SERUM APOLIPOPROTEIN AI AND B ARE STRONGER BIOMARKERS OF DIABETIC RETINOPATHY THAN TRADITIONAL LIPIDS

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ABSTRACT

Introduction: Diabetic Retinopathy (DR) is a prevalent microvascular consequence of diabetes and is the primary cause of blindness and visual impairment on a global scale. Dyslipidemia is the primary metabolic disturbance in diabetes mellitus. Several research have indicated a favourable correlation between conventional serum lipids and the risk of DR, however other investigations have not consistently demonstrated the same relationships. **Materials and Methods:** This study was a case-control study conducted in the ophthalmology department, involving 100 patients with type 2 diabetes (T2D). The levels of fasting blood sugar and post-prandial blood sugar were measured using the glucose oxidase-peroxidase (GOD-POD) method, as well as HbA1C. The autoanalyzer was used to evaluate the serum lipid profile.The levels of SerumApo-A1 and Apo-B were quantified using a fully automated nephelometry method. The Apo-B/A1 ratio was computed. The grading of retinopathy was conducted based on the criteria established by the Early Treatment of Diabetic Retinopathy Study (ETDRS). **Results:** The levels of Serum Apo-A1 showed a substantial decrease as the severity of DR increased ($p<0.001$). Significant elevations in Apo B ($p<0.001$) and the Apo B-to-Apo-A1 ratio ($p<0.001$) were observed, and these findings were substantially correlated with the severity of diabetic retinopathy (DR). The severity of DR was found to have a negative correlation with HDL cholesterol levels ($p<0.05$). The triglyceride level showed a strong correlation with both the higher occurrence and greater severity of diabetic retinopathy in patients with type 2 diabetes, while LDL or total cholesterol did not exhibit such an association. **Conclusion:** The relationship between serum apolipoproteins (Apo-A1, Apo B, and the ApoB/Apo-A1 ratio) and the advancement and severity of diabetic retinopathy (DR) in individuals with type 2 diabetes (T2D) is more significant than that of traditional lipids.Despite the limited usage of apolipoprotein measures in

clinical practise, further research is necessary to understand its protective mechanism. This will help reduce the prevalence of eye conditions and provide more comprehensive eye care.

Keywords: Dyslipidemia,Lipidprofile,Retinopathy, Type2diabetes

INTRODUCTION

Diabetic retinopathy is a known and serious long-term consequence that affects the small blood vessels in the eyes of all individuals with diabetes mellitus, leading to vision loss.[1] Diabetes continues to be the primary factor contributing to death and impairment among employed individuals. Nevertheless, this state of being susceptible to disease or death may be mostly avoided and cured. By promptly addressing the issue, the overall well-being can be maintained.[2 - 4.]

The Diabetic Retinopathy (DR) exerts a substantial influence on the global healthcare system, leading to the annual occurrence of blindness in more than 10,000 individuals suffering from diabetes. In the absence of immediate intervention, the population of individuals with diabetic retinopathy (DR) is projected to rise from 126.6 million in 2010 to 191.0 million by 2030. Additionally, it is anticipated that the number of individuals with vision-threatening DR (VTDR) will escalate from 37.3 million to 56.3 million.[5]

The primary determinant linked to the occurrence of DR is the length of time the disease persists and the presence of long-term high blood sugar levels. Nevertheless, several patients do not exhibit diabetic retinopathy (DR) despite having inadequate glycemic control and a prolonged history of diabetes, while others who maintain strict glycemic control and have a very short duration of diabetes still experience the progression of DR[6]. Dyslipidemia is a prominent metabolic alteration commonly seen in individuals with diabetes mellitus. Studies investigating the correlation between DR and conventional lipids have yielded inconsistent findings.[7] Increased lipid levels result in endothelial dysfunction through a localised inflammatory reaction involving the release of cytokines and growth factors. This triggers oxygen-sensitive biological changes in the walls of blood vessels, leading to an elevation in LDL oxidation and a reduction in nitric oxide levels.

Currently, there is a growing interest in studying the link between Apo A-I and Apo B with DR. There is limited research demonstrating a correlation between apolipoproteins and DR.[3, 6.]

