cm)	0.86 ± 0.12	0.88 ± 0.19	0.71 ± 0.08*	0.90 ± 0.11	0.78 ± 0.07*	1.15 ± 0.09*	1
LV WT (cm)/mass ^{0.25}	0.317 ± 0.041	0.298 ± 0.051	0.252 ± 0.028*	0.307 ± 0.038	0.274 ± 0.027*	0.362 ± 0.025*	0.105
LV RWT	0.366 ± 0.067	0.350 ± 0.065	0.302 ± 0.048*	0.356 ± 0.040	0.320 ± 0.033*	0.427 ± 0.054*	0.78
Mid LV WT (cm)	1.02 ± 0.25	0.89 ± 0.22	0.74 ± 0.09*	0.88 ± 0.25	0.78 ± 0.07*	1.18 ± 0.09*	0.073
Mid LV WT (cm)/mass ^{0.25}	0.376 ± 0.089	0.307 ± 0.053	0.264 ± 0.033*	0.323 ± 0.039*	0.274 ± 0.026*	0.372 ± 0.024	<0.001
Mid LV RWT	0.427 ± 0.113	0.366 ± 0.074	0.317 ± 0.058*	0.378 ± 0.050	0.322 ± 0.035*	0.452 ± 0.058	0.02
EDV (ml)	99 ± 17	147 ± 39	106 ± 17	125 ± 17*	185 ± 20*	171 ± 28*	<0.001
EDV (ml)/mass ^{1.0}	1.8 ± 0.3	2.0 ± 0.5	1.7 ± 0.3	1.7 ± 0.3	2.8 ± 0.3*	1.7 ± 0.2	0.201
ESV (ml)	49 ± 15	59 ± 20	38 ± 9*	46 ± 9	80 ± 11*	69 ± 14*	0.027
ESV (ml)/mass ^{1.0}	0.91 ± 0.24	0.79 ± 0.29	0.63 ± 0.14*	0.63 ± 0.15*	1.20 ± 0.18*	0.68 ± 0.12*	0.095
LV Length (cm)	7.60 ± 0.56	8.80 ± 0.69	8.72 ± 0.45*	8.21 ± 0.60*	9.45 ± 0.58*	8.81 ± 0.49*	<0.001
LV length (cm)/mass ^{0.25}	2.80 ± 0.20	3.00 ± 0.29	3.11 ± 0.15*	2.79 ± 0.20	3.31 ± 0.20*	2.78 ± 0.16	<0.001
LV OT (cm)	1.96 ± 0.19	2.14 ± 0.16	2.12 ± 0.14*	2.05 ± 0.14	2.19 ± 0.12*	2.18 ± 0.19*	<0.001
LV OT (cm)/mass ^{0.25}	0.720 ± 0.063	0.728 ± 0.055	0.755 ± 0.043	0.698 ± 0.046	0.768 ± 0.039*	0.688 ± 0.043	1
Sphericity index	1.68 ± 0.15	1.85 ± 0.17	1.90 ± 0.16*	1.84 ± 0.11*	1.93 ± 0.15*	1.72 ± 0.17	<0.001
LV Systolic Function							
SV (ml)	50 ± 11	89 ± 22	68 ± 12*	78 ± 13*	105 ± 44*	102 ± 22*	<0.001
SV (ml)/mass ^{1.0}	0.926 ± 0.219	1.192 ± 0.304	1.109 ± 0.194*	1.057 ± 0.194	1.578 ± 0.205*	1.006 ± 0.197	<0.001
S' (cm/s)	0.0624 ± 0.0134	0.1114 ± 0.0157	0.1015 ± 0.0157*	0.1143 ± 0.0093*	0.1067 ± 0.0146*	0.1235 ± 0.0133*	<0.001
S'/LVL	0.0082 ± 0.0018	0.0127 ± 0.0021	0.0117 ± 0.0020*	0.0140 ± 0.0015*	0.0113 ± 0.0016*	0.0140 ± 0.0014*	<0.001
Peak LV Twist (°) [†]	6.4 ± 3.3	17.4 ± 6.2	18.0 ± 6.3*	18.4 ± 5.7*	14.4 ± 5.2*	16.5 ± 7.4*	<0.001
LV Diastolic Function							
E (cm/s)	0.77 ± 0.17	0.83 ± 0.14	0.85 ± 0.20	0.84 ± 0.11	0.88 ± 0.13	0.77 ± 0.09	0.241
A (cm/s)	0.39 ± 0.15	0.51 ± 0.15	0.51 ± 0.15*	0.48 ± 0.11*	0.39 ± 0.09	0.65 ± 0.14*	<0.001
E/A	2.21 ± 0.73	1.82 ± 0.69	1.79 ± 0.65	1.82 ± 0.44	2.38 ± 0.73	1.26 ± 0.38*	0.029
E' (cm/s)	0.101 ± 0.024	0.136 ± 0.032	0.152 ± 0.030*	0.115 ± 0.014	0.168 ± 0.013*	0.107 ± 0.018	<0.001
E'/LVL	0.013 ± 0.003	0.015 ± 0.003	0.017 ± 0.003*	0.014 ± 0.002	0.018 ± 0.002*	0.012 ± 0.002	0.009
A' (cm/s)	0.062 ± 0.018	0.073 ± 0.025	0.102 ± 0.020*	0.051 ± 0.012	0.065 ± 0.011	0.074 ± 0.020	0.017
A'/LVL	0.008 ± 0.003	0.008 ± 0.003	0.012 ± 0.002*	0.006 ± 0.002*	0.007 ± 0.001	0.008 ± 0.002	1
LV UTV (°/s) [‡]	-53 ± 23	-109 ± 38	-109 ± 39*	-121 ± 44*	-109 ± 27*	-96 ± 36*	<0.001

