Published in final edited form as:
*Curr Opin Pediatr*. 2011 April ; 23(2): 173–178. doi:10.1097/MOP.0b013e3283423f35.

# Current update on retinopathy of prematurity: screening and treatment

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## Abstract

**Purpose of review—**Despite current treatments, retinopathy of prematurity (ROP) remains a major cause of blindness in premature infants and the incidence is increasing with increased survival of infants born at very early gestational ages. This review summarizes the recent literature on ROP with a special focus on recent advances in treatment options as well as newly developed methods for disease screening.

**Recent findings—**Genetic studies find a genetic predisposition to ROP linking genes in the Wnt pathway with development of severe ROP. With regard to diagnosis, a new screening method has been developed that allows prediction of ROP risk based on postnatal body weight gain alone. Formerly weight gain postnatally in combination with insulin-like growth factor levels was found to predict treatable ROP. New treatment options for severe cases of ROP have been proposed targeting vascular endothelial growth factor (VEGF). Whether anti-VEGF treatment is safe in preterm infants, however, has to be further evaluated in controlled clinical trials. Finally, new reports from the early treatment ROP group suggest that early laser treatment for type 1 but not type 2 high-risk pre-threshold ROP improves visual acuity outcomes at 6 years of age.

**Summary—**With the increasing survival of premature infants and increased incidence of ROP, it is important to screen for ROP risk and treat at-risk patients in a timely manner to preserve their visual function and reduce complications.

### Keywords

Retinopathy of prematurity; postnatal weight gain; laser photocoagulation; vascular endothelial growth factor; insulin-like growth factor

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## Introduction

Retinopathy of prematurity (ROP) is a major cause of blindness in children in the developing and developed world despite current surgical and laser treatment in the late-stage of the disease[1]. ROP was first connected with premature birth in the 1940s and slightly later to oxygen supplementation in these premature infants[2].Although current ablation treatments can reduce the incidence of blindness by ~25% in infants with late-stage ROP, the patients often still have poor visual acuity after treatment; and the life-long impact of the disease on eye development and vision remains significant. Improved understanding of the ROP disease process and the development of new screening tools to predict ROP much earlier with the possibility of new preventative treatments are highly desirable. This review summarizes the recent advances in ROP studies with a focus on new screening and prediction methods as well as potential new treatment options.

## Pathogenesis of ROP: two phases

ROP is a biphasic disease consisting of an initial phase of vessel growth retardation followed by a second phase of vessel proliferation. Infants born prematurely have incompletely vascularized retinas with a peripheral avascular zone. This first phase of ROP occurs from birth to postmenstrual age approximately 30–32 weeks. As the infant matures, the non-vascularized retina becomes increasingly metabolically active, leading to tissue hypoxia. The second phase of ROP is characterized by hypoxia-induced retinal neovascularization. This second, vaso-proliferative phase begins around 32–34 weeks postmenstrual age. Hypoxia stimulates upregulation of proangiogenic growth factors such as vascular endothelial growth factor (VEGF) and erythropoietin, leading, in severe cases, to uncontrolled vascular growth into the vitreous. Because of the critical role of VEGF in inducing pathologic neovascularization, anti-VEGF treatment has now been suggested by some clinicians as a potential treatment option for severe ROP (see details below).

## Risk factors of ROP

The association between ROP and excessive oxygen was recognized shortly after the initial description of the disease[3, 4]. This led to improved and better-controlled supplemental oxygen protocols to maintain adequate blood levels of oxygen without inducing more hyperoxia to the premature retina than necessary[5]. However, even with monitored oxygen use, the incidence of ROP has increased further, probably due to the increased survival of infants with very low birth weight[6]. Although oxygen use and gestational age/ birth weight are major risk factors for ROP[7–9], other factors reflecting the postnatal changes in the overall health of the baby, such as sepsis, anemia and chronic lung disease are also positively associated with ROP development[10, 11]. Recently postnatal weight gain and insulin-like growth factor 1 (IGF-1) levels, as well as hyperglycemia, were identified as very important predictors for ROP risk, as important as birth weight and gestational age at birth[12–15].

