Fetuin-A and type II diabetes mellitus

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Background
The pathophysiology of type II DM is complex; in addition to impaired insulin secretion from Beta-cells, reduced insulin sensitivity was found to play a predominant role in the pathogenesis of the disease. Fetuin-A is a hepatic secretory protein that binds the insulin receptor and inhibits insulin action both in vivo and in vitro.

Objective
Our aim was to investigate whether serum fetuin-A levels predict the incidence of insulin resistance in type II DM.

Patient and methods
The present study included 40 patients who had type II diabetes mellitus served as patients group and 40 apparently normal individuals served as control group. All patient and control groups were subjected to the following: full medical history and thorough physical examination, fasting & post prandial blood glucose, urea, creatinine, lipid profile, CRP, insulin and fetuin-A.

Results
There was highly significant increase in serum insulin, serum fetuin A and HOMA-IR in diabetic group compared with control group. There was significant positive correlation between serum fetuin A and serum insulin, FBG, HbA1c and serum CRP. Also a significant positive correlation between HOMA-IR and serum fetuin A, serum insulin and HbA1c were found.

Conclusion
We concluded that fetuin-A may play a role in the pathogenesis of type II DM, and high serum fetuin-A has a strong association with IR and glycemic control in type II diabetic patients. Future studies are recommended to establish the possibility of using fetuin-A as a predictor of insulin resistance in type II diabetic patients.

Keywords:
C-reactive protein, diabetes mellitus, fetuin-A, insulin resistance

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Introduction

Diabetes mellitus (DM) is one of the most serious diseases [1]. The pathophysiologies of type II DM are reduced insulin sensitivity and increased insulin resistance associated with enhanced hepatic glucose output and impaired insulin secretion due to a progressive decline of β-cell function [2].

Several circulating proteins have been shown to be involved in the regulation of insulin sensitivity, such as fetuin-A, which is an endogenous inhibitor of the insulin-stimulated receptor tyrosine kinase; high levels of fetuin-A are associated with insulin resistance [3–6].

Fetuin-A located on chromosome 3q27; this region was mapped as a type II DM and metabolic syndrome susceptibility locus [7].

Aim of the work

Our aim was to investigate whether serum fetuin-A levels predict the incidence of insulin resistance in type II DM.

Patients and methods

The present study included 40 patients who had type II DM (20 females and 20 males) who served as the patients group; their ages ranged from 37 to 63 years with mean age of 47.45 ± 6.70 years. Forty apparently healthy individuals served as the control group (20 females and 20 males); their ages ranged from 37 to 58 years with mean age of 45 ± 6.03. All patients were selected from Al-Zhraa University Hospital and Almokatam Hospital during the period between January 2012 and May 2012. Written consent was obtained from all participants before the start of the research in addition to approval of ethical committee of Faculty of Medicine, Al-Azhar University.

All patients and control group were subjected to the following: full medical history and thorough physical examination were performed. The following laboratory parameters were estimated in blood samples: fasting and postprandial blood glucose, glycosylated haemoglobin (HbA_{1c}), urea, creatinine, lipid profile (total cholesterol, triglyceride, HDL, LDL), C-reactive protein (CRP), insulin, and fetuin-A. Abdominal ultrasound was performed to all patients.

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Exclusion criteria

Individuals were excluded if they had a known history of cardiovascular disease, stroke or transient ischemic attacks, uncontrolled hypertension, liver disease, renal disease, severe dyslipidemia (triglycerides >600 mg/dl or cholesterol >350 mg/dl); pregnant diabetic women and individuals taking lipid-lowering agents during the last 3 months, glucocorticoids, antineoplastic agents, psychoactive agents, or bronchodilators on a regular basis were also excluded.

Fasting venous blood samples of 5 ml was taken from each participant in the study and divided into two parts: the first part (1 ml) of venous blood was added to a tube containing EDTA for determination of HbA_{1c} by cation exchange resin [8]. The rest of blood was left to clot (the second part) and centrifuged at 3000 xg for 5 min for separation of serum. Fasting blood glucose (FBG) was determined immediately using the glucose oxidase method on Hitachi 912 autoanalyzer (Hitachi, Roche, Japan). The rest of serum was stored at −20°C for determination of CRP, urea, creatinine, cholesterol, triglyceride, insulin, and fetuin-A.

