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HbA1c and Serum Level of VEGF in Diabetic Retinopathy Patients

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Abstract

Objective: This study aims to determine whether HbA1c level has a correlation with vascular endothelial growth factor (VEGF) serum level in diabetic retinopathy (DR) patients.

Methods: This is an analytical cross-sectional study of patients with DR due to type 2 diabetes, with a purposive sampling technique. Patients were grouped into non-proliferative DR (NPDR) and proliferative DR (PDR). HbA1c and VEGF serum levels were assessed by taking the patient's venous blood.

Results: A total of 82 samples were included, in which the mean HbA1c levels were 8.17% ± 1.91% and the median VEGF levels were 85.78 ng/L (range 38.23-149.43 ng/L). A total of 23 out of 35 NPDR patients were female (65.7%), while 29 out of 47 PDR patients were male (61.7%). Approximately 61.7% of PDR patients had a DM duration of more than 10 years, while 62.9% of NPDR patients had DM duration of less than 10 years. There was an increase in the mean of HbA1c levels in the PDR group compared to NPDR, although it was not statistically significant (p = 0.214), and there was no difference in the median VEGF levels of the two groups. Spearman's correlation analysis revealed no correlation between HbA1 and VEGF levels in diabetic retinopathy, in both the NPDR and PDR groups (correlation coefficient 0.183 and -0.022 respectively).

Conclusion: No statistically significant correlation was found between HbA1c and VEGF serum levels in diabetic retinopathy patients. In this study poor glycemic control were not proven for their implications for VEGF progression. VEGF serum levels may not be used as a marker of DR severity.

Keywords: HbA1c levels; VEGF serum levels; diabetic retinopathy; HbA1c-VEGF correlation.

Introduction

Retinopathy is a microvascular complication due to chronic hyperglycemia. This complication occurs in about 42-60% of patients with type 2 diabetes, and 99% of patients with type 1 diabetes.[1-3] Retinopathy has been known to cause blindness in productive age (20-64 years old).[4] The prevalence of type 2 diabetes mellitus (DM) in Indonesia is around 2.1% (5.5 million population)[5]while the prevalence of diabetic retinopathy (DR) were 43.1%.[2,5,6]

Vascular endothelial growth factor (VEGF) is a protein that plays a role in the angiogenesis process that occurs in DM and DR as a response to hypoxia and hyperglycemia. VEGF can stimulate endothelial

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cells to degrade the extracellular matrix in the process of forming new blood vessels. VEGF plays an important role in the early stages of retinopathy, and intraocular VEGF can be an important marker.[7-9]

HbA1c levels, which represent glycemic control in 3 months, have been associated with complications of DM. According to The Diabetes Control and Complications Trial Research Group, a reduction in HbA1c from 9% to 7% over 6.5 years can reduce the risk of retinopathy by 76%.[10]

This study was conducted to determine the correlation between HbA1c and VEGF serum levels in DR patients, in both non-proliferative diabetic retinopathy (NPDR) and proliferative diabetic retinopathy (PDR).

Methods

This research protocol has been approved by the Research Ethics Commission Faculty of Medicine, Udayana University. A total of 82 DR patients who came to the retina eye clinic of the Bali Provincial Government Eye Hospital were sampled. The study was conducted from August 2020 to January 2021. The inclusion criteria in this study were type 2 DM patients who were over 40 years old and had a retina dilated pupil examination to confirm the diagnosis of DR. Diabetic retinopathy in this study was grouped into 2, namely NPDR if microaneurysms were found without neovascularization, and PDR if there was neovascularization in the retina or optic disc. The exclusion criteria in this study were patients who had undergone avastin injection in the last 3 months, and had undergone vitrectomy surgery. Other exclusion criteria were patients on immunosuppressant drugs and patients with a history of malignancy. Subjects who met the inclusion and exclusion criteria then signed the informed consent. Data regarding the duration of DM, history of hypertension, and body mass index were recorded in the research sheet. The patient then drew 3 ml of EDTA tube venous blood for HbA1c examination by Immunoassay-Turbidimetry, and 3 ml of plain tubes for VEGF examination using the Elisa (Bioassay Technology Laboratory) method.

Statistics

All numerical data were tested for normality by Kolmogorov-Smirnov. Numerical data that were not normally distributed were subjected to a non-parametric test using the Mann-Whitney test. Chi-Square test was used for proportions of catagorical data. The relationship between 2 numerical variables was analysed by the Spearman's correlation test.

Results

A total of 82 type 2 DM patients were included in this study. It was found that there were equal numbers of male and female, as many as 41 subjects. The range of age in this study was 43-76 years old, with a median was 56 years old. The characteristics of the Body Mass Index (BMI) in this study were mostly with a normal BMI (48.8%), followed by overweight as much as 37.8%. Most of the research subjects had hypertension as many as 49 people (59.8%). Mostly it was found that the DR patients had experienced type 2 diabetes for ≥ 10 years, which was 51.2%. DR patients in this study were grouped into 2 groups, namely the PDR group and the NPDR group based on the presence or absence of retinal neovascularization after evaluation of the retina in the dilated pupil. The PDR group was found to be more numerous, which were 47 people (57.3%). The mean HbA1c levels were 8.168 ± 1.91%. The median serum VEGF level was 85.779 ng/L with a range of 38,227-149,430 ng/L.

Twenty three out of 35 subjects in the NPDR group were female (65.7%), while the PDR group there were mostly male, as many as 29 (61.7%) from a total of 47 patients. There was a significant difference between these two groups with p = 0.014. In NPDR group was 45-75 years, with a median 60 years. While in the PDR group, it was 43-72 years old, with a median of 55 years. There was a statiscally that PDR patients were younger than the NPDR group (p=0.013).

