Original Article

Association of sFas and sFas Ligand with Progression of Type 2 Diabetes Mellitus in Basrah Province

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Abstract

Objective: This study aimed to assess the role of Fas/Fas ligand (FasL) system in the progression of type 2 diabetes mellitus (T2DM). **Materials and Methods:** This study included 100 participants – 30 persons as a control group and 70 patients with T2DM (35 males and 35 females) and their ages were ranged from 40 to 70 years. The patients were distributed into two groups according to gender and duration of the disease: newly diagnosed group for short duration ≤5 years and chronic diagnosed group for long duration >5 years. Serum sFas and sFasL levels were measured by enzyme-linked immunosorbent assay technique, and also, lipid and glucose profile were measured by COBAS analyzer. **Results:** The results revealed a significant ($P \leq 0.05$) decrement in the levels of FasL in T2DM than controls while the levels of Fas were increasing significantly ($P \leq 0.05$) in T2DM than controls. Hemoglobin A1c (HbA1c) and fasting blood glucose were negatively correlated with FasL, while high-density lipoprotein was positively correlated with it, and whereas HbA1c positively correlated with Fas, the gender and duration of disease did not show any correlation with the disease. **Conclusion:** Our findings suggest that hyperglycemia causes increase in Fas levels which lead to dysfunction of pancreatic β-cell in T2DM.

Keywords: Enzyme-linked immunosorbent assay, sFas, sFas ligand, type 2 diabetes mellitus

Introduction

Diabetes mellitus (DM) is a set of metabolic disorders known by hyperglycemia resulting from defects in insulin secretion or action or both.[1] DM caused many complications such as macrovascular and microvascular complications.[2] The International Diabetes Federation has shown that 371 million people suffer from diabetes worldwide, and it may increase up to 552 million by 2030, meaning that, there are three new cases every second.[3,4]

DM had been classified into three major types which include Type 1 diabetes mellitus (T1DM) which is characterized by β-cell destruction, usually leading to absolute insulin deficiency; T2DM, which is the most predominant type and is characterized by insulin resistance; and gestational DM. Type 2 diabetes is an international health problem featured by a defect in insulin secretion and/or a decrease in sensitivity to insulin, also termed insulin resistance.[5] T2 DM is caused by a failure of the β-cells to preserve normal glycemia when insulin resistance is present, the capacity to secrete sufficient amounts of insulin depends on the function and the mass of β-cell. Increasing evidence indicates that the reduction in β-cell mass leads to an increase of β-cell apoptosis in T2DM.[6]

There are many studies that showed that the Fas/Fas ligand (FasL) system has an important role in the development of T2DM. Cosson *et al*.[7] pointed that Fas-mediated apoptosis is involved in type 2 diabetes and might be associated with hypertension and/or its vascular consequences. Blüher *et al*.[8] showed that increased Fas expression may contribute to impaired insulin sensitivity and adipose tissue dysfunction in obesity. Maedler *et al*.[9] and Maedler and Donath[10] noticed that increased Fas expression on β-cell has also been recorded in type 2 diabetic patients. In addition to Mahfouz *et al*.[11] who revealed that a dysregulation of apoptosis, as expressed by higher levels of sFas which may be useful for the early diagnosis of type 2 diabetic patients study. Furthermore,

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Hussian *et al*.[12] study whom concluded in their study which performed in diabetic foot patients that the apoptotic pathway in the development of diabetic foot increases by means of the Fas/FasL, and the development of new treatment against apoptosis may play an important role in the management of diabetic foot lesions. Stoynev *et al*.[13] showed that the values of sFasL reduced in persons with impairment in glucose tolerance; in addition, this decrease is detected in hypertensive patients with T2DM. Hence, current work aimed to study the association between T2DM progression with sFas and sFasL levels in patients of Basrah province.

Materials and Methods

Subjects

This study included 100 participants – 30 healthy persons as a control group and 70 patients with T2DM (35 males and 35 females) and their ages were ranged from 40 to 70 years. The patients were selected from Al-Mawani Hospital Specialized Center for Diabetes and Endocrinology in Basrah city during the period from February 2016 to February 2017. In the current study, the patients were distributed into two groups according to gender and duration of the disease: newly diagnosed group for short duration ≤5 years and chronic diagnosed group for long duration >5 years. Exclusion criteria involved those who were diagnosed with T1DM or who are taking insulin or patients suffering from other diseases and pregnancy.

