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The relationship between levels of apolipoprotein A1 and B in aqueous and serum with stage of diabetic retinopathy

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ABSTRACT

Aim: To determine the association between serum and aqueous apolipoprotein (Apo) A1 and Apo B levels and Apo B/A1 ratio in diabetic retinopathy.

Material and Method: This cross-sectional prospective study included 63 diabetic patients with or without retinopathy and 38 control subjects who underwent cataract surgery. The study groups were as follows; healthy subjects (Group 1), diabetic patients without retinopathy (Group 2), with non-proliferative diabetic retinopathy (Group 3), and with proliferative diabetic retinopathy (Group 4). Serum and aqueous Apo A1 and Apo B levels were determined by using an enzyme-linked immunosorbent assay.

Results: The amount of Apo B was determined in aqueous samples of all (100%) patients in Group 4 and 77.7% of patients in Group 3.The mean serum Apo B/A1 ratio was significantly higher in Group 2, Group 3 and Group 4 compared with Group 1 ($p=0.002$, $p=0.037$ and $p<0.001$, respectively). The aqueous Apo B level, aqueous and serum Apo B/A1 levels were significantly correlated with the severity of retinopathy in diabetic patients (all $p<0.001$).

Conclusion: Higher serum and aqueous Apo B/A1 ratio were significantly associated with the stage of diabetic retinopathy.

Keywords: Aqueous humor, apolipoprotein A1, apolipoprotein B, diabetic retinopathy

INTRODUCTION

Diabetic retinopathy (DR) is one of the major cause of morbidity in diabetic patients. The prevalence of DR increases with the duration of diabetes (1). Diabetes duration, glycemic control, microalbuminuria and hypertension are well-known modifiable major risk factors for the progression of DR (2,3). The role of dyslipidemia as a potential risk factor for DR is of interest. There was no consensus on the relation with DR and dyslipidemia that some studies have interpreted the role of dyslipidemia with traditional lipid markers like total cholesterol, high density lipoprotein (HDL), low density lipoprotein (LDL) and triglycerides on the development and progression of DR (4,5) while several studies have reported no association (6).

Though the association of traditional lipid parameters with the initation or progression of DR is contraversial, there is evidence that the severity of DR is associated with the precursors of soluble lipoproteins apolipoprotein A1 (Apo A1) and apolipoprotein B (Apo B) levels in diabetic patients (4,7-9). Apo A1 is a HDL constituent and Apo B is found in very low density lipoprotein (VLDL), intermediate-density lipoprotein and LDL. Unlike traditional lipid parameters, these apolipoproteins are not affected by the prandial status (10). Apo A1 is a good indicator of lipid accumulation in peripheral tissues (8) and also has anti-inflammatory and antioxidant properties (11,12), it specifically inhibits oxidation of LDL which may damage vascular endothelial, anti-platelet and anti-inflammatory functions. Apo B is present as an extravasated form and was detected in the retinal layers of human eyes with DR (13) which may play a potential role in the pathophysiology of DR. Also, serum Apo B/A1 ratio was also shown to be related with the severity of DR in several studies (14-18).

To the best of our knowledge, there have been no studies published about Apo B levels and ApoB/A1 ratio in aqueous samples of diabetic patients. In this study, we aimed to determine the Apo A1 and B levels

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and the Apo B/A1 ratio in aqueous humor and serum of diabetic patients with or without retinopathy and compared with healthy controls.

MATERIAL AND METHOD

The study was carried out with the permission of Kırıkkale University Medical Faculty Clinical Researches Ethics Committee (Date: 2012, Decision No: 12/58). All research procedures were performed in accordance with the Declaration of Helsinki.

This cross-sectional prospective study included 63 patients with type 2 diabetes mellitus (DM) and 38 healthy age and gender matched control subjects who admitted for decreased vision due to cataract. Informed consents which included serum and aqueous sampling were obtained from all subjects before study participation.

Patients with macular edema, vitreous hemorrhage and retinal detachment, previous laser photocoagulation or intravitreal therapy within six months, previous vitreoretinal or glaucoma surgery history, dense cataract obscuring visibility of retina, systemic diseases other than DM and vitreoretinal disorders other than DR were excluded from the study. Patients scheduled for cataract surgery with no systemic or ocular diseases were recruited in the study as control group.

