

The Relevance of Adiponectin and Resistin Levels with Oxidative Stress in Insulin Resistant Type 2 Diabetes

العلاقة بين مستويات الأديبونكتين والريزستين مع الإجهاد التأكسدي في مرضى السكري من النوع الثاني الغير مستجيب للأنسولين

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الخلاصة :

الهدف : تم تصميم هذه الدراسة للتحقق من التغييرات لمستويات (اديبونكتين (ADN)، ريزستين) والإجهاد التأكسدي (MDA) مع مستويات مقاومة الأنسولين لدى مرضى السكري النوع الثاني .

المنهجية : ولتحقيق هذا الهدف أختيرت العينة من المرضى المسجلين في تحقيق هذا الهدف حيث كانت عينة البحث تتكون من 60 مريضاً مع مرضى السكري من النوع الثاني (24 من الذكور و 36 من الإناث) من الذين تتراوح أعمارهم (53.9 ± 12.4) ، بالإضافة إلى 30 شخصاً غير مصابين (13 من الذكور و 17 من الإناث)، تم قياس تركيز السكر في الدم لجميع العينة وكانوا جميعهم (صيام) ، الدهون الثلاثية، والأنسولين، اديبونكتين، ريزستين والمستويات الإجهاد التأكسدي (MDA). وقد تم قياس تركيز الأنسولين، واديبونكتين ريزستين بواسطة جهاز (ELISA)، وتم قياس مستوى كلوكوز الدم ومستويات الدهون الثلاثية بالطرق الأنزيمية. وتم قياس مستوى Malondialdehyde (MDA) بطريقة spectrophotometric. تم تقييم مقاومة الأنسولين بواسطة أربع طرق. وهي تشمل على طريقة (HOMA)، و (QUICKI)، و (MAC) وطريقة (FI).

النتائج : وأشارت النتائج إلى أن من بين عينة البحث لمرضى (60) ، 49 (81%) ، 45 (75%) ، 33 (55%) و 32 (53%) كانت مقاومتهم عدم الاستجابة للأنسولين عندما تم تقييمهم من قبل الطرق الأربعة (HOMA ، QUICKI ، MCA ، FI) على التوالي . وجرى تقييم مقاومة الأنسولين لـ (49) مريضاً بالسكري من نوع الثاني (IRP) التي تم الحصول عليها من خلال أسلوب HOMA للتغيير في مستويات اديبونكتين، ريزستين و (MDA). وكشف تحليل البيانات عن انخفاض كبير في المجموعة IRP عند مقارنتها مع تلك التي حصلنا عليها من المرضى الذين لديهم تحسس للأنسولين (ISP) والغير المرضى.

الاستنتاجات : لوحظ ارتفاع كبير في مستويات الريزستين في المجموعة IRP فيما يتعلق بتلك المراقبة وأظهرت مستويات MDA ارتفاعاً معنوياً في المجموعة IRP عند مقارنتها مع مجموعة ISP والغير المصابين . تم العثور على تغييرات في مستوى اديبونكتين، ريزستين و (MDA) لتكون مستقلة عن جنس المرضى . وقد لوحظ أن للوزن (BMI) تأثير في تغيير مستويات الاديبونكتين، الريزستين و (MDA). كما ولوحظ ان عمر المريض لم يسجل تأثير على مستويات الاديبونكتين، الريزستين و (MDA).

ولقد وجد أن العلاقة ما بين مقاومة الأنسولين مع مستوى ريزستين و (MDA) تغيراً إيجابياً وبشكل ملحوظ وعكسياً مع مستويات اديبونكتين لدى المرضى المصابين المقاومين للأنسولين لوحظ ان علاقة اديبونكتين علاقة عكسية مع ريزستين وعلاقة عكسية مع (MDA) ، في حين أن مستوى ريزستين مع (MDA) أظهر علاقة إيجابية لدى مرضى السكري من النوع الثاني المقاوم للأنسولين.

التوصيات : دراسة تأثير الأحماض الشحمية على فعالية الأنزيمات المسؤولة عن تحطيم الأنسولين لدى الأشخاص البدنيين .

Abstract:

Objectives: The present study was designed to verify the changes of adiponectin and resistin with oxidative stress in insulin resistant in diabetes mellitus type 2.

Methodology: To achieve this aim 60 patients with type 2 diabetes mellitus (24 male and 36 female) of ages, 53.9 ± 12.4 years in addition to 30 apparently healthy individuals (13 males and 17 females) were enrolled .

