Association of Body Fluid Expansion With Optical Coherence Tomography Measurements in Diabetic Retinopathy and Diabetic Macular Edema

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Purpose. To evaluate associations of body fluid status with optical coherence tomography measurements in patients with diabetic retinopathy (DR).

Methods. This prospective, cross-sectional study enrolled a total of 104 eyes from 104 patients with diabetes mellitus for fundus evaluations of DR and optical coherence tomography examinations. DR severity was graded via fundus photography. Systemic body fluid status was recorded via a body composition monitor with output values of total body water, extracellular water (ECW), intracellular water, and overhydration (OH). Relative overhydration (ROH) was defined as OH/ECW. Volume overload was defined as ROH $\geq$7%. Correlations of central subfield thickness (CST) with body fluid status were analyzed by partial correlation with adjustment for age, sex, and body mass index (BMI). Logistic regression analysis was used to evaluate factors associated with diabetic macular edema (DME).

Results. Higher levels of ECW, OH, and ROH were correlated with thick CST in patients with DR ($P = 0.006$, 0.021, and 0.008, respectively), but not in those without any DR (all $P > 0.05$), after adjusting for age, sex, and BMI. Patients with DME ($n = 31$) had higher OH than DR patients without DME ($n = 28$) or those without any DR ($n = 45$) ($P = 0.002$ and $P < 0.001$, respectively). Multiple regression model showed that volume overload was the independent factor for the presence of DME (odds ratio, 9.532; 95% confidence interval, 2.898–31.348; $P < 0.001$).

Conclusions. While both ECW and OH reflect CST in patients with DR, overhydration had particularly strong associations with DME. This study provides a novel insight into our current understanding regarding the pathogenesis for DME.

Keywords: central subfield thickness, diabetes mellitus, diabetic retinopathy, diabetic macular edema, relative overhydration, volume overload

Diabetic retinopathy (DR) and its sequelae are a major contributor to visual loss worldwide.[1] These conditions occur basically owing to a prolonged hyperglycemic state, and an increase in oxidative stress that causes vascular damage. Diabetic macular edema (DME), however, appears to be more complex and multifactorial.[2] Clinically, it is characterized by the formation of intraretinal cysts around the fovea, with or without the presence of hard exudate and subretinal fluid, which can be easily visualized via modern optical coherence tomography (OCT).[3] Thickening of the fovea leads to a decrease in visual acuity. Although concepts of DME occurrence have continued to evolve over the past few years,[4] the pathogenesis has not yet been fully established. Retinal capillary nonperfusion and ischemia upregulate the expression of vascular endothelial growth factors (VEGFs) and various inflammatory cytokines, which subsequently increase vascular permeability and lead to the breakdown of the blood-retinal barrier.[5,6] Based on these considerations, intravitreal anti-VEGF agents and steroid injections have been the mainstay of treatment.[7,8] While current treatment modalities address the components of the blood-retinal barrier, edema may not resolve sometimes even after repeated injections of these agents.

It is worth noting that retinal edema may not necessarily develop merely as a result of barrier breakdown in the absence of excessive fluid accumulation, suggesting other factors beyond VEGF may also be involved.[9] Extracellular water has been thought to be the major fluid compartment in retinal parenchyma. The idea of fluid trafficking in the pathogenesis of DME has emerged recently. Starling's rule plays an important role in determining the direction of fluid movement, which is largely driven by the differences between hydrostatic pressure and osmotic pressure across a semipermeable membrane.[4,10,11] The application of this rule to the flow of extracellular fluid possibly sheds light on previous observations that tight glycemic control does not seem to have a positive impact on the resolution of edema.[12,13] At a cellular level, previous research has suggested that intracellular edema may precede the development of macular edema.[9] Retinal ischemia may alter the expression of water channel proteins, particularly aquaporin-4, and potassium channels, in Müller cells, causing osmotic and water imbalance.[14,15] Swelling of these cells leads to the disruption of the retinal structure, and failure of the glial cell support may lead to excessive fluid accumulation.

