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Emerging Treatments for Retinopathy of Prematurity

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Abstract

Retinopathy or prematurity (ROP) is a leading cause of potentially preventable blindness in children. With increased survival of infants born at earlier gestational ages the number of infants at risk from vision loss from ROP has increased. Current treatments consist of close monitoring of oxygen saturation levels, peripheral retinal ablation by cryotherapy or laser photocoagulation, and vitreoretinal surgery. Research in the area of angiogenesis has lead to numerous breakthroughs. Emerging treatments for ROP are targeting the Vascular Endothelial Growth Factor (VEGF) and Insulin-Like Growth Factor 1 (IGF-1) pathways, as well as dietary supplementation with omega-3-polyunsaturated fatty acids.

Keywords

retinopathy of prematurity; ROP; vascular endothelial growth factor; insulin-like growth factor 1; VEGF; IGF-1; omega-3-polyunsaturated fatty acids; polyunsaturated fatty acids

INTRODUCTION

Retinopathy of prematurity (ROP) is a leading cause of potentially preventable blindness in children, especially in developed and emerging countries. Originally described by Terry in 1942,¹ its association with excessive oxygen use at that time was noted soon after. With increased survival of infants born at earlier gestational ages the number of infants at risk for vision loss from ROP has increased over the years.

RISK FACTORS

Major risk factors for the development of ROP are birth weight and gestational age.^{2,3} Supplemental oxygen treatment has also been identified as a major modifiable risk factor

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Declaration of Interest

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

and as a result of close oxygen monitoring and titration of oxygen levels the incidence of vision loss from ROP has decreased. Other known risk factors for ROP that reflect the overall state of the infant's health include sepsis, hypercarbia, hypocarbia, anemia, chronic lung disease, bradycardia, and intraventricular hemorrhages.^{4–8} It has also been established recently that postnatal IGF-1 levels and postnatal growth (weight gain) are important risk factors for the development of ROP and can be used to predict more specifically which infants will develop ROP.⁹

EPIDEMIOLOGY

From January 1986 until November 1987 the CRYO-ROP study evaluated 4099 babies with birth weight less than 1250 g, which was thought to represent 15% of babies in the United States with that birth weight. Patients underwent serial eye examinations and 65.8% of them developed ROP. ROP was negatively correlated with birth weight and gestational age. Most ROP regressed without causing significant ocular or visual sequelae. However, 6% of infants with birth weight less than 1250 g developed threshold disease, which was defined as stage 3 ROP with plus disease (dilation and tortuosity of posterior pole retinal vessels) in zone I or II, involving at least five contiguous clock-hour sectors or at least eight interrupted clock-hour sectors.^{10,11}

In the Early Treatment for Retinopathy of Prematurity (ETROP) study, infants who were identified to have prethreshold ROP were risk stratified to high- or low-risk prethreshold ROP by using the RM-ROP2 risk model. Prethreshold ROP was defined as any ROP in zone I that was less than threshold; or zone II, stage 2 with plus disease, or zone II, stage 3 disease without plus disease, or stage 3 with plus disease but fewer than the required clock hours for threshold disease. Infants with high-risk prethreshold ROP were randomized to early retinal ablative treatment or conventional timing of treatment at threshold. Evaluation of high-risk prethreshold eyes that were randomized to conventional management demonstrated that zone I, stage 1 or 2 without plus disease eyes, as well as zone II, stage 3 without plus disease eyes, had lower rates of progressing to threshold or unfavorable outcomes than the other eyes in the group. This led to the development of a clinical algorithm in which treatment is to be considered for “Type 1” eyes with zone I, any stage ROP with plus disease; eyes with zone I, stage 3 ROP without plus disease; and eyes with zone II, stage 2 or 3 with plus disease. Using this early clinical treatment algorithm 8% of infants weighing less than 1,251 g at birth would require treatment.¹²

CURRENT TREATMENTS

Treatment of ROP today consist of close monitoring of oxygen saturation levels, peripheral retinal ablation by cryotherapy or laser photocoagulation, and vitreoretinal surgery for retinal detachment repair.¹³