The Body Mass Index (BMI) in the NPDR and PDR group was mostly normal (48.8% and 51.1% respectively), followed by overweight (40% and 36.2% respectively). There was no statistically significant difference with the 2 groups (p=0.952).

It was found that 24 (68.6%) of NPDR patients and 25 subjects (53.2%) in PDR group had hypertension. In the NPDR group, it was found that most of the patients had type 2 diabetes for less than 10 years, as many as 22 people (62.9%). This is different from the PDR group, where it was found that the majority of subjects had type 2 diabetes mellitus for ≥ 10 years, as many as 29 patients (61.7%). This difference was statistically significant with p = 0.028.

The mean of HbA1c level in the NPDR group was 7.894% with a standard deviation of 1.961, while

in the PDR group it was 8.372% with a standard deviation of 1.869. There was an increase in the mean of HbA1c levels on the progression of DR, but it was not statistically significant (p=0.214).

The relationship between HbA1c and VEGF serum levels in DR patients, in both PDR and NPDR is shown in Table 3. The results showed no correlation between glycemic control and VEGF levels in the DR (p = 0.634), in the PDR group (p = 0.884), and in the NPDR group (p = 0.294).

Discussion

In the PDR group, most patients had been diagnosed with type 2 diabetes for more than 10 years. The duration of DM is associated with the onset of microvascular complications.[11,12] The duration of DM less than 10 years generally has a low prevalence of retinopathy which is mostly non-proliferative.[13]

Most of the DR patients in this study were found to have hypertension (59.8%). This is consistent with research from Lokesh and Shivaswamy, 2018; Yin et al, 2020.[14,15] Hypertension is associated with the incidence of DR, whether it is hypertensive conditions that are not under therapy or who have received therapy, but are not controlled.[16,17] Based on The Guideline for the Prevention, Detection, Evaluation, and Management of High Blood Pressure in Adult, 2017, suggests that a decrease in blood pressure of less than 130/80 mmHg, in DM patients, can reduce complications.[18] Well-controlled hypertension will reduce the risk of progressive retinopathy by 34%.[4]

Diabetic retinopathy patients in this study mostly had normal BMI, in both the PDR and NPDR groups. This study obtained the same results as Awata et al. 2002.[19] Research by Lim et al., 2010 was found BMI as a risk factor for DR, possibly because it involves platelet function, blood viscosity, aldose reductase activity and vasoproliferative parameters such as VEGF.[20,21]

The mean HbA1c levels were higher in the PDR group. This study found an increase in the mean HbA1c level in the PDR group compared to the NPDR group, although it was not statistically significant. Decreasing HbA1c levels will reduce the complications that occur. A reduction in HbA1c levels of less than 6.5% can prevent the incidence and progression of DR. HbA1c level is one of the indicators used to assess glycemic control, and is not affected by daily blood sugar fluctuations because it is bound to hemoglobin. Research conducted by Nakamura et al., 2008 found that HbA1c levels were associated with DR. However, several studies have found different results, that HbA1c levels not reflect the state of DR, and do not have a correlation with the severity of DR.[19,22-24]

Serum VEGF levels in this study were not significantly different in the NPDR and PDR groups. This is similar to studies conducted by Meleth et al. 2005 and Ozturk et al. 2009.[25,26] However, several studies have found serum VEGF levels to be associated with the progression of DR.[27,28] The study of Sarkar et al., distinguished the DR group into mild NPDR, moderate NPDR and PDR, and the mean serum VEGF levels increased with severity. Several studies regarding the differences between vitreous and serum VEGF levels have found that there was an increase in VEGF levels, in the diabetes group compared to normal in both the vitreous and serum.[25,29] In Murugeswari et al's study, vitreous VEGF levels were significantly higher than serum levels, indicating the occurrence of a local inflammatory process in DR.[30]

VEGF, is a proangiogenic glycoprotein, which increases vascular permeability in DR. VEGF is expressed in vivo and in vitro and is induced by hypoxia. VEGF levels in general can be influenced by various factors, such as tissue hypoxia, hyperglycemia, oxidative stress, and the environment. The results of serum VEGF levels will be different from VEGF levels in eye tissues, such as aqueos and vitreous humor. Increased VEGF production in hyperglycaemia occurs even before any clinical features appear on funduscopy.[31] The important role of VEGF in the pathogenesis of diabetic macular edema, makes intravitreal anti-VEGF as the main choice of therapy.[32] Administration of intravitreal anti-VEGF injection therapy has also shown a decrease in serum VEGF levels.[33]

This study found no relationship between HbA1c and VEGF levels. This is consistent with research from Ozturk et al., 2009.[26] This may occur because serum VEGF levels are influenced by many factors such as other inflammatory factors, epigenetic factors and differences in research methodology such as sampling techniques and sample processing. This study did not compare with the type 2 DM group without the complications of DR, and also with the healthy group so that it could not compare the results with the group without retinopathy This study is also

cross-sectional, so it does not describe whether the condition of retinopathy is acute or not, so the effect of serum VEGF may not be significant. Nevertheless, other studies have found a significant correlation between VEGF levels and HbA1c levels so that serum VEGF levels are considered to predict DR because they represent endothelial damage in diabetes.[27,28]

Conclusion

This study found no significant correlation between HbA1c and VEGF serum levels in DR patients. In this study poor glycemic control were not proven for their implications for DR and VEGF progression. The difference in the results obtained with other studies could be due to inflammatory and epigenetic factors, and this may also be due to differences in the research methodology. This can be used as a basis for considering a better research methodology for future study, in terms of looking for the correlation between HbA1c and serum levels VEGF.

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