Table 1. Comparisons of Diabetic Patients According to Severity of Diabetic Retinopathy

Clinical Factor	No DR, $n = 45$	DR Without DME, $n = 28$	DR With DME, $n = 31$	*P* Value
Age, y	54.9 ± 12.6	61.3 ± 10.1	59.4 ± 11.9	0.12
Male sex, *n* (%)	26 (57.8)	15 (53.6)	22 (71.0)	0.35
Body weight, kg	73.1 ± 17.5	69.0 ± 14.8	69.3 ± 15.0	0.36
BMI, kg/m^2	27.4 ± 5.9	26.9 ± 6.0	25.8 ± 4.2	0.37
Obesity, *n* (%)	17 (37.8)	8 (28.6)	8 (25.8)	0.5
HbA1c, %	8.0 ± 2.3	9.1 ± 2.6	8.0 ± 2.4	0.044
Serum creatinine, mg/dL	1.0 ± 0.4	1.6 ± 1.6	1.5 ± 1.2	0.08
eGFR, mL/min/1.73 m^2	81.5 ± 20.7	67.7 ± 32.9	63.9 ± 30.9	0.023
eGFR <60, *n* (%)	7 (15.6)	9 (32.1)	15 (48.4)	0.008
Hypertension, *n* (%)	6 (13.3)	4 (14.3)	4 (12.9)	0.99
Systolic BP, mm Hg	132 ± 19	137 ± 18	142 ± 21	0.08
Diastolic BP, mm Hg	78 ± 12	74 ± 14	79 ± 11	0.26
CAD, *n* (%)	9 (27.3)	9 (27.3)	15 (45.5)	0.033
DM >10 y, *n* (%)	5 (13.9)	15 (41.7)	16 (44.4)	<0.001
Metabolic control, *n* (%)				
OHA	40 (44.9)	23 (25.8)	26 (29.2)	0.69
Insulin dependence	11 (25.6)	16 (37.2)	16 (37.2)	0.009
LLD	31 (47.7)	16 (24.6)	18 (27.7)	0.5
Diuretics	1 (2.2)	3 (10.7)	6 (19.4)	0.044
Body fluid status				
TBW, l	41.6 ± 10.6	39.4 ± 7.7	43.7 ± 8.7	0.3
ECW, l	17.6 ± 3.7	16.7 ± 3.0	19.4 ± 4.1	0.04
ICW, l	24.0 ± 7.0	22.7 ± 5.1	24.2 ± 5.3	0.61
OH, l	−0.3 ± 1.7	−0.3 ± 2.0	1.9 ± 2.9	<0.001
ROH, OH/ECW, %	−1.5 ± 8.8	−2.3 ± 13.1	8.7 ± 12.6	<0.001
Volume overload, *n* (%)	5 (18.5)	5 (18.5)	17 (63.0)	<0.001
OCT values				
CST, μm	255 ± 20	255 ± 23	409 ± 130	<0.001
Cube volume, mm^3	9.9 ± 0.6	10.1 ± 0.8	12.9 ± 2.3	<0.001

DM, diabetes mellitus; LLD, lipid-lowering drug; OHA, oral hypoglycemic agent.

The differential body fluid compartments can be quantitatively measured by a body composition monitor with bioimpedance analysis.[16,17] This noninvasive device has been used in a number of clinical settings, such as determination of hydration status in patients with chronic kidney disease and the volume of fluid to be removed before hemodialysis for patients with end-stage renal disease.[18] Nevertheless, the association between various fluid compartments and retinal edema has not been explored yet from basic perspectives. On the basis of the hypothesis of volumetric effect on retinal edema mentioned above, we aimed in the present study to investigate the association between the body fluid status in a group of diabetic patients by using central retinal thickness and volume.