In addition, a recent study identified maternal risk factors associated with ROP development. Wu *et. al.* found in 144 Asian preterm infants that maternal age is a significant risk factor in addition to birth weight[16], suggesting a potentially race-dependent maternal risk factor for ROP that is different from that observed in Western population[17].

Furthermore, recent studies with genetic approaches in monozygotic twins and other clinical and experimental studies suggest a strong genetic predisposition to ROP[18, 19]. Three genes (Norrin, Frizzled 4 and Lrp5) involved in Wnt signaling pathways, a molecular pathway fundamentally important for development and disease[20–22], were found mutated in a small percentage of advanced forms of ROP in several studies[18, 23–29]. These genetic factors,

although they do not account for a substantial number of ROP patients overall, might help explain in part why ROP in some infants progresses to the most severe stage of retinal detachment despite timely intervention whereas others with similar ROP characteristics regress spontaneously.

## ROP screening and prediction

Timely screening of premature infants at risk of developing ROP is important in ROP management as early treatment can result in improved visual outcome. The current screening guideline of ROP in the United States calls for dilated fundus examination by indirect ophthalmoscopy for all premature infants below 30 week gestational age or less than 1500g birth weight with the first examination performed by 31 week postmenstrual age or by 4 weeks chronologic age, with additional examinations performed repeatedly thereafter to detect late stage ROP requiring treatment. Additional screening for older or larger babies is recommended at the discretion of the attending neonatologist. Fortunately, only about 10% of those screened require treatment eventually. This also suggests that there is room for improvement of the current screening protocols. Development of easier and more efficient screening and earlier prediction based on additional clinical criteria could help identify the high risk patients and also identify patients with no or low risk to reduce the number of unnecessary examinations. A major clinical problem in very preterm infants is weight loss and a delay of proper weight gain after premature birth. Poor early weight gain, as well as low serum levels of IGF-I during the first weeks/months after birth have been found to be strongly correlated with the later development of severe ROP[13–15]. These variables have now begun to be used successfully to predict early, the eventual development of severe ROP [30, 31].

In a prospective study, Lofqvist *et. al.* used a surveillance algorithm WINROP (Weight, IGF-I, Neonatal, ROP) to detect infants at risk for proliferative ROP[31]. WINROP is based on weekly measurements of body weight as well as serum IGF-1 levels from birth until postmenstrual age 36 weeks. In a group of 50 preterm infants with average postmenstrual age of 26 weeks, the WINROP algorithm correctly identified all children (100% sensitivity) who were diagnosed with proliferative ROP weeks later, while also successfully identifying infants with low ROP risk. WINROP was then validated, using postnatal weight gain only, in another Swedish population of 354 preterm infants with 100% sensitivity and 84.5% specificity[32]. To validate the same algorithm in a US cohort, Wu *et. al.* evaluated ROP development and weekly weight measurements for 318 US infants[30] and successfully predicted all infants who later developed severe ROP at a median of 9 weeks before ROP diagnosis. None of the infants who were graded as having no or a low ROP risk developed more than mild ROP. These findings suggest following longitudinal postnatal weight gain with WINROP as a useful method to complement the current ROP screening protocols. It might help to identify patients at high risk for closer monitoring, as well as patients at no or low risk to avoid stressful, time consuming, costly and often unnecessary examinations. This algorithm is currently being tested in a large multi-center multinational clinical trial.

The usefulness of IGF-1 in ROP prediction was independently confirmed in a similar prospective study. Pérez-Muñuzuri *et. al.* found in 74 preterm infant from a Spanish population that serum IGF-1 levels at three week post-partum have a 90% sensitivity in ROP prediction[33], suggesting IGF-1 to be a reliable prediction factor independent of gestational age at birth. In a separate study, Pieh *et. al.* recently identified plasma sE-selectin, an adhesion molecule, as a new surrogate marker for ROP that can be used in combination with gestational age to predict ROP [34]. In 42 preterm infants analyzed, plasma sE-selectin levels assessed 2 to 3 weeks after birth were significantly increased in ROP patients and sE-

selectin plasma levels were used successfully to predict ROP development, suggesting sE-selectin as another potentially useful clinical marker for ROP prediction.

