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Metabolic syndrome components can predict C reactive protein concentration in adolescents

Larissa Leandro da Cruz1, Luciane Daniele Cardoso2, Daniela Pala1, Heberth de Paula1, Joel Alves Lamounier1, Camilo Adalton Mariano Silva1, Ana Carolina Pinheiro Volp1 and Renata Nascimento de Freitas1,2


Abstract

Background: Metabolic syndrome (MS) is suggested to be associated with a low grade inflammation state, but the relationship between inflammation biomarkers and the components of metabolic syndrome in adolescents are still lacking.

Objective: To investigate the association between C-reactive protein (CRP) serum concentrations and metabolic syndrome components in adolescents.

Methods: A cross-sectional population based study was conducted. Anthropometric, biochemical and clinical data were collected from 524 adolescents (11-15 years old) randomly sampled from school population of Alegre city, Espírito Santo, Brazil. Data were analyzed by STATA version 9.0.

Results: Adolescents with higher values for BMI (p = 0.001) and higher body fat percentage (p = 0.003) had higher CRP concentrations than those with lower BMI and body fat percentage. CRP concentrations was directly correlated with BMI (r = 0.17, p = 0.0001), waist circumference (r = 0.15, p = 0.0005), HDL-c (r = 0.13, p = 0.003), fasting insulin (r = 0.12, p = 0.003) and systolic blood pressure (r = 0.11, p with = 0.01). In the multiple linear regression analysis BMI (r = 0.05, p = 0.002), fasting glucose (r = -0.01, p = 0.003) and HDL-c (r = 0.017, p < 0.001) were associated to CRP concentrations after adjusting for the other components of MS.

Conclusion: The association found between individual components of MS and CRP concentrations suggests that inflammation might be an early event in the development of metabolic disorders in adolescents.

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Key words: Adolescents. Risk factors. C reactive protein. Metabolic syndrome x. Obesity.

COMPONENTES DEL SÍNDROME METABÓLICO PUEDEN PREDECIR CONCENTRACIÓN DE PROTEÍNA C REACTIVA EN ADOLESCENTES

Resumen

Antecedentes: El síndrome metabólico (SM) se sugiere que está asociado con un estado de inflamación crónica de bajo grado, pero la relación entre biomarcadores de inflamación crónica de bajo grado, pero la relación entre biomarcadores de inflamación y los componentes del síndrome metabólico en adolescentes son escasos.

Objetivo: Investigar la asociación entre las concentraciones séricas de proteína C reactiva (CRP) y los componentes del síndrome metabólico en adolescentes.

Metodología: Hemos realizado una población basada en estudio de corte transversal. Los datos antropométricos, bioquímicos y clínicos se obtuvieron de 524 adolescentes (11-15 años de edad) seleccionados al azar de la población escolar de la ciudad Alegre, Espírito Santo, Brasil. Los datos fueron analizados por STATA versión 9.0.

Resultados: Los adolescentes, con valores más altos de IMC (p = 0,001) y mayor porcentaje de grasa corporal (p = 0,003) tuvieron mayores concentraciones de PCR que aquellos con menor IMC y porcentaje de grasa corporal. Las concentraciones de PCR se correlacionaron directamente con el IMC (r = 0,17, p = 0,0001), la circunferencia de la cintura (r = 0,15, p = 0,0005), HDL-c (r = 0,13, p = 0,003), la insulina en ayunas (r = 0,12, p = 0,003) y la presión arterial sistólica (r = 0,11, p = a 0,01). En el análisis de regresión lineal múltiple IMC (r = 0,05, p = 0,002), la glucosa en ayunas (r = -0,01, p = 0,003) y HDL-c (r = 0,017, p < 0,001) se asociaron con las concentraciones de PCR después de ajustar por los otros componentes de SM.

Conclusion: La asociación encontrada entre los componentes individuales de SM y las concentraciones de PCR sugiere que la inflamación podría ser un evento temprano en el desarrollo de trastornos metabólicos en los adolescentes.

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**Abbreviations**

- %BF: Body Fat Percentage.
- BMI: Body Mass Index.
- CRP: C-reactive protein.
- DBP: Diastolic Blood Pressure.
- HDL-c: High Density Lipoprotein cholesterol.
- LDL-c: Low Density Lipoprotein cholesterol.
- MS: Metabolic Syndrome.
- SBP: Systolic Blood Pressure.
- STATA: Statistical Software for Professionals.
- WC: Waist circumference.

