Anti VEGF (BEVACIZUMAB) as rescue therapy in sight threatening ROP

Dr. Shaila Sharmin[1], Dr. Sanyam Bajimaya[2], Dr. Eli Pradhan[3], Dr. Govinda Paudyal[4]

Abstract

Purpose: *The aim of the study was to observe the efficacy and safety of injection antiVEGF as rescue therapy in sight threatening threshold ROP as monotherapy or in combination with conventional laser therapy.*

Patients and methods: *This was a retrospective observational study. 20 eyes of 13 preterm infants suffering from ROP suffering from threshold ROP, and they were treated with intravitreal injection Bevacizumab alone or combinedly with conventional laser photocoagulation were included. All eyes had stage III zone 2 with 'plus disease'. All eyes had a follow up period of atleast 3 month following treatment. On follow up detail fundus examination with documentation and retinoscopy was done.*

Results : *The study included 20 eyes of 9 male infants and 4 female infants. The gestational ages ranged from 26 to 35 weeks (mean 29.15 weeks), and the birth weights ranged from 830 to 1600 g (mean1257 g). All 20 eyes had stage III zone 2 ROP with plus disease; All study eyes following one or both treatment modalities remained stable during followup. Except 2 eyes develop temporal macular dragging which were treated combinedly with injection Bevacizumab and laser photocoagulation. Retinoscopic value was mean -3.44 and -2.35 diopter spherical equivalent for Bevacizumab alone and combinedly treated group respectively.*

Conclusion: *Intravitreal injection Bevacizumab alone or combinedly with laser photocoagulation treatment is effective for sight threatening ROP and does not seem to be associated with serious ocular or systemic adverse events*

Keywords: *intravitreal Bevacizumab, retinopathy of prematurity, threshold ROP*

Introduction

Retinopathy of prematurity previously known as Retrolental fibroplasia (RLF). ROP is a vasoproliferative retinal disorder of childhood, which has its natural course of either spontaneous regression, or progression to macular dragging and retinal detachment and causing children to become blind in early childhood. It has become an emerging cause of blindness and visual morbidity in the surviving premature infants. Supplemental oxygen in higher concentration for longer duration with or without systemic co-morbidity such as neonatal septicemia, neonatal jaundice, respiratory distress syndrome and intraventricular hemorrhage in premature low birth weight infants provides important risk factors for development and progression of ROP.

The worldwide prevalence of blindness due to

1. FCPS. Consultant Medical Retina, Chittagong Eye Infirmary and Training Complex
2. MD, Consultant Vitreoretina Surgeon, Netradham Eye Centre, Nepal
3. MD, MRCSEd, Associate Prof, Tilganga Institute of Ophthalmology, Nepal
4. MD, Prof, Tilganga Institute of Ophthalmology, Nepal

Address of Correspondence: Dr Eli Pradhan, MD, MRCSEd Opth,UK, Consultant Medical Retina, Tilganga Institute of Ophthalmology, Associate Prof, NAMS, elipranjitkar@gmail.com

retinopathy of prematurity is 50,000.(1) In United states and other developed countries ROP accounts for 4% of childhood blindness, whereas in the developing ones it is higher. Near about 40% of it.[2] There it occurs primarily in infants of low birth weight(1250 g; mean, 700 g).(1)Their incidence of blindness in infants due to retinopathy of prematurityis about 1 case in 820 infants(3), which is relatively low because of good neonatal care and appropriate screening and treatment.(1) In developing countries it is a major cause of childhood blindness, develops in larger premature infants(birth weight 2000 g; mean, 1400 g).

Nepal is a developing country in South Asia. According to the Nepal Demographic Health Survey 2016 births in a health facility increased from 35% in 2011 to 57% in 2016 whereas Neonatal mortality dropped from 33% in 2011 to 21% in 2016. With the increase availability of neonatal intensive care a lot more babies born prematurely are surviving. The incidence of ROP in Nepal is not negligible.In two previous study incidence of ROP was 29.5% and 25.5%. And among them treatment requiring ROP was 3.8% and 5.5%.(4,5)

Increased vascular endothelial growth factor (VEGF) expression, altered insulin-like growthfactor-1 (IFG-1) expression and others play important role in the pathogenesis of retinopathy of prematurity.(6-8)ROP is a biphasic disorder. Phase 1 involves relative hyperoxia and decreased vascular endothelial growth factor (VEGF) levels; hyperoxia suppresses VEGF expression and results in vaso-obliteration. Meanwhile, in phase 2, VEGF expression is increased due to peripheral retinal hypoxia, leading to neovascularization.(10)

