The role of body fat in the relationship of cardiorespiratory fitness with cardiovascular risk factors in Brazilian children

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Abstract — Aims: To analyze the association between the percentage of body fat and cardiorespiratory fitness (CRF) with cardiovascular risk factors in children, and; to examine whether percentage of body fat acts as a mediator on the association between CRF and cardiovascular risk factor. Methods: This cross-sectional study included 128 children aged 7-11 years (Mean 8.54, SD: 1.42). The following variables were evaluated: the percentage of body fat, CRF, diastolic and systolic blood pressure, glucose, triglycerides (TG) and total cholesterol. For statistical analysis were performed Partial correlation and mediation analysis. All analysis was adjusted for sex, age and height. Results: CRF, the percentage of body fat, mean arterial pressure (MAP) and TG showed a correlation between each other. Percentage of body fat mediated the association between CRF and MAP (Indirect Effect= -0.008; IC: -0.0159 -0.0030), explaining 29% of this association. However, it was found that the percentage of body fat was not a mediator of the association between CRF and TG. Conclusion: The percentage of body fat mediates the association between CRF and MAP. Our findings show that the importance of a healthy body composition for the prevention of high blood pressure levels in childhood as well as the relevance of physical activity on these parameters.

Keywords: schoolers, health, arterial pressure, triglycerides

Introduction

The prevalence of cardiovascular disease (CVD) has been increasing all over the world¹. Although, the presence of risk factors for CVD in children and adolescents has more clearly been identified over the last 20 years². Initially, this kind of disease were considered a concern for the adult population, however studies have observed that many cardiometabolic abnormalities, including dyslipidemias, insulin resistance, and type 2 diabetes mellitus have its origin during childhood³. Thus, the early identification and intervention for children at risk of developing CVD would minimize the tracking of risk factors into adulthood⁴.

Cardiorespiratory fitness (CRF) has been considered an important health indicator⁵. Studies have shown that there is an association between low levels of cardiorespiratory fitness and CVD risk factors in children and adolescent⁶⁷. Furthermore, it has been suggested that moderate to high levels of CRF could attenuate some of the adverse metabolic consequences related to obesity⁸. Indeed, the general belief that normal weight is enough for being healthy could be wrong and it is now recognized that moderate to high levels of CRF plays an important role in health even for subject with body mass index (BMI) over 24.9 kg/m².

Therefore, studies have shown that both fatness and fitness are independently associated with cardiometabolic risk factors among youth. It has been reported that when adiposity is included in the models the magnitude of the association between fitness and cardiometabolic risk appears to be small to moderate⁹¹⁰. Furthermore, a mediation analysis revealed that BMI has a powerful influence on the relationship between CRF and metabolic syndrome in schoolchildren¹¹. The same kind of analysis indicated that BMI mediates the relationship between CRF and mean arterial pressure in Spanish schoolchildren¹².

Although BMI, an anthropometric index, is widely used in epidemiological studies, it is unable to distinguish fat and lean mass¹³. Also, most studies have explored BMI as an intermediate and confounding variable in the relationship between CRF and cardiovascular risk factor¹¹¹². Thus, considering percentage of body fat (%BF) could provide more precise information regarding adiposity influence on the association between CRF and CVD, the study aimed: 1) To analyze the association between percentage of body fat and CRF with cardiovascular risk factors in children; and 2) To examine whether percentage of body fat acts as a mediator on the association between CRF and cardiovascular risk factors.

Methods

Study design

This was a cross-sectional analysis using the baseline data from a longitudinal study that assessed the effects of a soccer intervention...
program on cognition-associated variables, metabolic syndrome and inflammatory markers in children. One hundred twenty-eight schoolchildren were included in this study (63 girls and 65 boys), aged 7-11 years, from a public school in the city of Porto Alegre-Brazil, selected by convenience. All children in the first to fifth grade were invited to participate in this project. The parents from those who agreed to participate signed the consent form as well as the children signed the assent form. The Ethics and Research Committee of Federal University of Rio Grande do Sul approved the study (2014997). The research received support from CNPq process number 401969/2016-9 'Universal Announcement'.

The minimum number of subjects in the sample was calculated through the software G*Power version 3.1. For a sample calculation, an effect size F of 0.15 (medium effect, corresponding to 1.7 in prevalence ratio) was used, as well as the level of significance of 0.05 and a statistical power of 0.95. Linear regression models have used with approximately five predictors and a 20% increase to cover for possible losses and refusals. Based on these criteria, the minimum sample size was 107 children. The sample size was calculated for linear regression, considering that mediation analysis consists of different regressions. Thus, the sample size is sufficient for this analysis.

