Metabolic Syndrome after Preeclampsia: A Cohort Study with a Mean Follow-Up of 14 Years

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Abstract

Objective: To investigate the occurrence and characterization of metabolic syndrome (MetS) in the long term after pregnancies with preeclampsia. Design: Retrospective cohort study. Setting: Assis Chateaubriand Maternity Teaching Hospital - Federal University of Ceará, Fortaleza, Ceará, Brazil. Sample: 68 patients who gave birth between 1992 and 2002 at the Maternity, 34 patients with a history of preeclampsia and 34 with no history of obstetric complications.

Methods: Blood pressure and body compositional indices were recorded. Fasting blood samples were tested for glucose, total cholesterol, high density lipoprotein-cholesterol, low density lipoprotein-cholesterol and triglycerides. A questionnaire was used to collect demographic data including family history of diseases associated with cardiovascular diseases. Criteria for metabolic syndrome were defined by International Diabetes Federation 2005 (IDF). Main outcome measures: Occurrence and characterization of MetS.

Results: There were 18 (52.9%) diagnoses of MetS in the group of women without a history of obstetric complications and 28 (82.3%) in the group of women with a history of preeclampsia, p=0.01 with a RR of 4.1 (CI 95%), 1.4 - 12.2, p=0.009). The number of components to characterize MetS were, respectively, 2.7 (± 1.3) and 3.3 (± 1.3), p=0.05.

Conclusions: Women with a history of preeclampsia have a higher prevalence of MetS 14 years after gestation.

Keywords: Metabolic Syndrome X; Preeclampsia; Cardiovascular Diseases; Retrospective studies

Introduction

Of all the conditions that affect pregnancy and childbirth, preeclampsia is one of the most responsible for high rates of maternal and perinatal mortality in many countries [1,2]. Studies show that women with a history of preeclampsia are at increased risk of cardiovascular disease (CVD) in the long term, as demonstrated by several studies with varied follow-up times and specific outcomes for cardiovascular events [3]. A meta-analysis that evaluated studies with a follow-up period of 10 to 14 years, found a relative risk (RR) of 3.70 for systemic arterial hypertension (SAH), 2.16 for acute myocardial infarction (AMI), 1.84 for strokes and 1.79 for deep venous thrombosis (DVT) in these patients [3]. Preeclampsia and CVD have common underlying mechanisms such as dyslipidemia, inflammation, hypercoagulability and insulin deregulation. These factors are components of the clinical framework of metabolic syndrome (MetS) [4,5].

MetS includes a number of metabolic risk factors including the abnormal distribution of body fat, insulin resistance, atherogenic dyslipidemia and elevated blood pressure [5]. Compared with individuals without MetS, those diagnosed with MetS have a higher incidence of cerebrovascular disease and a higher mortality from these causes [6]. Given the significant association between the two conditions, preeclampsia and metabolic syndrome, with the occurrence of CVD, this study aimed to evaluate the occurrence and characteristics of MetS in the long term in women with history of preeclampsia.

Methods

Study design

This is a retrospective cohort study conducted in the Assis Chateaubriand Maternity Teaching Hospital, Fortaleza, Ceará, Brazil, which cares for patients from the capital and the interior, with medium and high risk pregnancies. The population was composed of women who gave birth in the period from 1992 to 2002 in the aforementioned hospital and who resided in the state capital.

The study included women diagnosed with pure preeclampsia as documented in their medical records when discharged. Preeclampsia was defined as the presence of gestational hypertension and concomitant proteinuria in the second half of pregnancy, have been based on the criteria of the International Society for the Study of Hypertension in Pregnancy [7]. According to the International Society for the Study of Hypertension in Pregnancy criteria, gestational
hypertension is defined as diastolic blood pressure more than 90 mm Hg, systolic blood pressure more than 140 mm Hg, or both measured on two or more separate occasions at least 4 hours apart; proteinuria was diagnosed when there was more than 300 mg per 24 hours or when dipstick urinalysis was more than 2+ [7].

