Association of Serum Cystatin-C Level with Diabetic Retinopathy in a Sample of Iraqi Patients with Type 2 Diabetes Mellitus

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ABSTRACT

Background: Retinal complications of diabetes mellitus mainly Type 2 is known as diabetic retinopathy. Screening or early diagnosis of diabetic retinopathy involve time-consuming and costly clinical retinal examination and imaging, but they are crucial to prevent vision loss. Many studies have linked diabetic retinopathy with certain biochemical parameters including cystatin-C, suggesting that they could serve as biomarkers for diagnosis of diabetic retinopathy.

Aim To investigate the possible role of serum level of cystatin-C in type 2 diabetes as a biomarker to identify the presence diabetic retinopathy.

Materials Methods This cross-sectional study was caried out in three hospitals in Baghdad, Iraq. A total of 88 patients with type 2 diabetes mellitus ≥ 40 years were enrolled and assorted into three groups: 30 patients with no signs of diabetic retinopathy (control), 28 patients with Non-Proliferative Diabetic Retinopathy (NPDR), and 30 patients with Proliferative Diabetic Retinopathy (PDR). Serum levels of cystatin-C, cholesterol, triglycerides, high density lipoprotein cholesterol, as well as glycated hemoglobulin percentage were measured for all participants. Statistical tests of significance, correlation and receiver operating characteristic (ROC) analyses were used.

Results The mean level of cystatin-C was significantly higher in patients with diabetic retinopathy of all stages than in controls. The mean level of cystatin-C in all- stages diabetic retinopathy patients (NPDR and PDR) was 6.209 ± 3.528 ng/dl and in control group was 3.981 ± 2.927 ng/dl, (P= 0.004). The ROC curve analysis showed that cystatin-C level had a sensitivity of 96.6% and a specificity of 63.4% in discrimination of diabetic retinopathy patients from controls.

Conclusion Serum level of Cystatin-C is a useful biomarker to detect diabetic retinopathy in type 2 diabetic patients in early stage.

Key words: Cystatin-C; Type 2 diabetes mellitus; Screening for diabetic retinopathy

INTRODUCTION

Diabetes Mellitus (DM) is a metabolic disorder marked by chronic high blood sugar (hyperglycemia) due to defects in insulin secretion or action.[1] The DM global burden is due to its complications like heart disease, neuropathy, amputations, retinopathy and blindness causing high morbidity and mortality.[2]

Diabetic Retinopathy (DR) is the most common eye complication of DM, it is a major cause of blindness among working-age individuals in developed countries.[3] Global DR prevalence is projected to rise from 103 million in 2020 to 161 million in 2045 which will contribute 55.8% increase within 25 years.[4] In East Mediterranean Region, prevalence reaches 31% with a variation depending on healthcare systems.[5] Arabian Gulf countries showed a DR prevalence of 20.5% among patients with DM, where the highest rate was in Saudi Arabia while United Arab Emirates showed the lowest prevalence rate.[6] In Iraq, high DR prevalence was noted, with 71% in Baghdad, and 33.3% in Basra.[7, 8] These facts underscore the importance of early screening and examinations for DR. A comprehensive dilated eye examination using a slit lamp and Optical Coherence Tomography (OCT) provide reliable diagnostic tools for DR but both are time consuming and costly.[9]

Ophthalmologists classify DR into two main forms according to signs and degree of retinal damage; Non-Proliferative Diabetic Retinopathy (NPDR), and Proliferative Diabetic Retinopathy (PDR).[10] The NPDR is differentiated into three stages mild, moderate and severe.[11]

It is essential to detect DR as early as possible to prevent vision loss. Screening rates are still low, only 50% or less of patients with DM were detected in early stages of DR and were referred to receive proper ophthalmic care.[12]

In terms of pathophysiology, DR is a microvascular disease characterized by elevated vascular flow and vascular leakage due to the presence of vascular lesions, neovascularization and inflammation.[13] Oxidative stress, edema in tissues, adhesion molecules expression, cytokines, reactive glia, and apoptosis of inner retinal cell also play a role in the pathophysiology [figure 1].[14]

Cystatin-C (Cys-C) is a cysteine proteinase inhibitor protein that is found in various body fluids. The gene of this protein is belonging to the type 2 cystatin gene family on chromosome 20.[15] Clinically, Cys-C level estimates glomerular filtration rate due to its low molecular weight, complete kidney filtration with complete reabsorption and metabolism in proximal tubules.