The classification of DR was made using the Airlie House Classification system[19]with seven-field retinal photograph by two experienced ophthalmologists. Patients groups were divided as follows; those without retinopathy as Group 2, with non-proliferative DR (non-PDR) as Group 3 and with proliferative DR as Group 4. Healthy control subjects were assigned as Group 1.

Each study participant underwent a detailed ophthalmological examination including best-corrected visual acuity using Snellen chart, Goldman applanation tonometry, slit-lamp anterior segment evaluation and dilated fundus examination. Fundus fluorescein angiography (FFA; Topcon TRC; Topcon Co, Tokyo, Japan) and spectral domain optical coherence tomography (SD-OCT; RetinaScan Advanced RS-3000; NIDEK, Gamagori, Japan) imaging were also performed when indicated.

Venous blood samples for Apo A1 and B levels were obtained preoperatively and stored at - 80°C after centrifuged. All cataract surgeries were performed under topical anesthesia using 2% lidocaine. About 0,1-0,2 ml of aqueous humor was aspirated with a sterile injector capped with an anterior irrigation tip. The aqueous samples of all patients were collected and stored at -80°C until testing in the same batch. Inefficient amount of samples were not recruited in the study.

Serum and aqueous Apo A1 and B levels were determined by using an enzyme-linked immunosorbent assay (ELISA) kit (USCNlife, USCN life Science Inc., PRC) according to the manufacturer's instructions. Sensitivity of Apo A1 and Apo B kits were 55 ng/mL and 4.27 ng/mL. Serum concentration of triglyceride, cholesterol, LDL and HDL were evaluated by an automated analyzer using commercially available kits and serum HbA1c levels of diabetic patients were measured with high-performance liquid chromatography.

Statistical Analysis

Statistical analyses were performed using the SPSS program software (Version 21, International Business Machines Co,Armonk, NY). Demographic characteristics and aqueous and serum apo A1 and B concentrations of the patients among the 4 groups were compared. One-way analysis of variance, the Wilcoxon rank-sum test, and the Kruskall-Wallis test were used to compare numerical data, and the Fisher exact test was used to compare categorical variables. Correlations were analysed by using Spearman's correlation coefficient. All data were expressed as mean±Standard deviation (±SD). A p value less than 0.05 was considered as statistically significant.

RESULTS

In Group 1, the mean age was 68.11±9.5 years and 55.3% of 38 patients were male. The mean age and the percentage of male patients were, 68.76±8.65 years and 52.9% in Group 2, 68.74±8.42 years and 51.9% in Group 3, and 65.42±8.33 years and 52.6% in Group 4, respectively. There were no significant differences among the groups in terms of age and gender (p=0.994 and p=0.594) (**Table 1**).

The mean HbA1c level was 8.9±2.29% in Group 2, 8.53±1.76% in Group 3 and 8.87±2.24% in Group 4, respectively and no statistically significant difference was observed between the groups (p=0.876). The mean duration of DM was 11.4±4.7 years in Group 2, 12.2±7.9 years in Group 3 and 18.9±6 years in Group 4. There were statistically significant differences between groups for DM duration (p<0.001) and DM duration was longer in Group 4 than Group2 and 3 (p< 0.001 and p=0.001) (**Table 1**).

The mean serum triglyceride level was 140.12±20.65 mg/dL in Group 2, 124.56±26.99 mg/dL in Group 3, 131±35.48 mg/dL in Group 4 and 114.16±31.68 mg/dL in Group 1. Serum triglyceride level was lower in Group 1 than Group 2 and Group 4 (p=0.002 and p=0.020). The mean serum total cholesterol, LDL and HDL levels were 195.24±4.67 mg/dL, 128.12±15.84 mg/

dL and 48.82±11.29 mg/dL in Group 2; 185.11±19.65 mg/dL, 120.26±32.42 mg/dL and 51.04±12.09 mg/dL in Group 3; 192.16±18.06 mg/dL, 113.58±26.62 mg/dL and 44.37±9.96 mg/dL in Group 4; 178.74±25.6 mg/dL, 112.03±27.78 mg/dL and 48.05±6.88 mg/dL in the control group. There were no statistically significant differences in total cholesterol, LDL and HDL levels among all groups (p=0.194, p=0.182 and p=0.124).

Apo B was determined in aqueous samples of 40 (63.5%) diabetic patients that were 21 of 27 patients in group 3 (33.3%) and all patients in group 4 (30.2%). The mean aqueous Apo B level was significantly higher in Group 4 than in Group 3 (p<0.001). Nevertheless Apo A1 level was detected in aqueous samples of all diabetic patients and healthy subjects.

