1	PHYSIOLOGICAL DIFFERENCES BETWEEN ADVANCED CROSSFIT ATHLETES,
2	RECREATIONAL CROSSFIT PARTICIPANTS, AND PHYSICALLY-ACTIVE
3	ADULTS
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11	Running Title: Physiological characteristics among CrossFit athletes
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ABSTRACT

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3	This investigation examined anthropometric, hormonal, and physiological differences between
4	advanced (ADV; n = 8, 27.8 \pm 4.2 years, 170 \pm 11 cm, 79.8 \pm 13.3 kg) and recreational (REC; n
5	= 8, 33.5 \pm 8.1 years, 172 \pm 14 cm, 76.3 \pm 19.5 kg) CrossFit (CF) trained participants in
6	comparison to physically-active controls (CON; n = 7, 27.5 \pm 6.7 years, 171 \pm 14 cm, 74.5 \pm
7	14.3 kg). ADV and REC were distinguished by their past competitive success. REC and CON
8	were resistance-trained (>2 years) and exercised on 3-5 days wk ⁻¹ for the past year, but CON
9	utilized traditional resistance and cardiovascular exercise. All participants provided a fasted,
10	resting blood sample and completed assessments of resting metabolic rate, body composition,
11	muscle morphology, isometric mid-thigh pull strength, peak aerobic capacity, and a 3-minute
12	maximal cycle ergometer sprint across two separate occasions (separated by 3-7 days). Blood
13	samples were analyzed for testosterone, cortisol, and insulin-like growth factor-1. One-way
14	analysis of variance revealed ADV to possess lower body fat percentage (6.7-8.3%, $p = 0.007$),
15	greater bone and non-bone lean mass (12.5-26.8%, $p \le 0.028$), muscle morphology
16	characteristics (14.2-59.9%, $p < 0.05$), isometric strength characteristics (15.4-41.8%, $p < 0.05$),
17	peak aerobic capacity (18.8-19.1%, $p = 0.002$), and anaerobic performance (15.4-51.1%, $p \le 0.002$)
18	0.023) compared to both REC and CON. No differences were seen between REC and CON, or
19	between all groups for resting metabolic rate or hormone concentrations. These data suggest
20	ADV possess several physiological advantages over REC and CON, whereas similar
21	physiological characteristics were present in individuals who have been regularly participating in
22	either CF or resistance and cardiovascular training for the past year.

- 1 KEYWORDS: HIFT, hormones, resting metabolic rate, peak aerobic capacity, critical power,
- 2 strength

3

INTRODUCTION

4

5 CrossFit® (CF) is a form of high-intensity functional training that combines resistance 6 exercises, gymnastics, and traditional aerobic modalities (e.g., cycling, rowing, running) into 7 single workouts that vary by day to elicit general physical preparedness (1, 2). This training form 8 is enjoyed recreationally by participants of varying levels of fitness, training experience, age, and 9 lifestyles (3) and also exists as its own sport. The primary CF competition is the Reebok CrossFit 10 GamesTM (the Games) which awards individual winners the title of "Fittest on EarthTM". Historically, this competition has consisted of several stages designed to narrow the initial 11 12 participant pool (>400,000 athletes) down to the top athletes within each category (i.e., adult 13 men, adult women, teenagers, Masters, teams). Although the existence, format and relevance of 14 each stage have undergone changes throughout the competition's existence (4, 5), the presence 15 of an initial online qualifying round (e.g., the CrossFit OpenTM) has remained. This round 16 typically involves multiple workout challenges that are completed over the course of several 17 weeks. Competitors who complete all workouts and rank high enough will progress to the next 18 stage of the competition (i.e., historically regionals, currently the Games or Sanctioned 19 competitions that lead to the Games). Regardless of which stage, it is expected that each workout 20 will consist of a set of challenges that will require some combination of strength, power, 21 endurance, and/or sport-specific skill (1). However, little is known about which physiological 22 factors are most influential of CF competition performance or distinguish competitive status. 23 Body mass (6), strength and anaerobic power (6-10), aerobic capacity (9), sport-specific 24 skill (8, 10), and experience (9) have all been associated with either workout performance or 25 competitive ranking. Collectively, these data imply that athletes must train to be proficient in

26 each to perform well in competition. However, several limitations exist among these studies that 27 prevent making such a conclusion. For instance, Serafini et al. (2018) reported that higher 28 ranking competitors of the 2016 Open were stronger, more powerful, and more proficient at 29 short-duration, sprint-type CF workouts. Among regional competitors, final ranking was 30 positively related to 400-m sprint time and time-to-completion in longer, benchmark workouts 31 (i.e., Filthy-50) (r = 0.69 - 0.77), and negatively related to maximal weight lifted in the Olympic 32 lifts (r = -0.39 to -0.42) (10). Although these studies involved participants who have successful 33 competitive records, the measures used to distinguish rank were all self-reported. As such, the 34 authenticity and actual data of measurement (self-reported data were obtained from an online 35 resource) cannot be verified. In contrast, others have measured a variety of physical parameters 36 and related them to CF-style workouts performed in a controlled, laboratory setting (6, 7, 9). 37 While these studies have also included successful CF athletes, laboratory workouts do not 38 adequately emulate the competitive setting and may influence the physiological response to CF 39 training (11-14). Thus, questions remain about the distinguishing characteristics of successful CF 40 athletes.

41 In more traditional sports (e.g., football, baseball, basketball, etc.), identifying the key 42 physiological and athletic characteristics that distinguish performance is common (15-18). The 43 practice enables strength and conditioning professionals to develop sport-specific training 44 programs that are more effective in translating adaptations to in-game performance. However, 45 CF is unique in that typical training session workouts mirror those that appear in competition. 46 Moreover and consistent with its primary purpose (1, 2), chronic participation in CF training has 47 been documented to improve a variety of fitness parameters (19). Though it might be assumed 48 that CF training represents an ideal training strategy for developing the physiological

characteristics that distinguish performance in the sport, such a conclusion would be prematurebased on the available data.

51 Evidence of CF training being more advantageous towards developing a variety of fitness 52 outcomes in comparison to alternative training strategies (e.g., resistance training, high-intensity 53 interval training) is equivocal (19-25). This is likely because most comparative training studies 54 have utilized untrained or novice (to CF) participants, which is problematic because they do not 55 require a very specific or intense training stimulus to elicit adaptations compared to experienced 56 trainees (26). It is possible that either a longer training duration or more advanced participants 57 are necessary to observe the advantages or disadvantages of the CF strategy. Unfortunately, elite 58 competitors rarely share their training strategies and anecdotal evidence suggests that they 59 incorporate more than what commonly occurs during a typical CF training session. To the best of 60 our knowledge, only one well-controlled study exists where a variety of physiological 61 parameters were examined in trained participants (27). In that cross-sectional investigation, men 62 with at least on year of CF training experience outperformed their resistance-trained (> 1 year) 63 counterparts in a multi-stage shuttle run test and possessed a higher aerobic capacity; all other 64 measures were statistically similar. While this study provides evidence in favor of CF training, 65 there was no aerobic training requirement for the resistance-trained group, and the actual 66 experience of the CF group was unclear beyond their having participated in the strategy for at 67 least one year. It is possible that multiple physiological differences exist when experience is considered. Therefore, the purpose of this study was to examine anthropometric, hormonal, and 68 69 physiological differences between advanced CF athletes, recreational CF practitioners, and 70 physically-active adults who regularly participate in both resistance and cardiovascular training. 71 Since adaptations are specific to the training modality and effort (26), we hypothesized that body

