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Article

Metabolic Profile and Body Composition in Twins Concordant and Discordant for Physical Exercise

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Abstract

The aim of this study was to evaluate the differences in the metabolic profile and body composition of monozygotic (MZ) twins concordant and discordant for the practice of physical exercise. The sample consisted of 92 MZ twins (72.5% female and 27.5% male, mean age 25.4 ± 5.69 years), registered with the Brazilian Registry of Twins, residing in Natal, Brazil. Data collection was carried out between the years 2016 and 2018. On day 1, subjects underwent a whole-body fitness evaluation, including measures of weight, height, body composition by Dual-Energy X-ray Absorptiometry and the Cardiorespiratory Exercise Test. On day 2, 10 ml blood samples were collected (overnight fasting) to determine the lipid profile and fasting glucose. The sample was separated into three groups: Active Concordant twins (Concordant A, $n = 44$ subjects), Inactive Concordant twins (Concordant I, $n = 22$ subjects) and Discordant pairs for Physical Exercise (Discordant PE, $n = 26$ subjects). The results demonstrated a difference between the discordant twins for exercise and also between the active versus sedentary groups, indicating a causal effect of exercise on the fat percentage, maximum oxygen consumption (VO_2max) and second ventilatory threshold variables. Between groups, a difference was also observed between the groups in ventilatory threshold, very low-density lipoprotein and triglycerides. We concluded that, regardless of genetics, the practice of physical exercise was sufficient to generate alterations in body composition and VO_2max in MZ twins, but not in the lipid profile or fasting glucose.

Keywords: lipid profile; twins; fasting glucose; maximum oxygen consumption; physical activity

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An active lifestyle promotes improvements in physical fitness and health (Ekelund et al., 2012; Jermendy et al., 2011). Modification of these variables is among the goals of most physical exercise practitioners (Patel & Abate, 2013; Silva et al., 2012). However, improvement and relief of symptoms in response to exercise vary substantially between individuals, and this interindividual variability is supported, in part, by genotypic differences (Dias et al., 2011). This makes studies with twins an important tool, since each monozygotic (MZ) pair comes from a single zygote and the twins present genetically identical elements (Neale & Maes, 1992), reducing genotypic differences in phenotypic responses and making it possible to identify variations between individuals caused by physical exercise.

Cardiorespiratory fitness is seen as a variable with significant genetic effects (50%–67%; Bouchard et al., 1999; Miyamoto-Mikami et al., 2018); however, it is also influenced by factors such as age, sex and level of physical activity (Queiroga et al., 2013). There is evidence that the best cardiorespiratory fitness caused by physical exercise exhibits a dose-response relationship, implying improvements in the lipid and glycemic profiles and body composition, as well as generating general health benefits (Dias et al., 2011; McKinney et al., 2016; Oliveira et al., 2014). Queiroga et al. (2013) demonstrated that MZ twins discordant for cardiorespiratory fitness presented differences in glycemic metabolism, regardless of genetic factors. However, despite the significant number of studies that have analyzed these factors, few studies have used twins discordant for physical exercise and evaluated the effects on metabolic risk factors (Queiroga et al., 2013).

Therefore, the aim of the current study is to investigate differences in the metabolic profile and body composition of adult MZ twins who are concordant and discordant for physical exercise.

Methods

Sample

The nonprobabilistic sample was obtained from twins registered at the I Twin Festival of Rio Grande do Norte, held in 2016 at the Federal University of Rio Grande do Norte, in partnership with the Brazilian Twin Registry (Ferreira et al., 2016). Individuals with physical disabilities or musculoskeletal limitations that prevented walking or running, subjects on obesity-related drug treatment, twins of different sexes and individuals with positive responses to the Physical Activity Readiness Questionnaire (PAR-Q; Warburton et al., 2011) were excluded from the study.

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Given the exclusion criteria, a sample of 92 MZ twins was used (72.5% female and 27.5% male, mean age 25.4 ± 5.69 years).

