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Relation Between Insulin Resistance and Hematological Parameters in a Brazilian Sample

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ABSTRACT

It has already been demonstrated that insulin resistance (IR) is associated with the stimulation of erythroid progenitors and with increased levels of inflammation markers. Therefore, IR should also be associated with increased red blood cell (RBC) and white blood cell (WBC) count. The aim of this study is to demonstrate that IR is independently associated with altered hematological parameters in a Brazilian sample. We analyzed laboratorial exams from 925 subjects. All data on hematological parameters, insulin resistance (Homeostasis Model Assessment (HOMA)) and lipid levels were included in the analysis. Demographic information included age and gender. HOMA correlated positively with RBC (r= 0.17, p< 0.001), plasma hemoglobin concentrations (r= 0.14, p< 0.001), hematocrit value (r= 0.15, p< 0.001) and WBC (r= 0.17, p< 0.01). Subjects in the upper quartile of IR had higher levels of plasma glucose, fasting insulin, triglycerides, hematocrit, hemoglobin, RBC and WBC count than those in the lower quartile. In conclusion, IR seems to be associated with alterations in several hematological parameters. These hematological alterations may be considered an indirect feature of the IR syndrome. **(Arq Bras Endocrinol Metab 2006;50/1:114-117)**

Keywords: Insulin resistance; Hyperinsulinemia; Hemoglobin; Red blood cells; White blood cells

RESUMO

Relação Entre Resistência à Insulina e Parâmetros Hematológicos em Uma Amostra da População Brasileira.
Já foi demonstrado que a resistência à insulina (RI) está associada com a estimulação de progenitores eritrocitários e com níveis aumentados de marcadores inflamatórios. Desta maneira, a RI pode também estar associada o aumento de hemácias e leucócitos. Esse estudo objetivou demonstrar que a RI está independentemente associada com a alteração de parâmetros hematológicos em uma amostra da população brasileira. Nós analisamos exames laboratoriais de 925 indivíduos, incluindo todos os dados de parâmetros hematológicos, resistência insulínica (*homeostasis model assessment* - HOMA) e níveis lipídicos. Informação demográfica incluiu idade e gênero. HOMA correlacionou-se positivamente com a contagem de eritrócitos (r= 0,17; p<0,001), a concentração de hemoglobina (r= 0,14; p<0,001), o hematócrito (r= 0,15; p<0,001) e a contagem de leucócitos (r= 0,17; p<0,01). Indivíduos no quartil superior de RI tinham níveis mais elevados de glicemia, insulina de jejum, triglicérides, hematócrito, hemoglobina e contagem de leucócitos e eritrócitos do que aqueles no quartil inferior. Em conclusão: a RI parece estar associada com alterações em diversos parâmetros hematológicos. Essa alterações hematológicas podem ser consideradas um achado indireto da síndrome de resistência insulínica. **(Arq Bras Endocrinol Metab 2006;50/1:114-117)**

Descritores: Resistência insulínica; Hiperinsulinemia; Hemoglobina; Eritrócitos; Leucócitos.

THE METABOLIC SYNDROME (MS) is currently described as the association of insulin resistance with obesity (mainly visceral), hypertension, and dyslipidemia (increased triglycerides and/or decreased HDL-cholesterol). According to WHO criteria (1), insulin resistance (IR) is the main determinant of the syndrome and hyperinsulinemia, its major clinical expression, is directly related to all other minor criteria. Patients with MS are at increased risk for cardiovascular death and hyperinsulinemia seems to be an independent factor for a cardiovascular event (2).

The association between hyperinsulinemia and cardiovascular disease is partially explained by the effects of insulin on cell growth. Insulin has been shown to promote growth of vascular cells and consequently to induce atherosclerosis. Moreover, several authors have already demonstrated that insulin also regulates erythropoiesis *in vitro* (3,4). Recently, it was suggested that the effects of hyperinsulinemia in erythroid progenitors could also lead to an increase in red blood cell (RBC) count (5). Therefore, the alterations in hematological parameters could be included as a new and indirect feature of the IR. We aimed at demonstrating that IR is independently associated with alterations in hematological parameters and dyslipidemia in a sample of the Brazilian population.

PATIENTS AND METHODS

For the purpose of this study, we analyzed the results of laboratorial exams from 1,045 subjects sequentially selected between 18 and 80 years old. All patients had a blood sample collected at a private laboratory (Laboratório Sérgio Franco) to perform exams for the evaluation of haematological parameters, lipid profile and insulin resistance. One hundred and twenty one patients were excluded: 112 subjects because of fasting glucose levels higher than 126 mg/dl (i.e. Diabetes Mellitus) and 9 subjects because of insulin levels higher than 100 μUI/ml. The final analysis included 223 men and 702 women with a mean age of 43.8 ± 15.3 years old. The protocol was approved by the Ethics Committee of the State Institute of Diabetes and Endocrinology (IEDE) and by the Execute Board of Laboratório Sérgio Franco.