Blood sampling

Five milliliters of venous blood was drawn by disposable syringe. Each blood sample was divided into 3 ml placed in a sterile plane tube and allowed to clot; then, serum was separated by centrifugation. Serum was stored at −20°C. These sera were used for estimating sFas and sFasL, lipid profile, and fasting blood glucose (FBG). The remaining 2 ml of blood were put directly in EDTA tube for estimating hemoglobin A1c (HbA1c).

Methods

Lipid profile and HbA1c were measured by COBAS automated analyzer, while sFas and sFasL were measured by sandwich enzyme-linked immunosorbent assay kit which provided from MyBioSource Company, USA.

Statistical analysis

Data were processed and analyzed using the Statistical Package of the Social Science (SPSS 19, IBM, Armonk, NY, United States of America). All results were expressed as mean ± standard deviation. Quantitative variables were expressed as (mean ± SD) and compared using student t-test. The linear relationship between variables was assessed by Pearson's correlation coefficient (r). $P < 0.05$ was considered statistically significant.

Results

Recorded data represented that there were no significant differences between study groups related to gender, age, and duration of disease as shown in Table 1. The concentrations of FBG and HbA1c and the lipid profile (total cholesterol, triglyceride, and low-density lipoprotein [LDL]) were significantly increasing ($P \leq 0.05$) in T2DM patients compared with the controls whereas the mean of high-density lipoprotein (HDL) was significantly lower ($P \leq 0.05$) in T2DM patients than controls as shown in Table 2. Recent results which are documented in Table 3 displayed a significant decrease ($P \leq 0.05$) in the levels of sFasL in T2DM in diabetic patients as compared with the controls, while sFas represented a significant elevation ($P \leq 0.05$) in diabetic patients when compared with the controls. Tables 4 and 5 revealed that sFasL and sFas concentrations did not show any significant difference among diabetic groups depending on gender and duration of diabetes.

Pearson correlation analysis revealed a significant positive correlation between HbA1c and sFas, while sFasL exhibited a significant negative correlation with FBG and HbA1c, but HDL correlated positively with FasL as shown in Table 6.

Discussion

The Fas/FasL system is an important regulating system accountable for the activation of apoptosis in several cell types, comprising cellular components of the vessel wall.[14] The present study showed a significant decrease ($P \leq 0.05$) in the concentrations of sFasL in T2DM compared with the controls. This decreasing may be explained that the higher

Table 1: Distribution of the studied groups according to gender, age, and duration of disease

Parameters	Controls (n=30)	T2DM (n=70)		P
Gender (n)%				
Male	(15) 50%	(35) 50%		1
Female	(15) 50%	(35) 50%		
Age (years)	29.71±6.019	54.991±8.182		0.99
BMI (kg/m^2)	23.98±1.14	24.60±1.43		0.08
Duration of disease (years) %	-	<5 years	More than 5 years	1
	-	35	35	
	-	50%	50%	

Table 2: Glucose and lipid profile in type 2 diabetes mellitus patients and control group

Parameters	Mean±SD		P
	Controls	Patients	
HbA1c	5.573±0.238	9.598±2.190	0.00**
FBG	98.811±11.064	229.342±97.18	0.00**
Total cholesterol (mg/dl)	160.534±26.168	186.841±60.239	0.05*
Triglyceride (mg/dl)	110.318±23.387	188.977±111.871	0.028*
HDL (mg/dl)	47.796±6.336	37.730±11.077	0.00*
LDL (mg/dl)	91.131±31.583	151.044±50.152	0.00*

*Significant difference at $P \leq 0.05$, **Significant difference at $P \leq 0.01$. HbA1c: Glycated hemoglobin, FBG: Fasting blood glucose, HDL: High-density lipoprotein, LDL: Low-density lipoprotein, SD: Standard deviation

Table 3: The concentrations of (sFas ligand and sFas) (pg/ml) in patients with type 2 diabetes mellitus and controls