The determination of fasting and postprandial blood glucose, serum cholesterol, serum triglyceride, serum urea, and serum creatinine was carried out on Hitachi 912 autoanalyzer (Hitachi) by colorimetric methods. For determination of HDL-cholesterol, phosphotungstic acid and magnesium ions are used for precipitating all lipoprotein except the HDL fraction, which was present in the supernatant and measured by autoanalyzer 912. LDL-cholesterol was calculated using the Friedwald formula [9].

Direct detection of serum CRP was performed in serum by rapid latex agglutination procedure [10].

Serum insulin was determined using radioimmunoassay [11]. Insulin resistance was calculated as homeostasis model assessment of insulin resistance (HOMA-IR) using the following equation: HOMA-IR=fasting blood glucose (mg/dl)×fasting serum insulin (mIU/ml)/405 [12].

The determination of serum fetuin-A was carried out using quantitative sandwich enzyme immunoassay technique [13], and the kit was supplied from R & D Systems Europe Ltd (19 Barton Lane, UK).

Statistical analysis

The results were performed using statistical package for social science software, version 17.0 (SPSS Inc., Chicago, Illinois, USA). Continuous variables were expressed as mean ± SD. Comparison between two sets of patients was performed by the independent *t*-test, but more than two sets of patients were compared by one-way analysis of variance. Pearson correlation coefficient '*r*' was used to describe the association between serum fetuin-A and the variables of interest. *P* values less than 0.05 were considered statistically significant.

Results

The present study was carried out on 80 Egyptian persons and were classified into two groups.

Group 1 included 40 healthy age-matched and sex-matched individuals who served as the control group (20 females and 20 males). Their mean age was 45 ± 6.03 years.

Group 2 included 40 type II diabetic patients (20 females and 20 males). Their mean age was 47.45 ± 6.70 years.

Table 1 reveals highly significant increase in FBG, HbA_{1c}, serum urea, cholesterol, and CRP in the diabetic group compared with the control group ($P < 0.01$). There was nonsignificant difference with respect to age and systolic and diastolic blood pressure.

Table 2 shows highly significant increase in serum insulin, serum fetuin-A, and HOMA-IR in the diabetic group compared with the control group ($P < 0.01$).

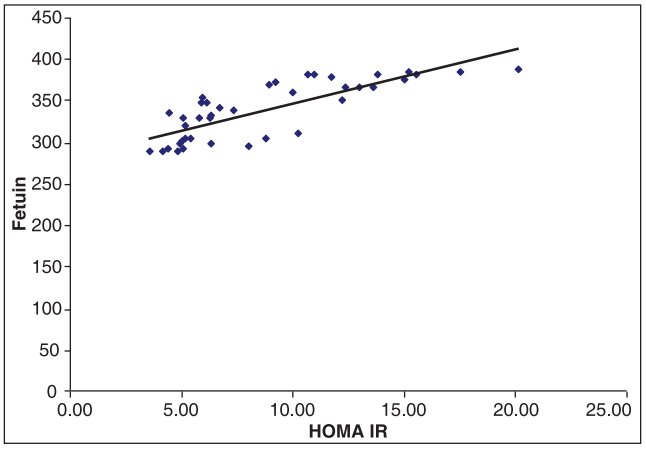
Tables 3 and 4 and Figs 1–3 shows a significant positive correlation between HOMA-IR and serum fetuin-A, serum insulin, and HbA_{1c} in diabetic patients. In

Table 1 Clinical and laboratory data of diabetic patients (group 2) and the control group (group 1) (mean ± SD)