The role of Cys-C in DR may be attributed to the following aspects. First, DR is considered as a vascular complication of diabetes involving changes in blood vessels and the potential connection of Cys-C to endothelial health might impact the blood vessels of the retina and affecting the progression of the condition.[16] Second, Cys-C is associated with inflammation and endothelial dysfunction which are processes implicated in the development and progression of DR. Chronic inflammation and damage to blood vessel walls are known features of DR.[17]

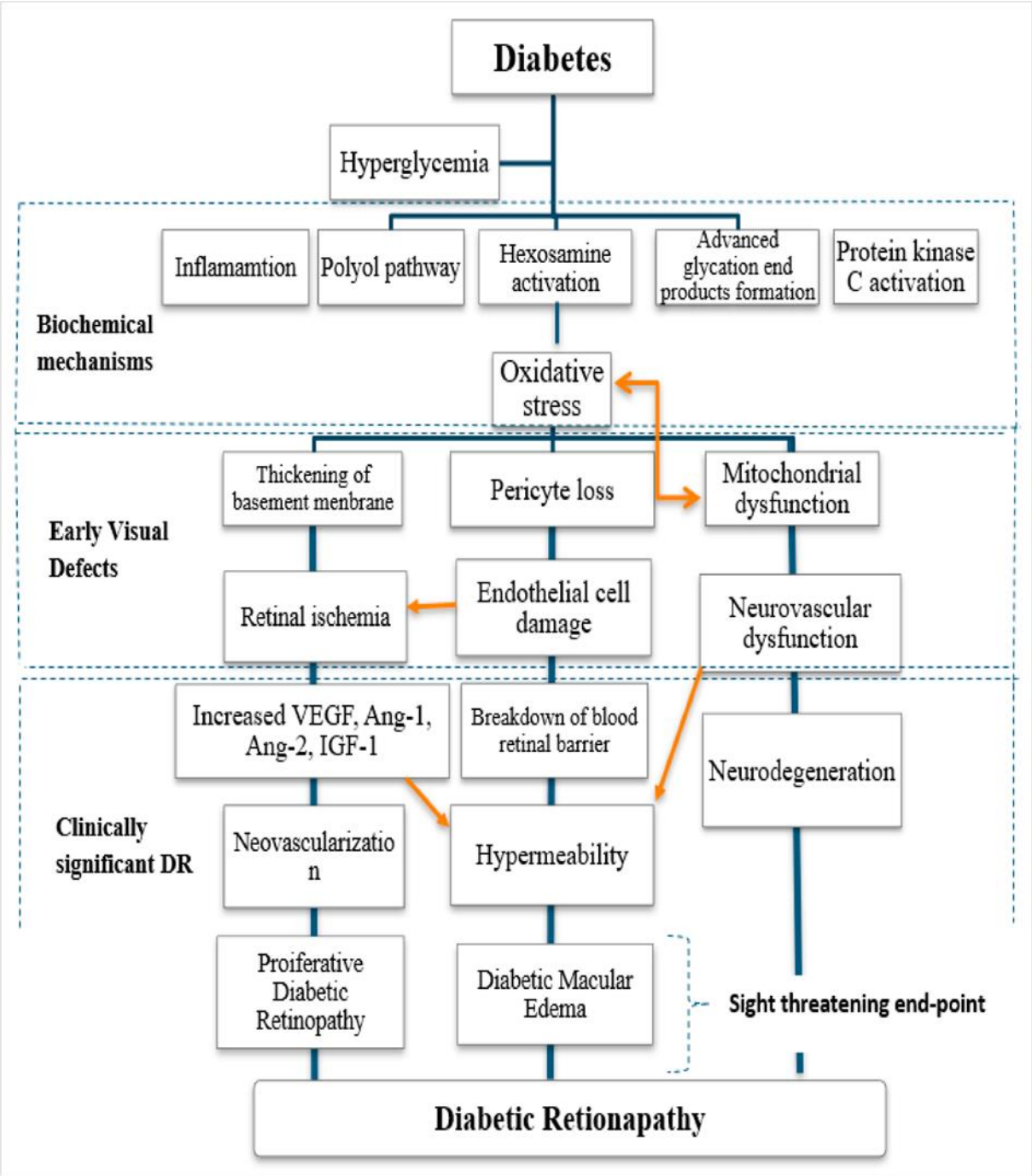

Figure.1: Diagrammatic synopsis of pathogenesis and pathophysiology of diabetic retinopathy[14]

Patients and Methods

This cross-sectional study was carried out at the department of chemistry and biochemistry, college of medicine, Mustansiriyah University. A sample of 88 patients ≥ 40 years old with Type 2 Diabetes Mellitus (T2DM), who attended ophthalmology outpatient clinics in three hospitals in Baghdad, Iraq were enrolled in the study. They were consecutively selected and assorted, according to their clinical examination and clinical investigations into three main groups: **PDR group:** Involved 30 patients with features of active PDR in the worse eye, either newly diagnosed or being treated during the last 2 years. **NPDR group:** Involved 28 patients with features of NPDR in the worse eye. The patients in the NPDR group involved two sub groups; 14 patients with mild signs of DR (Mi- NPDR subgroup), and 14 with moderate to severe signs of DR (Mo to S-NPDR subgroup). The total patients in the PDR group and the two subgroups of the NPDR group (58 patients) comprised patients with all-stages diabetic retinopathy or total DR patients. The third group is the **control group,** it involved 30 patients with T2DM but no DR in both eyes. During selection of participants in this study, sex and age matching in the three study groups was taken into consideration.

Inclusion criteria: Patients with T2DM, aged 40 years old and above.

Exclusion criteria: Patients with type1 DM, chronic renal diseases, chronic illness and malignancies, uncontrolled hypertension, retinal diseases other than DR, pregnant women and smoking.

