

Prevalence of Leukocytes in Type 2 Diabetic Patients in Erbil City

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Abstract

Background: Diabetes mellitus (DM) has become a global health concern affecting children, youth, and adults. Type 2 DM (T2DM) is considered an inflammatory disease as inflammation can trigger the insulin signaling, assumingly by promoting beta cell death. **Objectives:** This comparative study has been conducted on white blood cells in diabetic patients to examine the relationship between inflammation and blood glucose levels. **Methods:** Blood samples from both males and females were obtained randomly and measured at Rizgary Teaching Hospital and Media Laboratory in Erbil City during October 2015 till March 2016. Blood glucose levels and different types of leukocytes were measured in T2DM patients and compared with nondiabetic patients. **Results:** The results showed an increment in the percentage of monocytes and neutrophils in diabetic females and a decrement in the percentage of lymphocytes as compared to controls, in contrast to diabetic males where an increment in the percentage of monocytes and lymphocytes was found. **Conclusions:** It has been proven by this study that leucocyte count is elevated in diabetic patients, in reference to the hyperactivated immune system in such people. However, the results revealed that the inflammatory system showed a significant difference between diabetic and nondiabetic people, reflecting on the importance of such system in the response of a body to the lack of control over blood glucose.

Keywords: Insulin, leukocytes, type 2 diabetes mellitus

INTRODUCTION

Diabetes mellitus (DM) is a metabolic disease characterized by elevated blood glucose levels (hyperglycemia) resulting from defects in insulin secretion, insulin action, or both. Insulin is a hormone manufactured by the beta cells of the pancreas, which is required to utilize glucose from digested food as an energy source. Chronic hyperglycemia is associated with microvascular and macrovascular complications that can lead to visual impairment, blindness, renal failure, nerve damage, and amputations.^[1]

Diabetes is classified into two major classes: Type 1 DM (T1DM) and Type 2 DM (T2DM). T1DM is the classical and most common form of diabetes which is mostly diagnosed during childhood. About 5%–10% of people with diabetes have T1DM, a condition characterized by when a person's pancreas makes little or no insulin and they cannot survive without insulin treatment, requiring daily injections of insulin to bring the glucose levels in the blood to a normal level. This is mainly caused either due to autoimmune or idiopathic destruction of the pancreatic beta cells by activated CD4⁺ and CD8⁺ T cells and

macrophages infiltrating the pancreatic islets.^[2] T1DM can also be termed as insulin-dependent DM and juvenile diabetes.^[3,4]

T2DM is the result of failure to produce sufficient insulin and insulin resistance. It is characterized by hyperglycemia, insulin resistance, decreased number of beta cells, systemic inflammation, elevated cytokines, hypercoagulation, and endothelial cell dysfunction.^[5] The elevated blood glucose levels are managed with reduced food intake, increased physical activity, and eventually oral medication or insulin. About 90% of all cases of diabetes are of T2DM.^[6]

T2DM is divided into two subgroups: diabetes with obesity and without obesity. The obese T2DM patients usually develop

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Submission: 04-01-2020 **Accepted:** 26-01-2020 **Published Online:** 17-03-2020

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How to cite this article: Ubeid MH. Prevalence of leukocytes in type 2 diabetic patients in Erbil City. *Med J Babylon* 2020;17:19-24.

resistance to endogenous insulin due to alteration in cell receptors, and this is associated with distribution of abdominal fat. In nonobese T2DM patients, there is some insulin resistance at the postreceptor levels in addition to a deficiency in insulin production and release. T2DM is the more common one, and it appears in adulthood. It may go unnoticed and undiagnosed for years. In this situation, the pancreas is not making enough insulin or there is insulin resistance whereby the cells that are normally targeted by insulin show a decreased response to insulin.^[7]

