# Intravitreal Bevacizumab at the End of Diabetic Vitrectomy for Prevention of Postoperative Vitreous Hemorrhage: A Comparative Study

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***Objective:*** *To study the role of intravitreal injection of bevacizumab (Avastin) at the end of vitrectomy for prevention of postoperative vitreous hemorrhage in diabetic eye disease.*
***Material and Method:*** *The authors conducted a retrospective, comparative, nonrandomized study comparing patients with diabetic eye disease who received a 1.25 mg bevacizumab injection at the end of vitrectomy to a group with diabetic eye disease who underwent vitrectomy but did not receive this injection. For statistical analysis, each patient was assigned to one of four groups according to the hemostatic modalities used (group 1, none; group 2, only long acting gas; group 3, only intraoperative intravitreal bevacizumab; group 4, both long acting gas and intraoperative intravitreal bevacizumab). The primary outcome measure was the incidence of early and late postoperative vitreous hemorrhage (POVH). The secondary outcome measure was visual acuity (VA) at 1 and 6 months.*
***Results:*** *The present study included 87 eyes from 78 patients. The 87 eyes were categorized in the four groups described above. The incidence of early postoperative VH was lowest in group 3 (5%), followed by group 1 and group 4 (13%) and group 2 (16%) (p = 0.49). No eyes in group 1 or group 4 had late postoperative VH. Group 3 had the lowest incidence of VA below 20/200 (11%) in the first postoperative month, followed by group 1 (22%), group 4 (33%) and group 2 (50%) (p = 0.44). Group 3 also had the best visual recovery (VA above 20/50) with 38% at the end of six months compared with 13% in group 1, 7% in group 4 and no eyes in group 2 (p = 0.03).*
***Conclusion:*** *Patients with intravitreal injection of 1.25 mg bevacizumab at the end of diabetic vitrectomy had the lowest incidence of early postoperative vitreous hemorrhage with no statistical significance. However, they had significantly the best visual recovery at the end of six months.*

***Keywords:*** *Avastin, Bevacizumab, Diabetes mellitus, Proliferative diabetic retinopathy, Vitrectomy, Vitreous hemorrhage*

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Postoperative vitreous hemorrhage (POVH) is a common problem after diabetic vitrectomy, with an incidence from 29 to 75%[1-3]. The cause is not precisely known. Early POVH may be caused by the remnants of fibrovascular tissue, by the dissolution of a blood clot trapped in the remaining antero-peripheral vitreous gel, or by reactivation of retinal sites that bled intraoperatively. Late POVH may be caused by anterior hyaloidal fibrovascular proliferation or neovascularization originating from the sclerotomy sites[4].

Par Planar Vitrectomy (PPV), the modern surgery in diabetic retinopathy, increases the concentration of angiogenic factors such as vascular endothelial growth factor (VEGF) in response to surgical trauma and inflammation. These angiogenic factors can stimulate complications such as iris neovascularization and fibrovascular proliferation, leading to recurrent POVH[5-10] Bevacizumab (Avastin, Genentech Inc., South San Francisco, California) is a full-length humanized antibody that binds to all subtypes of VEGF and is successfully used in cancer therapy as a systemic drug[11]. Various modalities have been used to prevent POVH after diabetic vitrectomy but the results are generally unsatisfying. Intravitreal injections of bevacizumab (IVB) injected preoperatively have recently been demonstrated to be effective in reducing early POVH. Possible mechanisms include improvement

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in the integrity of retinal blood vessels and regression of neovascularization[10,12]. However, intraoperative bevacizumab injection reportedly cannot prevent rebleeding in eyes undergoing PPV for treatment of diabetic vitreous hemorrhage[13].

The aim of the present retrospective interventional study was to assess and compare the role of bevacizumab in prevention of POVH when injected at the end of surgery for diabetic eye disease.

**Material and Method**

The authors conducted a retrospective chart review from 87 eyes of 79 patients who received IVB injection at the end of diabetic vitrectomy at Maharaj Nakorn Chiang Mai University Hospital from January 2006 to December 2008. The treatment protocol was determined by one retina specialist (NI).

The present study was done in accordance with the Declaration of Helsinki and the present study protocol was approved by the Research Ethics Committee, Faculty of Medicine, Chiang Mai University. All patients were informed of the procedure and informed consent was obtained. Particularly, the off-label use of the drug and its potential risks and benefits were discussed extensively with patients.

