

# Evaluation of plasma eosinophil count and mean platelet volume in patients with coronary slow flow

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**OBJECTIVE:** The pathophysiology of coronary slow flow has not been clearly defined, although multiple abnormalities including arteritis, endothelial dysfunction, and atherothrombosis, have been reported. It is known that eosinophils play an important role in inflammation, endothelial dysfunction, and thrombosis. We aimed to compare the eosinophil counts of coronary slow flow patients *versus* healthy controls.

**METHODS:** This study included 50 coronary slow flow patients (19 males, mean age  $65.6 \pm 13.7$  years) and 30 healthy controls (10 males, mean age  $57.86 \pm 11.6$  years). These participants were evaluated using concurrent routine biochemical tests as well as neutrophil, lymphocyte, and eosinophil counts and mean platelet volume (MPV), which were obtained from the whole blood count. These parameters were compared between groups.

**RESULTS:** The baseline characteristics of the study groups were comparable. The coronary slow flow patients had a higher mean platelet volume and eosinophil count than the control group ( $8.38 \pm 0.86$  vs  $6.28 \pm 1.6$  fL and  $0.31 \pm 0.42$  vs  $0.09 \pm 0.05$ ;  $p < 0.001$  and  $0.008$ , respectively).

**CONCLUSION:** Our study demonstrated a relationship between eosinophil count and MPV in patients with coronary slow flow.

**KEYWORDS:** Coronary Slow Flow; Eosinophil; Mean Platelet Volume; Inflammation; Endothelial Dysfunction.

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## INTRODUCTION

Coronary slow flow (CSF) is characterized by the delayed opacification of coronary arteries in the absence of obstructive coronary artery disease (CAD) on coronary angiography (1).

Several mechanisms have been proposed for the etiology of CSF, including microvascular and endothelial dysfunction, small vessel disease, diffuse atherosclerosis, and inflammation. However, its etiopathogenesis remains unclear (2,3). Coronary atherosclerosis, disturbed microvascularization functions, and endothelial dysfunction have been reported to be closely related to CSF (4,5). A significant relationship has also been reported between inflammatory markers and coronary flow rate (6,7).

Eosinophils are known to play an important role in endothelial dysfunction, inflammation, and thrombosis (8,9). Additionally, vascular anomalies, such as aneurysms,

have been noted in patients with hypereosinophilic syndromes (10,11).

The powerful vasoactive and procoagulant effects of eosinophils led us to hypothesize that a correlation exists between eosinophil concentrations and CSF. To our knowledge, to date, no studies have examined the relationship between blood eosinophil concentration and CSF. In our study, we compared eosinophil counts between CSF patients and healthy controls.

## MATERIALS AND METHODS

### Patients

The study group included 50 patients (19 males, mean age  $65.6 \pm 13.7$  years) with isolated CSF without any stenotic lesions under visual assessment. The control group consisted of 30 age- and gender-matched subjects (10 males, mean age  $49.16 \pm 9.2$  years) with normal coronary angiograms.

In both groups, the indication for coronary angiography was the presence of typical angina or positive or equivocal results for myocardial ischemia on noninvasive screening tests.

Both groups underwent medical history reviews and physical, blood biochemistry, and transthoracic echocardiographic examinations to exclude systemic diseases. The following exclusion criteria were applied: obstructive CAD

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(coronary stenotic lesions >20%), chronic renal failure, chronic liver disorders, chronic lung disease, moderate or severe valvular disease, hypertension, diabetes mellitus, congenital heart disease; left ventricular systolic dysfunction on echocardiography (EF <50%), anemia, pregnancy, obstructive sleep apnea, hematological disorders, known malignancy, thyroid dysfunction, hypercholesterolemia, electrolyte imbalance, and a drug history that included anti-gout, anti-inflammatory (steroid or nonsteroidal), antiaggregant, anticoagulant, or antihistamine agents or any medication that could potentially interfere with the measurement of eosinophil counts. Patients with recent histories of acute infection and/or a high body temperature (>38°C) or an inflammatory or allergic disease were also excluded from the study.

Patients with systolic blood pressure ≥140 mmHg and/or diastolic blood pressure ≥90 mmHg and those taking antihypertensive drugs were considered to be hypertensive. Diabetes was defined as a fasting blood glucose level >126 mg/dl or current use of a diet or medication to lower blood glucose. Current cigarette smoking was defined as >10 cigarettes/day at the time of diagnosis.