The group without a history of preeclampsia was composed of women who gave birth during the same period as the exposed women, selected so that there was one non-exposed patient to each exposed patient. The sample was calculated using STATA® statistical software, version 12.0 (Stata Corp, USA), considering a significance level of 5%, 80% power and the averages of the mean systolic arterial blood pressures (SBP) found in the two groups, which was the main outcome found in previous studies, resulting in a sample size equal to 60 with 30 patients in each group. Women with a history of other complications associated with the pregnancy-index (Placental abruption, gestational diabetes and placenta previa) or who were pregnant or postpartum at the time of the invitation for evaluation were excluded from the study. The chart shows how the final number of patients was obtained at the end of the study (Figure 1).

Clinical evaluation was done through the collection of anthropometric and laboratory variables. All analyses were performed by technicians blinded for study group and unaware of the underlying hypothesis. The verification protocol was defined according to international consensus [5,8,9]. Blood pressure was evaluated using the Micro life BP 3BTO-H® semi-automatic monitor following the recommendations of American Heart Association [9]. The weight and percentage of body fat were measured using the bio impedance technique with the Wiso® W835 digital analyzer scale with high precision sensors that use infrared and ultrasound technology to analyze body composition to show the percentages of lean body mass, fat, water and bone weight. Height was measured using the Seca® stadiometer with a scale of 0 to 220 cm and 0.1 cm accuracy. To calculate the Body Mass Index (BMI), the weight in kilograms was divided by the height in meters squared and the evaluation followed the classification of American Heart Association [8].

For the measurement of waist circumference (WC), the patient was requested to remain in a supine position, with their arms relaxed along their sides, wearing light clothing without a belt, taking as reference the last rib and the iliac crest and performing the measurement at the midpoint between them. To measure the abdominal circumference (AC) the same recommendations were followed using the umbilicus as reference. For Caucasian women the International Diabetes Federation sets a waist circumference measurement equal to or above 80 cm as the cut-off point for increased cardiovascular risk [5]. The measurement of the hip circumference (HC) was performed using the greatest curvature of the hip as the reference. For these three measurements non-extendable WCS® measuring tapes were used with variation in mm and a length of 150 cm.

The waist/hip ratio (WHR), which is the ratio between the circumference of the waist and the hip in cm was calculated using as the cut-off point for women the value of 0.85. The waist-to-height (WHR), which is the ratio between waist circumference and height in cm, used the cut-off value of 0.53 as presented in the work of Pitanga and Lessa [10]. The Conicity Index (CI) was also calculated, determined by the formula described by Valdez [11], dividing the WC in cm by the constant 0.109, multiplied by the square root of the body weight in kilograms divided by the height in meters using 1.18 as the cutoff point as indicated by Pitanga and Lessa [10].

In addition, the bicipital fold was measured using Innovare 2 Cescorf® calipers, with the patient in a supine position with her right arm relaxed and extended along the body. The fold was ascertained in the direction of the longitudinal axis of the inside of the arm, precisely at the point of the greatest apparent circumference of the center of the biceps or located at the midpoint between the acromion and the olecranon. The laboratory evaluation included measurements of total cholesterol and HDL, LDL and VLDL, triglycerides and fasting glucose levels. The patients were instructed to fast for 12 hours and abstain from alcohol the day before venipuncture. The Tender enzymatic method was used to analyze the samples, fractions of total cholesterol and triglyceride levels and the enzymatic method was employed for the analysis of blood glucose.

To determine MetS, the criteria set by the International Diabetes Federation (IDF) [5], were used, namely: a AC equal to or greater than 80 cm for women, characterized by central obesity and the presence of

Figure 1: Organization Chart of patient selection for the study, MRF-Metropolitan Region of Fortaleza

Data collection

Interviews were used to collect data on obstetric history (maternal age at the time of delivery, gestational age according to the date of the last menstrual period or by ultrasound before the 20th gestational week in the medical records, type of delivery, number of pregnancies, deliveries and miscarriages and any complications in other pregnancies), socio-demographic data (race, current age, education, income, marital status and occupation), history of current morbidities and lifestyle (diagnosed conditions and/or receiving treatment, medication use, physical activity, and smoking) and a family history of CVD.

two or more components, specifically; triglyceride values equal to or greater than 150 mg/dL, HDL less than 50 mg/dL for women; SBP equal to or greater than 130 mmHg and/or a diastolic blood pressure (DBP) equal to or greater than 85 mmHg, or treatment for SAH; fasting glucose equal to or greater than 100 mg/dL, or treatment for diabetes mellitus (DM). Besides the characterization of MetS, the number of components to characterize the MetS was calculated and the frequency of alteration of each component.