T2DM is considered a metabolic and an inflammatory disease.^[8] Individuals with impaired glucose metabolism are at an increased risk of fatal ischemic heart disease,^[9] and there is firm evidence that inflammation is an indication in the pathogenesis of atherosclerosis^[10] and hyperglycemia is associated with increased markers of inflammation.^[11]

Chronic inflammation, characterized by the increased production of cytokines and the activation of inflammatory signaling networks, may be involved in the pathogenesis of T2DM.^[12] Various markers of inflammation have been shown to predict the future risk of diabetes, including interleukin-6 (IL-6) and C-reactive protein.^[13]

Obesity is a strong risk factor for T2DM associated with inflammation as the fat tissue releases inflammatory cytokines.^[14] Inflammation on its own can affect insulin signaling, indirectly increasing the risk of T2DM, without the presence of obesity. Furthermore, inflammation is thought to promote beta-cell death. However, there is considerable uncertainty about the direction of causality of the relationship between inflammation and T2DM.^[15]

Only few studies have been conducted on the relationship between the blood parameters with the level of blood sugar, especially those people whom are DM independent on insulin intake. Therefore, this comparative study was conducted on some types of leukocytes and the blood glucose level in insulin-dependent DM patients in the city of Erbil.

MATERIALS AND METHODS

Patient selection

All the patients gave informed consent in accordance with the local ethics committee recommendations. A total of sixty insulin-dependent diabetic patients of both sexes (females 32 and males 28) were recruited in this study. The samples were taken randomly from different areas over the city of Erbil. The blood collection and glucose test were carried out at the Media lab and the Medical lab of Rizgary Teaching Hospital in Erbil, during the period from November 2015 till March 2016. All the patients were compared with twenty healthy nondiabetic people that were used as controls. The control samples in this study were also randomly selected.

Random blood sugar test

There are two different types of glucose blood screening tests for diabetes: random blood sugar (RBS) and fasting blood sugar. Patients are randomly chosen, and the amount of 2–3 mm of

blood is drawn by a technician from the patients' vein. Samples are left to clot at room temperature for a period of 5–10 min and are later centrifuged for 10 min to obtain the serum which is then placed inside an autoanalyzer device. Results of the test appear on the computer's screen and are reported in mg/dl or in mmol/l. The normal result for unaffected healthy people is between 60 and 120 mg/dl or 3.2 and 6.5 mmol/l.

Complete blood count

Complete blood count was performed by normal procedure, listing the number of many important values. Typically, it includes the following:

- White blood cell (WBC) count (or leukocyte count)
- WBC differential count.

Some of these blood parameters have been used in the analysis of the result to find the relationship between any of them with the blood sugar level of the diabetic patients.

Statistical analysis

All the results were analyzed using the increasing and decreasing frequency in the amount of blood parameters according to the standard level of the normal sample measurements. Statistical analysis was conducted to find the relationship between blood sugar levels (in diabetic insulin-dependent patients) and some of the WBC parameters in female adults. The results were analyzed statistically using Student's *t*-test to compare the significance of the differences between the means of test and control studies. The results were expressed as mean $\pm$ standard deviation. $P < 0.05$ was considered statistically significant.

Ethical consideration

The study was conducted in accordance with the ethical principles that have their origin in the Declaration of Helsinki. It was carried out with patients' verbal and analytical approval before sample was taken. The study protocol, the patient information, and consent form were reviewed and approved by a local ethics committee.

RESULTS

Table 1 describes the differences in the total number of leukocytes between males and females in both diabetic and nondiabetic patients.

WBC count differs between diabetic and nondiabetic patients to a significant degree. As presented in Table 2, the mean of WBC count in diabetic patients is about $8.39 \times 10^3/\mu\text{l}$ in females and $8.22 \times 10^3/\mu\text{l}$ in males, whereas in nondiabetic patients, it is $9.15 \times 10^3/\mu\text{l}$ and $8.74 \times 10^3/\mu\text{l}$. Figures 1 and 2 show the WBC count in diabetic and nondiabetic females and males, respectively, indicating that the leukocytes in both male and female diabetic patients are less compared to their controls.

