Are we there yet? Bevacizumab therapy for retinopathy of prematurity

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SUMMARY
The publication of the BEAT-ROP study of bevacizumab (Avastin) treatment for Zone I and II retinopathy of prematurity (ROP) has raised hopes that there might now be a simpler, cheaper and more effective treatment than laser therapy, the current standard of care. However, we would urge caution at this point in time. We review the scientific background to the use of intravitreal anti-vascular endothelial growth factor (VEGF) and subsequently its various isotopes, aimed at the eradication of VEGF. Antiangiogenesis was initially shown to be effective in inhibiting retinal vessel growth in ROP. More recently, there has been exciting progress for severe ROP were undertaken in Japan in the early 1970s and consisted of photocoagulation, initially of the neovascular tufts and later of the avascular retina. In a situation that parallels that with bevacizumab treatment, there followed a number of case series of photocoagulation and cryotherapy before a large multicentre trial demonstrated the effectiveness of the latter and also some of the longer-term issues.

ROP is recognised as progressing through two phases. Phase I begins with premature delivery and the relative retinal hypoxia of the extrauterine environment. Supplemental oxygen therapy further suppresses hypoxia-inducible factor and VEGF, which, together with the lack of the non-oxygen inducible insulin-like growth factor-1 (IGF-1) formerly secreted by the placenta and required for VEGF signalling, leads to the cessation of retinal vessel development. In Phase II, further growth of the neural retina and its high metabolic rate causes relative hypoxia, upregulation of VEGF and other growth factors from the avascular retina, and potential abnormal vessel development.

In Phase I, therapeutic interest has focused on the physiological replacement of IGF-1. A trial of rhIGF-1 as a complex with its binding protein, rhIGF-1BP3 (IPLEX), to prevent ROP and other complications of preterm birth is underway (ClinicalTrials.gov Identifier: NCT01096784).

In Phase II of ROP, the focus has shifted from laser panretinal photocoagulation to direct inhibition of VEGF. Antiangiogenesis was initially developed as a therapeutic tool to target the vigorous capillary growth that accompanies most solid tumour metastases. Bevacizumab is a full monoclonal antibody, which also binds to all VEGF isoforms and which received the United States Food and Drug Administration (FDA) approval for the systemic treatment of cancer of the colon in 2004 and subsequently for other cancers.

An intravitreal anti-VEGF neutralising antibody was reported to be effective in inhibiting retinal vasoproliferation in a murine model in 1995. Following clinical trials, pegaptanib (Macugen), an aptamer which specifically binds VEGF, first received FDA approval for intravitreal use in adults with neovascular age-related macular degeneration. However, for ROP, there are concerns about the potential systemic risks of using systemically directed drugs in the developing preterm infant because vascular growth factors play a critical role in organogenesis. We conclude that bevacizumab should be reserved for exceptional circumstances and compassionate use pending further studies.

INTRODUCTION
“Are we nearly there yet?” Parents of young children will be familiar with this refrain on many a journey. How tempting to answer “almost” when we know some way still lies ahead. Following the recent publication of the eagerly awaited Bevacizumab Eliminates the Angiogenic Threat of Retinopathy of Prematurity (BEAT-ROP) study, how tempting to say bevacizumab (Avastin) should be the treatment of choice for Zone I, stage 3+ ROP, and by extension other forms of retinopathy of prematurity (ROP), when a more cautious analysis will suggest we have not yet arrived at that point.