### Coronary angiography

Coronary angiograms were performed with a femoral approach using the Judkins technique, without the use of nitroglycerin, adenosine, or calcium channel blockers. All patients in the study population underwent elective coronary artery angiography using a Siemens Axiom Artis DFC (Siemens AG, Forchheim Germany) following appropriate patient preparation. Coronary angiograms were judged by smooth appearance, luminal wall irregularities, epicardial local or diffuse caliber reduction, and stenosis. Coronary arteries were observed in at least four views of the left coronary system using 6-French left coronary catheters, and two views of the right coronary artery were obtained using 6-French right coronary catheters with a 15 fps rate in the same cardiac catheterization laboratory. Coronary blood flow was measured quantitatively using the thrombolysis in myocardial infarction frame count (TFC). The initial frame count is defined as the frame in which concentrated dye occupies the full width of the proximal coronary artery lumen, touching both borders of the lumen, and exhibiting forward motion down the artery. The final frame is designated when the leading edge of the contrast column initially arrives at the distal end. The distal end was defined as the distal bifurcation for the left anterior descending (LAD) artery, the distal bifurcation of the segment with the longest total distance for the circumflex artery (CX), and the first branch of the posterolateral artery for the right coronary artery (RCA). The LAD coronary artery is usually longer than the other major coronary arteries; the TFC for this vessel is often higher. To obtain the corrected TFC for the LAD coronary artery, the TFC was divided by 1.7. The mean TFC for each patient and control subject was calculated by adding the TFC for the LAD, CX, and RCA and then dividing three by the obtained value. Because of the different durations required for normal visualization of the coronary arteries, the corrected cutoff values were 36.28 ± 2.6 frames for the LAD, 22.28 ± 4.1 frames for the CX, and 20.48 ± 3 frames for the RCA, as previously reported in the literature (12). All participants with a TFC greater than two standard deviations of the previously published range for a particular vessel were considered to have CSF. Any

values obtained above these thresholds in one of the three coronary arteries (not all three) were considered to indicate CSF in our study. Coronary angiograms and TFC were analyzed by two experienced interventional cardiologists who were blinded to the clinical status and laboratory measurements of the subjects.

### Laboratory tests

Biochemical parameters were analyzed spectrophotometrically using an Architect C16000 (Abbott, USA) Auto-Analyzer following an enzymatic-colorimetric assay.

For the whole blood count (i.e., eosinophil count, hematocrit, hemoglobin, MCV, MPV, leukocytes, neutrophils, lymphocytes, and platelets), blood samples were collected into tubes containing ethylenediaminetetraacetic acid (EDTA) and analyzed within 2 hours after blood sampling with a CELL-DYN 3700 (Abbott, USA) device using the impedance and optic scatter method.

### Statistical analysis

The SPSS 16.0 statistical program (SPSS Inc., Chicago, IL, USA) was used for the statistical analyses. All values are given as the mean ± standard deviation. The mean values of continuous variables were compared between groups using Student's t test or the Mann-Whitney U test, based on whether the values were normally distributed, as indicated by the Kolmogorov-Smirnov test. To define the relationship between CSF and possible confounding factors, logistic regression analysis was used and *p* < 0.05 was considered to be significant.

## RESULTS

The basic clinical and demographic characteristics were compared, and there were no statistically significant between-group differences in terms of age, gender distribution, body mass index, smoking status, or biochemical parameters (Table 1).

The TFC for all epicardial coronary arteries and the mean TFC were significantly greater in the CSF group than in the

**Table 1 - A comparison of the basic clinical and biochemical features of the patients and controls.**

	Patients (n = 50)	Controls (n = 30)	<i>p</i> value
Age (years)	65.6 ± 13.7	57.86 ± 11.6	NS
Sex (n, %) males	19 (38%)	10 (33%)	NS
Body mass index (BMI) (kg/m <sup>2</sup> )	29.9 ± 6.3	28.5 ± 4.6	NS
Smoking	10 (20%)	6 (20%)	NS
Fasting glucose (mg/dl)	92 ± 7	97.6 ± 8.5	NS
Creatinine (mg/dl)	0.78 ± 0.3	0.72 ± 0.2	NS
Total cholesterol (mg/dl)	202 ± 55	181 ± 36	NS
Triglycerides (mg/dl)	166.5 ± 52	151.9 ± 41	NS
TSH (μIU/mL)	1.8 ± 0.5	1.6 ± 0.4	NS
cLAD TFC	37.2 ± 6.9	18.1 ± 4.6	<0.001
Cx TFC	31.5 ± 6.1	19.3 ± 3.6	<0.001
RCA TFC	27.6 ± 5.1	18.5 ± 3.3	<0.001
Mean TFC	31.1 ± 3.3	18.6 ± 2.3	<0.001

TSH: thyroid-stimulating hormone; NS: nonsignificant; LAD: left anterior descending artery; cLAD: corrected TIMI frame count for LAD; TFC: TIMI frame count, Cx: circumflex artery, RCA: right coronary artery.



control group. The mean TFC of all CSF patients was greater than that of the control group (Table 1).

Regarding the blood count parameters, in the CSF patient group, the blood eosinophil count and MPV were significantly higher compared with those in the control group. There was no statistically significant difference between the two groups with regard to the leukocyte count, platelet count, hemoglobin, or hematocrit level (Table 2).

Using logistic regression analysis, eosinophil count and MPV were found to be significantly associated with CSF (Table 3).

## DISCUSSION

The main finding of this study was that the eosinophil count and MPV increased in the CSF patients compared with the healthy controls. To the best of our knowledge, our study is the first report to focus on the relationship between the eosinophil count and CSF.