After signing the Informed Consent Form, an anamnesis questionnaire was applied to analyze health status and pre-existing diseases, and the PAR-Q to analyze readiness to perform physical activities. To determine the practice of physical exercise, the pairs of twins answered a previously prepared questionnaire with information on physical exercise. Twins with more than 1 month of regular practice of any sport or recreational modality (e.g., weight training, running, cycling and working out in gyms) were considered active. Twins considered inactive did not exercise at all or exercised with a frequency below 2 days a week. From this information, the sample was separated into three groups: Active Concordant twins (Concordant A) where both siblings are active; Inactive Concordant twins (Concordant I) where both siblings are sedentary and Discordant Pairs for Physical Exercise (Discordant PE) where one sibling is active and the other sedentary. Zygosity in twins was determined using a validated zygosity questionnaire with 93.3% accuracy (Ooki & Asaka, 2004).

Physical Assessment

The subjects were instructed not to drink caffeine or alcohol-based drinks on the day of the tests, to consume a light meal 2 h before the test and to avoid vigorous physical efforts the day before the tests. All tests were applied in the laboratory at a temperature maintained between 20°C and 25°C.

On day 1, subjects underwent a whole-body fitness evaluation, including measures of weight, height and body composition, using Dual-Energy X-ray Absorptiometry (Marfell-Jones et al., 2006), through which we evaluated the percentage of android fat (% AF), percentage of gynoid fat (% GF) and percentage of total fat (% TF).

A Cardiorespiratory Exercise Test was also applied, adopting a ramp protocol with an initial speed of 3 km/h for sedentary individuals and 4 km/h for physically active subjects and 0% incline. Speed and incline were gradually increased during the test up to the subject's voluntary resignation. All tests lasted between 8 and 12 min (Guazzi et al., 2016).

Ventilatory variables were measured using a metabolic gas analyzer (model Metalyzer 3B, MICROMED®), using the breath-by-breath method with a 10-s window for later analysis using the Metasoft program. The Cortex unit was calibrated by the closed-loop method, using calibration gas (original 16% O_2 and 5% CO_2 cylinder, supplied by the manufacturer), which allowed a new calibration before each test. The characterization of the test as maximum effort was adopted from the acceptance of, at least, three of the following criteria: (a) voluntary exhaustion; (b) maximum heart rate (HR) reached at least 90% of that predicted for age (220 age); (c) respiratory exchange ratio >1.1; (d) maximum oxygen consumption, observed by the plateau concept, when the VO_2 stabilizes without a difference of ≤150 ml/kg per min between the values of the last 30 s of the test, or the VO_2 peak was considered maximum when the plateau did not occur (Nelson & Asplund, 2016; Thompson et al., 2013). The ventilatory thresholds of each patient were determined using the V-slope method (Binder et al., 2008).

On day 2, 10 ml blood samples were collected (overnight fasting) to determine the serum fasting glucose (GLU), total cholesterol (COL), high-density lipoproteins (HDL), very low-density lipoprotein (VLDL), low-density lipoprotein (LDL) and triglycerides (TG). All dosages were performed using enzymatic-colorimetric assays and commercial kits (Labtest Diagnóstica-SA®) in Bio 2000 equipment (Bioplus®.Barueri/SP). The concentrations of LDL and VLDL cholesterol were obtained by applying the Friedewald formula (Friedewald et al., 1972).

Statistical Analysis

In the Concordant A and Concordant I groups, the intrapair mean within each group was calculated and the difference between the groups was verified by the analysis of variance test ($p < .001$). In the discordant PA group, the Wilcoxon test ($p < .05$) was applied for intrapair analysis, that is, comparing the active twin versus the inactive twin.

To verify data reliability, Spearman's intrapair correlation of the MZ twins was also performed, separated into concordant and discordant. We used the Cohen parameter (Cohen, 1992), assuming $r = .10-.29$ (weak correlation), $r = .30-049$ (moderate correlation) and $r = .50-1$ (strong correlation).

Cardiorespiratory fitness can be classified into levels, as indicated by the American College of Sports Medicine (ACSM; Thompson et al., 2013). In the present study, we classified the sample according to the categories poor, regular and good cardiorespiratory fitness (Thompson et al., 2013), after which the chi-square test and Spearman's correlation were applied between the categorical variables, level of cardiorespiratory fitness and physical exercise (active and sedentary). All analysis was conducted using the statistical program SPSS®, version 20, and the normality and homoscedasticity of the data distribution were verified by the Shapiro–Wilk test.