**Introduction**

In the last three decades, obesity in the pediatric population became a public health problem worldwide. In Brazil, the occurrence of the nutritional transition in recent decades has shown a significant decrease in the prevalence of malnutrition and a significant increase of overweight in both sexes. Overweight is diagnosed in about one fifth of Brazilian adolescents and it exceeded six times the frequency of underweight.

The high prevalence of obesity is a concern since the excess of body adiposity, especially the visceral, is related to changes in the lipid profile, on glucose metabolism and an increased blood pressure. These conditions occurring simultaneously characterize the Metabolic Syndrome (MS) and increase the risk of cardiovascular disease and type 2 diabetes.

Despite still lacking a universal diagnostic criterion for MS, what can be observed is that, regardless of the institution, the parameters considered to diagnose the syndrome have few variations. Also, regardless of the criteria used, the scientific literature shows a high correlation between obesity and the prevalence of MS, even during childhood. MS is very prevalent among overweight and obese children and adolescents.

The inflammation state that accompanies obesity is considered to be a chronic low-grade inflammation, generally associated with high concentrations of leukocytes, fibrinogen and other inflammatory biomarkers, such as C reactive protein Reactive Protein (CRP).

CRP is an acute phase reactant protein produced by hepatocytes as part of the inflammatory response to tissue damage induced by infection, trauma and malignancies. Some researchers have demonstrated an association between mild elevations of serum CRP and risk of developing cardiovascular disease, type 2 diabetes, cancer and MS.

There are many studies characterizing the risk variables and evaluating the prevalence of MS among Brazilian adolescents. However, few studies with adolescents have searched for the association between metabolic syndrome components and inflammatory biomarkers. Thus, we aimed to evaluate the associations among MS components and CRP concentrations in Brazilian adolescents.

**Methods**

A cross-sectional population based study was conducted in Alegre city, Espírito Santo state, in Brazil Southeast region. Clinical and anthropometric data were obtained from the subjects. Blood samples for biochemical analyzes were also collected. Data collection was performed in schools by researchers or trained and qualified undergraduate students. Samples were collected from November 2010 to May 2011, except during school holidays. This study was approved by the Ethics Committee of Federal University of Espírito Santo, and a written informed consent term was obtained from the parents of each participant.

**Data collection**

Weight and body fat percentage (%BF) were determined by a bipolar electrical impedance monitor (Tanita Ironman). Height was obtained using a portable stadiometer (Alturaexata). Waist circumference (WC) was measured to the nearest 0.1 cm according to WHO recommendations.

Body Mass Index (BMI) was calculated by dividing weight in kilograms (kg) by height in square meters (m²). Subjects were categorized by BMI as normal, overweight and obese according to the IOTF and to WHO definitions.

Subjects were also categorized by body fat percentage according to sex, adopting the cutting points suggested by Williams et al.: body fat percentage was considered high for girls when above 30% and for boys when above 25%.

Blood pressure measurement was taken according to the recommendations of the 6th Brazilian Guidelines...
on Hypertension. Systolic and diastolic blood pressure was measured in a sitting position and three times after a 1-minute rest.\textsuperscript{30} 

**Laboratory measurements**

Blood samples were collected after a 12 hours fasting. Lipids (total cholesterol, HDL cholesterol, triacylglycerols), glucose and high sensitivity CRP were measured in a spectrophotometer (Model 700 plus Femto\textsuperscript{2}®) using specific commercial kits (Labtest\textsuperscript{®}). LDL cholesterol concentrations were calculated using the equation proposed by Friedewald et al.,\textsuperscript{21} since no value found for the triglycerides was $> 400\text{mg/dl}$. Insulin was determined using a specific immunoassay kit (IRI Siemens\textsuperscript{®}). Insulin resistance was assessed by the homeostasis model assessment of insulin resistance (HOMA-IR) calculated by the formula: HOMA-IR = fasting plasma insulin ($\mu\text{U/ml}$) x fasting plasma glucose (mmol/L)/22.5.\textsuperscript{31}

**Metabolic syndrome components**

Four categories of metabolic syndrome components were considered: body composition (assessed by BMI and WC), glucose metabolism (assessed by fasting glucose, fasting insulin and HOMA-IR), lipids metabolism (assessed by total cholesterol, HDL-c, LDL-c and triglycerides) and blood pressure.