Parameters	Groups		
	Diabetic patients (group 2)	Control (group 1)	*P*
Age (years)	47.45 ± 6.70	45.03 ± 6.03	>0.05
Systolic blood pressure (mmHg)	121.67 ± 5.87	119.5 ± 6.05	>0.05
Diastolic blood pressure (mmHg)	87.67 ± 3.43	78.5 ± 3.66	>0.05
FBG (mg/dl)	111.93 ± 27.64	82.10 ± 13.50	<0.01
HbA_{1c} (%)	6.64 ± 2.96	5.23 ± 0.12	<0.01
Urea (mg/dl)	31.92 ± 12.33	23.11 ± 8.67	<0.01
Creatinine (mg/dl)	0.83 ± 0.19	0.77 ± 0.18	>0.05
Cholesterol (mg/dl)	208.08 ± 45.43	175.43 ± 47.92	<0.01
Triglycerides (mg/dl)	125.10 ± 64.77	127.70 ± 62.44	>0.05
LDL (mg/dl)	128.24 ± 40.49	107.23 ± 42.72	>0.05
HDL (mg/dl)	51.40 ± 14.52	49.28 ± 18.20	>0.05
CRP (mg/l)	7.65 ± 9.31	0.30 ± 1.90	<0.01

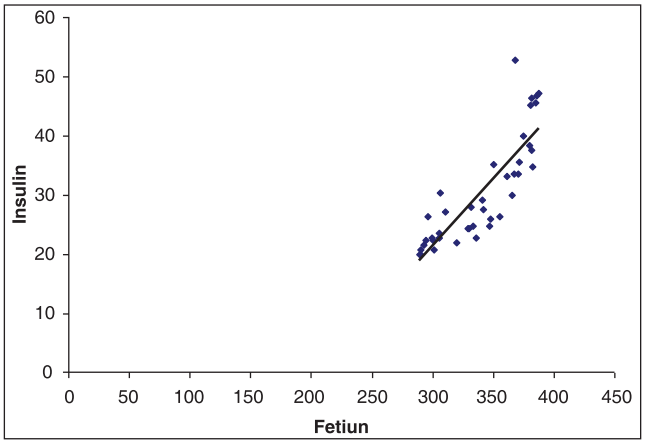
CRP, C-reactive protein; FBG, fasting blood glucose; HbA_{1c}, glycosylated hemoglobin or the hemoglobin A_{1c}; $P > 0.05$ = nonsignificant; $P < 0.01$ = highly significant.

Figure 1



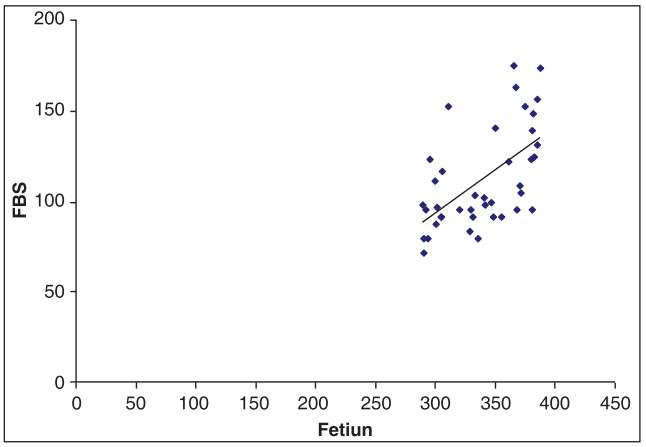
The correlation between HOMA-IR and serum fetuin-A in the diabetic group. HOMA-IR, homeostasis assessment model for insulin resistance.

Figure 2



The correlation between serum fetuin-A and serum insulin in the diabetic group.

Figure 3



The correlation between serum fetuin-A and FBG in the diabetic group. FBG, fasting blood glucose.

Table 2 Serum insulin, serum fetuin-A, and HOMA-IR in different groups (Mean ± SD)

Parameters	Groups		
	Diabetic group	Control group	*P*
Insulin (mIU/ml)	30.48 ± 8.99	9.05 ± 1.65	<0.01
Fetuin-A (μg/ml)	339.59 ± 33.91	284.33 ± 14.60	<0.01
HOMA-IR	8.77 ± 4.23	1.82 ± 0.37	<0.01

HOMA-IR, homeostasis assessment model for insulin resistance.

Table 3 Correlation between HOMA-IR and other laboratory parameters in the diabetic group

Diabetic group	HOMA-IR	*P*
Fetuin-A	0.796	<0.01
Insulin	0.889	<0.01
Urea	0.000	>0.05
Creatinine	0.191	>0.05
FBG	0.872	>0.05
HbA_{1c}	0.561	<0.01
Cholesterol	0.113	>0.05
Triglyceride	−0.136	>0.05
HDL	0.188	>0.05
LDL	0.003	>0.05

FBG, fasting blood glucose; HbA_{1c}, glycosylated hemoglobin or the hemoglobin A_{1c}.