Results

The results are shown in Table 1, separated by groups (Concordant A, Concordant I and Discordant PE). Intrapair differences were observed in discordant twins (Active twin vs. Inactive twin) and between groups (Concordant A vs. Concordant I) for the variables body composition and metabolic profile, including maximum oxygen consumption (VO_2max), oxygen consumption corresponding to the first and second ventilatory thresholds, lipid profile and fasting blood glucose (see Table 2).

An association was found between the level of cardiorespiratory fitness and the practice of physical activity (see Table 3). Of the twins who were active, 51.1% presented regular or good cardiorespiratory fitness and 37% regular or poor cardiorespiratory fitness. Of those who were sedentary, 35.8% presented poor or regular fitness.

Discussion

In our findings, the practice of physical exercise was shown to be an important component of interindividual differences for the percentages of body fat (%AF, % GF and % TF), demonstrated by the intrapair differences in the discordant twins for physical exercise and by the difference between the active twin group and the inactive twin group, indicating a causal effect of the practice of physical exercise. Thus, we can infer that genetics does not guarantee equivalence of MZ twins with respect to percentage of body fat as a consequence of physical exercise. Previous studies indicate heritability of body fat measures with significant genetic effects (58%–86% of heritability; Hopkins et al., 2010; Mustelin et al., 2011; Oliveira et al., 2014). However, the current study demonstrates that physical exercise is able to generate changes regardless of genetics. This occurs because the practice of physical activities is

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Table 1. Differences in body composition and metabolic variables in pairs of monozygotic twins concordant and discordant for physical activity expressed as mean ± standard deviation

	Concordant A ($n = 44$ subjects)	Concordant I ($n = 22$ subjects)		Discordant PE ($n = 13$ pairs)		
	Active twins group	Inactive twins group	p**	Active twin	Inactive twin	p*
Age	25.9 ± 6.3	25.6 ± 4.8		23.8 ± 4.1		
Body composition						
Weight (kg)	62.9 ± 12.1	70.9 ± 24.7	.08	69.1 ± 13.5	70.9 ± 19.5	.35
% AF	24.7 ± 9.1	39.2 ± 11.7	.001	31.8 ± 10.7	35.7 ± 10.1	.03
% GF	32.3 ± 7.9	43.9 ± 6.9	.001	33.7 ± 9.6	36.9 ± 8.2	.01
% TF	28.3 ± 7.1	39.1 ± 8.3	.001	30.5 ± 7.9	33.5 ± 6.9	.02
Cardiorespiratory variables						
VO_2L1	20.9 ± 5.5	15.9 ± 2.9	.001	18.9 ± 2.9	18.2 ± 3.8	.54
VO_2L2	34.8 ± 6.8	25.0 ± 5.3	.001	33.7 ± 7.6	28.9 ± 5.5	.01
VO_2Max	36.9 ± 6.6	27.2 ± 6.2	.001	37.0 ± 7.4	30.9 ± 4.9	.001
Metabolic variables						
COL	167.6 ± 33.9	167.3 ± 48.0	.98	156.1 ± 44.4	163.2 ± 41.8	.08
HDL	49.4 ± 22.8	48.1 ± 14.6	.81	38.6 ± 14.1	38.2 ± 13.2	.86
VLDL	14.2 ± 6.8	21.4 ± 13.5	.01*	17.6 ± 7.8	24.8 ± 18.4	.07
LDL	104.0 ± 34.6	97.8 ± 42.3	.55	99.9 ± 41.0	100.2 ± 34.6	.60
TG	73.4 ± 35.4	102.7 ±70.5	.03*	87.9 ± 39.2	123.9 ± 91.9	.07
GLU	85.4 ± 8.9	86.5 ± 11.6	.69	90.2 ± 10.5	95.1 ± 23.9	.69

Concordant A, Active Concordant twins; Concordant I, Inactive Concordant twins; Discordant PE, Discordant pairs for physical exercise; % AF, percentage of android fat; % GF, percentage of gynoid fat; % TF, percentage of total fat; VO_2L1, Maximum oxygen consumption corresponding to the first ventilatory threshold (ml/kg per min); VO_2L2, maximum oxygen consumption corresponding to the second ventilatory threshold (ml/kg per min); VO_2Max, maximum oxygen consumption (ml/kg per min); COL, total cholesterol; HDL, high-density lipoproteins; VLDL, very low-density lipoprotein; LDL, low-density lipoprotein; TG, triglycerides; GLU, fasting glucose.
**$p \leq .001$; *$p \leq .05$.

able to modify the action of the genes responsible for the predisposition to obesity (Graff et al., 2011).