The patients were divided into four groups according to the hemostatic modalities used. Group 1 eyes received neither IVB nor long-acting gas tamponade. Group 2 eyes received long-acting gas tamponade only. Group 3 eyes received only IVB at the end of diabetic vitrectomy. Group 4 eyes received both long-acting gas tamponade and IVB at the end of diabetic vitrectomy. The inclusion criteria were: non-clearing vitreous hemorrhage, proliferative diabetic retinopathy (PDR) with documented tractional retinal detachment (TRD) and the availability of clear chart records showing details of postoperative conditions. The exclusion criteria were: history of IVB injection less than 12 months before the surgery, previous ocular surgery (except cataract surgery), eyes with iris neovascularization or neovascular glaucoma, other consequences of TRD, the presence of major intraoperative complications such as suprachoroidal hemorrhage or other situations leading to uncompleted surgery, use of silicone oil for intravitreal tamponade at the end of surgery, and postoperative follow-up of < 1 month or loss of follow-up before the complete absorption of intraocular long-acting gas.

Medical records were reviewed for age, gender, follow-up periods, lens status, preexisting complications of diabetic retinopathy and postoperative complications. Each patient underwent complete preoperative ophthalmic examinations including best-corrected visual acuity (BCVA) using the Snellen chart, slit-lamp biomicroscopy, intraocular pressure (IOP) measurement using applanation tonometry, fundus examination by indirect ophthalmoscopy and B-scan ultrasonography, if indicated. The review occurred during follow-up examinations at postoperative months 1, 3 and 6. For statistical analysis, the severity of pre- and postoperative vitreous hemorrhage (VH) was classified into four grades as follows: none (no vitreous hemorrhage), mild (most of the optic disc and retinal vessels were visible), moderate (optic disc or retinal vessels were barely visible) and severe (VH was too dense to allow visualization of the optic disc). Early POVH was defined as VH occurring within 1 month after the surgery and late POVH was defined as VH occurring between 4 weeks and 6 months after surgery. The long-acting gas comprised 20% sulphurhexafluoride (SF6) and 16% perfluoropropane (C3F8). Snellen visual acuity was converted to the logarithm of the minimum angle of resolution (logMAR). Counting fingers vision was defined as 2.0 logMAR and hand movement vision was defined as 3.0 logMAR[14] BCVAs were classified into three groups for comparison of the distribution of final BCVA: 1) group 1 (good visual acuity) represented visual acuities of 20/50 or better, 2) group 2 (fair visual acuity) represented visual acuities of 20/200 or better, but less than 20/50 and 3) group 3 (poor visual acuity) represented visual acuities less than 20/200.

The surgical procedures were done by a single surgeon (NI). The techniques of PPV were standardized using three port pars plana sclerotomies, removing the vitreous up to the vitreous base, peeling off membranes, removing the posteror vitreous surface, and performing panretinal endolaser photocoagulation of the retina. At the end of the surgical procedure, intravitreal injection of 1.25 mg (0.05 ml) bevacizumab (Avastin, Genentech, South San Francisco, CA) through the closed infertemporal sclerotomy site was done in the bevacizumab groups (groups 3 and 4). No intravitreal injection was done in the non-bevacizumab groups (groups 1 and 2). The postoperative regimen included topical tobramycin eye drops four times per day for 1 week and 1% topical prednisolone acetate eye drops at a dose of four times per day that was usually tapered off over 4 weeks. Patients who had fluid gas exchange were instructed to remain face down for 7 to 10 days.

***Statistical analysis***

Means were used for description of quantitative data, and percentages were used for qualitative data. A Mann-Whitney U test were used to compare the means for parametric and nonparametric numerical data. Differences between groups were determined using a chi-square analysis or Fishers' Exact test for categorical data. Statistical analyses were done using SPSS statistical software (version 13.0; SPSS Inc., Chicago, IL). For all statistical tests, $p < 0.05$ was considered significant.

**Results**

A total of 88 eyes were enrolled in the present study. One was excluded due to a postoperative complication of choroidal and retinal detachment. A total of 87 eyes from 78 patients met the inclusion criteria initially, but only 53 eyes from 48 patients met the inclusion criteria at 6 months. The mean age was 52.2 $\pm$ 10.3 years (range: 23 to 74 years). Thirty-five eyes in 33 patients (19 men, 14 women; mean age 53.7 years) received diabetic vitrectomy without intraoperative IVB. Fifty-two eyes in 48 patients (19 men, 29 women; mean age 51.1 years) received diabetic vitrectomy with intraoperative IVB. Demographics and clinical data of the patients in both groups are summarized in Table 1. There are no statistically significant differences in the demographic data.