The concentration of fasting blood glucose , triglyceride , insulin , adiponectin, resistin and malondialdehyde (MDA) levels were measured. Fasting insulin concentration, adiponectin and resistin was measured by enzyme linked immunosorbent assay (ELISA) . Fasting blood glucose and triglyceride levels were determined by enzymatic methods. Malondialdehyde (MDA) levels were measured by spectrophotometric methods . Insulin resistance was evaluated by four methods .

Results : The results indicated that out of 60 patients , 49 (81%) , 45 (75%) , 33 (55%) and 32 (53%) were insulin resistant when they were evaluated by Homeostasis model assessment (HOMA) , Quantitative insulin sensitivity check index (QUICKI) , McAuley's index (MCA) and fasting insulin (FI) indices respectively . The 49 insulin resistant type 2 diabetic patients (IRP) that obtained through the HOMA method were assessed for the alteration in the levels adiponectin , resistin and MDA . The analysis of the data revealed a significant decrease of adiponectin in the IRP group when compared with those of the insulin sensitive patients (ISP) and the control groups . Significant elevation of resistin levels were observed in the IRP group with respect to those of the control groups .

Conclusion: Insulin resistance was found to be significantly positively correlated with resistin, MDA and significantly negatively correlated with adiponectin levels in diabetic patients . Adiponectin exhibited insignificant negative correlation with resistin and significant negative correlation with MDA levels, while the level of resistin showed a significant positive correlation with MDA level in insulin resistant type 2 diabetics .

Recommendations: A study on of the effect free fatty acid on the activity of insulin degrading enzymes in obese individual .

Key word: Diabetes patient, healthy group , blood parameter .

INTRODUCTION:

The adiponectin gene is located on chromosome in a region mapped as a susceptibility locus for type 2 diabetes and adiposity and is thought to potentially link obesity to insulin resistance. Adiponectin was first characterized in mice as a transcript over expressed in preadipocytes⁽¹⁾ (precursors of fat cells) differentiating into adipocytes⁽²⁾ .

Resistin is a cysteine-rich protein that was discovered in 2001⁽³⁾. Resistin is regarded as an important marker for obesity mediated insulin resistance and Type 2 diabetes mellitus⁽⁴⁾ .

Diabetes mellitus is a disease recognized more than 2000 years ago and is characterized by chronic hyperglycemia, excessive urine production with a compensatory thirst⁽⁵⁾ .

Type 2 is the most common form, accounts for 85-90% of all diabetes cases, and results from a combination of insulin resistance and impaired insulin secretion⁽⁶⁾ .

Diabetes mellitus is characterized by absolute or relative deficiencies in insulin secretion and/or insulin action associated with chronic hyperglycemia and disturbances of carbohydrate, lipid and protein metabolism⁽⁷⁾. Changes in human behaviour and lifestyle over the last century have resulted in a dramatic increase in the incidence of diabetes worldwide^(8, 9) .

Type 2 diabetes is rapidly becoming pandemic, and although the origin of this disease is not entirely clear, it is accepted that insulin resistance is important in its pathogenesis and that defects in insulin secretion by pancreatic β -cells lead to hyperglycemia and the onset of diabetes^(10,11) .

OBJECTIVES:

The main objectives in this theses can be summarized into the following points :

- 1- To determine the difference between serum levels of adiponectin, resistin and MDA in type 2 diabetic patients and the control group ;
- 2- To evaluate insulin resistance in type 2 diabetes mellitus;
- 3- To explore the relevance of insulin resistance with adiponectin, resistin and MDA in type 2 diabetic patients;.
- 4- To evaluate the correlation of adiponectin ,resistin and MDA levels in insulin resistant type 2 diabetic patients .

METHODOLOGY:

The study was conducted on 60 type 2 diabetic patients (24 male and 36 female) attending at diabetes mellitus center at Al-Sadder Teaching Hospital in al-Najaf province from December 2010 to April 2011.

A group of 30 healthy subjects (13 males and 17 females) were included as a control group. The ages of the apparently healthy individuals were ranged from (21-81) years. They were collected from my family , medical staff and relatives who were free from signs and symptoms of any chronic diseases like diabetes , hypertension, and others.

Venous fasting blood samples (5-6 ml) were collected from the patients and healthy volunteers after an over night fasting. The samples were put in tubes containing no

anticoagulant. Disposable syringes and needles were used for blood collection. After allowing the blood to clot at 37°C for about 15 min.