A study questionnaire was designed to collect data by direct personal interview of all participants. The data collected included general information regarding sex, age, duration of DM, smoking and medical history (chronic illness, malignancy, renal disease), as well as taking blood pressure.

A fasting venous blood sample was collected for measurement of Cys-C by an Enzyme Linked Immuno-Sorbent Assay (BIO-RAD, Germany), HbA1c% by an auto-analyzer (Lifotronic GH-900 Plus, China), lipid profile of triglyceride, total cholesterol, high density lipoprotein cholesterol (HDL-C) and renal function tests by an automated analyzer (SPINREACT Spin 200, Spain).

Ethical approval

This study was approved by the ethical committee of department of chemistry and biochemistry, College of Medicine, Mustansiriyah University. In addition, official approvals were obtained from Baghdad Al-Karkh and Baghdad Al-Rusafa Health Directorates to enroll patients from ophthalmology out-patient clinics in hospitals they manage. All patients had the right to participate willingly in the study and no enticements were offered to the participants in return for their participation. Explanation of the information and the tests needed for the purpose of the study was given to patients. Informed written consents were obtained from patients to use their information and test results in the study. Data obtained were treated as confidential and used only for the purpose of this study.

Statistical Analysis

All statistical analyses were carried out using the IBM SPSS software. Data collected by study questionnaire and readings obtained from biochemical analyses were statistically analyzed to calculate simple measures of frequency, percentages, means, ranges and standard deviations. The significance of the differences between the study groups were calculated using the suitable statistical tests such as t-test, chi square, ANOVA and Kruskal Wallis tests. Correlation and receiver operating characteristic (ROC) curve analyses were done.

Results

The 88 participants in this study constituted 58 males and 30 females. Males constituted 70% of control group, 60.7% of NPDR group and 66.7% of PDR group, while females represented 30%, 39.3% and 33.3% respectively [Figur 2].

