

Do DPP-4 enzyme inhibitors affect hemoglobin, leucocyte and thrombocyte levels in patients with type 2 diabetes mellitus?

F. AVCI MERDIN¹, A. MERDIN²

¹University of Health Sciences, Ankara Dr. Abdurrahman Yurtaslan Oncology Training and Research Hospital, Internal Medicine Clinic, Ankara, Turkey

²University of Health Sciences, Hematology Clinic and Bone Marrow Transplantation Unit, Ankara Dr. Abdurrahman Yurtaslan Oncology Training and Research Hospital, Ankara, Turkey

Abstract. – OBJECTIVE: Dipeptidyl peptidase-4 (DPP-4) enzyme inhibitors are used to increase the effect of incretins in the treatment of type 2 diabetes mellitus (DM). This study aimed to explore possible effects of DPP-4 enzyme inhibitors, which are widely used for blood sugar regulation in patients with type 2 DM, on hemoglobin, leukocyte (leucocyte), mean corpuscular volume (MCV) and thrombocyte levels.

PATIENTS AND METHODS: The study included 110 patients aged over 18 and diagnosed with type 2 diabetes mellitus, who applied to the Internal Medicine Polyclinic of Ankara Dr. Abdurrahman Yurtaslan Oncology Training and Research Hospital between 01.01.2019 and 31.08.2019 for routine control. Patients using metformin for DM and patients using metformin combined with DPP-4 enzyme inhibitors were divided into 2 groups. The data on patient hemogram (complete blood count) parameters were recorded retrospectively from the electronic patient file system. Patients with nutritional anemia were not included in the study. Besides, patients with known additional diseases (such as liver disease, kidney failure and malignancy) that may affect hemogram parameters, and patients with known infectious or inflammatory diseases were not included. Pregnant women were also excluded from the study. In addition, patients using angiotensin-converting enzyme (ACE) inhibitor class drugs were not included in the study.

RESULTS: There were no statistically significant differences between the two groups in terms of hemoglobin levels, MCV levels, leukocyte counts and thrombocyte counts ($p>0.05$).

CONCLUSIONS: Abnormalities in hemogram parameters in type 2 DM patients using DPP-4 enzyme inhibitors should not be immediately related to DPP-4 enzyme inhibitors, and non-drug etiologies that may cause abnormal levels in hemogram parameters should be carefully investigated.

Key Words:

Thrombocyte, Hemoglobin, Leukocyte, DPP-4 enzyme inhibitor, Effect.

Introduction

Diabetes Mellitus (DM) is a chronic metabolic disease characterized by hyperglycemia, which may occur due to defects in insulin secretion, insulin action, or both of these factors. Classification of DM can be performed based on the pathogenesis of the disease and is important in that it can guide the treatment. It is mainly classified into categories including type 1 diabetes mellitus, type 2 diabetes mellitus, various specific types of diabetes mellitus and gestational diabetes mellitus. In type 2 DM, partial insulin deficiency can be observed along with insulin resistance. At the same time, the decrease in the levels of two incretin hormones glucagon-like peptide-1 (GLP-1) and gastric inhibitory peptide (GIP), in the human body and the inability to inhibit glucagon secretion are among the reasons that may be associated with type 2 DM. Deacon et al¹ suggested in their study that the main enzyme that degraded the glucagon-like peptide-1 protein *in vitro* was dipeptidyl peptidase-IV. Sitagliptin, vildagliptin, saxagliptin, linagliptin are diabetes mellitus drugs known to block the Dipeptidyl peptidase-4 (DPP-4) enzyme. Mentlein et al² showed that growth-hormone-releasing factor (1-29)-amide, gastric inhibitory peptide with terminal Tyr-Ala, glucagon-like peptide-1 (7-36)-amide and peptide histidine methionine (PHM) with terminal His-Ala proteins are hydrolyzed by dipeptidyl-peptidase IV purified from the human placenta.