Nondiabetic female patients have a larger percentage of lymphocyte cells (37.81%) than female diabetic patients (29.87%) as shown in Figure 3. In contrast with the male patients, the percentage of lymphocytes was slightly higher in

Table 1: The differences in the total number of leukocytes between genders (females and males) in diabetic and nondiabetic patients

Gender	n	Mean	SD	SEM	t-test, P	Decision
Age						
Female	32	38.56	19.038	3.365	0.285	NS
Male	28	33.75	15.501	2.929		
Leukocyte ($\times 10^3/\mu\text{l}$)						
Female	32	8.3875	4.85916	0.85899	0.876	NS
Male	28	8.2214	3.26501	0.61703		
Lymphocyte (%)						
Female	32	29.8719	10.04731	1.77613	0.023	S
Male	28	35.0429	7.00256	1.32336		
Monocyte (%)						
Female	32	10.0344	2.18331	0.38596	0.341	NS
Male	28	9.1893	4.16256	0.78665		
Neutrophil (%)						
Female	32	60.7094	11.14371	1.96995	0.033	S
Male	28	54.9429	9.28589	1.75487		
RBS						
Female	32	174.3750	35.22073	6.22620	0.454	NS
Male	28	183.1786	52.09468	9.84497		

SD: Standard deviation, SEM: Standard error of mean, RBS: Random blood sugar, NS: Not significant, S: Significant

Table 2: The mean of white blood cells as total and differential count and random blood sugar in both diabetic and control (nondiabetic) group

	Gender, mean									
	Female					Male				
	Leukocyte ($\times 10^3/\mu\text{l}$)	Lymphocyte (%)	Monocyte (%)	Neutrophil (%)	RBS	Leukocyte ($\times 10^3/\mu\text{l}$)	Lymphocyte (%)	Monocyte (%)	Neutrophil (%)	RBS
Diabetics	8.39	29.87	10.03	60.71	174.38	8.22	35.04	9.19	54.94	183.18
Control	9.15	37.81	8.01	54.18	110.25	8.74	34.24	9.06	56.70	109.17

RBS: Random blood sugar

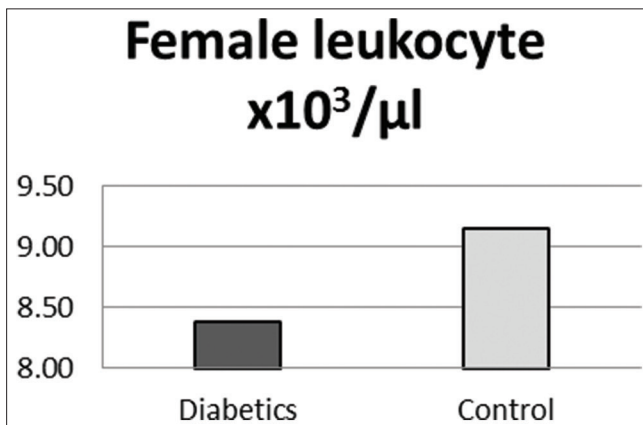


Figure 1: White blood cell count in diabetic and nondiabetic females

diabetic patients (35.04%) compared to nondiabetics (34.24%) as shown in Figure 4.