The mean serum Apo A1 levels were significantly decreased in Group 4 compared with Group 1 and Group 3 (p=0.002 and p=0.001). The mean serum Apo B levels were significantly increased in Group 2, 3 and 4 compared with Group 1 (p values; 0.001, 0.002 and 0.008, respectively). Aqueous Apo A1 level was significantly higher in Group 3 and Group 4 in comparison to patients in Group 2 (p=0.001 and p=0.005). Aqueous Apo A1 level was also decreased in Group 1 when compared with Group 2, Group 3 and Group 4 (p=0.004, p<0.001 and p<0.001).

The mean serum Apo B/A1 ratio was higher in Group 2, Group 3 and Group 4 than control group (p=0.002, p=0.037 and p<0.001). The mean serum and aqueous Apo B/A1 ratios were higher in Group 4 compared with Group 3 (p=0.013 and p<0.001). (**Table 2**)

Serum LDL and cholesterol levels were significantly correlated with serum Apo B levels (p<0.001, Spearman correlation: 0.503, CI:0.328-0.662 and p<0.000, Spearman correlation: 0.602, CI:0.422-0.745). Aqueous Apo B levels were significantly correlated with the duration of DM and the severity of retinopathy (p<0.001, Spearman correlation: 0.478, CI:0.263-0.669 and p<0.001, Spearman correlation: 0.861, CI:0.816-0.908).

Higher aqueous Apo B levels and Apo B/A1 ratios had significant strong correlation with the severity of diabetic retinopathy (p<0.001, Spearman correlation: 0.809, CI:0.758-0.856 and p<0.001, Spearman correlation: 0.816, CI:0.754-0.867). Lower serum Apo A1 levels, higher serum Apo B levels and Apo B/A1 ratios had significant moderate correlation with the severity of diabetic retinopathy (p<0.001, Spearman correlation: -0.534, CI:-0.424-0.199, p=0.001, Spearman correlation: 0.417, CI:0.147-0.382 and p<0.001, Spearman correlation: 0.457, CI:0.307-0.599). Serum and aqueous humor Apo A1 and B levels and B/A1 ratios were not significantly correlated with HbA1c levels (p>0.05, in each comparison).

DISCUSSION

This study demonstrated that serum Apo B levels and Apo B/A1 ratio were increased in all diabetic patients with and without retinopathy than in control group. Serum Apo A1 levels were decreased in patients with PDR in comparison to patients with non-PDR and control subjects. Serum Apo B/A1 ratio was also higher in patients with PDR than in patients with non-PDR.

Table 1. Demographic features and serum lipid parameters in diabetic patients and control subjects

Parameters	Group 1 (control group)	Group 2 (without DR)	Group 3 (non-PDR)	Group 4 (PDR)	P value
Number, n	38	17	27	19	
Gender (F/M)	17/21	8/9	13/14	9/10	0.994
Age (years)	68.11±9.5	68.76±8.65	68.74±8.42	65.42±8.33	0.594
Total cholesterol (mg/dL)	178.74±25.6	195.24±4.67	185.11±19.65	192.16±18.06	0.194
Triglyceride (mg/dL)	114.16±31.68	140.12±20.65	124.56±26.99	131±35.48	0.005*
LDL (mg/dL)	112.03±27.78	128.12±15.84	120.26±32.42	113.58±26.62	0.182
HDL (mg/dL)	48.05±6.88	48.82±11.29	51.04±12.09	44.37±9.96	0.124
DM duration (years)		11.4±4.7	12.2±7.9	18.9±6	<0.001
HbA1c (%)		7.9±1.29	7.53±1.76	7.87±1.24	0.876

DM: Diabetes Mellitus, DR: diabetic retinopathy, F: female, HbA1c: hemoglobin A1c, HDL: high-density lipoprotein, LDL: low-density lipoprotein, M: male, PDR: proliferative diabetic retinopathy, *p< 0.05

Table 2. Comparison of serum and aqueous Apo A1 and B levels in diabetic patients and control subjects

