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Monocyte chemoattractant protein 1 and macrophage migration inhibitory factor in children with type 1 diabetes

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Abstract

Background: Serum monocyte chemoattractant protein 1 (MCP-1) and macrophage migration inhibitory factor (MIF) could be involved in the pathophysiological process of diabetes. The aim of the study was to evaluate MCP-1 and MIF in patients with diabetes mellitus type 1 (T1DM) and to assess its relation to diabetic control.

Methods: The study included 39 patients with type 1 diabetes and 38 healthy volunteers. Blood sample was taken for assessment of glycosylated hemoglobin, serum MIF and MCP-1.

Results: Serum MIF and MCP-1 were significantly higher in diabetic cases than in healthy controls. HbA_{1c} levels, were significantly higher in cases than in controls. Serum MIF had a significant positive correlation with serum MCP-1 (r=0.361, p=0.03). No other significant correlation with glycosylated hemoglobin or duration of diabetes was detected.

Conclusions: A significant increase of serum level of MIF and serum MCP-1 was found in patients with T1DM. These results support that MCP-1 and MIF could be a therapeutic target to treat diabetes and to prevent its complications.

Keywords: diabetes mellitus type 1 (T1DM); HbA_{1c}; macrophage migration inhibitory factor (MIF); monocyte chemoattractant protein 1 (MCP-1).

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Introduction

Diabetes mellitus type 1 (T1DM) is a multifactorial, organ-specific autoimmune disease that occurs in genetically susceptible individuals [1] and causes many acute and chronic complications [2]. It is the result of the autoimmune destruction of pancreatic islet β cells (insulitis); this occurs due to a failure in immune tolerance because the organism has had contact with specific viruses or with food molecules that caused molecular mimicry [3]. The common autoantigens recognized T1DM are insulin, glutamate decarboxylase 65 (GAD65), and the islet antigens IA-2 and IA-2β [4, 5]. During insulitis, high levels of pro-inflammatory cytokines, including, MIF are secreted by effector T cells to trigger the β cell destruction process [1].

MIF is a T cell derived cytokine that inhibited the random migration of macrophages in vitro and promoted macrophage accumulation during delayed-type hypersensitivity reactions [6, 7]. It is stored in intracellular pools, so it does not require immediate synthesis before secretion. MIF is released from cells through a nonconventional protein-secretion pathway [8]. It is involved in immunological and inflammatory diseases [9, 10] such as chronic diseases including bowel disease [11], obesity [12, 13], and diabetes [14]. More recently, MIF was proposed as a diagnostic biomarker for autoimmune diseases such as diabetes [15].

Monocyte chemotactic protein-1 (MCP-1), called CCL2 chemokine, is belonging to the CC chemokine family. It is one of the key chemokines that regulate migration and infiltration of monocytes/macrophages. MCP-1 has been demonstrated to be induced and involved in various diseases. It exerts its pro-inflammatory activity in the course of diabetes. MCP-1 also plays a critical role in angiogenesis, mainly by its chemotactic activity. It should be emphasized that MCP-1 exerts a direct effect on the vascular endothelium [16, 17].

The aim of the study was to evaluate serum MIF and MCP-1 in patients with T1DM and to assess its relation to diabetic control.

Materials and methods

The study included 39 patients with T1DM among those attending the pediatrics clinic at the National Research Centre. The control group consisted of 38 age- and sex-matched healthy normal volunteers. Inclusion criteria included patients between the ages 8 and 18 years. Exclusion criteria included patients during acute diabetic complications, for example, diabetic ketoacidosis (DKA) or hypoglycemia; patients suffering from other autoimmune diseases as thyroiditis, bronchial asthma, obesity, malignancy or receiving immunosuppressive drugs.

Study design and protocol

It is a prospective cross-sectional observational study done after obtaining approval from the Ethical Committee of the National Research Centre, Cairo, Egypt. Written informed consent was obtained from all the patients, their parents, and controls after full discussion about the aim of the study. This study is a part from a project done in the National Research Centre, Cairo, Egypt.

All the studied patients were subjected to the following: Full clinical history and clinical examination.

Anthropometric measurements in the form of weight, height, waist circumference (WC), and hip circumference (HC) were taken for each participant. The weight and height of the participants were measured up to 0.01 kg and 0.1 cm using a Seca Scale Standing Balance and a Holtain Portable Anthropometer (Holtain Ltd, Crymych, Wales, UK). Body mass index (BMI) was calculated as weight (in kg) divided by height (in m) squared [18]. The landmarks, instruments used, and techniques followed were those recommended by the international biological program [19].

MCP-1 in serum and MIF as well as serum hemoglobin A_{1c} (HbA_{1c}) were measured after overnight fasting simultaneously in all patients and controls. Their MIF and MCP-1 levels were measured using commercially available ELISA kits (MIF: Hangzhou East Biopharma Co. Ltd., China; MCP-1: Sun Red Biological Technology, China) according to the manufacturer's instructions. Glycosylated hemoglobin (HbA_{1c}) was measured using high-pressure liquid chromatography (Nichols Institute, Van Nuys, CA, USA).

