

Correlation of Plasma RBP4-to-Vitamin A Ratio with Severity of Diabetic Retinopathy

Hadi Rostamkhani^{1,2} , Arezo Karimimoghaddam¹ , Jalal Hejazi^{1,3} ,
Hossein Chiti¹ , Ali Awsat Mellati^{1,2*} 

1. Zanjan Metabolic Diseases Research Center, Zanjan University of Medical Sciences, Zanjan, Iran
2. Dept. of Clinical Biochemistry, School of Medicine, Zanjan University of Medical Sciences, Zanjan, Iran
3. Dept. of Nutrition, School of Medicine, Zanjan University of Medical Sciences, Zanjan, Iran

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Corresponding Information:

Ali Awsat Mellati,

Dept. of Clinical Biochemistry, School of Medicine, Zanjan University of Medical Sciences, Zanjan, Iran

E-Mail: mellati@zums.ac.ir

ABSTRACT

Background & Objective: Diabetic retinopathy (DR) is a common microvascular complication of type 2 diabetes mellitus (T2DM) and the leading cause of vision loss in working-age adults. Vitamin A (retinol) has a role in the mechanism of vision process and retinol binding protein-4 (RBP4), is a carrier of vitamin A, and as an adipokine may be associated with increased risk for insulin resistance and DR. This case-control study was aimed to determine and analyze plasma RBP4-to-vitamin A ratio in relation with terms of DR severity.

Materials & Methods: In the present analytical cross-sectional study, 51 T2DM patients, aged 48-73 years old, were enrolled from those attending to the Ophthalmology Center of Vali-e Asr Hospital, Zanjan, Iran. Patients were categorized as non-retinopathy diabetic patients (NRDP) without any eye problem, those with mild non-proliferative DR (mild NPDR) (n=12), those with severe non-proliferative diabetic retinopathy (severe NPDR) (n=12), and those with proliferative DR (PDR) (n=12); a control group (n=15) was also considered. Anthropometric parameters, BMI, and WHR were determined and blood sample were taken from each participant after overnight fasting (12-14h) to measure their biochemical parameters. Serum RBP4 and vitamin A levels were measured via ELISA and C18 reverse-phase HPLC methods, respectively.

Results: Plasma RBP4 concentration was significantly higher in three different stages of DR than that of the control group suffering from diabetes (77.0 ± 11.0 , 81.7 ± 10.9 and 88.3 ± 11.9 vs. 71.4 ± 12.3 , respectively; $P=0.004$). The ratio of plasma RBP4-to-retinol in DR groups was found to be significantly higher than that in the control group suffering from diabetes (0.21 ± 0.06 , 0.27 ± 0.12 and 0.28 ± 0.07 vs. 0.16 ± 0.14 , respectively; $P=0.001$).

Conclusion: Higher plasma RBP4-to-vitamin A ratio was related to DR severity. Further experimental studies with larger scales are recommended.

Keywords: Diabetic retinopathy, RBP4, RBP4 delivery kinetic, Vitamin A



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Introduction

Chronic hyperglycemia which is the main feature of diabetes mellitus is associated with microvascular (due to small vessels) and macrovascular (due to large vessels) complications. These vascular complications are responsible for impaired quality of life, morbidity, and premature mortality, which occur in the patients with diabetes mellitus (1). Diabetic retinopathy (DR) is one of the severe microvascular complications of diabetic mellitus, which is the leading cause of vision loss in middle-aged and elderly adults worldwide (2, 3). About 15% of patients have some degrees of diabetic retinopathy when their diabetes is diagnosed

for the first time, whereas most of those suffering from diabetes will develop retinopathy after 20 years (4).

Marofizadeh *et al.* found out that the prevalence of DR was 43.1% among the Iranian patients suffering from diabetes mellitus (5). Therefore, new diagnostic tools and efficient strategies are necessary to detect DR in the early stages before reaching the advanced stages.

DR is clinically divided into non-proliferative diabetic retinopathy (NPDR) and proliferative diabetic retinopathy (PDR) stages on the basis of abnormal blood vessel growth (neovascularization) and presence

of visible ophthalmological changes (3,6,7). NPDR is the initial stage of DR, which is characterized by microaneurysms and dot blot hemorrhages (8). The NPDR can be classified into mild, moderate, and severe, depending on the mentioned changes. PDR is the advanced stage of DR, which is characterized by neovascularization and growth of abnormal vessels (4, 9). Principal risk factors for developing DR are hypertension, glycemic control, and diabetic duration (10); also, risk rises with worsening glycemic control. Various biochemical pathways and circulating biomarkers have been proposed which underlie the etiology of DR (3, 8, 11).