Numerous studies have looked at the effects of oxygen saturation levels and ROP.^{14,15} The earliest studies showed that neonates developed less ROP if supplemental oxygen was restricted; unfortunately, this was extrapolated and inappropriately lead to a widespread decrease in oxygen supplementation in neonatal units. Even though there was a significant reduction in blindness from ROP, neurological complications and mortality from oxygen

restriction increased.^{15–20} Since then, other studies have shown modest reductions in oxygen supplementation and control of fluctuations by reducing oxygen saturation alarm limits can result in improved outcomes from ROP and chronic lung disease without significant adverse effects.^{21–24} However, there has been no study to date that establishes the best oxygen level to balance ROP development and adequate brain oxygenation, and guidelines amongst Neonatal Intensive Care Units vary widely.²² Increasing supplemental oxygen for infants with prethreshold ROP was not found to provide significant benefit in reducing progression to threshold in the STOP-ROP trial,²⁵ though trends for reduced progression to threshold were seen for infants in the HOPE-ROP and BOOST trials.^{26,27}

No proven benefit from vitamin E supplementation or light reduction has been shown.^{28–30}

Peripheral retinal ablation by cryotherapy was confirmed to significantly decrease unfavorable outcomes from ROP in the CRYO-ROP study, and later studies have shown that laser ablation is similarly effective as cryotherapy. In the CRYO-ROP study treatment with cryotherapy at threshold ROP was noted to decrease the number of infants with vision 20/200 or worse by age 3.5 years by 26% but did not improve the chances of good visual acuity (>20/40) in these patients.^{10,11} In the ETROP study visual acuity was the primary outcome, but structural outcomes were also evaluated. Unfavorable visual outcomes were reduced from 19.8% to 14.3% with earlier ablative treatment, and unfavorable structural outcomes (posterior retinal folds involving the macula, a retinal detachment involving the macula, or a retrolental tissue or “mass” obscuring the view of the posterior pole) were reduced from 15.6% to 9.0%. Additional sub-group analysis supported a “wait and watch” approach to type II ROP (zone I, stage 1 and 2 without plus disease, or zone II, stage 3 without plus disease).¹² Currently laser photocoagulation is the treatment of choice.

In cases where ROP progresses, resulting in vitreous traction and retinal detachment, scleral buckling and/or vitrectomy are often offered. However, the visual outcomes of these patients are generally poor even in cases of successful retinal reattachment.

Complications of ROP include vision loss, retinal folds, dragging of the macula, retinal tears and detachments, iris neovascularization, glaucoma, high myopia, photoreceptor dysfunction, visual field loss.^{31–35}

EMERGING TREATMENTS

Vascular Endothelial Growth Factor (VEGF)

ROP pathogenesis is based on the fact that retinal vascularization is incomplete in premature infants and the normal retinal vascular maturation is disrupted [refer to Heidary et al. in this issue]³⁶ and numerous agents have been implicated in the modulation of these pathways. ROP includes 2 phases, with I: cessation of normal retinal vascular growth and vasoobliteration, and II: retinal neovascularization.^{3,36–39} The extent of phase II ROP is determined by the extent of phase I ROP. Current understanding of ROP pathogenesis is that hypoxia is a major driving force for Phase II ROP by modulating the expression of vascular endothelial growth factor (VEGF),^{39–41} and experimental animal models have shown that hypoxia stimulates VEGF expression prior to neovascularization. Elevated levels of VEGF

have been found in vitreous samples from patients with retinal neovascularization, and VEGF inhibition has been used as a treatment for ocular neovascular disease, including neovascular age related macular degeneration, diabetic retinopathy, and neovascular glaucoma.⁴²

