Turkish Journal of Endocrinology and Metabolism, (2002) 3 : 101-104 **ORIGINAL ARTICLE**

Correlation Between Insulin-Receptor Binding and Insulin Resistance Measured By the Homeostasis Model Assessment

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The study was designed to investigate the correlation between insulin-receptor binding and insulin resistance in obesity, type 2 diabetes mellitus and in a group of healthy normal-weight subjects. 121 subjects were enrolled in the study - 32 subjects with different degrees of obesity (mean age 44.1±12.1 years); 43 newly-diagnosed type 2 diabetic patients (mean age 49.8±9.5 years) and 46 healthy controls (mean age 47.7±10.8 years). Insulin-receptor binding was studied on circulating mononuclear blood cells. Results are presented as the number of total and high-affinity receptors per cell and receptor affinity. Insulin resistance was estimated using the homeostasis model assessment (HOMA-IR). We found a significant negative correlation between the total number of insulin receptors per cell and HOMA-IR ($r=-0.71$, $p<0.0001$) and the number of high-affinity insulin receptors and HOMA-IR ($r=-0.61$, $p=0.001$) and no correlation between receptor affinity and HOMA-IR ($r=0.07$, $p>0.1$) in the whole study population. When analysing the groups separately we found the strongest correlation between insulin receptors and HOMA-IR in the obese subjects ($r=-0.84$, $p<0.0001$) compared with the type 2 diabetic patients ($r=-0.58$, $p=0.001$) and the healthy controls ($r=-0.51$, $p=0.001$). Our results demonstrate that there is a significant correlation between the number of insulin receptors, measured on mononuclear blood cells, and insulin resistance estimated by HOMA index in type 2 diabetic patients, in obese subjects and in healthy controls.

Key words: Insulin receptor, insulin resistance, HOMA, type 2 diabetes, obesity

Introduction

Insulin resistance is present in a number of conditions, such as type 2 diabetes mellitus and obesity. It can be due to prereceptor, receptor or post-receptor defects (1).

Type 2 diabetes is characterized by alterations in both insulin secretion and insulin sensitivity. The relative importance of the two defects is stil controversial. Studies on insulin-receptor binding have shown different results. Some authors have reported a decrease in insulin-receptor binding, due to a reduction in the number of insulin receptors (1,2), while others have not found any alterations at the receptor level and relate insulin resistance to postreceptor defects in insulin action (3,4).

Obesity is a heterogenous disorder and it is the most powerful risk factor for type 2 diabetes mellitus. Data concerning the relationship between obesity and fasting hyperinsulinemia have been demonstrated and a strong negative correlation between the number of receptors per cell and the fasting insulin level have been reported (5). So the question is raised as to whether hyperinsulinemia is

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the cause or the consequence of the reduced insulin receptors. In contrast to the authors, who have reported a decrease in insulin receptors (1,4,5), others have found only a dilution of receptors on the surface of the enlarged adipocytes, their number remaining unchanged. Another group of investigators have reported normal insulin-receptor binding in obesity and have attributed insulin resistance to a postreceptor defect in insulin action (6).

A lot of methods have been applied to estimate insulin resistance, the most frequently used recently being the homeostasis model assessment (HOMA). There is evidence that this method of assessment strongly correlates with the results achieved by means of the euglycaemic hyperinsulinemic clamp technique (r=-0.82), which appears to be the "gold standard" in estimating insulin resistance (7). This method has also been recommended in epidemiological studies as it is a reliable and easily performed one (8,9).

The aim of the present study was to evaluate the correlation between the parameters of insulin-receptor binding, measured on circulating mononuclear blood cells, and insulin resistance, estimated by the HOMA index, in healthy controls, obese subjects and newly-diagnosed type 2 diabetic patients.