Retinol binding protein-4 (RBP4), 21 kDa plasma protein, is a specific carrier for vitamin A (retinol) in the blood. The liver is clearly the dominant expression level of RBP4; but, other tissues also synthesize this protein. However, an important secondary source is adipose tissue which expresses RBP4 as an adipokine (adipose-derived cytokine) (12, 13). RBP4 circulates in a combination with transthyretin (TTR) and, thus, prevents its glomerular filtration (14). It is taken up by peripheral tissue cells that have an RBP4-specific receptor, STRA6, which is the RBP4-specific membrane receptor and vitamin A (retinol) channel, and conducts vitamin A from RBP4 to the tissue cells including eyes (15). The concomitant of triple (GLUT4, RBP4, and STRA6) is probably implicated in the pathogenesis of type 2 diabetes mellitus (T2DM). In this axis, down-regulation of GLUT4 leads to the elevated levels of RBP4 and this increases in RBP4 expression causes insulin resistance through STRA6 (16). Circulating RBP4 is increased in obese individuals, which is associated with impaired glucose tolerance and insulin resistance (17, 18). It has been suggested that obesity is associated with significantly higher apo-RBP4 (RBP4 without vitamin A), but normal holo-RBP4 (RBP4 attached vitamin A) (19), and that the RBP4-to-retinol ratio is elevated in the patients with T2DM (20).

The role of RBP4 in DR is controversial. Few studies (10, 21, 22) have suggested that RBP4 may be involved in the process of DR. In one study (23), RBP4 was reported to be significantly elevated in the patients with PDR than those with NPDR. Akbay (24) suggested that serum RBP4 levels in the patients who had DR was similar to those in the patients who did not.

Vitamin A (all-trans-retinol) with its two bioactive isoforms, retinal and retinoic acid, constitute the groups of retinoid. Retinal, a precursor of rhodopsin, is an essential part of retina visual cycle and retinoic acid and, via binding nuclear receptors, regulates the transcription of more than 300 genes and is a major regulator of growth and development (25, 26). Vitamin A (retinol) protects against hazards of hyperglycemia, which occur in patients with diabetes and initiate cell proliferation in retinopathy (27).

RBP4 is responsible for the distribution of vitamin A to various tissues, especially retina. In previous studies

that have explained the association between serum RBP4 levels and DR, the relation of serum vitamin A levels with severity of DR has not been considered and the role of RBP4 in the pathogenesis of DR is not completely clear. Therefore, this study aimed to evaluate the association of serum RBP4 and vitamin A levels, on one hand, and severity of DR, on the other hand, in comparison with the patients without diabetic non-retinopathy.

Materials and Methods

Participants

This analytical cross-sectional study was performed on 51 patients with T2DM (17 males and 34 females) referring to Ophthalmology Center of Vali-e Asr Hospital, Zanjan, Iran. First, complete eye examinations was conducted on each case by a retina specialist and, accordingly, the patients with T2DM were classified into four groups: mild NPDR (mild non-proliferative diabetic retinopathy) (n = 12); severe NPDR (severe non-proliferative diabetic retinopathy) (n = 12); PDR (patients with proliferative diabetic retinopathy) (n = 12); and NRDP (non-retinopathy T2DM as the control group) (n=15). DR and its stage were diagnosed and certified by an eye specialist. Ethical approval was obtained from Research Committee of Zanjan University of Medical Sciences, Zanjan, Iran (ZUMS.REC.1395.259). Search related to the current study was conducted in accordance with the ethical principles stated in "Declaration of Helsinki". Conventional dietary intake was assessed with the use of a 148-item semi-quantitative food frequency questionnaire (FFQ). The validity and reliability of the FFQ were described in detail another study (Mirmiran *et al.*, 2010). The FFQ contains a list of Iranian food and drink items with standard portion sizes commonly consumed by Iranians. All the participants were asked to report their frequency of intake of a given serving of each item during the previous year. Then, consumption of vitamin A-containing food items was compared in different studied groups in order to control vitamin A intake as a confounding variable (28). The mean age of the participants was 60 years old. Inclusion criteria were as follows: having mean age of 60 (50–70) years old and having body mass index (BMI) <35 kg/m². The exclusion criteria were having nephropathy, heart disease or thyroid dysfunction, inflammatory diseases, cancers, and history of stroke, smoking and consuming dietary supplements.