Materials and Methods

Study Population

This prospective, cross-sectional study enrolled a total of 104 eyes from 104 patients, from January 2017 to March 2018. For statistical reasons, if both eyes from each patient were eligible for the study, only the eye with greater central subfield thickness was included for analysis. The inclusion criterion was diabetes mellitus with or without DR or DME. Serum creatinine and glycosylated hemoglobin (HbA1c) data for the most recent month were available. Estimated glomerular filtration rate (eGFR) was calculated from serum creatinine by using the Modification of Diet in Renal Disease (MDRD) formula.[19] Obesity was defined by a value of body mass index (BMI) of 28 kg/m^2 or more, which has been validated in Chinese populations.[20] Hypertension was defined by values of both systolic and diastolic blood pressure (BP) of 140/90 mm Hg or more. Status of coronary artery disease (CAD) was evaluated and obtained from medical records including ICD-9 coding 414.01 (equivalent to ICD-10 I25.1 and its subcategories). Use of diuretics was also reviewed from medical records to investigate possible associations with macular edema. We excluded patients with any previous intraocular surgery as well as intravitreal injections of anti-VEGF or steroids; previous panretinal photocoagulation within the prior 6 months; severe vitreous hemorrhage that obscured the fundus from evaluation; or other ocular pathology such as epiretinal membrane, retinal vein occlusion, macular degeneration, or uveitis. To minimize fluctuations of body fluid that may potentially cause inaccuracy, we also excluded patients under hemodialysis, and patients with diabetic ketoacidosis and other conditions requiring intravenous fluid challenge. Informed consent was obtained from each patient. This research adhered to the tenets of the Declaration of Helsinki and was approved by the Institutional Review Board of Taipei Tzu Chi Hospital, Buddhist Tzu Chi Medical Foundation.

Measurements of DR Severity and DME

Dilated fundus photography was performed to evaluate the status of retinopathy according to the Early Treatment Diabetic Retinopathy Study (ETDRS) grading system described in the previous literature.[21] Macular exudate was defined by the presence of hard exudate within the anatomical area between

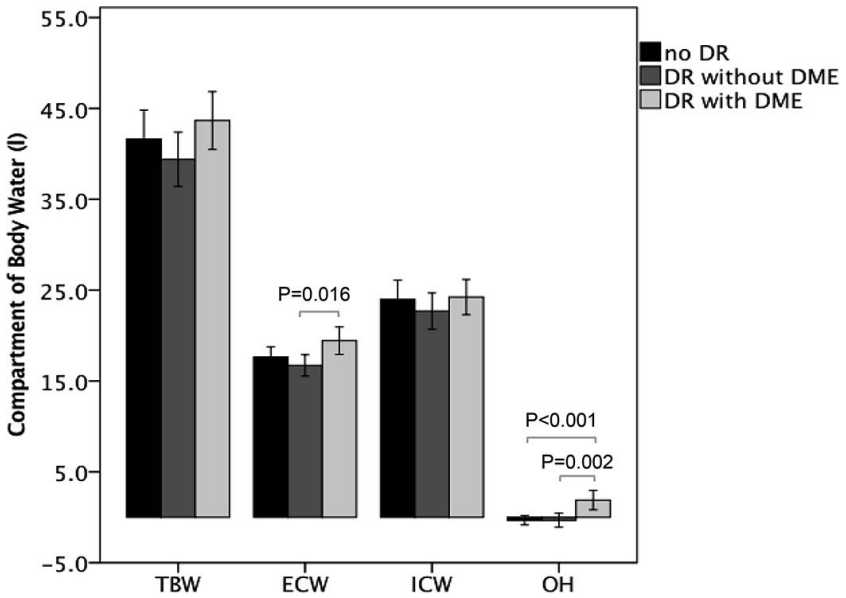

FIGURE 1. Body fluid compartments by DR status. Eyes with DR and DME had higher level of ECW than those with DR but without DME ($P = 0.04$). Levels of OH were significantly higher in those with DME than in those without any DR, or those with DR but without DME ($P < 0.001$ and $P = 0.002$, respectively).

the superior and inferior arcade. OCT (Cirrus HD-OCT 400; Carl Zeiss Meditec, Dublin, CA, USA) was used to record CST and cube volume, which were displayed via a thickness map produced by this instrument with nine grids defined by the previous ETDRS.[21] The CST was calculated in the central 1-mm grid, and the cube volume was calculated as total volumes in all the nine grids. DME was defined by a central subfield thickness of more than 250 μm, with presence of either intraretinal cysts or subretinal fluid, or both, as described in the previous literature.[22] All patients were categorized into one of three groups: no apparent DR; DR without DME; or DR with DME. A group consisting of no DR with DME was not found in this study after excluding all the potential nondiabetic causes of edema as mentioned above.