Abnormal retinal vasculature is the hallmark of ROP.(11)The International Classification of Retinopathy of Prematurity (ICROP), published in1984, defined ROP in terms of location (zones I–III),severity (stages I–V), extent (clock hours 1–12), and vascular dilatation and tortuosity (plus disease). Staging of ROP is crucial to decide when to treat. ROP in zone I is the most difficult to treat and has a high incidence of recurrence warranting additional treatment. Stage III, in which a ridge with neovascularization extends into the vitreousgel, is the ideal time for treatment.(10)The mainstay of treatment for threshold ROP is ablation of peripheral avascular retina through cryotherapy or laser. However threshold ROP is defined as stage III ROP in zone I or II involving at least five contiguous or eight cumulative clock hours with plus disease. Plus disease, emerge as a sign of high-risk prethreshold ROP that benefits from early laser ablation of the peripheral avascular retina.(11)

In 1988, cryotherapy (freezing from the externalocular surface, affecting the sclera, choroid, andthe full thickness of the retina) was recommended for threshold ROP(12)In the 1990s, there was a slow transition of treatment from cryotherapy to laser therapy (in which a laser is applied through the dilated pupil to the internal retinal surface). Both these treatments destroy the avascular retina that produce vascular endothelial growth factor (VEGF) in the retina.Conventional laser therapy for zone I retinopathy ofprematurity is successful in inducing regressonof ROP in 50% of case but inevitably causes permanent loss of the peripheral visual field and often induces clinically significant myopia. When multiple applications of conventional laser therapy fail to induce regression of retinopathy of prematurity, vitrectomy is required.(13)

As ablative treatments often requires intubation and anesthesia in preterm neonates with comorbid conditions, to find out an alternative treatment, Off-label use of intravitreal bevacizumab (IVB) therapy for ophthalmologic neovascular disorders began shortly after its approval by the Food and Drug Administration (FDA) in 2004 for non ophthalmic purposes. Cases and series of stage III, IV, and V ROP with plus disease had been treated with IVB, both as monotherapy and in combination with conventional laser therapy or vitrectomy or both.(10,14,15)

In this retrospective observational study, we observe the efficacy and safety of inj antiVEGF as

rescue therapy in sight threatening threshold ROP as monotherapy or in combination with conventional laser therapy.

Methodology

Study included the babies who were treated and followed up within 1st july 2018 to 30thjune 2019. In this study we retrospectively observed 20 eyes of 13 preterm infants suffering from threshold ROP, and they were treated with intravitreal inj Bevacizumab or combinedly with conventional laser as disease process demands. All cases underwent treatment in vitreo-retina department of Tilganga Institute Of Ophthalmology. The ROPstage and plus disease were defined on the basis of theinternational classification scheme (ICROP).(13)The detection of peripheral neovascular activity duringthe preoperative examinations and follow-up was performed using indirect ophthalmoscope. Documentation of fundus pathology was done by using FORUS Retcam (3 Nethra).

Inclusion criteria were preterm infants having bilateral or unilateral ROP stage III with plus disease affecting zone II treated with inj Bevacizumab monotherapy or combinedly with conventional laser in atleast one eye. Exclusion criteria were eyes with 4/5 stage of retinal detachment or congenitalocular anomaly or eyes that needed surgery. Data were collected from patient's hospital record.Data were collected for each baby regarding sex, , gestational age at birth, birth weight, the stage of ROP, the affected zone, and presence of risk factor like oxygenand surfactant given or not, presence of common problems of prematurity and stay of infant in the Intensive Care Unit of the Neonatology Department, given treatment(inj Bevacizumab as monotherapy or combined with conventional laser), fundus status at 1stand 3rd month, complication of treatment, retinoscopy value after 3rd month.

Pupil was dilated using topical mydriatic drops (1%tropicamide and 2.5% phenylephrine) and was instilled twice in both eyes10 min apart, at least 30 min before the treatment. The treatment was performed under anaesthetics uper vision using topical proparacaine hydrochloride (0.5%) applied for both eyes. Indirect laser photocoagulation was used. It was applied to the 360°avascular retina, extending to the oraserrata with confluent spots with less than half burn width apart. This was performed using 810-nm infrared diode laser. This laser is mounted on indirect ophthalmoscope (Heine, Germany). The procedure usually lasts from 15 to 20 minutes for each eye depending on the extent of treatment required and the zone of the disease.