Statistical analysis

Statistical analysis was conducted using Statistical Package for Social Science (SPSS) program version 12.0 (Chicago, IL, USA). All numeric variables were expressed as mean±SEM. Comparison of different variables in various groups was carried out using the Student t-test and the Mann-Whitney U-test for normal and nonparametric variables, respectively. Pearson's and Spearman's correlation tests (r=correlation coefficient) were used for correlating normal and nonparametric variables, respectively. For all tests, a p-value of <0.05 was considered significant.

Results

The study included 39 patients with type 1 diabetes (13 males and 26 females) and 38 healthy volunteers (14 males and 24 females). All the patients with diabetes were on intensive insulin therapy regimen. Table 1 shows the descriptive data of T1DM cases including: (minimum and maximum reading, mean and SD) of age, BMI, disease duration, HbA_{1c}, insulin dose.

Table 2 shows comparison between anthropometric and laboratory data of patients and controls.

Figure 1 shows the levels of serum chemokines, MCP-1 and MIF in T1DM children and controls. Both were significantly higher incases than in healthy controls (p=0.000). Serum HbA_{1c} levels, were significantly higher in patients with diabetes than in controls (p=0.000). Table 3 presents percentiles levels of MCP-1 and MIF of T1DM cases and controls. Both MCP-1 pg/mL and MIF ng/mL showed higher serum levels in all percentiles in cases than controls.

Correlations among MCP-1 pg/mL and MIF ng/mL with other data of patients with diabetes are presented in (Table 4). Serum MIF had a significant positive correlation with serum MCP-1 (r=0.361, p=0.03). No other significant correlation was detected. Figure 2: Relation between serum MCP-1 and MIF in T1DM cases. IT shows a positive relation.

Discussion

Knowing that insulitis marks the beginning of the T1DM and it is an autoimmune inflammatory process, we

Table 1: Descriptive data of T1DM cases.

	Minimum	Maximum	Mean	Std. deviation
Age, years	8.00	19.00	14.60	2.63
BMI, kg/m^2	16.00	24.20	23.51	4.14
Systolic blood pressure, mmHg	80.00	135.00	105.38	12.74
Diastolic blood pressure, mmHg	50.00	90.00	68.07	9.77
Duration of T1DM, years	1.00	15.00	5.36	3.32
Insulin dose, IU/kg/day	0.50	1.50	0.97	0.24
Glycosylated hemoglobin	4.60	13.00	8.35	2.07

Table 2: Comparison between anthropometric, and laboratory data of patients and controls.

	1=T1DM cases 2=Controls	n	Mean	Std. deviation	Sig. (2-tailed)
Age, years	1	39	14.60	2.63	NS
	2	38	14.02	3.51	
BMI, kg/m^2	1	39	23.51	4.14	NS
	2	38	23.14	3.97	
Systolic blood pressure, mmHg	1	39	105.38	12.74	NS
	2	38	103.55	12.13	
Diastolic blood pressure, mmHg	1	39	68.07	9.77	NS
	2	38	66.57	6.98	
Total cholesterol, mg/dL	1	39	189.16	60.51	0.020
	2	38	162.97	28.05	
Triglyceride, mg/dL	1	39	104.72	50.38	NS
	2	38	89.13	31.90	
HDL, mg/dL	1	39	43.94	14.36	10^{-3}
	2	38	62.97	15.36	
LDL, mg/dL	1	39	108.12	64.56	NS
	2	38	98.36	28.07	
MCP-1, pg/mL	1	39	69.61	18.18	10^{-3}
	2	39	6.18	3.43	
MIF, ng/mL	1	39	9.23	1.96	10^{-3}
	2	39	6.40	3.23	

BMI, body mass index; HbA_{1c}, glycosylated hemoglobin; LDL, low-density lipoprotein; HDL, high-density lipoprotein.

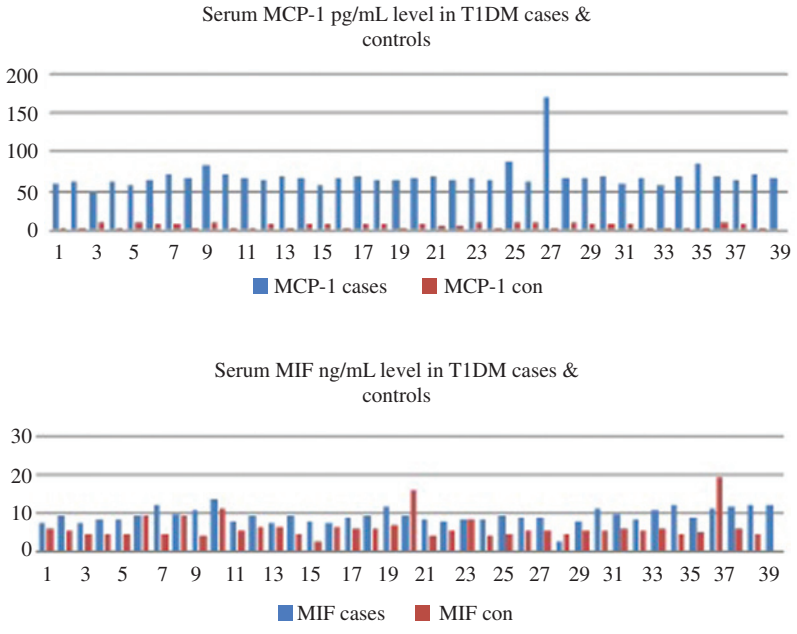

Figure 1: Serum MCP-1 and MIF levels in T1DM cases and controls.