Anthropometric and Biochemical Measurements

Anthropometric parameters such as height, weight, as well as waist and hip circumference were measured by WHO approved methods (29). For BMI evaluation, each participant, in light clothes and no shoes, was weighed (in kilogram, using a digital scale, to nearest 0.1 Kg) and measured for height (in meters, using a stadiometer, to the nearest 0.5 cm). BMI was calculated as weight in kg divided by height in m squared (kg/m²).

Waist and hip circumferences were measured at the halfway between the lower rib to the iliac crest and bumps, respectively.

Blood samples of all the participants were taken after overnight fasting at 7–10 a.m. Serum was separated and frozen at -80°C until the analysis time. Fasting blood glucose, C-reactive protein, and lipid profiles (cholesterol, triglyceride, HDL, and LDL) were measured by enzymatic methods (Bionic Co. Kit; Tehran, Iran) and Hitachi biochemical autoanalyzer.

Serum RBP4 Levels

Serum RBP4 levels were determined by enzyme-linked immunosorbent assay (ELISA) using a commercial kit (Abcam, ab108897-Retinolbinding protein4 (RBP4) Human ELISA kit) with the sensitivity of 3.4 ng/mL, and the inter- and intra-assay variabilities were 3.1% and 5.4%, respectively.

Serum Vitamin A Levels

Serum vitamin A levels were determined by a C18 reverse-phase HPLC/uv isocratic system (Knauer, Germany) with 250 mm×4.6 mm column and 3µm particle size, using a procedure previously described (30) along with some modifications. All the used chemicals, methanol (cat. no. 1060072500), ethanol (cat. no. 1117272500), acetonitrile (cat. no. 1000302500), and distilled water (cat. no. 773218), were purchased from Merck. All-trans-retinol (cat. no. R7632) as external standard, retinyl acetate (cat. no. 46958) as internal standard, and hexane (cat. no. 110543) were obtained from Sigma-Aldrich. All the chemicals were HPLC purified grade. Stock solutions of retinol (20mg/L; 105 µmol/L) and retinyl acetate (20 mg/L; 70 µmol/L) were prepared in ethanol. All the stock solutions were stored at -20°C (maximum of three months). Work solutions of vitamin A (retinol) and internal standard were prepared on the daily basis. Briefly, serum was isolated from fasting blood samples by refrigerator centrifuge (3500g) for 10 min. Serum solution was allocated in a number of microtubes, covered with aluminum foil, and filled by N₂ gas to minimize light exposure and vitamin A oxidation. The internal standard (retinyl acetate) solution (100 µL), 150 µL of methanol, and 200 µL of ethanol were pipetted into a well-capped polypropylene tube; 200 µL of serum was added and the content was mixed on a vortex for 10 s. Then, 500 µL of hexane was added and the solution was vortexed for 60 s. Each sample solution was centrifuged (5000 RPM, 5 min, 4°C) and supernatants were transferred to other microtubes. Repeatedly, the samples were centrifuged (5000 RPM) for 5 min and each isolated supernatant was added to the microtube of isolated supernatant of the previous step. The combined supernatant was evaporated to dryness under nitrogen gas at 45-50°C. Then, the dried hexane phase was resuspended in 200 of methanol and vortexed for 10 s. Afterwards, 100 µL of the vortexed solution was injected into the HPLC machine, using a reverse-phase 3 µm column (250 mm [Length] and 4.6

mm [inner diameter]), at the isocratic flow rate of 0.8 mL/min; the pressure was 53 bar, and the column's temperature was 30°C. The serum vitamin A (retinol) levels were detected by using an (all-trans-retinol)-retinyl acetate standard curve at 325 nm (30). To ensure reproducible retention on the column, the column was washed with methanol (1 mL/min for 60 min at 45°C) prior to loading the next sample.

Statistical Analysis

Statistical tests were performed by SPSS version 20 (SPSS Inc., Chicago Ill., USA). All the data were reported as mean ± SD. ANOVA and LSD post-hoc tests were run to compare differences between the groups. Kolmogorov-Smirnov test was performed to check for the normal distribution of the variables. Spearman and Pearson's correlation coefficients were employed to investigate the relationship between different parameters. The levels of significance was set at P-value<0.05.