**Statistical analysis**

All the statistical analysis was performed using the Statistical Software for Professionals (STATA) for Windows (version 9.0). A p value $\leq 0.05$ was considered. The Shapiro-Wilk test was applied to assess the assumption of normality for the data. Data were reported as mean and standard deviation or median and interquartile range, for parametric and nonparametric variables, respectively. Frequencies, means and medians were compared by using Chi-square ($\chi^2$), t Student test and Mann-Whitney U test, respectively. Spearman correlation was used to track the association between CRP and MS components. Multiple linear regression models were designed to identify predictors of CRP. The selection of variables was done by backward stepwise procedure and the final model was composed by variables that showed a p value $\leq 0.05$, after adjusting for all the interest variables. Then the adequacy of the models was verified by performing residual analysis.

**Results**

Final sample consisted of 247 (47.14\%) boys and 277 (52.86\%) girls. The mean age of the total population was 13 years ($\pm 1.29$) and did not differ between genders (p = 0.56) (data not shown). Table I shows the mean or median values for the MS components of the participants according to gender. Regarding body composition, there was no statistical difference between BMI and WC among boys and girls. On the other hand, girls showed a higher %BF than boys (p $< 0.0001$).

Mean values of total cholesterol its fractions and triacylglycerols were similar in both genders. Fasting plasma glucose median value was higher in boys (p = 0.019). On the other hand the fasting insulin (p $< 0.0001$), HOMA-IR (p $< 0.0001$) and CRP (p = 0.023) medians were higher among girls. Diastolic blood pressure (DBP) was similar (65.66 mmHg) between boys and girls while boys presented higher systolic blood pressure (SBP) than girls (p = 0.04).

Table II shows the median CRP concentrations among the adolescents categorized by BMI and %BF. Adolescents presenting higher BMI values (p = 0.001) and also %BF (p = 0.0026), also had significantly higher CRP concentrations than those with normal BMI and %BF.

CRP concentrations was positively and significantly correlated with BMI (p = 0.0001), WC (p = 0.0005), HDL-c (p = 0.003), fasting insulin (p = 0.003), and SBP (p = 0.01), as shown in table III. A negative and significant correlation was observed between CRP concentrations and fasting glucose (p = 0.05), total cholesterol (p = 0.05) and LDL-c (p = 0.0007).

Based on the correlation analysis, a multiple linear regression analyses was performed to assess the predictability of the CRP concentrations by the MS components. The best model explained 7\% the variation in CRP concentrations and it was defined by the following regression equation: CRP = 0.05$^*$ BMI + (- 0.01)$^*$glucose + 0.017$^*$ HDL-c (p $< 0.035$), as shown in table IV.

**Discussion**

In this cross-sectional study CRP concentrations were higher in overweight and obese adolescents (considering BMI) and also in those adolescents whose %BF was higher.

Most of the previous studies with children and adolescents from Brazil and other countries\textsuperscript{7,12,23} showed a positive association between CRP concentrations and BMI. However, it is well known that BMI is not the best way to evaluate adiposity. So, our finding of a similar relation between CRP and %BF, reinforce the association of high adiposity and low grade inflammation in adolescents. An apparent reason for these findings is that the excess of adipose tissue is able to secret inflammatory adipokines such as interleukin-6 and a tumor necrosis factor that acting on the liver would be able to stimulate the CRP production.\textsuperscript{24,25}
Table I

<table>
<thead>
<tr>
<th></th>
<th>Total (n = 524)</th>
<th>Girls (n = 277, 52.86%)</th>
<th>Boys (n = 247, 47.14%)</th>
<th>p**</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Body composition</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>BMI (kg/m², n = 523)</td>
<td>20.3 ± 3.8</td>
<td>20.6 ± 3.9</td>
<td>20.1 ± 3.6</td>
<td>0.14</td>
</tr>
<tr>
<td>WC (cm, n = 521)</td>
<td>69.75 ± 9.30</td>
<td>69.25 ± 9.22</td>
<td>70.31 ± 9.38</td>
<td>0.19</td>
</tr>
<tr>
<td><strong>Glucose metabolism</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Glucose (mg/dL, n = 523)*</td>
<td>78.07</td>
<td>76.87</td>
<td>78.96</td>
<td>0.019</td>
</tr>
<tr>
<td>Insulin (µU/mL, n = 512)*</td>
<td>7.96</td>
<td>9.17</td>
<td>6.79</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>HOMA IR (n = 511)*</td>
<td>1.52</td>
<td>1.77</td>
<td>1.32</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td><strong>Lipids metabolism</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TC (mg/dL, n = 523)</td>
<td>130.75 ± 37.02</td>
<td>129.04 ± 36.92</td>
<td>132.67 ± 37.11</td>
<td>0.26</td>
</tr>
<tr>
<td>HDL-c (mg/dL, n = 523)</td>
<td>47.31 ± 16.31</td>
<td>47.79 ± 15.89</td>
<td>46.98 ± 16.79</td>
<td>0.58</td>
</tr>
<tr>
<td>LDL-c (mg/dL, n = 523)</td>
<td>67.99 ± 36.71</td>
<td>65.36 ± 35.79</td>
<td>70.93 ± 37.56</td>
<td>0.066</td>
</tr>
<tr>
<td>TG (mg/dL, n = 523)</td>
<td>74.08 ± 46.17</td>
<td>76.55 ± 42.22</td>
<td>71.31 ± 50.15</td>
<td>0.19</td>
</tr>
<tr>
<td><strong>Blood pressure</strong></td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>SBP (mmHg, n = 521)*</td>
<td>103.33</td>
<td>100</td>
<td>103.33</td>
<td>0.04</td>
</tr>
<tr>
<td>DBP (mmHg, n = 521)</td>
<td>65.66 ± 7.66</td>
<td>65.62 ± 7.37</td>
<td>65.67 ± 7.99</td>
<td>0.93</td>
</tr>
<tr>
<td>CRP (mg/L, n = 523)*</td>
<td>0.7</td>
<td>ND-1.98</td>
<td>0.92</td>
<td>0.023</td>
</tr>
</tbody>
</table>