Materials and Methods

121 subjects (69 males and 52 females) were enrolled in the study - 32 subjects with different degrees of obesity (mean age 44.1±12.1 years; mean BMI 34.7±4.9 kg/m^2); 43 newly-diagnosed type 2 diabetic patients (diagnosed according to World Health Organization criteria (10,11), of mean age 49.8±9.5 years; mean BMI 28.0±3.9 kg/m^2) and 46 healthy controls (mean age 47.7±10.8years; mean BMI 24.5±3.1 kg/m^2). According to the degree of obesity the obese subjects were divided into three groups: I degree (BMI 30.0-34.9 kg/m^2) (n=11) - mean age 40.1±7.8 years; mean BMI 32.5±2.4kg/m^2; II degree (BMI 35.0-39.9 kg/m^2) (n=11) - mean age 42.3±12.1 years; mean BMI 36.7±2.3 kg/m^2; III degree (BMI>40.0 kg/m^2) (n=10) - mean age 45.8±4.5 years; mean BMI 43.0±2.9 kg/m^2. The diabetic patients were divided according to their BMI into obese (n=23, mean age 43.3±10.1 years; BMI>30 kg/m^2, mean BMI 32.8±4.3 kg/m^2) and nonobese (n=20, mean age 48.1±9.8 years; BMI<30 kg/m^2, mean BMI 23.5±2.1 kg/m^2).

Insulin-receptor binding was studied on circulating mononuclear blood cells, isolated according to the method of Boyum (12), by incubating them with A14-monoiod [^{125}I]-insulin (Amersham, 0.2 ng/ml) and increasing concentrations of unlabelled insulin (NovoNordisk, from 0 to 10^5 ng/ml) in Hepes (pH 7.8) (13). Data were corrected for nonspecific binding (in the presence of 10^5 ng/ml unlabelled insulin) and were analysed by a computer program, based on mathematical analysis of a Scatchard curve (14,15). Results are presented as the number of total and high-affinity insulin receptors per cell and insulin receptor affinity. Insulin receptor affinity was estimated on the basis of the association constant (K_a).

Insulin resistance was estimated using the homeostasis model assessment (HOMA) index, according to the formula: HOMA-IR = plasma glucose (mmol/l) x fasting serum insulin (mU/L) /22.5 (16).

Plasma glucose was measured with a Glucose Analyzer (Beckman), and serum insulin level by RIA (Hungarian kit) (reference range 5-25 mU/L).

All the patients gave their written consent to participate in the study after full explanation of the nature of the study according to the Helsinki Declaration.

Statistical analysis was performed with a SPSS 9.01 package. Repeated measures analysis of variance and linear regression and correlation analysis were used. Results are presented as means ± SEM.

Results

The parameters of insulin-receptor binding - total number of insulin receptors per cell, number of high-affinity receptors per cell and receptor affinity of the different groups as well as HOMA-IR are presented in Table 1. We have compared the parameters of insulin-receptor binding between the groups with different degrees of obesity and the results are presented in Table 2.

Table 1. Parameters of insulin receptor binding of the different groups (total number of insulin receptors, number of high-affinity receptors and insulin receptor affinity). Values are means±SEM.

Parameter/Group	Control group (n=46)	Obese subjects (n=32)	Type 2 diabetic patients (n=43)
Total number of receptors	13543 ± 465	6767 ± 298*	6243 ± 311*
Number of high-affinity receptors	382 ± 95	243 ± 64*	259 ± 79*
Insulin receptor affinity	2.82 ± 0.52	3.26 ± 0.78	2.67 ± 0.7
HOMA-IR	2.04 ± 0.8	7.4 ± 2.4*	6.8 ± 2.1*

* p<0.0001 as compared to the healthy controls

Table 2. Parameters of insulin receptor binding in the subjects with different degrees of obesity (total number of insulin receptors, number of high-affinity receptors and insulin receptor affinity). Values are means±SEM.

Parameter/Group	Control group (n=46)	I degree obesity (n=11)	II degree obesity (n=11)	III degree obesity (n=10)
Total number of receptors	13543 ± 465	8603 ± 276**	5661 ± 199**#	5063 ± 218**#
Number of high-affinity receptors	382 ± 95	286 ± 97**	197 ± 67**#	175 ± 82**#
Insulin receptor affinity	2.82 ± 0.52	3.51 ± 0.46*	3.89 ± 0.5*	2.31 ± 0.7

* p<0.05 as compared to the healthy controls
** p<0.0001 as compared to the healthy controls
# p<0.001 as compared to I degree obesity