While both female and male diabetic patients showed an increment in the percentage of monocytes in comparison with their controls, in diabetic females, it appeared to be 10.03% and in diabetic males to be 9.19% compared to nondiabetic

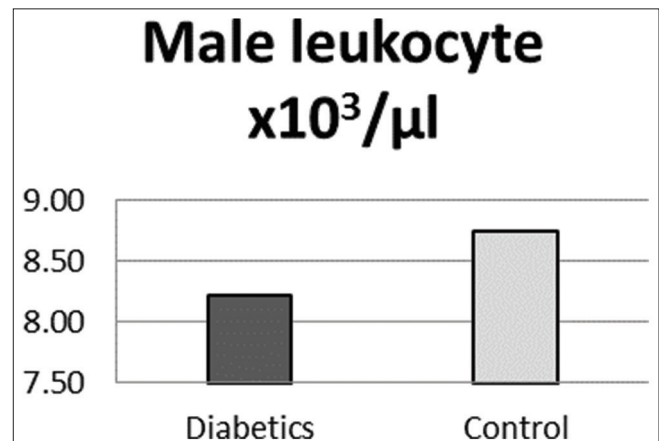


Figure 2: White blood cell count in diabetic and nondiabetic males

females to be 8.01% and nondiabetic males to be 9.09%, as shown in Figures 5 and 6, respectively.

The percentage of neutrophil type of WBCs showed some differences between diabetic and nondiabetic patients, appearing to be higher in diabetic females (60.71%) than in nondiabetics

(54.18%), as shown in Figure 7. This percentage contrasted in diabetic males where it showed to be 54.94% compared to the nondiabetic males (56.70%), as presented in Figure 8.

DISCUSSION

Type 2 diabetes used to be called adult-onset diabetes, but with the epidemic of obese and overweight kids, more teenagers

are now developing Type 2 diabetes. Type 2 diabetes is often a milder form of diabetes than Type 1. Nevertheless, Type 2 diabetes can still cause major health complications, such as heart disease and stroke. Therefore, this study attempts to find an indication to the relationship between the level of blood glucose in diabetic patients and their WBCs.

Evidence from epidemiological studies suggests an association between total WBC count, a nonspecific marker of inflammation,