Thus, it is important that physical activity is encouraged in the population to facilitate alterations in the percentage of body fat, as well as to improve other aspects of physical health. Since the distribution of body fat is related to the risk of developing cardiovascular diseases (Despres et al., 2008), as well as dyslipidemia and insulin resistance, among other comorbidities that are associated with elevated body fat (Berntzen et al., 2018; Ji et al., 2014; Lundbom et al., 2016; Rottensteiner et al., 2016). Studies with pairs of MZ twins discordant for physical activity (Bathgate et al., 2018; Rottensteiner et al., 2016) found that the inactive twins presented markedly higher percentages of body fat and fat mass than their active twins. In addition, a low level of physical activity is associated with high BMI and fat percentage (Silventoinen et al., 2009). Thus, individuals with a higher genetic risk for obesity benefit from increased physical activity (Hasselbalch et al., 2010; Segal et al., 2009; Silventoinen et al., 2009; Waller, 2010a).

The VO_2 at the ventilatory threshold (threshold 1) did not demonstrate any difference in the discordant PA; that is, the practice of physical exercise was not enough to generate intrapair differences at this exercise intensity. This behavior suggests a possible effect of heritability on this variable, as indicated in previous studies (Feitosa et al., 2002; Gaskill et al., 2001), where the ventilatory threshold was shown to have a heritability of 54% and 58% for Black and White sedentary individuals, respectively, adjusted for age and body mass; however, this genetic influence does not seem to influence the training of the ventilatory threshold.

At the anaerobic threshold (threshold 2), there was a difference both in the concordant and in the discordant PA groups; that is, the practice of physical exercise generated intrapair differences in individuals genetically equal at this exercise intensity. Threshold 2 is an anaerobic phase of effort and is modified by interventions. In addition, other factors can influence this component, such as the type of physical activity, intensity, duration of effort and types of muscle fibers (Binder et al., 2008; Guazzi et al., 2016; Thompson et al., 2013); however, these variables were not controlled in the current study.

The VO_2max presented differences in all groups, suggesting that regardless of genetics, this variable can be modified by the practice of physical exercise. Further, as physical inactivity also promotes changes, generating loss in cardiorespiratory fitness, having 'privileged' genetics does not guarantee good VO_2max levels. Physical inactivity, in addition to all the health hazards (Waller, 2010b), will not maintain inherited levels of cardiorespiratory fitness. Therefore, it is essential to stimulate physical activity so that the aerobic component is also developed, as good levels of aerobic conditioning are associated with a reduction of chronic diseases, in addition to promoting healthy cognitive and psychosocial function (McKinney et al., 2016).

Cardiorespiratory fitness demonstrated a significant association with the practice of physical exercise ($p = .001$), confirming the development of the aerobic component. In our data, of the twins who were active, 37% presented with regular or weak cardiorespiratory fitness, indicating that they were insufficiently active to perform physical activities, with little use of the aerobic component to generate the necessary gains for good cardiorespiratory fitness.

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Table 2. Intrapair correlations of monozygotic twins for body composition and metabolic profile variables, separated into concordant and discordant, for physical exercise

	Concordant (66 subjects)		Discordant (26 subjects)	
	$r_{(s)}$	*P*	$r_{(s)}$	*p*
Body composition				
Weight (kg)	.94	.001	.97	.001
% AF	.79	.001	.84	.001
% GF	.92	.001	.98	.001
% TF	.88	.001	.77	.002
Cardiorespiratory variables				
VO_2L1	.48	.005	.17	.57
VO_2L2	.80	.001	.66	.01
VO_2Max	.91	.001	.79	.001
Metabolic variables				
COL	.82	.001	.89	.001
HDL	.77	.001	.91	.001
VLDL	.71	.001	.86	.001
LDL	.73	.001	.87	.001
TG	.75	.001	.86	.001
GLU	.72	.001	.76	.003