## Treatment options

In addition to the research on ROP screening and prediction, significant effort has gone into identification of new and improved treatment options for ROP in order to provide fast resolution of neovessels with minimal complications and maximal preservation of neurosensory structure and function.

The treatment of choice for ROP has long shifted from cryotherapy to peripheral diode laser photocoagulation soon after clinical studies showed that laser therapy is superior to cryotherapy [35, 36]. However, acute risks of laser photocoagulation include corneal edema, intraocular hemorrhage and cataract formation. Recently Parvaresh *et. al.* report in a retrospective study effective outcomes using transscleral diode laser without conjunctival incisions instead of transpupillary laser treatment for threshold ROP[37]. Of 103 treated eyes, 96% showed complete ROP regression and favorable outcomes with minimal adverse effects. The authors conclude that transscleral laser treatment may be technically easier than the transpupillary approach, especially for the treatment of the retinal periphery with possibly fewer anterior segment complications such as cataract formation.

In the last two decades, research in ROP pathogenesis identified VEGF as one of the major angiogenic factors responsible for ROP[38–40]. In several clinical studies, significantly elevated VEGF levels were measured in the vitreous of patients with vasoproliferative ROP [41–43]. Recently Sato *et. al.* assessed the vitreous levels of 27 cytokines in ROP eyes[41] and found VEGF to correlate most strongly with vascularly active ROP. This study also identified several other factors including several members of the interleukin protein family, fibroblast growth factor (FGF) and granulocyte colony-stimulating factor (G-CSF) as well as granulocyte macrophage colony-stimulating factor (GM-CSF) elevated in ROP. The upregulation of these immune regulatory proteins especially for macrophage activation indicates participation of an inflammatory response in the eye that contributes to the complex process of ROP development in addition to known angiogenic growth factors such as VEGF.

Based on the extensive research on VEGF in ROP animal models that show suppression of neovascular disease with VEGF inhibition[39, 40], several smaller clinical treatment trials targeting VEGF-inhibition have been undertaken. In other ocular neovascular diseases, most notably exudative age-related macular degeneration (AMD), anti-VEGF therapy has been used successfully to reduce pathological neovessel formation[44, 45]. Its potential use in diabetic retinopathy is also being currently investigated in clinical trials. With regard to ROP, several reports exist that report on the off-label use of an anti-VEGF monoclonal antibody bevacizumab (Avastin; Genentech Inc, South San Francisco, California, USA) as anti-angiogenic therapy, either used alone or in conjunction with other treatments.

Nonobe *et.al.* assessed concentration of aqueous humor VEGF in ROP eyes after intravitreal injection of bevacizumab[46]. In eight patients with stage 4 or 5 ROP, intravitreal injection of bevacizumab resulted in a marked decrease in the unbound VEGF concentration. Although the patient number is small in this study and a direct comparison before and after injection is not possible, the authors suggest that bevacizumab injection may be useful to reduce the risk of bleeding from neovessels during vitrectomy. In a similar study by Law *et. al.*, 13 eyes of 7 infants were injected with bevacizumab prior to laser therapy or vitrectomy [47]. The authors report that bevacizumab treatment improved intra-operative visualization of the retina without obvious ocular or systemic complications, suggesting that bevacizumab may be a potentially useful adjunct to vitrectomy. However, larger studies are needed to establish

anti-VEGF therapy as a safe compound in ROP (both locally and systemically) before general clinical use can be suggested.