THE ROLE OF GROWTH FACTORS IN THE PATHOGENESIS OF ROP
It was suspected by early investigators that some factor derived from the retina anterior to the advancing vessels might orchestrate the abnormal vessel growth in ROP. But it was not until the 1980s that vascular endothelial growth factor (VEGF), and subsequently its various isoforms, were identified. Well before this understanding, however, the first attempts at ablative treatment...
in 2004 but it is expensive and its use has been limited. Ranibizumab (Lucentis) is a monoclonal antibody fragment that binds to all VEGF isoforms, which was developed for the treatment of eye diseases and was first used in trials to treat exudative age-related macular degeneration (AMD) in 2003. Ranibizumab was approved by the FDA for this indication in 2006. Bevacizumab, which is much cheaper than ranibizumab, has been used to treat AMD off-label from 2005. These two drugs have subsequently been shown to have highly beneficial, and largely equivalent, effects so that the treatment of exudative AMD has been transformed in recent years, albeit with the need for frequent follow-up to re-treat the recurrences. Given this experience, some ophthalmologists began to give intravitreal bevacizumab off-label to treat other neovascular eye diseases, including diabetic retinopathy and ROP.

TREATMENT OF ROP WITH INTRAVITREAL ANTVASCULAR GROWTH FACTORS

Intravitreal anti-VEGF agents have an appeal in the treatment of severe ROP because of a number of potential advantages. Although laser therapy, which has superseded cryotherapy, is usually effective (in the Early Treatment for Retinopathy of Prematurity Trial (ETROP) unfavourable visual acuity outcomes were seen at corrected age 6–9 months in 14.5% eyes treated with laser), ablative treatment is to some extent destructive of the peripheral retina. The recent revision of the International Classification of Retinopathy of Prematurity recognised an uncommon but rapidly progressive form of ROP, called aggressive posterior ROP (AP-ROP), which is typically located in Zone 1, and which exhibits four quadrants of severe plus disease despite a rather featureless peripheral proliferative retinopathy. ETROP did not use the AP-ROP designation and there is limited evidence on the effectiveness of treatment with laser. Laser therapy is difficult to undertake when there is poor visualisation of the retina from vitreous haze or inadequate pupil dilatation. An intravitreal injection is a shorter procedure than laser therapy, requires no special equipment and can be carried out with topical anaesthesia only, so avoiding intubation and a general anaesthetic which is sometimes preferred when laser is used. Add to these observations that bevacizumab is cheap and seemingly has few short-term side effects, and it is understandable that some ophthalmologists who treat retinal vascular disease in adults should consider using such treatment in eyes with severe ROP. However, this ocular benefit may ignore the particular systemic issues inherent in treating premature infants.

Micelli reviewed publications on the off-label use of bevacizumab in preterm infants up to March 2009 (77 eyes in 48 infants). The first six published case reports from 2007 to 2008 came from around the world. Most treatment was initiated for Zone I disease or AP-ROP, either after failed laser treatment or combined with laser as primary therapy, although in one case laser was considered contraindicated ‘as clinical status was poor’. Some reports did include cases where intravitreal bevacizumab was used as primary monotherapy. Micelli noted that there were two reports of worsening retinal detachment after anti-VEGF treatment and commented that there were many uncertainties as to dose, timing and frequency of treatment, as well as possible side effects and long-term outcomes. These uncertainties still exist.

TRIALS OF INTRAVITREAL BEVACIZUMAB

The BEAT-ROP trial was a prospective, randomised, non-blinded Phase II study of intravitreal bevacizumab (0.625 mg in 0.025 ml) versus diode laser as the primary treatment for stage 3+ ROP in Zone I or posterior Zone II. Both eyes were randomised to the same treatment. The primary outcome was originally the absence of recurrence of stage 3 plus disease before 54 weeks postmenstrual age (PMA). Before the data were first analysed, this primary outcome was changed to ‘ROP recurrence needing treatment before 54 weeks PMA’.

One hundred and fifty infants were enrolled, with seven dying before 54 weeks PMA and not included in the primary analysis. Overall, ROP requiring retreatment occurred in 470 infants (6%) treated by bevacizumab and 1973 (26%) treated by laser (OR 0.17 (95% CI 0.05 to 0.53) p=0.002). Significant benefit was seen in eyes with Zone I disease (OR 0.09 (95% CI 0.02 to 0.43) p=0.003) but not in eyes with Zone II disease (OR 0.39 (95% CI 0.07 to 2.11) p=0.27). The timing of recurrences needing treatment was later following bevacizumab (19.2±8.6 weeks) compared with laser (6.4±6.7 weeks).