The laser parameters depending on fundus pigmentation and area to be treated, ranged between140mW to 240mW for 150ms to 200ms with the repeat mode set at 200ms. The intensity of burns was grayishwhite rather than white and placement of spots was nearly confluent.

Bevacizumab was obtained from the commercially available Avastin (Genentech/Roche Inc., South San Francisco, California, USA). Sterile gloves, speculum, and forceps wereutilized while administering the injections. Sterilizationof skin was performed using povidone-iodine 10%. Aspeculum for premature infants was placed between the lids. A drop of povidone-iodine (5%) ophthalmic solution was placed into the conjunctival sac for 1minute with the excess removed by a sterile cotton tipapplicator from the temporal lid margin. The eye was stabilized with sterile cotton tip applicator while the dose of bevacizumab 0.025 ml (0.625 mg) was injected in the superior nasal quadrant behind the lens. The needle, aimed posteriorly (toward the optic nerve), entered the sclera through the conjunctiva 1.5 mm behind the limbus and was advanced approximately two-third of the length of the needle (not to the hub), and the syringe

was emptied completely into the central vitreous. After the injection antibiotic ointment(ciprofloxacin) was placed intothe conjunctival sac and pad bandage was given for 3 hours. In addition, the ophthalmic antibiotic (ciprofloxacin) drops were prescribed for both eyes to begin immediately and to be continued every 6 h for 7 days. The babies were examined using indirect ophthalmoscope on the following

day to specifically look for any sign of vitreous infection and signs of ROP progression or regression.

Following laser or injection, post-treatment examination took place 5–7 days after treatment and continued at least weekly for signs of decreasing activity and regression and to determine whether further intervention required or not. The following characteristics of regression was searched for during follow up:

(1) Lack of increase in severity.

(2) Partial resolution progressing toward complete

(3) Transgression of vessels through the demarcation line.

(4) Regression of plus sign and replacement of active ROP lesions by scar tissue.

When signs of improvement appear, stable retina was seen then follow up was done every 3 monthly. The primary ocular outcome was response to modality of treatment (monotherapy or combined).

In addition, at the end of the 3-month follow-up examinations, refraction was performed not to skip any possible refractive error and findings were noted.

Statistical analysis was done using SPSS.

Results

The study includes 20 eyes of 13 preterm babies with threshold ROP. The study comprised 9 male and 4 female. The mean gestational age (GA) at birth was 29.15±2.48 week and the mean birth weight was 1257±225.14 gm. Among 13 babies all were admitted in NICU after birth, 8 babies had history of Oxygen therapy and 3 babies had history of NNJ and 1 baby had history of RDS. All eyes showed Zone II Stage 3 ROP with plus disease. As first modality treatment 69.2% (16 eyes)received intravitreal injection Bevacizumab and 4 eyes received conventional laser (Fig:2).Among 16 eyes that received injection, 38.5%(10 eyes) needed laser treatment subsequently. And 4 eyes that received laser as initial treatment, needed injection subsequently that is combined treatment (Fig:3). So 6 eyes were treated solely with anti VEGF Bevacizumab injection. And combined treatment were given in 14 babies. If we look for complication, there was no immediate complication of injection or laser. On post treatmaent follow up all the treated eyes were vascularized or lasered stable except 2 eyes develop temporal macular dragging who were treated combinedly (Fig;3) which is statistically significant(Wilcoxon sign ranks test,Z value=-4.714; $p<0.05$).

Table I : *Baseline characteristics of infants with ROP treated with intravitreal injection of Bevacizumab monotherapy or combinedly with laser*

