An Updated Study of the Use of Bevacizumab in the Treatment of Patients with Prethreshold Retinopathy of Prematurity in Taiwan

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• PURPOSE: **To investigate the effectiveness and complications associated with the use of bevacizumab, an anti–vascular endothelial growth factor agent, in the treatment of prethreshold retinopathy of prematurity (ROP).**

• DESIGN: **A multicenter, retrospective case series.**

• METHODS: **Data from patients who had received intravitreal bevacizumab (IVB) injections for the treatment of ROP were collected from 4 medical centers in Taiwan. The main outcome measures were the regression of ROP and the complications that were associated with the IVB injections.**

• RESULTS: **In total, 162 eyes from 85 patients were included in the study. After receiving IVB injections, 143 eyes (88%) exhibited ROP regression. Fourteen eyes (9%) required additional laser treatment for ROP regression after the absence of a positive response to the IVB injections. Three eyes (2%) progressed to stage 4 ROP and required vitrectomies to reattach the retinas. Two eyes (1%) received 1 additional IVB injection to decrease persistent plus disease. All of the eyes (100%) had attached retinas after the various treatments that they received. The major ocular complications that were associated with IVB injections included vitreous or preretinal hemorrhage in 2 eyes (1%); cataract in 1 eye (1%); and exotropia in 1 eye (1%). No notable systemic complications related to the IVB injections were observed.**

• CONCLUSIONS: **IVB injection seems to be an effective and well-tolerated method of treating prethreshold ROP. Laser therapy may still be required as a backup treatment for patients who do not respond to an IVB injection or for those in whom ROP worsens after an IVB injection. (Am J Ophthalmol 2013;155:150–158. © 2013 by Elsevier Inc. All rights reserved.)**

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RETINOPATHY OF PREMATURITY (ROP) REMAINS ONE of the leading causes of childhood blindness. In later stages of ROP, neovascularization of the retina develops as a result of retinal immaturity. Neovascularization in turn leads to retinal traction, retinal detachment, hemorrhage, and the development of a funnel configuration of the retina, which eventually affects visual function. Neovascularization is driven mainly by vascular endothelial growth factor (VEGF).[1] Currently, the recommended treatment for type 1 ROP is laser ablation in the periphery. The timing of treatment has been moved to an earlier stage of the disease, which was documented by the Early Treatment for Retinopathy of Prematurity Study.[2] Although laser treatment effectively prevents stage 3 ROP from progressing to stage 4 ROP in 90% of patients, ablative laser treatments actually destroy approximately two thirds of the retina, and some patients still progress to retinal detachment despite receiving laser or cryotherapy. Moreover, the functional outcomes in stage 4B or stage 5 ROP still are not satisfactory, even after the patient has been treated with a vitrectomy or scleral buckle.[3–5] Therefore, a new treatment that could decrease the need for either laser treatment or vitreoretinal surgery is necessary.

Bevacizumab (Avastin; Genentech Inc, San Francisco, California, USA) is a humanized anti-VEGF monoclonal antibody.[6] It is the first antiangiogenic agent to be approved for the treatment of metastatic colorectal cancer. It also has been shown to produce favorable outcomes in the treatment of many retinopathies that are associated with VEGF upregulation, including age-related macular degeneration,[7,8] diabetic retinopathy,[9–11] vitreous hemorrhage,[12,13] neovascular glaucoma,[14] and retinal vascular occlusion.[15–17] Previous studies have shown that VEGF levels in the vitreous fluid often are highly elevated in ROP patients.[18,19] These results suggest that bevacizumab is a potential candidate for use in the treatment of ROP.

Previously, Mintz-Hittner and associates published the results of a randomized trial of ROP therapies (Bevacizumab Eliminates the Angiogenic Threat of Retinopathy of Prematurity [BEAT-ROP] study) and found that bevacizumab was effective in the treatment of ROP; it was more effective than laser treatment in zone 1 ROP cases in particular.[20] We also published the results of a multicenter study in Taiwan in which 27 patients (49 eyes) were enrolled.[21] Thus far, however, the number of studies on the

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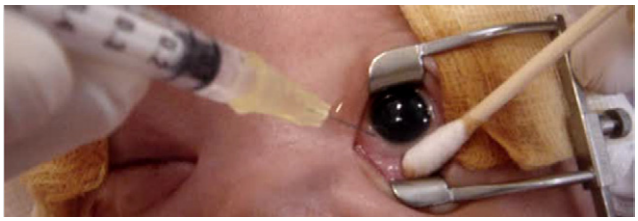

FIGURE 1. Photograph showing intravitreal injection of bevacizumab at the pars plicata for a patient with retinopathy of prematurity. After aseptic procedures, the injection was performed with a 30-gauge needle directed almost perpendicularly to the globe initially and then slightly directed toward the center of the eyeball after the needle passed the lens equator to avoid damaging the lens or retina.

use of bevacizumab in the treatment of ROP remains limited.[22–34] The present study aimed to update the findings of our previous study in Taiwan by including more patients and monitoring those patients for a longer follow-up period. Patients with prethreshold ROP (as defined by criteria that were established in the Early Treatment for Retinopathy of Prematurity Study) who were treated with bevacizumab (patients with stage 4 and stage 5 ROP were excluded) were included in this study. Refraction data, which were collected after the injection of bevacizumab and after the regression of ROP in some patients, were analyzed. Treatment effects, ocular complications, and systemic complications also were analyzed.