SBP=systolic blood pressure; DBP=diastolic blood pressure; CO=cardiac output; LV=left ventricular; WT=wall thickness; RWT=relative wall thickness; EDV=end diastolic volume; ESV=end systolic volume; OT=outflow tract; SV=stroke volume; S'=systolic ventricular wall velocity; LVL=left ventricular length; E=early diastolic trans-mitral filling velocity; A=late diastolic trans-mitral filling velocity; E:A=the ratio of early to late diastolic trans-mitral filling velocities; E'=early diastolic ventricular wall velocity; A'=late diastolic ventricular wall velocity; UTV=untwisting velocity. [§] Comparison of chimpanzees to all humans using 2-tailed unpaired Welch's *t*-tests for unequal variances. *P*-values are adjusted for family-wise error (37 tests) using the sequential Bonferroni method. * *P* < 0.05 for comparison of chimpanzees to individual human groups using 2-tailed unpaired Welch's *t*-tests for unequal variances. *P*-values are adjusted for family-wise error (148 tests) using the sequential Bonferroni method. [†] For two angular variables (Peak LV Twist and LV untwisting velocity) a circular-linear model with a von Mises distribution for the response was used to compare means across groups. [^] Calculated via the summation of lower limb and crown to rump measurements. N=207 for all analyses except: EDV (198); EDV/mass^{1.0} (198); Simpson's ESV (76); ESV (198); ESV/mass^{1.0} (198); SV (198); SV/mass^{1.0} (198); CO (198); CO/mass^{0.75} (198); Sphericity index (204); MVE (206); MVA (206); MV E/A (206); Mean E' (206); E'/LVL (206); Mean A' (206); A'/LVL (206); Mean S' (205); S'/LVL (205); Twist Peak (97); Untwisting velocity (97).

Table S2. Physical activity of the Tarahumara based on Accelerometer data collected over a 14-day assessment period.

Physical Activity Variable	Value \pm SD (% of total)
Total steps per day (n)	18,800 \pm 4500
Sedentary activity, min/day	83 \pm 40 (9)
Light physical activity, min/day	480 \pm 70 (53)
Moderate physical activity, min/day	340 \pm 90 (38)
Vigorous physical activity, min/day	0 (0)
Very Vigorous physical activity, min/day	0 (0)

Table S3. Echocardiographic variables in Tarahumara versus Chimpanzees – sensitivity analysis using sample-specific allometric modelling