FDA-approved VEGF inhibitors for intraocular use include Macugen® (pegaptanib sodium) and Lucentis® (ranibizumab). Avastin® (bevacizumab) has also been used off label for intraocular injection with similar results. None of these have been approved for pediatric use. However, there have been case reports describing the use of VEGF inhibitors in patients with ROP who continued to have aggressive disease with neovascularization of the iris (NVI), anterior fibrovascular proliferation despite laser treatment,⁴³ or could not be treated with laser photocoagulation because of their poor clinical status.⁴⁴ In these case reports there was regression of the fibrovascular proliferation, the NVI, and the ROP, without adverse systemic or ocular side effects being noted. Mintz-Hittner and Kuffel (2008) studied the use of bevacizumab in the treatment of moderate and severe stage 3 ROP in eyes that had never received laser treatment. 11 infants (22 eyes) received bevacizumab intravitreal injections and were followed up for a mean period of 48.5 weeks. All eyes were considered to have been successfully treated after 1 injection, without the development of retinal detachment, macular ectopia, high myopia or anisometropia, and no systemic or ocular complications were encountered.⁴⁵

It seems apparent that modulation of VEGF levels in the premature retina has a role in the treatment of ROP, and an 11 center randomized controlled trial is underway to carefully evaluate ocular outcomes, ocular and systemic adverse events, and to establish a safe dose for the treatment of ROP.⁴⁶ As with any new treatment during this fragile period of newborn development it will be important to closely follow these infants to evaluate for potential long-term neurological and ophthalmic consequences, as well as to compare visual outcomes with current laser photocoagulation treatments.

Insulin-Like Growth Factor 1 (IGF-1)

IGF-1 may be an additional target for prevention of ROP. During in utero development IGF-1 is provided by the placenta and the amniotic fluid; after birth IGF-1 levels fall precipitously.^{47,48} It has been postulated that interaction of IGF-1 and VEGF controls proliferation of blood vessels in infants, as a minimum level of IGF-1 is required for VEGF signaling. Therefore premature birth leads to early suppression of VEGF and decreasing levels of IGF-1, but increasing levels of VEGF during the proliferative phase of ROP along with rising levels of IGF-1 in the maturing infant further promotes abnormal neovascularization.^{3,47-49}

However, if IGF-1 levels can better match VEGF levels and allow normal vascular growth by supplementation in phase I then the effect of abnormal neovascularization in phase II may be reduced. It has been established that low IGF-1 is associated with the degree of ROP.^{48,49} A clinical trial is currently underway to restore IGF-1 levels to those found in utero in premature infants to see if this will prevent ROP.

Polyunsaturated Fatty Acids (PUFA)

In addition to the role of IGF-1 and VEGF in the development of retinopathy of prematurity, more recently the role of omega-3- and omega-6-polyunsaturated fatty acids in angiogenesis is starting to be evaluated.^{50,51} The major retinal PUFA are docosahexaenoic acid (DHA) and arachidonic acid; both are found in neural and vascular cell membrane phospholipids.⁵² In addition, eicosapentaenoic acid (EPA), which is the DHA precursor, is found in the retinal vascular endothelium.⁵³ Bioactive intermediaries are produced from PUFA; these include eicosanoids from arachidonic acid, neuroprotectins from DHA, D series resolvins from DHA, and E series resolvins from EPA.^{54–56}

Recently the relationship of PUFA and oxygen-induced retinopathy was evaluated using a transgenic mouse model in which overexpression of the *C. elegans fat-1* gene converts omega-6-PUFA to omega-3-PUFA, resulting in elevated tissue levels of omega-3-PUFA.^{57,58} The effect on oxygen-induced retinopathy in mice by dietary supplementation of omega-3-PUFA was also studied.⁵⁸ It was found that both the transgenic *Fat-1* mice and that received diets rich in omega-3-PUFA had elevated tissue levels of omega-3-PUFA, including EPA, docosapentaenoic acid (DPA)-omega-3 and (DHA). More importantly, it was noted that a lower omega-6/omega-3-PUFA ratio was protective against pathologic angiogenesis. *Fat-1* mice also showed increased retinal omega-3-PUFA levels and inhibited neovascularization. Similar protection from vasoobliteration and neovascularization was noted when mice without omega-3-PUFA supplementation received low intraperitoneal doses of resolvinD1 (RvD1), resolvinE1 (RvE1) or neuroprotectinD1 (NPD1). RvD1, RvE1 and NPD1 had been shown to be present in retinas of mice that had received omega-3-PUFA diets, but not in mice that had received omega-6-PUFA diets. The protective effect was noted to be secondary to increased regrowth of vessels and not due to decreased oxygen-induced vessel loss during hyperoxia.⁵⁸ Western diets are often deficient in omega-3-PUFA, and premature infants lack the important transfer from the mother to the infant of omega-3-PUFA that normally occurs in the third trimester of pregnancy.⁵⁹