Apolipoprotein A-I is the primary antiatherogenic apolipoprotein found in HDL cholesterol, and it has a significant impact on HDL metabolism. The enzyme lecithin cholesterol acyltransferase, found in high-density lipoprotein (HDL), is activated by Apo A-I. This activation leads to the catalysis of a process that results in the formation of cholesterol esters (CEs). This high-density lipoprotein (HDL) that is rich in cholesterol esters performs the function of removing cholesterol from peripheral tissues and transporting it to the liver, a process known as reverse cholesterol transport."[8]".

Apolipoprotein B is a prominent apolipoprotein found in very low-density lipoprotein (VLDL) and LDL, which are lipoproteins that contribute to the development of atherosclerosis. It aids in dissolving the cholesterol inside the LDL complex, hence enhancing the ability of LDL to carry cholesterol for later accumulation in the artery wall. Each lipoprotein particle contains only one molecule of Apo B. Therefore, the amount of Apo B serves as a precise indicator of VLDL and LDL particles. Because there are significant differences in the levels of cholesterol in these lipoproteins, measuring Apo B is more accurate in determining the concentration of atherogenic lipoprotein particles compared to LDL cholesterol or non-HDL cholesterol levels. While the routine use of apolipoprotein measures is not common, they appear to have clear and substantial connections with diabetic retinopathy (DR) compared to standard lipids. "[9]".

Nevertheless, the degree to which these apolipoprotein measures can be utilised to ascertain the susceptibility of diabetic patients to diabetic retinopathy is currently uncertain.

Therefore, the objective of this study was to compare the association of DR with the serum levels of Apo- A1, Apo B, Apo B/Apo-A1 ratio and with traditional lipid profile in Type 2 Diabetes Mellitus (T2D) patients.

MATERIALS AND METHODS

The current study was a case-control study carried out with a cohort of 300 individuals who sought medical care at both the outpatient and inpatient departments of Ophthalmology. The patients provided informed consent. This study examined people who had been diagnosed with type 2 diabetes (T2D). The study participants were chosen from the age bracket of 35 to 80 years. Relevant clinical data, such as gender and duration of diabetes, were collected from both medical records and personal interviews with the patients.

Patients who had Type 1 diabetes, secondary diabetes, gestational diabetes, severe hypertension, acute infections, known cardiovascular and renal diseases, liver dysfunction, severe anaemia, thyroid disorders, a history of glaucoma, previous vitreo-retinal surgery, and/or a dense cataract were not included.

The study was carried out with 100 T2D patients and was divided into 2 groups and they were compared with 50 subjects of age and sex matched healthy controls.

Group I-Con: 100 subjects of age and sex matched healthy controls.

Group II-Non-DR: 88 subjects with T2D for more than 5 years without signs of DR

Group III: DR-112 subjects with T2D for more than 5 years with signs of DR.

Participants underwent a standardised clinical assessment and a dilated fundus examination. The diagnosis and grading of diabetic retinopathy were performed using a slit lamp in the presence of dilated fundus. The patients with retinopathy (Group III) were categorised into two subgroups: non-proliferative diabetic retinopathy (NPDR) and proliferative diabetic retinopathy (PDR). NPDR was divided into three sub-classifications: mild, moderate, and severe. Mild NPDR includes mild

microaneurysm or intra-retinal haemorrhage. Moderate NPDR includes moderate microaneurysm or intra-retinal haemorrhage, early Intra-Retinal Microvascular Abnormality (IRMA), and the presence or absence of hard/soft exudates. Severe NPDR includes severe microaneurysm or intra-retinal haemorrhage in all four quadrants, venous beading in only two quadrants, and IRMA in only one quadrant.

Blood samples were obtained from each person. Approximately 5 millilitres of blood were extracted from patients who had fasted overnight using EDTA-coated containers for the analysis of Fasting Blood Sugar (FBS), Post-Prandial Blood Sugar (PPBS), and HbA1C. Additionally, the serum was isolated in order to quantify the lipid profile and apolipoprotein levels.