METHODS

THIS WAS A MULTICENTER, RETROSPECTIVE STUDY OF THE use of bevacizumab in the treatment of patients with ROP. The data were collected from medical centers in 4 cities across Taiwan: the National Taiwan University Hospital in Taipei, the Changhua Christian Hospital in Changhua, the Chang Gung Memorial Hospital in Kaohsiung, and the Chang Gung Memorial Hospital in Taoyuan. The data were collected between January 2006 and February 2011. Data from patients with prethreshold ROP (ROP that had not yet reached stage 4 or 5) who were treated with an intravitreal bevacizumab (IVB) injection were collected from each center and were pooled for analysis. The indications for treatment were patients whose retinopathy met the criteria that were established by the Early Treatment for Retinopathy of Prematurity Study.[2] Patients whose disease progression and treatment outcomes were monitored for fewer than 6 months were excluded. Numerical variables are presented as means ± standard deviations.

Eyes were prepared in a standard fashion using 5% povidone–iodine and topical antibiotics, after which 0.625 mg (0.025 mL) bevacizumab was injected intravitreally via the pars plicata under intravenous sedation. The injection was performed with a 30-gauge needle that initially was directed along an angle that was perpendicular to the globe and that then was redirected slightly toward the center of the eyeball after the needle had passed the equator of the lens. This technique was used to avoid damaging either the lens or the retina (Figure 1).[35] This technique was different from the technique that we used to perform intravitreal injections in adults. After the injection had been administered, the intraocular pressure and retinal artery perfusion of the injected eye were checked, and patients received topical antibiotics for 7 days. If the patients did not respond positively to the IVB injection or if the ROP worsened, the patients were treated using conventional laser treatment or an additional IVB injection. A positive response included the disappearance of the tunica vasculosa lentis, pupil dilation, the disappearance of or a decrease in retinal vessel tortuosity and neovascularization, and the presence of vessels that continued to vascularize toward the peripheral retina. The worsening of ROP was defined as follows: the reappearance of plus disease, the persistence or reappearance of neovascularization arising from the retinal vessels, and progression to retinal detachment. All of these conditions required retreatment. After treatment, patients were monitored every 1 or 2 weeks until full vascularization of the retina was observed. Full vascularization was defined as follows: vascularization as far as it would develop without an active component or clinically significant tractional elements. All of the screeners and treating physicians were retina specialists. In certain cases, photographs were obtained before and after the injection of bevacizumab using the RetCam Imaging System (Clarity Medical System, Pleasanton, California, USA). In some patients, examinations were performed under anesthesia after the IVB injection. In these examinations, intraocular pressure, corneal size, and axial length were measured, and slit-lamp ophthalmoscopy, indirect ophthalmoscopy, and cycloplegic refraction were performed. The cycloplegic agent that was used was either 1% cyclopentolate or 1% tropicamide.

RESULTS

THE CURRENT STUDY IS AN EXTENSION OF A PRIOR STUDY[21] that enrolled more patients and involved a longer follow-up period. The patients with prethreshold disease (excluding stage 4 and 5 ROP) who had received an IVB injection in the prior study (41 eyes of 23 patients) were pooled together with the 121 eyes of 62 new patients in the current study for the outcome analysis. In total, 162 eyes from 85 patients (57 male and 28 female) were included in the study. Data were collected from 15 patients from the National Taiwan University Hospital in Taipei, 14 patients from the Changhua Christian Hospital in Changhua, 23 patients from the Chang Gung Memorial Hospital in Kaohsiung, and 33 patients from the Chang Gung

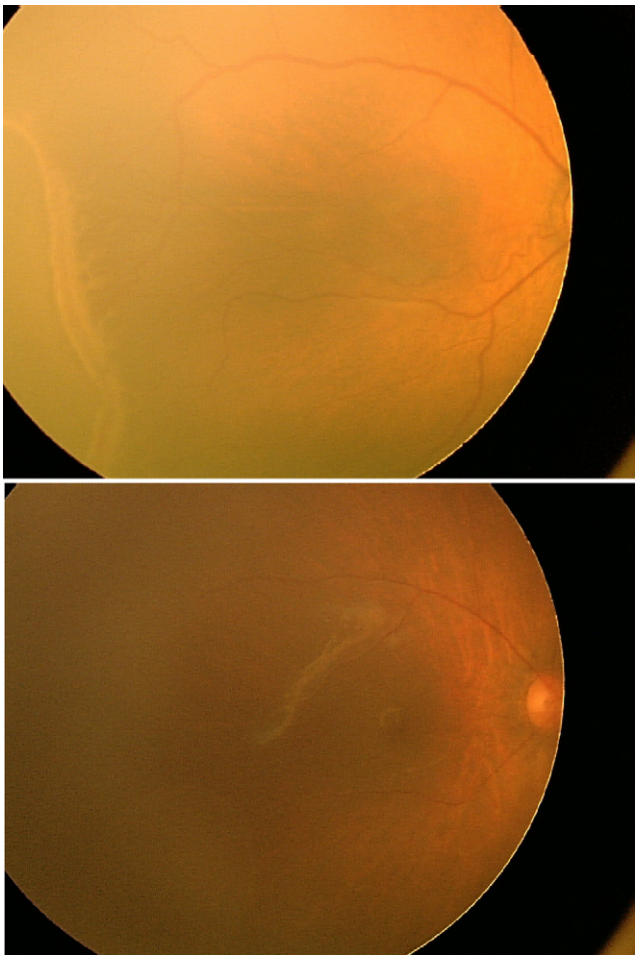

FIGURE 2. Fundus photographs showing stage 3 retinopathy of prematurity (Top) before and (Bottom) after the injection of bevacizumab. (Top) Before the injection of bevacizumab, neovascularization is seen with avascular retina in the periphery. (Bottom) After the injection of bevacizumab, neovascularization regressed and the retinal vascularization continued toward the retinal periphery.

