Research Article

Correlation between Imaging Morphological Findings and Laboratory Biomarkers in Patients with Diabetic Macular Edema

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Purpose. To investigate the potential association between peripheral blood biomarkers and morphological characteristics of retinal imaging in patients with diabetic macular edema (DME). *Methods.* Participants in this cross-sectional study were 36 consecutive patients (36 eyes) with treatment-naïve DME, who underwent spectral domain-optical coherence tomography (SD-OCT), fundus photography, and fundus fluorescein angiography (FFA). In addition, peripheral blood samples were taken to evaluate full blood count and biochemical parameters. Correlation between imaging characteristics and laboratory parameters was examined. *Results.* Eyes with central subfield thickness greater than $405\,\mu$m presented significantly higher neutrophils/lymphocytes ($p = 0.043$) and higher lipoprotein (a) compared to eyes with CST $< 405\,\mu$m ($p = 0.003$). Presence of hyperreflective foci on SD-OCT was associated with significantly higher white blood cell count ($p = 0.028$). Ellipsoid zone disruption was associated with significantly lower hematocrit ($p = 0.012$), hemoglobin ($p = 0.009$), and red blood cell count ($p = 0.026$), as well as with higher lipoprotein (a) ($p = 0.015$). Macular ischemia on FFA was associated with significantly higher monocytes ($p = 0.027$) and monocytes/HDL ($p = 0.019$). No significant associations were found between laboratory parameters and subretinal fluid, intraretinal fluid, exudates, cysts, disorganization of inner retinal layers, epiretinal membrane, and external limiting membrane condition. *Conclusion.* Specific imaging morphological characteristics were found to be associated with laboratory parameters in patients with DME. These findings may shed light on the pathophysiology of DME and its correlation with the development of specific clinical signs.

1. Introduction

Diabetic macular edema (DME) is the most common cause of visual impairment in patients with diabetes mellitus (DM), characterized by exudation and accumulation of extracellular fluid in the macula [1, 2]. The overall prevalence of DME in patients with DM has been estimated to be about 7-14%, while it varies from 0% to 3% in patients with recent DM diagnosis and increases to 28% in patients with DM for more than 20 years [1–5].

In the pathogenesis of DME, chronic hyperglycemia promotes a cascade of biochemical pathways and consequent structural alterations in the retinal blood vessels' wall, including the loss of pericytes and the breakdown of the blood-retinal-barrier, leading to retinal vascular permeability [6, 7]. This breakdown is mainly driven by the production of inflammatory cytokines, with vascular endothelial growth factor (VEGF) to be the most prominent [6, 7]. Moreover, it has been shown that patients with DME have increased levels of proinflammatory mediators in aqueous humor com-

pared to non-DME cases [8], while there is a controversy whether the pathophysiology of DME is mainly attributed to such systemic affection or to a local intraocular response. Of note, several studies have shown that elevated serum lipids, including cholesterol and low-density lipoprotein (LDL), demonstrated a significant association with retinal hard exudates and formation of DME [9–12], while elevated IL-6 has also been correlated with diffuse retinal thickness or severity of DME [13, 14].

Nowadays, there is a great development in retinal imaging, especially with the advent of spectral domain-optical coherence tomography (SD-OCT) and swept source-OCT (SS-OCT) [15, 16], as well as OCT angiography [17, 18]. Both OCT and OCTA are noninvasive techniques, enabling the identification of specific morphological characteristics of DME, while OCTA allows the quantification of the foveal avascular zone (FAZ) area besides vessel density [18].

Given the current understanding of DME pathogenesis, although much reported information exists on intraocular biomarkers in patients with DME, literature is scarce regarding systemic biomarkers and their correlation with morphological characteristics in retinal imaging. Ghosh et al. examined the relationship between different OCT patterns of DME and systemic risk factors in patients with DME and did not identify any modifiable systemic factor for any of the OCT patterns in DME [19].

Based on the above, the purpose of the present study was to investigate the potential association between peripheral blood biomarkers and morphological characteristics of retinal imaging in patients with DME.

2. Methods

Participants in this observational, cross-sectional study were 36 consecutive patients with DM type 2 and treatment naïve DME, who were diagnosed and treated at the 2nd Department of Ophthalmology, National and Kapodistrian University of Athens, Athens, Greece, between 1st September 2019 and 31st March 2020. The study protocol adhered to the tenets of the Declaration of Helsinki and was approved by the Institutional Review Board. Written informed consent was obtained from all participants.