72	composition, muscle morphology, aerobic and anaerobic performance, and strength would be
73	different between groups. Specifically, the advanced CF athletes would outperform the other
74	groups whereas recreational CF practitioners and physically-active adults would be similar.
75	However, because resting hormonal concentrations do not typically change through training (14),
76	it was hypothesized that these would be similar between groups.
77	
78	MATERIALS AND METHODS
79	
80	Experimental Design
81	For this cross-sectional study, physically-active adults were recruited and assigned into
82	groups based on their experience with CF training and performance during specific CF
83	competitions. Participants who possessed CF training experience (> 2 years) were classified as
84	advanced (ADV) if they had previously qualified for the regional round of the Games
85	competition. Otherwise, they were classified as recreational (REC) because they had never
86	progressed beyond the opening round of the competition (i.e., The Open) but still trained on 3 –
87	5 days per week for at least the previous year. Individuals who did not possess CF training
88	experience but possessed resistance training experience (> 2 years) and participated in both
89	resistance and cardiovascular training on $3-5$ days per week for at least the previous year, were
90	assigned to the physically-active control (CON) group. All participants reported to the Exercise
91	Physiology Laboratory on two separate occasions, within one month of the onset of the Open, to
92	complete all testing. During the first visit, each participant provided a fasted blood sample before
93	completing assessments of muscle morphology and peak aerobic capacity. Participants returned
94	to the Exercise Physiology Lab for the second visit (within 3 – 7 days of the first visit) to

95 complete assessments of resting metabolic rate, body composition, strength, and anaerobic 96 performance. All testing sessions occurred in the morning ($\sim 6:00 - 10:00$ a.m.) with the 97 participants having abstained from unaccustomed physical activity and alcohol for 24 hours, 98 caffeine for 12 hours, and fasted for 8 hours. Participants completed all measurements while 99 wearing comfortable athletic clothing and were able to consume a light snack prior to 100 performance testing (i.e., peak aerobic capacity, strength, and anaerobic performance). Prior to 101 leaving the laboratory on the first visit, participants were asked to complete a 24-hour dietary 102 recall, retain a copy, and follow a similar diet prior to their second visit. Comparisons were made 103 between groups for all anthropometric, biochemical, and physiological measures.

104

105 **Participants**

106 Twenty-three physically-active adults $(29.7 \pm 6.8 \text{ years}, 171 \pm 12 \text{ cm}, 76.9 \pm 15.4 \text{ kg})$ 107 agreed to participate in this study. All participants were free of any physical limitations 108 (determined by medical and physical-activity history questionnaire and PAR-O+) and had been 109 regularly participating (at the time of recruitment) in their chosen exercise form (i.e., CrossFit 110 training or Resistance/Cardiovascular training) for a minimum of 2 years. Participants in ADV (n 111 $= 8,27.8 \pm 4.2$ years, 170 ± 11 cm, 79.8 ± 13.3 kg) reported having regularly participated in 112 resistance training for 11.5 ± 5.8 years and CF training for 6.4 ± 5.6 years $(6 - 7 \text{ sessions} \cdot \text{week}^{-1})$ 113 ¹). As individual competitors, the highest rank these participants ever achieved in the Open was 114 $659^{\text{th}} \pm 991^{\text{st}}$ (range: $19^{\text{th}} - 3,052^{\text{nd}}$) within their respective divisions worldwide. While each of 115 these athletes qualified for this study by having competed as members of a team in regional (highest average rank = $11^{\text{th}} \pm 13^{\text{th}}$) and Games competition (highest average rank = $20^{\text{th}} \pm 9^{\text{th}}$). 116 117 three competed individually in their respective regions with one having progressed to the Games

118 on multiple occasions. REC participants (n = 8, 33.5 ± 8.1 years, 172 ± 14 cm, 76.3 ± 19.5 kg) 119 reported having regularly participated in resistance training for 8.1 ± 7.9 years and CF training 120 for 3.3 ± 1.7 years (4 – 5 sessions week⁻¹). The highest rank these participants had ever achieved 121 in the Open was 22,306th $\pm 14,028$ th (range: 5,466th - 44,315th) within their respective divisions 122 worldwide. Participants in CON (n = 7, 27.5 ± 6.7 years, 171 ± 14 cm, 74.5 ± 14.3 kg) reported 123 having 7.6 ± 4.8 years of regular resistance training experience and incorporated 3.7 ± 1.3 124 sessions and 3.6 ± 1.0 sessions of resistance and cardiovascular training per week. Although two 125 participants in CON reported having previously participated in CF-style workouts, these did not 126 occur with regularity (≤ 3 sessions week⁻¹) or for an extended duration (≤ 1 year) and they had 127 never competed in the Open at the time of data collection. Following an explanation of all 128 procedures, risks and benefits, each participant provided his or her written informed consent to 129 participate in the study. The study was conducted in accordance with the Declaration of Helsinki, 130 and the protocol was approved by the Kennesaw State University Institutional Review Board 131 (#17-501).

132

133 Blood sampling and biochemical analysis

Blood samples were obtained on the first visit prior to any physical activity. All samples were obtained from an antecubital vein using a needle by a research team member who was trained and experienced in phlebotomy. Approximately 15 mL of blood was drawn into SST tubes (for serum collection) and EDTA-treated Vacutainer® tubes (for plasma). SST tubes were allowed to clot for 10 minutes prior to centrifugation, while EDTA treated tubes were centrifuged immediately for 10 minutes at 3600 rpms at 4 °C. The resulting serum and plasma were aliquoted and stored at -80°C until analysis.

141	Circulating concentrations of testosterone (T; in ng·dL ⁻¹), cortisol (C; in μ g·dL ⁻¹), and
142	insulin-like growth factor (IGF-1; in ng·mL ⁻¹) were assessed via enzyme-linked immunosorbent
143	assays (ELISA) via a 96-well spectrophotometer (BioTek, Winooski, VT) using commercially
144	available kits. To eliminate inter-assay variance, all samples for each assay were thawed once
145	and analyzed in duplicate in the same assay run by a single technician. Samples were analyzed in
146	duplicate, with an average coefficient of variation of 1.63% for T, 6.88% for C, and 2.00% IGF-
147	1.

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149 Muscle morphology

150 Non-invasive skeletal muscle ultrasound images were collected from the right thigh and 151 arm locations of all participants. This technique uses sound waves at fixed frequencies to create 152 in vivo, real time images of the limb musculature. Prior to image collection, all anatomical 153 locations of interest were identified using standardized landmarks for the rectus femoris (RF), 154 vastus medialis (VM), vastus lateralis (VL), biceps brachii (BB), and triceps brachii (TB) 155 muscles. The landmarks for the thigh musculature were identified along the longitudinal distance 156 over the femur. The RF and VM were respectively assessed at 50% and 20% of the distance from 157 the proximal border of the patella to the anterior, inferior suprailiac crest. The VL was assessed 158 at 50% of the distance from the lateral condyle of the tibia to the most prominent point of the 159 greater trochanter of the femur. VL measurement required the participant to lay on their side. 160 Landmark identification of the BB and TB required the participant to sit upright on the 161 examination table and extend their arm to rest upon the shoulder of the researcher. Both muscles 162 were assessed along the humerus at a position equal to 40% of the distance from the lateral 163 epicondyle to the acromion process of the scapula (28). Subsequently, the participant resumed

laying supine on the examination table for a minimum of 5 – 10 minutes to allow fluid shifts to
occur before images were collected (29). The same investigator performed all landmark
measurements for each participant.