Memorial Hospital in Taoyuan. The mean gestational age and birth weight of the entire cohort were 26.3 ± 2.6 weeks and 930.1 ± 359.3 g, respectively. All of the treated eyes had prethreshold ROP without retinal detachment.

Among the treated eyes, 160 eyes had stage 3 ROP (99%), and 2 eyes (1%) had stage 2 ROP. In addition, 17 eyes (10%) had zone 1 ROP, and 145 eyes (90%) had zone 2 ROP. Among the eyes with stage 2 or 3 ROP, bevacizumab was used as the primary treatment for ROP in 147 eyes (91%), and it was used as a salvage treatment in 15 eyes (9%) after a lack of response to prior laser treatment. Patients who had undergone previous laser treatment had undergone either 1 or 2 treatment sessions (4 eyes [27%] had undergone 2 laser treatment sessions, and 11 [73%] eyes had been treated only once). Among eyes with prethreshold ROP, the mean age at injection was 36.6 ± 3.2weeks postmenstrual age (PMA). One to 3 days after the injection, the tunica vasculosa lentis had diminished, and pupil dilation was noted. Decreased vessel tortuosity was observed in most eyes within 1 week of receiving an IVB injection. The regression of neovascularization took anywhere from several weeks to months. After the regression of neovascularization, the continuation of normal vascularization toward the periphery of the retina could be observed. Of the 162 eyes, the use of bevacizumab in the treatment of ROP resulted in the regression of ROP after a single IVB injection in 143 eyes (88%; Figure 2). Additional laser treatment was required to regress ROP in 14 eyes (9%), in which either no positive response to IVB injection or worsening of the ROP was observed. The additional laser treatment occurred at a mean age of PMA 40 ± 2.8 weeks (range, 36 to 44 weeks PMA). The average time between laser treatment and IVB injection was 6 ± 3.4 weeks. Three eyes (2%) progressed to stage 4 ROP and required vitrectomies to reattach the retinas. The time to vitrectomy was 35, 41, and 69 weeks PMA in these 3 eyes. Two eyes (1%) received 1 additional IVB injection at 39 and 41 weeks PMA to decrease persistent plus disease. If we pooled all of the cases with a worsening of ROP after IVB, the average time between retreatment (including laser, vitrectomy, additional IVB injection) and initial IVB injection was 7.6 ± 9.4 weeks. On average, patients were monitored for 13.7 ± 5.8 months. All of the eyes (100%) had attached retinas at the final follow-up appointments.

Examinations under anesthesia were performed on 28 patients (33%). All of the eyes had normal intraocular pressure, corneal size, and clear media, except one, who had a cataract. Refraction data were available in 53 eyes from 28 patients with regressed ROP after receiving an IVB injection. The mean spherical power was 0.8 ± 2.6 diopters (D; range, −6.3 to 7.3 D), the mean cylindrical power was −2.1 ± 1.1 D (range, −5.3 to −0.3 D), and the mean axial length was 20.7 mm (range, 19.8 to 22.3 mm). The mean spherical equivalent of refractive error among these patients was −0.1 ± 1.8 D (range, −8.75 to 6.55 D). Only 4 eyes (8%) had spherical equivalents that were more severe than −5 D. Most eyes (42/53; 79%) had with-the-rule astigmatism. The mean age at examination was 17.8 ± 7.6 months (range, 8 to 35 months).

Major ocular complications that were associated with IVB injection included vitreous or preretinal hemorrhage, which occurred in 2 eyes (1%); cataract, which occurred in 1 eye (1%); and exotropia, which occurred in 1 eye (1%). Vitreous or preretinal hemorrhage eventually resolved in all of the eyes. The cause of cataract in 1 patient was not related to the trauma caused during the intravitreal injection. That patient underwent cataract surgery at the age of 19 months. No notable systemic complications that were related to the IVB injection were observed during the follow-up period.

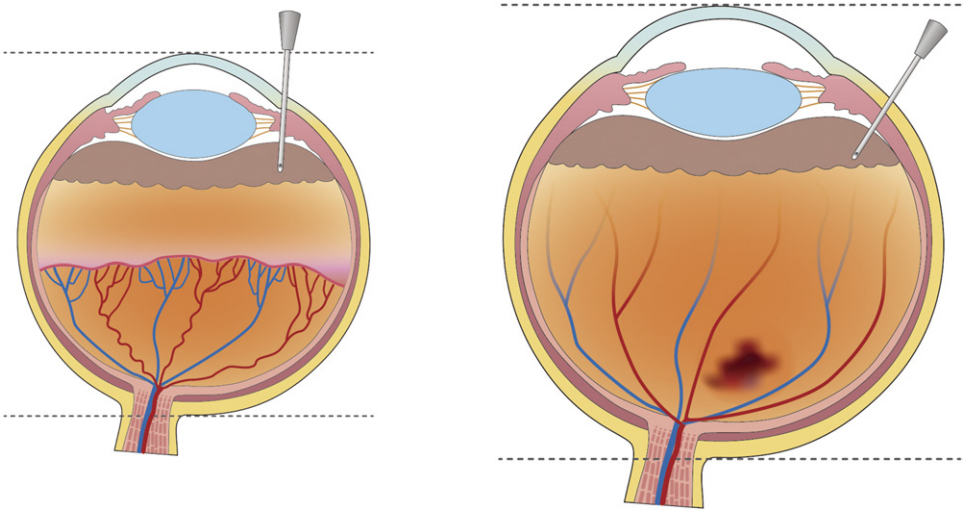

FIGURE 3. Diagrams showing a comparison of the intravitreal injection angle in (Left) newborns and (Right) adults. (Left) For the intravitreal injection of bevacizumab in newborn patients with retinopathy of prematurity, the injection was performed at the pars plicata because the pars plana is not fully developed in newborns. Therefore, the injection angle is almost perpendicular to the globe. (Right) For the intravitreal injection of bevacizumab in adult patients with age-related macular degeneration, the injection was performed at the pars plana, and the injection angle was directed toward the center of the globe. Currently, the injected volume of bevacizumab in infants (0.625 mg [0.025 mL]) is half of the adult dose (1.25 mg [0.05 mL]).

