Evaluation of the Systemic Pro-Inflammatory Response Associated with Diabetic Retinopathy as a Limiting Factor on the Efficacy of Intravitreal Treatment for Diabetic Macular Edema

Avaliação da Resposta Pró-Inflamatória Sistêmica Associada à Retinopatia Diabética como Fator Limitante da Eficácia do Tratamento Intravítreo para Edema Macular Diabético

Pedro S. Brito[1,2] , Jorge Correia-Pinto[2,3] , Rufino M. Silva[4,5,6]

[1] Opthalmology Department, Hospital de Braga, Braga, Portugal
[2] Life and Health Sciences Research Institute (ICVS), School of Medicine, University of Minho, Campus Gualtar, Braga, Portugal
[3] ICVS/3B's - PT Government Associate Laboratory, Braga/Guimarães, Portugal
[4] Department of Ophthalmology. Centro Hospitalar e Universitario de Coimbra (CHUC), Coimbra, Portugal
[5] Coimbra Institute for Clinical and Biomedical Research. Faculty of Medicine.University of Coimbra (iCBR- FMUC, Coimbra, Portugal)
[6] Association for Innovation and Biomedical Research on Light and Image (AIBILI), Coimbra, Portugal

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ABSTRACT

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RESUMO

PALAVRAS-CHAVE: Edema Macular; Fator A de Crescimento do Endotélio Vascular; Retinopatia Diabética

Diabetic retinopathy (DR) is estimated to occur in about 34% of all diabetic patients,[1] and represents a significant cause of global visual impairment due to the occurrence of complications such as diabetic macular edema (DME), vitreous hemorrhage or tractional retinal detachment. The pathogenesis of DME and DR is complex and still not fully understood. However, we do know that sustained hyperglycemia is the major trigger[2] and it can induce a series of biochemical reactions which lead to accumulating advanced glycation endproducts, activation of protein kinase C, increased oxidative stress and chronic inflammation.[3] In fact, several cytokines and growth factors are known to be increased in DR. Some of the most important include intercellular adhesion molecule 1 (ICAM1)[4] and monocyte chemoattractant protein 1 (MCP1),[5] both implicated in the occurrence of leukostasis, a hallmark feature of DR. Also of major relevance, is vascular endothelial growth factor A (VEGF-A) due to its potent pro-angiogenic and increased vascular permeability actions.[6] In fact, several clinical trials documented significant improvements in visual acuity (VA) and macular thickness with intravitreal injections of anti-VEGF agents: bevacizumab,[7] ranibizumab[8] or aflibercept.[9] Despite such clinical breakthrough, there are still challenges in optimizing DME treatment. In fact, a significant percentage of patients will have suboptimal outcomes even with prolonged monthly injections.[8,9] Additionally, the treatment frequency seen in randomized trials is not readily replicated in day-to-day clinical practice, meaning that real-world results are substantially inferior.[10]

Considering the growing evidence that patients with DR have increased systemic[11] or ocular[12] levels of pro-inflammatory factors, it is possible that different biochemical profiles are associated with different response patterns to anti-VEGF treatment. Therefore, we devised a research project intending to analyze the possible effect of metabolic and pro-inflammatory factors, known to be associated with the pathogenesis of diabetes, on the clinical response to anti-VEGF treatment for DME. Our first publication, reported the results in 30 patients with nonproliferative DR and DME. All cases underwent a laboratory work-up including indicators of cardiovascular risk, renal function, lipid profile and glucose control. Additionally, the serum fraction of peripheral blood samples was isolated, in order to measure the levels of 4 pro-inflammatory factors (VEGF-A, MCP-1, ICAM-1 and tumor necrosis factor-α). Treatment was initiated with intravitreal bevacizumab. To our knowledge, we first reported that patients with a limited macular response (less than 10% decrease in central retinal thickness (CRT)) at the 3rd month of follow-up, had significantly higher levels of high-sensitivity C-reactive protein (hsCRP) (p=0.007) as well as ICAM-1 (p=0.012). On the other hand, cases with a CRT decrease of at least 25% had significantly lower levels of MCP-1 (p=0.015)[13] (Table 1). Interestingly, cases achieving macular edema resolution (CRT < 330 μm) had significantly lower levels of hsCRP (p=0.021) and MCP1 (p=0.004). On a subsequent publication[14] we reported that patients achieving resolution of DME with 6 or less injections of anti-VEGF, had significantly lower values of creatinine (p = 0.003) and hsCRP (p=0.016). On the other hand, cases with persistent DME at the 6th month of follow-up, with a decrease in CRT of less than 20%, were found to have significantly higher baseline hsCRP (p=0.020), higher creatinine (p=0.016) and lower VEGF-A (p=0.018), comparing with cases achieving a CRT decrease of ≥20% (Table 2). Interestingly, hsCRP was the analytic variable found to have the most significant correlation with total number of intravitreal injections (p=0.002, r2=0.200).

Finally, in a more overall analysis, considering 1-year of follow-up and 67 cases,[15] we verified that 41.8% of cases could be classified as early responders (≥20% decrease in CRT at the third month), however 22.4% of cases were classified as poor responders (persistent DME with < 20% CRT decrease) at the 12th month of follow-up. Early responders were more likely to have severe nonproliferative DR (p=0.009), while poor responders had significantly lower baseline serum VEGF-A (54.96±22.52 pg/mL) comparing with early (95.57±53.37 pg/mL) or late responders (114.34±76.29 pg/mL) (p=0.009). Also interesting, cases progressing from bevacizumab to aflibercept anti-VEGF switch, and then on to combination treatment with triamcinolone, were found to have significantly higher values of hsCRP (p=0.002) and MCP-1 (p=0.033) comparing with cases maintained on anti-VEGF monotherapy. Additionally, increasing baseline hsCRP level was again found to be predictive of a low (<10%) CRT response at the 6th month, and such association was independent of any other clinical or analytical variables (p=0.004, OR=2.46). Regarding the outcome of obtaining acceptable resolution of DME (CRT<330 μm), hsCRP was a significant negative predictor of such outcome (p=0.005, OR=0.270) (Table 3). A similar trend of lower hsCRP in cases with CRT <330 μm was maintained until the 12th month (1.30 ± 1.39 vs 2.21 ± 1.70, p=0.005), but no longer significant in the regression model.