167 A 12 MHz linear probe scanning head (General Electric LOGIQ S7 Expert, Wauwatosa, 168 WI, USA) was coated with water soluble transmission gel to optimize spatial resolution and used 169 to collect all ultrasound images. Collection of each image began with the probe being positioned 170 on (and perpendicular to) the surface of the skin to provide acoustic contact without depressing the dermal layer. Subsequently, two consecutive images were collected in the extended field of 171 172 view mode (Gain = 50 dB; Image Depth = 5 - 6 cm) using a cross-sectional sweep in the axial 173 plane to capture panoramic images of each muscle. At the same sites, two consecutive images 174 were collected with the probe oriented longitudinal to the muscle tissue interface using 175 Brightness Mode (B-mode) ultrasound (30). Each of these images included a horizontal line 176 (approximately 1 cm), located below the image, which was used for calibration purposes when 177 analyzing the images offline (31). To capture images of the RF and VM, the participant remained 178 in the supine position, with their legs extended but relaxed. A rolled towel was placed beneath 179 the popliteal fossa of the dominant leg, allowing for a 10° bend in the knee as measured by a 180 goniometer, and the dominant foot secured (32). For the VL, the participant was placed on their 181 side with their legs together and the rolled towel between their needs. Once again, the legs were 182 positioned to allow a 10° bend in the knees, as measured by a goniometer (32). Measurement of 183 the BB and TB required the participant to sit upright with their arm extended, resting on the 184 shoulder of the researcher. The same investigator positioned each participant and collected all 185 images.

186 After all images were collected, the ultrasound data were transferred to a personal 187 computer for analysis via Image J (National Institutes of Health, Bethesda, MD, USA, version 188 1.45s) by the same technician. All panoramic images were used to measure cross-sectional area 189 (CSA) and echo intensity. For these measures, the polygon tracking tool in the ImageJ software 190 was used to isolate as much lean muscle as possible without any surrounding bone or fascia (30). 191 Subsequently, Image J calculated the area contained within the traced muscular image and 192 reported this value in centimeters squared (± 0.1 cm²). Concurrently, echo intensity was 193 determined by grayscale analysis using the standard histogram function in ImageJ (30) and 194 expressed as an arbitrary unit (au) value between 0 - 255 (0: black; 255: white) with lower 195 values reflecting more contractile tissue within each muscle (30, 33). Mean echo intensity values 196 were then corrected for subcutaneous fat thickness (SFT; averaged from the SFT values obtained 197 at the medial, midline, and lateral sites of each muscle) using Equation 1 (34). All B-mode 198 images were used to measure muscle thickness (± 0.01 cm; perpendicular distance between the 199 superficial and deep aponeuroses) and pennation angle ($\pm 0.1^{\circ}$; intersection of the fascicles with 200 the deep aponeurosis). Fascicle length (± 0.1 cm) across the deep and superficial aponeuroses 201 was estimated from muscle thickness and pennation angle using Equation 2. Intraclass 202 correlation coefficients (ICC_{3 k} = 0.77 - 0.99) for determining muscle thickness, pennation angle, 203 CSA and echo intensity was previously determined in ten active, resistance-trained men $(25.3 \pm$ 204 2.0 years, 180 ± 7 cm, 90.8 ± 6.8 kg) using the methodology described above. The methodology 205 for determination of FL has a reported estimated coefficient of variation of 4.7% (35). 206 Corrected echo intensity (EI) = Raw echo intensity + (SFT x 40.5278) Equation 1: 207 Equation 2: Fascicle length = Muscle thickness $\cdot \sin (\text{pennation angle})^{-1}$ 208

209 Peak aerobic capacity

210 Peak aerobic capacity (VO₂peak; ml·kg⁻¹·min⁻¹), respiratory compensation threshold 211 (RCT; ml·kg⁻¹·min⁻¹), and gas exchange threshold (GET; ml·kg⁻¹·min⁻¹) were assessed using a 212 continuous, ramp exercise protocol performed on an electromagnetic-braked cycle ergometer 213 (Lode Excalibur Sport, Lode., B.V., Groningen, The Netherlands). Prior to testing, each 214 participant completed a standardized warm-up that consisted of riding a cycle ergometer for 5 215 minutes at the participant's preferred resistance and cadence followed by 10 body weight squats, 216 10 alternating lunges, 10 walking knee hugs and 10 walking butt kicks. Participants were then 217 permitted to continue their warm-up with any additional practices that would help them feel 218 comfortable entering the test. Participants were fitted with a heart rate (HR) monitor (Team², 219 Polar, Lake Success, NY), a nose clip, and a 2-way valve mask connected to a metabolic 220 measurement system (True One 2400, ParvoMedics Inc., Salt Lake City, UT) to measure expired 221 gases. The cycle ergometer seat height and handlebar distance were adjusted to the participant's 222 comfort. The participants initially completed a 3-minute warm-up period with the resistance set 223 at 50 W before starting the test at 75 W. During testing, the participants were asked to maintain a 224 self-selected pedaling rate (> 50 rpm's) while power output was increased by 25 W every minute 225 until volitional fatigue or pedaling rate dropped below 50 rpm's for longer than 15 seconds. 226 Upon completion of the test, each participant immediately progressed to a 3-minute active 227 recovery period where they continued to pedal at their own cadence against a 50 W load. HR was 228 assessed on each minute of the 3-minute recovery period. Participants were then removed from 229 the cycle ergometer and asked to rest in a chair for an additional two minutes. 230 Relative oxygen consumption values (i.e., $VO_2 \cdot kg^{-1}$) collected on each breath were

averaged using the 11-breath averaging technique (36) and used to determine the highest value

232	achieved during the test (i.e., VO ₂ peak). RCT, also known as the second ventilatory threshold,
233	was identified as the VO_2 value at which the increase in ventilation- VO_2 relationship was
234	accompanied by an increase in the ventilation-VCO ₂ relationships (37). The GET was
235	determined using the V-slope method described by Beaver et al. (38). The GET was defined as
236	the VO ₂ value corresponding to the intersection of two linear regression lines derived separately
237	from the data points below and above the breakpoint in the CO ₂ produced (VCO ₂) versus the
238	VO_2 relationship (39).
239	

240 **Dietary recall**

241 Participant's dietary intake was tracked for the 24-hour period preceding each visit via a 242 paper dietary food recall form. All participants were instructed on how to properly log their food, 243 snacks and drinks via the paper form. Specifically, following their enrollment on their first visit, 244 participants were asked to record their food intake (breakfast, lunch, dinner, drinks and snacks) 245 for the previous 24 hours prior. Prior to leaving the laboratory on the first visit, the participants 246 were given a copy of their food recall form and asked to consume a similar diet during the 24 247 hours prior to their second visit. Each form was visually inspected to confirm dietary 248 compliance.

249

250 **Resting metabolic rate assessment**

Resting metabolic rate (RMR, kcals·day⁻¹) assessment was completed in a quiet room
with minimal lighting (e.g., only light from the RMR machine) located within the Exercise
Physiology Laboratory. Prior to their arrival, participants were informed of all pre-test guidelines
as outlined by Compher et al. (40). These included: 1) avoiding alcohol consumption 24 hours

255	prior to testing, 2) no food or caffeine ingestion 8 and 12 hours prior to testing, respectively, and
256	3) discontinuing unaccustomed physical activity 24 hours prior to testing. Resting metabolic rate
257	was measured via a metabolic measurement system (Parvo Medics TrueOne 2400, ParvoMedics
258	Inc., Salt Lake City, UT) utilizing a ventilated hood. Participants were asked to rest in the supine
259	position with the ventilated hood placed over their face and neck for a maximum of 30 minutes.
260	RMR determination was based on a 5-minute interval of measured volume of oxygen
261	consumption (VO ₂) with a coefficient of variation less than 10% (40). The average coefficient of
262	variation was 6.36%.
263	
264	Body composition assessments
265	Initially, height (\pm 0.1 cm) and body mass (\pm 0.1 kg) were determined using a stadiometer