DISCUSSION

IN THIS STUDY, WE FOUND THAT USING AN IVB INJECTION (0.625 mg) as either a primary treatment or salvage treatment after laser therapy resulted in the regression of neovascularization and full vascularization of the retina in 88% of eyes with prethreshold ROP. Nine percent of eyes showed no response to bevacizumab or worsening in the degree of ROP after receiving an IVB injection and required additional laser treatment. In addition, 2% of eyes progressed to stage 4 ROP and required vitrectomies to reattach the retinas; 1% of eyes required a second bevacizumab injection to decrease the plus disease. Ocular complications did occur in some patients (preretinal or vitreous hemorrhage, 1%; cataract, 1%; and exotropia, 1%), but in none of the patients did endophthalmitis or corneal opacities develop that required corneal transplants in the laser treatment arm, as was reported in the BEAT-ROP study.[20] No apparent systemic toxicity that was related to the injection was found between the injection date and the date of the last follow-up examination. These data were consistent with the data in our previous publication.[21] Because VEGF concentrations are highly elevated in advanced ROP and because VEGF has been found to be a driving force for neovascularization,[18,19,36] blocking VEGF with anti-VEGF agents, such as bevacizumab, seems to be a reasonable approach to the treatment of ROP. Increasingly, more patients worldwide have received anti-VEGF treatment for ROP, and therefore, data on the effectiveness and complications of this treatment method are urgently needed.

Mintz-Hittner and Kuffel showed that a single injection of bevacizumab prevented progression to retinal detachment in all eyes with posterior zone 1 ROP, even when laser ablation was not necessary.[23] In their recent randomized trial, the BEAT-ROP study, Mintz-Hittner and associates found that compared with conventional laser therapy, there was a significant benefit of IVB monotherapy in infants with stage 3+ retinopathy for zone 1 disease, but not for patients with zone 2 disease.[20] The development of peripheral retinal vessels continued after treatment with IVB, whereas conventional laser therapy led to permanent destruction of the peripheral retina.[20] The results are encouraging, because approximately 27% to 47% of posterior zone 1 patients progress to retinal detachment even with the application of laser ablation in the peripheral retina.[37–39] In our case series, 17 eyes (10%) with ROP were classified as having zone 1 disease. None of these eyes progressed to retinal detachment after receiving IVB. These results seem to reflect a better outcome after IVB injection as compared with laser treatment in zone 1 ROP patients. Other potential benefits of anti-VEGF therapy relative to ablative treatments include a simpler procedure that requires less time to complete, no need for the special equipment that is required for laser or cryotherapy, a procedure that is less destructive to the retina, the regression of tunica vasculosa lentis and the dilation of pupils (which facilitates subsequent treatments or follow-up examinations easier), and the elimination of various complications that are associated with ablative treatments, such as refractive errors or visual field loss.[40–42]

TABLE 1. Comparison of the Current Study and the Bevacizumab Eliminates the Angiogenic Threat of Retinopathy of Prematurity Study

	Current Study	BEAT-ROP[a]
Country conducted	Taiwan	United States
No. of study sites	4	15
Population type	Asians	Mixed
Total no. of eyes treated with IVB	162	140
Mean BW ± SD (g)	930.1 ± 359.3	615.2 ± 139.5 in zone 1 ROP; 689.2 ± 111.3 in zone 2 ROP
Mean PMA ± SD at IVB injection (wks)	36.6 ± 3.2	NA
No. of eyes with zone 1 ROP (%)	17 (10%)	62 (44%)
Mean ± SD time of recurrence or no response requiring retreatment after IVB injection (wks)	7.6 ± 9.4	16.0 ± 4.6
No. of eyes with retreatment after IVB injection (%)	19 (12%)	6 (4%)
No. of eyes with late recurrence at or after 53 weeks PMA (%)	1 (0.6%)	6 (4%)

BEAT-ROP = Bevacizumab Eliminates the Angiogenic Threat of Retinopathy of Prematurity; BW = birth weight; IVB = intravitreal bevacizumab; NA = not available; PMA = postmenstrual age; ROP = retinopathy of prematurity; SD = standard deviation.

[a]From Mintz-Hittner et al.[20]

However, long-term study is necessary to establish the validity of these advantages.

The injection site, depth, and angle that were used in pediatric eyes are different from those that have been used in adult eyes. These modifications seek to avoid damaging either the lens or the retina (Figure 3).[35] The injected volume that is required for treating infants also is much lower than the appropriate volume for adults. Surgeons need to be very familiar with the various injection techniques before applying this treatment to newborns.[43] Infants need to be monitored closely for potential complications that are associated with intravitreal injection, including cataract, endophthalmitis, and retinal detachment, after receiving an IVB injection.