Group 1, 2, 3 and 4 included 23, 12, 37 and 15 eyes, respectively. Clinical features and surgical methods used are depicted for each group in Table 2. Eyes in group 3 had the lowest incidence of early POVH (5%), followed by group 1 (13%), group 4 (13%) and group 2 (16%) ($p = 0.49$). Also, there was no difference in the incidence of late POVH between groups ($p = 0.42$).

The preoperative BCVA did not differ between groups ($p = 0.25$). At 1-month postoperative review, there was no different of BCVA between groups ($p = 0.44$). In subgroups analysis, group 3 had the lowest incidence of BCVA below 20/200 (11%), which differed significantly from the 50% in group 2 ($p < 0.01$), but was not statistically significant when compared with the 22% in group 1 ($p = 0.28$) and 33% in group 4 ($p = 0.07$). At 6-month postoperative review, the incidence of BCVA above 20/50 was highest in group 3 (38%), followed by group 1 (13%) and group 4 (7%). No eyes in group 2 reached BCVA 20/50. There was a significant difference between groups ($p = 0.03$). Groups 1, 2 and 4 had a significantly lower incidence of BCVA above 20/50 than group 3 (group 3 versus group 1, $p < 0.01$; group 3 versus group 2, $p < 0.01$; group 3 versus group 4, $p = 0.04$).

The severity of POVH is shown in Fig. 1. Only 2 eyes (5%) with a mild degree of early POVH occurred in group 3 compared with 1 eye (4%) of mild degree and 2 eyes (9%) of moderate degree in group 1, 1 eye (8%) of mild and 1 eye (8%) of severe degree in group 2 and 1 eye (7%) of mild and 1 eye (7%) of moderate degree in group 4 ($p = 0.67$). Also, there was no difference in grading severity of late POVH between groups ($p = 0.50$) (Fig. 2).

**Discussion**

Postoperative vitreous haemorrhage (POVH) after diabetic vitrectomy is a common event, with an occurrence of 29-75% reported in various studies[1-3] The etiology is diverse. Reactivation of fibrovascular tissue remnants at the retinal sites that bled intraoperatively may be causes of early POVH. And anterior hyaloidal fibrovascular proliferation or the sclerotomy sites neovascularization may be the causes of late POVH[4].

Multiple methods have been used for

**Table 1.** Demographic data for patients in the bevacizumab and control groups

| | Non-bevacizumab group | Bevacizumab group | p |
|---|---|---|---|
| Number of patients/eyes | 33/35 | 48/52 | |
| Age $\pm$ SD | 53.7 $\pm$ 11.1 | 51.1 $\pm$ 9.7 | 0.95 |
| Sex (male/female) | 19/14 | 19/29 | 0.09 |
| Lens status | | | |
| Phakic | 33 | 46 | 0.48 |
| Aphakic/pseudophakic | 2 | 5 | |
| Preoperative BCVA logMAR: mean $\pm$ SD | 1.5 $\pm$ 0.8 | 1.5 $\pm$ 0.9 | 0.99 |