% AF, percentage of android fat; % GF, percentage of gynoid fat; % TF, percentage of total fat; VO_2L1, maximum oxygen consumption corresponding to the first ventilatory threshold (ml/kg per min); VO_2L2, maximum oxygen consumption corresponding to the second ventilatory threshold (ml/kg per min); VO_2Max, maximum oxygen consumption (ml/kg per min); COL, total cholesterol; HDL, high-density lipoproteins; VLDL, very low-density lipoprotein; LDL, low-density lipoprotein; TG, triglycerides; GLU, fasting glucose.
Note: $r_{(s)}$, Spearman correlation; $r = .10–.29$ (weak correlation); $r = .30–.49$ (moderate correlation); $r = .50–1$ (strong correlation). $p \leq .001$.

Table 3. Association between cardiorespiratory fitness level and physical exercise in monozygotic twins

	Cardiorespiratory fitness level				
	Poor (%)	Regular (%)	Good (%)	Total (%)	*p* Value
Active	8.70	28.30	22.80	59.80	.001*
Sedentary	21.70	14.10	4.40	40.20	
Total	30.40	42.40	27.20	100.00	

Note: *Spearman's correlation ($p < .05$); $N = 92$ monozygotic twins.

The ACSM (Thompson et al., 2013) argues that healthy adults should achieve an energy expenditure equivalent to 150 min/week of moderate intensity aerobic activity; 75 min/week of vigorous intensity aerobic activity; or a combination of the two, to obtain substantial health benefits. These guidelines indicate a cause-effect relationship, detailing that additional health benefits are obtained with 300 min/week or more of moderate intensity aerobic activity.

In the present study, the practice of physical exercise was not enough to generate intrapair changes in the lipid profile and fasting glucose, demonstrated in the discordant PA pairs, suggesting that the components inherited or shared between the twins may influence the behavior of the lipid profile and fasting blood glucose. Corroborating these results, Marson et al. (2016) observed that physical training in general was not associated with a reduction in fasting blood glucose levels, but rather was associated with reductions in fasting insulin levels, a variable not analyzed in the current study. In addition, other lifestyle factors (e.g., age, sex, diet, smoking, drinking and obesity; Bogl et al., 2019) are responsible for modifying these variables.

However, concordant A versus concordant I groups presented differences in VLDL and TG, with lower values in the active group, suggesting that physical exercise has an effect on these variables. This behavior was also demonstrated in the study by Bathgate et al. (2018), where the trained MZ twin presented lower COL, LDL, TG and plasma glucose levels when compared to the untrained MZ twin. Therefore, an active lifestyle and healthy habits may be able to modify the action of the genes responsible for the predisposition to obesity and cardiovascular risk (Graff et al., 2011).

Despite the indications of modifications in the lipid profile and fasting glucose due to exercise (McKinney et al., 2016), the variables COL, HDL, LDL and GLI did not present differences in any group analyzed. As the current work deals with genetically identical individuals, we can infer that heritability or shared genetic factors have an influence on these variables and it is not possible to observe the effect of exercise on them. Corroborating these data, the serum levels of GLU, COL, HDL cholesterol and LDL cholesterol are strongly influenced by genetic factors with 55%–64% of heritability, even after adjustment for sex and age (Borges et al., 2018; Chen et al., 2004; Jedrusik et al., 2003).

The metabolic and body composition differences in the MZ twins from physical exercise, presented in this study, prove that physical exercise stands out from the heritability of these components through positive changes in the cardiorespiratory component and in the percentage of body fat. These alterations can occur in different proportions, which corroborates biological individuality (Thompson et al., 2013), and confirm that the response to exercise can vary substantially between individuals. Therefore, physical training for benefits of body composition and gains in cardiorespiratory fitness needs to be individualized, even in twins, as the environment generates different responses in genetically similar people.

We conclude that based on the practice of physical exercise, MZ twins present differences in % AF, % GF and % TF, as well as in VO_2max and in the second ventilatory threshold. However, the practice of physical exercise was not enough to generate alterations in the lipid profile or fasting glucose, regardless of genetics.

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Conflict of interest. None.