Variable	Mass exponent (95% CI)	Tarahumara	Chimpanzees	Difference (95% CI)
Wall thickness (cm)	0.27 (0.16 to 0.39)	0.73	0.92	-21 (-25 to -16) %
Simpson's EDV (ml)	0.56 (0.41 to 0.72)	114.0	114.6	-0.5 (-8 to 8) %
RWT ^a	0.16 (0.01 to 0.31)	0.306	0.381	-20 (-25 to -14) %
LV length (cm)	0.15 (0.08 to 0.22)	8.9	7.9	13 (9 to 16) %
Simpson's ESV (ml)	0.59 (0.37 to 0.82)	41.5	57.8	-28 (-36 to -19) %
Heart Rate ^b	N/A	59	58	2 (-4 to 8) %
MWT (cm)	0.27 (0.08 to 0.46)	0.77	1.10	-30 (-36 to -24) %
Simpson's SV (ml)	0.56 (0.36 to 0.76)	73	57	28 (14 to 43) %
Simpson's CO (l/min)	0.70 (0.46 to 0.94)	4.31	3.47	24 (8 to 43) %
LVOT (cm)	0.30 (0.22 to 0.37)	2.19	2.10	4 (1 to 7) %

Values are means with differences expressed as a percentage (ratio).

^aTheoretically, RWT is independent of body size, but in our data, there is a small positive exponent

^bHeart rate was independent of body size in our data, so the comparison of the raw, unscaled variable is shown here in contrast to the minus quarter-power body mass scaling in Table 1.

Table S4. Demographic, hemodynamic and left ventricular structure and function data from five adult male gorillas.

Gorilla (n=5)	
<i>Demographics</i>	
Age (years)	19 ± 9
Body Mass (kg)	170 ± 10
<i>Hemodynamics</i>	
Heart Rate (b/min)	53 ± 9
Heart Rate/mass^{-0.25}	191 ± 34
Cardiac Output (l/min)	5.09 ± 0.37
Cardiac Output (l/min)/mass^{0.75}	0.110 ± 0.007
<i>Left Ventricular Structure</i>	
LV Wall Thickness (cm)	1.28 ± 0.22
LV Wall Thickness (cm)/mass^{0.25}	0.35 ± 0.06
LV Relative Wall Thickness	0.46 ± 0.11
Mid LV Wall Thickness (cm)	1.30 ± 0.23
Mid LV Wall Thickness (cm)/mass^{0.25}	0.360 ± 0.063
Mid LV Relative Wall Thickness	0.52 ± 0.13
End Diastolic Volume (ml)	170 ± 15
End Diastolic Volume (ml)/mass^{1.0}	1.0 ± 0.05
End Systolic Volume	72 ± 14
End Systolic Volume (ml)/mass^{1.0}	0.43 ± 0.06
LV Length (cm)	9.54 ± 0.93
LV length (cm)/mass^{0.25}	2.6 ± 0.2
LV Outflow Tract (cm)	2.71 ± 0.2
LV Outflow Tract (cm)/mass^{0.25}	0.75 ± 0.2
Sphericity index	1.72 ± 0.19
Jeni trabecular criteria score	1.52 ± 0.20
<i>Left Ventricular Systolic Function</i>	
Stroke Volume (ml)	98 ± 13
Stroke Volume (ml)/mass^{1.0}	0.586 ± 0.086
S' (cm/s)	0.0711 ± 0.01
S'/LVL	0.0072 ± 0.001
<i>Left Ventricular Diastolic Function</i>	
E (cm/s)	0.82 ± 0.14
A (cm/s)	0.27 ± 0.04
E/A	3.05 ± 0.49
E' (cm/s)	0.122 ± 0.02
E'/LVL	0.017 ± 0.006
A' (cm/s)	0.064 ± 0.02
A'/LVL	0.007 ± 0.002

Table S5: Population pairwise mean pressure differences.

Contrast	Δ^*	SE	df	LCL	UCL	<i>t</i>-value	<i>P</i>-value**
CHI - TAR	3.77	0.28	190	2.97	4.58	13.29	<0.001
CHI - LDR	3.63	0.28	190	2.83	4.42	12.99	<0.001
CHI - SAM	2.23	0.28	190	1.43	3.03	7.92	<0.001
CHI - AFL	0.11	0.28	190	-0.69	0.91	0.39	1
TAR - LDR	-0.15	0.27	190	-0.91	0.62	-0.54	1
TAR - SAM	-1.54	0.27	190	-2.31	-0.77	-5.65	<0.001
TAR - AFL	-3.66	0.27	190	-4.44	-2.89	-13.45	<0.001
LDR - SAM	-1.39	0.27	190	-2.15	-0.63	-5.21	<0.001
LDR - AFL	-3.52	0.27	190	-4.27	-2.76	-13.16	<0.001
SAM - AFL	-2.12	0.27	190	-2.89	-1.35	-7.85	<0.001

* Estimate of mean difference in pressure between populations (units are standard deviations).