266 (WB-3000, TANITA Corporation, Tokyo, Japan) with the participants standing barefoot, with 267 feet together, in their normal daily attire. Subsequently, body composition was assessed by three 268 common methods (i.e., dual energy X-ray absorptiometry [iDXA, Lunar Corporation, Madison, 269 WI], air displacement plethysmography [BodPod, COSMED USA Inc., Chicago, IL], and 270 bioelectrical impedance analysis [770 Body Composition and Body Water Analyzer, InBody, 271 Seoul, South Korea]) using standardized procedures. Briefly, iDXA scanning required 272 participants to remove any metal or jewelry and lay supine on the iDXA table prior to an entire 273 body scan in "standard" mode using the company's recommended procedures and supplied 274 algorithms. Quality assurance was assessed by daily calibrations performed prior to all scans 275 using a calibration block provided by the manufacturer. All iDXA measurements were 276 performed by the same researcher using standardized subject positioning procedures. For air 277 displacement plethysmography, the device and associated scale were calibrated daily using a

278 known volume and mass provided by the manufacturer. During testing, participants were asked 279 to wear a tight-fitting bathing suit or compression shorts and swim cap before entering the 280 device. Two trials were performed for each participant to obtain two measurements of body 281 volume within 150 mL. A third trial was performed if body volume estimates from the first two 282 trials were not within 150 mL, and values from the two closest trials were averaged. Thoracic 283 lung volume was estimated (41). Bioelectrical impedance analysis required participants to stand 284 barefoot on two metal sensors located at the base of the device and hold two hand grips for 285 approximately 30-60 seconds. Prior to stepping onto the device, participants cleaned the soles 286 of their feet with alcohol wipes provided by the manufacturer. 287 Following testing, body mass, bone mineral content (BMC; from iDXA), body volume 288 (from BodPod), and total body water (from bioelectrical impedance analysis) were entered into a 289 4-compartment model, Equation 3 to estimate body fat percentage (BF%) (42), fat mass (± 0.1 290 kg), and fat-free mass (± 0.1 kg). These values, along with regional (arms [sum of each arm], 291 legs [sum of each leg], and trunk [sum of spine and pelvis]) estimates of bone mineral content (± 292 0.1 kg) and non-bone lean mass (\pm 0.1 kg) obtained from iDXA following manual demarcation 293 of these regions of interest were used for all group comparisons. Intraclass correlation 294 coefficients (ICC_{3,1} = 0.74 - 0.99) for manually determining regional estimates of bone mineral 295 content and non-bone lean mass had been previously found in 10 healthy, physically-active 296 adults $(25.1 \pm 2.4 \text{ years}; 176 \pm 7 \text{ cm}, 81.1 \pm 18.5 \text{ kg})$. $BF\% = \frac{(2.748 \, x \, volume) - (0.699 \, x \, water) + (1.129 \, x \, BMC) - (2.051 \, x \, Body \, Mass)}{R_{\rm eff} + M_{\rm eff}} \times 100$ 297 Equation 3: Bodv Mass 298

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- 300

301 Strength assessment

302 Following RMR and body composition assessments, strength was assessed by an 303 isometric mid-thigh pull test. Prior to testing, each participant completed the same standardized 304 warm-up described for the first visit (i.e., 5 minutes of cycling, dynamic stretching, additional 305 self-selected warm-up practices) followed by a protocol specific to the isometric mid-thigh pull 306 test. The specific component included three isometric efforts on an immobilized barbell 307 positioned at approximately the mid-thigh using a perceived intensity of 50, 70, and 90% of 308 maximum effort, interspersed with a one-minute recovery. The specific warm-up and isometric 309 mid-thigh pull test were completed within a power rack (Rogue Fitness, Columbus, OH) while 310 standing upon a portable force plate (Accupower, AMTI, Watertown, MA). While standing on 311 the force plate, the mid-thigh position was determined for each participant before testing by 312 marking the midpoint distance between the knee and hip joints. Each participant was instructed 313 to assume their preferred second pull power-clean position by self-selecting their hip and knee 314 angles. The height of the barbell was adjusted to a position approximately equal (± 2.54 cm) to 315 the mid-thigh. The participants were then asked to use an overhand, hooked grip on the barbell. 316 The hook grip was selected for this test because all participants reported having had experience 317 with the technique and it is commonly used among CF athletes during competition. Participants 318 were also allowed to wrap their thumbs with athletic training tape and use chalk. Upon the 319 researcher's "3, 2, 1, Go!" command, the participants were instructed to pull upwards on the 320 barbell as hard and as fast as possible and to continue their maximal effort for 6 seconds. All 321 participants were instructed to relax before the command "GO!" to avoid precontraction and 322 were allotted three maximal attempts. The portable force plate measured the ground reaction 323 forces, imposed onto the plate by the participant, as he/she pulled upon the bar. Peak force (F; in

N) production, peak and average rate of force development (RFD_{PEAK} , RFD_{AVG} ; in N·s⁻¹), and F and RFD across specific time bands (i.e., 0–30, 0–50, 0–90, 0–100, 0–150, 0–200, and 0–250

326 milliseconds) were subsequently calculated, as previously described (43).

327

328 Anaerobic performance assessment

329 Following the strength assessment, anaerobic performance was assessed via a 3-minute 330 maximal sprint on an electromagnetic-braked cycle ergometer (Lode Excalibur Sport, Lode., 331 B.V., Groningen, The Netherlands). Prior to the test, seat height and handlebar positions were 332 adjusted to mirror their positions during the peak aerobic capacity test, and participants were 333 provided with time ($\sim 3 - 5$ minutes) to acclimate to the cycle ergometer. A 5-minute rest period 334 was then allotted before initiating the testing protocol, which has been previously described in 335 detail elsewhere (44). Briefly, the test began with a 1-minute baseline period that involved 55 336 seconds of unloaded cycling at 90 rpm and then accelerating up to approximately 110 rpm over 337 the last 5 seconds of the minute. The protocol immediately transitioned to the 3-minute testing 338 period where the participants attempted to maintain cadence as high as possible throughout its 339 entirety. Resistance for the test was set using the linear mode of the cycle ergometer (linear 340 factor = power / [preferred cadence]²). That is, the linear factor was calculated as the power 341 output halfway between the VO₂peak and GET, divided by the preferred cadence of untrained 342 cyclists (70 rpm²) (45-47). To prevent pacing and ensure an all-out effort, participants were not 343 informed of the elapsed time and strong verbal encouragement was provided. After 3 minutes, 344 the participants progressed to a 3-minute recovery stage at 50 Watts at their preferred cadence. 345 Peak power (± 1 W), critical power (CP; average power over the final 30 seconds of the test; ± 1

W) (46), and anaerobic work capacity (AWC; work done above CP; $(\pm 0.1 \text{ kJ})$ (47) were calculated based upon performance during the 3-minute sprint test.

348

349 Statistical analysis

350 Data were modeled using both a frequentist and Bayesian approach. The frequentist 351 approach involved a two-way (Group x Sex) analysis of variance (ANOVA) for each dependent 352 variable. Assumptions of normality and equal variance were verified by Shapiro-Wilk and 353 Levene's tests, respectively. Significant interactions and main effects were further examined 354 using Tukey's post-hoc analysis. Criterion alpha was set at p < 0.05. To further assess the 355 likelihood (or the effect of group and/or sex) of the data under the alternative hypothesis 356 compared to the null hypothesis, a two-way Bayesian ANOVA was performed with default prior 357 scales (48). Likelihood was represented in the form of Bayes factors (i.e., BF_{10}) and were 358 interpreted according to the recommendations of Wagenmakers et al. (49). That is, data were 359 interpreted as evidence in favor of the null hypothesis when $BF_{10} < 1$. Otherwise, it was 360 interpreted as "anecdotally" ($1 \le BF_{10} \le 3$), "moderately" ($3 \le BF_{10} \le 10$), "strongly" ($10 \le BF_{10}$) 361 < 30), "very strongly" ($30 < BF_{10} < 100$), or "extremely" ($BF_{10} > 100$) in favor of the alternative 362 hypothesis. All statistical analyses were performed using JASP 0.10.2 (Amsterdam, the 363 Netherlands). All data are reported as mean \pm standard deviation. 364 365 366 367