All patients had nonproliferative diabetic retinopathy (NPDR) and treatment naïve DME, confirmed on SD-OCT, revealing central subfield thickness (CST) $\geq 320\,\mu$m. One eye of each patient was included. In cases of bilateral DME, the right eye was chosen, so as to avoid selection bias. Patients with other vitreoretinal diseases, uveitis, media opacities, previous vitreoretinal surgery, previous laser photocoagulation, or ocular surgery in the previous 6 months were excluded.

Demographic data of patients (age, gender) were recorded, along with the duration of DM. All participants underwent a complete ophthalmologic examination, including best-corrected visual acuity (BCVA) measurement by means of Snellen's charts (converted to logMAR scale), slit-lamp examination, dilated fundoscopy, color fundus photography using Topcon TRC-50DX (Topcon Corporation), SD-OCT, and fundus fluorescein angiography (FFA) using Spectralis (Spectralis HRA+OCT, Heidelberg Engi-

neering, Heidelberg, Germany). SD-OCT was obtained using a standard acquisition protocol; six radial scans 3 mm long were performed at equally spaced angular orientations centered on the foveola. The OCT volume scan was performed on a 20×20 degree cube, consisted of 49 horizontal B-scans with 20 averaged frames per B-scan centered over the fovea. The following SD-OCT variables were recorded at baseline: CST (μm), presence of intraretinal fluid (IRF), subretinal fluid (SRF), cysts, hyperreflective foci (HF), and disorganization of the inner retinal layers (DRIL) and epiretinal membrane (ERM). Ellipsoid zone (EZ) and external limiting membrane (ELM) condition were also assessed. In addition, the presence of exudates on color fundus photography was recorded. The severity of DR was based on color fundus photography and on FFA and was graded according to the international diabetic retinopathy disease severity scale [20]. Moreover, macular ischemia was evaluated on FFA and defined as disruption and enlargement of foveal avascular zone (FAZ). Two investigators (IC, ED) independently evaluated qualitatively the SD-OCT images, the fundus photographs, and the FFA images. The interobserver agreement ranged from very good to perfect for all SD-OCT parameters ($k = 0.999$ for IRF; $k = 0.999$ for SRF; $k = 0.872$ for HF; $k = 0.851$ for DRIL; $k = 0.999$ for ERM; $k = 0.902$ for EZ condition; and $k = 0.883$ for ELM condition), as well as for DR severity assessment on fundus photographs ($k = 0.901$) and ischemia evaluation on FFA ($k = 0.935$).

At the same day and following an eight hour overnight fast, all patients underwent a forearm venous puncture for peripheral blood extraction and serum was separated. Full blood count was measured on a Sysmex XE-2100 analyzer (Sysmex Corp. Kobe, Japan), while all biochemical analyses were performed on a Roche Cobas 8000 (Roche, Chicago, IL, USA) in the laboratory of Attikon University Hospital. Specifically, we analyzed the following parameters: glucose, glycated Hb (HbA1c), urea, creatinine, cholesterol, HDL, LDL, triglycerides, apolipoprotein A, apolipoprotein B, lipoprotein (a), homocysteine, vitamin D, and IL-6.

2.1. Statistical Analysis. For the description of patients' characteristics, descriptive statistics were calculated; mean ± standard deviation (SD) was used for continuous variables, while relative frequencies and percentages for categorical variables were reported. All variables were tested for normal distribution with the Shapiro–Wilk test. The associations between laboratory and imaging variables were evaluated with the Mann–Whitney–Wilcoxon test (MWW), as appropriate. In addition, Pearson's chi-squared test (P) or Fisher's exact test (F) were also appropriately implemented. According to the *a priori* power calculation, a sample size of 36 eyes was adequate to achieve 80% power for the detection of an effect size larger or equal to 1.05 (in simple terms, a difference of $1.05 * $SD between the compared subgroups), assuming an application of the two-tailed Mann–Whitney–Wilcoxon test for equally sized subgroups at the 5% level of significance. The sample size calculation was performed with G * Power 3.1.9.2 software (University of Dusseldorf, Germany). Statistical analysis was performed using STATA/SE 13 statistical software (Stata Corporation, College

TABLE 1: Demographic and clinical characteristics of the study sample ($n = 36$ patients with diabetic macular edema).