Table 4 Correlation between fetuin-A and other laboratory parameters in the diabetic group

Diabetic group	Fetuin-A	*P*
Insulin	0.842	<0.01
Urea	−0.015	>0.05
Creatinine	0.118	>0.05
FBG	0.576	<0.01
HbA_{1c}	0.485	<0.01
CRP	0.786	<0.01
Cholesterol	−0.053	>0.05
Triglyceride	−0.263	>0.05
HDL	−0.214	>0.05
LDL	−0.127	>0.05

CRP, C-reactive protein; FBG, fasting blood glucose; HbA_{1c}, glycosylated hemoglobin or the hemoglobin A_{1c}.

addition, there was significant positive correlation between serum fetuin-A and serum insulin, FBG, HbA_{1c}, and serum CRP.

Discussion

Type II diabetes is characterized by inadequate insulin secretion and insulin resistance in the target tissues. Insulin mediates its action through phosphorylation of the insulin receptor. Fetuin-A inhibits insulin receptor autophosphorylation [14].

The present study showed highly significant increase in serum insulin, serum fetuin-A, and HOMA-IR in the diabetic group compared with the control group. Our results showed significant positive correlations

between fetuin-A levels and both fasting insulin levels and HOMA-IR in patients with type II diabetes and this agreed with the studies conducted by Jung *et al.* [15] who demonstrated that serum fetuin-A is significantly associated with IR, and Graham *et al.* [4] who showed that fetuin-A is positively correlated with insulin resistance.

These results were previously reported by Wallace *et al.* [12], who demonstrated that fetuin-A levels were correlated with fasting insulin levels and HOMA-IR in obese patients, suggesting a potential link between fetuin-A and insulin resistance. Stefen *et al.* [16] had demonstrated that fetuin-A was correlated with insulin resistance and fat accumulation in the liver. Li *et al.* [17] reported that fetuin-A, which is predominantly secreted by the liver, is found to be related to the accumulation of fat in the liver, insulin resistance, type II diabetes, and cardiovascular diseases.

Dasgupta and colleagues reported that the liver-secreted protein fetuin-A induces insulin resistance, and circulating fetuin-A is elevated in insulin resistance and fatty liver in humans. In agreement with these data, Emoto *et al.* [18] had shown that high levels of circulating fetuin-A are associated with insulin resistance in humans, suggesting that fetuin-A may represent a mechanism involved in the pathophysiology of type II diabetes.

Looking at lipid profile and their relationship with fetuin-A, our study showed highly significant increase in total cholesterol level in group 2 when compared with group 1 and insignificant difference in serum triglycerides, LDL, and HDL levels between two groups. Kotronen and Yki-Järvinen [20] showed that fetuin-A levels were negatively correlated with HDL-cholesterol.

Khalil and Kuobaili [21] reported that elevated serum fetuin-a levels found in type II diabetic patients were significantly associated with atherogenic dyslipidemia, thus indicating that fetuin-a may be one of the contributing factors to the increased incidence of coronary heart diseases in type II diabetic patients. Ix *et al.*, [22] reported that higher level of fetuin-A was associated with higher triglycerides, LDL-cholesterol, BMI, and insulin resistance.

In our results, CRP also showed marked increase in group 2 when compared with group 1. This agrees with a study reported by Kotronen and Yki-Järvinen [20], which showed that serum fetuin-A levels were increased in diabetic patients when compared with case–control individuals and demonstrated a positive correlation between serum fetuin-A and CRP levels. These results agreed with our results, as there was a positive correlation between serum fetuin-A and CRP levels ($r = 0.786$, $P < 0.01$).

These data further suggest a potential role for fetuin-A as a marker associated with inflammation in both type II DM and obesity [20]. Baumann and colleagues found that fetuin-A participates in the inflammatory response. In support of its inflammatory profile, fetuin-A has been shown to increase transcriptional events leading to an increased expression of several proinflammatory cytokines including interleukin-1, interleukin-6, interleukin-12, and tumor necrosis factor-A [23].

Acknowledgements

Conflicts of interest

There are no conflicts of interest.

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