Of the seven deaths before 54 weeks, three had Zone I disease and four Zone II, five were treated with bevacizumab (6.6%) and two with laser (2.6%), but this difference was not significant. Four of five bevacizumab group and one of two laser group deaths were from respiratory causes.

There are a number of concerns about the trial design of the BEAT-ROP study and interpretation of the results. The treatment criteria were not those established by the ETROP trial (see box 1). The BEAT-ROP treatment criteria (stage 3+ in Zone I or posterior Zone II) included only a subset of eyes with Type I disease. The primary outcome was not masked. Treated eyes were assessed by the treating ophthalmologist with a second ophthalmologist confirming the need for retreatment. Although RetCam photographs were taken, these were only for the purposes of a post-hoc independent review. Visual outcomes were not reported, but are eagerly awaited along with studies of retinal electrophysiology.

Recurrence of ROP may have occurred after the end point. There is some evidence to suggest that anti-VEGF agents might damp down acute disease, only for it to recur at a later time. As Moshfegi has pointed out from the timing of recurrences observed in the bevacizumab group, it is possible that some recurrences may have occurred after the 54-week PMA cut-off point. One major implication is that bevacizumab treatment requires frequent retinal examinations to detect later recurrences, thereby adding cost to patients and providers.

A relatively high dose of bevacizumab was given. The dose used in the BEAT-ROP trial, 0.625 mg, is half the adult dose; yet the vitreous volume of an infant at 54 weeks is less (1.6 ml vs 4.0 ml), as is the retinal surface area (450 mm² vs 1240 mm²), and the body weight is about one-fiftieth of an adult. Sears has estimated that 0.5–1.0 mg bevacizumab is 10 000 times the concentration needed to neutralise the highest measured concentration of VEGF. Case series are already being reported where a lower dose of bevacizumab (0.575 mg) has produced...
regression of retinal neovascular changes and future studies should explore this further.

The outcome following laser treatment was worse than expected. The recurrence rate after laser of 26% was much higher than in the published series of 11–14%,22 No data are given on the laser pattern and burn intensity but there is good evidence that more dense and near confluent patterns are associated with better outcomes,32 including in cases with AP-ROP.24 In addition, 4 of 40 cases with Zone II disease had treatment complications that normally would be exceptionally rare (one case of corneal opacity, three of lens opacity—all of which required surgery).

### POTENTIAL HARMFUL EFFECTS OF INTRAVITREAL BEVACIZUMAB

#### Local

Local complications of bevacizumab related to intravitreal injection include endophthalmitis, retinal haemorrhage, cataract and retinal detachment.18–33 Bevacizumab is marketed in a preservative-free solution and preparation of aliquots for intravitreal injection must be under sterile conditions.34 There are several reports of delayed retinal detachment following anti-VEGF therapy.29 35 36 Mintz-Hittner37 has commented that if bevacizumab is given too late, then this might exacerbate any pre-existing traction retinal detachment (a well-recognised complication of the treatment of proliferative diabetic retinopathy). Knowledge of the anatomy of the eye in a premature infant is essential to give an injection safely.

#### Systemic

In adults treated with intravenous bevacizumab, increased risks of cerebral vascular accident, myocardial infarction and systemic hypertension have been reported.16 The role of bevacizumab in combination with chemotherapy as intravenous treatment for some cancers is being questioned because there is emerging evidence of increased treatment-related mortality, the most common events leading to death being haemorrhage, neutropenia and gastrointestinal perforation.38 39 In adults treated with intravitreal bevacizumab systemic complications are not easy to quantify16 but there is emerging evidence of increased risks of haemorrhagic stroke.40

Although potential systemic effects of intravitreal bevacizumab use in preterm infants are the greatest concern, they are also the hardest to determine. The BEAT-ROP trial was not powered to assess the systemic safety. Mintz-Hittner4 argues that because bevacizumab is a large molecule and the preterm vitreous highly viscous, it unlikely to escape the eye, and the long intravitreal half-life (5–10 days) is therefore seen as an advantage. In the editorial accompanying the BEAT-ROP study, Reynolds4 states ‘it seems reasonable to assume that intravitreal bevacizumab is safe’. But is this the case?