Supplementing omega-3-PUFA intake may be of benefit in preventing retinopathy. Prenatal vitamin supplementation is considered standard of care in the United States and expectant mothers already receive multivitamin supplementation. Since 1998 the United States Food and Drug Administration has mandated that enriched cereals and grains have folic acid fortification. Since the mandatory folic acid fortification the incidence of spina bifida has decreased 21–34%.^{60–62} If additional research supports decrease of retinopathy of prematurity adverse outcomes by omega-3-PUFA supplementation an approach similar to that of folic acid prenatal supplementation can be undertaken. However, further supplementation after birth of the premature infants is also needed. At present total parenteral nutrition (TPN) given to these infants contains only omega-6-PUFA with no omega-3-PUFA. A trial is in the planning stages to supplement infants who have no other source of these essential lipids with omega-3-PUFA either via diet or TPN.

SUMMARY

In summary, the area of angiogenesis and pathogenesis of prematurity is under intense investigation and numerous breakthroughs have occurred in recent years with important

therapeutic implications. Current treatments of retinal cryotherapy and laser photocoagulation have limitations and even after successful treatment patients can still have long-term effects from ROP and/or the ROP treatment. New treatment modalities are underway; currently the most promising are therapies that target the VEGF/IGF1 pathway, as well as dietary supplementation with omega-3-PUFA. The ideal treatment for ROP would create an environment as similar as that of the maternal uterus and allow for normal retinal vascular maturation to occur.

REFERENCES

1. Terry TL. Retrolental fibroplasia in the premature infant: V. Further studies on fibroplastic overgrowth of the persistent tunica vasculosa lentis. *Trans Am Ophthalmol Soc.* 1944; 44:383–396.
2. Luty GA, Chan-Ling T, Phelps DL, et al. Proceedings of the third international symposium on retinopathy of prematurity: An update on ROP from the lab to the nursery (November 2003, Anaheim, California). *Mol Vis.* 2006; 12:532–580. [PubMed: 16735995]
3. Smith LE. Pathogenesis of retinopathy of prematurity. *Semin Neonatol.* 2003; 8:469–473. [PubMed: 15001119]
4. Tasman W, Patz A, McNamara JA, et al. Retinopathy of prematurity: The life of lifetime disease. *Am J Ophthalmol.* 2006; 141:167–174. [PubMed: 16386993]