Age (mean ± SD, years)	64.2 ± 8.5
Gender (n, %)	
Male	21 (58.3%)
Female	15 (41.7%)
Duration of diabetes mellitus (mean ± SD, years)	11.7 ± 4.7
HbA1c (mean ± SD, %)	8.4 ± 1.9
Stage of diabetic retinopathy (n, %)	
Mild	10 (27.8%)
Moderate	17 (47.2%)
Severe	9 (25.0%)
Best-corrected visual acuity (mean ± SD, logMAR)	0.51 ± 0.29
Imaging characteristics	
Central subfield thickness (mean ± SD, μm)	439.2 ± 79.1
Intraretinal fluid (n, %)	36 (100%)
Subretinal fluid (n, %)	9 (25%)
Cysts (n, %)	2 (5.6%)
Hyperreflective foci (n, %)	15 (41.7%)
Exudates (n, %)	13 (36.1%)
Disorganization of inner retinal layer (n, %)	6 (16.7%)
Epiretinal membrane (n, %)	2 (5.6%)
Ellipsoid zone condition (n, %)	
Intact	23 (63.9%)
Disrupted	13 (36.1%)
External limiting membrane condition (n, %)	
Intact	26 (72.2%)
Disrupted	10 (27.8%)
Macular ischemia (n, %)	9 (25%)

Station, TX, USA). A p value < 0.05 was considered statistically significant.

3. Results

The demographic and clinical characteristics of the study sample are shown in Table 1. The mean age of patients was 64.2 ± 8.5 years. 58.3% of patients were male and 41.7% female. The mean duration of DM was 11.7 ± 4.7 years. The mean BCVA was 0.51 ± 0.29 logMAR, while the mean CST was 439.2 ± 79.1 μm.

Regarding the potential association between imaging characteristics and laboratory variables, no significant correlations were found for IRF, SRF, exudates, cysts, DRIL, ERM, and ELM condition.

Table 2 shows the comparison of laboratory parameters between eyes with HF ($n = 15$) and without HF ($n = 21$). Eyes with HF presented significantly higher white blood cell (WBC) count compared to those without HF ($p = 0.028$, MWW).

Table 3 shows the comparison of laboratory parameters between eyes with intact EZ ($n = 23$) and those with disrupted EZ ($n = 13$). Eyes with disrupted EZ presented significantly lower hematocrit ($p = 0.012$, MWW) and hemoglobin

TABLE 2: Association between laboratory variables and hyperreflective foci on optical coherence tomography. Laboratory variables are summarized as median (IQR: interquartile range).

	HF present ($n = 15$)	HF absent ($n = 21$)	p value (MWW)
Red blood cells ($10^6/\mu$l)	4.65 (0.61)	4.73 (0.91)	0.824
White blood cells ($10^3/\mu$l)	8.54 (2.10)	7.29 (2.26)	**0.028**
Neutrophils ($10^3/\mu$l)	5.24 (2.08)	4.37 (1.41)	0.124
Lymphocytes ($10^3/\mu$l)	2.30 (1.01)	1.91 (0.61)	0.096
Monocytes ($10^3/\mu$l)	0.63 (0.20)	0.51 (0.27)	0.260
Platelets ($10^3/\mu$l)	248 (37)	219 (78)	0.073
Hematocrit (%)	40.6 (4.1)	40.7 (3.5)	0.547
Hemoglobin (g/dl)	13.9 (1.5)	13.4 (1.5)	0.419
Monocytes/lymphocytes	0.27 (0.14)	0.29 (0.19)	0.962
Neutrophils/lymphocytes	2.15 (0.92)	2.25 (1.36)	0.937
Monocytes/HDL	0.013 (0.006)	0.011 (0.007)	0.516
Glucose (mg/dl)	198 (87)	161 (81)	0.084
HbA1c (%)	8.8 (2.5)	7.8 (1.8)	0.281
Urea (mg/dl)	37.8 (10.8)	37.5 (23.6)	0.635
Creatinine (mg/dl)	0.8 (0.2)	0.9 (0.7)	0.168
Cholesterol (mg/dl)	172 (61)	147 (56)	0.384
LDL (mg/dl)	94 (56)	80 (44)	0.310
HDL (mg/dl)	45 (14)	47 (17)	0.975
Triglycerides (mg/dl)	170 (168)	122 (89)	0.059
IL-6 (pg/ml)	3.5 (3.7)	3.8 (2.2)	0.680
Apolipoprotein A (mg/dl)	145 (35)	141 (37)	0.334
Apolipoprotein B (mg/dl)	89 (44)	77 (34)	0.228
Lipoprotein (a) (nmol/l)	28.7 (38.6)	15.4 (107.3)	0.975
Homocysteine (μmol/l)	14.7 (7.4)	17.8 (11.2)	0.141
Vitamin D (ng/ml)	19.8 (12.7)	21.6 (18.1)	0.506