The age of control group ranged from 40 to 76 years old with a mean of 56.8 years. The NPDR group had approximately similar age range (48 to 80 years) with a mean age of 60.3 years. The PDR group had an age range between 49 and 72 years with a mean of 59.7 years which is approximately the same as the other two groups. There was no significant difference in sex and age between groups. Duration of DM varied between 3 to 30 years. Mean duration of DM in control group was 10.6 years (range 3-25 years), the NPDR had a mean DM duration of 15.4 years (range 10-20 years) and PDR mean DM duration was 16.2 years (range 5-30 years). Table 1 showed that the duration of DM was significantly different between the groups. [Table1]

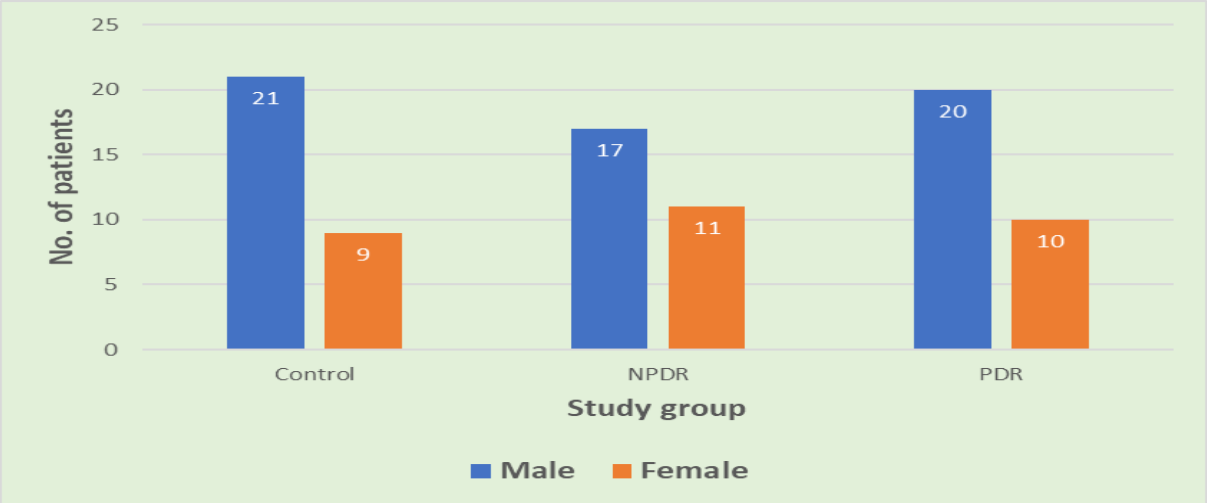

Figure.2: Sex distribution among study groups

Table 1: Comparison of basic demographic characteristics of study groups

Variable		Control N=30	NPDR N=28	PDR N=30	P-value*
Sex N (%)	Male	21 (70.0%)	17 (60.7%)	20 (66.7%)	0.753 Chi^2 test
	Female	9 (30.0%)	11 (39.3%)	10 (33.3%)	
Age (years) (Mean ± SD)		56.77 ± 9.87	60.29 ± 7.67	59.67 ± 6.26	0.209 ANOVA test
Duration of DM (years) (Mean ± SD)		10.63 ± 6.78	15.43 ± 2.43	16.23 ± 2.32	0.0001 Kruskal Wallis test

*P-value: Significance at ≤ 0.05

The comparison of biochemical parameters of study groups revealed that PDR group had the highest levels of HbA1c%, total cholesterol and triglyceride while control group had the highest level of HDL-C. In spite of these results, no statistically significant differences in all biochemical parameters among study groups were found [Table 2].

Levels of Cys-C were compared between controls in which mean Cys-C was (3.98 ± 2.93 ng/dl) and total DR patients (NPDR and PDR) in whom mean Cys-C was (6.21 ± 3.53 ng/dl). The comparison revealed statistically significant higher levels of Cys-C in patients with DR than in controls (P= 0.004) as shown in table 3.