Quantification of Body Fluid Status

The body fluid status of each patient was measured with a body composition monitor (BCM; Fresenius Medical Care, Bad Homburg, Germany) with input variables including height, weight, age, sex, and BP. Basically, the principle of the BCM is based on bioimpedance analysis, which is composed of resistance caused by total body water (TBW), and reactance caused by cell membrane capacitance. Details of how bioimpedance works have already been mentioned elsewhere.[23] The accuracy of this method in determining body fluids has been shown, compared with other gold standard reference methods in the previous literature, with excellent accordance and high agreement.[24] The impedance was measured by having the electrodes of the monitor attached to the hand and foot on the same side of the body after the patient had been in a supine position for 5 minutes. Data were then calculated with built-in equations that produce the following output: TBW, extracellular water (ECW), intracellular water (ICW), and overhydration (OH). Relative overhydration (ROH) was calculated as OH value divided by ECW, and volume overload was defined as ROH greater than or equal to 7%. All these body fluid determinations as well as the same reference range for ROH have been validated in Taiwanese populations without hemodialysis.[25,26] OCT was then performed soon after BCM evaluation was done at outpatient settings.

Statistical Analysis

The Shapiro-Wilk test was used to test the normality of data distributions among three groups (no DR; DR without DME; or DR with DME). However, only eGFR, systolic BP, and diastolic BP followed normal distributions among all the three groups. Parametric test with 1-way analysis of variance was used if the data followed the normal distribution, otherwise nonparametric method with Kruskal-Wallis test was used. As for each body fluid compartment, further comparisons between groups were done by using Mann-Whitney U test and were considered statistically significant if a P value was less than 0.017 under these conditions. Others were considered statistically significant if a P value was less than 0.05. Additionally, significant factors were appropriately selected to adjust for possible confounding effect for OCT values and body fluid compartments across the three groups. This could be achieved by analysis of covariance model using duration of diabetes mellitus as a covariate and presence of CAD as a fixed factor. Partial correlations with control for possible confounding factors, such as age, sex, and BMI, were used to explore correlations between body fluid compartments and OCT measurements, including CST and cube volume. Categorical variables for group comparisons were evaluated via the χ^2 test or Fisher's exact test. Levels of DR severity could be treated either as a continuous variable or a categorical variable. When treated as a continuous variable, Spearman's rank correlation was performed to study the correlation between ROH and CST. When treated as a categorical variable, however, an appropriate reference group was given by combining no-DR group with mild to moderate non-proliferative diabetic retinopathy (NPDR) group to calculate the odds ratio (OR) for DME.

Binary logistic regression was used to determine correlating factors for DME. Variables with P value less than 0.05 in the univariate model were included into the multivariate model. In the initial univariate analysis, factors with $P < 0.05$ in body fluid status (ECW, OH, ROH, and volume overload) were highly correlated with each other, and volume overload was selected as a factor of interest for multivariate analysis. The DR severity was not selected into the multivariate model owing to high correlation with volume overload. SPSS

Table 2. Correlations Between Body Fluids and Optical Coherence Tomography Measurements According to the Status of Diabetic Retinopathy

Body Fluid Compartment	Central Subfield Thickness				Cube Volume			
	Absence of DR		Presence of DR		Absence of DR		Presence of DR	
	Coefficient	*P* Value	Coefficient	*P* Value	Coefficient	*P* Value	Coefficient	*P* Value
TBW	0.16	0.31	0.25	0.06	0.239	0.13	0.248	0.07
ECW	0.117	0.46	0.364	0.006	0.269	0.08	0.362	0.006
ICW	0.177	0.26	0.099	0.47	0.212	0.18	0.098	0.47
OH	−0.245	0.12	0.309	0.021	−0.024	0.88	0.306	0.022
ROH	−0.245	0.12	0.353	0.008	0.023	0.89	0.32	0.016

(version 24; SPSS, Inc., Chicago, IL, USA) was used for all the statistical analyses.