5. Campbell K. Intensive oxygen therapy as a possible cause of retrolental fibroplasia; A clinical approach. *Med J Aust.* 1951; 2:48–50. [PubMed: 14874698]
6. Patz A, Hoeck LE, De La Cruz E. Studies on the effect of high oxygen administration in retrolental fibroplasia. I. Nursery observations. *Am J Ophthalmol.* 1952; 35:1248–1253. [PubMed: 12976495]
7. Ashton N, Ward B, Serpell G. Role of oxygen in the genesis of retrolental fibroplasia; A preliminary report. *Br J Ophthalmol.* 1953; 37:513–520. [PubMed: 13081949]
8. Kinsey VE, Arnold HJ, Kalina RE, et al. PaO₂ levels and retrolental fibroplasia: A report of the cooperative study. *Pediatrics.* 1977; 60:655–668. [PubMed: 578921]
9. Lofqvist C, Andersson E, Sigurdsson J, et al. Longitudinal postnatal weight and insulin-like growth factor I measurements in the prediction of retinopathy of prematurity. *Arch Ophthalmol.* 2006; 124:1711–1718. [PubMed: 17159030]
10. The Cryotherapy for Retinopathy of Prematurity Cooperative Group. Incidence and early course of retinopathy of prematurity. *Ophthalmology.* 1991; 98:1628–1640. [PubMed: 1800923]
11. The Cryotherapy for Retinopathy of Prematurity Cooperative Group. Multicenter Trial of Cryotherapy for Retinopathy of Prematurity: One-year outcome—Structure and function. *Arch Ophthalmol.* 1990; 108:1408–1416. [PubMed: 2222274]
12. Good WV. Early Treatment for Retinopathy of Prematurity Cooperative Group. Final results of the Early Treatment for Retinopathy of Prematurity (ETROP) randomized trial. *Trans Am Ophthalmol Soc.* 2004; 102:233–250. [PubMed: 15747762]
13. Hubbard GB 3rd. Surgical management of retinopathy of prematurity. *Curr Opin Ophthalmol.* 2008; 19:384–390. [PubMed: 18772670]
14. Silverman, WA.; Flynn, JT. Contemporary Issues in Fetal and Neonatal Medicine. Retinopathy of Prematurity. Boston: Blackwell Scientific; 1985.
15. Silverman WA. A cautionary tale about supplemental oxygen: The albatross of neonatal medicine. *Pediatrics.* 2004; 113:394–396. [PubMed: 14754955]
16. Kinsey VE. Retrolental fibroplasia; Cooperative study of retrolental fibroplasia and the use of oxygen. *AMA Arch Ophthalmol.* 1956; 56:481–543. [PubMed: 13361620]
17. Avery ME. Recent increase in mortality from hyaline disease. *J Pediatr.* 1960; 57:553–559. [PubMed: 13685272]
18. McDonald AD. The aetiology of spastic diplegia. A synthesis epidemiological and pathological evidence. *Dev Neurol.* 1964; 11:277–285.
19. Cross KW. Cost of preventing retrolental fibroplasia? *Lancet.* 1973; 2:954–956. [PubMed: 4126572]