BMI: Body mass index; CRP: C reactive protein; WC: Waist circumference; SBP: Systolic blood pressure; DBP: Diastolic blood pressure; TC: Total cholesterol; HDL-c: High density lipoprotein cholesterol; LDL-c: Low density lipoprotein cholesterol; TG: Triacylglycerols; HOMA-IR: Homeostasis model assessment-insulin resistance.

Results are presented as mean and standard deviation for parametric variables or as *median and interquartile interval for nonparametric variables.

**P value for comparison between gender. Student’s t test for mean (parametric variables) and Mann-Whitney U test for medians (nonparametric variables).
Besides, girls presented higher %BF and also higher CRP concentration than boys. Fernandes et al. (2009) evaluated the association between measures of adiposity and serum concentrations of CRP. They observed that women presented higher mean values of BMI and CRP, when compared to men. Whether or not the higher concentration of CRP found in woman or girls is related to higher adiposity compared to men or boys, remains to be investigated.

We observed a correlation between CRP concentrations and MS components: BMI, WC, glucose, total cholesterol, HDL-c, LDL-c, insulin and SBP. However, in the multiple linear regression analysis the components of MS that possibly explained 7% of the variation in CRP concentrations were BMI, glucose and HDL-c. So, these MS components were able to weakly predict the concentrations of CRP. Other researchers, evaluating adults or children and adolescents, found an association between CRP concentrations, BMI and glucose metabolism markers (insulin, HOMA-IR or fasting glucose itself) and lipid metabolism (HDL-c and triacylglycerols). The mechanisms underlying these associations are not clearly elucidated, but there are possible explanations. During the development of obesity occurs an “adipose tissue remodeling,” i.e., there is an increase in white adipose tissue, caused by a hyperplasia and, or adipocyte hypertrophy, an increased infiltration of immune cells such as lymphocytes and macrophages and an imbalance between production of pro-inflammatory and anti-inflammatory adipokines. The presence of the macrophages infiltration in adipose tissue makes it a source of inflammatory proteins.

### Table II

| Nutritional indicator | Status (n = 523) | Median (mg/L) | Interquartile range | p
|----------------------|-----------------|---------------|---------------------|-------|
| Body Mass Index      | **Normal Weight** (n = 409, 78.20%) | 0.64 | ND-1.86 | 0.02*
|                      | **Overweight** (n = 88, 16.83%) | 1.16 | 0.04-2.39 | 0.02*
|                      | **Obese** (n = 26, 4.97%) | 1.04 | 0.09-2.41 | 0.02*
| Body Mass Index      | **Normal Weight** (n = 365, 69.8%) | 0.63 | ND-1.84 | 0.02*
|                      | **Overweight** (n = 117, 22.4%) | 1.08 | 0.03-2.14 | 0.001*
|                      | **Obese** (n = 41, 7.8%) | 1.43 | 0.26-2.38 | 0.001*
| Body Fat Percentage  | **Normal** (n = 445, 85.1%) | 0.64 | ND-1.88 | 0.0026**
|                      | **High** (n = 78, 14.9%) | 1.37 | 0.09-2.41 | 0.0026**

ND = Not detectable.

*P value of Kruskal-Wallis test for comparison between categories.

