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Effect of Intravitreal Injection of Ranibizumab on Macular Perfusion by Optical Coherence Topography Angiography

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Abstract

Background: **Diabetic retinopathy (DR), diabetic microangiopathy, is characterized by capillary nonperfusion, which increases the expression of vascular endothelial growth factor (VEGF). Intravitreal ranibizumab injection was approved for the treatment of diabetic macular edema.** ***Objective:*** **Assessment of the efficacy of intravitreal injection of ranibizumab on macular perfusion.** ***Patients and Methods:*** **Quasi-experimental interventional study was undertaken at Suez Canal University hospitals, Ophthalmology Department on patients who were diagnosed as having diabetic macular edema by optical coherence topography. A total of 60 participants were evaluated by taking a complete ophthalmologic history, examination, and investigations by the use of a pre-designed checklist, and intravitreal injection of anti-VEGF was done.** ***Results:*** **Our study showed that the study group has consisted of 71.4% females and 28.3% males. The mean ± SD of the average superficial foveal avascular zone (sFAZ) area was 263.33 ± 61.80 µm2 pre-injection, 208.33 ± 40.41 post-injection with an improvement of 55.0 ± 21.39 (20.22%). The mean ± SD of the average deep foveal avascular zone (dFAZ) area was 1287.83 ± 220.22 µm2 pre-injection, 624.0 ± 65.35 post-injection with an improvement of 663.83 ± 281.0 (49.54%).** ***Conclusion:*** **Intravitreal injection of anti-VEGF was very effective in diabetic macular edema. Analysis of vascular density changes following anti-VEGF treatment for DME using Optical coherence tomography angiography (OCTA) was remarkably effective.**

Keywords: Anti-VEGF, sFAZ, dFAZ, Optical coherence tomography angiography (OCTA)

Introduction

Approximately 347 million people worldwide have diabetes mellitus (DM)[1]. The worldwide prevalence of DM is predicted to grow to 430 million patients by 2030[2]. Diabetic retinopathy (DR), diabetic microangiopathy, is characterized by microaneurysms (MAs), capillary nonperfusion, and ischemia within the retina[3]. It may cause several complications, such as diabetic macular edema (DME) and diabetic macular ischemia (DMI)[4]. In particular, capillary nonperfusion impairs the nutrition of the neuroglial tissues in the retinal parenchyma, and the resultant hypoxia increases the expression of vascular endothelial growth factor (VEGF) which is believed to be a key mediator in the pathogenesis of DME. It promotes angiogenesis and causes a breakdown in the BRB by damaging the tight junctions between ret-

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inal endothelial cells[5]. Intravitreal ranibizumab injection was first approved by the FDA in 2006 for wet age-related macular degeneration. Since then, it has been approved for the treatment of macular edema following retinal vein occlusion and diabetic macular edema. Most recently, it was approved in 2015 for patients with diabetic retinopathy[6]. The introduction of Optical Coherence Tomography Angiography (OCTA) provides qualitative and quantitative data regarding the retina, choroid, and vascular structures; there are promises that the OCTA may play a unique role in predicting, screening, grading, following up, and also guiding the treatment of DR. The OCTA in contrast to FA, is not invasive. The OCTA uniquely can visualize the changes in superficial and deeper layers, separately[7,8]. The OCTA is believed to be more accurate than the FA to demarcate and measure the FAZ; as it is not obscured by leakages, pooling, or staining from fluorescein. Additionally, the OCTA enables us to measure and define FAZ characteristics across the three superficial capillary plexuses (SCP), middle capillary plexuses (MCP), and deep capillary plexuses (DCP) which could not be assessed by means of the FA or SD-OCT[9].

Patients and Methods

Quasi-experimental interventional study was undertaken at Suez Canal University hospital, Ophthalmology Department on patients who were diagnosed as having diabetic macular edema. We excluded patients with type 1 diabetes, history of vitreoretinal surgery, intravitreal injection of any drugs, macular edema not due to diabetes, presence of media opacities such as vitreous hemorrhage or cataract, cases with motion artifacts preventing the accurate analysis of the micro vascularization and patients' refusal of intravitreal injection of anti-VEGF. Enrolled patients were evaluated by taking a complete ophthalmologic history, examination, and investigations using a pre-designed checklist in conjunction with a designed database computerized program for data entry and analysis.

1-Examination

i) Visual acuity assessment, ii) Slit-Lamp biomicroscopic examination (SL-D7 Topcon, Tokyo, Japan), iii) Intra-ocular pressure measurement, iv) Fundus examination: Indirect ophthalmoscope

2-Investigations

Oct angiography: This analysis was performed for 2 layers in both the 3 × 3-mm and 6 × 6-mm scans for each subject. The OCTA enables us to measure and define FAZ characteristics across the three superficial (SCP) and deep (DCP).