With regard to potential effects of anti-VEGF therapy on neovessel formation in ROP, a recent retrospective study by Lee *et. al.* on 15 premature Korean infants with stage 3 ROP reported regression of plus disease and a more rapid development of the peripheral retinal vascular bed after intravitreal bevacizumab injection combined with laser photocoagulation. The authors report no significant increase in systemic or ocular complications, compared with patients receiving laser treatment alone [48].

In a meta analysis of VEGF therapy in ROP, Micieli *et. al.* analyzed systemic off-label use of bevacizumab in ROP[49] and found considerable variability in dosing, timing, treatment frequency and whether it is used alone or in conjunction with other treatments among the studies analyzed. Overall, considerable concerns remain as to the safety of anti-VEGF treatment in ROP, especially regarding to the correct dosage, timing of injection and potential local complications such as lens damage, infection and adverse effects on retinal neurosensory development. It should also be noted that intraocular injections of these extrinsic factors around term has unknown systemic effects in this population of children with already persistent subnormal growth, impaired development and function of the central nervous system and other tissues. Therefore, randomized control trials following carefully local and systemic effects are needed before reliable statements can be made regarding both the safety and efficacy of bevacizumab treatment in ROP.

In addition to VEGF, erythropoietin (Epo) is another growth factor that is found to promote retinal angiogenesis similarly to VEGF *in vitro* and in animal studies[50–52]. Recombinant Epo is known to promote red blood cell formation and is used to treat anemia in premature infants in a few centers. Sato *et. al.* examined 40 eyes from 27 infants with stage 4 ROP and found that Epo levels were significantly elevated in the vitreous from infants with vascularly active ROP in correlation with VEGF[43], suggesting not only VEGF but also Epo as a contributer to ROP. Suk *et. al.* examined the relationship between rhEPO treatment and ROP in 264 patients in a retrospective study (that did not take into account phase of ROP). The authors identified both increased dose and later starting age of rhEpo treatment are significant risk factors of ROP. However, in another retrospective study of 85 preterm infants, Shah *et. al.* found no significant correlation between Epo treatment and ROP incidence and severity[53]. At present, further investigation is needed to determine the role of Epo in Phase I and Phase II in ROP development.

Recently numerous breakthroughs in angiogenesis research also suggest a number of new ways to potentially intervene in ROP progression such as targeting the IGF-1 pathway[13, 40, 54, 55] and dietary supplementation with omega-3 polyunsaturated fatty acids (PUFA)[56]. Serum IGF-1 is substantially reduced after preterm birth[57], due to interruption of the maternal/fetal interaction. In animal models of ROP, IGF-1 is essential for vascular growth through regulation of VEGF signaling[40, 55]. Therefore, supplementing IGF-1 in phase I of ROP would hypothetically normalize vascular growth and prevent phase I and prevent abnormal vascular proliferation in phase II. A phase I study administering IGF-I to preterm infants have been preformed[58] and a clinical trial is currently underway to investigate the possibility of preventing ROP in premature infant by restoring IGF-1 to the levels found *in utero*[59].

More recently the role of omega-3 and omega-6 essential PUFA was evaluated in ROP animal models. Dietary omega-3 PUFAs protect against pathologic neovascularization in ROP[56, 60]. Western diets are often deficient in omega-3 PUFAs and premature infant lack the important transfer of omega-3 PUFAs from their mother in the third trimester.

Supplementing omega-3 PUFAs intake to premature infants may therefore be of benefit in preventing retinopathy if additional research supports the observations found in animal studies. Currently a trial is in the planning stage to supplement premature infants with omega-3 PUFAs either via diet or total parenteral nutrition (TPN) which at present contains only omega-6 but no omega-3 PUFAs.

At this point, laser photocoagulation remains the only well-established therapies for severe cases of neovascular ROP. Whether medical approaches will complement these surgical strategies in the future remains an exciting topic to observe over the coming years.