propose the hypothesis that MIF plays an important role in insulitis onset or development. In our study, serum MIF was significantly higher in patients with T1DM which coincides with the result of Stosic-Grujicic et al. They reported that MIF participates in T1DM by controlling the functional activities of monocytes/macrophages and T cells and modulating their abilities to secrete pro-inflammatory molecules [20]. MIF is an important molecule to the development of T1DM complications such as cardiac dysfunction, and diabetic foot disease [21, 22]. It is known to promote inflammatory cytokine and palmitic acid-induced pancreatic islet apoptosis [23, 24].

In the current study, serum MIF had no significant correlation with BMI, HbA_{1c} or the duration of diabetes.

Table 3: Percentiles levels of MCP-1 and MIF in T1DM cases and controls.

		T1DM cases		Controls	
		MCP-1, pg/mL	MIF, ng/mL	MCP-1, pg/mL	MIF, ng/mL
n	Valid	39	39	38	38
Mean		69.61	9.23	6.18	6.40
Std. dev.		18.18	1.96	3.43	3.23
Percentiles	25	64.50	8.07	2.50	4.50
	50	66.50	9.05	7.80	5.50
	75	69.00	10.37	9.00	6.50

Table 4: Correlation between MCP-1 and MIF with other data of diabetes patients.

		MCP-1, pg/mL	MIF, ng/mL
BMI	Correlation coefficient (r)	0.235	0.156
	Sig. (2-tailed)	0.155	0.370
	n	39	39
MCP-1, pg/mL	Correlation coefficient (r)	1.000	0.361[a]
	Sig. (2-tailed)	.	0.030
	n	39	39
MIF, ng/mL	Correlation coefficient (r)	0.361[a]	1.000
	Sig. (2-tailed)	0.030	.
	n	39	39
Duration of T1DM, years	Correlation coefficient (r)	0.188	0.003
	Sig. (2-tailed)	0.257	0.985
	n	39	39
HbA_{1c},%	Correlation coefficient (r)	0.103	0.063
	Sig. (2-tailed)	0.593	0.753
	n	39	39

[a]Correlation is significant at the 0.05 level (2-tailed).

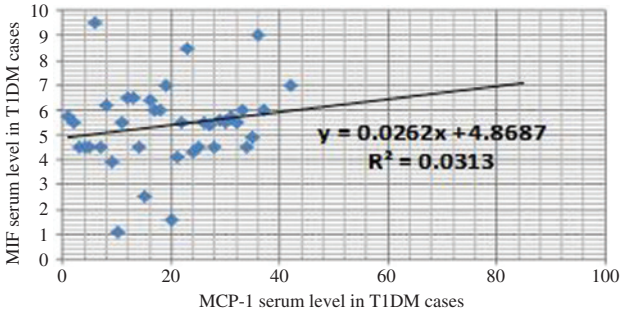

Figure 2: Relation between serum MCP-1 and MIF in T1DM cases.

This indicates that the increased MIF serum level in diabetes involves in the pathophysiological process of the disease not to uncontrolled or duration of diabetes.

In the present study, MCP-1 serum levels were significantly higher in T1DM cases than in healthy controls. Our result is in concordance with a study of Panee [25]. MCP-1 main function is to chemotactic monocyte/macrophages and T lymphocytes resulting in islet beta cell destruction. While another study reported that MCP-1 attracts monocytes to the inflammatory sites of vascular sub-endothelial space, initiating migration of monocytes into the arterial wall to form excessive macrophage-derived foam cells. So, diabetes with elevated MCP-1 was found to correlate with the association with accelerated rates of atherosclerosis [26, 27].

Bruun et al., demonstrated that MCP-1 release is higher in obese compared with lean subjects. MCP-1 is correlated with adiposity. In our study obesity being an exclusion criterion, BMI was not correlated with MIF serum level [28].

We conclude that a significant increase of serum level of MIF and serum MCP-1 was found in patients with T1DM. We demonstrated that Serum MIF had a significant positive correlation with serum MCP-1. These results support the statement that MCP-1 and MIF could be therapeutic target to treat diabetes and to prevent its complications.

There were few limitations of the study. We did not correlate our results with diabetic complications such as microalbuminuria, renal, ocular and cardiac complications. Also, we did not look for a possible association between chemokines levels and autoantibody status.

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