Ethical standards. The study was approved by the Research Ethics Committee of the Onofre Lopes University Hospital. Natal/RN – Brazil, CEP/HUOL, CAAE 35573214.1.0000.5292, according to Resolution 466/12 of the National Health Council.

References

Bathgate, K. E., Bagley, J. R., Jo, E., Talmadge, R. J., Tobias, I. S., Brown, L. E., . . . Galpin, A. J. (2018). Muscle health and performance in monozygotic twins with 30 years of discordant exercise habits. *European Journal of Applied Physiology*, *118*, 2097–2110.

Berntzen, B., Jukarainen, S., Kataja, M., Hakkarainen, A., Lundbom, J., Lundbom, N., . . . Pietiläinen, K. H. (2018). Physical activity, cardiorespiratory fitness, and metabolic outcomes in monozygotic twin pairs discordant for body mass index. *Scandinavian Journal of Medicine & Science in Sports*, *28*, 1048–1055.

Binder, R. K., Wonisch, M., Corra, U., Cohen-Solal, A., Vanhees, L., Saner, H., & Schmid, J.-P. (2008). Methodological approach to the first and second lactate threshold in incremental cardiopulmonary exercise testing. *European Journal of Cardiovascular Prevention & Rehabilitation*, *15*, 726–734.

Bogl, L. H., Mehlig, K., Intemann, T., Masip, G., Keski-Rahkonen, A., Russo, P., . . . Johnson, L. (2019). A within-sibling pair analysis of lifestyle behaviours and BMI z-score in the multi-centre I. Family study. *Nutrition, Metabolism and Cardiovascular Diseases*, *29*, 580–589.

Borges, M. V. d. O., de Sousa, E. C., Alonso, L., Juvanhol Lopes, L., Medeiros, V., Maciel, R., . . . Moreira, P. (2018). Heritability of biochemical and anthropometric parameter markers of cardiovascular risk: A study with twins. *Sports Medicine Journal/Medicina Sportivâ*, *14*, 2954–2960.

Bouchard, C., An, P., Rice, T., Skinner, J. S., Wilmore, J. H., Gagnon, J., . . . Rao, D. (1999). Familial aggregation of VO(2 max) response to exercise training: Results from the HERITAGE Family Study. *Journal of Applied Physiology*, *87*, 1003–1008.

Chen, T., Ji, C., Pang, Z., Wang, S., Hu, Y., & Qin, Y. (2004). Heritability of serum lipids and lipoproteins and its related factors in twins aged 5 to 19 years of China]. *Zhonghua yu fang yi xue za zhi [Chinese Journal of Preventive Medicine]*, *38*, 237.

Cohen, J. (1992). Statistical power analysis. *Current Directions in Psychological Science*, *1*, 98–101.

Despres, J., Lemieux, I., Bergeron, J., Pibarot, P., Mathieu, P., Larose, E., . . . Poirier, P. (2008). Abdominal obesity and the metabolic syndrome: Contribution to global cardiometabolic risk. *Arteriosclerosis, Thrombosis, and Vascular Biology*, *28*, 1039–1049.

Dias, R. G., Gowdak, M. M., & Pereira, A. C. (2011). Genetics and cardiovascular system: Influence of human genetic variants on vascular function. *Genes & Nutrition*, *6*, 55–62.

Ekelund, U., Luan, J. A., Sherar, L. B., Esliger, D. W., Griew, P., & Cooper, A. (2012). Moderate to vigorous physical activity and sedentary time and cardiometabolic risk factors in children and adolescents. *JAMA*, *307*, 704–712.

Feitosa, M. F., Gaskill, S. E., Rice, T., Rankinen, T., Bouchard, C., Rao, D., . . . Leon, A. S. (2002). Major gene effects on exercise ventilatory threshold: The HERITAGE Family Study. *Journal of Applied Physiology*, *93*, 1000–1006.

Ferreira, P. H., Oliveira, V. C., Junqueira, D. R., Cisneros, L. C., Ferreira, L. C., Murphy, K., . . . Teixeira-Salmela, L. F. (2016). The Brazilian twin registry. *Twin Research and Human Genetics*, *19*, 687–691.