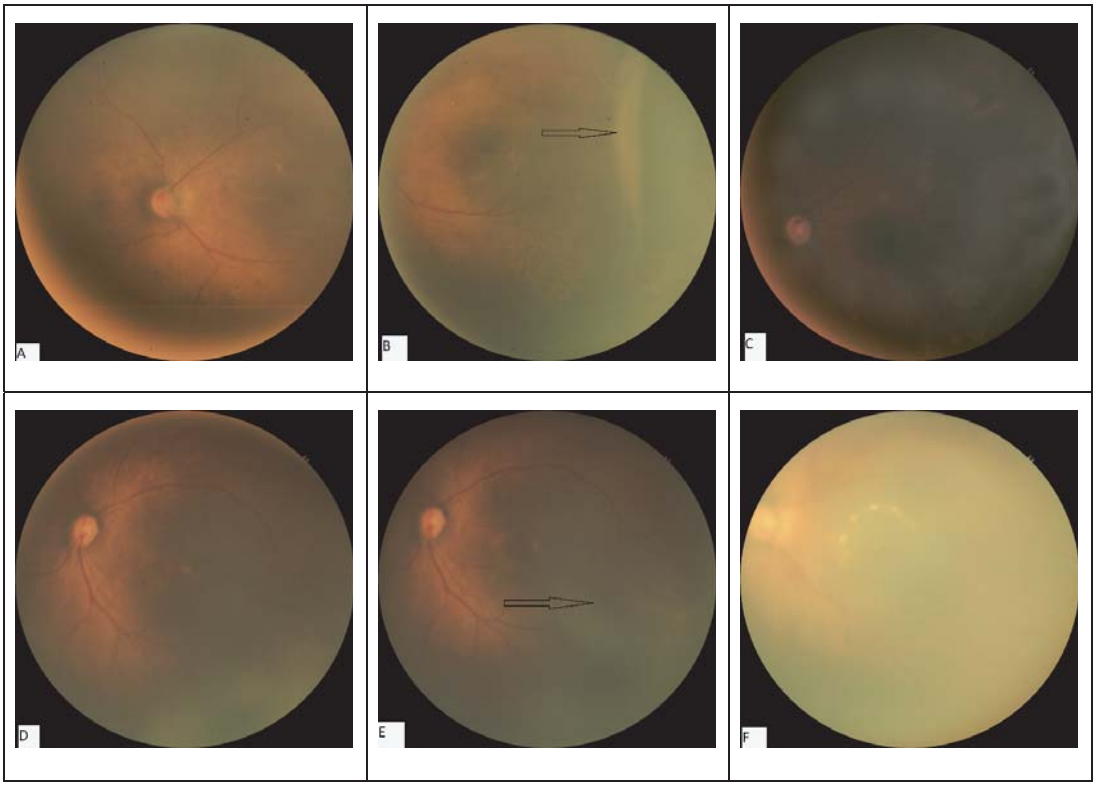
Parameters	No
Number of patients/eyes	13/20
M/F	9/4
BW, mean±SD g	1257 ±225.14
GA, mean±SD , wk	29.15 ±2.48 (26-32)
Stage/Zone	3/2
BW: Birth weight; GA: Gestational age;	

Table II : *First modality treatment*

	Frequency	Percent
BE Inj. Bevacizumab 0.625mg	7	53.8
R/E Inj. Bevacizumab 0.625mg	1	7.7
L/E Inj. Bevacizumab 0.625mg	1	7.7
R/E laser	3	23.1
L/E laser	1	7.7
Total	**13**	**100**

Table III : *Subsequent Treatment*

	Frequency	Percent
B/E Laser	5	38.5
R/E Inj. Bevacizumab 0.625mg	3	23
L/E Inj. Bevacizumab 0.625mg	1	7.7
None	4	30.80
Total	**13**	**100.0**



Patient 1 information: male, GA: 27wk, BW 1100 g, Zone II Stage 3+ ROP before treatment (A, B) and 1month after intravitrealbevacizumabinjection followed by laser develop left eye temporal macular dragging (C); Patient 2 information: male, GA: 26wk, BW 1320g, Zone II Stage 3+ ROP before treatment (D,E) and vascularized stable retina 1month after injbevacizumabmonotherapy.(F)

Patient 1 information: male, GA: 27wk, BW 1100 g, Zone II Stage 3^{+} ROP before treatment (A, B) and 1month after intravitreal bevacizumabinjection followed by laser develop left eye temporal macular dragging (C); Patient 2 information: male, GA: 26wk, BW 1320g, Zone II Stage 3^{+} ROP before treatment (D,E) and vascularized stable retina 1month after inj bevacizumab monotherapy.(F)

On retinoscopic value, the 6 eyes that were treated only with injection Bevacizumab for threshold ROP had a mean -3.44 ±3.04 diopter spherical equivalent value.And 10 eyes that were treated combinedly with injection Bevacizumab and laser had a mean -2.35 ±2.73 diopter spherical equivalent value.