Bevacizumab has been reported to enter the systemic circulation following intravitreal injection in animal models and adults. Following an intravitreal injection of 1.25 mg into a monkey eye, radioactive labelled bevacizumab was detectable in the serum 1 day after the injection and 5% of the injected dose was measurable in the blood on day 7.41 In adult macaques (weighing 3.9–5.5 kg), one intravitreal injection of 1.25 mg bevacizumab into one eye gave peak serum concentrations of the antibody at 1 week and still half this at 8 weeks.42 There are similar data in a rabbit model43 and in newborn rabbits higher serum concentrations were seen when the intravitreal injection was given earlier rather than later in the neonatal period.44 Avery reported therapeutic effects on the fellow eye when an intravitreal injection was given to one eye for proliferative diabetic retinopathy and a similar bilateral response after a unilateral injection has been reported in a child with uveitic cystoid macular oedema.46 Matsuyama47 found in adults with diabetic retinopathy that plasma concentrations of VEGF were still significantly reduced 1 month after a single injection of 1.25 mg bevacizumab and there are other similar reports.48

Studies of eyes with acute ROP using fluorescein angiography have clearly demonstrated vascular leakage and breakdown of the blood ocular barrier.49 Despite an urgent need to measure systemic concentrations of circulating VEGF and the antibody after intravitreal bevacizumab injections in infants with ROP, the only published report to date50 comes from infants who had previously been treated with laser. Bevacizumab concentrations reached a peak at 2 weeks postinjection while VEGF concentrations were lower 1 week after the injection and negatively correlated with bevacizumab concentrations.

VEGF is crucial for the survival in newborn animal models and for the growth of many organs.50 Preterm infants are still undergoing organogenesis at the time of ROP treatment late in the third trimester. VEGF has actions beyond vasculogenesis and angiogenesis.2 In the eye, VEGF is required for normal retinal development independent of angiogenesis.12 In the brain, it is neurotropic and neuroprotective and helps maintain the blood-brain barrier.51 Reviews on the complex processes of brain development do not focus extensively on the development of the vasculature although it is clear that this does play a crucial role.52 53 In the lungs, VEGF has important roles in alveolisation54 as well as surfactant synthesis.55 VEGF is also critical for glomerulogenesis and skeletal growth.9

We have noted the increase in mortality in the BEAT-ROP study in the bevacizumab group compared with the laser group (6.6% vs 2.6%).1 Although not significant in this study, mortality will need to be closely monitored in future studies and as increased numbers of infants are treated with intravitreal bevacizumab. Four of the five deaths in the treatment group (and one of the two for the laser group) were pulmonary deaths. In a newborn rat model, a single dose of a VEGF receptor inhibitor (Su-5416) impaired pulmonary vascular growth and postnatal alveolisation, causing pulmonary hypertension which was long lasting.56 Irreversible pulmonary hypertension has also been reported in adults with ovarian cancer treated with repeat doses of intravenous bevacizumab.57

### TREATMENT OF ROP IN A WORLD CONTEXT

The increasing proportion of ROP in middle-income countries, where it has been called the ‘third epidemic of ROP’, has been well documented.58 59 While most of this is potentially preventable with current knowledge, health systems are often not able to deliver the high level of neonatal care required. In these countries, there are frequently too few ophthalmologists to examine at risk infants and few facilities to treat with laser therapy. Ophthalmologists are often not or only poorly compensated for their time screening and treating ROP. In these circumstances, a treatment that could be easy to administer in the intensive care unit would have obvious advantages.