HF: hyperreflective foci; MWW: Mann–Whitney–Wilcoxon's test.

(Hb) ($p = 0.009$, MWW), as well as lower red blood cell (RBC) count ($p = 0.026$, MWW), while they had significantly higher lipoprotein (a) compared to eyes with intact EZ ($p = 0.015$, MWW).

Table 4 shows the comparison of laboratory parameters between eyes with macular ischemia ($n = 9$) and those without macular ischemia ($n = 27$). Eyes with macular ischemia had significantly higher monocytes ($p = 0.027$, MWW) and monocytes/HDL ($p = 0.019$, MWW) compared to eyes without macular ischemia.

Table 5 shows the comparison of laboratory parameters between eyes with CST above or equal to median (405 μm) and those with CST below median. Eyes with CST ≥ 405 μm presented higher neutrophils/lymphocytes ($p = 0.043$, MWW) and higher lipoprotein (a) compared to eyes with CST < 405 μm ($p = 0.003$, MWW).

Figures 1 and 2 show correlation between laboratory and morphological findings in patients with DME.

TABLE 3: Association between laboratory variables and ellipsoid zone condition on optical coherence tomography. Laboratory variables are summarized as median (IQR: interquartile range).

	EZ intact ($n = 23$)	EZ disrupted ($n = 13$)	p value (MWW)
Red blood cells ($10^6/\mu l$)	4.75 (0.71)	4.38 (0.53)	**0.026**
White blood cells ($10^3/\mu l$)	7.81 (3.01)	8.09 (2.28)	0.417
Neutrophils ($10^3/\mu l$)	4.64 (2.03)	5.29 (3.02)	0.174
Lymphocytes ($10^3/\mu l$)	2.04 (0.62)	2.18 (1.00)	0.846
Monocytes ($10^3/\mu l$)	0.54 (0.22)	0.57 (0.33)	0.818
Platelets ($10^3/\mu l$)	246 (71)	241 (31)	0.737
Hematocrit (%)	41.2 (4.4)	38.7 (3.7)	**0.012**
Hemoglobin (g/dl)	14.0 (1.2)	12.9 (1.3)	**0.009**
Monocytes/lymphocytes	0.27 (0.14)	0.28 (0.22)	0.659
Neutrophils/lymphocytes	2.25 (1.25)	2.44 (1.53)	0.548
Monocytes/HDL	0.013 (0.006)	0.013 (0.007)	>0.999
Glucose (mg/dl)	177 (87)	113 (105)	0.133
HbA1c (%)	7.6 (2.3)	9.0 (1.5)	0.331
Urea (mg/dl)	36.2 (17.4)	42.3 (18.3)	0.274
Creatinine (mg/dl)	0.8 (0.2)	1.0 (0.7)	0.143
Cholesterol (mg/dl)	153 (64)	166 (39)	0.584
LDL (mg/dl)	86 (55)	87 (43)	0.902
HDL (mg/dl)	45 (19)	50 (11)	0.584
Triglycerides (mg/dl)	131 (149)	138 (120)	0.596
IL-6 (pg/ml)	3.4 (2.1)	4.5 (8.0)	0.377
Apolipoprotein A (mg/dl)	142 (36)	143 (13)	0.750
Apolipoprotein B (mg/dl)	82 (42)	84 (25)	0.724
Lipoprotein (a) (nmol/l)	14.7 (34.8)	89.5 (188.4)	**0.015**
Homocysteine ($\mu mol/l$)	15.2 (8.0)	18.9 (8.7)	0.197
Vitamin D (ng/ml)	23 (18.1)	25.4 (9.8)	0.090

EZ: ellipsoid zone; MWW: Mann–Whitney–Wilcoxon's test.