Results

Baseline Characteristics

Of the 104 patients initially enrolled in this study, 9 patients had ocular pathology not eligible for analysis in one of their eyes. These included epiretinal membrane (three eyes), retinal detachment (one eye), previous intravitreal bevacizumab injections (one eye), corneal scar (one eye), severe vitreous hemorrhage (one eye), age-related macular degeneration (one eye), and foveal scar (one eye). For the rest of the 95 patients, only the eye with greater central subfield thickness was analyzed. The mean age of these patients was 58.0 ± 20.0 years, and 63 (60.6%) were male. The mean HbA1c was 8.3% ± 2.4%, and the mean duration of diabetes mellitus was 10.3 ± 7.4 years. Table 1 demonstrates the baseline and other clinical data, by degree of DR severity.

Body Fluid Status in Relation to Diabetic Retinopathy

Regarding body fluid status, there were significant differences in ECW and OH between patients with no DR, DR without DME, and DR with DME ($P = 0.04$ and $P < 0.001$, respectively) (Table 1). The significance remained even after adjustment with analysis of covariance model (Supplementary Table S1).

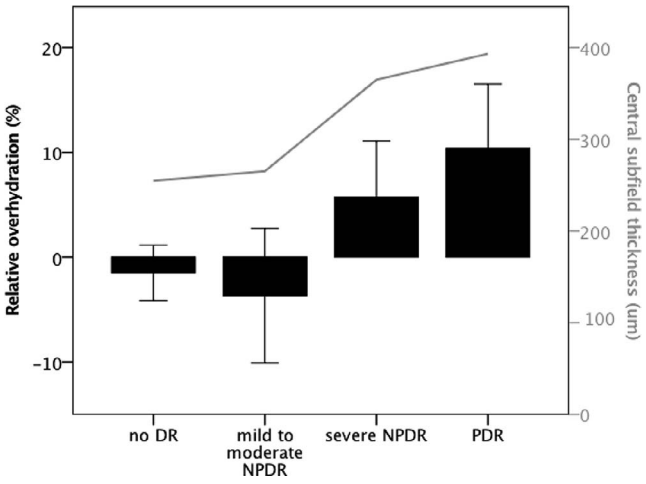

Figure 2. Overall, higher levels of ROH were correlated with thick CST ($r = 0.338$, $P = 0.001$, after adjusting for age, sex, and BMI). When correlated with DR severity, high ROH and thick CST were also associated with severe degree of DR (Spearman's $\rho = 0.345$, $P = 0.001$ and $\rho = 0.551$, $P < 0.001$, respectively).

Further analysis revealed that patients with DME had significantly more ECW than those with DR but without ME (Fig. 1). Also, patients with DME had the highest level of OH, compared with those without DME ($P < 0.001$ in no-DR group and $P = 0.002$ in DR group, respectively). There were no significant differences in TBW and ICW between patients with no DR, DR without DME, and DR with DME.

Correlations Between ROH and CST

Table 2 shows the correlations between each body fluid compartment category and OCT value, including CST and cube volume. When analyzed by the presence ($n = 59$) or absence of DR ($n = 45$), ROH was associated with thick central subfield thickness in patients with DR ($r = 0.353$, $P = 0.008$, after adjusting for age, sex, and BMI) but not in those without DR ($r = -0.245$, $P = 0.12$, after adjusting for age, sex and BMI). When correlated with DR severity, both high ROH and thick CST were associated with severe degree of retinopathy (Spearman's $\rho = 0.345$, $P = 0.001$ and $\rho = 0.551$, $P < 0.001$, respectively) (Fig. 2). When DR severity was treated as a categorical variable with "no DR to moderate NPDR" as a reference group, both severe NPDR and PDR were significant correlating factors for the presence of DME (OR 31.000 with 95% confidence interval of 8.331–115.352, and OR 27.280 with 95% confidence interval of 6.757–110.143, respectively, both $P < 0.001$).