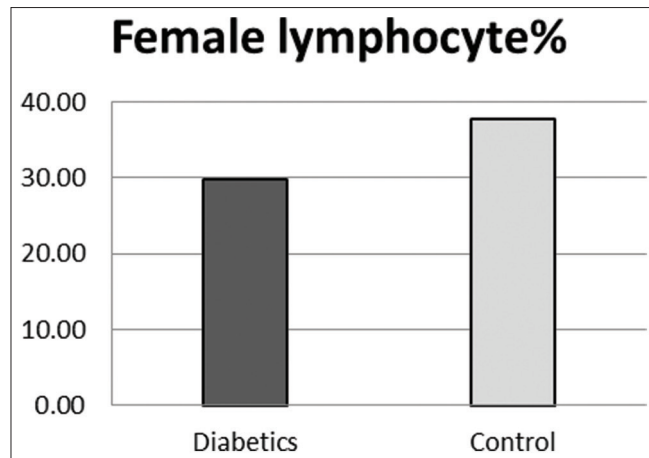


Figure 3: Lymphocyte % in diabetic and nondiabetic females

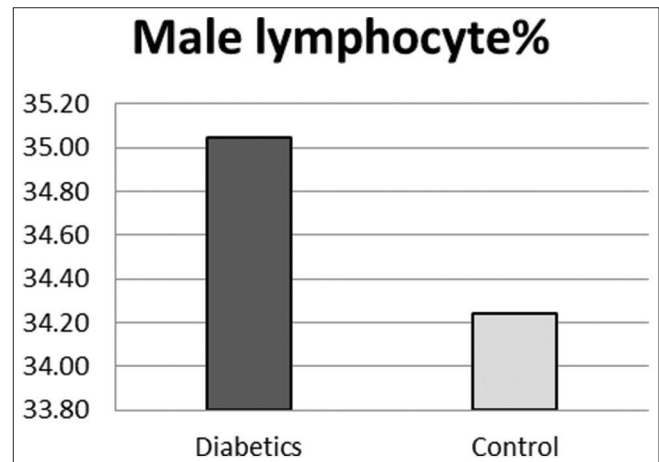


Figure 4: Lymphocyte % in diabetic and nondiabetic males

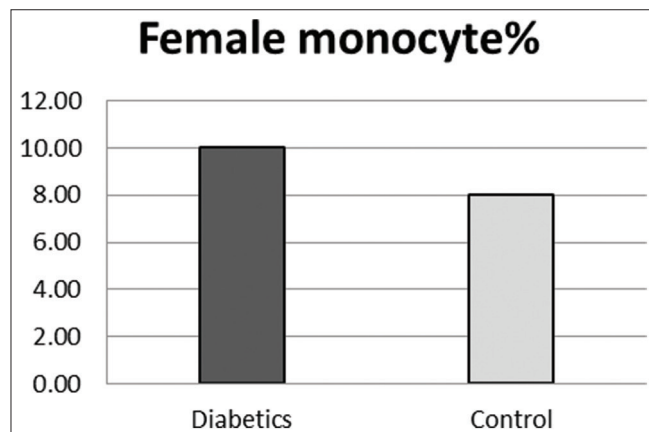


Figure 5: Monocyte % in diabetic and nondiabetic females

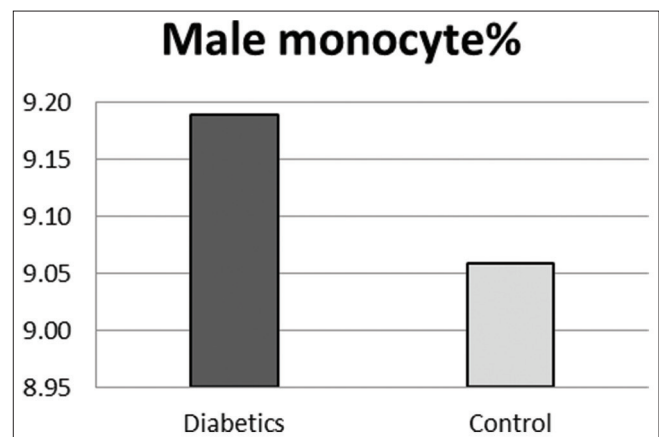


Figure 6: Monocyte % in diabetic and nondiabetic male

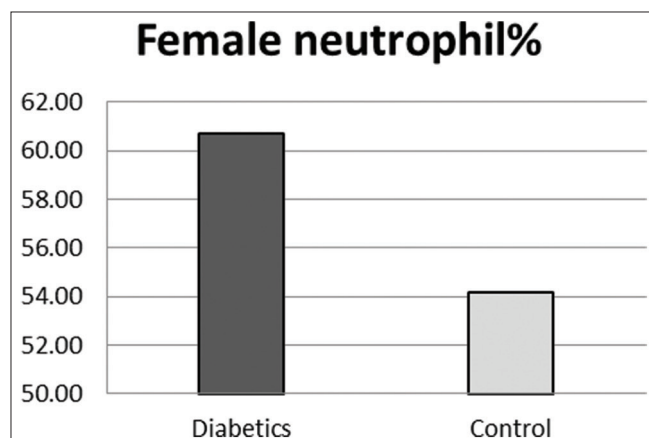


Figure 7: Neutrophil % in diabetic and nondiabetic females

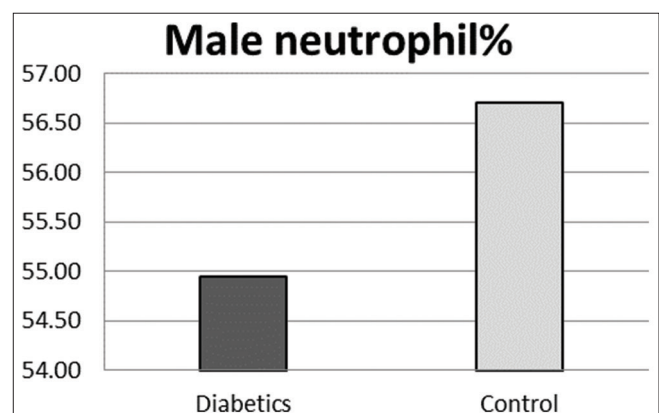


Figure 8: Neutrophil % in diabetic and nondiabetic males

and diabetes risk. Although a number of studies have been published, still considerable information is lacking.^[16]