Results

In total, 51 patients (17 men, 34 women) aged 48-73 years old were enrolled in the present study. Among those patients, 36(70%) were diagnosed with DR. Their basal characteristics as well as anthropometric and biochemical data after stratification into four groups are listed in Table 1. The mean age had no significant difference among all the groups. The levels of anthropometric indices (BMI and WHR) were similar in those groups. The plasma FBG levels were similar at high levels in all the groups, which indicated the groups were suffering from diabetes. As shown in Table 1, patients with severe NPDR had higher levels of TG and TC than the control group ($P=0.059$ and $P=0.047$, respectively). No significance difference was found in plasma HDL-c in four groups.

The data suggested that mean plasma RBP4 concentration was significantly higher in three different grades of DR than the control group (77.0 ± 11.0 , 81.7 ± 10.9 and 88.3 ± 11.9 vs. 71.4 ± 12.3 , respectively; $P=0.004$) (Table 1). For each unit (1 ng/mL) increase in the plasma level of RBP4, the adjusted risk of PDR would be increased by 18% (with the odd ratio of 1.184 (CI 95% (1.018 – 1.378) $P=0.028$).

The plasma vitamin A levels were significantly decreased in DR subgroups compared with the control group suffering from diabetes. The ratio of plasma RBP4 to retinol in DR groups was found to be significantly higher than that in the control groups (0.21 ± 0.06 , 0.27 ± 0.12 and 0.28 ± 0.07 vs. 0.16 ± 0.14 , respectively; $P=0.001$) (Table 1, Figure 1). Post-hoc analysis indicated that, regarding RBP4, there was only a significant difference between control and PDR groups

($P=0.003$) and, in the case of vitamin A levels, there was significant differences between the control group and

severe retinopathy and PDR groups ($P=0.03$ and $P=0.016$, respectively) (Table 1).

Table 1. Baseline characteristics and biochemical parameters of DR groups and control (NRDP) patients

Variables	Controls (n=15)	Mild NPDR (n=12)	Severe NPDR (n=12)	PDR (n=12)	P-value*
Age (year)	58.5±9.6	62.3±10.2	61.1±9.8	56.7±5.8	0.41
Weight (kg)	70.5±11.8	70.1±8.8	69.4± 8.7	68.0 ±10.9	0.93
BMI (kg/m ²)	25.1±0.7	28.7±0.8	27.76±0.56	26.56±0.64	0.32
WHR	1.0±0.99	1.0±0.98	0.99±0.84	1.0±0.93	0.52
FBG (mg/dL)	148±7.12	138.3±8.9	193.1±13.4	180.2±12.1	0.187
Tg (mg/dL)	138.0±56.0	162.2±51.2	207.2±94.0	137±75.0	0.059
TC (mg/dL)	164.8±29.0	174.2±32.6	205.3±56.0	164.3±40.0	0.047
LDL-c (mg/dL)	96.5±29.0	87.3±22.1	122±40.5	85±32.6	0.024
HDL-c (mg/dL)	41.3±14.0	36.2±8.7	35.5±9.5	34.3±7.7	0.32
CRP (mg/dL)	3.06±2.2	2.5±1.8	3.1±2	3.4±1.9	0.73
†Vitamin A (µg/dL)	44.7±9.1	39.1±12	33.7±9.9	32.7±7.8	0.01
†RBP4 (mg/mL)	71.4±12.3	77.0±11.0	81.7±10.9	88.3±11.9	0.004
RBP4/vitamin A ratio	0.16±0.04	0.21±0.06	0.27±0.12	0.28±0.07	0.002
SBP (mmHg)	12.3±0.81	12.3±1.15	12.8±0.87	13.1±1.2	0.17
DBP (mmHg)	8.3±0.87	8.0±0.76	8.3±0.76	8.1±0.83	0.77
Duration of Disease	10.0±6.37	13.1±6.3	15.4±7.25	14.9±7.2	0.168

DR, Diabetic retinopathy; NRDP, Non-retinopathy diabetic patients; Data are expressed as mean; BMI, Body mass index; WHR, Wist-to-hip ratio; FBS, Fast blood glucose; TG, Triglyceride; TC, Total cholesterol; LDL-C, Low density lipoprotein-cholesterol; HDL-C, High density lipoprotein- cholesterol; CRP, C-reactive protein; RBP4, Retinol binding protein4; SBP, Systolic blood pressure; DBP, Diastolic blood pressure; *, $P<0.05$ denotes statistically significant difference; † denotes post-hoc analysis which indicates significant difference of RBP4 between control and PDR groups ($P=0.03$) and significant differences of vitamin A between control, severe NPDR, and PDR groups ($P=0.03$ and 0.016 , respectively).