Parameters	Group 1 (control group)	Group 2 (without DR)	Group 3 (non-PDR)	Group 4 (PDR)	P value
Serum Apo A1(mg/dL)	125.11±19.81	126.76±23.42	130.78±20.85	109.74±14.78	0.004*
Serum Apo B (mg/dL)	92.89±19.71	113.53±17.6	110.78±22.49	110±20.81	0.001*
Serum Apo B/A1	0.74±0.13	0.91±0.18	0.85±0.18	1.01±0.19	<0.001
Aqueous Apo A1 (ng/mL)	210.82±170.34	324.12±175.25	559.44±289.19	519.95±243.53	<0.001
Aqueous Apo B (ng/mL)	-	-	99.19±42.91	280.53±83.85	<0.001
Aqueous Apo B/A1	-	-	0.20±0,10	0.57±0.14	<0.001

Apo A1: apolipoprotein A1, apo B: apoliprotein B, DR: diabetic retinopathy, PDR: proliferative diabetic retinopathy, *p< 0.05

There were no studies investigating Apo B in human aqueous samples whereas prior studies demonstrated Apo A1 in aqueous samples of diabetic patients and healthy subjects. Apo A1 and B were detected as an extravasated form in vitreous samples and also in the retinal layers of human eyes with DR (8,9,13).

When considering the molecules involved in the pathogenesis of DR simultaneously appear in aqueous and vitreus samples of the same eyes (20), Apo B, detected in the aqueous samples of non-PDR and PDR patients, may serve as a biomarker to show the severity of DR. Aqueous humor does not make a direct contact with the retina whereas functional barriers between vitreus humor and aqueous humor are also not strict. Thus Apo B detected in aqueous samples should be released from retina and may leak into cilio-retinal circulation through the disrupted blood-retinal barrier or enter aqueous humor through blood aqueous barrier by diffusion.

Apo B was not identified in all aqueous samples of diabetic patients while Apo A1 was detected in all aqueous samples of both diabetic patients and control group. Apo B was detected in 63.4% of diabetic patients including 77.7% of non-PDR patients and all PDR patients whereas detected in none of the diabetic patients without retinopathy and healthy controls. Previous studies have found that large molecules can be exchanged between the vitreus and aqueous humor through vitreus-aqueous humor barrier (21-23). Apo A1 and B have different molecular weights; Apo A1 has a lower molecular weight of 28.1 kD, the molecular weight of Apo B is 540 kD. The higher molecular weight Apo B particules leaking into the aqueous were detected in severe forms of diabetic retinopathy but in none of the patients without retinopathy. Wu et al. (13) measured Apo B levels in postmortem retinas of diabetics and non-diabetics; found that the amount of extravasated Apo B was correlated with the severity of DR.These findings were consistent with our study and suggest that the severity and duration of diabetes may be significantly and positively correlated with existence and detection of Apo B in aqueous samples.

Intraocular Apo A1 was investigated by different methods in ocular tissues. Kawai et al. (24) detected Apo A1, the major component of HDL particles by Western blot in the tear films of healthy individuals and diabetic patients and the presence of Apo A1 was associated with leakage from the capillary vessels of the main lacrimal glands as a result of vascular damage in diabetic angiopathy and the increased amount of Apo A1 was related to the severity of DR. Simo et al. (8) determined overexpresion of Apo A1 in the retinas of diabetic patients without microvascular abnormalities in the two years preceeding the death, not only in its mRNA levels, but also in terms of protein content in their postmortem study and overexpression of Apo A1 was considered as an early finding in the retina of diabetic patients before the initiation of DR. Over-production and higher mRNA expression of Apo A1 in the retinas of PDR patients than in non-diabetics were also reported by using fluid proteomic analysis of human vitreus fluid (9,25). Apo A1 levels in aqueous were not significanly different in diabetic patients with or without retinopathy and healthy controls in our study. It may be related with sample size in the study, homogeneity of DR groups, and DM regulation that HbA1c levels were similar between the groups.

The Apo B/A1 ratio is of interest that the ratio reflects vascular endothelial damage in vascular disorders. Hu et al. (14) and Ankit et al. (17) exhibited a significant correlation between severity of DR and lower Apo A1 levels and decreased Apo A1/B ratio in serum samples. Sasongko et al. (15) determined a negative correlation between the development and the severity of DR and serum Apo A1 levels and a positive correlation with serum Apo B levels and Apo B/A1 ratio. Crosby-Nwaobi et al. (26) reported that an increase in serum Apo B and higher Apo B/A1 ratio may be associated with increased risk of PDR and clinically significant diabetic macular edema (CSME). Chung et al. (18) reported the association of serum Apo A1 and the ApoB/A1 ratio with presence of diabetic retinopathy. These findings were in agreement with our study and suggest that lower serum Apo A1, higher serum Apo B level and Apo B/A1 ratio were related with the severity of DR.