BCVA: best corrected visual acuity

**Table 2.** Clinical features and surgical methods, by group

| | Group 1 | Group 2 | Group 3 | Group 4 | p-value |
|---|---|---|---|---|---|
| Number of eyes | 23 | 12 | 37 | 15 | - |
| Preoperative BCVA, n(%) | - | - | - | - | 0.25 |
| < 20/200 | 11 (47) | 9 (75) | 19 (51) | 5 (33) | - |
| 20/200 to 20/50 | 12 (52) | 3 (25) | 16 (43) | 9 (60) | - |
| > 20/50 | 0 (0) | 0 (0) | 2 (5) | 1 (7) | - |
| 1-month postoperative BCVA, n(%) | - | - | - | - | 0.44 |
| < 20/200 | 5 (22) | 6 (50) | 4 (11) | 5 (33) | - |
| 20/200 to 20/50 | 10 (43) | 6 (50) | 21 (57) | 9 (60) | - |
| > 20/50 | 8 (35) | 0 (0) | 12 (32) | 1 (7) | - |
| 6-month postoperative BCVA, n(%) | - | - | - | - | 0.03** |
| < 20/200 | 1 (4) | 2 (17) | 3 (8) | 1 (7) | - |
| 20/200 to 20/50 | 12 (52) | 7 (58) | 4 (11) | 4 (27) | - |
| > 20/50 | 3 (13) | 0 (0) | 14 (38) | 1 (7) | - |
| Simultaneous cataract extraction, n(%) | 9 (39) | 3 (25) | 14 (38) | 3 (20) | 0.88 |
| Long-acting gas tamponade, n(%) | - | - | - | - | 0.13* |
| SF6 | - | 6 (50) | - | 3 (20) | - |
| C3F8 | - | 6 (50) | - | 12 (80) | - |
| Early postoperative VH, n(%) | 3 (13) | 2 (16) | 2 (5) | 2 (13) | 0.49 |
| Late postoperative VH, n(%) | 0 (0) | 1 (8) | 3 (8) | 0 (0) | 0.42 |

* Comparing the difference between group 2 and group 4 eyes
** Statistically significant comparing the difference between each groups
BCVA = best corrected visual acuity; VH = vitreous haemorrhage

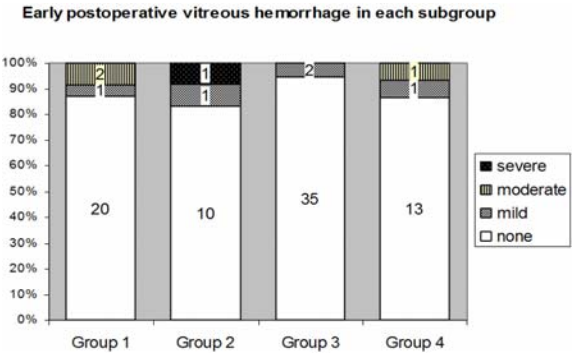


**Fig. 1** Severity of early postoperative vitreous hemorrhage in eyes with different hemostatic modalities. Group 1, none; group 2, only long-acting gas; group 3, intraoperative intravitreal bevacizumab; group 4, both long-acting gas and intraoperative intravitreal bevacizumab

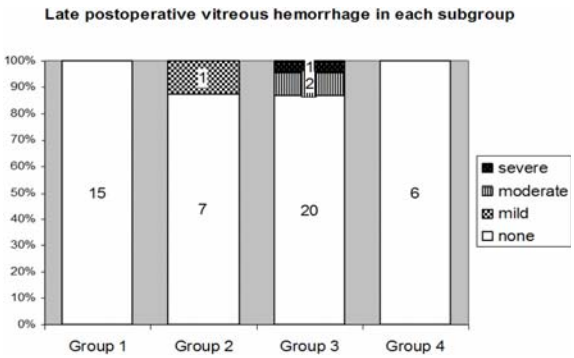


**Fig. 2** Severity of late postoperative vitreous hemorrhage in eyes with different hemostatic modalities. Group 1, none; group 2, only long-acting gas; group 3, intraoperative intravitreal bevacizumab; group 4, both long-acting gas and intraoperative intravitreal bevacizumab

prevention of POVH, but the results are unsatisfactory. De Bustros et al and Koutsandrea et al reported systemic administration of antifibrinolytic medications and intravitreal infusion of short-acting gas[15,16] Yang et al reported that 10% C3F8 is effective in preventing early postoperative VH by restoring the integrity of blood vessels after surgical injury. This technique may take more than 2-3 weeks, as intraocular gas acts as a mechanical tamponade on the fragile vessels during this period[17].

VEGF is the major angiogenic factor in proliferative diabetic retinopathy. It is well established that intraocular surgery in diabetic patients, including PPV, results in a rising concentration of angiogenic

factors such as VEGF in response to surgical trauma and inflammation[5-9]. A VEGF surge acts as a stimulant for iris neovascularization or fibrovascular proliferation that may lead to VH[7]. Bevacizumab is the full-length humanized antibody that binds to all subtypes of VEGF. Many studies have been proposed regarding the biological effect of intravitreal bevacizumab (IVB) in diabetic retinopathy[18-21]. One possible mechanism is regression of neovascularization by improving the integrity of retinal blood vessels and hence reducing the risk of postoperative bleeding from the fragile retinal vessels, while another is related to vessel caliber[13].