Determination of fasting blood glucose concentration(FBG) and Triglyceride concentration (TG) by enzymatic methods, Malondialdehyde(MDA) by spectrophotometric.

Insulin, adiponectin and resistin level in Patient and control group by (ELISA). Insulin resistance evaluate by four methods. "HOMA", "Quicki", "MCA", "FI".

RESULTS :

Table 1 : Level of fasting serum glucose, triglycerides, insulin, adiponectin , resistin and, MDA in patient and control groups

Parameter	Patients		Control		P value
	Mean±SD	Range	Mean±SD	Range	
Fasting blood glucose(mmol/L)	11.5±4.9	4.6-27.4	4.2±0.88	3.8-10.6	<0.001
TG(mmol/L)	2.4±1.2	1.2-7.8	1.3±0.5	0.28-2.25	<0.001
Insulin:µIU/ml	20.0±17.2	1.8-62.4	8.3±3.7	1.7-14.1	<0.01
ADN(µg/ml)	3.2±0.66	1.6-12.9	5.2±2.0	4.5-17.9	<0.001
Resistin(ng/ml)	17.1±2.6	10.5-23.8	15.2±3.6	9.8-19.85	0.005
MDA(µM)	13.15±4.5	5.3-23.1	10.1±4.2	2.6-19.51	<0.05

The results, Glucose, triglycerides, insulin, adiponectin, resistin and MDA increased significantly in diabetic patients when compared with control group . However, adiponectin significantly decreased during a comparable evaluation.

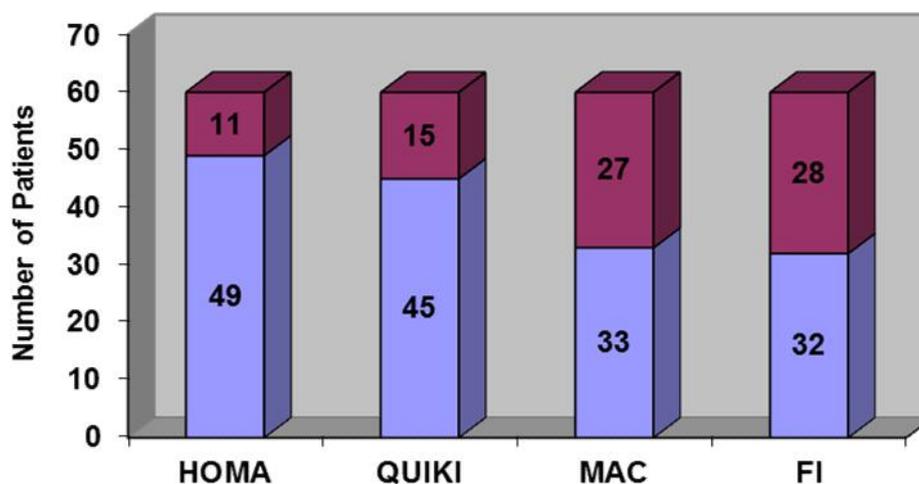


Fig 1: Insulin resistance (IR) among type 2 diabetes mellitus by indirect methods . The number of patients who are insulin resistance by HOMA ,QUIKI , MCA and FI indices were shown.

The result 32 of 60 (53%) patients were IR by FI, 33 out of 60(55%) are IR by MCA index . 45 out of 60 (75%) patients were IR by QUIKI index . 49 out of 60 (81%) were IR by HOMA index .

Table 2: The Incidence of insulin resistance and sensitivity in diabetic and the control groups.

Index	Insulin resistance subject		Insulin sensitivity subject	
	Patients	Control	Patient	Control
HOMA	49 (81%)	4 (13%)	11 (18%)	26 (87%)
QUICKI	45 (75%)	6 (20%)	15 (25%)	24 (80%)
MCA	33 (55%)	4 (13%)	27 (45%)	26 (87%)
FI	32 (53%)	8 (27%)	28 (46%)	22 (73%)

As shown in Table 2, 49(81%) , 45 (75%) , 33 (55%) and 32 (53%) of the 60 patients were demonstrated to be insulin resistant when they were evaluated by HOMA , QUICKI , MCA and FI indices respectively . However , 4(13%) and , 6 (20%) , 4 (13%) and 8 (27%) of the control group (30) with respect to the four methods of estimation of insulin resistance, revealed that the highest resistance was obtained with the use of HOMA index , while the lowest was indicated with the FI index.

Table 3 : Adiponectin, resistin and MDA levels in insulin resistant , insulin sensitive type 2 diabetic patients and the control groups .