** *P*-values adjusted for multiple comparisons using the sequential Bonferroni method.

Table S6: Population pairwise mean volume differences.

Contrast	Δ^*	SE	df	LCL	UCL	<i>t</i>-value	<i>P</i>-value**
CHI - TAR	-1.36	0.25	190	-2.06	-0.66	-5.54	<0.001
CHI - LDR	-4.45	0.24	190	-5.14	-3.77	-18.41	<0.001
CHI - SAM	-0.18	0.24	190	-0.88	0.51	-0.74	0.92
CHI - AFL	0.15	0.24	190	-0.55	0.84	0.60	0.92
TAR - LDR	-3.09	0.23	190	-3.75	-2.43	-13.25	<0.001
TAR - SAM	1.18	0.24	190	0.51	1.85	5.01	<0.001
TAR - AFL	1.51	0.24	190	0.84	2.18	6.40	<0.001
LDR - SAM	4.27	0.23	190	3.61	4.93	18.44	<0.001
LDR - AFL	4.60	0.23	190	3.94	5.26	19.86	<0.001
SAM - AFL	0.33	0.23	190	-0.34	0.99	1.40	0.49

* Estimate of mean difference in volume between populations (units are standard deviations).

** *P*-values adjusted for multiple comparisons using the sequential Bonferroni method.

Table S7: Demographic and hemodynamic data for the longitudinal training cohorts of long-distance runners and American-football linemen

<i>Demographics</i>	Long-Distance Runners (n= 42)			Football Linemen (n=40)		
	Pre-Training	Post-Training	<i>P</i> -value	Pre-Training	Post-Training	<i>P</i> -value
Age (y)	19.6 ± 2.1	-----	-----	19.0 ± 0.9	-----	
Mass (kg)	67.2 ± 4.6	66.7 ± 4.7	0.005	99.9 ± 12.6	101.9 ± 14.0*	0.003
Height (cm)	179.0 ± 5.8	-----	-----	185.4 ± 8.7	-----	-----
<i>Hemodynamics</i>						
SBP (mmHg)	111 ± 8	110 ± 8	1	123 ± 7	127 ± 9*	0.003
DBP (mmHg)	63 ± 8	60 ± 8	<0.001	72 ± 10	74 ± 13*	0.137
Heart Rate (beats/min)	59 ± 8	56 ± 8	<0.001	66 ± 6	66 ± 6*	1
<i>Training Data</i>						
<i>Preseason</i>						
Total Training (hours/week)	5.4 ± 2	-----	-----	6.1 ± 1.1	-----	-----
Endurance (hours/week)	4.9 ± 4	-----	-----	5.6 ± 1.2	-----	-----
Strength (hours/week)	0.5 ± 1.1	-----	-----	0.7 ± 1.0	-----	-----
<i>Study Period</i>						
Total Training (hours/week)	-----	13.2 ± 0.9	-----	-----	14.1 ± 1.3	-----
Endurance (hours/week)	-----	12.6 ± 0.4	-----	-----	13.6 ± 0.3	-----
Strength (hours/week)	-----	0.5 ± 0.2	-----	-----	0.6 ± 0.1	-----

P-values reflect within group (i.e. pre vs. post) significance of difference as derived using 2-tailed paired *t*-tests and are adjusted for table-wide (Tables S7-9) family-wise error (42 tests) using the sequential Bonferroni method.

* = $P < 0.05$ compared to post season long-distance runners as derived using 2-tailed unpaired *t*-tests, adjusted for table-wide (Tables S7-9) family-wise error (21 tests) using the sequential Bonferroni method.