Discussion

Bevacizumab got it's approval by the Food and Drug Administration (FDA) for it's use in nonophthalmic purposes in 2004.Shortly after that Off-label use of intravitreal Bevacizumab (IVB) therapy for ophthalmologic neovascular disorders began. Retinopathy of prematurity is not out of this trend anymore. Vascular endothelial growth factor (VEGF) is the potent pro-angiogenic factor in the pathologic angiogenesis of ROP, and blocking the action of VEGF by anti VEGF could be expected to reduce vascular activity. It will help to maintain normal anatomy of eye. Anti VEGF therapy has been showing promising result in treating sight threatening threshold ROP. Initially it was mainly used for Zone I ROP, AP-ROP, and failed ROP after laser treatment.(17,18) The BEAT-ROP also showed benefit of injection Bevacizumab over conventional laser in case of zone 1 ROP, not for zone 2.(10) But many recent studies are showing good response for zone2 ROP. Mintz-Hittner et al.(19) reported a case series of stage III ROP in zone I or posterior zone II treated

by bilateral IVB which included 11 infants with the mean weight of 706.4 g and mean gestational ages of 24.3 weeks who received IVB. All 22 eyes were treated with only one injection and they develop complete retinal vascularization. None of the eyes require laser later on. No ocular or systemic complications were encountered. Xiu-Mei Yang et al(20) has reported good efficacy of antiVEGF in stage3 zone2 ROP. Here one week after intravitreal injection, 32 eyes (37.21%) had achieved complete regres-sion of ROP and a partial regression of ROP was observed in 54 eyes (62.79%). That partially regressed eyes were treated either with second dose of injection or laser photocoagulation.

In our study we retrospectively observed 20 eyes of 13 preterm infant with stage3 zone2 threshold ROP who were treated with injection Bevacizumab only or combinedly with conventional pan retinal laser. 16 eyes (69.2%) received intravitreal injection Bevacizumab and 4 eyes received conventional laser initially. Among 16 eyes that received injection,10 (38.5%) babies needed laser treatment subsequently. And 4 eyes that received laser as initial treatment, needed injection subsequently that is combined treatment.

CRYO-ROP published in 1988 recommended Cryotherapy for threshold ROP. (12) In the 1990s, there was introduction of laser therapy. Both these treatments destroy the avascular retina that produce vascular endothelial growth factor (VEGF) in the retina. CRYO-ROP established the benefit of peripheral retinal ablation for threshold ROP. It showed a decrease of more than 40% in unfavorable structural outcomes and a decrease of 30% in unfavorable visual acuity in treated eyes compared with observed eyes .But 44% of children with threshold ROP treated with cryotherapy had vision less than 20/200 at10 years.(12) Hence, the Early Treatment of Retinopathy of Prematurity (ET-ROP) trials were conducted in the early 2000s which demonstrated the benefit of earlier treatment with laser of high-risk prethreshold ROP .(21) Though the ETROP study showed that laser performed for high risk prethreshold disease resulted in reduction of unfavorable visual acuity outcomes from 19.8% to 14.3% and unfavorable anatomic outcomes from 15.6% to 9.0%. (22) But it does not eliminate the risk of peripheral visual field loss, high myopia. Thus the success of retinal ablation in treating ROP, is outweighted by it's disadvantages in a newborn such as cataract formation, anterior segment and vitreous hemorrhage, anterior segment ischemia, iris adhesions to the lens, and fluctuating intraocular pressures. In the long term, ablative therapy can lead to loss of peripheral vision, strabismus, and marked myopia. Intravitreal injection are lack of these ocular side effects and gives a favorable outcome. The BEAT ROP study not only showed significant benefit for zone 1 but also showed that development of peripheral retinal vessels continued after treatment with intravitreal injection bevacizumab, but conventional laser therapy led to permanent destruction of the peripheral retina.(10) in a study of 26 patients with stage 3 ROP treated with bevacizumab in whom involution of neovascularization, flattening of demarcation lines, and growth of differentiated vessels into capillary-devoid areas was noted.(22)Additionally, a more recent retrospective study showed that bevacizumab with or without zone I sparing laser reduced unfavorable outcomes (i.e., retinal folds, disc dragging, retinal detachments, early refractive errors, and retro-lental tissue obscuring the view of the posterior pole) from 22.7% to 0% as compared to laser alone.(24) A recent systemic review suggests that intravitreal anti-VEGF agents reduce the risk of refractive errors (high myopia) during childhood.(25)