	Groups	N	Mean± SD	Range	p value
ADN (µg/ml)	Patients(IRP)	49	3.1 ± 0.5	1.6 - 4	<0.001
	Patients(ISP)	11	4.9 ± 2.6	2.4 – 12.9	N S
	Control (IS)	26	6.77 ± 2.6	4.5 – 17.9	
	IRP vs. ISP				< 0.05
Resistin(ng/ml)	Patients(IRP)	49	17.6 ± 2.3	10.5– 23.8	<0.05
	Patients(ISP)	11	16.6± 3.3	10.5 – 20.5	N S
	Control (IS)	26	14.9 ± 0.7	9.8 – 19.8	
	IRP vs. ISP				NS
MDA(µM)	Patients(IRP)	49	12.6 ± 4.0	5.3 – 23.1	< 0.05
	Patients(ISP)	11	9.2 ± 2.2	6.38 – 15.05	N S
	Control (IS)	26	10.4 ± 4.5	2.6 – 19.5	
	IRP vs. ISP				< 0.05

The 2 groups of insulin sensitive and insulin resistant type 2 diabetic patients were compared with the insulin sensitive healthy subjects for the difference of the adiponectin, resistin and MDA . The 2 groups of insulin sensitive and insulin resistant type 2 diabetic patients were age-matched (p>0.05) .

The results pointed out a significant decrease of adiponctin levels in the IRP group when compared with those of the ISP (p<0.05) and the control (p<0.01) groups. Significant (p<0.05) elevation of resistin levels in the IRP group were found when compared with those of the control groups .

In addition, a significant rise(p<0.05) of MDA values was observed in IRP when compared with those of ISP and IS control groups.

DISCUSSION:

According to the results of the present investigation, the HOMA method was implicated to select diabetics of insulin resistance. Two factors have strongly led us to HOMA implication .

The first is the wide use of HOMA in the previous work mentioned in literatures. The second constitutes about 81% of the enrolled patients who were overweight or obese . Thus, the data of the HOMA method was highly suggestive to be used for selection of insulin resistant type 2 diabetic. So 49 out of 60 patients were categorized as insulin resistant and used in the present investigation .

The results in Table 1, Glucose, triglycerides, insulin, adiponectin, resistin and MDA increased significantly in diabetic patients when compared with control group . However, adiponectin significantly decreased during a comparable evaluation. These results are in agreements with those reported previously ⁽¹²⁾ .

In Table2. The significant decrease of adiponectin levels in insulin resistant type 2 diabetic may be due to interaction between genetic and environmental factors , such as a high-fat diet and sedentary life style. Such factors may play a crucial role in the development of insulin resistance, metabolic syndrome and diabetes mellitus⁽¹³⁾. The role of adiponectin as insulin sensitizer was evidenced in previous reports . It has been stated that low plasma adiponectin is an independent risk factor for the future development type 2 diabetes mellitus⁽¹⁴⁾ .

The measurement of resistin level indicated significant($p < 0.05$) increase when compared with those of the ISP and the control groups, Some studies have found significantly increased serum resistin concentration in association with development of insulin resistance and type 2 diabetes ⁽¹⁵⁾. Many studies have demonstrated a marked oxidative stress in diabetes mellitus as an expression of increased free radical production.⁽¹⁶⁾

On the other hand, lipid profile is altered in the serum of diabetic patients ⁽¹⁷⁾, where lipid peroxidation was evaluated by measuring serum levels of MDA. In agreement with other studies, a significant increase in MDA levels was observed in serum of diabetic patients when compared to normal control group ⁽¹⁸⁾. Excessive production of free radicals observed in diabetic patients and its insufficient removal results in damage to cellular proteins, membrane lipids, and nucleic acids. The presence of high levels of MDA in the present study may relate to the glycemic control.

CONCLUSIONS:

1. Most of type 2 diabetic patients are presented with insulin resistance.
2. Oxidative stress reflected by higher levels of MDA is involved in insulin resistance in type 2 diabetic patients .
3. The decreased levels of the anti-inflammatory Adipocytokine (adiponectin) and increase resistin are involved in insulin resistance in type 2 diabetes mellitus .
4. Changes of adiponectin and resistin associated the elevated oxidative stress in insulin resistance of type 2 diabetes.

RECOMMENDATION:

1. A study on the effect of free fatty acid on the activity of insulin degrading enzymes in obese individual .
2. A study on the effect of elevated cholesterol and triglyceride level on insulin activity.

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