TABLE 4: Association between laboratory variables and macular ischemia on fluorescein angiography. Laboratory variables are summarized as median (IQR: interquartile range).

	Macular ischemia presence ($n = 9$)	Macular ischemia absence ($n = 27$)	p (MWW)
Red blood cells ($10^6/\mu l$)	4.65 (0.73)	4.70 (0.82)	0.932
White blood cells ($10^3/\mu l$)	9.0 (2.49)	7.57 (1.96)	0.096
Neutrophils ($10^3/\mu l$)	5.24 (1.96)	4.57 (2.03)	0.074
Lymphocytes ($10^3/\mu l$)	2.33 (1.87)	2.03 (0.46)	0.718
Monocytes ($10^3/\mu l$)	0.64 (0.38)	0.51 (0.19)	**0.027**
Platelets ($10^3/\mu l$)	243 (86)	244 (72)	0.430
Hematocrit (%)	40.5 (6.4)	40.7 (5.3)	0.503
Hemoglobin (g/dl)	13.2 (1.4)	13.9 (1.4)	0.345
Monocytes/lymphocytes	0.29 (0.21)	0.27 (0.14)	0.159
Neutrophils/lymphocytes	2.15 (2.95)	2.25 (0.99)	0.718
Monocytes/HDL	0.016 (0.010)	0.011 (0.004)	**0.019**
Glucose (mg/dl)	198 (219)	167 (62)	0.606
HbA1c (%)	9.4 (3.0)	7.4 (1.8)	0.074
Urea (mg/dl)	40 (16.8)	35.8 (20)	0.154
Creatinine (mg/dl)	0.8 (0.5)	0.8 (0.2)	0.889
Cholesterol (mg/dl)	172 (80)	147 (55)	0.216
LDL (mg/dl)	98 (68)	80 (43)	0.287
HDL (mg/dl)	43 (16)	48 (19)	0.390
Triglycerides (mg/dl)	211 (190)	128 (70)	0.198
IL-6 (pg/ml)	4.2 (6.2)	3.0 (2.5)	0.149
Apolipoprotein A (mg/dl)	141 (33)	143 (34)	0.606
Apolipoprotein B (mg/dl)	93 (63)	81 (33)	0.363
Lipoprotein (a) (nmol/l)	34.7 (92.6)	15.4 (59.6)	0.363
Homocysteine ($\mu mol/l$)	17.8 (6.8)	15.7 (8)	0.731
Vitamin D (ng/ml)	17.9 (13)	21.6 (12.3)	0.668

MWW: Mann–Whitney–Wilcoxon's test.

4. Discussion

The principal message of the study is that most of the imaging morphological characteristics studied herein were not correlated with laboratory parameters in patients with DME, suggesting that DME may be mainly attributed to a local response more than a systemic effect. However, CST and specific OCT biomarkers, i.e., HF and EZ conditions, as well as macular ischemia on FFA, were found to be associated with laboratory findings in patients with DME, shedding light on the pathophysiology of DME and its correlation with the development of these specific clinical characteristics.

Firstly, lipoprotein (a) was found to be associated with high CST and with EZ disruption. Previous studies have shown that high lipoprotein (a) concentration was independently associated with the presence and severity of DR in patients with type 2 DM, regardless of glycemic control [21–25]. Specifically, lipoprotein (a) can affect vascular tone

and perfusion, oxidize lipids, and enhance oxidative stress via the generation of reactive oxygen species and inflammatory actions on the vascular wall [26, 27]. Moreover, lipoprotein (a) has been associated with endothelial dysfunction, suggesting that it could be an independent risk factor for diabetic microvascular complications [28, 29]. Although the exact mechanism behind the potential causal relationship between lipoprotein (a) and DME, and especially EZ disruption, remains unclear, it has been hypothesized that elevated lipoprotein (a) concentrations may play a causative role in DME by damaging the microcirculation [30]. In addition, lipoprotein (a) has been involved in the activation of acute inflammation and may be related to more severe DR, including DME severity with higher CST and EZ disruption [31]. Ellipsoid zone disruption has also been associated with decreased RBC count, decreased Hb, and consequently, decreased hematocrit. In patients with DM, RBC have been

TABLE 5: Association between laboratory variables and central subfield thickness on optical coherence tomography. Laboratory variables are summarized as median (IQR: interquartile range).

