Published in final edited form as:

Int Ophthalmol Clin. 2011 ; 51(1): 77–91. doi:10.1097/IIO.0b013e3182010f29.

Current Update on Retinoblastoma

Samuel K. Houston, MD,

Bascom Palmer Eye Institute, University of Miami, Miami, FL

Timothy G. Murray, MD, MBA, FACS,

Bascom Palmer Eye Institute, University of Miami, Miami, FL

Stacey Quintero Wolfe, MD, and

Department of Neurological Surgery, University of Miami/Jackson Memorial Hospital, Miami, FL

Cristina E. Fernandes, MD

Department of Pediatrics, University of Miami, Miami, FL

Introduction

Treatment of retinoblastoma has undergone significant advancements over the past few decades, as globe-salvaging therapies with chemoreduction and focal consolidation are favored over external beam radiation and enucleation. With current chemotherapy protocols and focal laser, survival in the United States and other developed countries has now climbed to almost 100% with many children maintaining functional vision. However, in other continents such as Africa, survival rates drop significantly.^{1, 2} Additionally, children with advanced tumors often require enucleation. Novel therapeutic directions are actively being pursued for these advanced retinoblastoma tumors, as well as development of treatments that decrease associated risks of systemic chemotherapy. Additionally, there are a number of experimental adjuvant therapies that have been shown to be efficacious in animal models, and may be part of the future armamentarium of cancer treatment. Finally, novel imaging modalities are being developed that may aid in the diagnosis and management of retinoblastoma in the future. The following review discusses current updates on retinoblastoma.

Genetics

Retinoblastoma tumors can be either heritable and associated with a germline mutation of the RB1 gene, or non-heritable. Heritable mutations typically present in the 1st year of life with bilateral disease. In comparison, the non-heritable form typically presents slightly later and is primarily unilateral. In 1971, Knudson proposed his “two-hit” hypothesis for the development of retinoblastoma tumors.³ Following his seminal work, the retinoblastoma gene was the first tumor suppressor gene discovered in the human genome.⁴ Over the decades, the RB gene has been shown to be a key regulator of the cell cycle through inhibitory effects on the transcription factor elongation factor-2 (EF2) and cellular differentiation through effects on tissue-specific transcription factors. Recent evidence suggests a decision-making role for the RB gene, mediating proliferation, apoptosis, and differentiation.⁵

Knudson's "two-hit" hypothesis has recently been challenged, as it has been proposed that the previously held belief of RB gene mutation leading to retinoblastoma tumors simplifies a complex genetic process, with genomic instability and aneuploidy more likely responsible.⁶ Using microarray analysis, other investigators analyzed the genomic expression of human retinoblastoma tumors. They found that 1004 genes were upregulated and 481 genes were downregulated compared to control eyes with clusters of differentially expressed genes identified on chromosomes 1, 6, and 17.⁷ Other investigators have shown that in unilateral, sporadic tumors may be subclassified according to the presence or absence of loss of heterozygosity on chromosome 13, as well as novel regions of amplification or loss on a number of other chromosomes.⁸

More recently, using the LH_{BETA}T_{AG} retinoblastoma murine model, microarray analysis showed that regional and temporal variations in genomic expression were evident in retinoblastoma tumors. More advanced tumors showed dysregulated genes involved in networks involved in angiogenesis, hypoxia, and cellular metabolism.⁹ Genomic expression analysis provides a greater understanding of the pathogenesis of retinoblastoma tumors, identifying potential gene targets and signaling pathways for adjuvant treatments. However, further studies are needed to correlate animal studies with human tumors.

Epidemiology

A recent editorial on the epidemiology of retinoblastoma made the following comment: "retinoblastoma is the most frequent primary intraocular cancer and is gaining importance rapidly."¹⁰ Based on the epidemiologic studies of several investigators, retinoblastoma affects approximately 1 in 16,000 – 18,000 births, for an incidence of 7000 – 8000 new cases annually worldwide.^{11, 12} It is estimated that there is an annual incidence of 3.5 per million children younger than 15,¹³ and 11.8 per million children younger than 5.¹¹ The estimated cumulative incidence is 53 – 62 per million children younger than 14.^{10, 13} Survival rates in the US approach 100%, with survival in other continents, primarily developing nations, much lower. Survival rates in developed Latin America countries has been reported to be 80 – 89%,^{14–16} 83% in Iran,¹⁷ 81% in China,^{18, 19} 48% in India,²⁰ and as low as 20 – 46% in Africa.^{1, 2} As a result, it has been estimated that 3000 – 4000 deaths occur annually worldwide.¹⁰ Amazingly, if survival rates worldwide approached those of developed countries such as those in Europe, the US, and Japan, retinoblastoma deaths could potentially be reduced by 88%, for approximately 400 children. These epidemiologic studies stress the importance of early detection and referral to tertiary care centers. Additionally, with the growing populations, especially in Asia and Africa, retinoblastoma is "gaining importance rapidly."¹⁰ Retinoblastoma is a highly curable cancer, and there is still much progress to made worldwide to combat this pediatric ocular malignancy.