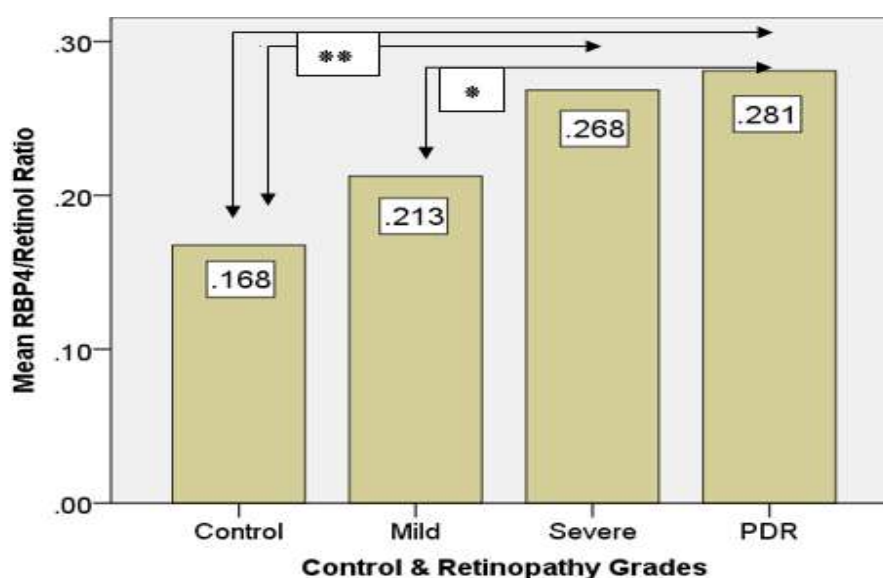


Figure 1. Ratio of plasma RBP4-to-retinol versus control and three groups of diabetic retinopathy (DR). Control, Non-retinopathy diabetic patients; Mild, Mild non-proliferative diabetic retinopathy (NPDR); Severe, Severe non-proliferative diabetic retinopathy; PDR, Proliferative diabetic retinopathy. Plasma RBP4-to-retinol ratio in DR groups is significantly higher than that of the control group (*, $P<0.05$; **, $P<0.01$).

Discussion

Several studies have reported the link between RBP4 and diabetic retinopathy. However, most of these studies have not considered the status of vitamin A (retinol) levels, which is needed to maintain visual process, and the change of which may lead to ocular complications such as DR.

In the present study, the association of RBP4-to-vitamin A ratio with different grades or severity of DR was investigated in comparison with the patients with NRDM as the control group. The results indicated significant elevation in the plasma levels of TG and TC in the patients with different grades of DR compared with the control group, which was similar to that found in previous studies (3, 21). These investigations have shown that severity of DR is positively correlated with increasing triglyceride and plasma triglyceride level and so with plasma HDL-c level in T2DM patients. There was no correlation between the plasma RBP4 levels and biochemical and anthropometric parameters, which is similar to the results obtained by Akbay *et al.* (24).

To the best of our knowledge, there is no study for assessing RBP4-to-vitamin A ratio in the various stages of DR. Our results indicated significant elevation of RBP4 level in serum of DR patient groups compared with NRDP control group. Similar observations have been made by some studies and they have suggested increased plasma RBP4 levels has a causative role in retinal degeneration and vision loss, which may lead to DR. Hence, they have demonstrated elevated RBP4 levels induce inflammation in human retinal endothelial cells through JNK and P38 MAPK signaling (10, 23, 31). However, plasma vitamin A levels were negatively related to severity of DR, the levels of which decreased in the patients with PDR and NPDR compared with NRDP (control group). Hence, our results represented RBP4-to-vitamin A ratio was significantly increased in correlation with severity of DR compared with the control group (Figure 1). It is clear that vitamin A is needed to maintain the visual process. Decreased level of vitamin A is associated with an adverse profile of oxidative stress, which has a role in the pathogenesis of DR, as suggested by some authors (32, 33). Therefore, at lower serum levels of vitamin A, the risk of DR and its severity could increase.