	Central subfield thickness $\geq 405\,\mu$m ($n = 18$)	Central subfield thickness $< 405\,\mu$m ($n = 18$)	p (MWW)
Red blood cells ($10^6/\mu$l)	4.6 (0.48)	4.79 (0.76)	0.097
White blood cells ($10^3/\mu$l)	7.7 (3.31)	7.87 (1.95)	0.728
Neutrophils ($10^3/\mu$l)	5.0 (3.0)	4.64 (1.74)	0.174
Lymphocytes ($10^3/\mu$l)	1.97 (0.76)	2.08 (1.02)	0.282
Monocytes ($10^3/\mu$l)	0.57 (0.29)	0.54 (0.20)	0.824
Platelets ($10^3/\mu$l)	241 (82)	251 (68)	0.359
Hematocrit (%)	40.5 (4.7)	40.9 (4.1)	0.728
Hemoglobin (g/dl)	13.3 (1.5)	13.9 (1.0)	0.516
Monocytes/lymphocytes	0.28 (0.20)	0.27 (0.17)	0.268
Neutrophils/lymphocytes	2.74 (1.34)	1.95 (0.72)	**0.043**
Monocytes/HDL	0.012 (0.007)	0.013 (0.006)	0.950
Glucose (mg/dl)	159 (92)	180 (97)	0.179
HbA1c (%)	8.2 (2.3)	7.6 (3.4)	0.924
Urea (mg/dl)	37 (19)	38.9 (17.1)	0.812
Creatinine (mg/dl)	0.9 (0.5)	0.8 (0.2)	0.608
Cholesterol (mg/dl)	163 (47)	153 (64)	0.658
LDL (mg/dl)	95 (43)	84 (55)	0.716
HDL (mg/dl)	48 (12)	45 (20)	0.924
Triglycerides (mg/dl)	137 (90)	122 (207)	0.693
IL-6 (pg/ml)	3.4 (3.6)	3.8 (1.9)	0.764
Apolipoprotein A (mg/dl)	142 (32)	144 (43)	0.937
Apolipoprotein B (mg/dl)	88 (25)	75 (56)	0.359
Lipoprotein (a) (nmol/l)	60.0 (162.1)	10.1 (17.4)	**0.003**
Homocysteine (μmol/l)	18.3 (10.9)	15.2 (6.9)	0.624
Vitamin D (ng/ml)	21.7 (13.0)	21.3 (18.7)	0.776

MWW: Mann–Whitney–Wilcoxon's test.

shown to present variations in size and diameter [32]. Chronic hyperglycemia can lead to nonenzymatic glycation of RBC membrane proteins that would accelerate RBC aging due to negative surface electrical charge [32, 33]. The RBC count has been found to be reduced in patients with microvascular complications, especially in those with longer DM duration [32–34]. Since Hb levels are directly correlated with RBC count, reduced RBC would reflect on Hb concentration and on hematocrit. In our case with DME, hyperglycemia induces the rearrangement of proteins in the plasma membrane, while cytoskeleton proteins also appear to be heavily glycosylated, affecting membrane stability, as it has been mentioned in RBC membrane proteins [32]. Therefore, reduced RBC could represent more severe DR and DME, which may be reflected on EZ disruption, as a result of cytoskeleton weakening.

An interesting finding of our study was the association between HF and WBC. Many theories have attempted to explain the pathophysiology of HF, but their precise nature remains elusive. Framme et al. suggested that HF can be leucocytes or RPE cells, indicating retinal inflammation [35]. Coscas et al. supported this concept and postulated that HF

are microglia cells activated by inflammation [36]. White blood cells and their subtypes are the biomarkers of inflammatory response because their activation leads to the synthesis of inflammatory cytokines, as it has been previously identified in patients with DR [37, 38]. Therefore, our finding that HF presence was associated with increased WBC is consistent with the hypothesis that an inflammatory component is implemented in HF pathogenesis [39–41].