369	RESULTS
370	
371	Resting hormone concentrations
372	No interactions were observed for T, C, IGF-1. However, a trend for an interaction (F =
373	2.87, $p = 0.090$) driven by a main sex effect was seen for T (F = 6.11, $p = 0.027$) with anecdotal
374	differences between sexes being 2.058 times likely compared to the null hypothesis. Specifically,
375	women in ADV tended to exhibit lower T concentrations ($p = 0.83$) than ADV men. Male and
376	female hormone concentrations are illustrated in Fig 1.
377 378 379	Fig 1. Male and female resting concentrations in A) testosterone, B) cortisol, and C) IGF-1.
380	Muscle morphology
381	Measures of muscle morphology for each group and sex are presented in Table 1.
382	Significant ($p < 0.05$) group x sex interactions were observed for BB fascicle length and EI for
383	each muscle, though the likelihood of these interactions favored the null hypotheses (BF ₁₀ < 1).
384	Rather, the observed interactions were primarily driven by <i>anecdotal</i> -to- <i>strong</i> evidence ($1.7 <$
385	$BF_{10} < 30.0$) of main effects for sex and group. The observed interaction for BB fascicle length
386	was primarily driven by a main effect for sex where women were 8.8 times more likely to
387	possess shorter fascicles than men, specifically REC women compared to the men of REC ($p =$
388	0.029) and CON ($p = 0.012$). Though the underlying causes for the interactions seen for EI
389	varied with each muscle, <i>anecdotal</i> -to- <i>moderate</i> evidence indicated that men were $1.7 - 5.5$
390	times more likely to possess a lower EI than women. Specifically, women in REC possessed
391	higher EI (p < 0.05) than men in ADV (RF, VL, and TB; a trend [$p = 0.056$] for VM) and REC
392	(RF, VM, VL, and TB; a trend [$p = 0.087$] was noted for BB), and tended ($p < 0.10$) to be higher

393	than men in CON (RF, VL, and TB). Even though a main effect was not seen, the effect of group
394	was 2.4 – 30.0 times likely to influence EI. Specifically, post-hoc analysis of the interaction
395	showed that women in REC possessed higher EI than their counterparts in ADV (RF, VM, VL,
396	and TB).
397	Significant group effects were found for muscle thickness (VL and BB), pennation angle
398	(BB and TB), fascicle length of TB, and CSA (VM and VL). Compared to CON, ADV possessed
399	greater muscle thickness in VL ($p = 0.013$, BF ₁₀ = 3.0) and in BB ($p = 0.012$, BF ₁₀ = 2.2), larger
400	BB pennation angle ($p = 0.007$, BF ₁₀ = 21.9), and greater CSA in VM ($p = 0.050$, BF ₁₀ = 2.1)
401	and VL ($p = 0.009$, BF ₁₀ = 0.7). Compared to REC, ADV possessed greater muscle thickness in
402	VL ($p = 0.026$, BF ₁₀ = 1.9), larger pennation angle in TB ($p = 0.009$, BF ₁₀ = 3.2), longer fascicles
403	in TB ($p = 0.019$, BF ₁₀ = 3.9), and greater CSA in VL ($p = 0.009$, BF ₁₀ = 0.8); a tendency for
404	greater muscle thickness in BB was also noted for ADV compared to REC ($p = 0.086$, BF ₁₀ =
405	0.9). No differences were seen between REC and CON. Morphological comparisons are
406	presented in Table 1.

		A	DV	REC		CON		Group		Sex		Group x Sex	
		Women	Men	Women	Men	Women	Men	р	\mathbf{BF}_{10}	р	\mathbf{BF}_{10}	р	\mathbf{BF}_{10}
Muscle thickness (cm)	Rectus femoris	2.48 ± 0.36	3.28 ± 0.50	2.32 ± 0.27	2.86 ± 0.20	2.46 ± 0.25	2.61 ± 0.43	0.155	8.2	0.004	6.7	0.248	0.5
	Vastus medialis	3.44 ± 0.84	4.35 ± 0.58	2.77 ± 0.08	4.28 ± 0.61	3.41 ± 0.62	4.26 ± 0.22	0.439	>100	0.001	39.8	0.502	0.3
	Vastus lateralis	1.92 ± 0.49	2.47 ± 0.39	1.49 ± 0.25	1.97 ± 0.19	1.63 ± 0.19	1.67 ± 0.28	0.009	11.9	0.017	7.9	0.288	1.7
	Biceps brachii	3.32 ± 0.60	4.29 ± 0.87	2.62 ± 0.19	3.74 ± 0.71	2.43 ± 0.08	3.31 ± 0.17	0.013	>100	0.001	40.6	0.910	1.3
	Triceps brachii	2.51 ± 0.45	3.05 ± 0.76	2.44 ± 0.64	2.91 ± 0.58	1.98 ± 0.24	3.05 ± 0.43	0.674	7.1	0.008	2.1	0.541	0.3
Pennation angle (°)	Rectus femoris	12.7 ± 3.5	15.5 ± 0.5	10.0 ± 3.0	14.5 ± 1.2	13.3 ± 5.6	16.4 ± 5.5	0.375	2.9	0.036	1.4	0.894	0.5
	Vastus medialis	19.2 ± 3.9	26.2 ± 9.3	17.2 ± 3.4	24.9 ± 6.0	27.7 ± 10.8	24.0 ± 3.5	0.417	0.7	0.216	0.4	0.229	0.2
	Vastus lateralis	14.3 ± 3.1	14.8 ± 2.9	10.7 ± 3.4	12.4 ± 5.8	12.3 ± 0.4	13.2 ± 3.5	0.286	0.6	0.502	0.5	0.947	0.1
	Biceps brachii	13.6 ± 3.1	17.5 ± 2.2	12.8 ± 2.1	12.3 ± 5.0	10.5 ± 2.2	9.7 ± 1.5	0.009	7.0	0.489	3.2	0.249	0.4
	Triceps brachii	17.9 ± 5.1	26.8 ± 7.8	11.1 ± 3.2	16.8 ± 4.5	14.2 ± 2.6	20.3 ± 4.1	0.012	34.5	0.004	13.8	0.791	2.4
Fascicle length (cm)	Rectus femoris	12.2 ± 5.1	12.3 ± 2.2	14.1 ± 3.8	11.5 ± 0.8	12.8 ± 7.6	10.3 ± 4.5	0.852	0.6	0.365	0.3	0.786	0.1
	Vastus medialis	10.6 ± 2.4	11.2 ± 4.8	9.6 ± 1.8	10.6 ± 2.7	7.9 ± 2.4	10.6 ± 1.1	0.552	0.6	0.280	0.3	0.758	0.1
	Vastus lateralis	7.7 ± 0.4	9.9 ± 2.1	8.3 ± 1.5	10.5 ± 4.6	7.6 ± 0.6	7.7 ± 2.2	0.399	0.8	0.163	0.4	0.643	0.2
	Biceps brachii	14.5 ± 2.8	14.2 ± 1.3	$12.0 \pm 1.1^{\rm df}$	$19.0\pm4.8^{\rm c}$	13.8 ± 2.8	$19.9\pm2.7^{\circ}$	0.271	9.8	0.002	8.8	0.043	0.5
	Triceps brachii	8.7 ± 2.8	7.3 ± 2.9	12.8 ± 1.7	10.5 ± 3.1	8.2 ± 0.9	9.1 ± 2.2	0.018	4.3	0.370	2.4	0.448	0.5
Cross-sectional area (cm ²)	Rectus femoris	10.8 ± 2.4	17.8 ± 3.3	8.8 ± 1.5	15.6 ± 1.8	11.2 ± 1.3	15.0 ± 2.8	0.216	>100	0.000	>100	0.364	0.4
	Vastus medialis	24.3 ± 6.6	29.8 ± 4	17.0 ± 3.9	27.8 ± 6.4	18.2 ± 2.1	23.7 ± 1.2	0.046	22.3	0.002	12.0	0.441	0.8
	Vastus lateralis	29.8 ± 4.1	44.9 ± 4.1	24.4 ± 2.6	38.2 ± 4.5	24.1 ± 1.7	37.9 ± 3.0	0.004	>100	0.001	>100	0.912	0.5
	Biceps brachii	8.4 ± 1.7	17.7 ± 9.2	7.4 ± 2.0	14.9 ± 2.5	7.9 ± 0.2	12.9 ± 1.0	0.464	77.5	0.001	32.6	0.622	0.3
	Triceps brachii	10.5 ± 1.2	18 ± 4.3	7.0 ± 1.4	17.0 ± 5.7	8.9 ± 1.4	14.0 ± 3.6	0.273	>100	0.000	>100	0.417	0.3
Echo intensity (au)	Rectus femoris	$116 \pm 26^{\circ}$	$113 \pm 11^{\circ}$	174 ± 33^{abd}	$97\pm14^{\text{ce}}$	151 ± 16^{d}	129 ± 14	0.061	30.0	0.001	5.5	0.008	0.6
	Vastus medialis	$105\pm16^{\rm c}$	108 ± 11	153 ± 39^{ad}	$104 \pm 12^{\circ}$	116 ± 11	134 ± 13	0.093	2.4	0.289	0.8	0.012	0.4
	Vastus lateralis	$111\pm20^{\circ}$	$113 \pm 15^{\circ}$	171 ± 42^{abd}	$107 \pm 19^{\circ}$	134 ± 16	123 ± 10	0.087	4.3	0.023	1.7	0.028	0.7
	Biceps brachii	123 ± 26	140 ± 9	170 ± 45	115 ± 29	148 ± 9	142 ± 17	0.585	0.6	0.206	0.5	0.044	0.2
	Triceps brachii	$83 \pm 17^{\circ}$	$89 \pm 18^{\circ}$	145 ± 43^{abd}	$78 \pm 13^{\circ}$	114 ± 16	100 ± 6	0.079	7.2	0.014	1.8	0.012	0.6