RESULTS

The grades of diabetic retinopathy in patients with type 2 diabetes are provided in Table/Figure 1.Out of the 112 patients, 28 (25%) had mild non-proliferative diabetic retinopathy (NPDR), 48 (43%) had moderate NPDR, 22 (20%) had severe NPDR,The prevalence of diabetic retinopathy (DR) was seen to be high in 54 (48.2%) individuals aged 61-70 years and low in 4 (3.5%) patients aged 35-50 years.[Table/Fig-2] The data demonstrated a positive correlation between the duration of the disease and the incidence of DR, indicating that as the disease duration increased, the occurrence of DR also increased. Groups II and III had elevated levels of FBS compared to Group I. Additionally, when comparing Group III to Group II, it was revealed that patients in Group III had greater FBS levels.Patients diagnosed with Group III, which includes severe non-proliferative diabetic retinopathy (NPDR) and proliferative diabetic retinopathy (PDR), exhibited elevated levels of Hb

Table1: GradesofDRaccordingtoagegroupinT2Dpatients.

AgeinYears	DR(n=112)				Total
	MildNPDR(n=28)25%	ModerateNPDR(n=48)43%	SevereNPDR (n=22)20%	PDR (n=14)12%	
35-50	4	-	-	-	4
51-60	8	24	-	-	32
61-70	14	20	14	6	54
71-80	2	4	8	8	22
Total	28	48	22	14	112

Table-2 : Association of duration of diabetes, fasting blood sugar and HbA1c in T2D patients retinopathy.

Parameters	Group I CON	Group II Non-DR	Group –III {DR(n=112)}				p-value
			Mild NPDR	Moderate NPDR	Severe NPDR	PDR	
Durationof Diabetes	-	8.6±0.63	7.8± 0.7	11.4± 1.2	13.6± 1.8	15.4± 2.4	<0.001**

(years)							
FBS (mg/dl)	96± 15.1	148±23	120 ±18	128± 27	156± 30	173± 34	<0.04*
Hb A1C(%)	5.6±0.34	7.9±0.5	7.8± 0.5	8.1± 0.6	11.1± 1.2	13.8± 1.4	<0.001**

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*p<0.05statisticallysignificant

**p<0.001statisticallyhighlysignificant ,p- valuesarecalculatedusingone wayanova

There is a correlation between poor glycemic control (elevated HbA1c levels) and the prevalence of diabetic retinopathy. Retinopathy was observed in the fundus images of patients with diabetic retinopathy.

[Table 3] displays the conventional lipid profile and apolipoproteins in patients with type 2 diabetes (T2D), both with and without diabetic retinopathy (DR). Elevated levels of triglycerides (TG) were reported in Group II and Group III, particularly in cases of severe non-proliferative diabetic retinopathy (NPDR) and proliferative diabetic retinopathy (PDR), when compared to Group I.When comparing Groups II and III, TG levels were significantly higher in the severe NPDR and PDR of the DR group compared to the non-DR group (P<0.001).The level of HDL-C showed an inverse association with the severity of diabetic retinopathy (p<0.05).There was no significant association of.

Table-3 Association of apolipoproteins and serum lipids in T2D patients with retinopathy.

Parameters	Group-I CON	Group-II Non-DR	DR (n=112)				p-value
			MildNPDR	Moderate	Severe	PDR	
Triglycerides (mg/dl)	134±16	165±37.2	121±13.3	132±14	169±42	202±70	<0.001**
TotalCholesterol(mg/dl)	158±25.3	202±34.3	188±27.1	204±33	209±31	213±36.3	0.38
HDL Cholesterol (mg/dl)	58±12	51±11	52±13	50±11.3	48±10.8	46±8.2	<0.05*
LDL Cholesterol (mg/dl)	109±18.4	128±26.8	115±19	117±19.6	120±20	129±28	0.42
Apo- A1 (mg/dl)	285±13.4	230±43	212±25	193±19	136±17	118±14	<0.001**
Apo-B (mg/dl)	102±9.8	120±19.2	125±20	149±32.4	174±43.4	197±48.8	<0.001**
Apo B/A po-A1 ratio	0.72±0.05	0.94±0.1	0.78±0.08	0.8±0.07	1.27±0.2	1.78±0.45	<0.001**

:*p<0.05statisticallysignificant

**p<0.001statisticallyhighlysignificant

LDL-C and total cholesterol were higher in Group III compared to Groups I and II.The research suggests that TG (triglycerides) are linked to a higher occurrence and severity of diabetic retinopathy (DR) in individuals with type 2 diabetes (T2D), rather than LDL (low-density lipoproteinAdditionally, both blood Apo-B levels and the ApoB-to-ApoA1 ratio were found to be greater in individuals with diabetic retinopathy (DR) compared to the non-DR group. This indicates a significant and close relationship between apolipoproteins and the progression and severity of DR ($P < 0.001$).