## ROP outcome

The potential visual and developmental impact of ROP requires lifelong follow-up of affected patients. Although the complications are more prevalent in infants with severe stages of ROP that required treatment, continued follow-ups are also recommended for children with mild or moderate disease that regresses spontaneously. Myopia, for example, is more prevalent in premature infants, affecting approximately 70% of infants, with ROP, during the first year after birth[61]. Similarly, the incidence of strabismus is significantly increased in ROP infants affecting up to 20%, of patients. Finally, astigmatic refractive errors are increased in ROP patients affecting up to 40% of eyes with a history of ROP[61].

Recently the Early Treatment For ROP Study published its final visual acuity results at 6 years of age, comparing eyes that received early treatment for high-risk prethreshold ROP with conventionally managed eyes[62]. The group found early treatment for Type 1 but not Type 2 high-risk prethreshold eyes improved visual acuity at 6 years of age, suggesting in clinical practice Type 1 eyes, but not Type 2 eyes, should be treated early. Early-treated eyes also showed a significantly better structural outcome compared to conventionally managed eyes, with no greater risk of ocular complications. These results are particularly important considering that 52% of Type 2 high-risk prethreshold eyes underwent regression of ROP without requiring treatment. Nevertheless, Type 2 ROP eyes still require close examination, even if early treatment might not be as pressing as in Type 1 eyes. Christiansen *et. al.* found that approximately 22% of Type 2 ROP eye progress to Type 1 ROP. The risk of progression was greatest between 33 and 36 weeks' postmenstrual age, indicating the requirement for thorough examinations throughout this period[63].

## Conclusion

Improved survival of very premature infants has resulted in an increasing number of babies with ROP requiring close screening and potential treatment. Newly developed screening and prediction methods for ROP will likely allow fewer stressful examinations and more cost-effective screening as well as early identification of high risk patients. The current treatment of laser ablation therapy has limitations with regard to acute and long term complications. However, novel treatment approaches, suppressing the neovascularization of phase II, like anti-VEGF therapies have not yet been sufficiently evaluated to be broadly recommended for clinical treatment. Nevertheless, the ongoing studies investigating the safety and efficacy of anti-VEGF therapies for ROP treatment might provide valuable treatment options in the future. To prevent the vessel loss of phase I, other emerging treatments targeting the IGF-1 pathway[13, 40, 54, 55] as well as dietary supplementation with omega-3 polyunsaturated fatty acids[56], both of which are deficient in preterm infants, might provide further benefits. These treatments aim for prevention of ROP by promoting the normal vascular growth of an *in utero* environment by normalizing factors missing from the third trimester after preterm birth. In general, improving functional vascularization of the avascular parts of the postnatal retina without inducing pathologic neovessel formation would be a very appealing approach

to treat ROP since the extent of the second destructive phase of ROP is determined by the amount of avascular retina in phase I. Our increasing understanding of ROP pathogenesis also illustrates that timing is critical in any medical or surgical intervention, since the two phases of ROP require very different approaches. Finally, it has to be noted that in the fragile neonate, the advantages and risks of any intervention must be weighed very carefully, with both disease progression and treatment effects monitored very closely and much more frequently than in adult patients, due to the rapid developmental changes in these infants.

**Key points**

- Newly developed ROP screening and prediction methods based on post-natal weight gain and IGF-1 levels can successfully predict infants who are at high risk for ROP. These infants may be monitored more closely while those identified to be at low risk may be spared unnecessary diagnostic procedures.
- The safety (both locally and systemically) and efficacy of anti-VEGF therapy in ROP still awaits clarification from randomized clinical trials which are currently underway.
- Early treatment of Type 1 ROP is beneficial with regard to long-term visual outcome. No improvement of visual acuity was found for Type 2 ROP with early treatment.
- Further research on angiogenesis and ROP may shed light on potential future therapies using IGF-1 replacement and omega-3 PUFA supplement in a timely coordinated fashion.

## Acknowledgments

This work was supported in part by the: NIH EY017017, V. Kann Rasmussen Foundation, Roche Foundation for Anemia Research, MacTel Foundation (LEHS) and Juvenile Diabetes Research Foundation International (JC).

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