Table 2: Comparison of basic biochemical parameters among study groups

Parameter (mean ± SD)	Control N=30	NPDR* N=28	PDR† N=30	P-value‡
HbA1c (%)	7.78 ± 1.17	8.14 ± 1.60	8.19 ± 1.33	0.456 ANOVA test
Cholesterol (mg/dl)	184.38 ± 56.73	171.26 ± 49.22	187.53 ± 32.51	0.338 Kruskal Wallis test
Triglyceride (mg/dl)	130.41 ± 54.67	137.90 ± 59.45	147.55 ± 85.48	0.662 Kruskal Wallis test
HDL-C (mg/dl)	49.81 ± 10.43	46.65 ± 12.27	44.46 ± 13.30	0.231 Kruskal Wallis test

* NPDR= non-proliferative diabetic retinopathy. † PDR= Proliferative diabetic retinopathy.
‡P-value: Significance at ≤ 0.05.

Table 3: Comparison of cystatin-C level between all-stages diabetic retinopathy patients and control group

	Control N=30	Total DR* Patients N= 58	P-value†
Cystatin- C (ng/dl) (mean±SD)	3.98 ± 2.93	6.21 ± 3.53	0.004 Independent t-test

*DR= Diabetic retinopathy. †P value: Significance at ≤ 0.05

Comparison of the level of Cys-C among the main study groups is shown in table 4. The comparison revealed a statistically significant difference in levels of Cys-C between groups (P= 0.014). Patients' Mean Cys-C in NPDR and in PDR groups were 5.97 ± 3.11 ng/dl and 6.43 ± 3.9 ng/dl respectively, they were noticeably higher than in control group (3.89 ± 2.93 ng/dl).

Table 5 shows a comparison of Cys-C levels according to the stage of DR. The comparison revealed statistically non-significant difference (P= 0.89) among the mean of Cys-C levels .

Table 4: Comparison of Cystatin-C level among main study groups

	Control N=30	NPDR* N=28	PDR† N=30	P-value‡
Cystatin-C (ng/dl) (mean ± SD)	3.98±2.93	5.97±3.12	6.43±3.92	0.014 Kruskal Wallis test

*NPDR= Non-proliferative diabetic retinopathy. † PDR= Proliferative diabetic retinopathy.
‡P-value: Significance at ≤ 0.05.

Table 5: comparison of cystatin-C level according to the stage of diabetic retinopathy

	Mild NPDR* N=14	Moderate to severe NPDR N=14	PDR† N=30	P-value‡
Cystatin-C (ng/dl) (mean ± SD)	5.96 ± 3.09	5.98 ± 3.24	6.43 ± 3.92	0.89 Kruskal Wallis test

*NPDR= Non-proliferative diabetic retinopathy. †PDR= Proliferative diabetic retinopathy.
‡P-value: Significance at ≤ 0.05

Correlation analysis to investigate the association of Cys-C level with the basic biochemical parameter was carried out. Table 6 showed that HbA1c% value had a significant positive correlation with Cys-C level, while Serum levels of total cholesterol, triglyceride and HDL-C were found to have no significant correlation with Cys-C level.

The Cys-C level was evaluated for its performance in discrimination of patients with DR from control group patients using ROC analysis [Table 7]. They showed an area under the curve (AUC) of 0.70 with a diagnostic accuracy of 80%. At an optimum cut-off value of 2.29 ng/dl, Cys-C level had 96.6% sensitivity and 63.4% specificity in differentiation of DR patients.

Table 6: Correlation of cystatin-C level with basic biochemical parameters

		Cystatin-C
HbA1c %	R	0.378
	P-value*	0.0001
Cholesterol	R	0.015
	P-value	0.886
Triglyceride	R	- 0.012
	P-value	0.912
High Density Lipoprotein Cholesterol	R	- 0.045
	P-value	0.680
Cystatin-C	R	
	P-value	

*P-value: Significance at ≤0.05

Table 7: Receiver operating characteristic curve analysis for cystatin-C level to discriminate patient with diabetic retinopathy from patients without retinopathy (controls)

	AUC	Cut-off value	Sensitivity	Specificity	Accuracy	P-value
Cystatin-C	0.70	2.29 ng/dl	96.6%	63.4%	80%	0.003

AUC = area under the curve

DISCUSSION

Diabetic retinopathy is one of the most common complications of DM and is one of the leading causes of vision loss.[3] Early diagnosis and management are essential in preventing such destiny and screening for DR is recommended for all patients with T2DM at time of diagnosis and during follow-up. About half of patients with DM are not screened for DR because of the time consuming and costly examination procedures.[12] Finding rapid and relatively affordable test is important to improve DR screening rates in patients with T2DM and to send only high-risk patients to confirmation the examinations.