DPP-4 enzyme protein has long been known to be found in several different tissues and cells. Lojda³ has shown that DPP-4 can be found in vascular endothelium. Mentlein et al⁴ found that DPP-4 is a marker that could be found on T lymphocyte surfaces. Bathon et al⁵ demonstrated that there is dipeptidyl(amino)peptidase IV on the surfaces of synovial cells. Raynaud et al⁶ reported that DPP-4 was also found in fibroblasts in the skin. In their study conducted on rats, Mitro and Lojda⁷ showed DPP-4 activity in the capillary endothelium of the choroid plexus and leptomeninges. Bernstein et al⁸ reported that DPP-4 was also present in the human brain, especially in the immature human brain. In addition, Iwaki-Egawa et al⁹ purified dipeptidyl peptidase IV from human serum, but the enzyme purified from the human serum in their study lacked the transmembrane part of the whole enzyme. Iwaki-Egawa et al⁹ suggested that the transmembrane domain of the dipeptidyl peptidase IV was not necessary to bind adenosine deaminase.

Scholz et al¹⁰ reported a relation between dipeptidyl peptidase IV activity and interleukin (IL)-2 production. The enzymatic activity of proteins formed due to genetic transcription may cause different effects depending on the cell and tissue type. In this study, we aimed to evaluate the possible effects of DPP-4 enzyme inhibitors, which are widely used for blood sugar regulation in patients with type 2 DM, on hemoglobin, leukocyte, MCV and platelet (thrombocyte) levels.

Patients and Methods

The study included 110 patients aged over 18 and diagnosed with type 2 diabetes mellitus, who applied to the Internal Medicine Polyclinic of Ankara Dr. Abdurrahman Yurtaslan Oncology Training and Research Hospital between 01.01.2019 and 31.08.2019 for routine control. Type 2 DM patients using metformin and type 2 DM patients using metformin + DPP-4 enzyme inhibitors were included in the study. Patients using metformin but not using DPP-4 enzyme inhibitors formed a group, while patients using metformin combined with DPP-4 enzyme inhibitors formed the comparison group. Type 2 DM patients using metformin without using DPP-4 enzyme inhibitors for at least 3 months or using combined therapy of metformin + DPP-4 enzyme inhibitors for at least 3 months were included in the study. Those who received treatment for less

than 3 months were not included. Patients with known nutritional anemia and hypothyroidism were excluded from the study. Patients with comorbidities such as liver disease, kidney failure and malignancy that could affect complete blood count parameters were not included in the study. In addition, patients with known infectious or inflammatory diseases that could affect complete blood count parameters were also not included in the study. Women known to be pregnant, patients with known diagnosis of type 1 DM, patients using ACE inhibitor class drugs and patients under 18 years of age were also excluded from the study. The data on complete blood count parameters were recorded retrospectively from the electronic patient file system.

Statistical Analysis

Statistical analyses were performed on a software package called SPSS for Windows; Version 24.0 released 2016 (IBM Corp., Armonk, NY, USA). Quantitative data were presented as mean±standard deviation and median. “Independent Sample” *t*-test (*t*-table value) statistics were used to compare the measurement values of two independent groups with normally distributed data. The “Mann-Whitney U” test (*Z*-table value) statistics were used to compare the measurement values of two independent groups in the data that did not have a normal distribution. A *p*-value lower than 0.05 (*p*<0.05) was considered statistically significant.

Results

While the median age was 58.5 years in the metformin group, it was 59 years in the group using metformin+DPP-4 enzyme inhibitor treatments, and there was no statistically significant difference in median age between the two groups (Table I).

When both groups were compared in terms of hemoglobin levels, no statistically significant difference was found between the groups (*p*>0.05) (Table I). There was also no statistically significant difference between the two groups in terms of thrombocyte counts (*p*>0.05) (Table I). In addition, there was no statistically significant difference between the two groups with respect to leukocyte counts (*p*>0.05) (Table I). Lastly, there was no statistically significant difference in MCV levels between the two groups (*p*>0.05) (Table I).

Table I. Comparison of type 2 DM patients using metformin and type 2 DM patients using metformin combined with DPP-4 enzyme inhibitors in terms of age, hemoglobin level, MCV, leukocyte and thrombocyte levels.