Friedewald, W. T., Levy, R. I., & Fredrickson, D. S. (1972). Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clinical Chemistry*, *18*, 499–502.

Gaskill, S. E., Rice, T., Bouchard, C., Gagnon, J., Rao, D., Skinner, J. S., . . . Leon, A. S. (2001). Familial resemblance in ventilatory threshold: The HERITAGE Family Study. *Medicine and Science in Sports and Exercise*, *33*, 1832–1840.

Graff, M., North, K. E., Monda, K. L., Lange, E. M., Lange, L. A., Guo, G., & Gordon-Larsen, P. (2011). The combined influence of genetic factors and sedentary activity on body mass changes from adolescence to young adulthood: The National Longitudinal Adolescent Health Study. *Diabetes/Metabolism Research and Reviews*, *27*, 63–69.

Guazzi, M., Arena, R., Halle, M., Piepoli, M. F., Myers, J., & Lavie, C. J. (2016). 2016 focused update: Clinical recommendations for cardiopulmonary exercise testing data assessment in specific patient populations. *European Heart Journal*, *39*, 1144–1161.

Hasselbalch, A. L., Heitmann, B. L., Kyvik, K. O., & Sorensen, T. I. (2010). Associations between dietary intake and body fat independent of genetic and familial environmental background. *International Journal of Obesity*, *34*, 892–898.

Hopkins, N., Stratton, G., Maia, J., Tinken, T. M., Graves, L. E., Cable, T. N., & Green, D. J. (2010). Heritability of arterial function, fitness, and physical activity in youth: A study of monozygotic and dizygotic twins. *Journal of Pediatrics*, *157*, 943–948.

Jedrusik, P., Januszewicz, A., Busjahn, A., Zawadzki, B., Wocial, B., Ignatowska-Switalska, H., . . . Januszewicz, W. (2003). Genetic influence on blood pressure and lipid parameters in a sample of Polish twins. *Blood Pressure*, *12*, 7–11.

Jermendy, G., Horvath, T., Littvay, L., Steinbach, R., Jermendy, A., Tarnoki, A., . . . Osztovits, J. (2011). Effect of genetic and environmental influences on cardiometabolic risk factors: A twin study. *Cardiovascular Diabetology*, *10*, Article no. 96.

Ji, F., Ning, F., Duan, H., Kaprio, J., Zhang, D., Wang, S., . . . Silventoinen, K. (2014). Genetic and environmental influences on cardiovascular disease risk factors: A study of Chinese twin children and adolescents. *Twin Research and Human Genetics*, *17*, 72–79.

Lundbom, J., Bierwagen, A., Bodis, K., Szendrödi, J., Kaprio, J., Rissanen, A., . . . Pietiläinen, K. H. (2016). Deep subcutaneous adipose tissue lipid unsaturation associates with intramyocellular lipid content. *Metabolism*, *65*, 1230–1237.

Marfell-Jones, M., Olds, T., Stewart, A., & Carter, L. (2006). *International Standards for Anthropometric Assessment*. International Society for the Advancement of Kinanthropometry.

Marson, E. C., Delevatti, R. S., Prado, A. K. G., Netto, N., & Kruel, L. F. M. (2016). Effects of aerobic, resistance, and combined exercise training on insulin resistance markers in overweight or obese children and adolescents: A systematic review and meta-analysis. *Preventive Medicine*, *93*, 211–218.

McKinney, J., Lithwick, D. J., Morrison, B. N., Nazzari, H., Isserow, S. H., Heilbron, B., & Krahn, A. D. (2016). The health benefits of physical activity and cardiorespiratory fitness. *British Columbia Medical Journal*, *58*, 131–137.

Miyamoto-Mikami, E., Zempo, H., Fuku, N., Kikuchi, N., Miyachi, M., & Murakami, H. (2018). Heritability estimates of endurance-related phenotypes: A systematic review and meta-analysis. *Scandinavian Journal of Medicine & Science in Sports*, *28*, 834–845.

Mustelin, L., Latvala, A., Pietiläinen, K. H., Piirilã, P., Sovijãrvi, A. R., Kujala, U. M., . . . Kaprio, J. (2011). Associations between sports participation, cardiorespiratory fitness, and adiposity in young adult twins. *Journal of Applied Physiology*, *110*, 681–686.