Though the effects of lipids on the initation or progression of DR were controversial in literature, our study suggested that higher serum Apo B levels and higher Apo B/A1 ratio had a significant moderate correlation and higher aqueous Apo B level and Apo B/A1 ratio had significant strong correlation with the severity of DR. Miljanovic et al. (27) observed an association between the serum lipid levels and increased risk of CSME and hard exudates, but not with progression of DR and PDR formation. In the Fenofibrate Intervention and Event Lowering in Diabetes Study (FIELD) no associations were declared between the serum lipid levels and the formation and the progression of DR (28). Several studies emphasized that the mechanisms of intraretinal lipid transport may be more prominent than the serum lipid levels in pathogenesis of DR (29,30). Aqueous humor exchanges substances, proteins and ions through capillaries by direct and indirect contact with ocular tissues. The contents of aqueous humor including proteins were used to determine the health status of blood vessels and/or ocular tissues. Aqueous humor and vitreus were evaluated for determining the formation or progression of DR (31). All these findings indicate that the mechanisms regulating the aqueous

lipid transport, rather than serum lipid levels, may have a more significant role in the pathogenesis of DR and Apo A1 and Apo B should be an ocular fluid biomarker for predicting progression of DR.

CONCLUSION

To conclude, in this study, higher aqueous Apo B/A1 ratios and higher aqueous Apo B levels in diabetic patients may help to detect and monitor the severity of DR and can be used as a potential biomarker. As best we know, this is the first study to investigate Apo B in aqueous samples of diabetic patients with any stages of DR. Further population-based longitudinal studies are needed to evaluate the role of various apolipoproteins in the pathogenesis of DR and the origin of them in intraocular fluids.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of Kırıkkale University Medical Faculty Clinical Researches Ethics Committee (Date: 2012, Decision No: 12/58).

Informed Consent: All patients signed the free and informed consent form.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

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Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