In our study the 6 eyes that were treated only with injection Bevacizumab for threshold ROP had a mean -3.44 ±3.04 diopter spherical equivalent value.And 14 eyes that were treated combinedly with injection Bevacizumab and laser had a mean -2.35 ±2.73 diopter spherical equivalent value on retinoscopy. There was no significant difference in retinoscopic value between the only injection treated eyes and combinedly treated eyes. The value were not high enough to define as high myopia. Which can be explain by that Anti VEGF reduce vascular aggressiveness and decrease the need for extensive laser spots. So chance of laser

induced high myopia is reduced.

Spandau et al(26) discussed treatment modalities for AP-ROP in 16 eyes of eight infants who were treated with laser and/or IVB. Two eyes (one infant) were only treated with laser, and six eyes (three infants) were treated with laser therapy or cryopexy. And because of lack of regression, bevacizumab was used as rescue therapy. Eight eyes (four infants) were treated with a first-line IVB and four of these eyes (two infants) required additional laser ablation for continued disease progression in zoneII. One eye that was primarily treated with laser develop macular dragging. They concluded that, to avoid the high complication rate of the extensive laser treatment, it is now time to consider anti-VEGF treatment as an alternative treatment.

Our treatment complication match with the result of this study. In our study, two eyes develop temporal macular dragging which were treated combinedly with injection Bevacizumab and laser photocoagulation. Rest of the 18 eyes had stable retina without any complication.

But Wu et al. (27), reported in his study major ocular complications that were associated with IVB injections included vitreous or preretinal hemorrhage in two (1%) eyes, cataract in one (1%) eye, and exotropia in one (1%) eye.No notable systemic complications related to the IVB injections were observed in his study. Lee et al. (28)reported that, three of five eyes develop tractional retinal detachment from fibrous traction membrane that had arisen along the major vascular arcades after the regression of ROP. Those eyes were treated with inection Bevacizumab as initial therapy. They concluded that vision threatening complication can occur even In eyes treated with injection Bevacizumab as initial therapy. Therefore, ROP patients who received bevacizumab treatment should be closely followed up. The follow up of injection cases should also be continued long time to detect recurrence. Many studies has reported late recurrence with intravitreal injection.(29,30,) Because there is slow vascularization after intravitreal injection of anti VEGF.

Besides ocular side effects of anti VEGF, there is controversy about systemic side effect on preterm infants. VEGF is vital in angiogenesis, in maintaining organ health and in the development of various vital organs in the body. The inhibition of VEGF may lead to abnormal organogenesis or neurodevelopment because, possibly this inhibition effect these important physiologic effects associated with VEGF. (31) After intra-vitreal injection, VEGF inhibitors may enter the systemic circulation and decrease systemic VEGF levels as the blood retinal barrier is compromised in premature infants. Sato et al. found that bevacizumab could escape from the eye into the systemic circulation and reduce the serum VEGF level at least 2 weeks after intravitreal injection of bevacizumab (IVB) in infants with ROP.(32) Higher mortality rate of children was observed in the Bevacizumab treated eyes(6.6%)in comparison to laser treated eyes(2.6%).(33) Canadian Neonatal Network and the Canadian Neonatal Follow-Up Network Investigators showed that Preterm infants treated with bevacizumab versus laser had higher odds of severe neurodevelopmental disabilities.(34) On the other hand Wei-Chi Wu et al found no difference on neurodevelopment for those who received only bevacizumab versus only laser treatment even two years after laser and/or intravitreal injections of bevacizumab for ROP.(35) SO babies that are treated with injection Bevacizumab need to be further evaluated and monitored to detect any systemic side effect.

The limitation of our study was it's small small sample size. And it was an retrospective observational study. We had to depend on patient's hospital record.We could not compare our samle with only lase treated eyes. So our study result did not has the power to completely evaluate the efficacy and safety of Bevacizumab alone or combinedly with laser photocoagulation. We had fundus photograph as evidence but no fluorescein angiography was done for evaluation of ROP patients. Even though clinically we found that all the treated eyes had complete retinal vascularization after treatment by funduscopy, but since fluorescein angiography was not available it

remains unknown whether vascularization was normal or abnormal Further prospective studies with larger study populations are required to stablish the safety and effecacy of Bevacizumab for ROP treatment.