Statistical analysis

The collected data were tabulated and analyzed using the STATA* Program, version 12.0 (StataCorp, USA) and the mean and standard deviation (SD) of clinical and metabolic variables were calculated. The Kolmogorov-Smirnov test was used to test the normality of the variables and the analysis of the differences between exposed and non-exposed women used the T-test for variables with normal distribution and the Mann-Whitney test for non-normal distribution. Pearson’s X2 test or the Fisher exact test were used for categorical data. The significance level of p<0.05 with a confidence interval (CI) of 95% was considered. The calculation of Relative Risk (RR) was carried out for variables that showed p<0.10 in the first analysis.

The study was approved by the MEAC Ethics Committee under Opinion No. 83/2011.

Results

The study included 68 patients, 34 with a history of preeclampsia and 34 with no history of obstetric complications. The mean age at the time of delivery was 25.7 ± 7.3 years. The average years of follow-up of the unexposed group was 14.8 ± 3.7 years and among the exposed group it was 13.4 ± 2.9, no difference was found between the groups for this variable (p=0.31).

Table 1 presents the characteristics of the current socio-demographic variables. The groups did not differ in age, race, marital status, occupation, education level and family income. The patients were mostly non-white, living with a partner, in paid work either as self-employed or employed formally, with an average of 7.6 ± 3.4 years of schooling and an income of 1.6 ± 6.8 minimum wages.

<table>
<thead>
<tr>
<th>Exposure</th>
<th>Non-exposed</th>
<th>Exposed</th>
<th>p†</th>
</tr>
</thead>
<tbody>
<tr>
<td>Current age (years) (mean ± DP) (n)</td>
<td>39.1 ± 8.4</td>
<td>40.6 ± 8.1</td>
<td>0.45*</td>
</tr>
<tr>
<td>Race (n,%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>7</td>
<td>20.6</td>
<td>9</td>
</tr>
<tr>
<td>Non-white</td>
<td>27</td>
<td>79.4</td>
<td>25</td>
</tr>
<tr>
<td>Civil Status (n,%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>With a partner</td>
<td>22</td>
<td>64.7</td>
<td>25</td>
</tr>
<tr>
<td>Without a partner</td>
<td>12</td>
<td>35.3</td>
<td>9</td>
</tr>
<tr>
<td>Occupation (n,%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Works outside home/Self-employed</td>
<td>17</td>
<td>50</td>
<td>20</td>
</tr>
<tr>
<td>Unemployed/Housewife</td>
<td>17</td>
<td>50</td>
<td>14</td>
</tr>
</tbody>
</table>

Table 2: Current clinical and obstetric history for women with and without preeclampsia – Average follow-up of 14 years, MEAC-UFC, Fortaleza, 2012

† Different statistical tests were used in the analysis * T-Student; †X 2 Test; ‡ Fisher Test.

The patients cited one or more morbid conditions: Systemic arterial hypertension, diabetes mellitus, acute myocardial attack, stroke, congestive heart failure.

CVE= Cardiovascular Event; CVD= Cardiovascular Disease.

GHS= Gestational Hypertensive Syndrome
The most common condition, with a prevalence of 47.1% among the exposed group and 17.6% in the non-exposed group, with a RR of 4.1 (CI 95% 1.4 to 12.5, p=0.012). The family history of CVD was similar between the groups, which can thus be excluded as a factor for confusion in the analysis of the outcomes. Although all the patients who were currently smokers were in the group with a history of preeclampsia, difference was not found between groups on this variable.

The groups were also similar regarding doing regular physical activity, with the majority of patients (72.1%) reporting that they did not do any physical activity. As for obstetric history, it was found that the women with a history of preeclampsia reported more complications in other pregnancies than non-exposed patients, with a RR of 3.4 (95% CI 1.2 to 9.1, p=0.01).

Table 3 presents the clinical and metabolic findings.