In this study, participants in all groups (control, NPDR and PDR) have been considered for sex and age-matching to eliminate the confounding effect of these factors. Sex was reported to be a predisposing factor for DR development in T2DM patients. Some studies showed that women are more likely to develop DR and sight loss than men and female sex is associated with higher prevalence of DR in all stages.[18,19] Age also plays a role in development of DR in diabetic patient, some studies have reported that old age is an important risk factor.[20,21]

Duration of DM has strong association with the progression of DR, so it was unlikely to match participants of the study groups for this factor. This association was proved by Amelia and colleagues in a study carried out in Indonesia 2021 and in Malaysia by Naserrudin and colleagues 2022.[20,22]

Metabolic changes play a role in the development of DR and so this study has investigated four biochemical parameters which are known to have the most important role, these parameters are HbA1c, cholesterol, triglyceride and HDL-C.

Participants in the three main groups had close levels of the four important biochemical parameters with no significant difference as shown in table 2, which eliminates any confounding effect of these biochemical parameters on the biomarker of this study. A study in Saudi Arabia had linked high levels of HbA1c with the development of DR.[23] Another study found that increased levels of HbA1c is likely to be associated with progression to PDR.[24] Levels of serum cholesterol and TG are also found to be associated with the occurrence of DR, positive association between total cholesterol/TG ratio and the presence of DR in Chinese individuals has been found.[25] Association of base line levels of cholesterol and TG with the occurrence of DR was found.[26] Positive association was found between levels of HDL-C and the risk to develop DR.[27] There is an inversed relationship between serum levels of HDL-C and the risk of DR development in T2DM patients, lower levels of HDL-C were found to be significantly associated with advanced forms of DR.[28,29]

Cystatin-C serum level showed a significant elevation in T2DM patients with DR than those without DR (P= 0.004) indicating an association between elevation of Cys-C levels and DR development in T2DM patients. Qureshi and colleagues in a study carried out in Lahore, Pakistan found the same relation and concluded that level of Cys-C can be useful biomarker in detecting DR and its progression.[30] The results of this study also agrees with a meta-analysis study caried out in China that found the same relation of Cys-C with DR and recommended Cys-C as a screening marker and even a prognostic indicator for DR.[31]

The serum level of Cys-C was found by the researcher to be highly sensitive (96.6%) and moderately specific (63.4%) in differentiating diabetic patients with DR from those without DR, so it is a good and affordable biomarker for screening to find diabetic patients with DR even in early stage.

Conclusions

According to the above findings in addition to the high sensitivity of Cys-C level in discrimination of DR patients from DM patients with no DR, it could be concluded that serum Cys-C level is a useful biomarker to detect DR patients in early stage.

References

1. Petersmann A, Müller-Wieland D, Müller UA, Landgraf R, Nauck M, Freckmann G, et al. Definition, Classification and Diagnosis of Diabetes Mellitus. Experimental and Clinical Endocrinology and Diabetes. 2019;127.

2. Zimmet P, Alberti KG, Magliano DJ, Bennett PH. Diabetes mellitus statistics on prevalence and mortality: Facts and fallacies. Nature Reviews Endocrinology. 2016;12(10).

3. Querques G. Eye complications of diabetes. Acta Diabetologica. 2019;56(9).

4. Tan TE, Wong TY. Diabetic retinopathy: Looking forward to 2030. Front Endocrinol [Internet]. 2023 Jan 9 [cited 2024 Aug 25];13. Available from: https://www.frontiersin.org/journals/endocrinology/articles/10.3389/fendo.2022.1077669/full

5. Heiran A, Azarchehry SP, Dehghankhalili S, Afarid M, Shaabani S, Mirahmadizadeh A. Prevalence of diabetic retinopathy in the Eastern Mediterranean Region: a systematic review and meta-analysis. J Int Med Res. 2022 Oct 1;50(10):03000605221117134.