IVB injection seems to be an effective treatment for prethreshold ROP, including zone 1 ROP. However, the drug is not able to cure ROP in all patients. The BEAT-ROP study observed late recurrent disease in 2 of 62 eyes in infants with zone 1 disease and in 4 of 78 eyes in infants with posterior zone 2 disease after treatment with IVB. Considering all of the eyes with recurrent ROP together, the mean ± standard deviation time to recurrence was 16.0 ± 4.6 weeks in 6 eyes that received an IVB injection, compared with 6.2 ± 5.7 weeks in 32 eyes that received conventional laser therapy. Although more eyes required additional therapies after failing to respond to an IVB injection or because of to recurrence, we did not observe ROP recurrence at the aforementioned 16.0 ± 4.6-week mean follow-up after IVB injection. Only 1 eye progressed to stage 4A ROP at 69 weeks PMA. The difference in late recurrence after IVB injection between the current study and the BEAT-ROP is shown in Table 1. Various hypotheses are proposed to explain the difference between the BEAT-ROP study results and ours. First, we believe that the phenomenon may be related to the early additional laser treatment (performed at mean of 6 ± 3.4 weeks after IVB injection) that was required for patients who did not exhibit an obvious response to bevacizumab treatment. IVB may be able to neutralize VEGF that is already present in the vitreous cavity, but may not be able to inhibit the continual production of VEGF in the avascular area. Thus, a late recurrence is possible if VEGF is expressed continually in the avascular retina after the degradation of bevacizumab and the subsequent loss of its therapeutic effects. If laser therapy also is used in the treatment of patients who are not responsive to IVB, the VEGF level in the eye may be inhibited further by destroying the avascular retina that is responsible for continued VEGF expression. Thus, laser therapy may be used to treat patients who do not benefit clearly from IVB. This therapy in turn may lead to a decrease in the late recurrence of ROP that was observed in the BEAT-ROP study. Second, there was a high proportion of zone 1 ROP eyes in the BEAT-ROP study compared with that found in our study (44% in the BEAT-ROP study vs 10% in the current study). Moreover, the mean birth weight of patients in the BEAT-ROP study is much lower than that observed among our patient cohort (615.2 ± 139.5 g for zone 1 ROP and 689.2 ± 111.3 g for zone 2 ROP in the BEAT-ROP study; 930.1 ± 359.3 g in the current study). If a higher proportion of zone 1 ROP patients were enrolled in the BEAT-ROP study, it is likely that full vascularization of the retina after IVB would have taken a longer time because more avascular retina is present in zone 1 ROP than in zone 2 ROP. Notably, the recurrence of ROP occurs at a later time if there is a delayed rebound in VEGF levels. Third, the BEAT-ROP study is a prospective study that used stricter criteria for retreatment, whereas researchers in our study may have been more aggressive in retreating patients if no prompt positive response was seen after IVB injection. Thus, the current study used retreatments more frequently and earlier for the patients who did not respond or who exhibited recurrences after IVB injection and involved fewer late recurrences compared with the BEAT-ROP study. Finally, there are interracial differences in the observed cases of ROP.[44,45] VEGF is not the only growth

TABLE 2. Comparison of Refractions after Intravitreal Injection of Bevacizumab, Laser, and Cryotherapy for the Treatment of Retinopathy of Prematurity

Authors	Year of Publication	Country	Treatment	Mean SE of Refractive Error (D)	Mean Age at Examination (y)
Khwarg and associates[56]	1995	South Korea	Cryotherapy	−4.1	0.5
White and associates[57]	1997	United States	Laser	−6.6	3
			Cryotherapy	−7.6	3
Laws and associates[58]	1997	United Kingdom	Laser	−0.5	1
			Cryotherapy	−5.3	1
Pearce and associates[59]	1998	United Kingdom	Laser	−3.7	2
			Cryotherapy	−7.9	2
Connolly and associates[60]	1998	United States	Laser	−3.1	5.8
			Cryotherapy	−5.1	5.8
Fallaha and associates[61]	2002	United States	Laser	−4.5	1.7
Al-Ghamdi and associates[62]	2004	Canada	Laser	−1.8	3
			Cryotherapy	−9.2	3
Sahni and associates[63]	2005	United Kingdom	Laser	−2.4	3
			Cryotherapy	−6.5	3
Current study	2012	Taiwan	IVB injection	−0.1	1.5

D = diopters; IVB = intravitreal bevacizumab; SE = spherical equivalent; y = years.

factor with highly elevated concentrations in ROP. Insulin-like growth factor 1 and other growth factors also may play roles in the pathogenesis of ROP.[18,19,36,46–48] The inhibition of VEGF alone by bevacizumab may not be sufficient to regress all of the ROP cases. Because the effect of bevacizumab injection in ROP eyes remains under investigation, standard laser treatment could function as a backup method of treatment for the nonresponsive cases, and a vitrectomy may be required in the event that a patient progresses to the retinal detachment stage.

Although both of our studies and others that used IVB for the treatment of ROP seem to offer promising results for the use of anti-VEGF therapies in the treatment of ROP, recent studies in both animals and human beings have found evidence of systemic bevacizumab exposure after intravitreal injections.[49–52] In addition, Sato and associates found that systemic VEGF levels were depressed for as long as 2 weeks after the administration of either 1 or 0.5 mg IVB in ROP patients.[52] However, no adverse systemic events were observed in these patients after the administration of IVB. Concerns about the safety of using IVB in newborn individuals have been raised because of a lack of supportive data from either large animals or humans.[53] Among the studies in which IVB is used to treat ROP, the BEAT-ROP study is the only prospective randomized study of the use of bevacizumab in the treatment of ROP. That study showed that IVB is an effective treatment for ROP, but did not address the issue of drug safety; such a study would require a large patient population (2800 infants), which is difficult to achieve in a clinical setting. Thus, there are at present no definite conclusions regarding the way in which bevacizumab should be used in the treatment of ROP patients.[54] Although a study of the pathologic features of the eyes of a very low-birth weight infant (350 g) born at 22 weeks gestational age showed an absence of local toxic effects to the retina and the continuation of retinal differentiation and vascularization after 2 IVB injections (0.50 mg in 0.02 mL of solution),[26] more randomized control trials that include long-term monitoring of the patients in conjunction with systemic evaluations are warranted.