Clinical Features and Classification

Retinoblastoma can present in a many different ways, with common presentations including leukocoria (Figure 1) and strabismus. Initial evaluation includes echography which can be important in differentiating RB from other disease entities. Differential diagnosis of RB includes Coat's disease, Persistent Fetal Vasculature (PFV), and toxocariasis, as well as other pathologies. In a recent analysis of 111 cases referred for possible RB, 68% proved to be retinoblastoma, while 32% were found to be other diseases. Of the 32% with an alternate diagnosis, 31% had PFV and 29% had Coat's disease.²¹

Retinoblastoma appears as a whitish retinal mass that may be solitary or multifocal. Depending on the growth pattern, tumors may have several characteristic features. Endophytic tumors arise in the inner retinal layers and extend into the vitreous, with the potential to produce vitreous seeding (Figure 2). Exophytic tumors arise in the outer retinal

layers and extend into the subretinal space beneath the retina, with a propensity for subretinal fluid and retinal detachments. Finally, diffuse tumors do not result in a mass, but rather diffuse plaque-like thickening of the retina.

Classification of retinoblastoma has undergone changes as treatment strategies have evolved. In the 1960s, Reese and Ellsworth developed the Reese-Ellsworth classification which served to predict globe-salvage in an era where external beam radiation was the primary form of treatment.²² However, the R-E classification failed to incorporate vitreous and subretinal seeding, and with adoption of chemoreduction and local consolidation treatment, a modified classification was developed to better predict success. As a result, the International Classification of Retinoblastoma was developed, placing an emphasis on focal and diffuse vitreous and subretinal seeds (Table 1).^{23, 24}

Treatment

Management of retinoblastoma tumors is complex, requiring coordinated care at a tertiary care center with discourse between the ocular oncologist, pediatric oncologist, pediatrician, interventional radiologist, and ocular pathologist. Management is individualized for each child, considering factors such as laterality, size and location of tumors, the risk for metastasis, vitreous and subretinal seeds, as well as the tumor relationship to surrounding tissues.²⁵ Additionally, as is evident in some cultures, family desires play an important role in retinoblastoma management. In Honduras, following a national campaign for retinoblastoma education, extraocular retinoblastoma was reduced from 73% to 35%; however, 33% of families were not compliant with treatment.²⁶ Retinoblastoma treatment must be aimed at child survival, followed by globe salvage and preservation of vision.

Small tumors may be amenable to local ablative therapy based on several criteria, including location greater than 3 mm from the fovea, greater than 1.5 mm from the optic disk, and size less than 3 mm in basal diameter and height. Ablative therapy can be performed with laser photocoagulation, cryotherapy, or TTT, inducing regression in 86% of tumors.²⁷ Laser treatments may be repeated every 3 – 4 weeks, until evidence of complete tumor regression and inactivity. Determination of clinical inactivity may be determined by indirect funduscopy showing calcified, inactive tumor, along with the absence of new tumor foci, evidence of tumor recurrence, or evidence of subretinal fluid, subretinal seeds, or vitreous seeds.²⁸ For advanced tumors that are not amenable to local ablative therapy, intravenous chemotherapy is utilized (chemoreduction).

Chemotherapeutic regimens generally use 6 – 10 cycles of CVE (carboplatin, vincristine, etoposide) depending on the institution. Bilateral or hereditary cases routinely need chemotherapy as these children have an 8 – 10% risk of pinealblastoma,²⁹ with risk decreased with the widespread use of chemoreduction protocols.³⁰ Focal consolidation therapy with laser is performed prior to each cycle, with some institutional protocols treating every 3 – 4 weeks until evidence of tumor inactivity. Shields et al.³¹ has proven the efficacy of chemoreduction with local consolidation, with successful tumor control varying between 47 – 100% from Group D A, respectively. Combined treatment has been shown to be more efficacious for tumor control, as chemotherapy alone resulted in tumor control rates (Reese-Ellsworth (R-E) group I – IV) of 51 – 65%^{32, 33}, compared to 62 – 100% with combined treatment^{28, 31}. For more advanced tumors (R-E group V), chemotherapy alone results in tumor recurrence in 63 – 75%^{32, 33}, compared to recurrence in 17 – 57% of tumors treated with chemoreduction combined with local consolidation^{28, 31, 34}. Recently, chemoreduction with aggressive local consolidation of foveal and extrafoveal portions of tumors was shown by Scheffler et al.²⁸ showed successful tumor control in 83% of R-E group V tumors. Additionally, in group I – IV eyes, 100% achieved tumor control. Despite ablative foveal

laser treatment, visual acuity remained 20/400 or better in 86% of eyes, and 20/80 or better in 56%.