In a case-control study on 287 patients suffering from diabetes, Li (22) described that elevated levels of RBP4 in plasma were associated with DR in Chinese patients with T2DM and suggested that lowering RBP4 could be a new strategy for treating T2DM with DR. This was supported by some other research in different

populations (21, 23). Takebayashi *et al.* (23) found significantly elevated plasma RBP4 levels in the patients with PDR versus NPDR. In contrast, in another study (24), negative conclusion was made about the relationship of RBP4 and DR and it was explained that no effect of serum RBP4 was observed on DR or cardiovascular complications in T2DM. In an experimental study on transgenic mice, Du *et al.* (31) described that over-expressing RBP4 developed progressive retinal degeneration by a retinoid-independent mechanism. They suggested that elevation of RBP4 levels could be a risk factor for retinal damage and vision loss in patients with and without diabetes, regardless of vitamin A. Similarly, two other works (34, 35) have suggested retinol-free RBP4 (apo-RBP4) is a potent as retinol-bound RBP4 (holo-RBP4) in terms of inducing endothelial inflammation during microvascular complications of diabetes. However, Osman *et al.* (32) conducted a case-control study (on 25 healthy persons and 60 patients with DR) and concluded that the deficiency of vitamin A could affect the process of retinal cell proliferation.

Circulating RBP4 is an adipokine and is also secreted by the liver which transports vitamin A. RBP4 has been implicated in the pathogenesis of complications associated with insulin resistance such as DR. What proportion of plasma RBP4 is secreted from liver, and what proportion from adipose tissue, is yet under discussion. Fedders *et al.* (36) used adeno-associated viruses (AAV) that comprise a liver-specific promoter to drive the expression of murine RBP4 in the livers of adult mice to test whether RBP4 impairs glucose homeostasis. Their findings indicated that liver-secreted RBP4 did not affect glucose homeostasis. However, these researchers suggested that plasma RBP4 was exclusively derived from hepatices. Nevertheless, several other studies (37, 38) have indicated that RBP4 which is derived from adipose tissue acts as an adipokine and contributes in metabolic syndrome mechanisms.

Totally, the main finding of the present study showed RBP4-to-vitamin A ratio increased in correlation with the severity of DR. This indicated increased plasma RBP4 levels and low level of plasma vitamin A in DR compared with NRDP. For this result, two hypotheses can be proposed; firstly, plasma RBP4 released from adipose tissue probably has high affinity binding and slower vitamin A delivery kinetics than RBP4 secreted from the liver. Secondly, plasma RBP4 released from the adipose tissue has lower affinity for STRA6 than the liver RBP4. STRA6 mediates the translocation of vitamin A from plasma RBP4 to the intracellular

acceptor in peripheral tissues. Proving these hypotheses needs further verification by large-scale studies and more accurate assay methods which could determine the proportion of RBP4 secreted from the liver and adipose tissue, and RBP4 delivery kinetics in diabetic complications. The major limitation of this study was the sample size of the participants, which was small, and the strength was that it was the first time the plasma RBP4-to-vitamin A ratio was examined in relation to DR severity. No reports could be found to assess RBP-to-vitamin A ratio in the various stages of DR.

Conclusion

In conclusion, according to the data of the present study, RBP4-to-vitamin A ratio is positively correlated with DR severity, which indicated elevated plasma RBP4 levels and decreased plasma vitamin A levels in the process of DR. Since vitamin A is required to maintain visual process and decreased levels of vitamin A is associated with an adverse profile of oxidative stress, elevated RBP4 may have a role in the pathogenesis of DR complications. Therefore, regarding the importance of RBP4 delivery kinetic mechanisms in the process of DR, further experimental studies with larger scales and more accurate methodological issues are required.

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Conflict of Interest

Authors declared no conflict of interest.