Members of our group have been conducting workshops to improve ROP programmes in Central and South America and Southeast Asia for a number of years, and we know from personal experience that intravitreal anti-VEGF agents are being used extensively in many countries. Many infants who are
treated are bigger and more mature than those in more developed countries and do not have Zone 1 disease, but do have significant growth and development ahead of them. In these countries, AP-ROP may sometimes occur, presumably because of the die back of vessels already in Zone II. The latter can, however, be effectively treated with laser therapy.60

Following publication of the BEAT-ROP study, we are concerned that the trend to treat with bevacizumab will increase and that treatment might be given even before ETROP criteria are reached. Given that on average five of six babies reaching such criteria will have spontaneous resolution without treatment,23 it is imperative that the treatment is very safe and ‘does no harm’. Even in the absence of systemic side effects, the additional follow-up that may be required following anti-VEGF agents may significantly reduce any purported cost advantage.

WHAT IS THE WAY FORWARD?

By urging that more evidence is required before considering intravitreal bevacizumab as the treatment of choice, even for only some of the more severe forms of ROP, are we denying a therapy that should be used? Many argue that it took too long for antenatal steroids to be universally recommended for threatened preterm delivery.61 62 despite the fact that these agents had been studied on several thousand patients. But neonatology also has its share of therapies being adopted before there was good evidence of the lack of harm, several with disastrous consequences.63 64 Notable examples include the uncontrolled use of oxygen in the 1940s, which led to the first epidemic of ROP, and the higher rates of cerebral palsy in premature infants given the systemic steroids to shorten ventilatory support in the 1980s to 1990s.

Should we change the practice now based on anecdotal evidence and one small randomised controlled trial (RCT), which had a number of design issues and was underpowered to assess safety? We contend the answer to this is clearly ‘no’. The BEAT-ROP study is an important advance, but there is still serious work to be done. This should include additional basic science (further work with animal models), careful examination and follow-up of infants who are currently being treated, including obtaining serum VEGF and bevacizumab concentrations and exploring the pharmacodynamics, and rigorously designed RCTs with masked ocular (posterior pole images) and longer-term outcomes (visual acuity and neurodevelopment). International collaboration is well established in the field of neonatology, making multicentre RCTs enrolling thousands of infants a feasible proposition.65–67

Mintz-Hittner1 reported that 2800 infants would be required to detect a significantly increased death rate in the bevacizumab group compared with laser (1.5 times as high as the 5.4% death rate at least 9 months in the ETROP trial). Although around two-thirds of very preterm infants (VP; gestation <28 weeks) now survive, some 20% of survivors have moderate or severe disability at 2 years.68 In the Australian and New Zealand Neonatal Network in 2006, the mortality for VP infants from 28 days of age until hospital discharge was 6.8%,69 hence a composite figure for late neonatal deaths or moderate/severe disability is 27%. To detect an absolute 10% increase in the composite outcome at 90% power (α one-tailed=0.05), a total of 742 infants would be required. Safety data sought should include specific ocular, neurological, pulmonary, renal or bone complications.

Legitimate questions are what level of certainty of safety is required and what should be done when an unproven therapy is being widely adopted despite the lack of evidence? Even accepting the arguments put forward for not adopting intravitreal bevacizumab as the standard treatment for Zone 1 disease at this time, there clearly is a case for exceptional or compassionate use, such as after failed laser treatment or poor visualisation of the posterior retina. Treatment of AP-ROP with bevacizumab remains controversial and is hampered by a lack of good evidence. In circumstances where treatment with bevacizumab is recommended, parents should understand that such treatment remains experimental and that the long-term outcomes are unknown. We agree with Mintz-Hittner1 that a register of cases and outcomes should be established and suggest that this could be through the auspices of the major neonatal networks around the world, most of which are currently collecting data on the treatment of ROP with bevacizumab.

CONCLUSIONS

We have come a long way on this journey with the goal of arriving at a safe, cheap and simple therapy for acute severe ROP. The BEAT-ROP study is a significant step on the way but it is only one step. It is important that we do not now take a wrong turn that may mean infants are denied a proven effective therapy (laser) which has a good outcome in a majority of cases and are exposed instead to potential but yet unknown harms.

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REFERENCES

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