<table>
<thead>
<tr>
<th>Clinical and metabolic characteristics</th>
<th>Exposure</th>
<th>p††</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Anthropometric Variables</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>68.2 ± 9.7</td>
<td>71.4 ± 14.6</td>
</tr>
<tr>
<td>Height (m)</td>
<td>1.53 ± 0.1</td>
<td>1.52 ± 0.1</td>
</tr>
<tr>
<td>BMI (kg / m²)</td>
<td>28.2 ± 6.1</td>
<td>28.9 ± 9.2</td>
</tr>
<tr>
<td>SBP (mmHg)</td>
<td>120.4 ± 14.3</td>
<td>131.0 ± 23.8</td>
</tr>
<tr>
<td>DBP (mmHg)</td>
<td>79.5 ± 10.7</td>
<td>85.1 ± 14.9</td>
</tr>
<tr>
<td>AC (cm)</td>
<td>92.9 ± 8.9</td>
<td>95.6 ± 10.4</td>
</tr>
<tr>
<td>WC (cm)</td>
<td>86.5 ± 8.7</td>
<td>87.7 ± 11.2</td>
</tr>
<tr>
<td>HC (cm)</td>
<td>102.5 ± 7.7</td>
<td>106.1 ± 10.7</td>
</tr>
<tr>
<td>WHR</td>
<td>0.8 ± 0.1</td>
<td>0.7 ± 0.2</td>
</tr>
<tr>
<td>Index C</td>
<td>1.2 ± 0.2</td>
<td>1.1 ± 0.3</td>
</tr>
<tr>
<td>WHIR</td>
<td>0.54 ± 0.1</td>
<td>0.54 ± 0.2</td>
</tr>
<tr>
<td>Bicipital fold (mm)</td>
<td>14.2 ± 5.1</td>
<td>15.4 ± 6.9</td>
</tr>
<tr>
<td>Body fat (%)</td>
<td>42.9 ± 6.3</td>
<td>41.9 ± 7.7</td>
</tr>
<tr>
<td><strong>Metabolic variables</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total cholesterol</td>
<td>182.6 ± 41.8</td>
<td>190.5 ± 33.3</td>
</tr>
<tr>
<td>Triglycerides</td>
<td>143.7 ± 98.6</td>
<td>157.8 ± 62.4</td>
</tr>
<tr>
<td>LDL cholesterol</td>
<td>109.7 ± 36.1</td>
<td>122.4 ± 31.3</td>
</tr>
<tr>
<td>HDL cholesterol</td>
<td>43.5 ± 12.3</td>
<td>41.1 ± 8.4</td>
</tr>
<tr>
<td>Fasting glucose</td>
<td>95.0 ± 11.1</td>
<td>104.1 ± 38.7</td>
</tr>
</tbody>
</table>

Table 3: Clinical and metabolic characterization of women with and without preeclampsia – Average follow-up of 14 years, MEAC-UFC, Fortaleza, 2012

†Different statistical tests were used in the analysis: * T-test; **Mann-Whitney.


The patients’ average weight was 69.8 ± 12.4 kg, but there was no difference between the two groups with respect to this variable. The mean BMI of 28.6 ± 7.8 kg / m² indicates a profile of patients classified as overweight or obese, but without a statistical difference between groups.

The anthropometric measurements of AC and WC had averages above the recommended classification for low cardiometabolic risk, presenting 94.3 ± 9.7 cm and 87.1 ± 9.9 cm, respectively. A difference was detected between the groups in terms of mean SBP, with the group of women with a history of preeclampsia having the highest average. There was a trend towards higher values of DBP, triglycerides and LDL cholesterol in the exposed group. The clinical and metabolic variables were also categorized according to their cut-off points for altered values, there was a higher frequency of change in the group with a history of preeclampsia for the measurements of total cholesterol (values greater than 160 mg/dL) with a RR of 4.9 (95% CI 1.3 to 18.3, p=0.01).

Table 4 shows the data for the characterization of MetS according to the diagnosis, the number of altered components identified in each group and frequency of abnormal factors.