ROH and Varying Features of DME

Subanalysis of patients with DME is shown in Figure 3 and Supplementary Table S2. DME was categorized according to presentation of OCT features and funduscopic macular hard exudate. Patients with both intraretinal cysts and subretinal fluid tended to have higher levels of mean ROH and, similarly, a higher proportion of volume overload, than patients of any other group.

Volume Overload as a Predictor for DME

Patients with volume overload had higher proportions of intraretinal cysts, subretinal fluid, and macular hard exudate ($P < 0.001$, $P < 0.001$, and $P = 0.001$, respectively). As for systemic conditions, lower HbA1c (<7%) and the use of diuretics were significantly present in patients with volume overload ($P = 0.024$ and 0.018, respectively, χ^2 test) (Table 3). In Table 4, both univariate and multivariate regression models showed that volume overload was the independent factor for DME (OR, 7.457; 95% confidence interval, 2.493–22.309; $P < 0.001$). The significance of volume overload was still observed after additional adjustment for age and sex (OR, 9.532; 95% confidence interval, 2.898–31.348; $P < 0.001$). Univariate analysis of comprehensive clinical factors for DME is shown in Supplementary Table S3.

Table 3. Associations of Clinical Features With and Without Volume Overload

Clinical Condition	Presence of Volume Overload, $n = 27$	Absence of Volume Overload, $n = 77$	*P* Value
Ophthalmic features, *n* (%)			
Intraretinal cyst	17 (63.0)	14 (18.2)	<0.001
Subretinal fluid	8 (29.6)	3 (3.9)	<0.001
Macular hard exudate	12 (44.4)	10 (13.0)	0.001
Systemic conditions, *n* (%)			
Obesity	5 (18.5)	28 (36.4)	0.09
Hypertension	3 (11.1)	11 (14.3)	1.0
HbA1c <7%	15 (55.6)	24 (31.2)	0.024
eGFR <60	11 (40.7)	20 (26.0)	0.16
CAD	9 (33.3)	24 (31.2)	0.84
Insulin dependence	14 (51.9)	29 (37.7)	0.2
Use of diuretics	6 (22.2)	4 (5.2)	0.018
Use of LLD	16 (59.3)	49 (63.6)	0.69
Use of OHA	22 (81.5)	67 (87.0)	0.48

Discussion

To the best of our knowledge, this is the first study to objectively measure body fluid status and report its associations with severity of DR. The present study showed that greater central subfield thickness in diabetic patients is significantly correlated with higher levels of systemic ECW and OH, but not with ICW. Furthermore, subanalysis found that the correlation exists particularly in patients with DR, but not in those without DR. Explanations might be proposed from the perspective of varying barrier properties in DR, as well as systemic fluid status that becomes overhydrated in a number of systemic conditions.[25–27] We infer that both volume and barrier functions contribute to the anatomical consequences. Breakdown of the blood-retinal barrier, as seen in DR, may enhance the role of fluid movement between the retinal parenchyma and intravascular space. Clinically significant macular edema develops when there is frank leakage of intravascular fluid and protein into the interstitial tissue in the retina. While the current concept regarding the pathogenesis of DME has focused on impaired barrier properties caused by ischemic cascades with an increase in the production of VEGFs and various inflammatory cytokines,[5] our study supports the idea that in addition to barrier dysfunction, extravasation in retinal edema may be driven by the force of increasing capillary hydrostatic pressure due to volume expansion. It is worth mentioning that unlike normal vasculature, these diabetic retinal capillaries may lose their autoregulation in response to such volume fluctuations.[12,28]

Patients with DME have significantly higher levels of OH than those without DME or without any DR. This finding sheds light on some clinical situations that have previously remained largely unexplained. For instance, some patients with proliferative DR may rarely develop macular edema, which implies that production of VEGF per se may not necessarily lead to the accumulation of fluid. Not uncommonly, we may also observe cases of macular edema refractory to repeated intravitreal injections of anti-VEGFs or corticosteroids.[29] Beyond VEGFs and inflammatory cytokines, ROH could be a novel risk factor for DR, particularly in the presence of macular edema. The distinct association of volume expansion with DME may also be illustrated by a few cases showing resolution of edema after systemic furosemide, which has been used to treat fluid overload.[30,31] This also helps explain why we observed a higher proportion of diuretic use in patients with DME in the present study, in which the association may be confounded by volume overload in diuretic users.