Neale, M. C., & Maes, H. H. M. (1992). *Methodology for genetic studies of twins and families*. Kluwer Academic Publishers B.V.

Nelson, N., & Asplund, C. A. (2016). Exercise testing: Who, when, and why? *PM&R*, *8*, S16–S23.

Oliveira, M. V., Sousa, E. C., Cabral, B. G. A., Sánchez, D. S., Alonso, L. V. S., Dantas, P. M. S., & Lemos, T. M. A. M. (2014). Heredabilidad de los indicadores antropométricos relacionados con obesidad en gemelos de ambos sexos entre 8 a 26 años de Brasil [Heritability of anthropometric indicators related to obesity in twins of both sexes between 8 to 26 years of age in Brazil]. *Archivos de Medicina del Deporte*, *31*, 14–23.

Ooki, S., & Asaka, A. (2004). Zygosity diagnosis in young twins by questionnaire for twins' mothers and twins' self-reports. *Twin Research and Human Genetics*, *7*, 5–12.

Patel, P., & Abate, N. (2013). Body fat distribution and insulin resistance. *Nutrients*, *5*, 2019–2027.

Queiroga, M. R., Barbieri, R. A., Ferreira, S. A., Luchessi, A. D., Silbiger, V. N., Hirata, R. D. C., . . . Kokubun, E. (2013). Metabolismo de glicose em gêmeos monozigóticos discordantes para aptidão cardiorrespiratória [Glucose metabolism in discordant monozygotic twins for cardiorespiratory fitness]. *Revista Paulista de Pediatria*, *31*, 77–82.

Rottensteiner, M., Leskinen, T., Järvelä-Reijonen, E., Väisänen, K., Aaltonen, S., Kaprio, J., & Kujala, U. M. (2016). Leisure-time physical activity and intra-abdominal fat in young adulthood: A monozygotic co-twin control study. *Obesity*, *24*, 1185–1191.

https://doi.org/10.1017/thg.2020.58 Published online by Cambridge University Press

Sant'Anna, M., Priore, S., & Franceschini, S. (2009). Métodos de avaliação da composição corporal em crianças [Methods of body composition evaluation in children]. *Revista Paulista de Pediatria*, *27*, 315–321.

Segal, N. L., Feng, R., McGuire, S. A., Allison, D. B., & Miller, S. (2009). Genetic and environmental contributions to body mass index: Comparative analysis of monozygotic twins, dizygotic twins and same-age unrelated siblings. *International Journal of Obesity*, *33*, 37–41.

Silva, V. S., Petroski, E. L., Souza, I., & Silva, D. A. S. (2012). Prevalência e fatores associados ao excesso de peso em adultos do Brasil: um estudo de base populacional em todo território nacional [Prevalence and factors associated with overweight in adults in Brazil: A population-based study nationwide]. *Revista de Educação Física e Ciências do Esporte*, *34*, 713–726.

Silventoinen, K., Hasselbalch, A. L., Lallukka, T., Bogl, L., Pietilainen, K. H., Heitmann, B. L., … **Kaprio, J.** (2009). Modification effects of physical activity and protein intake on heritability of body size and composition. *American Journal of Clinical Nutrition*, *90*, 1096–1103.

Thompson, P. D., Arena, R., Riebe, D., & Pescatello, L. S. (2013). ACSM's new preparticipation health screening recommendations from ACSM's guidelines for exercise testing and prescription. *Current Sports Medicine Reports*, *12*, 215–217.

Waller, K. (2010a). Effect of physical activity on health in twins: A 30-yr longitudinal study. *Medicine & Science in Sports & Exercise*, *42*, 658–664.

Waller, K. (2010b). Physical activity, morbidity and mortality in twins: A 24-year prospective follow-up. *Eur J Epidemiol*, *25*(10), 731–739.

Warburton, D. E., Jamnik, V. K., Bredin, S. S., & Gledhill, N. (2011). The physical activity readiness questionnaire for everyone (PAR-Q+) and electronic physical activity readiness medical examination (ePARmed-X+). *The Health & Fitness Journal of Canada*, *4*, 3–17.

https://doi.org/10.1017/thg.2020.58 Published online by Cambridge University Press