<table>
<thead>
<tr>
<th>Diagnosis of MetS</th>
<th>Non-exposed</th>
<th>Exposed</th>
<th>p††</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of MetS components (mean ± SD)</td>
<td>2.7 ± 1.3</td>
<td>3.3 ± 1.3</td>
<td>0.05**</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Factors of altered MetS</th>
<th>Exposure</th>
<th>Non-exposed</th>
<th>Exposed</th>
<th>p††</th>
</tr>
</thead>
<tbody>
<tr>
<td>AC ≥80 cm</td>
<td>31</td>
<td>91.2</td>
<td>33</td>
<td>97.1</td>
</tr>
<tr>
<td>Triglycerides ≥ 150 mg/dL</td>
<td>13</td>
<td>38.2</td>
<td>17</td>
<td>50</td>
</tr>
<tr>
<td>HDL-c&lt; 50 mg/dL</td>
<td>26</td>
<td>76.5</td>
<td>28</td>
<td>84.8</td>
</tr>
<tr>
<td>SBP ≥ 130 mmHg</td>
<td>9</td>
<td>26.5</td>
<td>16</td>
<td>47.1</td>
</tr>
<tr>
<td>DBP ≥ 85 mmHg</td>
<td>12</td>
<td>35.3</td>
<td>17</td>
<td>50</td>
</tr>
<tr>
<td>Fasting glucose ≥ 100 mg/dL</td>
<td>10</td>
<td>29.4</td>
<td>14</td>
<td>41.2</td>
</tr>
</tbody>
</table>

Table 4: Components and characteristics of Metabolic Syndrome in women with and without a history of preeclampsia – Average follow-up of 14 years MEAC-UFC, Fortaleza, 2012

†† Different statistical tests were used in the analysis: ** Mann-Whitney; † Test X2; †† Fisher test.

MetS=Metabolic Syndrome; AC=Abdominal Circumference; SBP=Systolic Blood Pressure; DBP= Diastolic Blood Pressure; HDL=High-Density Lipoprotein.

The majority (67.7%) of patients were classified as having metabolic syndrome, and there was a higher prevalence in patients with a history of preeclampsia, with a RR of 4.1 (CI 95% 1.4 to 12.2, p=0.009). There was higher frequency of alteration
of the variables for AC (94.1%) and HDL cholesterol (80.1%) in the total population.

Discussion

Main findings

An unfavourable metabolic profile was found among the patients evaluated, with changes in the anthropometric characteristics of central obesity, a major cardiovascular risk factor, and the biochemical measurements that contribute to aggravating this risk, with the majority characterized as having MetS. There was a higher frequency in the group of patients with a history of preeclampsia.

Both groups were alike in their baseline characteristics at the start of the follow up in terms of age, the number of pregnancies, parity, occurrence of miscarriages and smoking status, factors that may influence the outcomes being assessed and demonstrate a recognized association with CVD [4,12,13].

Physical exercise was one important variable analyzed as a possible contributing factor to the composition of MetS, without a difference between groups. There was a low frequency of physical activity in the sample studied, comparable to the results of Ekelund et al. [14] in which the majority of women (59.1%) did not do physical activity or were moderately inactive. The high SBP averages found this study are consistent with the results of other studies that show higher rates of this measurement in women with a history of GHS [15-17].

The AC and WC measurements had averages above the amount considered appropriate; however, this difference was not significant between groups. When analyzing the biochemical blood findings, there was an unfavourable lipid profile, in which all the measurements were altered more frequently in the exposed group. The mean HDL-C values were lower than the amounts recommended as ideal parameters for the female population; this was the second most altered measurement among the exposed patients (84.8%). The presence of factors that characterize metabolic syndrome according to the IDF [5] was evaluated and there were an excessive number of altered values in both groups. In this study, most patients (67.7%) were characterized as having MetS. An analysis of components of MetS showed that AC, an obligatory factor for a classification of MetS, and HDL-C, was one of the most frequent alterations.

Strengths and limitations

It must be emphasized that in the present study, patients with other diagnoses of GHS, such chronic hypertension and preeclampsia superimposed on chronic hypertension were excluded, thereby seeking to evaluate the influence of preeclampsia as a specific disorder of pregnancy predisposing long term metabolic changes.

It is still uncertain whether preeclampsia predisposes women to future CVD mediated by MetS or if it is a manifestation of a subclinical susceptibility to future MetS that predisposes women to preeclampsia [18], which methodologically, cannot be evaluated through retrospective cohort studies, since many factors such as BMI, lipid and glycemic profile analysis and pre-pregnancy anthropometric values are required, which was not possible in this study.

Although some authors discuss the validity of the characterization of MetS, several studies have demonstrated the value of this classification and its association with the occurrence of future cardiovascular events, thus it is important to consider this syndrome when assessing patients who may have differentiated future cardiometabolic risk factors, such as women with an adverse obstetric history [19].