Preparation and administration of IVT treatment

We applied a single-use topical anesthetic to the eye. We instilled 5% povidone iodide onto the ocular surface and allow adequate time (3-5 minutes) prior to injection. We marked the scleral injection site using the mm gauge (the entry site of the needle should be 3.5-4.0 mm). We injected the appropriate volume (maximum 0.1 ml) of the therapeutic agent slowly and carefully.

Post-operative follow-up

OCT scanning and OCT angiography was done for all the patients after one month from the injection. We Compared finding pre- and post-injection.

Statistical analysis

Collected data were coded, entered, and analyzed using Microsoft Office Excel

(2007) software. Data were then imported into Statistical Package for the Social Sciences (SPSS) version 20.0 (IBM SPSS Ver. 20.0) and MedCalc version 12.1.3.0 software for (SPSS INC. CHICAGO IL USA) analysis. Baseline characteristics of the study population were presented as frequencies and percentages (%) in qualitative data or mean values and standard deviations (SD) in quantitative data.

Results

Sixty participants (60 eyes) were 71.4% (43) females and 28.3% (17) males. The mean age of the group was 46.95 ± 7.82 years (range: 32- 63), The mean DM duration in the group was 10.48 ± 4.70 years (range: 5 – 20) (Table 1).

Table 1: Demographic data of the study cases (n= 60)

Variables		No.	%
Gender	Male	17	28.3
	Female	43	71.7
Age (years)	Min. – Max.	32.0 – 63.0	
	Mean ± SD.	46.95 ± 7.82	
	Median (IQR)	46.50(40.0 – 52.0)	
Duration (years)	Min. – Max.	5.0 – 20.0	
	Mean ± SD.	10.48 ± 4.70	
	Median (IQR)	10.0(6.0 – 14.50)	

By comparing the foveal avascular zone (superficial) pre-and post-intravitreal injection of Anti VEGF, it was found that the mean ± SD of sFAZ VD was 598.33 ± 126.28 µm pre-injection, 516.67 ± 96.14 post-injection with the improvement of 81.67 ± 35.99 (13.11%). The mean ± SD of sFAZ HD was 623.33 ± 99.94 µm pre-injection, 474.0 ± 69.55 post-injection with an improvement of 149.33 ± 30.67 (23.80%). The mean ± SD of the average sFAZ area was 263.33 ± 61.80 μm^2 pre-injection, 208.33 ± 40.41 post-injection with an improvement of 55.0 ± 21.39 (20.22%). There was statistically significant (Table 2). By comparing the foveal avascular zone (deep) pre and post-intravitreal injection of Anti VEGF, It found that the mean ± SD of dFAZ VD was 1237.0 ± 136.85 µm pre-injection, 1010.83 ± 83.20 post-injection with improvement 226.17 ± 213.52 (16.73%). The mean ± SD of dFAZ HD was 1322.08 ± 188.41 µm pre-injection, 925.0 ± 66.70 post-injection with an improvement of 397.08 ± 247.98 (28.16%). The mean ± SD of the average dFAZ area was 1287.83 ± 220.22 μm^2 pre-injection, 624.0 ± 65.35 post-injection with an improvement of 663.83 ± 281.0 (49.54%). There was statistically significant (Table 3). Table (4) shows improvement in FAZ in deep capillary plexuses more than in superficial capillary plexuses. Improvement in sFAZ VD was 81.67 ±35.99 (13.11%) while in dFAZ was 226.17 ± 213.52 (16.73%), Improvement in sFAZ HD was 149.33 ± 30.67 (23.8%) while in dFAZ was 397.08 ± 247.98 (28.16%) and finally Improvement in sFAZ average was 55.0 ± 21.39 (20.22%) while in dFAZ Was 663.83 ± 281.0 (25.54%).

Discussion

Our study showed that the mean ± SD of sFAZ VD was 598.33 ± 126.28µm pre-injection, and 516.67 ± 96.14 post-injection with an improvement of 81.67 ± 35.99 (13.11%). The mean ± SD of sFAZ HD was 623.33 ± 99.94µm pre-injection, 474.0 ±

69.55 post-injection with an improvement of 149.33 ± 30.67 (23.80%).