**P value of Mann-Whitney U test for comparison between categories.

### Table III

| Metabolic syndrome components | r       | P*
<table>
<thead>
<tr>
<th></th>
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</thead>
<tbody>
<tr>
<td><strong>Body composition</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>0.17</td>
<td>0.0001</td>
</tr>
<tr>
<td>WC (cm)</td>
<td>0.15</td>
<td>0.0005</td>
</tr>
<tr>
<td><strong>Glucose metabolism</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Glucose (mg/dL)</td>
<td>-0.08</td>
<td>0.05</td>
</tr>
<tr>
<td>Insulin (µU/mL)</td>
<td>0.12</td>
<td>0.003</td>
</tr>
<tr>
<td>HOMA-IR</td>
<td>0.08</td>
<td>0.07</td>
</tr>
<tr>
<td><strong>Lipid metabolism</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TC (mg/dL)</td>
<td>-0.083</td>
<td>0.05</td>
</tr>
<tr>
<td>HDL-c (mg/dL)</td>
<td>0.13</td>
<td>0.003</td>
</tr>
<tr>
<td>LDL-c (mg/dL)</td>
<td>-0.15</td>
<td>0.0007</td>
</tr>
<tr>
<td>TG (mg/dL)</td>
<td>0.03</td>
<td>0.51</td>
</tr>
<tr>
<td><strong>Blood pressure</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SBP (mmHg)</td>
<td>0.11</td>
<td>0.01</td>
</tr>
<tr>
<td>DBP (mmHg)</td>
<td>0.07</td>
<td>0.11</td>
</tr>
</tbody>
</table>

BMI: Body mass index; WC: Waist circumference; SBP: Systolic blood pressure; DBP: Diastolic blood pressure; TC: Total cholesterol; HDL-c: High density lipoprotein cholesterol; LDL-c: Low density lipoprotein cholesterol; TG: Triacylglycerols; HOMA-IR: Homeostasis model assessment-insulin resistance.

*P value for Spearman correlation between metabolic syndrome components and serum C reactive protein concentration.

### Table IV

| Model | Coefficient β ± SD | p
<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Intercept</td>
<td>0.16 ± 0.52</td>
<td>0.761</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>0.05 ± 0.017</td>
<td>0.002</td>
</tr>
<tr>
<td>Glucose (mg/dL)</td>
<td>-0.01 ± 0.003</td>
<td>0.003</td>
</tr>
<tr>
<td>HDL-c (mg/dL)</td>
<td>0.017 ± 0.004</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

BMI: Body mass index; HDL-c: High density lipoprotein cholesterol.

Variables included in the model: body mass index, waist circumference, glucose, insulin, HOMA-IR, total cholesterol, HDL-c, LDL-c, triacylglycerols, systolic and diastolic blood pressure (r² = 0.07, p < 0.0001).

Besides, girls presented higher %BF and also higher CRP concentration than boys. Fernandes et al. (2009) evaluated the association between measures of adiposity and serum concentrations of CRP. They observed that women presented higher mean values of BMI and CRP, when compared to men. Whether or not the higher concentration of CRP found in woman or girls is related to higher adiposity compared to men or boys, remains to be investigated.
signals release that act in the adipose tissue metabolism itself and control metabolic changes associated with obesity.30,31 Adipokines, besides acting in metabolic functions, are able to promote a major impact on several body functions such as control of food intake and energy balance, immune system, angiogenesis, blood pressure, lipid metabolism, body homeostasis and insulin sensitivity.32,33

Indeed, some studies35,36 suggest that the obesity and the inflammation of the adipose tissue are the central component of the MS, and not the insulin resistance, as previously postulated by Reaven.37 Even in studies involving an adult population, in whom the metabolic sequela, in clinical terms, could already have been established, obesity, measured by BMI, had the central role in the development of MS when compared to all factors.35,36

The limitations of this study are related to the sectional design and the inflammatory biomarker used. A cross-sectional study does not establish a causal relationship, since the exposure and outcome are evaluated at the same period. Regarding the biomarker assessed, it is not a specific marker for subclinical chronic inflammation related to adipose tissue metabolism. However, even with these limitations, what can be highlighted is that this is a population-based study that produced original results that are consistent with scientific literature.

Therefore, this study contributes to demonstrate associations between individual components of MS and CRP concentrations in adolescents. The association between fasting glucose, BMI, HDL-c and CRP shows a close relationship of these alterations with inflammation which might imply in a high risk for developing cardiovascular disease and type 2 diabetes later in life.

Acknowledgements

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