407 Table 1. Measures of muscle morphology by group and sex.

Note: ^a = Significantly (p < 0.05) different from ADV women; ^b = Significantly (p < 0.05) different from ADV men; ^c = Significantly

(p < 0.05) different from REC women; ^d = Significantly (p < 0.05) different from REC men; ^e = Significantly (p < 0.05) different from 410 CON women; ^f = Significantly (p < 0.05) different from CON men.

411 **Peak aerobic capacity**

412	No significant group x sex interactions were observed for VO ₂ peak (F = 1.09, p = 0.358,
413	$BF_{10} = 10.1$), RCT (F = 0.32, p = 0.730, $BF_{10} = 1.7$), or GET (F = 0.05, p = 0.949, $BF_{10} = 1.1$).
414	However, moderate-to-strong evidence were found in favor of main group effects for each
415	variable. VO ₂ peak (F = 9.10, $p = 0.002$, BF ₁₀ = 17.0) and RCT (F = 5.56, $p = 0.014$, BF ₁₀ = 4.5)
416	were significantly greater in ADV compared to REC (p \leq 0.039) and CON (p \leq 0.020), while
417	GET (F = 5.29, p = 0.016, BF ₁₀ = 5.7) was significantly greater in ADV compared to CON (p =
418	0.016) and tended to be greater compared to REC ($p = 0.087$). No differences were seen between
419	REC and CON. Further, the percentage of VO2peak for GET and RCT were similar between
420	ADV (GET = 55.2 ± 11.2%; RCT = 71.7 ± 7.5%), REC (GET = 55.9 ± 6.8%; RCT = 73.5 ±
421	5.9%), and CON (GET = 53.9 \pm 4.3%; RCT = 74.6 \pm 7.7%). Group differences in absolute
422	measures of aerobic performance are illustrated in Fig 2.
423 424 425 426	Fig 2. Group differences in aerobic performance measures. <i>Note</i> : $* =$ Significantly (p < 0.05) different from ADV. $# =$ Different (p < 0.10) from ADV.
427	Resting metabolic rate
428	Neither a group x sex interaction (F = 0.21, p = 0.817, BF ₁₀ = 0.2) or main group effect
429	(F = 1.67, $p = 0.220$, BF ₁₀ = 0.1) was observed for RMR recordings in ADV (1788 ± 232
430	kcal·day ⁻¹), REC (1768 ± 407 kcal·day ⁻¹), and CON (1572 ± 356 kcal·day ⁻¹).
431	Body composition
432	No significant group x sex interactions were observed for any measure of body
433	composition (presented in Table 2). However, the evidence was strongly-to-extremely in favor of
434	main group effects for body density, regional and total BMC, regional and total lean mass, and
435	BF%. Compared to the REC, ADV possessed greater body density ($p = 0.004$), greater BMC of

- 436 the arms (p = 0.009), greater lean mass (i.e., total and regional; $p \le 0.035$), lower BF% (p =
- 437 0.009), and tended to possess more BMC (total-body: p = 0.066; legs: p = 0.060) and less fat
- 438 mass (p = 0.064). Compared to CON, ADV possessed greater body density (p = 0.006), greater
- 439 BMC throughout the body ($p \le 0.024$), lean mass throughout the body ($p \le 0.009$), and lower
- 440 BF% (p = 0.023). No differences were observed between REC and CON.