Variable	DPP4 + Metformin		Metformin		Statistical analysis* Probability
	$\bar{x} \pm s.s.$	Median [IQR]	$\bar{x} \pm s.s.$	Median [IQR]	
Age (years)	60.07±11.23	59.0 [20.0]	59.75 ± 9.87	58.5 [13.0]	Z = -0.025 p = 0.980
Hemoglobin level (mg/dL)	13.58 ± 1.39	13.4 [1.8]	13.67 ± 1.38	13.4 [1.9]	Z = -0.046 p = 0.963
MCV (femtolitres)	85.15 ± 5.10	85.0 [5.0]	85.81±3.87	86.0 [5.0]	Z = -0.352 p = 0.725
Leukocyte count/mm ³	8,295.64 ± 2,430.33	7,990.0 [3,420.0]	7,578.11 ± 1,676.23	7,300.0 [2,660.0]	t = 1.780 p = 0.078
Platelet count/mm ³	263,072.73 ± 58,144.73	266,000.0 [87,000.0]	267,716.98 ± 60,589.95	270,000.0 [70,000.0]	t = -0.406

*“Independent Sample” *t*-test (*t*-table value) statistics were used to compare the measurement values of two independent groups with normally distributed data. The “Mann-Whitney U” test (*Z*-table value) statistics were used to compare the measurement values of two independent groups in the data that did not have a normal distribution.

Discussion

Insulin secretion can also be regulated by the serum concentrations of absorbable digestive products such as glucose, amino acids, and fatty acids, as well as by insulin-secreting hormones called incretins, which are secreted from intestinal enteroendocrine cells and form a branch of the enteroinsular axis. The DPP-4 enzyme can inactivate incretin hormones. DPP-4 enzyme inhibitors are widely used in order to increase the effect of incretins in the treatment of type 2 DM. While DPP-4 enzyme inhibitors may provide effective glycemic control, they are frequently preferred in the treatment of type 2 DM because they may not cause hypoglycemia and may not cause weight gain.

The dipeptidyl peptidase-4 protein is also known as cluster of differentiation 26 (CD26). Hopsu-Havu and Glenner¹¹ discovered this enzyme. Wang et al¹² found that sitagliptin helped improve radiation induced hematopoietic injury in mice. On the other side, Pitocco et al¹³ reported two type 2 DM cases with severe leucopenia that might be associated with sitagliptin. Christopherson et al¹⁴ reported that inhibition of CD26 resulted in increased responsiveness to chemokine CXCL12, also known as stromal cell-derived factor-1 (SDF-1), for unfractionated cord blood, bone marrow, or granulocyte colony-stimulating factor-mobilized peripheral blood mononuclear cells. Broxmeyer et al¹⁵ suggested that DPP-4 might be able to cleave human and mouse growth factors. O'Leary et al¹⁶ and Broxmeyer et al¹⁵ suggested that DPP-4 could have a regulatory role in hematopoiesis. In their study, Aono and Sato¹⁷ reported that the DPP-4 inhibitor linagliptin might reduce the dosage of erythropoiesis-stimulating agents (ESAs) in hemodialysis patients.

In our study, however, it was observed that there were no statistically significant differences between the two groups in terms of hemoglobin levels, leukocyte counts and platelet counts. Although there were no statistically significant differences in terms of hemoglobin, leukocyte, MCV and thrombocyte levels between the groups in our study, larger prospective human studies are needed on this topic. Since our study is a retrospective study, analyses were made in line with the currently registered patient data. In addition, patients using type 2 DM treatments other than metformin or DPP-4 enzyme inhibitors were not considered as inclusion or exclusion factors in the study. Further prospective studies that will com-

pare the hemogram parameters before and after the start of DPP-4 enzyme inhibitor therapy in this area will reveal more accurate results.

Conclusions

When there are abnormalities in complete blood count parameters in type 2 DM patients using DPP-4 enzyme inhibitors, the current picture should not be directly linked to the drug with DPP-4 enzyme inhibitor, and non-drug etiologies that may cause abnormal levels in complete blood count (CBC) parameters should be carefully investigated first.

Conflict of Interest

The Authors declare that they have no conflict of interests.

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Ethics Approval

A written approval for the study was obtained from the local Ethics Committee of the Hospital (Sağlık Bilimleri Üniversitesi Dr. Abdurrahman Yurtarslan Onkoloji Sağlık Uygulama ve Araştırma Merkezi Klinik Araştırmalar Etik Kurulu). Ethics committee decision No. 2019-10/419; decision date: 16.10.2019.

Availability of Data and Materials

Data are available upon reasonable request to the corresponding author. De-identified data are available upon reasonable request to the corresponding author. De-identified data can only be shared with the consent of all authors and with the permission of the Ankara Dr. Abdurrahman Yurtarslan Oncology Training and Research Hospital. De-identified data will not be saved more than two years by the researchers.

ORCID ID

Fatma Avcı Merdin: 0000-0003-4753-1354; Alparslan Merdin: 0000-0003-1689-630X.

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