Discussion

There is a significant decrease in the total number of insulin receptors and the high-affinity receptors per cell in type 2 diabetic patients (p<0.0001) as well as in obese subjects (p<0.0001) compared with healthy controls. The difference between the obese subjects and the diabetic patients was not significant (Table 1). Type 2 diabetes and obesity are characterized by alterations in both insulin sensitivity and insulin secretion. We have estimated insulin resistance on the basis of the fasting plasma glucose and insulin concentration, using the HOMA index. Our results demonstrate that insulin resistance is significantly higher in newly-diagnosed type 2 diabetic patients (6.8±2.1 vs 2.04±0.8 mmol/l. mU/L, p<0.0001) and in the group of obese nondiabetic subjects (7.4±2.4 vs 2.04±0.8 mmol/l. mU/L, p<0.0001) as compared with the healthy normal-weight controls; the difference between diabetic patients and obese subjects being not significant (p>0.1) (Table 1). It appears that this insulin resistance is at least partly due to the alterations at the level of insulin-receptor binding. We have studied insulin receptor binding on circulating mononuclear blood cells as there is evidence that the insulin binding characteristics of these cells are similar to those of the target cells (11). We have found a significantly lower number of the total (p<0.0001) and high-affinity receptors per cell (p<0.0001) in the newly diagnosed diabetic patients as compared with the healthy controls; their receptor affinity being similar to that of the control group (p>0.1) (Table 1). Obese subjects also present a significantly lower number of both total and high affinity receptors per cell as compared with the control group (p<0.0001), and rather similar parameters to those of the newly-diagnosed diabetic patients (p>0.1). The subjects with I and II degree of obesity demonstrate a significant decrease in the number of insulin receptors per cell - both of the total number and the number of high-affinity receptors as compared with the healthy controls (p<0.0001). Nevertheless their receptor affinity is higher (p<0.05), thus compensating for the lower receptor number. In the group of extremely obese subjects (BMI>40kg/m^2) there is a significant decrease in the total number of insulin receptors per cell as well as in the number of high-affinity receptors per cell (p<0.0001). In contrast to the other degrees of obesity, their receptor affinity doesnot differ from that of the healthy controls, thus not compensating for the decreased receptor number (Table 2). These results resemble strongly the parameters of insulin receptor binding of the newly-diagnosed type 2 diabetic patients. Probably the compensatory mechanism of increase of receptor affinity is lost in very obese subjects, thus exposing them at higher risk for the development of diabetes.

When dividing the type 2 diabetic subjects into obese and nonobese, we found no significant difference in the total number and number of high-affinity receptors per cell as well as in the receptor affinity between the two groups ($p>0.1$); showing that diabetes but not body weight is responsible for their insulin receptor alterations. The aim of the present study was to correlate insulin receptor binding on circulating mononuclear blood cells (which was performed a number of years ago) with HOMA-IR index. We have found a strongly significant negative correlation between the total number of insulin receptors per cell of all the studied subjects ($n=121$) and insulin resistance, measured by the HOMA-IR ($r=-0.71$, $p<0.001$) as well as between the number of high affinity insulin receptors per cell of all the subjects and HOMA-IR ($r=-0.61$, $p=0.001$). There is no correlation between insulin receptor affinity of the studied subjects and HOMA-IR ($r=0.07$, $p>0.1$). We have also searched for correlation between the total number of insulin receptors per cell and HOMA-IR of the three different groups. The strongest correlation exists in the group of the obese nondiabetic subjects- $r=-0.84$, $p<0.0001$, followed by the group of newly-diagnosed type 2 diabetic patients - $r=-0.58$, $p<0.001$ and the group of healthy controls - $r=-0.51$, $p<0.001$. There could also be postreceptor defects in both type 2 diabetes and obesity, contributing to the insulin resistance, but the established significant correlation between insulin-receptor binding and insulin resistance proves the reliability and specificity of the two methods used - insulin receptors on mononuclear blood cells and insulin resistance (HOMA-IR).

In conclusion, the results from this study show that there is a significant negative correlation between insulin resistance measured by the homeostasis model assessment (HOMA) index and the total number of insulin receptors and the number of high-affinity receptors per cell, measured on circulating mononuclear blood cells in healthy controls, subjects with different degrees of obesity and newly-diagnosed type 2 diabetic patients; the strongest correlation being found in the obese group; in all the groups the correlation being significant.

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