	ADV				REC			CON				Group x Sex	
	Women	Men	Total	Women	Men	Total	Women	Men	Total	р	\mathbf{BF}_{10}	р	BF ₁₀
<u>Anthropometric</u>													
Height (cm)	160 ± 13	177 ± 3	170 ± 11	161 ± 4	183 ± 8	172 ± 14	158 ± 4	180 ± 9	171 ± 14	0.785	>100	0.526	0.3
Weight (kg)	68.3 ± 5.0	91.5 ± 5.1	79.8 ± 13.3	59.0 ± 2.0	93.5 ± 9.5	76.3 ± 19.5	60.8 ± 6.3	84.9 ± 7.3	74.5 ± 14.3	0.127	>100	0.169	0.3
BMI (kg·m ⁻²)	26.0 ± 3.5	29.2 ± 1.9	27.6 ± 3.1	22.9 ± 1.2	28.0 ± 3.4	25.5 ± 3.6	24.4 ± 3.4	26.1 ± 0.7	25.4 ± 2.2	0.163	6.6	0.456	0.6
Density (kg·L ⁻¹)	1.07 ± 0.01	1.07 ± 0.01	1.07 ± 0.01	1.05 ± 0.01	1.06 ± 0.01	$1.05\pm0.01*$	1.04 ± 0.02	1.06 ± 0.01	$1.05\pm0.02*$	0.002	13.8	0.159	1.1
Bone Mineral Content (kg)													
Total	3.05 ± 0.38	3.75 ± 0.13	3.45 ± 0.44	2.42 ± 0.16	3.62 ± 0.41	$3.02\pm0.70\#$	2.43 ± 0.14	3.29 ± 0.41	$2.92\pm0.55*$	0.012	>100	0.299	0.7
Arms	0.45 ± 0.07	0.62 ± 0.05	0.55 ± 0.11	0.32 ± 0.03	0.57 ± 0.05	$0.44 \pm 0.14*$	0.30 ± 0.01	0.48 ± 0.06	$0.40 \pm 0.11*$	0.001	>100	0.266	1.2
Legs	1.12 ± 0.13	1.44 ± 0.11	1.30 ± 0.20	0.82 ± 0.05	1.38 ± 0.17	$1.10 \pm 0.32 \#$	0.81 ± 0.03	1.31 ± 0.22	$1.09 \pm 0.31*$	0.022	>100	0.255	0.5
Trunk	0.95 ± 0.11	1.16 ± 0.03	1.07 ± 0.13	0.79 ± 0.11	1.11 ± 0.14	0.95 ± 0.21	0.82 ± 0.08	0.97 ± 0.09	$0.90\pm0.11*$	0.028	>100	0.271	0.8
<u>Non-bone fat-free mass (kg)</u>													
Arms	7.15 ± 0.89	11.12 ± 1.22	9.42 ± 2.35	4.87 ± 0.49	10.02 ± 0.56	$7.45 \pm 2.79*$	4.83 ± 0.42	9.04 ± 1.14	$7.24\pm2.40*$	0.001	>100	0.400	0.6
Legs	18.4 ± 1.4	25.4 ± 1.6	22.4 ± 4.0	14.3 ± 1.0	24.4 ± 1.1	$19.3\pm5.4*$	14.7 ± 0.7	22.5 ± 3.2	$19.2\pm4.7*$	0.008	>100	0.252	0.5
Trunk	27.7 ± 2.9	35.2 ± 2.0	32.0 ± 4.5	20.3 ± 1.2	33.5 ± 1.4	$26.9\pm7.2*$	21.4 ± 2.2	30.1 ± 3.7	$26.4 \pm 5.5*$	0.001	>100	0.073	0.8
4-compartment model													
Body fat percentage (%)	11.9 ± 2.4	11.0 ± 2.6	11.4 ± 2.3	23.3 ± 2.4	16.1 ± 6.2	$19.7\pm5.8*$	23.9 ± 8.4	13.7 ± 3.2	$18.1\pm7.6*$	0.007	16.1	0.183	2.7
Fat-free mass (kg)	60.2 ± 3.5	81.3 ± 3.4	72.3 ± 11.7	45.2 ± 2.3	78.1 ± 5.3	$61.7 \pm 18.0*$	45.9 ± 1.6	73.3 ± 8.4	61.6 ± 15.9*	0.001	>100	0.097	0.5
Fat mass (kg)	8.2 ± 2.1	10.2 ± 2.7	9.3 ± 2.5	13.8 ± 1.4	15.4 ± 7.0	14.6 ± 4.7#	14.9 ± 6.7	11.5 ± 2.2	13.0 ± 4.5	0.069	1.5	0.436	0.3

441 <u>Table 2. Group differences in measures of body composition.</u>

442 Note: * = Significantly (p < 0.05) different from ADV, # = Different (p < 0.10) from ADV.

443 Strength

444	No significant group x sex interactions were observed for variables obtained from the
445	isometric mid-thigh pull assessment. Extreme evidence suggested significant main group effects
446	for F (F = 3.89, p = 0.042, BF ₁₀ = 667,577) and RFD at 200 ms (F = 3.67, p = 0.049, BF ₁₀ =
447	12,676), as well as tendencies for group differences in F at 150 ms (F = 2.80, p = 0.091, BF ₁₀ =
448	1,898), F at 200 ms (F = 3.50, $p = 0.055$, BF ₁₀ = 17,296), F at 250 ms (F = 3.14, $p = 0.071$, BF ₁₀
449	= 21524), RFD at 150 ms (F = 2.94, p = 0.082, BF ₁₀ = 1,868), and RFD at 250 ms (F = 3.37, p =
450	0.060, $BF_{10} = 20,187$). According to post-hoc analysis, ADV produced a higher peak F than
451	CON ($p = 0.036$) and expressed greater RFD at 200 ms than REC ($p = 0.049$). ADV also tended
452	to produce greater F at 200 ms ($p = 0.062$) and 250 ms ($p = 0.097$) compared to REC. No other
453	specific differences were seen between groups. Group differences in F and RFD production
454	across time are illustrated in Fig 3.
455 456 457 458 459	Fig 3. Group differences in A) force and B) rate of force production during an isometric mid- thigh pull. <i>Note</i> : $* =$ Significantly ($p < 0.05$) different from ADV. $# =$ Different ($p < 0.10$) from ADV.
460	Anaerobic performance
461	No significant group x sex interactions were observed. <i>Extreme</i> evidence in favor of a
462	significant group main effect for CP (F = 7.56, $p = 0.005$, BF ₁₀ = 267) indicated that ADV
463	possessed a higher CP than REC ($p = 0.029$) and CON ($p = 0.005$). Although extreme evidence
464	was also seen for AWC (F = 4.79, $p = 0.023$, BF ₁₀ = 247), post-hoc analysis did not reveal
465	specific group differences. No other differences were observed. Group differences in anaerobic
466	performance are illustrated in Fig 4.
467 468	Fig 4. Group differences in A) anaerobic work capacity, B) peak power, and C) critical power. <i>Note</i> : $* =$ Significantly (p < 0.05) different than ADV

469

DISCUSSION

470

471 The primary objectives of this study were to examine anthropometric, hormonal, and 472 physiological differences between advanced CF athletes, recreational CF participants, and 473 resistance and cardiovascular trained adults. Previously, only one other cross-sectional 474 investigation has made physiological comparisons between individuals with at least one year of 475 CF or resistance-training experience (27). The authors reported no differences between the 476 groups except for the CF-trained group possessing greater aerobic ability. This outcome, 477 however, is not surprising considering that the resistance-trained group was not required to also 478 have been performing aerobic exercise. Typical CF training workouts will concurrently 479 incorporate strength and conditioning elements into training (1, 2, 50). Although the conditioning 480 component varies in intensity and duration for each workout, it is important that alternative 481 exercise strategies include both elements to make a fair comparison. The present study builds 482 upon this limitation by having required participants in the CON group to have been participating 483 in both resistance and cardiovascular training on at least 3 days per week each; a similar training 484 frequency was expected of the recreational CF group (i.e., training on at least 3 days per week). 485 Another important aspect of CF training worth consideration is that it includes a wide variety of 486 traditional resistance and aerobic training exercises, along with simple-to-complex gymnastic 487 movements. Proficiency in these movements cannot be assumed after only a year of training and 488 would likely necessitate frequent workout modification. Recently, our group has reported 489 different physiological responses and recovery rates to CF workouts that are completed as 490 prescribed versus those that are modified (i.e., scaled) (11). Thus, CF-trained participants were 491 required to possess at least two years of experience and they were further divided into ADV and

492 REC based upon evidence of their skill as CF athletes (i.e., their previous success in CF 493 competition). Within these contexts, advanced CF athletes were observed to have a more 494 favorable body composition and muscular morphological characteristics, as well as greater 495 aerobic capacity, strength, and ability to sustain high-intensity effort compared to recreational CF 496 participants and physically-active adults. In contrast, no differences were observed between 497 recreational CF participants and physically-active adults in any measure and no differences were 498 seen in resting hormone concentrations or metabolic rate across all groups. This is the first 499 investigation to make comparisons among CF practitioners based on their competitive rank and 500 relative to resistance- and cardiovascular-trained, active adults. 501 Most competitive CF workouts require athletes to perform 2 or more exercises in a circuit 502 or listwise fashion for several repetitions and rounds, and to do so as quickly as possible or to 503 complete as much work as possible within a given time limit (1, 2, 50). Athletes who can 504 maintain a faster pace or rapidly recover between minimal rest periods would appear to be best 505 positioned to excel in this sport. A recent study in advanced CF athletes, as determined by their 506 performance in a common benchmark workout (i.e., "Fran"), supports this idea (51). Feito et al. 507 (2018) found that the best predictor of repetitions completed during a 15-minute CF workout was 508 the amount of work the athletes could perform on the final trial of four maximal Wingate sprints 509 separated by 90 seconds of rest. In the present study, the ADV group possessed a lower