ROP patients seem to have only minimal refractive errors after receiving IVB. The patients in our study had a mean spherical power of 0.8 D and a mean cylindrical power of −2.1 D with a mean axial length of 20.7 mm at 18 months of age. These observations seem to coincide with the refractive data from subthreshold ROP patients who have not received laser or cryotherapy.[55] The refractive errors after IVB injection seem to be less severe than those observed in the eyes treated either with laser or cryotherapy. The difference in refractive error between the current study and other studies using laser or cryotherapy for ROP is shown in Table 2.[56–63] However, the refractive error data were preliminary and were available only for a limited number of patients. More studies are necessary to confirm this finding.

One of the limitations of this study is that it was retrospective; it lacks a uniform protocol for patient management. For example, fundus photography was not available in all of the study centers. However, some useful information still can be retrieved from the present study. The effects of IVB for ROP were evaluated for a large number of eyes in an Asian population. Data were collected from several treatment centers that were located all

over the country, and the mean follow-up period for the participating patients was longer than 1 year. However, many questions remain unanswered. For example, the optimal dosing of bevacizumab for the treatment of ROP, the optimal injection time, the value of combined treatments, and the long-term safety and efficacy of using IVB in a child's eye all must be determined. Further study is necessary to answer these questions.

In conclusion, the effects and complications of using bevacizumab in the treatment of ROP are understood better because the present study includes a larger number of patients than previous reports and these patients were monitored for a longer follow-up period. Tunica vasculosa lentis and plus disease both respond quickly to treatment with bevacizumab. A single injection of bevacizumab resulted in the regression of ROP in 88% of the prethreshold ROP patients who were included in this multicenter study. Early laser intervention seems to decrease the incidence of late recurrences of ROP in patients who do not respond to bevacizumab therapy. The refractive errors observed after treatment with IVB are mild, and no obvious systemic complications are noted during the follow-up period. Although this initial experience with the use of IVB in the treatment of ROP offers promising results, ocular complications do occur after the administration of IVB in some cases. Close monitoring of ROP patients is necessary after anti-VEGF treatment because there is a possibility that a given patient will not respond to bevacizumab treatment, so doctors should be ready to intervene with the use of a back-up treatment.

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REFERENCES

1. Alon T, Hemo I, Itin A, et al. Vascular endothelial growth factor acts as a survival factor for newly formed retinal vessels and has implications for retinopathy of prematurity. *Nat Med* 1995;1(10):1024–1028.
2. Early Treatment For Retinopathy Of Prematurity Cooperative Group. Revised indications for the treatment of retinopathy of prematurity: results of the early treatment for retinopathy of prematurity randomized trial. *Arch Ophthalmol* 2003;121(12):1684–1694.
3. Repka MX, Tung B, Good WV, et al. Outcome of eyes developing retinal detachment during the Early Treatment for Retinopathy of Prematurity Study (ETROP). *Arch Ophthalmol* 2006;124(1):24–30.
4. Gilbert WS, Quinn GE, Dobson V, et al. Partial retinal detachment at 3 months after threshold retinopathy of prematurity. Long-term structural and functional outcome. Multicenter Trial of Cryotherapy for Retinopathy of Prematurity Cooperative Group. *Arch Ophthalmol* 1996;114(9):1085–1091.
5. Wu WC, Drenser KA, Lai M, et al. Plasmin enzyme-assisted vitrectomy for primary and reoperated eyes with stage 5 retinopathy of prematurity. *Retina* 2008;28(3 suppl):S75–S80.
6. Hurwitz H, Fehrenbacher L, Novotny W, et al. Bevacizumab plus irinotecan, fluorouracil, and leucovorin for metastatic colorectal cancer. *N Engl J Med* 2004;350(23):2335–2342.
7. Bashshur ZF, Haddad ZA, Schakal AR, et al. Intravitreal bevacizumab for treatment of neovascular age-related macular degeneration: the second year of a prospective study. *Am J Ophthalmol* 2009;148(1):59–65.
8. Spaide RF, Laud K, Fine HF, et al. Intravitreal bevacizumab treatment of choroidal neovascularization secondary to age-related macular degeneration. *Retina* 2006;26(4):383–390.
9. Avery RL, Pearlman J, Pieramici DJ, et al. Intravitreal bevacizumab (Avastin) in the treatment of proliferative diabetic retinopathy. *Ophthalmology* 2006;113(10):1695.e1–e15.
10. Mason JO 3rd, Nixon PA, White MF. Intravitreal injection of bevacizumab (Avastin) as adjunctive treatment of proliferative diabetic retinopathy. *Am J Ophthalmol* 2006;142(4):685–688.
11. Spaide RF, Fisher YL. Intravitreal bevacizumab (Avastin) treatment of proliferative diabetic retinopathy complicated by vitreous hemorrhage. *Retina* 2006;26(3):275–278.
12. Jonas JB, Libondi T, von BS, Vossmerbaeumer U. Intravitreal bevacizumab for vitreous haemorrhage. *Acta Ophthalmol* 2008;86(5):585–586.
13. Ruiz-Moreno JM, Montero JA, Lugo F, et al. Intravitreal bevacizumab in recurrent diabetic vitreous haemorrhage after vitrectomy. *Acta Ophthalmol* 2008;86(2):231–232.
14. Iliev ME, Domig D, Wolf-Schnurrbursch U, et al. Intravitreal bevacizumab (Avastin) in the treatment of neovascular glaucoma. *Am J Ophthalmol* 2006;142(6):1054–1056.
15. Jaissle GB, Leitritz M, Gelisken F, et al. One-year results after intravitreal bevacizumab therapy for macular edema secondary to branch retinal vein occlusion. *Graefes Arch Clin Exp Ophthalmol* 2009;247(1):27–33.
16. Prager F, Michels S, Kriechbaum K, et al. Intravitreal bevacizumab (Avastin) for macular oedema secondary to retinal vein occlusion—12-month results of a prospective clinical trial. *Br J Ophthalmol* 2009;93(4):452–456.
17. Kriechbaum K, Michels S, Prager F, et al. Intravitreal Avastin for macular oedema secondary to retinal vein occlusion: a prospective study. *Br J Ophthalmol* 2008;92(4):518–522.
18. Sato T, Kusaka S, Shimojo H, Fujikado T. Simultaneous analyses of vitreous levels of 27 cytokines in eyes with retinopathy of prematurity. *Ophthalmology* 2009;116(11):2165–2169.