Table S8: Left Ventricular Structure in long-distance runners and American-football linemen pre-post completion of 90 days training intensification

	Long-Distance Runners (n= 42)			Football Linemen (n=40)		
	Pre-Training	Post-Training	<i>P</i> -value	Pre-Training	Post-Training	<i>P</i> -value
Basal Wall Thickness (mm)	7.9 ± 0.7	7.8 ± 0.7	1	10.5 ± 0.7	11.5 ± 0.9*	<0.001
Mid LV Wall Thickness (mm)	7.8 ± 0.7	7.8 ± 0.7	1	11.2 ± 0.7	11.8 ± 0.9*	<0.001
LV Length (mm)	90.8 ± 5.6	94.5 ± 5.8	<0.001	88.9 ± 4.6	88.1 ± 4.9*	1
EDV (mL)	173 ± 21	185 ± 20	<0.001	173 ± 31	170 ± 28	1
ESV (mL)	70 ± 11	80 ± 11	<0.001	70 ± 12	69 ± 14*	1
LVOT (mm)	21.9 ± 1.2	21.9 ± 1.2	1	21.8 ± 1.5	21.8 ± 1.9	1
Relative Wall Thickness	0.33 ± 0.03	0.32 ± 0.03	0.178	0.40 ± 0.04	0.43 ± 0.05*	<0.001
LV Sphericity Index	1.90 ± 0.16	1.93 ± 0.15	<0.001	1.73 ± 0.16	1.72 ± 0.17*	0.831

P-values reflect within group (i.e. pre vs. post) significance of difference as derived using 2-tailed paired *t*-tests and are adjusted for table-wide (Tables S7-9) family-wise error (42 tests) using the sequential Bonferroni method.

* = $P < 0.05$ compared to post season long-distance runners as derived using 2-tailed un-paired *t*-tests, adjusted for table-wide (Tables S7-9) family-wise error (21 tests) using the sequential Bonferroni method.

Table S9: Left Ventricular Function in long-distance runners and American-football linemen pre-post completion of 90 days training intensification

	Long-Distance Runners (n= 42)			Football Linemen (n=40)		
	Pre-Training	Post-Training	P-value	Pre-Training	Post-Training	P-value
Stroke Volume (mL)	103 ± 16	105 ± 14	0.374	104 ± 26	102 ± 22	1
Cardiac Output (L/min)	6.0 ± 1.2	5.8 ± 1.1	0.238	6.9 ± 1.9	6.8 ± 1.7*	1
Ejection Fraction (%)	59 ± 5	57 ± 4	<0.001	59 ± 7	60 ± 6	1
Average S' (cm/s)	10.5 ± 1.4	10.7 ± 1.5	0.473	12.6 ± 1.4	12.4 ± 1.3*	1
E-wave velocity (cm/s)	83 ± 11	88 ± 13	<0.001	79 ± 9	77 ± 9*	0.070
A-wave velocity (cm/s)	37 ± 9	39 ± 9	0.473	53 ± 12	65 ± 14*	<0.001
E/A	2.42 ± 0.80	2.38 ± 0.73	1	1.59 ± 0.46	1.26 ± 0.38*	<0.001
Average E' (cm/s)	15.0 ± 1.2	16.8 ± 1.3	<0.001	11.4 ± 1.7	10.7 ± 1.8*	<0.001
Average A' (cm/s)	6.2 ± 1.2	6.6 ± 1.2	<0.001	6.2 ± 1.6	7.3 ± 2.0	<0.001

P-values reflect within group (i.e. pre vs. post) significance of difference as derived using 2-tailed paired *t*-tests and are adjusted for table-wide (Tables S7-9) family-wise error (42 tests) using the sequential Bonferroni method.

* = $P < 0.05$ compared to post season long-distance runners as derived using 2-tailed un-paired *t*-tests, adjusted for table-wide (Tables S7-9) family-wise error (21 tests) using the sequential Bonferroni method.