Prospective studies on Pima Indians and other populations support the hypothesis that altered markers of inflammation, such as high WBC, plasma fibrinogen, plasminogen activator inhibitor-1 (PAI-1), gamma globulin, and lower albumin concentrations, are associated with the development of Type 2 diabetes. However, the mechanisms by which these markers contribute to the development of Type 2 diabetes are largely unknown.^[17]

Interestingly, both acute and chronic diabetic complications are correlated with elevated WBC count. Some studies have indicated that an elevated WBC count, even within the normal range, is associated with both macro- and microvascular complications in Type 2 diabetes.^[18] This coincides with the results of this study where the number of monocytes and neutrophils increased in diabetic patients compared to their controls. There is some evidence from prospective studies on Pima Indians and other populations to support the hypothesis that altered markers of inflammation, such as high WBC, plasma fibrinogen, PAI-1, gamma globulin, and lower albumin concentrations, are associated with the later development of Type 2 diabetes. However, the mechanisms by which these markers of inflammation contribute to the development of Type 2 diabetes are largely unknown.^[17]

People who are obese more than 20% over their ideal body weight for their height are at particularly high risk of developing Type 2 diabetes. Obese people have insulin resistance, meaning that the pancreas must work very hard to produce more insulin. But even then, there is not enough insulin to keep sugars normal. This study has excluded this parameter and even the data that were randomly collected did not consider the body weight and age of the patients.

Findings from the present study reveal that the levels of RBS reach approximately 183.18 mg/dl in diabetic male patients compared with diabetic females at 174.38 mg/dl, as demonstrated in Table 1. The count of RBS is generally higher in diabetic patients in both males and females than that in nondiabetic males and females.

Furthermore, we report that the number of neutrophils and lymphocytes is significantly higher in male diabetic patients compared to females. Our data show that although females have less favorable T2DM disease risk, they have higher WBC counts than males [Table 1].

Female diabetic patients have higher monocyte and neutrophil counts and lower lymphocyte counts than male diabetic patients [Table 2]. However, further studies are needed to determine whether lifestyle differences are related to WBC variability across sex groups.

Both a higher WBC and insulin resistance reflect an underlying activation of the immune system. For instance, IL-6, a potent WBC differentiation factor that is produced mostly in adipose tissue, is associated with insulin resistance.

This is supported by the fact that a single-nucleotide polymorphism in the IL-6 gene is shown to be associated with an increased WBC and lower insulin sensitivity.^[19] It is likely that an activation of the immune system that results from inflammation could increase the number of WBCs leading to cytokine production, such as IL-6, and hence may decrease insulin sensitivity.^[20]

The present study demonstrated that the total and differential leukocyte counts were significantly altered in patients with hyperglycemia from a stable condition to ketoacidosis. This results as acute infection increased the elevating extent of the counts of total WBC and neutrophils.

CONCLUSIONS

It has been proven by this study that WBC count is elevated in diabetic patients, with reference to the hyperactivated immune system in such people. Such increase is linked to a rise in the leukocyte count along with neutrophils and other WBCs. The inflammatory response is an essential characteristic of those with diabetes, serving as a marker to detect such incidence. Furthermore, the study was carried out on WBC count in both males and females, shedding more light on the association between diabetes and WBCs in both genders. Overall, the results did show that the inflammatory system shows a significant difference between diabetic and nondiabetic people, reflecting on the importance of such system in the response of a body to the lack of control over blood glucose.

Acknowledgments

The whole practical work was carried out with the great help of the medical lab technicians of Rizgary Teaching Hospital and Media Laboratory, in Erbil City.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

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