Conclusion

In conclusion we can say that injection Bevacizumab alone or combinedly with laser photocoagulation treatment is effective for sight threatening ROP and does not seem to be associated with serious ocular or systemic adverse events. It helps to avoid anatomical damage of ocular structure when used alone or combinedly. However, the increased risk of recurrence and systemic side effects according to the reported results with bevacizumab necessitates a longer follow-up period with more frequent monitoring.

Referrences

1. Gilbert C. Retinopathy of prematurity:a global perspective of the epidemics,population of babies at risk and implicationsfor control. Early Hum Dev 2008;84:77-82.
2. Good WV, Carden SM. Retinopathyof prematurity. British Journal of Ophthalmology. 2006;90(3):254-255.DOI: 10.1136/bjo.2005.081166
3. Lad EM, Nguyen TC, Morton JM,Moshfeghi DM. Retinopathy of prematurityin the United States. Br J Ophthalmol2008;92:320-5. [Erratum, Br J Ophthalmol2010;94:1268.]
4. Shrestha JB, Bajimaya S, Sharma A, Shrestha JK et al. Incidence of ROP in a Neonatal Intensive Care Unit in Nepal. Journal of Pediatric Ophthalmology & Strabismus. Vol. 47, No.5, 2010
5. Adhikari S1, Badhu BP, Bhatta NK, Rajbhandari RS, Kalakheti BK.Retinopathy of Prematurity in a tertiary care hospital in eastern Nepal. JNMA J Nepal MedAssoc. 2008;47(169):24-7
7. Hellstrom A, Peruzzi C, Ju M, et al. Low IGF-I suppressesVEGF-survival signaling in retinal endothelial cells: direct correlationwith clinical retinopathy of prematurity. Proc NatlAcad Sci U S A 2001;98:5804–5808.5
8. Connor KM, SanGiovanni JP, Lofqvist C, et al. Increased dietary intake of omega-3-polyunsaturated fatty acids reducespathological retinal angiogenesis. Nat Med 2007;13:868–873.
9. Gerhardt H, Golding M, Fruttiger M, et al. Article VEGF guides angiogenic sprouting utilizing endothelial tip cell filopodia.J Cell Biol 2003;161:1163–1178.
10. Mintz-Hittner HA, Kennedy KA, Chuang AZBEAT-ROP CooperativeGroup. Efficacy of intravitreal bevacizumab for stage 3+ retinopathy of prematurity. N Engl J Med 2011; 364:603–615.
11. Gelman R, Martinez-Perez ME, Vanderveen DK, Moskowitz A, Fulton AB.Diagnosis of plus disease in retinopathy of prematurity using Retinal Image multiScale Analysis. Invest Ophthalmol Vis Sci 2005; 46:4734–4738.
12. Cryotherapy for Retinopathy of Prematurity Cooperative Group. Multicenter trial of cryotherapy for retinopathy of prematurity:preliminary results. Arch Ophthalmol 1988;106:471-9.
13. 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;124:24-30.
14 Travassos A, Teixeira S, Ferreira P, Regadas I, Travassos AS, Esperancinha FE, et al. Intravitreal bevacizumab in aggressive posterior retinopathy of prematurity. Ophthalmic Surg Lasers Imaging 2007; 38:233–237.
15. Wu WC, Yeh PT, Chen SN, Yang CM, Lai CC, Kuo HK. Effects and complications of bevacizumab use in patients with retinopathy of prematurity:a multicenter study in Taiwan. Ophthalmology 2011; 118:176–183.
16. International Committee for the Classification of Retinopathy of Prematurity. The International Classification of Retinopathy of Prematurity revisited. Arch Ophthalmol 2005; 123:991–999.
17. Darlow BA, Ells AL, Gilbert CE, Gole GA, Quinn GE. Are we there yet? Bevacizumab therapy for retinopathy of prematurity. Arch Dis Child Fetal Neonatal Ed. 2013;98(2):F170–F174. [PubMed] [Google Scholar]
18. Eldweik L, Mantagos IS. Role of VEGF inhibition in the treatment of retinopathy of prematurity. Semin Ophthalmol. 2016;31(1-2):163–168. [PubMed] [Google Scholar]
19. 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; 28:831–838.
20.Xiu-Mei Yang, Yue-Xiang Zhao, Zong-HuaWang, and LuLiu.Effect of anti-VEGF

treatment on retinopathy of prematurity in Zone II Stage 3+. Int J Ophthalmol. 2018; 11(4): 641–644.