For more advanced retinoblastoma tumors, or those classified as Group E eyes, standard management utilizes enucleation. Globes are submitted for histopathologic analysis to determine need for further adjuvant therapy. High-risk features include anterior chamber seeding, iris infiltration, ciliary body infiltration, retrolaminar optic nerve invasion, invasion of optic nerve transaction, massive choroidal infiltration, scleral infiltration, and extrascleral extension.³⁵ Eagle et al.³⁶ evaluated 387 globes, finding that 55 (18%) contained high-risk features. Adjuvant therapy often consists of 4 – 6 cycles of chemotherapy (CVE), with studies showing that metastasis developed in 24% of children compared to 4% when treated with adjuvant chemotherapy.³⁷

Local Chemotherapy

Intravenous chemotherapy combined with local consolidation has shifted management of retinoblastoma in favor of globe-salvage. However, successful tumor control in advanced tumors still often requires enucleation or radiation therapy. Additionally, systemic chemotherapy has been associated with systemic toxicities such as neutropenia and infection, as well as risks for second malignancies. To avoid systemic toxicities, local administration of chemotherapy has been proposed.

Subconjunctival carboplatin has been investigated for Group C and D eyes with long-term results of single therapy showing high failure rates.³⁸ The Children's Oncology Group advocates the combined use of 20 mg of carboplatin as a sub-Tenon's injection with chemoreduction and consolidation for tumors classified as group C and D.²⁵ Additionally, Leng et al.³⁹ showed early retinoblastoma tumors that progress despite laser ablative therapy can be effectively controlled with adjuvant treatment using subconjunctival carboplatin (Figure 3). As a result, focal subconjunctival injection of carboplatin may potentially be used to enhance tumor control in advanced retinoblastoma tumors as well as supplementing laser therapy for early tumors. Side effects have been reported, some severe, including ocular motility changes, optic nerve necrosis and atrophy, and periorbital fat necrosis.^{40–42} Further studies are needed to determine the role subconjunctival chemotherapy will play in retinoblastoma treatment as well as the long-term risk profile. We anticipate an ongoing multicenter trial combined with systemic chemotherapy to answer some of these questions.

More recently, intra-arterial chemotherapy has been investigated after selective ophthalmic artery infusion was demonstrated in 2004. Yamane et al.⁴³ showed that with cannulation of the internal carotid artery with distal balloon occlusion, selective infusion of melphalan could be successfully administered to the ophthalmic artery 97.51% of the time. Additionally, in 187 patients undergoing 563 cannulations, there were no reported complications from cannulation, including hemorrhage, stroke, or death.⁴³ However, the study did not report on tumor control rates or visual outcomes, and intra-arterial chemotherapy was combined with other treatments. Following this pioneering technique, techniques were developed to selectively cannulate the ophthalmic artery, eliminating the need for balloon occlusion (Figure 4). In a phase I/II study, Abramson et al.⁴⁴ demonstrated that 7 of 9 children with advanced tumors (R-E V) could be spared enucleation, with evidence of tumor regression, including vitreous and subretinal seeds. Importantly, no severe side effects were observed, and all but 1 patient had stabilization or improvement in vision.

Since these initial studies, several investigators have reported on their experience with supraselective ophthalmic artery infusion of chemotherapy. Abramson et al.⁴⁵ reported on their 3-year experience on 28 eyes of 23 children newly diagnosed with retinoblastoma were

enrolled. The majority (25 eyes) were R-E stage V, with one each of stage II, III, IV, with zero receiving prior treatment. All children were successfully cannulized and were treated with 1 – 6 infusions (mean 3.2). Twelve patients were treated with melphalan, 7 with melphalan plus topotecan, 3 with melphalan plus topotecan and carboplatin, and 1 with melphalan plus carboplatin. Only 1 of 28 eyes required enucleation because of disease progression, with the remainder also avoiding systemic chemotherapy and radiation. Kaplan-Meier estimates for globe-salvage was 100% at 1 year and 89% at 2 years (95% CI, 43 – 98%). Ophthalmic complications were mild, including lid edema, forehead hyperemia, and eyelash loss. Importantly, there were no deaths, strokes or hemorrhages, and several grade 3 and one grade 4 neutropenia, with zero requiring hospitalization.⁴⁵ The same group also reported on 4 patients with bilateral, advanced retinoblastoma (R-E stage V) who were treated initially with bilateral infusions during the same session (tandem therapy). All 4 eyes avoided enucleation or radiation, with no adverse effects observed, except one grade 3 neutropenia. Tumors underwent focal ablative therapy with TTT or cryotherapy following chemosurgery.⁴⁶

Mutapcic et al.⁴⁷ at Bascom Palmer Eye Institute (BPEI) reported on a series of 10 children (12 eyes) who received selective ophthalmic artery infusion of melphalan. All patients were R-E class Vb (ICRB Group D) who previously failed management with systemic chemotherapy and laser consolidation. Cannulation was successful 100% of time, with resultant regression of retinoblastoma tumors, as well as vitreous and subretinal seeds. Following 6 months, 9 of 12 eyes (75%) showed tumor control with no evidence of progression. Three eyes (25%) were enucleated secondary to tumor progression. Side effects included neutropenia in 4 children, intraretinal hemorrhages and peripapillary cotton wool spots in 1 eye that resembled Purtscher's-like retinopathy, vitreous hemorrhages in 3 eyes, and periocular edema from myositis in 1 eye. Mutapcic et al.⁴⁸ at BPEI also reports on the histopathology of the three enucleated eyes that showed evidence of tumor progression. These were the first retinoblastoma cases documenting viable tumor after supraselective ophthalmic artery infusion. On histopathology, 2 of 3 eyes had viable tumor with high risk characteristics. Two of three had tumors with invasion of the optic nerve to the level or just above the lamina cribrosa. There remained vitreous seeding in all 3 eyes, as well as one case showing minimal choroidal invasion. However, there was no anterior segment or extraocular extension in any of the eyes analyzed.