Regarding VA outcomes, baseline VA was the most significant variable associated with 12th month visual acuity ($p < 0.001$) as well as with the probability of achieving ≥2 lines of VA improvement (p=0.009, OR = 20.54). Interestingly, patients improving 2 lines of VA were found to have significantly lower ICAM-1 values (722.72 ± 183.12 vs 910.09 ± 345.05, p=0.025). Additionally, increasing age (p=0.042, OR=0.95), increasing macular volume (p=0.002, OR=0.56) as well as more severe DR (p= 0.015, OR = 0.28) diminished the likelihood of achieving VA ≥ 20/40.

Overall, our research demonstrated that increased hsCRP was consistently associated with parameters indicating a limited macular response under anti-VEGF treatment. Additionally, lower serum VEGF was found in long-term poor anatomic responders. We could postulate that there is a molecular basis for different phenotypes of DME. In fact, we identified a predominantly ischemic form of DME characterized by severe DR, high serum VEGF-A and low levels of pro-inflammatory factors (hsCRP, ICAM-1, MCP-1), such patients exhibit an early and significant anatomic response to anti-VEGF treatment. On the other end of the spectrum, there is a predominantly inflammatory DME, characterized by increased pro-inflammatory factors (hsCRP, ICAM-1, MCP-1) and lower circulating VEGF-A. Such patients may present with less severe NPDR, but will have a limited anatomic response to anti-VEGF monotherapy. It is possible that in this phenotype, blood-retinal barrier breakdown (BRB) is not primarily mediated by increased VEGF, but instead, more dependent on increased leukostasis and inflammatory damage to the neurovascular unit.

In conclusion, there is a significant association between

increased systemic biomarkers of inflammation and limited response to anti-VEGF for DME, identifying a DME phenotype in which inflammatory dysfunction of the BRB is preponderant over VEGF-induced vasopermeability. In such cases anti-VEGF monotherapy does not provide optimal results, therefore prompt consideration of alternative treatment regimens is recommended to improve clinical outcomes.

Table 1 Systemic associations with 3rd month macular response

	<10% CFT change		*P* value	≥25% CFT change		*P* value
	Yes (n=11)	No (n=19)		Yes (n=12)	No (n=18)	
hsCRP (mg/L)	3.33±2.01	1.39±1.15	0.007	1.55±1.31	2.25±1.88	0.368
sICAM1 (pg/mL)	975.54±265.49	727.07±336.09	0.012	687.08±154.83	877.97±397.48	0.172
MCP1 (pg/mL)	347.71±124.22	261.66±52.32	0.094	242.42±48.96	315.93±93.14	0.015
VEGF-A (pg/mL)	60.66±32.68	77.97±35.34	0.188	82.44±37.61	66.62±32.39	0.281
HbA1c %	8.51±1.05	7.79±1.19	0.910	8.02±1.00	7.92±1.13	0.921

Table 2 Comparison of serum factors according to whether triamcinolone combination treatment was initiated due to persistent DME

Serum factors	Combination treatment		*P* value
	Yes (n=25)	No (n=33)	
HbA1c (%)	7.85±1.14	7.71±1.13	0.685
Glucose (pg/mL)	173.00±46.65	161.00±63.03	0.486
Total Chol. (mg/dL)	186.50±40.51	163.95±34.13	0.052
LDL Chol. (mg/dL)	106.24±34.60	90.07±29.76	0.105
HDL Chol. (mg/dL)	50.90±18.21	52.93±21.23	0.732
Creatinine (mg/dL)	0.94±0.21	0.76±0.27	0.016
B.U.N (mg/dL)	49.88±18.52	43.15±20.96	0.361
Homocysteine	14.94±4.69	13.35±5.19	0.080
hsCRP (mg/L)	2.84±2.25	1.19±0.94	0.020
ICAM1 (pg/mL)	841.15±353.64	770.20±255.13	0.540
MCP1 (pg/mL)	301.53±96.82	253.52±61.09	0.057
VEGF-A (pg/mL)	62.03±28.52	97.81±33.74	0.018

Table 3 Association of serum factors and 6th month macular outcomes

	<10% CFT change		*P* value	CFT < 330 μm		*P* value
	Yes (n=13)	No (n=54)		Yes (n=24)	No (n=43)	
hsCRP (mg/L)	3.39±1.84	1.49±1.38	<0.001	1.08±0.76	2.26±1.83	0.007
sICAM1 (pg/mL)	924.61±411.13	739.23±264.12	0.219	705.18±241.93	842.64±351.28	0.291
MCP1 (pg/mL)	288.54±66.11	263.62±91.12	0.515	263.75±79.58	291.75±88.61	0.263
VEGF-A (pg/mL)	53.63±28.77	79.23±33.99	0.164	80.59±41.53	64.44±28.20	0.556

ETHICAL DISCLOSURES

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Corresponding Author/
Autor Correspondente:

Pedro S. Brito
Hospital de Braga
R. das Comunidades Lusíadas 133
4710-243 Braga
pbritomd@hotmail.com

ORCID: 0000-0001-7760-2702