It may also be considered that the average age of the patients analyzed in this study (39.8 ± 8.2 years) is considered relatively young and may explain why greater changes have not manifested themselves yet, as postulated by Rich-Edwards [20]. Even with the small sample size, significant differences could be found in the profile of morbidities among the group of patients analyzed, showing the need for further evaluation of patients with preeclampsia that also involves the subclinical conditions that may characterize a future adverse cardiovascular profile, so that preventive measures can be taken early, reducing the complications associated with these changes.

Interpretation

Previous studies, with shorter follow-up periods, had already demonstrated this association but no study had ever addressed this assessment in a period over 10 years. Several studies have had longer follow ups for other outcomes, but not for metabolic syndrome [16,18]. A study by Pouta et al. (2004) presented the results of this evaluation with a pregnancy-index range interval up to an evaluation of five months to 11 years [21]. Studies show that women with a history of gestational hypertension syndrome (GHS) tend to have higher frequencies of hypertension, diabetes and use of antihypertensive and hypoglycemic medications, however, the differences between the groups for these variables has not been identified [22-24].

A study by Callaway et al. [25], found that among 191 women with a history of GHS, 32.5% were hypertensive 21 years after delivery, less than in the present study, which showed a frequency of SAH of 47.1% in exposed patients. Studies point to different relative risks for the occurrence of future hypertension in women with a history of GHS, depending on the severity of the condition. For women with a history of gestational hypertension (GH), the RR found by Wilson et al. [24] was 2.47, whereas for preeclampsia/eclampsia, the RR was 3.89.

The profile of the high frequency of changes in anthropometric and metabolic variables are in line with results of other studies, such as Canti et al. [15] that, when analyzing 40 women with a history of preeclampsia and 14 with a history of normotensive pregnancies after a follow-up period of 14.6 years for the exposed group and 15.9 years for the unexposed group, found increased BMI, WC and DBP values in women with a history of PE, with measurements consistent with the characterization of high cardiometabolic risk [8].

Studies show that overweight and obesity represent a serious health threat and are strongly associated with an increased risk of cardiovascular disease, type 2 diabetes mellitus and metabolic disorders [26]. A study conducted by Forest et al. [16] evaluated 168 pairs of women recruited between 1989 and 1997, 7.8 years after labour, 105 with a history of GH and 63 with a history of PE who were compared with 168 controls. They found higher BP averages in patients with a history of GHS and the SBP was equal to 115 mmHg, lower than that shown by the patients in the present study (131.0 ± 23.8 mmHg).

Jie et al. [18] analyzed the prevalence of MetS in 62 women one to three years after a preeclamptic pregnancy. They found that 39% of these women met the IDF criteria for this condition, lower than the findings in the present study. The authors found that abdominal obesity, high blood pressure and decreased HDL-c were the most

altered factors in women with history of preeclampsia, whereas fasting glucose was less altered.

Conclusion
An unfavorable clinical and metabolic profile was found, with the patients with a history of preeclampsia having a higher prevalence of MetS diagnoses and a tendency for a higher number of altered components for a characterization of MetS.

Practical recommendations
Reproductive factors are rarely taken into consideration when evaluating an individual’s risk for metabolic diseases. However, with an increasing body of evidence indicating that may exist a strong relationship between these conditions, further clinical consideration is required through increased monitoring, risk stratification, and follow-up of individuals with such adverse reproductive histories such as preeclampsia.

Researches recommendations
More research is needed to understand the biological etiology underlying these relationships, including genetic and epigenetic mechanisms, and analyzing larger samples.

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Contribution to Authorship
ACPTH: conception and design of the protocol, acquisition of data, drafting the article approval of the final version; FHCC: conception and design of the protocol, drafting the article, acquisition of data, and approval of the final version; HNF: drafting the article and approval of the final version; JCGB: acquisition of data, drafting the article and approval of the final version; LRMP: acquisition of data, drafting the article and approval of the final version; FELF: drafting the article and approval of the final version.

Details of Ethics Approval
The study was approved on August 30, 2011 by the MEAC Ethics Committee under Opinion No. 83/2011. All women who participated in the study did so voluntarily, having given their informed written consent and received all results of the evaluations.

References