Intra-arterial chemotherapy offers an exciting alternative treatment that proves efficacious in the short-term for tumor control in retinoblastoma. However, long-term studies are needed to determine safety profiles and long-term tumor control, as well as determine appropriate treatment protocols. Additionally, it is unclear whether trilateral retinoblastoma, or pinealblastoma, will have a greater incidence without the use of systemic chemotherapy.

Translational Research – Targeting Tumor Microenvironment

The paradigm of cancer treatment has experienced tremendous advancements over the past few decades, as treatments not only target the hyperproliferating neoplastic cells, but also target the dynamic tumor microenvironment. The tumor microenvironment is a unique milieu of cytokines, growth factors, extracellular proteins, tumor cells, endothelial cells, fibroblasts, and inflammatory cells. The tumor microenvironment plays a key role in tumor growth and development, resistance to treatment, and metastasis. Despite evolution in chemotherapeutic agents and the delivery of these agents, a greater understanding of the pathophysiology of retinoblastoma is needed to develop novel targeted treatments. Using the LHBETA_{TA}G mouse model for retinoblastoma, our understanding of tumorigenesis, tumor microenvironment, and genomic expression has expanded rapidly.

Hypoxia has been identified to be a major contributor to tumor progression and metastasis. As neoplastic cells outgrow their blood supply, the cells are exposed to decreasing oxygen partial pressures. An angiogenic switch occurs, resulting in a complex vasculature modulated through hypoxia inducible factor (HIF) and growth factors such as vascular endothelial growth factor (VEGF).^{49, 50} Retinoblastoma tumors have been shown to consist of a heterogeneous vasculature that is spatially distributed in the tumor, with mature vessels concentrated in the center, while immature neovessels predominate in the periphery. Vascular targeting agents, such as anecortave acetate, have proven efficacious in decreasing tumor vasculature with subsequent enhancement in tumor control.⁵¹

Hypoxia has also been associated with more aggressive tumor phenotypes as hypoxic regions consist of slow-growing cells that prove resistant to chemotherapy and radiation. Hypoxic stress leads to cellular adaptations allowing survival in the harsh conditions. Tumor cells adopt an anaerobic state by altering gene expression and increasing glycolytic machinery for energy production. Advanced retinoblastoma tumors in the LH_{BETA}T_{AG} model have been shown to consist of hypoxic regions in 26% of tumors.⁵² As a result, hypoxia and hypoxic cells, such as vitreous seeds that lack a definitive blood supply, may serve as important targets for adjuvant therapies. Utilizing glycolytic inhibitors, such as 2-deoxy-glucose (2-DG), hypoxic cells have been targeted, resulting in a decrease in hypoxia as well as enhanced tumor control.⁵² Combined with vascular targeting therapies that increase hypoxia, glycolytic inhibitors and anti-angiogenic agents may be used synergistically for a greater reduction in tumor burden.

The tumor microenvironment also consists of tumor-associated macrophages (TAMs) and matrix metalloproteinases (MMPs) that have been associated with tumorigenesis. MMPs are key components of the extracellular matrix, and contribute to angiogenesis, tumor growth, and metastasis. With reduction in the expression of MMPs in LH_{BETA}T_{AG} tumors with anecortave acetate, there was an associated decrease in tumor burden.⁵³ Similarly, tumor-associated macrophages have been associated with increased levels of MMPs and mature vessels in retinoblastoma models. When these tumors were depleted of macrophages with subconjunctival liposomal clodronate, MMP expression decreased and tumor control improved, highlighting the potential importance of macrophages in tumor progression.⁵⁴ As a result, macrophages and associated MMPs appear to be important targets for adjuvant treatments, and further investigations are needed to define the complex role macrophages play in tumor modulation.

Recent genomic expression analysis of retinoblastoma tumors using the LH_{BETA}T_{AG} model has validated much of the prior experimental work. Genomic expression was shown to be both regionally and temporally-dependent, with dysregulated genes serving as key components of networks involved in hypoxia, angiogenesis, and cellular metabolism.⁹ Future analysis of dysregulated genes may lead to additional novel targets for adjuvant therapies. The PI3K/Akt/mTOR pathway was recently demonstrated to be a potentially important dysregulated pathway in the genomic expression of human retinoblastoma.⁷ Other studies have validated this observation, as rapamycin, an mTOR inhibitor has been shown to enhance tumor control in the retinoblastoma animal model.⁵⁵

Current studies have also shown that the genomic expression of retinoblastoma tumors in the animal model is altered by treatment with the glycolytic inhibitor, 2-DG, which targets hypoxia.⁵⁶ These studies provide a greater understanding in the mechanism of action of glycolytic inhibitors, and may potentially identify escape mechanisms used by neoplastic cells when hypoxic regions are targeted. Further investigations are needed to define the genetic profile of retinoblastoma tumors and correlate following treatment with chemotherapy and other agents.