In the present study, eyes in group 3 had a 5% incidence of early POVH, which is the lowest compared with 13%, 16% and 13% in group 1, 2 and 4, respectively. This finding agrees well with that found by Cheema et al[10]. However, the authors found no statistically significant difference in the incidence of early POVH between groups ($p = 0.49$). In a randomized controlled trial, Ahn et al reported that the adjunctive use of IVB did not significantly reduce POVH incidence; intraoperative injection of IVB only showed a reduction of early POVH incidence, most of which was mild[22]. The severity of early POVH in present study has the similar result, with a 5% incidence of mild VH as can be expected in mild visual deterioration. The early POVH clears up much faster and has relatively small consequences for final visual outcome. Yang et al reported that 10% C3F8 is effective in preventing early POVH[17]. Koutsandrea et al used a prospective randomized study to show that SF6 did not have a hemostatic effect on POVH[15]. However, the incidence of early POVH after use of a long-acting gas for hemostasis was not different between groups.

There was no difference in the incidence of late POVH between groups ($p = 0.42$). This is probably due to the relatively short half-life of intravitreal bevacizumab, especially in vitrectomized eyes, in which the normal half-life is 4.32 days[23].

The postoperative BCVA improved in all groups, especially at the 6-month review. BCVA is significantly improved in group 3, when using bevacizumab injection at the end of diabetic vitrectomy. The authors propose that these improvements could be due to the progressive resolution of macular edema by dual actions of the vitrectomy and bevacizumab injection in improving the integrity of the retinal blood vessels. These results are difficult to compare with others, as studies evaluating visual acuity for longer than 6 months after IVB have rarely been done.

The Kaplan-Meier analysis showed a reduction in the rate of POVH in the 4 to 12 week period after diabetic vitrectomy for those patients receiving intravitreal bevacizumab (Fig. 3). This treatment is beneficial for patients, as it facilitates early visual rehabilitation and allows the ophthalmologist to detect fundus lesions and plan further interventions sooner after vitrectomy.

The present study reports outcomes of intraoperative intravitreal bevacizumab injection after diabetic vitrectomy, a topic which has not been extensively investigated previously. The limitations of the present study are its non-randomized retrospective design, the large number of dropout patients during follow-up until the end of six months and the use of the Snellen chart as a marker for visual acuity. However, the study has a large number of eyes with a consecutive nature of data, long term follow-up and a single surgeon. In an attempt to minimize selection bias, the present study excluded patients with silicone oil injections, as use of silicone oil is related to the severity of disease.

In conclusion, POVH after diabetic vitrectomy remains a major cause of morbidity, but advances in modern vitrectomy and adjunctive treatments seem to have an encouraging effect on its incidence. In the present series, the use of intraoperative bevacizumab seems to be safe and effective. However, future randomized prospective studies with large sample sizes

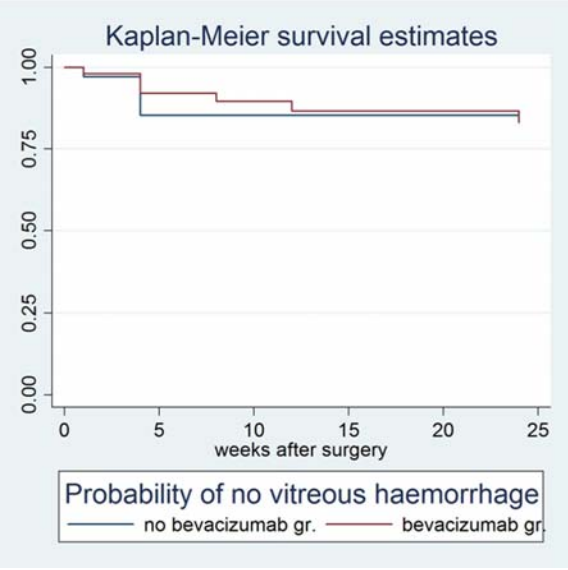


**Fig. 3** Kaplan-Meier estimate showing the influence of intraoperative intravitreal injection of bevacizumab on probability of no POVH

are necessary to fully evaluate the effectiveness of intraoperative bevacizumab on POVH and visual recovery as well as its long-term safety.

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**Potential conflicts of interest**

None.