In recent studies, neutrophil (an indicator of inflammation) to lymphocyte (an indicator of physiologic stress) ratio was evaluated in inflammatory diseases, such as coronary artery disease, noncardiac diseases, retinal vein occlusion, age-related macular degeneration and DR [42–47]. Of note, neutrophil-to-lymphocyte ratio is more powerful for prediction of inflammatory diseases than subtypes of WBC alone because it combines the predictive values of two parameters of WBC [44]. In DME, the chronic low-grade inflammation may lead to inflammatory cytokine release, which is commonly responsible for increased vascular permeability [7]. As a result, neutrophil-to-lymphocyte ratio may be defined as an indication of subclinical inflammation, especially in more severe DME, as it was found in our study, showing a
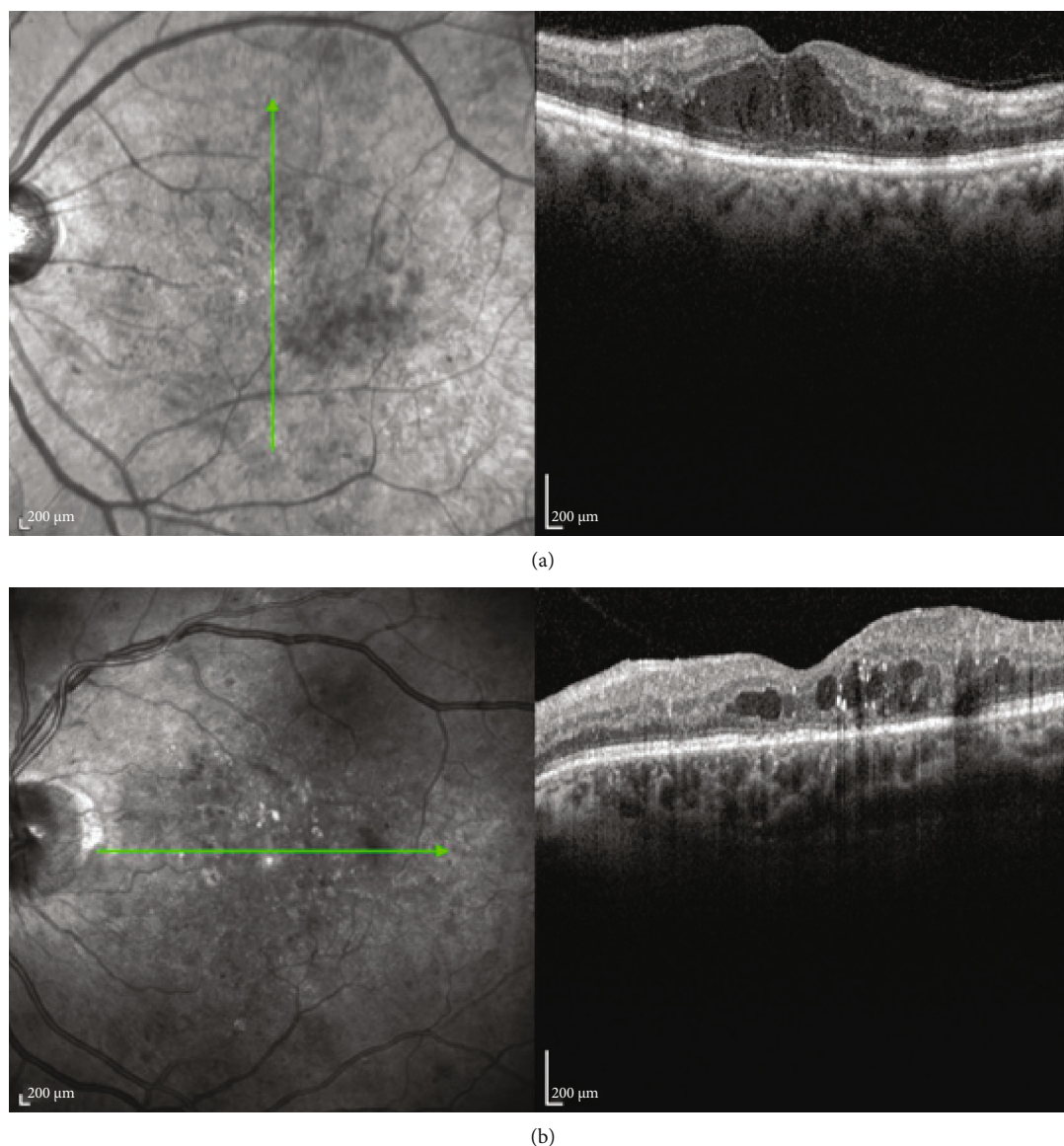
(a)

(b)

Figure 1: (A) Optical coherence tomography of a female patient with diabetic macular edema and elevated lipoprotein (a), as well as decreased hematocrit and red blood count, which was associated with the disruption of the ellipsoid zone and an increase in central subfield thickness. (B) Optical coherence tomography of a male patient with diabetic macular edema and elevated white blood cells, which were associated with the presence of hyperreflective foci.

significant association between increased neutrophil-to-lymphocyte ratio and higher CST.