Cancer treatment continues to evolve as many neoplasms are being treated with a combination of chemotherapy and adjuvant agents. With this shift in targeting the tumor microenvironment, elucidation of the mechanisms that drive tumorigenesis and resistance to treatment is imperative. As a result, continued translational research is important to develop treatments that achieve tumor control while minimizing treatment-related toxicities, as well as decreasing childrens' exposure to toxic systemic chemotherapies. With a greater understanding of genomic expression and the time-dependent nature of tumor growth, adjuvant therapies may be more optimally-timed to correspond to key time-points in tumor activity.

New Developments in Imaging

Intraoperative Optical Coherence Tomography (OCT)

Retinoblastoma screening occurs periodically in children with a history of retinoblastoma or a family history. Children undergo exam under anesthesia (EUA) to survey for new tumors, regression of treated tumors, or further progression of disease. Ocular oncologists' have a variety of imaging modalities in their armamentarium. Utilizing echography, wide-angle photography, and indirect ophthalmoscopy, practitioners base treatment decisions. A new spectral domain optical coherence tomography (SD-OCT) imaging platform with the ability to reliably and consistently image children in the operating room during EUAs has recently been developed and used at BPEI. The new platform uses the current Heidelberg SD-OCT, but suspends the unit vertically by mounting on an operating arm. This intraoperative platform allows images to be obtained consistently over time secondary to Heidelberg's eye-tracking technology. In addition to providing virtual biopsies of the retina, OCT imaging also provides the ability to obtain autofluorescence imaging. Initial reports have shown that intraoperative OCT images can be successfully obtained and provide detailed images of retinoblastoma tumors involving the macula, as well as peripheral lesions (Figure 5). Further studies are needed to compare SD-OCT with other common imaging modalities (echography, wide-angle photography, and indirect ophthalmoscopy). However, OCT may prove more sensitive for diagnosing subretinal seeds or determining active tumor growth. Intraoperative OCT imaging is an exciting new technology that may improve the diagnosis and management of retinoblastoma tumors. OCT may become the gold standard in intraoperative imaging in the future.

Conclusion

Retinoblastoma treatment has undergone tremendous advancements over the past few decades as EBRT has been replaced with chemoreduction and local consolidation as the primary treatment strategy. Concomitantly, survival has approached 100% in developed countries, with globe-salvaging possible in many children. However, progress is still needed in developing countries, notably areas of Africa and Asia, where death rates continue to be high. With the success of current treatment protocols, new investigations are evaluating local administration of chemotherapy, including intra-arterial and subconjunctival chemotherapy, to improve tumor control in advanced tumors, while also minimizing toxic systemic chemotherapy. Supersensitive ophthalmic artery infusion of chemotherapy has shown exciting preliminary results in control rates for retinoblastoma tumors, including advanced tumors destined for enucleation. Translational research continues to elucidate mechanisms of tumorigenesis, develop novel adjuvant therapies, as well as determine gene expression profiles of retinoblastoma tumors. Adjuvant therapies targeting hypoxia, angiogenesis, and cellular metabolism have been shown to be efficacious in tumor control and warrant further evaluation and trial in human retinoblastoma. Finally, we anticipate improved surveillance of tumor activity and growth with new imaging modalities such as intraoperative SD-OCT.

Acknowledgments

Supported by NIH center grant R01 EY013629, R01 EY12651, and P30 EY014801; by the American Cancer Society, Sylvester Comprehensive Cancer Center; and by an unrestricted grant to the University of Miami from Research to Prevent Blindness, Inc.