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## *การศึกษาเปรียบเทียบผลของการฉีดยา Bevacizumab เข้าช่องหลังลูกตา ในขณะสิ้นสุดการผ่าตัด diabetic vitrectomy เพื่อป้องกันภาวะเลือดออกในวุ้นตาภายหลังผ่าตัด*

***เชิง จิรวิศัลย์, นิมิตร อิทธิพันธุ์กุล***

***วัตถุประสงค์****: เพื่อศึกษาเปรียบเทียบผลของการฉีดยา Bevacizumab เข้าวุ้นตา ในขณะสิ้นสุดการผ่าตัด diabetic vitrectomy เพื่อป้องกันเลือดออกในวุ้นตาภายหลังผ่าตัด*

***วัสดุและวิธีการ*** *: ศึกษาย้อนหลังเพื่อเปรียบเทียบผลของการฉีดยา Bevacizumab 1.25 มิลลิกรัม เข้าวุ้นตาในขณะสิ้นสุดการผ่าตัด diabetic vitrectomy ในกลุ่มที่ผ่าตัดและได้รับยาและกลุ่มที่ไม่ได้รับยา Bevacizumab เพื่อดูผลการป้องกันเลือดออกในวุ้นตาภายหลังผ่าตัด โดยแบ่งผู้ป่วยเป็น 4 กลุ่ม ได้แก่ กลุ่มที่ 1 ได้รับการผ่าตัด diabetic vitrectomy, กลุ่มที่ 2 ได้รับการผ่าตัด diabetic vitrectomy ร่วมกับฉีด Long acting gas เข้าวุ้นตาในขณะสิ้นสุดการผ่าตัด, กลุ่มที่ 3 ได้รับการผ่าตัด diabetic vitrectomy ร่วมกับฉีดยา Bevacizumab เข้าวุ้นตาในขณะสิ้นสุดการผ่าตัด และกลุ่มที่ 4 ได้รับการผ่าตัด diabetic vitrectomy ร่วมกับฉีด Long acting gas และยา Bevacizumab เข้าวุ้นตาในขณะสิ้นสุดการผ่าตัด ศึกษาเปรียบเทียบอุบัติการณ์ของภาวะเลือดออกในวุ้นตาภายหลังผ่าตัด รวมถึงระดับสายตาภายหลังการผ่าตัด(final visual acuity)ที่เวลา 1 และ 6 เดือน*

***ผลการศึกษา*** *: ผู้ป่วยจำนวน 78 คน 87 ตา พบอุบัติการณ์ของภาวะเลือดออกในวุ้นตาภายหลังผ่าตัดที่ 1 เดือนต่ำที่สุดในกลุ่ม 3 (ร้อยละ 5) ตามด้วยกลุ่ม 1 และกลุ่ม 4 (ร้อยละ 13) และกลุ่ม 2 (ร้อยละ 16) (P=0.49) ไม่พบภาวะเลือดออกในวุ้นตาภายหลังผ่าตัดที่ 6 เดือนในกลุ่ม 1 และกลุ่ม 4 พบว่ากลุ่ม 3 มีผู้ป่วยที่ระดับสายตาแย่กว่า 20/200 ภายหลังผ่าตัด 1 เดือนต่ำสุด (ร้อยละ 11) ตามด้วยกลุ่ม 1 (ร้อยละ 22) กลุ่ม 4 (ร้อยละ 32) และกลุ่ม 2 (ร้อยละ 50) (P=0.44) พบระดับสายตาดีกว่า 20/50 ภายหลังผ่าตัด 6 เดือนสูงสุดในกลุ่ม 3 (ร้อยละ 38) ตามด้วยกลุ่ม 1 (ร้อยละ 13) และกลุ่ม 4 (ร้อยละ 7) ซึ่งไม่พบอุบัติการในกลุ่ม 2 (P=0.03)*

***สรุป*** *: ผู้ป่วยที่ได้รับการฉีดยา Bevacizumab เข้าช่องหลังลูกตาในขณะสิ้นสุดการผ่าตัด diabetic vitrectomy พบอุบัติการณ์ของภาวะเลือดออกในวุ้นตาภายหลังผ่าตัดที่ 1 เดือนต่ำที่สุด โดยไม่มีนัยสำคัญทางสถิติ แต่ผู้ป่วยมีระดับสายตาภายหลังผ่าตัดที่ระยะเวลา 6 เดือนดีขึ้นอย่างมีนัยสำคัญ*