In the present study, we identified an independent association between the eosinophil count and CSF. We demonstrated that CSF patients had an increased eosinophil count compared with the controls, who exhibited normal coronary arteries.

The pathophysiology of CSF has not been clearly identified, although multiple abnormalities, including inflammation, oxidative stress, endothelial dysfunction, vasculitis, platelet function disorder, and atherothrombosis, have been reported (13-15).

Previous studies have demonstrated that the C-reactive protein (CRP) and interleukin 6 levels and the neutrophil lymphocyte ratio (NLR) are greater in CSF patients than in healthy controls. The CRP and NLR increases may suggest that these markers can be used in clinical practice to assess the inflammatory statuses of CSF patients (1,16,17).

Eosinophils induce the activation of the coagulation system and platelets, as well as inflammation, endothelial dysfunction, and aneurysm. Additionally, eosinophils play a role in vascular injury (10).

To our knowledge, no studies in the literature have examined the association between CSF and eosinophil count.

In our study, we found significant MPV differences between the CSF patients and controls. Our findings align with those of previous studies (18,19). Additionally, when the two groups were compared in our study, the eosinophil counts of the CSF patients were significantly greater than those of the controls.

Eosinophils contain several granule-associated molecules that play a role in the occurrence of thrombosis and vascular injury. Eosinophils can increase the risk of thrombosis through leukocyte and platelet stimulation and the release

**Table 2 - A comparison of the whole blood count features of the patients and controls.**

	Patients (n = 50)	Controls (n = 30)	p value
Leukocyte (10 <sup>3</sup> /μl)	7.83 ± 5.5	7.72 ± 5.4	NS
Eosinophil count (10 <sup>3</sup> /μl)	0.31 ± 0.42	0.09 ± 0.05	0.008
Mean platelet volume (MPV) (fL)	8.38 ± 0.86	6.28 ± 1.6	<0.001
Hemoglobin (g/dL)	13.9 ± 1.71	13.59 ± 1.59	NS
Hematocrit (%)	41.3 ± 4.2	40.7 ± 3.49	NS
Platelet (10 <sup>3</sup> /μl)	266 ± 82	236 ± 73	NS

NS: nonsignificant.

**Table 3 - Multiple logistic regression analysis of CSF.**

	Odds Ratio	95% Confidence Interval	p value
Eosinophils	1.02	1.006-1.018	0.076
Mean platelet volume	4.2	2.20-8.42	<0.001
Leukocytes	0.95	0.91-1.0	0.130
Hemoglobin	1.007	0.07-1.20	0.734
Platelets	1.01	1.005-1.017	0.570

of tissue factor (20,21). These effects contribute to procoagulation by preventing the activation of thrombin and by inducing fibrin formation. Eosinophils store and release tissue factor and other cationic proteins. Major basic protein and eosinophilic cationic protein activate platelets and promote thrombus formation by inhibiting thrombomodulin in hypereosinophilic syndromes and allergic diseases. Reportedly, activated eosinophils and secreted eosinophil granule proteins are most evident within necrotic and later-stage thrombotic lesions and are found primarily within the areas of acute tissue damage in the endocardium and in the walls of small blood vessels. These findings suggest that eosinophil granule proteins are involved in vascular injury and that eosinophils may affect the cardiovascular system by inducing inflammatory cell infiltration (22,23).

Recent studies have reported that eosinophils are associated with arterial tortuosity, dilatation, thrombosis, cardiac syndrome X, and non-dipper hypertension (24,25).

Major basic protein, eosinophilic cationic protein, and eosinophil-derived neurotoxin are the primary mediators of eosinophil-associated toxicity to human tissue and may induce eosinophilic myocarditis, pneumonitis, dermatitis, neuropathy, and vasculitis (10).

The powerful vasoactive, inflammatory, and procoagulant effects of eosinophils led us to hypothesize that a correlation exists between the eosinophil concentration and CSF. In the literature, no studies have examined the association between CSF and eosinophils. Therefore, our study is of particular importance, as we investigated the association between eosinophil concentration and CSF in our patients.

The most important restriction of our study is the limited number of patients. Furthermore, the angiographic diagnosis of normal coronary arteries was based on axial contrast angiography of the vessel lumen, which underestimates the presence of atherosclerotic plaques. Additional studies are required to determine the relationship between eosinophil count and CSF.

The eosinophil count is increased in CSF patients, and it is significantly and independently associated with CSF. Our results may contribute to the understanding of the etio-pathogenesis of CSF and the pathophysiological mechanisms underlying the increased prevalence of cardiovascular morbidity and increased mortality risk in these patients. The increased concentration of eosinophils might be associated with the vascular destruction, endothelial dysfunction, and thrombosis observed in CSF patients.

## AUTHOR CONTRIBUTIONS

Demir M was responsible for the study design, statistical analysis, manuscript preparation and data interpretation. Coşar S was responsible for data collection and literature search. Melek M was responsible for data interpretation.



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