References

1. Wessels G, Hesselting PB. Outcome of children treated for cancer in the Republic of Namibia. *Med Pediatr Oncol*. 1996; 27:160–164. [PubMed: 8699993]
2. Bowman RJ, Mafwiri M, Luthert P, Luande J, Wood M. Outcome of retinoblastoma in east Africa. *Pediatr Blood Cancer*. 2008; 50:160–162. [PubMed: 17120241]
3. Knudson AG Jr. Mutation and cancer: statistical study of retinoblastoma. *Proc Natl Acad Sci U S A*. 1971; 68:820–823. [PubMed: 5279523]
4. Huang HJ, Yee JK, Shew JY, et al. Suppression of the neoplastic phenotype by replacement of the RB gene in human cancer cells. *Science*. 1988; 242:1563–1566. [PubMed: 3201247]
5. Khidr L, Chen PL. RB, the conductor that orchestrates life, death and differentiation. *Oncogene*. 2006; 25:5210–5219. [PubMed: 16936739]
6. Mastrangelo D, De Francesco S, Di Leonardo A, Lentini L, Hadjistilianou T. Retinoblastoma epidemiology: does the evidence matter? *Eur J Cancer*. 2007; 43:1596–1603. [PubMed: 17543516]
7. Chakraborty S, Khare S, Dorairaj SK, Prabhakaran VC, Prakash DR, Kumar A. Identification of genes associated with tumorigenesis of retinoblastoma by microarray analysis. *Genomics*. 2007; 90:344–353. [PubMed: 17604597]
8. Ganguly A, Nichols KE, Grant G, Rappaport E, Shields C. Molecular karyotype of sporadic unilateral retinoblastoma tumors. *Retina*. 2009; 29:1002–1012. [PubMed: 19491728]
9. Houston, SK.; Pina, Y.; Scott, WK., et al. Regional and Temporal Differences in the Genetic Expression of LHBETA TAG Retinoblastoma Tumors. Presented at the Association for Research in Vision and Ophthalmology (ARVO) Meeting; Ft Lauderdale, FL. May, 2010; 2010.
10. Kivela T. The epidemiological challenge of the most frequent eye cancer: retinoblastoma, an issue of birth and death. *Br J Ophthalmol*. 2009; 93:1129–1131. [PubMed: 19704035]
11. Broadus E, Topham A, Singh AD. Incidence of retinoblastoma in the USA:1975–2004. *Br J Ophthalmol*. 2009; 93:21–23. [PubMed: 18621794]
12. Seregard S, Lundell G, Svedberg H, Kivela T. Incidence of retinoblastoma from 1958 to 1998 in Northern Europe: advantages of birth cohort analysis. *Ophthalmology*. 2004; 111:1228–1232. [PubMed: 15177976]
13. MacCarthy A, Birch JM, Draper GJ, et al. Retinoblastoma in Great Britain 1963–2002. *Br J Ophthalmol*. 2009; 93:33–37. [PubMed: 18838413]
14. Schwartzman E, Chantada G, Fandino A, de Davila MT, Raslawski E, Manzitti J. Results of a stage-based protocol for the treatment of retinoblastoma. *J Clin Oncol*. 1996; 14:1532–1536. [PubMed: 8622068]
15. Carlos LL, Roberto RL, Victor TG, Carlos HG, Eduardo LP. Risk of dying of retinoblastoma in Mexican children. *Med Pediatr Oncol*. 2002; 38:211–213. [PubMed: 11836727]
16. Leal-Leal C, Flores-Rojas M, Medina-Sanson A, et al. A multicentre report from the Mexican Retinoblastoma Group. *Br J Ophthalmol*. 2004; 88:1074–1077. [PubMed: 15258028]
17. Naseripour M, Nazari H, Bakhtiari P, Modarres-zadeh M, Vosough P, Ausari M. Retinoblastoma in Iran: outcomes in terms of patients' survival and globe survival. *Br J Ophthalmol*. 2009; 93:28–32. [PubMed: 18952647]
18. Kao LY, Su WW, Lin YW. Retinoblastoma in Taiwan: survival and clinical characteristics 1978–2000. *Jpn J Ophthalmol*. 2002; 46:577–580. [PubMed: 12457919]
19. Chang CY, Chiou TJ, Hwang B, Bai LY, Hsu WM, Hsieh YL. Retinoblastoma in Taiwan: survival rate and prognostic factors. *Jpn J Ophthalmol*. 2006; 50:242–249. [PubMed: 16767380]
20. Swaminathan R, Rama R, Shanta V. Childhood cancers in Chennai, India, 1990–2001: incidence and survival. *Int J Cancer*. 2008; 122:2607–2611. [PubMed: 18324630]