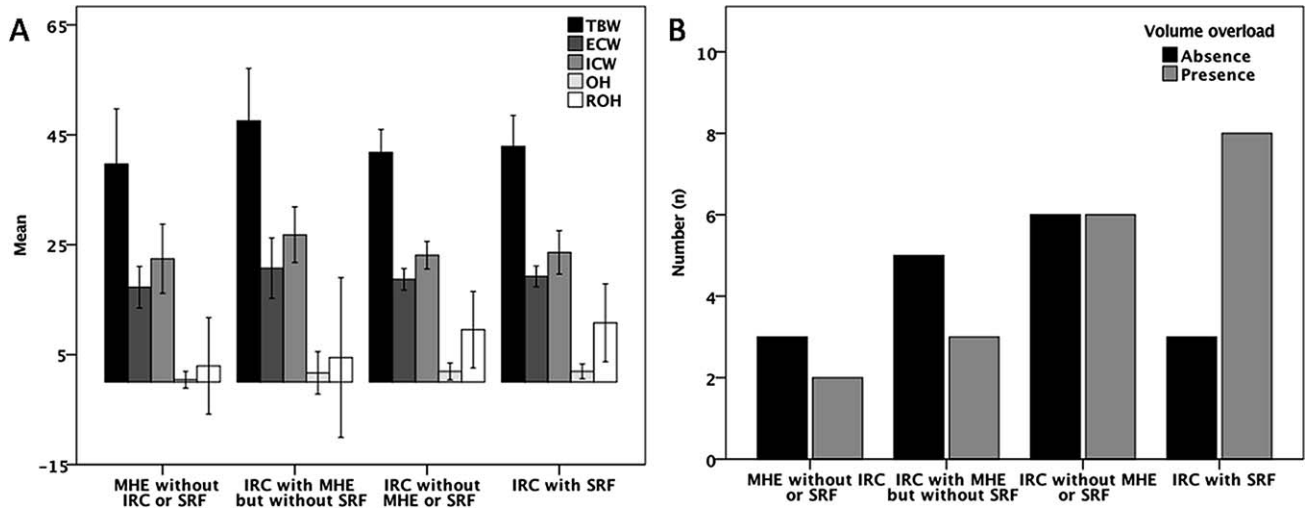

Figure 3. Mean levels of OH, ROH (**A**), and percentage of volume overload (**B**) were observed to be highest in eyes with presence of both intraretinal cysts (IRCs) and subretinal fluid (SRF), and lowest in those with macular hard exudate (MHE) but without IRCs or SRFs. See Supplementary Table S2 for precise numbers.

Table 4. Correlating Factors for Diabetic Macular Edema by Binary Logistic Regression Analysis

	Univariate Model		Multivariate Model			
Clinical Parameter	OR (95%CI)	*P* Value	OR (95%CI)	*P* Value	OR* (95%CI)	*P* Value*
DM >10 y	2.827 (1.182–6.762)	0.02	1.53 (0.509–4.604)	0.45	1.612 (0.513–5.069)	0.41
CAD	2.865 (1.185–6.926)	0.019	2.642 (0.856–8.151)	0.09	2.592 (0.799–8.409)	0.11
eGFR <60	3.340 (1.363–8.186)	0.008	1.896 (0.59–6.096)	0.28	1.631 (0.489–5.442)	0.42
Use of diuretics	4.140 (1.078–15.895)	0.038	1.401 (0.268–7.326)	0.69	1.404 (0.245–8.056)	0.7
Volume overload	7.650 (2.893–20.228)	<0.001	7.457 (2.493–22.309)	<0.001	9.532 (2.898–31.348)	<0.001

* Additional adjustment for age and sex.