References

1. Fowler MJ. Microvascular and macrovascular complications of diabetes. *Clin Diabet.* 2008;26(2):77-82. [DOI:10.2337/diaclin.26.2.77]
2. Cheung N, Mitchell P, Wong TY. Diabetic retinopathy. *Lancet (London, England).* 2010;376(9735):124-36. [DOI:10.1016/S0140-6736(09)62124-3]
3. Ting DSW, Cheung GCM, Wong TY. Diabetic retinopathy: global prevalence, major risk factors, screening practices and public health challenges: a review. *Clin Exp Ophthalmol.* 2016;44(4):260-77. [DOI:10.1111/ceo.12696]
4. Rodríguez ML, Pérez S, Mena-Mollá S, Desco MC, Ortega ÁL. Oxidative stress and microvascular alterations in diabetic retinopathy: Future Therapies. *Oxidat Med Cell Long.* 2019; 2019:1-18 [DOI:10.1155/2019/4940825]
5. Maroufizadeh S, Almasi-Hashiani A, Hosseini M, Sepidarkish M, Samani RO. Prevalence of diabetic retinopathy in Iran: a systematic review and Meta-analysis. *Int J of Ophthalmol.* 2017;10(5):782-802.
6. Mohamed Q, Gillies MC, Wong TY. Management of diabetic retinopathy: a systematic review. *JAMA.* 2007;298(8):902-16. [DOI:10.1001/jama.298.8.902]
7. Santiago AR, Boia R, Aires ID, Ambrósio AF, Fernandes R. Sweet stress: coping with vascular dysfunction in diabetic retinopathy. *Front Physiol.* 2018;9 (16):820-841. [DOI:10.3389/fphys.2018.00820]
8. Mitsch C, Fehre K, Prager S, et al. Clinical decision support for the classification of diabetic retinopathy: A comparison of manual and automated results. *Stud Health Technol Inform.* 2016;223:17-24.
9. Fong DS, Aiello L, Gardner TW, et al. Diabetic retinopathy. *Diabetes Care.* 2003;26(suppl 1):99-102. [DOI:10.2337/diacare.26.2007.S99]
10. Simó-Servat O, Simó R, Hernández C. Circulating biomarkers of diabetic retinopathy: an overview based on physiopathology. *J Diabet Res.* 2016;320-33. [DOI:10.1155/2016/5263798]
11. Mahajan N, Arora P, Sandhir R. Perturbed biochemical pathways and associated oxidative stress lead to vascular dysfunctions in diabetic retinopathy. *Oxidat Med Cell Long.* 2019;2019:1- 16 [DOI:10.1155/2019/8458472]
12. Von Eynatten M, Humpert PM. Retinol-binding protein-4 in experimental and clinical metabolic