Table 2: values of pre and post-VEGF injection according to sFAZ (n= 60)

sFAZ	Pre	Post	Improvement (%)	t	P
VD					
Min. – Max.	486.0 – 766.0	440.0 – 650.0	81.67 ± 35.99 (13.11%)	17.575*	<0.001*
Mean ± SD.	598.33 ± 126.28	516.67 ± 96.14			
HD					
Min. – Max.	530.0 – 750.0	408.0 – 560.0	149.33 ± 30.67 (23.80%)	37.717*	<0.001*
Mean ± SD.	623.33 ± 99.94	474.0 ± 69.55			
Average					
Min. – Max.	220.0 – 350.0	180.0 – 265.0	55.0 ± 21.39 (20.22%)	19.915*	<0.001*
Mean ± SD.	263.33 ± 61.80	208.33 ± 40.41			

*t: Paired t-test, p: p value for comparing between pre and post, *: Statistically significant at p ≤ 0.05*

Table 3: values of pre and post-VEGF injection according to dFAZ (n= 60)

Dfaz	Pre	Post	Improvement (%)	T	P
VD					
Min. – Max.	1122.0 – 1426.0	920.0 – 1100.0	226.17 ± 213.52 (16.73%)	8.205*	<0.001*
Mean ± SD.	1237.0 ± 136.85	1010.83 ± 83.20			
HD					
Min. – Max.	1173.0 – 1585.0	850.0 – 1000.0	397.08 ± 247.98 (28.16%)	12.403*	<0.001*
Mean ± SD.	1322.08 ± 188.41	925.0 ± 66.70			
Average					
Min. – Max.	1110.0 – 1598.0	540.0 – 750.0	663.83 ± 281.0 (49.54%)	18.299*	<0.001*
Mean ± SD.	1287.83 ± 220.22	624.0 ± 65.35			

t: Paired t-test, p: p-value for comparing between pre and post,
**: Statistically significant at p ≤ 0.05*

The mean ± SD of the average sFAZ area was 263.33 ± 61.80 µm2 pre-injection, 208.33 ± 40.41 post-injection with an improvement of 55.0 ± 21.39 (20.22%). Also, we reported that the mean ± SD of dFAZ VD was 1237.0 ± 136.85 µm pre-injection and 1010.83 ± 83.20 post-injection with an improvement of 226.17 ± 213.52 (16.73%). The mean ± SD of dFAZ HD was 1322.08 ± 188.41 µm pre-injection, 925.0 ± 66.70 post-injection with an improvement of 397.08±247.98 (28.16%). The mean ± SD of the average dFAZ area was 1287.83 ± 220.22 µm2 pre-injection, 624.0 ± 65.35 post-injection with an improvement of 663.83 ± 281.0 (49.54%). Many studies had discussed this topic demonstrating conflicting results with some studies showing stable or improved macular perfusion following treatment[10-12], other studies showed worsening of macular perfusion following treatment[13,14], and a study showed conflicting results. Factors that could result in retinal perfusion improvement after treatment with anti-VEGF antibodies include the reversal of leukostasis that is induced by excessive VEGF secretion in diabetics and results in increased capillary occlusion[15], restoration of the normal retinal architecture due to decreased intraretinal edema[16]. Factors that could justify retinal perfusion worsening following VEGF inhibition include inducing vasoconstriction of the retinal vas-

culature which was found following bevacizumab and ranibizumab injections for DME possibly due to nitric oxide inhibition which occurs with VEGF inhibition an also leads to systemic hypertension in case of systemic VEGF inhibition[17].

Table 4: Correlation between sFAZ and dFAZ (improvement)

FAZ	sFAZ Improvement	dFAZ Improvement	R	P
FAZ VD	81.67 ± 35.99 (13.11%)	226.17 ± 213.52 (16.73%)	0.601	<0.001*
FAZ HD	149.33 ± 30.67 (23.80%)	397.08 ± 247.98 (28.16%)	0.835	<0.001*
FAZ average	55.0 ± 21.39 (20.22%)	663.83 ± 281.0 (25.54%)	0.883	<0.001*

*r: Pearson coefficient, *: Statistically significant at p ≤ 0.05*

Inhibition of VEGF by bevacizumab also resulted in a decrease of the mean blood flow velocity of the central retinal, the temporal posterior ciliary, and the ophthalmic artery by about 10%, 20%, and 20%, respectively, 4 weeks after one bevacizumab injection in patients with exudative age-related macular degeneration[18,19].

Conclusion

sFAZ & dFAZ areas significantly enlarge in diabetic patients. Microvascular changes in DCP, such as dFAZ area enlargement, dFAZ perimeter irregularity, and presence of capillary drop-out areas, are associated with worsening of BCVA, significantly more than changes in SCP. Analysis of vascular density changes following anti-VEGF treatment for DME using OCTA could benefit from a unified scanning protocol for those patients. Anti-VEGF treatment was very effective in DME.

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