DISCUSSION

Carbohydrate metabolism dysfunction results in diabetes, which has reached epidemic levels in both developing and industrialised countries.This has resulted in one of the complications of diabetes, such as diabetic retinopathy, which is relatively frequent in the general population.

Studies have indicated that diabetic retinopathy (DR) can develop at any age, similar to diabetes. Furthermore, individuals who acquire the condition at a younger age have a higher likelihood of developing retinopathy [10].Subsequent research has revealed that the duration of diabetes is one of the risk variables associated with the development of diabetic retinopathy (DR) [11,12].In this study, the duration of diabetes has been identified as a significant factor in determining the severity of diabetic retinopathy, which is consistent with the existing literature.

Dysregulated glucose regulation is a significant indicator for type 2 diabetes (T2D), as seen by elevated fasting blood sugar (FBS) levels in the current investigation. Optimal metabolic control is crucial for effective ophthalmic care of individuals with diabetes.Several studies have documented a strong correlation between elevated HbA1c levels and the onset and advancement of diabetic retinopathy (DR) [13-15].The present study discovered a higher level of HbA1c in groups II and III compared to the control group. This suggests that inadequate control of blood sugar levels could impact not only the formation, but also the advancement of diabetic retinopathy.
Dyslipidemia can be primarily characterised by increased levels of triglycerides, which are thought to be a significant risk factor for the development of certain conditions.

Early detection of diabetic retinopathy (DR) using more effective biomarkers. Therefore, our results, although from a limited sample size, suggest that these apolipoprotein measures may possess the capability to act early and serve as superior indicators of diabetic retinopathy compared to standard lipid measurements.

Among the subjects, there were individuals diagnosed with diabetes.Previous studies have suggested that serum lipids may be risk factors for diabetic retinopathy (DR) [16-18]. However, the relationship between lipids and DR has not been extensively explored in relation to diabetes duration, HbA1c, and blood pressure. Our investigation showed that among the standard lipid measures, only elevated triglyceride levels (TG) and decreased high-density lipoprotein cholesterol (HDL) were linked to

diabetic retinopathy (DR).Our findings corroborate earlier studies indicating that traditional serum lipids do not exhibit a stronger or consistent correlation with DR [19-22].

There is a scarcity of literature demonstrating that apolipoproteins are superior diagnostic indicators for DR.While the precise underlying process remains uncertain, we have compelling evidence supporting the plausibility of our findings. Research has indicated that ApoA1, the primary structural protein of HDL, plays a crucial role in protecting the retina from lipid buildup and inflammation-induced lipotoxicity, which can lead to diabetic retinopathy (DR). Higher levels of ApoA1 are associated with these protective mechanisms [23,24]. Additionally, ApoA1 exhibits anti-inflammatory and anti-oxidant properties [25] and plays a crucial role in the transport of lipids inside the retina [26].Thus, it is feasible that a deficiency of this protective chemical may be a causal factor in promoting DR.The current investigation revealed reduced levels of serum ApoA1 in patients with diabetic retinopathy (DR). Wuetal. observed a correlation between increasing levels of ApoB and the severity of DR. They indicated that higher ApoB levels may result in increased synthesis of lipoprotein-related toxins, which might cause damage to the retinal vascular cells [27].Additionally, it has been shown that the reduced removal of LDL and TG is caused by an excessive generation of ApoB and a decrease in lipoprotein lipase activity. This has implications in the malfunctioning of endothelial cells and the local inflammatory response, leading to the release of cytokines and growth factors that promote the formation of new blood vessels in the retina [28].A literature search revealed that there was no positive association between ApoA1 and DR. However, the ApoB-to-ApoA1 ratio was found to be favourably correlated with DR in previous studies [20, 29, 30].Similar findings were recently reported by Prakash et al. and Ankit et al. [21,22].Our findings are highly congruent with the literature referenced earlier.