20. Bolton DP, Cross KW. Further observations on cost preventing retrolental fibroplasia. *Lancet*. 1974; 1:445–448. [PubMed: 4131442]
21. Tin W, Milligan DW, Pennefather P, Hey E. Pulse oximetry, severe retinopathy, and outcome at one year in babies of less than 28 weeks gestation. *Arch Dis Child Fetal Neonatal Ed*. 2001; 84:F106–F110. [PubMed: 11207226]
22. Anderson CG, Benitz WE, Madan A. Retinopathy of prematurity and pulse oximetry: A national survey of recent practices. *J Perinatol*. 2004; 24:164–168. [PubMed: 14999216]
23. Sun SC. Relation of target SpO₂ levels and clinical outcome in ELBW infants on supplemental oxygen. *Pediatr Res*. 2002; 51:350A.
24. CSMC Oxygen Administration Study Group. Can changes in clinical practice decrease the incidence of severe retinopathy of prematurity in very low birth weight infants? *Pediatrics*. 2003; 111:339–345. [PubMed: 12563061]
25. The STOP-ROP, Multicenter Study Group. Supplemental therapeutic oxygen for prethreshold retinopathy of prematurity (STOP-ROP), a randomized, controlled trial. I: Primary outcomes. *Pediatrics*. 2000; 105:295–310. [PubMed: 10654946]
26. HOPE-ROP Multicenter Group. Retinopathy of prematurity outcome in infants with prethreshold retinopathy of prematurity and oxygen saturation >94% in room air: The high oxygen percentage in retinopathy of prematurity study. *Pediatrics*. 2002; 110:540–544. [PubMed: 12205257]
27. Askie LM, Henderson-Smart DJ, Irwig L, Simpson JM. Oxygen-saturation targets and outcomes in extremely preterm infants. *N Engl J Med*. 2003; 349:959–967. [PubMed: 12954744]
28. Phelps DL, Rosenbaum AL, Isenberg SL, et al. Tocopherol efficacy and safety for preventing retinopathy of prematurity: A randomized, controlled, double-masked trial. *Pediatrics*. 1987; 79:489–500. [PubMed: 3547300]
29. Schaffer DB, Johnson L, Quinn GE, et al. Vitamin E and retinopathy of prematurity. Follow up at 1 year. *Ophthalmology*. 1985; 92:1005–1011. [PubMed: 3900846]
30. Reynold JD, Hardy RJ, Kennedy KA, et al. Lack of efficacy of light reduction in preventing retinopathy of prematurity. *N Engl J Med*. 1998; 338:1572–1576. [PubMed: 9603794]
31. Choi MY, Park IK, Yu YS. Long term refractive outcome in eyes of preterm infants with and without retinopathy of prematurity: Comparison of keratometric value, axial length, anterior chamber depth, and lens thickness. *Br J Ophthalmol*. 2000; 84:138–143. [PubMed: 10655187]
32. Knight-Nanan DM, Algawi K, Bowell R, O'Keefe M. Advanced cicatricial retinopathy of prematurity-outcome and complications. *Br J Ophthalmol*. 1996; 80:343–345. [PubMed: 8703887]
33. Maly E. Frequency and natural history of retinopathy of prematurity (ROP). A prospective study in a Swedish city 1986–1990. *Acta Ophthalmol Suppl*. 1993; 210:52–55. [PubMed: 8329955]
34. Kaiser RS, Trese MT, Williams GA, Cox MS Jr. Adult retinopathy of prematurity: Outcomes of rhegmatogenous retinal detachments and retinal tears. *Ophthalmology*. 2001; 108:1647–1653. [PubMed: 11535465]
35. Sneed SR, Pulido JS, Blodi CF, et al. Surgical management of late-onset retinal detachments associated with regressed retinopathy of prematurity. *Ophthalmology*. 1990; 97:179–183. [PubMed: 2326006]
36. Ashton N, Ward B, Serpell G. Effect of oxygen on developing retinal vessels with particular reference to the problem of retrolental fibroplasia. *Br J Ophthalmol*. 1954; 38:397–432. [PubMed: 13172417]
37. McLeod DS, Brownstein R, Luty GA. Vaso-obliteration in the canine model of oxygen-induced retinopathy. *Invest Ophthalmol Vis Sci*. 1996; 37:300–311. [PubMed: 8603834]
38. Smith LE, Wesolowski E, McLellan A, et al. Oxygen-induced retinopathy mouse. *Invest Ophthalmol Vis Sci*. 1994; 35:101–111. [PubMed: 7507904]
39. Donahue ML, Phelps DL, Watkins RH, et al. Retinal vascular endothelial growth factor (VEGF) mRNA expression is altered in relation to neovascularization in oxygen induced retinopathy. *Curr Eye Res*. 1996; 15:175–184. [PubMed: 8670726]
40. Stone J, Chan-Ling T, Pe'er J, et al. Roles of vascular endothelial growth factor and astrocyte degeneration in the genesis of retinopathy of prematurity. *Invest Ophthalmol Vis Sci*. 1996; 37:290–299. [PubMed: 8603833]