19. Sato T, Kusaka S, Shimojo H, Fujikado T. Vitreous levels of erythropoietin and vascular endothelial growth factor in eyes with retinopathy of prematurity. *Ophthalmology* 2009;116(9): 1599–1603.
20. Mintz-Hittner HA, Kennedy KA, Chuang AZ. Efficacy of intravitreal bevacizumab for stage 3+ retinopathy of prematurity. *N Engl J Med* 2011;364(7):603–615.
21. Wu WC, Yeh PT, Chen SN, et al. Effects and complications of bevacizumab use in patients with retinopathy of prematurity: a multicenter study in Taiwan. *Ophthalmology* 2011; 118(1):176–183.
22. Lalwani GA, Berrocal AM, Murray TG, et al. Off-label use of intravitreal bevacizumab (Avastin) for salvage treatment in progressive threshold retinopathy of prematurity. *Retina* 2008;28(3 suppl):S13–S18.
23. Mintz-Hittner HA, Kuffel RR Jr. Intravitreal injection of bevacizumab (Avastin) for treatment of stage 3 retinopathy of prematurity in zone I or posterior zone II. *Retina* 2008; 28(6):831–838.
24. Quiroz-Mercado H, Martinez-Castellanos MA, Hernandez-Rojas ML, et al. Antiangiogenic therapy with intravitreal bevacizumab for retinopathy of prematurity. *Retina* 2008; 28(3 suppl):S19–S25.
25. Kusaka S, Shima C, Wada K, et al. Efficacy of intravitreal injection of bevacizumab for severe retinopathy of prematurity: a pilot study. *Br J Ophthalmol* 2008;92(11):1450–1455.
26. Kong L, Mintz-Hittner HA, Penland RL, et al. Intravitreous bevacizumab as anti-vascular endothelial growth factor therapy for retinopathy of prematurity: a morphologic study. *Arch Ophthalmol* 2008;126(8):1161–1163.
27. Rishi E, Rishi P, Ratra D, Bhende M. Off-label use of bevacizumab in retinopathy of prematurity. *Retina* 2009; 29(2):284–285.
28. Travassos A, Teixeira S, Ferreira P, et al. Intravitreal bevacizumab in aggressive posterior retinopathy of prematurity. *Ophthalmic Surg Lasers Imaging* 2007;38(3):233–237.
29. Azad R, Chandra P. Intravitreal bevacizumab in aggressive posterior retinopathy of prematurity. *Indian J Ophthalmol* 2007;55(4):319.
30. Chung EJ, Kim JH, Ahn HS, Koh HJ. Combination of laser photocoagulation and intravitreal bevacizumab (Avastin) for aggressive zone I retinopathy of prematurity. *Graefes Arch Clin Exp Ophthalmol* 2007;245(11):1727–1730.
31. Harder BC, von BS, Jonas JB, Schlichtenbrede FC. Intravitreal bevacizumab for retinopathy of prematurity. *J Ocul Pharmacol Ther* 2011;27(6):623–627.
32. Raizada S, Al KJ, Al FA. Early experience with intravitreal bevacizumab combined with laser treatment for retinopathy of prematurity. *Middle East Afr J Ophthalmol* 2011;18(2):196–197.
33. Axer-Siegel R, Snir M, Ron Y, et al. Intravitreal bevacizumab as supplemental treatment or monotherapy for severe retinopathy of prematurity. *Retina* 2011;31(7):1239–1247.
34. Wutthiworawong B, Thitiratsanont U, Saovaprut C, et al. Combine intravitreal bevacizumab injection with laser treatment for aggressive posterior retinopathy of prematurity (AP-ROP). *J Med Assoc Thai* 2011;94(suppl 3):S15–S21.
35. Chan-Kai BT, Lauer AK. Transconjunctival, sutureless 25-gauge lens sparing vitrectomy for stage 4 retinopathy of prematurity-related retinal detachments. *Retina* 2009;29(6): 854–859.
36. Smith LE. Through the eyes of a child: understanding retinopathy through ROP the Friedenwald lecture. *Invest Ophthalmol Vis Sci* 2008;49(12):5177–5182.
37. Foroozan R, Connolly BP, Tasman WS. Outcomes after laser therapy for threshold retinopathy of prematurity. *Ophthalmology* 2001;108(9):1644–1646.
38. Kychenthal A, Dorta P, Katz X. Zone I retinopathy of prematurity: clinical characteristics and treatment outcomes. *Retina* 2006;26(7 suppl):S11–S15.
39. O'Keefe M, Lanigan B, Long VW. Outcome of zone 1 retinopathy of prematurity. *Acta Ophthalmol Scand* 2003; 81(6):614–616.
40. Davitt BV, Dobson V, Quinn GE, et al. Astigmatism in the Early Treatment for Retinopathy of Prematurity Study: findings to 3 years of age. *Ophthalmology* 2009;116(2):332–339.
41. Quinn GE, Dobson V, Davitt BV, et al. Progression of myopia and high myopia in the early treatment for retinopathy of prematurity study: findings to 3 years of age. *Ophthalmology* 2008;115(6):1058–1064.
42. Quinn GE, Dobson V, Hardy RJ, et al. Visual fields measured with double-arc perimetry in eyes with threshold retinopathy of prematurity from the cryotherapy for retinopathy of prematurity trial. The CRYO-Retinopathy of Prematurity Cooperative Group. *Ophthalmology* 1996;103(9):1432–1437.
43. Moshfeghi DM, Berrocal AM. Retinopathy of prematurity in the time of bevacizumab: incorporating the BEAT-ROP results into clinical practice. *Ophthalmology* 2011;118(7): 1227–1228.
44. Yang MB, Donovan EF, Wagge JR. Race, gender, and clinical risk index for babies (CRIB) score as predictors of severe retinopathy of prematurity. *J AAPOS* 2006;10(3):253–261.
45. Tadesse M, Dhanireddy R, Mittal M, Higgins RD. Race, Candida sepsis, and retinopathy of prematurity. *Biol Neonate* 2002;81(2):86–90.
46. Heidary G, Vanderveen D, Smith LE. Retinopathy of prematurity: current concepts in molecular pathogenesis. *Semin Ophthalmol* 2009;24(2):77–81.
47. Lashkari K, Hirose T, Yazdany J, et al. Vascular endothelial growth factor and hepatocyte growth factor levels are differentially elevated in patients with advanced retinopathy of prematurity. *Am J Pathol* 2000;156(4):1337–1344.
48. Lofqvist C, Niklasson A, Engstrom E, et al. A pharmacokinetic and dosing study of intravenous insulin-like growth factor-I and IGF-binding protein-3 complex to preterm infants. *Pediatr Res* 2009;65(5):574–579.
49. Miyake T, Sawada O, Kakinoki M, et al. Pharmacokinetics of bevacizumab and its effect on vascular endothelial growth factor after intravitreal injection of bevacizumab in macaque eyes. *Invest Ophthalmol Vis Sci* 2010;51(3):1606–1608.
50. Wu WC, Lai CC, Chen KJ, et al. Long-term tolerability and serum concentration of bevacizumab (Avastin) when injected in newborn rabbit eyes. *Invest Ophthalmol Vis Sci* 2010;51(7):3701–3708.
51. Qian J, Lu Q, Tao Y, Jiang YR. Vitreous and plasma concentrations of apelin and vascular endothelial growth factor after intravitreal bevacizumab in eyes with proliferative diabetic retinopathy. *Retina* 2011;31(1):161–168.
52. Sato T, Wada K, Arahori H, et al. Serum concentrations of bevacizumab (Avastin) and vascular endothelial growth factor in infants with retinopathy of prematurity. *Am J Ophthalmol* 2012;153(2):327–333.