Parameters	Groups	*n*	Mean±SD	SE	*P*
sFasL (pg/ml)	Patients	70	1.279±0.571	0.088	0.015*
	Controls	30	1.816±1.022	0.264	
sFas (pg/ml)	Patients	70	1.870±1.548	0.261	0.044*
	Controls	30	1.152±0.481	0.105	

*Significant difference at $P \leq 0.05$. SD: Standard deviation, SE: Standard error, sFasL: sFas ligand

Table 4: Comparison of sFas ligand and sFas (pg/ml) concentrations between diabetic groups depending on gender

Parameters	Groups	*n*	Mean±SD	SE	*P*
sFasL (pg/ml)	Male	35	1.020±0.579	0.105852	0.108
	Female	35	1.309±0.655	0.146639	
sFas (pg/ml)	Male	35	2.591±2.578	0.526	0.550
	Female	35	2.090±2.059	0.571	

SD: Standard deviation, SE: Standard error, sFasL: sFas ligand

Table 5: Comparison of sFas ligand and sFas (pg/ml) concentrations between diabetic groups depending on duration of disease

Parameters	Groups	*n*	Mean±SD	SE	*P*
sFasL (pg/ml) (years)	<5	35	1.339±0.560	0.149	0.55
	>5	35	1.418±0.604	0.161	
sFas (pg/ml) (years)	<5	35	2.013±1.922	0.466	0.864
	>5	35	2.259±1.709	0.414	

SD: Standard deviation, SE: Standard error, sFasL: sFas ligand

Table 6: Correlations between sFas and sFas ligand with lipid and glucose profile

Parameters	sFas		sFasL	
	r	*P*	*r*	*P*
HbA1c	0.289*	0.018*	−0.313*	0.011*
FBG	0.188	0.143	−0.244*	0.043*
Total cholesterol	−0.099	0.443	−0.118	0.359
Triglyceride	0.255	0.060	−0.219	0.114
HDL	0.108	0.466	0.273*	0.044*
LDL	0.197	0.145	−0.177	0.189

*Significant difference at $P \leq 0.05$. HbA1c: Glycated hemoglobin, FBG: Fasting blood glucose, HDL: High-density lipoprotein, LDL: Low-density lipoprotein, ssFasL: sFas ligand

expression of Fas receptor (which binding with sFasL to initiate the apoptosis) will lead to an increasing of consuming of FasL during the process of apoptosis which leads to reduction of FasL concentration in the serum.[15] These results are in line with Cosson *et al.*[7] and Stoynev *et al.*[13] who reported that sFasL was decreasing in serum sFasL levels in both hypertensive and normotensive participants with T2DM compared with the controls.

Recorded results demonstrate that the serum sFas concentrations in participants with T2DM were significantly increased ($P \leq 0.05$) compared to the controls. The reason of this increase is explained when the glucose levels are increasing (hyperglycemia). It would be caused an inflammation of β-cell and stimulating of elevating of Fas expression which correlates with β-cell inflammation and the islets become gradually sensitive to the apoptosis.[16] The induction, transcription, and modulation of Fas of expression are initiated by a number of pro-inflammatory cytokines such as interleukin (IL)-1α, IL-1β, interferon-gamma, nitric oxide, and tumor necrosis factor-alpha that cooperate with Fas to be the effector mechanisms of β-cell destruction.[17]

Documented results were in agreement with many studies worldwide,[7,11,17] but they were not consistent with the findings of other authors,[13,18] who stated that serum sFas showed nonsignificant differences in T2DM compared with controls.

Further, the higher concentrations of glucose impair the islet function by disturbing the metabolism of glucose in the mitochondria of β-cells and could induce apoptosis.[9]

Recorded results revealed that the total cholesterol, triglyceride, and LDL are increased significantly ($P \leq 0.05$) in T2DM patients while HDL is significantly ($P \leq 0.05$) decreased when compared with the controls. These results agreed with several authors.[19,20] The probable reason for the elevating concentrations of serum cholesterol may be referred to many factors such as obesity, increased calorie intake, and lack of muscular exercise or suppression of cholesterol catabolism.[21]

Conclusion

Our results suggest that hyperglycemia caused increase of Fas levels which lead to dysfunction of pancreatic β-cell in T2DM.

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Conflicts of interest

There are no conflicts of interest.

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