21. John VJ, Hess D, Berrocal MA. Avastin in ROP; the anti-VEGF drug has shown promise in the treatment of retinopathy of prematurity. Retinal Physician 2012; 9:42–45.
22. Good WV, Early Treatment for Retinopathy of Prematurity Cooperative G. Final results of the Early Treatment for Retinopathyof Prematurity (ETROP) randomized trial, Trans Am Ophthalmol Soc, 2004; 102:233–48; discussion 48–50
23. Henaine-Berra A, Garcia-Aguirre G, Quiroz-Mercado H, Martinez-Castellanos MA, Retinal fluorescein angiographic changes following intravitreal anti-VEGF therapy, J AAPOS, 2014;18:120-3.
24. Yoon JM, Shin DH, Kim SJ, et al., Outcomes after Laser VersusCombined Laser and Bevacizumab Treatment for Type 1Retinopathy of Prematurity in Zone I, Retina, 2017;37:88–96.
25. M. J. Sankar, J. Sankar, and P. Chandra, "Anti-vascular endothelialgrowth factor (VEGF) drugs for treatment of retinopathyof prematurity," Cochrane Database Syst Rev, vol. 1,2018.
26. Spandau U, Tomic Z, Ewald U, Larsson E, Akerblom H, Holmström G.Time to consider a new treatment protocol for aggressive posterior retinopathy of prematurity? Acta Ophthalmol 2012; 91:170–175.
27. Wu WC, Kuo HK, Yeh PT, Yang CM, Lai CC, Chen SN. An updated study of the use of bevacizumab in the treatment of patients with prethreshold retinopathy of prematurity in Taiwan. Am J Ophthalmol 2013; 155:150–158.
28. Lee BJ, Kim JH, Heo H, Yu YS. Delayed onset atypical vitreoretinal tractionband formation after an intravitreal injection of bevacizumab in stage 3 retinopathy of prematurity. Eye (Lond) 2012; 26:903–909. quiz 910
29 Hu J, Blair MP, Shapiro MJ, Lichtenstein SJ, Galasso JM, Kapur R.Reactivation of retinopathy of prematurity after bevacizumab injection. Arch Ophthalmol 2012; 130:1000–1006
30 Feng J, Qian J, Jiang Y, Zhao M, Liang J, Yin H, Chen Y, Yu W, Li X. Efficacy of primary intravitreal ranibizumab for retinopathy of prematurity in China. Ophthalmology. 2017;124(3):408–409. [PubMed] [Google Scholar]
31. L. S. Lim, C. M. G. Cheung, P. Mitchell, and T. Y. Wong, "Emerging evidence concerning systemic safety of anti-VEGF agents-should ophthalmologists be concerned?" American Journal of Ophthalmology, vol. 152, no. 3, pp. 329–331, 2011.
32. T. Sato, K. Wada, H. Arahori et al., "Serum concentrations of bevacizumab (Avastin) and vascular endothelial growth factor in infants with retinopathy of prematurity," American Journal of Ophthalmology, vol. 153, no. 2, pp. 327–333, 2012.
33. Darlow BA, Ells AL, Gilbert CE, Gole GA, Quinn GE. Are we there yet? Bevacizumab therapy for retinopathy of prematurity. Arch Dis Child Fetal Neonatal Ed. 2013;98(2):F170–F174. [PubMed] [Google Scholar
34. Morin J, Luu TM, Superstein R, Ospina LH, Lefebvre F, Simard MN Canadian Neonatal Network and the Canadian Neonatal Follow-Up Network Investigators, Neurodevelopmental Outcomes Following Bevacizumab Injections for Retinopathy of Prematurity. Pediatrics. 2016 Apr; 137(4). pii: e20153218. doi: 10.1542/peds.2015-3218
35. Reyin Lien, Mu-Hsien Yu , Kuang-Hung Hsu , Pei-Ju Liao, Yen-Po Chen, Chi Chun Lai, Wei-Chi Wu, Neurodevelopmental Outcomes in Infants with Retinopathy of Prematurity and Bevacizumab Treatment PLOS ONE | DOI:10.1371/journal.pone.0148019 January 27, 2016.