53. Hard AL, Hellstrom A. On safety, pharmacokinetics and dosage of bevacizumab in ROP treatment—a review. *Acta Paediatr* 2011;100(12):1523–1527.
54. Micieli JA, Surkont M, Smith AF. A systematic analysis of the off-label use of bevacizumab for severe retinopathy of prematurity. *Am J Ophthalmol* 2009;148(4):536–543.
55. Sahni J, Subhedar NV, Clark D. Treated threshold stage 3 versus spontaneously regressed subthreshold stage 3 retinopathy of prematurity: a study of motility, refractive, and anatomical outcomes at 6 months and 36 months. *Br J Ophthalmol* 2005;89(2):154–159.
56. Khwarg SI, Yu HG, Yu YS. Change of refraction in premature infants after cryotherapy for retinopathy of prematurity between the age of six months and three years. *Korean J Ophthalmol* 1995;9(2):111–116.
57. White JE, Repka MX. Randomized comparison of diode laser photocoagulation versus cryotherapy for threshold retinopathy of prematurity: 3-year outcome. *J Pediatr Ophthalmol Strabismus* 1997;34(2):83–87.
58. Laws F, Laws D, Clark D. Cryotherapy and laser treatment for acute retinopathy of prematurity: refractive outcomes, a longitudinal study. *Br J Ophthalmol* 1997;81(1):12–15.
59. Pearce IA, Pennie FC, Gannon LM, Weindling AM, Clark DI. Three year visual outcome for treated stage 3 retinopathy of prematurity: cryotherapy versus laser. *Br J Ophthalmol* 1998;82(11):1254–1259.
60. Connolly BP, McNamara JA, Sharma S, Regillo CD, Tasman W. A comparison of laser photocoagulation with trans-scleral cryotherapy in the treatment of threshold retinopathy of prematurity. *Ophthalmology* 1998;105(9):1628–1631.
61. Fallaha N, Lynn MJ, Aaberg TM Jr, Lambert SR. Clinical outcome of confluent laser photoablation for retinopathy of prematurity. *J AAPOS* 2002;6(2):81–85.
62. Al-Ghamdi A, Albiani DA, Hodge WG, Clarke WN. Myopia and astigmatism in retinopathy of prematurity after treatment with cryotherapy or laser photocoagulation. *Can J Ophthalmol* 2004;39(5):521–525.
63. Sahni J, Subhedar NV, Clark D. Treated threshold stage 3 versus spontaneously regressed subthreshold stage 3 retinopathy of prematurity: a study of motility, refractive, and anatomical outcomes at 6 months and 36 months. *Br J Ophthalmol* 2005;89(2):154–159.

Biosketch

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