6. Mohamed Z, Al-Natour M, Al Rahbi H. Prevalence of Diabetic Retinopathy Among Individuals with Diabetes in Gulf Cooperation Council countries: A Systematic Review and Meta-analysis. Oman Med J. 2024 Jan 31;39(1):e585.

7. Al Ashoor M, Al Hamza A, Zaboon I, Almomin A, Mansour A. Prevalence and risk factors of diabetic retinopathy in Basrah, Iraq. J Med Life. 2023 Feb;16(2):299–306.

8. Tawfeeq AS. Prevalence and risk factors of diabetic retinopathy among Iraqi patients with type 2 diabetes mellitus. IRAQI JOURNAL OF COMMUNITY MEDICINE [Internet]. 2015 [cited 2024 Sep 8];28(1). Available from: https://www.iasj.net/iasj/article/127204

9. Boyd K, Turbert D. American Academy of Ophthalmology. 2023 [cited 2024 Aug 31]. Diabetic Retinopathy: Causes, Symptoms, Treatment - American Academy of Ophthalmology. Available from: https://www.aao.org/eye-health/diseases/what-is-diabetic-retinopathy

10. Yang Z, Tan TE, Shao Y, Wong TY, Li X. Classification of diabetic retinopathy: Past, present and future. Front Endocrinol (Lausanne). 2022 Dec 16;13:1079217.

11. Qureshi I, Ma J, Abbas Q. Diabetic retinopathy detection and stage classification in eye fundus images using active deep learning. Multimed Tools Appl. 2021 Mar 1;80(8):11691–721.

12. Chong DD, Das N, Singh RP. Diabetic retinopathy: Screening, prevention, and treatment. CCJM. 2024 Aug 1;91(8):503–10.

13. Forrester JV, Kuffova L, Delibegovic M. The Role of Inflammation in Diabetic Retinopathy. Front Immunol [Internet]. 2020 Nov 6 [cited 2024 Sep 17];11. Available from: https://www.frontiersin.org/journals/immunology/articles/10.3389/fimmu.2020.583687/full

14. Ansari P, Tabasumma N, Snigdha NN, Siam NH, Panduru RVNRS, Azam S, et al. Diabetic Retinopathy: An Overview on Mechanisms, Pathophysiology and Pharmacotherapy. Diabetology. 2022 Mar;3(1):159–75.

15. Abrahamson M, Islam MQ, Szpirer J, Szpirer C, Levan G. The human cystatin C gene (CST3), mutated in hereditary cystatin C amyloid angiopathy, is located on chromosome 20. Hum Genet. 1989 Jun;82(3):223–6.

16. Wang Q, Yang A, Sun F, Zhang M, Xu X, Gao B. Correlation between retinal vascular parameters and cystatin C in patients with type 2 diabetes. Acta Diabetol. 2021 Oct 1;58(10):1395–401.

17. Kreslová M, Jehlička P, Sýkorová A, Rajdl D, Klásková E, Prokop P, et al. Circulating Serum Cystatin C as an Independent Risk Biomarker for Vascular Endothelial Dysfunction in Patients with COVID-19-Associated Multisystem Inflammatory Syndrome in Children (MIS-C): A Prospective Observational Study. Biomedicines. 2022 Nov;10(11):2956.

18. Ding J, Strachan MWJ, Reynolds RM, Frier BM, Deary IJ, Gerald FFR, et al. Diabetic Retinopathy and Cognitive Decline in Older People With Type 2 Diabetes: The Edinburgh Type 2 Diabetes Study. Diabetes. 2010 Aug 26;59(11):2883–9.

19. Xu Y, Wang A, Lin X, Xu J, Shan Y, Pan X, et al. Global burden and gender disparity of vision loss associated with diabetes retinopathy. Acta Ophthalmologica. 2021;99(4):431–40.