Table S10. Isometric Handgrip Testing (“Pressure Challenge”)

	Long-Distance Runners (n= 10)			Football Linemen (n=10)		
	Pre-IHG	Peak-IHG	Δ (%)	Pre-IHG	Peak-IHG	Δ (%)
Mass (kg)	67 \pm 6	----	----	103 \pm 5	----	----
Height (cm)	178 \pm 7	----	----	188 \pm 8	----	----
3-minute force (psi)	29 \pm 13	----	----	48 \pm 10	----	----
Heart Rate (beats/min)	54 \pm 11	66 \pm 8 [^]	24 \pm 14	67 \pm 3	78 \pm 4	17 \pm 9*
SBP (mmHg)	123 \pm 10	158 \pm 21 [^]	51 \pm 20	130 \pm 10	159 \pm 14 [^]	29 \pm 18
DBP (mmHg)	63 \pm 4	95 \pm 11 [^]	27 \pm 13	61 \pm 4	78 \pm 9 [^]	29 \pm 18
Stroke Volume (mL)	109 \pm 15	86 \pm 17	-21 \pm 11	92 \pm 8	88 \pm 8	-4 \pm 4*
Average E' (cm/s)	16.4 \pm 1.0	11.4 \pm 1.7	-30 \pm 12	9.9 \pm 0.8	9.5 \pm 0.7	-4 \pm 4*
Cardiac Output (L)	5.9 \pm 1.1	5.7 \pm 1.4	-4 \pm 13	6.2 \pm 0.7	6.9 \pm 0.8	13 \pm 11

[^] $P < 0.05$ for comparison with resting (i.e., pre-handgrip test) value using 2-tailed paired exact permutation tests. P -values are adjusted for table-wide (Tables S10-11) family-wise error (24 tests) using the sequential Bonferroni method.

* $P < 0.05$ compared to Δ observed among long-distance runners using 2-tailed unpaired exact permutation tests. P -values are adjusted for table-wide (Tables S10-11) family-wise error (12 tests) using the sequential Bonferroni method. 3-minute force represents 40% of peak 1sec maximal handgrip strength.

Table S11. Normal Saline Infusion (“Volume Challenge”)

	Long-Distance Runners (n= 8)			Football Linemen (n=8)		
	Pre-Saline	Peak-Saline	Δ (%)	Pre-Saline	Peak-Saline	Δ (%)
Mass (kg)	68 \pm 3	----	----	104 \pm 4	----	----
Height (cm)	173 \pm 6	----	----	188 \pm 7	----	----
Saline Volume (L)	2.16 \pm 0.04	----	----	2.79 \pm 0.04	----	----
Heart Rate (beats/min)	51 \pm 4	53 \pm 4	4 \pm 8	68 \pm 3	68 \pm 3	1 \pm 7
SBP (mmHg)	110 \pm 7	112 \pm 6	2 \pm 6	132 \pm 5	133 \pm 8	1 \pm 4
DBP (mmHg)	57 \pm 5	59 \pm 7	5 \pm 11	76 \pm 3	72 \pm 3	-3 \pm 5
Stroke Volume (mL)	105 \pm 9	121 \pm 8	16 \pm 4	86 \pm 6	90 \pm 8	6 \pm 5*
Average E' (cm/s)	15.8 \pm 1.0	17.7 \pm 0.8	13 \pm 4	9.8 \pm 0.7	10.0 \pm 0.8	3 \pm 3*
Cardiac Output (L)	5.3 \pm 0.4	6.4 \pm 0.7	21 \pm 10	5.2 \pm 0.3	5.6 \pm 0.3	6 \pm 6*

[^] $P < 0.05$ for comparison with resting (i.e. pre-saline test) value using 2-tailed paired exact permutation tests. P -values are adjusted for table-wide (Tables S10-11) family-wise error (24 tests) using the sequential Bonferroni method.

* $P < 0.05$ compared to Δ observed among long-distance runners using 2-tailed unpaired exact permutation tests. P -values are adjusted for table-wide (Tables S10-11) family-wise error (12 tests) using the sequential Bonferroni method. Saline infusions determined by 1200mL x BSA.