21. Maki JL, Marr BP, Abramson DH. Diagnosis of retinoblastoma: how good are referring physicians? *Ophthalmic Genet.* 2009; 30:199–205. [PubMed: 19852578]
22. Reese AB, Ellsworth RM. The evaluation and current concept of retinoblastoma therapy. *Trans Am Acad Ophthalmol Otolaryngol.* 1963; 67:164–172. [PubMed: 13973597]
23. Linn Murphree A. Intraocular retinoblastoma: the case for a new group classification. *Ophthalmol Clin North Am.* 2005; 18:41–53. viii. [PubMed: 15763190]
24. Shields CL, Shields JA. Basic understanding of current classification and management of retinoblastoma. *Curr Opin Ophthalmol.* 2006; 17:228–234. [PubMed: 16794434]
25. Shields CL, Shields JA. Retinoblastoma management: advances in enucleation, intravenous chemoreduction, and intra-arterial chemotherapy. *Curr Opin Ophthalmol.* 2010; 21:203–212. [PubMed: 20224400]
26. Leander C, Fu LC, Pena A, et al. Impact of an education program on late diagnosis of retinoblastoma in Honduras. *Pediatr Blood Cancer.* 2007; 49:817–819. [PubMed: 17009236]
27. Shields CL, Santos MC, Diniz W, et al. Thermotherapy for retinoblastoma. *Arch Ophthalmol.* 1999; 117:885–893. [PubMed: 10408452]
28. Scheffler AC, Cicciarelli N, Feuer W, Toledano S, Murray TG. Macular retinoblastoma: evaluation of tumor control, local complications, and visual outcomes for eyes treated with chemotherapy and repetitive foveal laser ablation. *Ophthalmology.* 2007; 114:162–169. [PubMed: 17070578]
29. De Potter P, Shields CL, Shields JA. Clinical variations of trilateral retinoblastoma: a report of 13 cases. *J Pediatr Ophthalmol Strabismus.* 1994; 31:26–31. [PubMed: 8195959]
30. Shields CL, Meadows AT, Shields JA, Carvalho C, Smith AF. Chemoreduction for retinoblastoma may prevent intracranial neuroblastic malignancy (trilateral retinoblastoma). *Arch Ophthalmol.* 2001; 119:1269–1272. [PubMed: 11545631]
31. Shields CL, Mashayekhi A, Au AK, et al. The International Classification of Retinoblastoma predicts chemoreduction success. *Ophthalmology.* 2006; 113:2276–2280. [PubMed: 16996605]
32. Rodriguez-Galindo C, Wilson MW, Haik BG, et al. Treatment of intraocular retinoblastoma with vincristine and carboplatin. *J Clin Oncol.* 2003; 21:2019–2025. [PubMed: 12743157]
33. Gombos DS, Kelly A, Coen PG, Kingston JE, Hungerford JL. Retinoblastoma treated with primary chemotherapy alone: the significance of tumour size, location, and age. *Br J Ophthalmol.* 2002; 86:80–83. [PubMed: 11801509]
34. Shields CL, Mashayekhi A, Cater J, Shelil A, Meadows AT, Shields JA. Chemoreduction for retinoblastoma: analysis of tumor control and risks for recurrence in 457 tumors. *Trans Am Ophthalmol Soc.* 2004; 102:35–44. discussion 44–35. [PubMed: 15747743]
35. Honavar SG, Singh AD. Management of advanced retinoblastoma. *Ophthalmol Clin North Am.* 2005; 18:65–73. viii. [PubMed: 15763192]
36. Eagle RC Jr. High-risk features and tumor differentiation in retinoblastoma: a retrospective histopathologic study. *Arch Pathol Lab Med.* 2009; 133:1203–1209. [PubMed: 19653710]
37. Honavar SG, Singh AD, Shields CL, et al. Postenucleation adjuvant therapy in high-risk retinoblastoma. *Arch Ophthalmol.* 2002; 120:923–931. [PubMed: 12096963]
38. Abramson DH, Frank CM, Dunkel IJ. A phase I/II study of subconjunctival carboplatin for intraocular retinoblastoma. *Ophthalmology.* 1999; 106:1947–1950. [PubMed: 10519590]
39. Leng T, Cebulla CM, Scheffler AC, Murray TG. Focal periocular carboplatin chemotherapy avoids systemic chemotherapy for unilateral, progressive retinoblastoma. *Retina.* 2010; 30:S66–68. [PubMed: 20419851]
40. Mulvihill A, Budning A, Jay V, et al. Ocular motility changes after subtenon carboplatin chemotherapy for retinoblastoma. *Arch Ophthalmol.* 2003; 121:1120–1124. [PubMed: 12912689]
41. Schmack I, Hubbard GB, Kang SJ, Aaberg TM Jr, Grossniklaus HE. Ischemic necrosis and atrophy of the optic nerve after periocular carboplatin injection for intraocular retinoblastoma. *Am J Ophthalmol.* 2006; 142:310–315. [PubMed: 16876514]
42. Kiratli H, Kocabeyoglu S, Bilgic S. Severe pseudo-preseptal cellulitis following sub-Tenon's carboplatin injection for intraocular retinoblastoma. *J AAPOS.* 2007; 11:404–405. [PubMed: 17306997]