41. Miller JW, Adamis AP, Shima DT, et al. Vascular endothelial growth factor/vascular permeability factor is temporally and spatially correlated with ocular angiogenesis in a primate model. *Am J Pathol.* 1994; 145:574–584. [PubMed: 7521577]
42. Andreoli CM, Miller JW. Anti-vascular endothelial growth factor therapy for ocular neovascular disease. *Curr Opin Ophthalmol.* 2007; 18:502–508. [PubMed: 18163003]
43. Shah PK, Narendran V, Tawansy KA, et al. Intravitreal bevacizumab (Avastin) for post laser anterior segment ischemia in aggressive posterior retinopathy of prematurity. *Indian J Ophthalmol.* 2007; 55:75–76. [PubMed: 17189897]
44. Kong L, Mintz-Hittner HA, Penland RL, et al. Intravitreal bevacizumab as anti-vascular endothelial growth factor therapy for retinopathy of prematurity: A morphologic study. *Arch Ophthalmol.* 2008; 126:1161–1163. [PubMed: 18695118]
45. 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. [PubMed: 18536599]
46. Sears JE. Anti-vascular endothelial growth factor and retinopathy of prematurity. *Br J Ophthalmol.* 2008; 92:14-37–14-38.
47. Smith LE, Shen W, Perruzzi C, et al. Regulation of vascular endothelial growth factor-dependent retinal neovascularization by insulin-like growth factor-1 receptor. *Nat Med.* 1999; 5:1390–1395. [PubMed: 10581081]
48. Hellstrom A, Perruzzi C, Ju M, et al. Low IGF-I suppresses VEGF survival signaling in retinal endothelial cells: Direct correlation with clinical retinopathy of prematurity. *Proc Natl Acad Sci USA.* 2001; 98:5804–5808. [PubMed: 11331770]
49. Hellstrom A, Engstrom E, Hard AL, et al. Postnatal serum insulin-like growth factor I deficiency is associated with retinopathy of prematurity and other complications of premature birth. *Pediatrics.* 2003; 112:1016–1020. [PubMed: 14595040]
50. Kermorvant-Duchemin E, Sennlaub F, Sirinyan M, et al. Trans-arachidonic acids generated during nitrate stress induce a thrombospondin-1-dependent microvascular degeneration. *Nat Med.* 2005; 11:1339–1345. [PubMed: 16311602]
51. Fierro IM, Kutok JL, Serhan CN. Novel lipid mediator regulators of endothelial cell proliferation and migration: Aspirin-triggered-15R-lipoxin A(4) and lipoxin A(4). *J Pharmacol Exp Ther.* 2002; 300:385–392. [PubMed: 11805195]
52. Fliesler SJ, Anderson RE. Chemistry and metabolism of lipids in the vertebrate retina. *Prog Lipid Res.* 1983; 22:79–131. [PubMed: 6348799]
53. SanGiovanni JP, Chew EY. The role of omega-3 long-chain polyunsaturated fatty acids in health and disease of the retina. *Prog Retin Eye Res.* 2005; 24:87–138. [PubMed: 15555528]
54. Calder PC. Polyunsaturated fatty acids and inflammation. *Prostaglandins Leukot Essent Fatty Acids.* 2006; 75:197–202. [PubMed: 16828270]
55. Salem N Jr, Litman B, Kim HY, Gawrisch K. Mechanisms of action of docosahexaenoic acid in the nervous system. *Lipids.* 2001; 36:945–959. [PubMed: 11724467]
56. Serhan CN, Savill J. Resolution of inflammation: The beginning programs the end. *Nat Immunol.* 2005; 6:1191–1197. [PubMed: 16369558]
57. Kang JX, Wang J, Wu L, Kang ZB. Transgenic mice: Fat-1 mice convert n-6 to n-3 fatty acids. *Nature.* 2004; 427:504. [PubMed: 14765186]
58. Connor KM, SanGiovanni JP, Lofqvist C, et al. Increased dietary intake of ω -3-polyunsaturated fatty acids reduces pathological retinal angiogenesis. *Nat Med.* 2007; 13:868–873. [PubMed: 17589522]
59. Crawford MA, Costeloe K, Ghebremeskel K, et al. Are deficits of arachidonic and docosahexaenoic acids responsible for the neural and vascular complications of preterm babies? *Am J Clin Nutr.* 1997; 66:1032S–1041S. [PubMed: 9322584]
60. Honein MA, Paulozzi LJ, Mathews TJ, et al. Impact of folic acid fortification of the US food supply on the occurrence of neural tube defects. *JAMA.* 2001; 285:2981–2986. [PubMed: 11410096]

61. Williams LJ, Mai CT, Edmonds LD, et al. Prevalence of spina bifida and anencephaly during the transition to mandatory folic acid fortification in the United States. *Teratology*. 2002; 66:33–39. [PubMed: 12115778]
62. Canfield MA, Collins JS, Botto LD, et al. Changes in the birth prevalence of selected birth defects after grain fortification with folic acid in the United States: Findings from a multi-state population-based study. *Birth Defects Res A Clin Mol Teratol*. 2005; 73:679–689. [PubMed: 16240378]