CONCLUSION

The results of our investigation indicate a strong correlation between serum apolipoproteins (ApoA1, ApoB, and the ApoB-to-Apo-A1 ratio) and the advancement and severity of diabetic retinopathy in patients with type 2 diabetes. In our investigation, it was found that standard serum lipid levels, with the exception of triglycerides (TG) and high-density lipoprotein (HDL) cholesterol, did not show a significant association with diabetic retinopathy (DR). Recently, the correlation between serum ApoA1 and B and DR has been disproven, which provides substantial support for our work. Although apolipoprotein measurements are considered emerging biomarkers for assessing diabetic retinopathy (DR), they have not been routinely investigated by clinicians. Further studies are needed to unravel the underlying mechanisms of their protective effects in order to decrease the prevalence of DR and provide comprehensive eye care to prevent blindness. Medical practitioners in different geographical areas are advised to conduct comparable investigations to determine the presence of such associations.Pending confirming results in larger research will assist ophthalmologists.Early detection of diabetic retinopathy (DR) using improved biomarkers. Therefore, our results, which are based on a limited number of participants, suggest that these apolipoprotein measures may have the potential to operate as early and superior biomarkers of diabetic retinopathy compared to standard lipid measurements.

REFERENCES

1. Brownlee, M.; Aiello, LP.; Cooper, ME., et al. Complications of diabetes mellitus. In: Melmed S, Larsen PR, Polonsky KS, et al., editors. Williams's textbook of endocrinology. 12th ed. USA: Saunders Elsevier publishing Division; 2008. pp. 1417-1442.
2. Zheng Y, He M, Congdon N. The worldwide epidemic of diabetic retinopathy. Indian J Ophthalmol 2012 Sep-Oct;60(5): 428-431.
3. Sasongko MB, Wong TY, Kawasaki R, Jenkins A, Shaw J, Wang JJ, Nguyen TT. Serum Apolipoprotein A-I and B are stronger biomarkers of diabetic retinopathy than traditional lipids. Diabetes Care 2011 Feb;34(2):474-479.
4. Kowluru RA, Chan PS Oxidative stress and diabetic retinopa- thy. Exp Diabetes Res 2007 Feb;2007(43603):1-12.
5. Rema M, Pradeepa R. Diabetic retinopathy: an Indian perspec- tive. Indian J Med Res 2007 Mar;125(3):297-310.
6. Hu A, Luo Y, Li T, Guo X, Ding X, Zhu X, Wang X, Tang S. Low serum apolipoprotein A1/B ratio is associated withproliferative diabetic retinopathy in type 2 diabetes. Graefes Arch Clin Exp Ophthalmol 2012 Jul;250(7):957-962.
7. Sinav S, Onelge MA, Onelge S, Sinav B. Plasma lipids and lipoproteins in retinopathy of type I (insulin-dependent) diabetic patients. Ann Ophthalmol 1993 Feb;25(2):64-66.
8. Thomas, L. Clinical laboratory diagnostics. 1st ed. Frankfurt: TH-Books Verlagsgesellschaft mb H; 1998.
9. Talmund. Apo B and TG levels predict CHD risk. ArteriosclerThrombVasc Biol 2002:1918-1922.
10. PrevalenceofdiabeticretinopathyandcorrelationwithsystemicriskfactorsinType2diabetesmellitusin atertiarycarehospital.SchJAppMedSci.2015;3(7C):2659-54.
11. Facts About Diabetic Eye Disease | National Eye Institute [Internet]. [cited 2017Jul 1]. Available from: https://nei.nih.gov/health/diabetic/retinopathy
12. NiaziMK,AkramA,NazMA,AwanS.DurationofdiabetesasasignificantfactorfOr retinopathy. Pak J Ophthalmol. 2010;26(4):182–86.
13. Fong DS, Aiello LP, Ferris FL, Klein R. Diabetic retinopathy. Diabetes Care.2004;27(10):2540–53.
14. Al-ShammariFK,Al-MeraghiO,NasifA,Al-OtaibiS.Theprevalenceofdiabeticretinopathyandassociatedriskfactorsintype2diabetesmellitusinA l-Naeemarea(Kuwait).MiddleEastJFamMed.2005;3(2):45–48.
15. Al-Bdour MD, Al-Till MI, Abu Samra KM. Risk factors for diabetic retinopathyamongjordaniandiabetics.MiddleEastAfrJOphthalmol.2008;15(2):77–80.
16. Khan AR, Wiseberg JA, Lateef ZAA, Khan SA. Prevalence and determinantsof diabetic retinopathy in Al hasa region of saudiarabia: primary health carecentrebasedcross-sectionalsurvey,2007-2009.MiddleEastAfrJOphthalmol.2010;17(3):257–63.
17. Klein BE, Moss SE, Klein R, Surawicz TS. The Wisconsin Epidemiologic Study ofDiabetic Retinopathy. XIII. Relationship of serum cholesterol to retinopathy andhard exudate. Ophthalmology. 1991;98(8):1261–65.