20. Naserrudin NA, Jeffree MS, Kaur N, Rahim SSSA, Ibrahim MY. Diabetic retinopathy among type 2 diabetes mellitus patients in Sabah primary health clinics–Addressing the underlying factors. PLOS ONE. 2022 Jan 28;17(1):e0261249.

21. Yun JS, Lim TS, Cha SA, Ahn YB, Song KH, Choi JA, et al. Clinical Course and Risk Factors of Diabetic Retinopathy in Patients with Type 2 Diabetes Mellitus in Korea. Diabetes Metab J. 2016 Oct 5;40(6):482–93.

22. Amelia R, Sari MD, Virgayanti V, Ariga RA, Harahap MS. Effect of duration of illness and lipid profile of type 2 Diabetes Mellitus patients on diabetic retinopathy. IOP Conf Ser: Earth Environ Sci. 2021 Mar;713(1):012058.

23. Almutairi NM, Alahmadi S, Alharbi M, Gotah S, Alharbi M, Almutairi NM, et al. The Association Between HbA1c and Other Biomarkers With the Prevalence and Severity of Diabetic Retinopathy. Cureus [Internet]. 2021 Jan 6 [cited 2024 Dec 31];13. Available from: https://www.cureus.com/articles/44085-the-association-between-hba1c-and-other-biomarkers-with-the-prevalence-and-severity-of-diabetic-retinopathy#!/

24. Perais J, Agarwal R, Evans JR, Loveman E, Colquitt JL, Owens D, et al. Prognostic factors for the development and progression of proliferative diabetic retinopathy in

people with diabetic retinopathy - Perais, J - 2023 | Cochrane Library. 2023 Feb 22 [cited 2025 Jan 3]; Available from: https://www.cochranelibrary.com/cdsr/doi/10.1002/14651858.CD013775.pub2/full

25. Zhong Y, Yue S, Wu J, Guan P, Zhang G, Liu L, et al. Association of the Serum Total Cholesterol to Triglyceride Ratio with Diabetic Retinopathy in Chinese Patients with Type 2 Diabetes: A Community-Based Study. Diabetes Ther. 2019 Apr 1;10(2):597–604.

26. Li Z, Yuan Y, Qi Q, Wang Q, Feng L. Relationship between dyslipidemia and diabetic retinopathy in patients with type 2 diabetes mellitus: a systematic review and meta-analysis. Syst Rev. 2023 Aug 24;12(1):148.

27. Xu W, Xu X, Zhang M, Sun C. Association between HDL cholesterol with diabetic retinopathy in diabetic patients: a cross-sectional retrospective study. BMC Endocr Disord. 2024 May 10;24(1):65.

28. Ezhilvendhan K, Sathiyamoorthy A, Prakash BJ, Bhava BS, Shenoy A. Association of Dyslipidemia with Diabetic Retinopathy in Type 2 Diabetes Mellitus Patients: A Hospital-Based Study. Journal of Pharmacy & Bioallied Sciences. 2021 Nov;13(Suppl 2):S1062.

29. ling J, Xie ZL, Luo X, Chen xingLin, Gao Y, zhuang jiayuan. The Association between High-Density Lipoprotein Cholesterol and Diabetic Retinopathy : A Cross-Sectional Study in patients with type 2 diabetic mellitus [Internet]. Research Square; 2025 [cited 2025 Jan 19]. Available from: https://www.researchsquare.com/article/rs-5733668/v1

30. Qureshi HJ, Masud S, Ul-Qamar M, Aslam MS, Gill M, Fazal A. Original Articles ASSOCIATION OF SERUM C- REACTIVE PROTEIN (CRP), CYSTATIN C AND HOMOCYSTEINE WITH DIABETIC RETINOPATHY IN PATIENTS OF TYPE 2 DIABETES MELLITUS. Journal of Akhtar Saeed Medical & Dental College. 2020;2(1):24–9.

31. Yang N, Lu YF, Yang X, Jiang K, Sang AM, Wu HQ. Association between cystatin C and diabetic retinopathy among type 2 diabetic patients in China: a Meta-analysis. Int J Ophthalmol. 2021 Sep 18;14(9):1430–40.