43. Yamane T, Kaneko A, Mohri M. The technique of ophthalmic arterial infusion therapy for patients with intraocular retinoblastoma. *Int J Clin Oncol*. 2004; 9:69–73. [PubMed: 15108036]
44. Abramson DH, Dunkel IJ, Brodie SE, Kim JW, Gobin YP. A phase I/II study of direct intraarterial (ophthalmic artery) chemotherapy with melphalan for intraocular retinoblastoma initial results. *Ophthalmology*. 2008; 115:1398–1404. 1404, e1391. [PubMed: 18342944]
45. Abramson DH, Dunkel IJ, Brodie SE, Marr B, Gobin YP. Superselective Ophthalmic Artery Chemotherapy as Primary Treatment for Retinoblastoma (Chemosurgery). *Ophthalmology*. 2010
46. Abramson DH, Dunkel IJ, Brodie SE, Marr B, Gobin YP. Bilateral superselective ophthalmic artery chemotherapy for bilateral retinoblastoma: tandem therapy. *Arch Ophthalmol*. 2010; 128:370–372. [PubMed: 20212212]
47. Mutapcic L, Murray TG, Aziz-Sultan MA, et al. Supraselective Intra-Arterial Chemotherapy: Evaluation of Treatment Related Complications in Advanced Refractory Retinoblastoma. *Clin Ophthalmol*. 2010 In press.
48. Mutapcic L, Murray TG, Aziz-Sultan MA, et al. Clinicopathologic Review of Enucleated Eyes After Intra-Arterial Chemotherapy with Melphalan for Advanced Retinoblastoma. *Arch Ophthalmol*. 2010 In press.
49. Pina Y, Boutrid H, Scheffler A, et al. Blood Vessel Maturation in Human and LHBETATAG Mouse Model Retinoblastoma Tumors: Spatial Distribution of Neovessels and Mature Vessels and Its Impact on Ocular Treatment. *Invest Ophthalmol Vis Sci*. 2008
50. Pina Y, Boutrid H, Scheffler A, et al. Blood vessel maturation in retinoblastoma tumors: spatial distribution of neovessels and mature vessels and its impact on ocular treatment. *Invest Ophthalmol Vis Sci*. 2009; 50:1020–1024. [PubMed: 18952925]
51. Boutrid H, Pina Y, Cebulla C, et al. Vessel Targeting Increases Hypoxia in a Murine Model of Retinoblastoma. *Invest Ophthalmol Vis Sci*. 2009
52. Boutrid H, Jockovich ME, Murray TG, et al. Targeting hypoxia, a novel treatment for advanced retinoblastoma. *Invest Ophthalmol Vis Sci*. 2008; 49:2799–2805. [PubMed: 18326690]
53. Bajenaru L, Pina Y, Murray T, et al. Gelatinase expression in Retinoblastoma: Modulation of LHBETATAG retinal tumor development by Anecortave Acetate. *Invest Ophthalmol Vis Sci*.
54. Pina Y, Boutrid H, Murray T, et al. Impact of Tumor-Associated Macrophages in LHBETATAG on retinal tumor progression: relation to macrophage sub-type. *Invest Ophthalmol Vis Sci*.
55. Murray, TG.; Pina, Y.; Houston, SK.; Hernandez, E.; Celdran, M.; Feuer, W. Retinoblastoma Tumor Burden Control: Rapamycin, an mTOR inhibitor, Decreases Tumor Burden in Advanced LHBETATAG Murine Retinoblastoma. Presented at the Association for Research in Vision and Ophthalmology (ARVO) Meeting; Ft Lauderdale, FL. May, 2010; 2010.
56. Pina, Y.; Houston, SK.; Scott, WK., et al. Retinoblastoma Molecular Genomics: Regional Differences in the Molecular Genomics Expression Following Treatment with 2-deoxy-D-glucose in LHBETATAG Retinal Tumors. Presented at the Association for Research in Vision and Ophthalmology (ARVO) Meeting; Ft Lauderdale, FL. May, 2010; 2010.

Figure 1.
Leukocoria in a child with retinoblastoma (left eye).

Figure 2. Larger macular retinoblastoma before treatment (left), and tumor after chemoreduction and laser consolidation (right).

Figure 3. Tumor control with sub-Tenon's carboplatin combined with focal laser ablation. Multifocal, unilateral retinoblastoma (left); following sub-Tenon's injection of carboplatin, drug is seen posterior to retina and tumor (middle); following laser and sub-Tenon's carboplatin, tumor control is evident at 5 year follow-up (right).

Figure 4.
Superselective ophthalmic artery cannulation resulting in local administration of chemotherapy.

Figure 5. Wide-angle photograph (top-left) of tumor activity in area of previous laser treatment. Intraoperative SD-OCT – 3-D image (top-right), fundus (bottom-left) and B-scan (bottom-right)

Table 1International Classification of Retinoblastoma (modified from Murphree et al.²³ and Shields et al.³¹)

Group	Subgroup	Quick Reference	Specific Features
A	A	Small Tumors	Rb $\leq$ 3mm
B	B	Larger Tumor	Rb > 3mm
		Macula	Location $\leq$ 3mm to foveola
		Juxtapapillary	Location $\leq$ 1.5mm from optic disc
		Subretinal Fluid	Clear subretinal fluid $\leq$ 3mm from Rb margin
C		Focal Seeds	
	C1		Subretinal seeds $\leq$ 3mm from Rb
	C2		Vitreous seeds $\leq$ 3mm from Rb
	C3		Both subretinal and vitreous seeds $\leq$ 3mm from Rb
D		Diffuse Seeds	
	D1		Subretinal seeds > 3mm from Rb
	D2		Vitreous seeds > 3mm from Rb
	D3		Both subretinal and vitreous seeds > 3mm from Rb
E		Extensive Rb	Occupying > 50% of globe or Neovascular Glaucoma Opaque media from hemorrhage in anterior chamber, vitreous, or subretinal space Invasion of postlaminar optic nerve, > 2mm of choroid, sclera, orbit, or anterior chamber