REFERENCES

1. Cheung N, Mitchell P, Wong TY. Diabetic retinopathy. Lancet 2010 Jul 10; 376: 124-36.
2. Vujosevic S, Aldington SJ, Silva P, et al. Screening for diabetic retinopathy: new perspectives and challenges. Lancet Diabetes Endocrinol 2020; 8: 337-47.
3. Ghamdi AHA. Clinical predictors of diabetic retinopathy progression; a systematic review. Curr Diabetes Rev 2020; 16: 242-7.
4. Lyons TJ, Jenkins AJ, Zheng D, et al. Diabetic retinopathy and serum lipoprotein subclasses in the DCCT/EDIC cohort. Invest Ophthalmol Vis Sci 2004; 45: 910-8.
5. Klein R, Sharrett AR, Klein BE, et al. The association of atherosclerosis, vascular risk factors, and retinopathy in adults with diabetes: the Atherosclerosis Risk in Communities Study. Ophthalmology 2002; 109: 1225–34.
6. Zhou Y, Wang C, Shi K, YinX. Relationship between dyslipidemia and diabetic retinopathy: A systematic review and meta-analysis. Medicine (Baltimore) 2018; 97: e12283.
7. Zhang Q, Hu J, Hu Y, et al. Relationship between serum apolipoproteins levels and retinopathy risk in subjects with type 2 diabetes mellitus. Acta Diabetol 2018; 55: 681-9.
8. Simo R, Garcia-Ramirez M, Higuera M, Hernandez C. Apolipoprotein A1 is overexpressed in the retina of diabetic patients. Am J Ophthalmol 2009; 147: 319-25 e1.
9. Simo R, Higuera M, Garcia-Ramirez M, Canals F, Garcia-Arumi J, Hernandez C. Elevation of apolipoprotein A-I and apolipoprotein H levels in the vitreous fluid and overexpression in the retina of diabetic patients. Arch Ophthalmol 2008; 126: 1076-81.
10. Davidson MH. Apolipoprotein measurements: is more widespread use clinically indicated? Clin Cardiol 2009; 32: 482-6.
11. Shah PK, Kaul S, Nilsson J, Cercek B. Exploiting the vascular protective effects of high-density lipoprotein and its apolipoproteins: an idea whose time for testing is coming, part II. Circulation 2001; 104: 2498-502.
12. Marcovina S, Packard CJ. Measurement and meaning of apolipoprotein AI and apolipoprotein B plasma levels. J Intern Med 2006; 259: 437-46.
13. Wu M, Chen Y, Wilson K, et al. Intraretinal leakage and oxidation of LDL in diabetic retinopathy. Invest OphthalmolVis Sci 2008; 49: 2679-85.
14. Hu A, Luo Y, Li T, et al. Low serum apolipoprotein A1/B ratio is associated with proliferative diabetic retinopathy in type 2 diabetes. Graefes Arch Clin Exp Ophthalmol 2012; 250: 957-62.
15. Sasongko MB, Wong TY, Nguyen TT, et al. Serum apolipoprotein AI and B are stronger biomarkers of diabetic retinopathy than traditional lipids. Diabetes Care 2011; 34: 474-9.
16. Sasongko MB, Wong TY, Nguyen TT, et al. Serum apolipoproteins are associated with systemic and retinal microvascular function in people with diabetes. Diabetes 2012; 61: 1785-92.
17. Ankit BS, Mathur G, Agrawal RP, Mathur KC. Stronger relationship of serum apolipoprotein A-1 and B with diabetic retinopathy than traditional lipids. Indian J Endocrinol Metab 2017; 21: 102-5.
18. Chung JO, Park SY, Cho DH, Chung DJ, Chung MY. Associations between serum apolipoproteins, urinary albumin excretion rate, estimated glomerular filtration rate, and diabetic retinopathy in individuals with type 2 diabetes. Medicine (Baltimore) 2019; 98: e15703.
19. Early Treatment Diabetic Retinopathy Study Research Group. Grading diabetic retinopathy from stereoscopic color fundus photographs—an extension of the modified Airlie House classification: ETDRS report number 10: Early Treatment Diabetic Retinopathy Study Research Group. Ophthalmology 1991; 98: 786-806.
20. Funatsu H, Yamashita H, Noma H, et al. Aqueous humor levels of cytokines are related to vitreous levels and progression of diabetic retinopathy in diabetic patients. Graef Arch Clin Exp Ophthalmol 2005; 243: 3-8.
21. Barsotti MF, Bartels SP, Freddo TF, Kamm RD. The source of protein in the aqueous humor of the normal monkey eye. Invest Ophthalmol Vis Sci 1992; 33: 581-95.
22. Maurice DM. Flow of water between aqueous and vitreous compartments in the rabbit eye. Am J Physiol 1987; 252: F104–8.
23. Fatt I. Flow and diffusion in the vitreous body of the eye. Bull Math Biol 1975; 37: 85-90.
24. Kawai S, Nakajima T, Hokari S, Komoda T, Kawai K. Apolipoprotein A-I concentration in tears in diabetic retinopathy. Annals Clin Bioch 2002; 39: 56-61.
25. Garcia-Ramirez M, Canals F, Hernandez C, et al. Proteomic analysis of human vitreous fluid by fluorescence-based difference gel electrophoresis (DIGE): a new strategy for identifying potential candidates in the pathogenesis of proliferative diabetic retinopathy. Diabetologia 2007; 50: 1294-303.
26. Crosby-Nwaobi R, Chatziralli I, Sergentanis T, Dew T, Forbes A, Sivaprasad S. Cross Talk between Lipid Metabolism and Inflammatory Markers in Patients with Diabetic Retinopathy. J Diabetes Res 2015; 2015: 191382.

27. Miljanovic B, Glynn RJ, Nathan DM, Manson JE, Schaumberg DA. A prospective study of serum lipids and risk of diabetic macular edema in type 1 diabetes. Diabetes 2004; 53: 2883-92.
28. Keech AC, Mitchell P, Summanen PA, et al. Effect of fenofibrate on the need for laser treatment for diabetic retinopathy (FIELD study): a randomised controlled trial. Lancet 2007; 370: 1687-97.
29. Tserentsoodol N, Sztein J, Campos M, et al. Uptake of cholesterol by the retina occurs primarily via a low density lipoprotein receptor-mediated process. Molecular Vision 2006; 12: 1306-18.
30. Tserentsoodol N, Gordiyenko NV, Pascual I, Lee JW, Fliesler SJ, Rodriguez IR. Intraretinal lipid transport is dependent on high density lipoprotein-like particles and class B scavenger receptors. Molecular vision 2006; 12: 1319-33.
31. Chiang SY, Tsai ML, Wang CY, et al. Proteomic analysis and identification of aqueous humor proteins with a pathophysiological role in diabetic retinopathy. J Proteomics 2012; 75: 2950-9.