**Measurements procedures**

The variables were measured at school by trained researchers. Weight was measured using a digital anthropometric scale, graded from 0 to 150 kg, with a resolution of 0.05 kg. The children should be lightly dressed and without shoes. Height was measured using a metric tape fixed on the wall and extended from the bottom upwards, with the children kept in a vertical position, with feet and trunk leaning against the wall. The described procedures followed the PROESP-BR standard. CRF was assessed by running and walking test in six minutes. The evaluated ones should accomplish the greatest number of turns, running or walking, in a sports court with the perimeter marked with 6 cones and the ground with indications of meters traveled (from 4 in 4 to close 54 meters). The measurement of the test was noted from the number of laps traveled, plus the meters in the case of those who at the end of the time did not complete a lap, so after multiplying the number of laps by the perimeter of meters covered was obtained the estimate of CRF.

Diatostic and systolic blood pressure levels (DPB and SBP) were determined by an automatic blood pressure monitor (Omron Digital Hem-7130), using different sized cuffs according to the circumference of the right arm. The children must be sitting, at least 5 minutes rest, with the arm supported. Mean arterial pressure (MAP) was calculated using the following formula: DBP+[0.333 × (SBP-DBP)]. We emphasize that a large number of published studies used MAP as an independent predictor of cardiovascular event. Besides, adiposity is similarly associated with SBP, DBP and MAP, indicating that independently of the blood pressure component, children with more adiposity are more likely to have a higher risk of hypertension.

After twelve hours of overnight fasting, capillary samples were collected. The fingertip was pierced using an automated lancet and the first drop of blood removed with a sterile cotton swab. This method was used because it allowed the dosage to be performed at school with fewer traumas and better acceptance between the children. Total cholesterol and triglycerides was determined by using Accutrend Plus, while glucose was evaluated by On Call Plus. Capillary samples was tested and authorized by Food and Drug Administration, and the coefficient of results variation (accuracy > 95% with an agreement of laboratory measures) are in according to the index established by National Cholesterol Education Program.

Percentage of body fat was assessed using dual-energy X-ray absorptiometry Lunar Prodigy Primo (General Electric Healthcare; Madison, WI), by the same well-trained professional. This measurement was taken at the physical activity laboratory of Federal University of Rio Grande do Sul (Brazil).

**Statistical Analysis**

Descriptive analysis was expressed considering the mean and standard deviations of all variables included in the study. All variables were checked for normality. Independent Two-tailed T-tests were used to examine the sex differences. Considering that the variables, triglycerides, mean arterial pressure glucose and cholesterol, did not show differences between sexes, all the statistical analysis were performed with both sexes together to increase statistical power. Pearson correlation was used to determine the relationship between %BF and CRF with cardiovascular risk factors.

To examine whether the association between CRF and TG and MAP was mediated by %BF, linear regression models were fitted using the PROCESS macro for the Statistical Package for Social Sciences (SPSS) version 24.0 (IBM Corp, Armonk, NY). The goal of this model was to investigate the total (c) and direct effects (a, b, c'), reflected by the unstandardized regression coefficient and significance between the independent and dependent variables in each model. The model also investigated the indirect effect obtained from the product of coefficients (a × b), which indicates the change in the TG or MAP for every unit change in the CRF that is mediated by the proposed mediator (i.e. %BF). The PROCESS macro used bootstrapping methods recommended by Preacher and Hayes (2008) for testing mediation hypotheses, using a resampling procedure of 10.000 bootstrap samples. Point estimates and confidence intervals (95%) were estimated for the indirect effect. The point estimate was considered significant when the confidence interval did not contain zero. Thus, the following criteria were used to establish mediation: (1) the independent variable (CRF) is significantly related to the mediator (%BF); (2) the independent variable (CRF) is significantly related to the dependent variable (MAP and TG); (3) the mediator (%BF ) is significantly related to the dependent variable (MAP and TG); and (4) the association between the independent and dependent variable is attenuated when the mediator is included in the regression model. The analysis was adjusted for age, sex and height.

All the analysis was carried out using the IBM SPSS 21 (SPSS, Inc., Chicago, Illinois, USA). The level of statistical significance was established at p < 0.05.
**Results**

Table 1 presents the descriptive characteristics of the sample. The results indicated that there were differences in mean values of CRF and %BF between girls and boys. Boys presented higher levels of CRF, while girls showed higher % in BF.

Partial correlations between %BF, CRF and cardiovascular risk factors are presented in Table 2. MAP and TG were the variables that showed an association with %BF and CRF. Also, there was an association between %BF and total cholesterol.