Moreover, monocyte-to-HDL ratio has been investigated as a new inflammation biomarker and is considered superior to subtypes of WBC, especially in patients with cardiovascular and cerebrovascular diseases, as well as in patients with branch retinal vein occlusion [48–50]. Monocytes are indicators of inflammation, since they are responsible for inflammatory cytokine secretion, while HDL has antioxidant and anti-inflammatory effects [51, 52]. In our study, patients with macular ischemia were found to have increased monocytes, as well as monocyte-to-HDL ratio, which is consistent with other studies in patients with myocardial infarction and limb

ischemia, suggesting monocyte-to-HDL ratio as a biomarker of ischemic conditions [53, 54].

A potential limitation of this study pertains to the relatively small sample size, although our *a priori* statistical power calculation showed that it was adequate to achieve 80% power for the detection of an effect size larger or equal to 1.05; further larger studies seem necessary to validate our results.

In conclusion, this study investigated the potential correlation between imaging morphological findings and laboratory biomarkers in patients with DME. Our results showed that specific inflammatory biomarkers, such as WBC, monocytes, monocyte-to-HDL, and neutrophil-to-lymphocyte
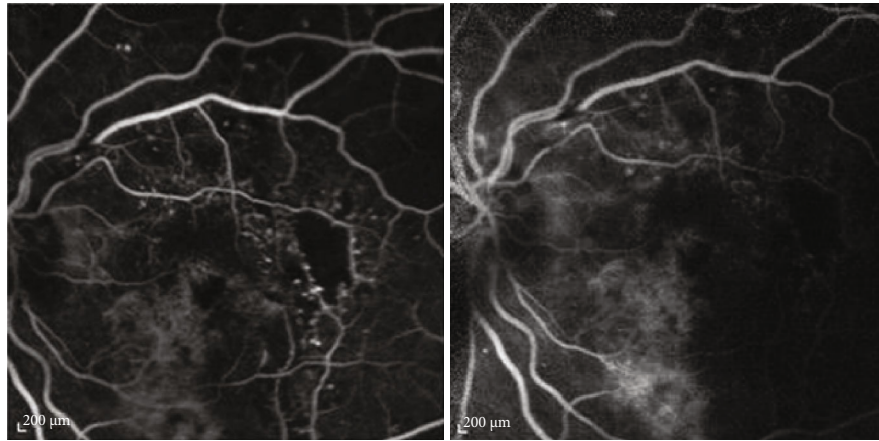
FIGURE 2: Fluorescein angiography of a male patient with diabetic macular edema, showing macular ischemia, which was associated with higher monocytes and higher monocytes/HDL.

ratios, were associated with more severe disease activity with higher CST and presence of HF and macular ischemia, while EZ disruption was found to be associated with increased lipoprotein (a) and decreased RBC, both of which were involved in microcirculation alterations in patients with DME. These findings may scrutinize the pathophysiology of DME and the pathogenesis of specific clinical signs. However, it should be noted that most of imaging biomarkers studied herein were not correlated with laboratory parameters in patients with DME, suggesting that DME may be mainly attributed to a local response more than a systemic effect. Further studies with a large sample size are needed to justify our results.

Data Availability

Data are available upon request from authors.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

Authors' Contributions

Eleni Dimitriou collected and interpreted data. Theodoros Sergentanis performed the statistical analysis and interpretation of data. Vaia Lambadiari, George Theodossiadis, and Panagiotis Theodossiadis collected data, interpreted data, and critically revised the manuscript. Irini Chatziralli conceived and designed the study; collected, analyzed, and interpreted data; drafted the manuscript; and supervised the study. Eleni Dimitriou and Theodoros Sergentanis have equal contribution.

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