510 percentage of body fat and greater non-bone fat-free mass compared to the REC and CON

511 groups. In sports, possessing an ideal ratio of skeletal muscle to fat mass may offer a competitive

512 advantage by improving efficiency, thermoregulation, and the ability to sustain effort (52). Aside

513 from their historical success in CF competition, the ADV group's performance during aerobic

and anaerobic testing provide evidence of this ability. ADV participants possessed a higher

515 VO₂peak than the other groups, which would imply that they were able to perform aerobic work 516 throughout a greater range of workloads (53, 54) but it does not completely explain their ability 517 to sustain effort at higher intensities (55). As the oxygen requirements of a workload exceed an 518 athlete's capacity to efficiently deliver oxygen, the ability to sustain effort may be further 519 explained by measures of anerobic performance and specific threshold points indicative of the 520 onset of fatigue (i.e., GET, RCT, and CP) (47, 55, 56). Participants in the ADV group were also 521 found to possess a higher GET, RCT, and CP, which are strongly correlated with each other CP 522 (56) and are thought (specifically RCT and CP) to demarcate the point in which exercise 523 transitions from 'heavy' to 'severe' (56, 57). Together, these data suggest that the ADV athletes 524 in this study had a greater capacity to produce energy aerobically, and that they were better 525 equipped to maintain efforts at higher absolute workloads and thus, be successful in their sport. 526 Skeletal mass and the morphological characteristics of muscle are suggestive of a greater 527 ability to produce force (58-61). That is, the size, architecture and quality of skeletal muscle 528 reflect the capability of activated muscle to produce force, whereas bone mass provides the 529 structural support and stability needed to effectively translate force production into human 530 movement. In the present study, ADV athletes possessed greater bone and muscle mass/size, 531 larger pennation angles, shorter fascicles, and better quality in the arm and quadriceps 532 musculature compared to the other groups. However, these only partially translated to greater 533 force production by ADV group participants during the IMTP test. IMTP performance was 534 highly variable until 0 to 200 - 250 ms, upon which ADV clearly produced greater force and at a 535 faster rate. The lack of uniformity across all strength measures might be explained by testing 536 specificity and the skillset of our sample. The importance of being able to rapidly activate muscle 537 (i.e., higher RFD) and the magnitude of IMTP force production varies across sports and athletic

538 activities. In weightlifters, significant relationships have been reported between one-repetition 539 maximums in the Olympic lifts and IMTP force (peak and from 0 to 100 - 250 ms) (62) but 540 relationships to RFD have either been limited to specific time bands (from 0 to 200 - 250 ms) 541 (62) or remain unclear in other athletes (63, 64). Although maximal strength in the Olympic and 542 power lifts can distinguish competitive ranking in CF athletes (8, 10), it is not a common 543 requisite of CF competition to maximally perform these lifts. Rather, most competitive workouts 544 either utilize submaximal loads that are performed for several repetitions or they require the 545 athlete to perform maximal (or near maximal) lifts after a fatiguing task (i.e., not a true measure of maximal strength) (50). It is also possible that the composition of the ADV group may help 546 547 explain the variability observed prior to 200 ms. While all ADV group participants ranked higher 548 than REC in the Open, their participation in later rounds of the Games competition had primarily 549 occurred as part of a team. Within this capacity, team members may be included based on their 550 skill set (e.g., strong/powerful athletes, gymnastically-skilled athletes, endurance athletes) to 551 minimize team weaknesses. This differs from individual competitors who must be proficient in a 552 broader set of skills to be competitive (8, 10). Currently, evidence documenting the physiological 553 differences between high-ranking individual and team competitors does not exist. 554 There is little evidence to suggest that consistent alterations will occur to resting 555 concentrations in T, C, or IGF-1 as a result of chronic training (14). Rather, their concentrations 556 generally reflect the current status of muscle tissue in response to the demands of training. An 557 overreaching period, marked by elevated training intensity or volume, might elicit transient 558 elevations in T and IGF-1 that typically return to baseline once training returns to 'normal' while

prolonged overreaching (or overstress) periods may elicit elevations in C (14). CF training is

560 characterized by an effort to maximize training density (i.e., complete a set amount of work as

561 quickly as possible, or maximize work completed within given time frame) within an unplanned 562 (i.e., non-periodized) training structure to promote general physical preparedness (1, 2). Further, 563 the 5-week Open is the most common avenue used by athletes to qualify for the Games (4, 5). 564 Prior to an important competitive event, athletes may elevate training intensity to promote peak 565 performance (65). Thus, the combination of the CF training strategy and the approach of an 566 important, extended competitive event could increase the likelihood of a prolonged period of 567 overstress. The occurrence of which might be identified by changes in resting hormonal concentrations, resting metabolic rate, performance, as well as a variety of other factors (14, 66, 568 569 67). However, the present investigation did not reveal any evidence of prolonged stress or 570 negative adaptations to training. Resting hormone concentrations and metabolic rates were 571 similar between groups and the physiological advantages demonstrated by the ADV group 572 appeared to reflect their reported training habits over the past six months (via medical and 573 physical activity history questionnaire). Excluding the conditioning component typically present 574 in CF workouts, members from each group reported using a similar number of sets per muscle 575 group (3-6), repetitions (3-12), and rest intervals (60-90 seconds) during the strength 576 component of their workouts. Only training frequency was reported to be different with the ADV 577 group utilizing a form of resistance exercise on approximately 5.3 days per week whereas the 578 REC and CON groups averaged 4.6 days per week and 3.7 days per week, respectively. 579 Although the greater training frequency seen in ADV would have theoretically provided more of 580 an opportunity to accumulate training volume and promote adaptations, it could have also 581 interfered with their recovery. Nevertheless, ADV possessed a more favorable body composition 582 and generally outperformed the other groups in each performance measure. Therefore, as of one-583 month prior to competition, adequate recovery appeared to be present in this group. Likewise,

the lack of differences seen between REC and CON, who were not actively training for the Open, also provides evidence of adequate recovery. Future investigations can expand on this by monitoring performance surrounding the extended Open competition.

587 The findings of this study suggest that advanced CF athletes possess a more favorable 588 body composition, greater bone and muscle mass, greater muscle quality and strength, greater 589 aerobic capacity, and a greater ability to sustain effort than recreational CF participants and 590 physically-active adults. The reasons for these differences remain unclear due to the cross-591 sectional design of this study but may be related to differences in training experience and recent 592 training habits. Although all participants in this study could be considered well-trained (68), 593 ADV group participants reported having more resistance training experience and having been 594 training more frequently over the past 6 months than the other groups. It is possible that their 595 advantages are simply the result of training for a longer amount of time or creating more 596 opportunities to increase their volume load throughout the week. Without documentation (i.e., 597 extensive, detailed training logs), however, it is only possible to speculate upon their potential 598 influence as unknown factors (e.g., training quality, genetic predisposition) would certainly 599 modulate resultant adaptations. Further, the influence of daily variations in the conditioning 600 components of CF workouts on effort and volume load, as well as how these might compare to 601 traditional aerobic exercise (utilized by CON), remains unclear. It is interesting to note that 602 despite the apparent differences in each training strategy (i.e., CF conditioning and traditional 603 aerobic exercise), no differences were seen between REC and CON. To be included in this study, 604 both had to have been regularly participating in their chosen training strategy on 3-5 days per 605 week for at least the past year. Nevertheless, REC and CON were found to possess similar 606 physiological characteristics. Future longitudinal investigations that document both the quality

- and quantity of these training forms may help to provide insight into whether an advantage exists
- 608 between these strategies or if they promote comparable adaptations among recreationally-active
- adults.

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