When we tested the mediator role of %BF in the association between CRF and MAP (Fig.1A), in the first regression equation, the association between CRF and %BF was negative (p=0.01). In the second equation, CRF was also negatively associated with MAP (p=0.002). Finally, in the third equation, when %BF and CRF were included simultaneously in the model, %BF was positively associated with MAP (p<0.001) and associated negatively with CRF (p=0.01). Furthermore, the association between CRF and MAP was attenuated when %BF was included in the model, indicating that %BF a mediator of this association (Indirect Effect=-0.008; IC: -0.0159 -0.0030), explaining 29% of this association. However, the analysis of the mediator role of %BF in the association between CRF and TG (Fig. 1B), showed that %BF is not a mediator of this association since the above-mentioned criteria for the mediation analysis was not observed.

| Table 1. Characteristics of the study samples according to sex in children. |
|-------------------|------------------|------------------|-------|
|                   | Total            | Boys             | Girls            |
|                   | Mean (SD)        | Mean (SD)        | Mean (SD)        | p    |
| Age (years)       | 8.82 (7.89)      | 8.36 (1.51)      | 8.43 (1.44)      | 0.39 |
| Weight (kg)       | 33.14 (10.59)    | 32.86 (9.30)     | 33.45 (11.83)    | 0.74 |
| Height (cm)       | 1.34 (0.10)      | 1.34 (0.09)      | 1.33 (1.11)      | 0.21 |
| Cardiorespiratory fitness (m) | 770.52 (136.82) | 798.10 (146.36) | 741.36 (119.45) | <0.001 |
| Percentage body fatness (%) | 32.73 (8.36)    | 30.34 (8.80)     | 35.15 (7.15)     | <0.001 |
| Systolic blood pressure (mmHg) | 103.49 (11.56) | 103.40 (11.35)  | 104 (11.82)      | 0.62 |
| Diastolic blood pressure (mmHg) | 60.75 (8.83)    | 60.78 (9.13)     | 69.28 (9.28)     | 0.29 |
| Triglycerides (mg/dL) | 116.69 (61.68)  | 108.83 (66.79)   | 125.49 (54.46)   | 0.07 |
| Glucose (mg/dL)   | 86.01 (7.46)     | 86.19 (7.23)     | 85.78 (7.79)     | 0.69 |
| Cholesterol (mg/dL) | 177.01 (22.34)  | 175.72 (23.15)   | 178.43 (21.46)   | 0.42 |
|                   | SD: Standard deviation; p ≤ 0.05 |

| Table 2. Partial correlation between body fat percentage and CRF with cardiovascular risk factors in children |
|---------------------------------|------------------|------------------|-------|
| Percentage of body fat          | Cardiorespiratory fitness |
|                                 | r   | p     | r   | p     |
| MAP                             | 0.38 | <0.001 | -0.45 | <0.001 |
| TG                              | 0.31 | 0.004 | -0.31 | 0.005 |
| Glucose                         | 0.06 | 0.56  | -0.09 | 0.35  |
| Total Cholesterol               | 0.27 | 0.009 | -0.05 | 0.62  |
|                                 | MAP: Mean arterial pressure; TG: Triglycerides; CRF: Cardiorespiratory fitness; All analysis were adjusted for age, sex and height. |

A B

Figure 1. (A) Percentage of body fat mediation model of the relationship between cardiorespiratory fitness and mean arterial pressure; (B) Percentage of body fat mediation model of the relationship between cardiorespiratory fitness and triglycerides; CRF: Cardiorespiratory fitness; MAP: Mean arterial pressure; %BF: Percentage of body fat; TG: Triglycerides. *p<0.001. The analysis was adjusted for age, sex and height.
Discussion

Our study shows that %BF and CRF were associated with TG and MAP. Moreover, the mediation analyses disclosed that %BF was a mediator in the relationship between CRF and MAP and the estimated percentage of total effect mediated by %BF was 29%. Our findings also showed a negative association between CRF and TG, independent of %BF.

Our data agree with a study developed with 7,821 children and adolescents indicated that %BF were associated with total cholesterol, HDL and LDL cholesterol and triglycerides21. Data from Brazil showed that individual between the ages of 6 and 19 years with excess of body weight, elevated %BF and waist circumference presented a positive correlation with alteration in the lipid profile22. Furthermore, Wheelock, Fufaa, Nelson, Hanson, Knowler, Sinha23 used BMI as an adiposity parameter and found that higher BMI was associated with blood pressure elevation and high TG levels in children and adolescents.

Likewise, our study found that CRF was associated with MAP and TG, which is in accordance with the previous study showing that the prevalence of dyslipidemia is directly related to both obesity and lower levels of CRF2. CRF was also associated with blood pressure in children24,25.