18. Ferris FL, Chew EY, Hoogwerf BJ. Serum lipids and diabetic retinopathy.EarlyTreatmentDiabeticRetinopathyStudyResearchGroup.DiabetesCare.1996;19(11):1291–93.
19. HuA,LuoY,LiT,GuoX,DingX,ZhuX,etal.LowserumapolipoproteinA1/Bratioisassociatedwithproliferativediabeticretinopathyintype2diabetes.GraefesArch Clin Exp Ophthalmol Albrecht Von Graefes Arch Klin Exp Ophthalmol.2012;250(7):957–62.
20. Sasongko MB, Wong TY, Nguyen TT, Kawasaki R, Jenkins A, Shaw J, et al.SerumapolipoproteinAIandBarestrongerbiomarkersofdiabeticretinopathythantraditional lipids. Diabetes Care. 2011;34(2):474–79.
21. Prakash G, Agrawal R, Satsangi SK, Prakash S. Comparison of serumapolipoproteinsandtraditionallipidsineyeswithdiabeticretinopathyinIndianpopulation:acaseseries.MiddleEastAfrJOphthalmol.2016;23(2):212–14.
22. Ankit BS, Mathur G, Agrawal RP, Mathur KC. Stronger relationship of serumapolipoproteinA-1andBwithdiabeticretinopathythantraditionallipids.IndianJEndocrinol Metab. 2017;21(1):102–05.
23. AgroiyaP,PhilipR,SaranS,GutchM,TyagiR,GuptaKK.etal.,Associationofserumlipidswithdiabeticretinopathyintype2diabetes.IndianJEndocrinolMetab.2013;17(Suppl1):S335–37.
24. SimóR,HigueraM,García-RamírezM,CanalsF,García-ArumíJ,HernándezC.et al., Elevation of apolipoprotein A-I and apolipoprotein H levels in the vitreousfluidandoverexpressionintheretinaofdiabeticpatients.ArchOphthalmolChicIll 1960. 2008;126(8):1076–81.
25. SimóR,García-RamírezM,HigueraM,HernándezC.ApolipoproteinA1 is overexpressed in the retina of diabetic patients. Am J Ophthalmol.2009;147(2):319–325.e1.
26. TserentsoodolN,GordiyenkoNV,PascualI,LeeJW,FlieslerSJ,RodriguezIR.et al., Intraretinal lipid transport is dependent on high density lipoprotein-likeparticles and class B scavenger receptors. Mol Vis. 2006;12:1319-33.
27. WuM,ChenY,WilsonK,ChirindelA,IhnatMA,YuY,etal.Intraretinalleakageand oxidation of LDL in diabetic retinopathy. Invest Ophthalmol Vis Sci.2008;49(6):2679-85.
28. JoussenAM,MurataT,TsujikawaA,KirchhofB,BursellS-E,AdamisAP.etal.,Leukocyte-MediatedEndothelialCellInjuryandDeathintheDiabeticRetina.Am JPathol.2001;158(1):147-52.
29. ChangY-C,WuW-C.Dyslipidemiaanddiabeticretinopathy.RevDiabetStudRDS.2013;10(2–3):121-32.
30. PusparajahP,LeeL-H,AbdulKadirK.MolecularMarkersofDiabeticRetinopathy:Potential Screening Tool of the Future? Front Physiol [Internet]. 2016 Jun 1[cited 2017 Jul 1];7. Available from: http://www.ncbi.nlm.nih.gov/pmc/articles/PMC4887489/