Supporting Information References

1. Farmer KH, *et al.* (2009) Pan African Sanctuary Alliance Operations Manual. (Pan African Sanctuary Alliance).
2. Wobber V & Hare B (2011) Psychological health of orphan bonobos and chimpanzees in African sanctuaries. *PLoS One* 6(6):e17147.
3. Pontzer H & Wrangham RW (2004) Climbing and the daily energy cost of locomotion in wild chimpanzees: implications for hominoid locomotor evolution. *J Hum Evol* 46(3):317-335.
4. Pontzer H, *et al.* (2014) Primate energy expenditure and life history. *Proc Natl Acad Sci U S A* 111(4):1433-1437.
5. Pontzer H, *et al.* (2016) Metabolic acceleration and the evolution of human brain size and life history. *Nature* 533(7603):390-392.
6. Carrasco D (2001) *The Oxford encyclopedia of Mesoamerican cultures : the civilizations of Mexico and Central America* (Oxford University Press, Oxford).
7. Bennett WC & Zingg RM (1976) *The Tarahumara, an Indian tribe of northern Mexico* (Rio Grande Press, Glorietta, N.M.).
8. Lumholz C (1902) *Unknown Mexico: A Record of Five Years' Exploration Among the Tribes of the Western Sierra Madre; In the Tierra Caliente of Tepic and Jalisco; and Among the Tarascos of Michoacan* (Macmillan and Co., London).
9. Irigoyen-Tascon F & Palma-Batista JM (1994) *Rarajipari: The Kick Ball Race of the Tarahumara Indians*. (Centra Librero de la Prensa, Sucursal Universidad, Chihuahua).
10. Sasaki JE, John D, & Freedson PS (2011) Validation and comparison of ActiGraph activity monitors. *J Sci Med Sport* 14(5):411-416.
11. Baggish AL, *et al.* (2008) The impact of endurance exercise training on left ventricular systolic mechanics. *Am J Physiol Heart Circ Physiol* 295(3):H1109-H1116.
12. Baggish AL, *et al.* (2008) Training-specific changes in cardiac structure and function: a prospective and longitudinal assessment of competitive athletes. *J Appl Physiol (1985)* 104(4):1121-1128.
13. Weiner RB, *et al.* (2013) Blood pressure and left ventricular hypertrophy during American-style football participation. *Circulation* 128(5):524-531.
14. Weiner RB, *et al.* (2010) The impact of endurance exercise training on left ventricular torsion. *JACC Cardiovasc Imaging* 3(10):1001-1009.
15. Wasfy MM, *et al.* (2015) Endurance Exercise-Induced Cardiac Remodeling: Not All Sports Are Created Equal. *J Am Soc Echocardiogr* 28(12):1434-1440.
16. Zipf G, *et al.* (2013) National health and nutrition examination survey: plan and operations, 1999-2010. *Vital Health Stat I (56)*:1-37.
17. Sengupta PP, Tajik AJ, Chandrasekaran K, & Khandheria BK (2008) Twist mechanics of the left ventricle: principles and application. *JACC Cardiovasc Imaging* 1(3):366-376.
18. Williams TM, Bengtson P, Steller DL, Croll DA, & Davis RW (2015) The Healthy Heart: Lessons from Nature's Elite Athletes. *Physiology (Bethesda)* 30(5):349-357.
19. Batterham AM, George KP, Whyte G, Sharma S, & McKenna W (1999) Scaling cardiac structural data by body dimensions: a review of theory, practice, and problems. *Int J Sports Med* 20(8):495-502.

20. Atencia R, *et al.* (2017) Heart Rate and Indirect Blood Pressure Responses to Four Different Field Anesthetic Protocols in Wild-Born Captive Chimpanzees (*Pan Troglodytes*). *J Zoo Wildl Med* 48(3):636-644.
21. Wei W, Tolle M, Zidek W, & van der Giet M (2010) Validation of the mobil-O-Graph: 24 h-blood pressure measurement device. *Blood Press Monit* 15(4):225-228.
22. Franssen PM & Imholz BP (2010) Evaluation of the Mobil-O-Graph new generation ABPM device using the ESH criteria. *Blood Press Monit* 15(4):229-231.
23. Jones CR, Taylor K, Chowienczyk P, Poston L, & Shennan AH (2000) A validation of the Mobil O Graph (version 12) ambulatory blood pressure monitor. *Blood Press Monit* 5(4):233-238.
24. Weiner RB, *et al.* (2010) Preload dependency of left ventricular torsion: the impact of normal saline infusion. *Circ Cardiovasc Imaging* 3(6):672-678.
25. Jenni R, Oechslin E, Schneider J, Attenhofer Jost C, & Kaufmann PA (2001) Echocardiographic and pathoanatomical characteristics of isolated left ventricular non-compaction: a step towards classification as a distinct cardiomyopathy. *Heart* 86(6):666-671.