Therefore, CRF and adiposity are considered an important health indicator in children26. Indeed, previous studies have been demonstrated a relationship between CRF and adiposity with cardiovascular risk factor in children and adolescents27. However, it is not established which indicator, CRF or adiposity, is the most important for health and the link between both remain controversial, especially in the child population. Barry, Baruth, Beets, Durstine, Liu, Blair28 suggested that high levels of CRF could counteract the negative effects of obesity morbidity and mortality. Based on this assumption, obesity compared to CRF, would be the less preponderant factor for health than is widely believed. On the other hand, Buchan, Young, Boddy, Baker29 found that adiposity, evaluated through BMI and non-CRF were independently associated with cardiometabolic risk factors.

Our mediation analysis offers new information into understanding the relationship between adiposity, CRF and MAP, suggesting that %BF parameter mediates the relationship between CRF and MAP. Thus, the effect of CRF on MAP seems to be minimized by the negative effect of body adiposity. These results were achieved after adjustment for sex, age and height and highlight that high CRF levels and a healthy body composition are important for maintaining adequate arterial blood pressure. Taking this aspect into consideration, physical activity program should be focused on reducing body fat and increasing CRF.

In accordance with our results, a study including 1,604 schoolchildren from Spain, indicated that adiposity assessed through BMI, mediates the relationship between CRF and MAP23. In the same line, Diez-Fernández, Sanchez-Lopez, Mora-Rodriguez, Notario-Pacheco, Torrijos-Nino, Martinez-Vizcaino1 showed that BMI mediated the association between CRF and metabolic syndrome, including MAP, with a percentage of effect mediated by BMI of 16%. Similar results were found in young Spanish adults in which different body composition variables such as BMI, waist circumference, fat mass percentage acted as mediators between CRF and blood pressure30. Likewise, in 935 Colombian children and adolescents, Garcia-Hermoso, Agostinis-Sobrinho, Mota, Santos, Correa-Bautista, Ramirez-Vélez31 showed that high levels of CRF might not counteract the negative consequences ascribed to adiposity on inflammation.

Thus, high blood pressure levels are related to low levels of CRF and adiposity32. Likely, the mechanisms involved in increased blood pressure are associated with the adipokines secreted by visceral adipocytes that have been linked to diminished insulin-sensitivity. Furthermore, the favorable impact of CRF on blood pressure is based on reduced sympathetic nervous system activity and improvement in endothelial function33.

Conversely, %BF was not a mediator in the relationship between CRF and TG. Our finding indicates that there was a direct association between these variables, independent of %BF, suggesting the necessity of highlighted aerobic fitness as a health indicator. Indeed, studies have been shown that low CRF in children and adolescents is independently associated with cardiovascular risk factors, including TG34.

A study developed with adults indicated that changes over time in both adiposity and physical fitness predicted the development of cardiometabolic risk factors, such as hypertension and dyslipidemia, but the impact of fitness appears somewhat better than did adiposity for future risk of this disorders35. In addition, increasing fitness was associated with reductions in all-cause of cardiovascular mortality, approximately 15%. Regarding BMI changes, no associations were found36. Thus, studies have indicated that a normal-weight is not synonymous with being healthy.

Therefore, the relative and combined contribution of physical fitness and fatness to health in children are scarce. Considering that obesity is a major public health, and physical fitness is related to cardiometabolic risk, we highlight that lifestyle intervention programs should focus on reducing weight/fat, but also increasing CRF. Several studies suggest that intervention programs developed at school environment that address diverse components, such as physical activity and nutritional guidance, appear to be extremely effective for promoting an active and healthy lifestyle37,38, even CRF39. Thus, effort should be concentrated to sustain an effective physical education curriculum, aimed to health promotion, during the key period of children and adolescents, considering that choices made in this period of life tend to remain in adulthood.

Our study has some limitations. The analysis had a cross-sectional design, so we cannot make cause-effect inferences, also the sample was selected by convenience. Furthermore, there are many factors that were not measured and influence cardiometabolic profile, especially in children, such as sexual maturity or diet. However, strengths of this study are the mediation analysis; one of the first studies that used this kind of analysis in Brazilian children; use of the gold standard to measure cardiovascular risk factors, including adiposity as a health indicator parameter. Our data add to previous findings reported in the literature and give additional strength towards the influence of %BF in the association between CRF and cardiovascular risk factors in Brazilian children. Future studies should also consider potential confounders, such as physical activity, exercise, and nutrition.
sedentary habits and socioeconomic status to better explaining the role of %BF. In fact, cardiometabolic risk is a complex issue, related to lifestyle and genetic factors.

In conclusion, %BF mediates the association between CRF and MAP. Our findings show that the importance of a healthy body composition for the prevention of high blood pressure levels in childhood as well as the relevance of physical activity on these parameters.

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