Venous blood samples were collected after a 12 hr fasting period. Blood glucose was measured by an enzymatic colorimetric assay using a glucose oxidase method and plasma insulin by a commercial double-antibody, solid phase radioimmunoassay. Commercial enzymatic tests were used for determining serum total- and HDL cholesterol and triglyceride concentrations. Serum LDL cholesterol concentrations were calculated by the Friedwald formula (6). Red blood cells (RBC) count, white blood cells (WBC) count, haemoglobin concentration and hematocrit were done using a haematology autoanalyzer.

The estimate of insulin resistance by homeostasis model assessment (HOMA) was calculated with the formula: [fasting serum insulin (μUI/ml) x fasting plasma glucose (mg/dl) x 0.0551] / 22.5 (7).

Statistical analysis was performed with GraphPad InStat 3.00 for Windows 95 (GraphPad Software, San Diego, California, USA). The strength of the linear relationship between two continuous variables was evaluated by means of the Spearman's rank order correlation coefficient. Multivariate linear regression analysis was used to test the independent association of age, gender and HOMA with the hematological parameters. The level of statistical significance was 5%.

RESULTS

In the whole group, HOMA was positively associated with plasma concentrations of triglycerides ($r= 0.36$, $p< 0.001$), total cholesterol ($r= 0.08$, $p= 0.01$) and negatively associated with concentrations of HDL-cholesterol ($r= -0.27$, $p< 0.001$). After adjustment for age, however, the association between HOMA and total cholesterol was not significant ($p= 0.17$).

A positive correlation between HOMA and the main hematological variables was also found. HOMA correlated with RBC count ($r= 0.17$, $p< 0.001$), plasma hemoglobin concentrations ($r= 0.14$, $p< 0.001$), hematocrit value ($r= 0.15$, $p< 0.001$) and WBC count ($r= 0.17$, $p< 0.01$). All these associations were still significant after adjustment for age (data not shown). HOMA was not associated with the number of platelets ($r= 0.02$, $p= 0.52$). Table 1 shows the correlation between HOMA and the main hematological parameters according to gender.

By dividing subjects into quartiles of HOMA, it was shown that subjects in the upper quartile had higher levels of plasma glucose, fasting insulin, triglycerides, hematocrit, hemoglobin, RBC and WBC count than those in the lower quartile. Levels of fasting insulin, triglycerides and RBC count were found to be even higher than those in the third quartile (table 2).

Table 1. Correlation between insulin resistance and the main hematological parameters according to gender.

	Male (n= 223)		Female (n= 702)	
HOMA and RBC count	r= 0.08	p= 0.19	r= 0.13	p< 0.01
HOMA and hemoglobin	r= 0.06	p= 0.32	r= 0.078	p= 0.038
HOMA and hematocrit	r= 0.12	p= 0.07	r= 0.10	p= 0.007
HOMA and WBC count	r= 0.15	p= 0.02	r= 0.16	p< 0.001
HOMA and platelets	r= -0.07	p= 0.29	r= 0.09	p= 0.01

HOMA= Homeostasis Model Assessment; RBC= Red Blood Cell; WBC= White Blood Cell

Table 2. Quartiles of insulin resistance.

	1st quartile (< 1.44)	2nd quartile (1.44–2.35)	3rd quartile (2.35–3.90)	4th quartile (> 3.91)
IR (HOMA)	0.93 ± 0.33	1.89 ± 0.26[a]	3.09 ± 0.45[a,b]	6.57 ± 2.83[a,b,c]
Glucose (mg/dl)	87.5 ± 9.4	92.0 ± 10.2[a]	94.7 ± 11.1[a]	99.1 ± 11.5[a,b,c]
Insulin (µU/ml)	4.3 ± 1.5	8.5 ± 1.4[a]	13.5 ± 2.6[a,b]	27.2 ± 11.7[a,b,c]
Chol. (mg/dl)	195.4 ± 44.2	192.6 ± 39.0	196.9 ± 34.4	198.8 ± 40.3
HDL-Chol. (mg/dl)	60.4 ± 16.8	53.3 ± 12.6[a]	52.5 ± 12.5[a]	48.9 ± 12.2[a,b,c]
Triglyc. (mg/dl)	94.0 ± 60.3	108.4 ± 62.1[a]	129.2 ± 76.7[a,b]	156.5 ± 91.2[a,b,c]
RBC (10^6/ml)	4.5 ± 0.4	4.6 ± 0.4	4.6 ± 0.4[a]	4.7 ± 0.4[a,b,c]
WBC (10^3/ml)	6.3 ± 1.8	6.3 ± 1.6	6.7 ± 1.9	7.0 ± 1.7[a,b]
Haemoglobin (g/dl)	13.6 ± 1.2	13.8 ± 1.3	13.9 ± 1.2	14.1 ± 1.4[a]
Hematocrit (%)	41.3 ± 3.3	41.8 ± 3.7	42.3 ± 3.4[a]	42.7 ± 3.9[a]
Platelet – 10^3/ml	257.4 ± 60.3	250.3 ± 52.6	262.4 ± 59.3	257.2 ± 63.2