We did not observe a significant correlation between high systolic BP and volume overload, which could be supported by the previous study.[32] Hypertension with volume overload usually occurs in patients with chronic kidney disease,[26] which was present in only 29.8% of cases in the present study. An interesting observation is that volume overload was associated with lower HbA1c values. Starling's rule may help to explain how fluid expansion may occur in the context of lowering glucose levels.[4,11] Aggressive glycemic control reduces the intravascular osmotic pressure that keeps fluid from draining out. With the loss of vascular autoregulation, increased vascular hydrostatic pressure eventually leads to fluid retention and volume overload. Also, as stated previously, increased capillary hydrostatic pressure creates an excessive flow of water into the interstitial tissue and subsequently causes macular edema. In line with the present result, one recent study[12] has observed that tight glycemic control may slow the resolution of subretinal fluid in DME after ranibizumab treatment at 3 months. Similarly, another report[33] has shown that glycosylated hemoglobin is inversely correlated with central macular thickness in the presence of macular edema. These findings have highlighted the complexity of DME, which is multifactorial and systemically related. We believe that volume expansion probably contributes in part to the occurrence of macular edema.

Consideration of morphologic features in DME that could possibly change with systemic body fluid can be meaningful. In DME, presence of intraretinal cysts with coexisting subretinal fluid was associated with the highest mean level of ROH. It is also interesting to observe that the absence of macular hard exudate was associated with higher levels of ROH than was the presence of hard exudate. However, we did not observe this trend for other body fluid compartments, including TBW, ECW, and ICW. Although there were a relatively small number of patients, potentially limiting significant analysis, there may be some reasons that can explain this. First, both the intraretinal and subretinal space are extravascular spaces, which have the ability to maintain balance between fluid entry and egress via the mechanisms of Starling's force and the pumping function of the retinal pigmented epithelium to keep the retina dry.[4,34] It is plausible that accumulation of fluid in both spaces could be a consequence of severe OH, where there is too much fluid to be removed from the deep capillary plexus or to be pumped out of the subretinal space. Second, it is known that precipitation of lipid exudate usually develops after resolution of chronic vascular leakage.[35] We infer that resorption of retinal fluid might somewhat be affected by the reduction of systemic OH. This hypothesis also supports our data that macular hard exudate without intraretinal cysts or subretinal fluid was associated with the lowest level of ROH. The relationship whereby CST did not significantly correlate with levels of ROH in patients with edema can also be attributed to the varying duration and chronicity of macular edema that existed at the time of observation.

The strength of this study was its prospective design and the novelty of building a connection between systemic body fluid status and DR. Our data also validated some other basic theories beyond VEGFs and inflammation that have long been implicated in the pathogenesis of DME. The limitation of this study was that we failed to clarify cause-and-effect relationships, owing to the nature of cross-sectional analysis. Whether volume expansion is a true causative factor for macular edema should be proposed with caution. In addition, there may be some other confounding factors that were not adjusted for in the regression model. Another limitation was that while we tried to minimize body fluid fluctuations as mentioned previously, we did not strictly control the oral intake of the participants before BCM evaluation. Recent intake of fluids directly influences body weight; however, the impact on distal bioimpedance is minimal.[36] Fluid change in the trunk, which has a large cross-sectional area, may contribute only modestly to the whole body impedance measured from distal areas.[37] Thus, little change in body fluid measurements could be expected. Despite these limitations, this observational study was clinically significant and may be an area of interest for potential therapeutic research in the future. A larger, randomized controlled trial with longitudinal follow-up is necessary to investigate the causal relationship between systemic OH and DR with or without DME.

In conclusion, OCT-measured retinal thickness in patients with DR is correlated with systemic OH. Volume overload is an independent factor for DME. Different levels of ROH may have a role in determining morphologic features that appear in DME.

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