- disease. *Expert Rev Molec Diag.* 2008;8(3):289-99. [DOI:10.1586/14737159.8.3.289]
13. Christou G, Tselepis A, Kiortsis D. The metabolic role of retinol binding protein 4: an update. *Hormone Metab Res.* 2012;44(01):6-14. [DOI:10.1055/s-0031-1295491]
 14. Pullakhandam R, Palika R, Ghosh S, Reddy GB. Contrasting effects of type 2 and type 1 diabetes on plasma RBP4 levels: the significance of transthyretin. *IUBMB Life.* 2012;64(12):975-82.
 15. Kelly M, Widjaja-Adhi MAK, Palczewski G, von Lintig J. Transport of vitamin A across blood-tissue barriers is facilitated by STRA6. *FASEB J.* 2016;30(8):2985-95. [DOI:10.1096/fj.201600446R]
 16. Nair AK, Sugunan D, Kumar H, Anilkumar G. Case-control analysis of SNPs in GLUT4, RBP4 and STRA6: Association of SNPs in STRA6 with type 2 diabetes in a South Indian population. *PLoS One.* 2010; 5(7): e11444. [DOI:10.1371/journal.pone.0011444]
 17. Graham TE, Yang Q, Blüher M, et al. Retinol-binding protein 4 and insulin resistance in lean, obese, and diabetic subjects. *New Eng J Med.* 2006;354(24):2552-63. [DOI:10.1056/NEJMoa054862]
 18. Kwanbunjan K, Panprathip P, Phosat C, et al. Association of retinol binding protein 4 and transthyretin with triglyceride levels and insulin resistance in rural thais with high type 2 diabetes risk. *BMC Endocrine Disorder.* 2018;18(1):26-35. [DOI:10.1186/s12902-018-0254-2]
 19. Mills JP, Furr HC, Tanumihardjo SA. Retinol to retinol-binding protein (RBP) is low in obese adults due to elevated apo-RBP. *Experiment Biol Med.* 2008;233(10):1255-61. [DOI:10.3181/0803-RM-94]
 20. Erikstrup C, Mortensen OH, Nielsen A, et al. RBP-to-retinol ratio, but not total RBP, is elevated in patients with type 2 diabetes. *Diabetes, Obesit Metab.* 2009;11(3):204-12. [DOI:10.1111/j.1463-1326.2008.00901.x]
 21. Li Z, Lu X, Liu J, Chen L. Serum retinol-binding protein 4 levels in patients with diabetic retinopathy. *J Int Med Res.* 2010;38(1):95-9. [DOI:10.1177/147323001003800111]
 22. Li JY, Chen XX, Lu XH, Zhang C-B, Shi Q-P, Feng L. Elevated RBP4 plasma levels were associated with diabetic retinopathy in type 2 diabetes. *Biosci Rep.* 2018;38(5): BSR20181100 [DOI:10.1042/BSR20181100]
 23. Takebayashi K, Suetsugu M, Wakabayashi S, Aso Y, Inukai T. Retinol binding protein-4 levels and clinical features of type 2 diabetes patients. *J Clin Endocrinol Metab.* 2007;92(7):2712-9. [DOI:10.1210/jc.2006-1249]
 24. Akbay E, Muslu N, Nayır E, Ozhan O, Kiykim A. Serum retinol binding protein 4 level is related with renal functions in Type 2 diabetes. *J Endocrinol Invest.* 2010;33(10):725-9. [DOI:10.1007/BF03346678]
 25. Daruwalla A, Choi EH, Palczewski K, Kiser PD. Structural biology of 11-cis-retinaldehyde production in the classical visual cycle. *Biochem J.* 2018;475(20):3171-88. [DOI:10.1042/BCJ20180193]
 26. Williams AL, Bohnsack BL. What's retinoic acid got to do with it? Retinoic acid regulation of the neural crest in craniofacial and ocular development. *Genesis.* 2019;57(7- 8):e23308. [DOI:10.1002/dvg.23308]
 27. Augustin A, Dick H, Koch F, Schmidt-Erfurth U. Correlation of blood-glucose control with oxidative metabolites in plasma and vitreous body of diabetic patients. *Eur J Ophthal.* 2002;12(2):94-101. [DOI:10.1177/112067210201200204]
 28. Mirmiran P, Hosseini Esfahani F, Mehrabi Y, et al. Reliability and relative validity of an FFQ for nutrients in the Tehran Lipid and Glucose Study. *Public Health Nutr.* 2010; 13, 654-62. [DOI:10.1017/S1368980009991698]
 29. Jelliffe D. The assessment of the nutritional status of the community. 1966;51:67-67.
 30. Kim Y-K, Quadro L. Reverse-phase high-performance liquid chromatography (HPLC) analysis of retinol and retinyl esters in mouse serum and tissues. *Retinoids: Springer;* 2010;651(5): 263-75. [DOI:10.1007/978-1-60327-325-1_15]
 31. Du M, Otalora L, Martin AA, et al. Transgenic mice overexpressing serum retinol-binding protein develop progressive retinal degeneration through a retinoid-independent mechanism. *Molec Cell Biol.* 2015;35(16):2771-89. [DOI:10.1128/MCB.00181-15]
 32. Osman ZM, Gomaa AM, Hussein HM, Soliman FH, El-Shobaki AF. Association between retinal metabolism and diabetic retinopathy. *Polish J Food Nutr Sci.* 2004;13(4):391-6.
 33. Rostamkhani H, Mellati AA, Tabaei BS, Alavi M, Mousavi SN. Association of serum zinc and vitamin A levels with severity of retinopathy in type 2 diabetic patients: a Cross-Sectional Study. *Biol Trace Element Res.* 2019;192(2):123-8. [DOI:10.1007/s12011-019-01664-z]
 34. Farjo KM, Farjo RA, Halsey S, Moiseyev G, Ma J-X. Retinol-binding protein 4 induces inflammation in human endothelial cells by an

- NADPH oxidase-and nuclear factor kappa B-dependent and retinol-independent mechanism. *Molec Cell Biol.* 2012;32(24):5103-15. [DOI:10.1128/MCB.00820-12]
35. Norseen J, Hosooka T, Hammarstedt A, et al. Retinol-binding protein 4 inhibits insulin signaling in adipocytes by inducing proinflammatory cytokines in macrophages through a c-Jun N-terminal kinase-and toll-like receptor 4-dependent and retinol-independent mechanism. *Molec Cell Biol.* 2012;32(10):2010-9. [DOI:10.1128/MCB.06193-11]
36. Fedders R, Muenzner M, Schupp M. Retinol binding protein 4 and its membrane receptors: a metabolic perspective. *Hormone molecular biology and clinical investigation.* 2015;22(1):27-37. [DOI:10.1515/hmbci-2015-0013]
37. Chang X, Yan H, Bian H, et al. Serum retinol binding protein 4 is associated with visceral fat in human with nonalcoholic fatty liver disease without known diabetes: a cross-sectional study. *Lipid Health Dis.* 2015;14(1):28. [DOI:10.1186/s12944-015-0033-2]
38. Karamfilova V, Gateva A, Alexiev A, et al. The association between retinol-binding protein 4 and prediabetes in obese patients with nonalcoholic fatty liver disease. *Arch Physiol Biochem.* 2019:1-6. [DOI:10.1080/13813455.2019.1673429]

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