IR= Insulin Resistance; HOMA= Homeostasis Model Assessment; Chol.= Cholesterol; Triglyc.= Triglycerides; RBC= Red Blood Count; WBC= White Blood Count. Data are means ± SD.
[a] p< 0.01 x 1st quartile; [b] p< 0.01 x 2nd quartile; [c] p< 0.01 x 3rd quartile.

DISCUSSION

We investigated whether IR is associated with dislypidemia and hematological parameters. To reach this goal, we systematically evaluated the results of biochemical exams from 925 subjects. Our most significant findings were the following: (1) IR presented a positive correlation with all hematological parameters (except platelets), especially in women; and (2) patients at the higher quartile of IR had higher levels of cardiovascular risk factors than those subjects at the lower quartile.

Hyperinsulinemia seems to exert its effects in erythropoiesis through different mechanisms. The presence of the insulin receptor (INS-R) in human erythropoietic cells during all stages of development suggests that insulin acts as a co-factor in erythropoiesis (8). Indeed, increased haematological parameters (i.e. polycythaemia) observed in newborn babies of diabetic mothers support the relationship of hyperinsulinemia and erythropoiesis *in vivo* (9). Furthermore, several authors have also demonstrated the growth-promoting effects of insulin in erythropoietic cells *in vitro* (3,4). Also, hyperinsulinemia seems to increase concentrations of hypoxia-inducible factor-1 alpha (HIF-1 alpha). HIF-1 alpha promotes the synthesis of erythropoietin and may also mediate intestinal iron absorption (10). Taken together, these mechanisms may help to explain the relation between IR and the erythropoietic parameters.

The increase in WBC and RBC count associated with IR may contribute to the increased cardiovascular mortality related to the MS. Blood viscosity is regulated by several factors, including the number of both white and red blood cells. The effects of insulin in erythropoiesis may lead to an increase in blood viscosity and to altered circulatory kinetics. Indeed, blood viscosity has already been shown to be an independent risk factor for stroke and myocardial infarction (11).

It has been suggested that the MS presents several features of an inflammatory disease. Several cytokines (e.g. TNF-α, IL-6) were positively associated with IR and with the formation of the atherosclerotic plaque. Moreover, other inflammation markers, such as C Reactive Protein, have recently been associated with cardiovascular disease and cardiovascular mortality (12). The association of IR with increased WBC count may provide further evidence for all those who believe that chronic inflammation is part of the

MS. WBC are an element necessary for plaque formation and growth. Therefore, increased WBC may reflect the inflammatory activation related to the MS.

In our study, subjects in the higher quartile of IR showed significantly higher levels of several independent cardiovascular risk factors. Insulin resistance was associated with higher levels of triglycerides, RBC and WBC count, hemoglobin, hematocrit and lower levels of HDL cholesterol than subjects at the lowest quartile. Although the relation among IR, dyslipidemia and cardiovascular morbidity is widely accepted, the inclusion of the hematological parameters as risk factors strengths the necessity of a more detailed approach of the patients with the MS. Further studies, however, are necessary to clarify the independent impact of the hematological parameters in cardiovascular morbidity and mortality.

Our study has some limitations. First, we could not include anthropometrical measurements (i.e. waist, waist-to-hip ratio, body mass index [BMI]) in the statistical analysis. Therefore, we could not investigate whether the hematological parameters were influenced by weight excess instead of IR. However, previously published reports demonstrated that the relation between IR and the hematological parameters persisted even after adjustment for waist and BMI (5,13,14). Second, we could not exclude patients who were under treatment with drugs that might interfere with erythropoiesis or with lipid levels. We believe that the strength of the relation would not be changed with the exclusion of these patients. Finally, it remains to be determined the reason why the relationship between IR and the haematological parameters (except for WBC count) could not be demonstrated in men. We believe that the small sample may be partially responsible for these findings. Further studies are necessary to clarify this issue.

In conclusion, IR seems to be associated with increased white and red cell count, hemoglobin and hematocrit. Hyperinsulinemia may increase erythropoiesis and consequently increase blood viscosity. The alterations in hematological parameters induced by IR may be partly responsible for the increased cardiovascular mortality related to the MS. Controlled studies are necessary to clarify the impact of the treatment of IR in the hematological parameters.

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