Retinoblastoma

Dr. Ashwin C Mallipatna
ashwin@narayananethralaya.com

Retinoblastoma is a rare and unique cancer that forms in the eyes of children, often before they are born. It is a complicated disease triggered by genetic mutations in one or more cells of the retina. The incidence of retinoblastoma is 1 in 15,000 live births, 1 with about 1500 children being diagnosed in India each year. It is the most common eye cancer in India. Untreated, retinoblastoma is fatal. With timely screening, diagnosis, referral, treatment, and follow-up delivered in a systematic way by a multidisciplinary team, 98% of children with retinoblastoma are cured, many with useful vision.

Pathogenesis:

Retinoblastoma is caused by the inactivation of both the alleles of the RB1 gene, located at the 13q14 locus (long arm of the 13^{th} chromosome). This gene is a tumor suppressor gene, is responsible for the formation of a protein called pRB which is responsible for regulating the cell cycle. **Knudson** used statistical probability to explain the difference in the age at presentation of a unilateral and bilateral retinoblastoma. His 'two-hit' hypothesis predicted that there are two mutational events which are responsible for the formation of retinoblastoma. **Comings** then proposed that these two events could be mutations on the two alleles of the RB1 gene. **Gallie** went on to show that the mutation of the RB1 gene was sufficient to form a retinoma, but its progression to retinoblastoma needed the inactivation of many more genes (such as the oncogenes KIF14, E2F3, DEK, MYCN and tumor suppressor gene CDH11).

The cell of origin of retinoblastoma is debatable. Many studies point to cone precursor cells as the origin of retinoblastoma. Once genetic changes in the precursor cell initiate tumor formation inside the eye, the tumor grows at a rapid pace. The growing tumor undergoes dystrophic calcification, which is pathognomonic of this tumor. Small tumors initially tend to be limited by the retinal boundaries (Bruch's membrane and the inner limiting membrane).

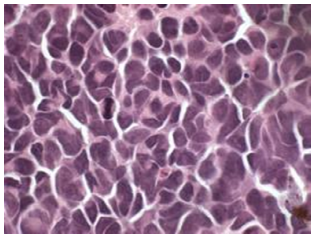
Two main patterns of tumor growth are seen in this disease. *'Endophytic'* retinoblastoma is described as the tumor pushing into the vitreous. When the inner limiting membrane breaks, "seeds" float into the vitreous which might eventually attach on to the retinal surface and grow. *'Exophytic'* tumors grow into the subretinal space, leading to exhudative retinal detachment.

Tumors may outgrow their blood supply leading to necrosis. Massive tumor necrosis may lead to significant ocular inflammation and aseptic orbital cellulitis. As the tumor progresses further, it can spread into the underlying choroid, optic nerve (beyond lamina cribrosa) or into the anterior segment causing a cataract, hypopyon, rubeosis iridis and glaucoma. Extraocular retinoblastoma is defined as tumor involvement outside the globe. The tumor can involve the central nervous system via the optic nerve or spread via the choroid, iris or trabecular meshwork into blood and disseminate to the rest of the body (commonly detected in bone marrow). The tumor could spread into the orbit transsclerally,

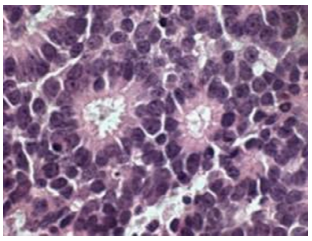
through inadvertent surgery or through the cut end of the optic nerve after an enucleation, which could result in regional lymph node involvement and systemic metastasis.

Pathology:

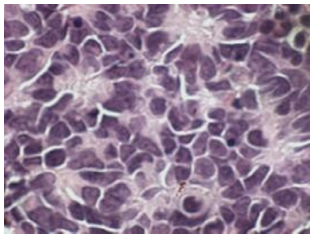
Retinoblastoma tumor is composed of cells with large hypochromatic nuclei and scanty cytoplasm, commonly with mitotic figures. More differentiated tumors consist of the typical Flexner-Wintersteiner rosettes, in which columnar cells are arranged in spheres around the lumen containing the primitive inner segments of photoreceptors. Homer-Wright rosettes are also seen within a retinoblastoma. Fleurettes are characteristic of retinomas, which could occur within areas with retinoblastoma.



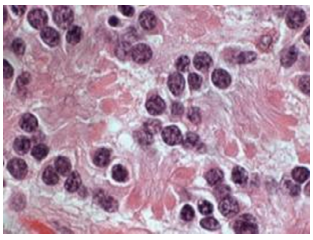
Undifferentiated RB



Flexner-Wintersteiner rosettes



Homer Wright rosettes



Fleurettes in Retinoma

Histopathology is important to pick up high-risk features, such as massive choroidal involvement and optic nerve involvement. Clinical care would require to be modified based on these findings.

Clinical Presentation:

Most cases present with a history of leukocoria (a white pupil). A white reflex might be noticed during routine red-reflex screening in pre-verbal children using distant direct ophthalmoscopy. Strabismus could result from a foveal tumor, and hence help pick up early disease.

Signs of late presentation might include buphthalmos, cataract, pseudo-hypopyon, aseptic orbital inflammation or proptosis. It is important that a child presenting with any of these signs be fully examined with the possibility of retinoblastoma in mind.

The tumor is characteristically a white- yellow mass in the retina, which is described as endophytic when it grows into the vitreous and exophytic when a detached retina covers it.

Classification:

Depending on the clinical features, retinoblastoma can be classified according to its prognosis with the current treatment modalities.

Intraocular retinoblastoma can be classified as follows:

International Classification for Intraocular Retinoblastoma

A	**Small tumour(s) confined to retina** • All tumours 3 mm or smaller in greatest dimension, confined to the retina and • All tumours located further than 3 mm from the foveola and 1.5 mm from the optic disc	• Laser • Freezing
B	**All remaining discrete tumours confined to the retina** • All tumours confined to the retina not in Group A • Any tumour-associated subretinal fluid less than 3 mm from the tumour with no subretinal seeding	• Chemotherapy • Laser • Freezing
C	**Discrete local disease with minimal subretinal or vitreous seeding** • Tumour(s) discrete • Subretinal fluid, present or past, without seeding, involving up to 1/4 retina • Local subretinal seeding, present or past, less than 3 mm (2 DD) from the tumour. • Local fine vitreous seeding close to discrete tumour	• Chemotherapy • Laser • Freezing • Removal of the eye if the other eye is normal
D	**Diffuse disease with significant vitreous or subretinal seeding** • Tumour(s) may be massive or diffuse • Subretinal fluid, present or past, without seeding, involving up to total retinal detachment • Diffuse subretinal seeding, present or past, may include subretinal plaques or tumour nodules • Diffuse or massive vitreous disease may include "greasy" seeds or avascular tumour masses	• Chemotherapy • Laser • Freezing • Removal of the eye if the other eye is normal
E	**Features of poor prognosis** • Tumour touching the lens • Neovascular glaucoma • Tumour anterior to anterior vitreous face involving ciliary body or anterior segment • Diffuse infiltrating retinoblastoma • Opaque media from hemorrhage • Tumour necrosis with aseptic orbital cellulites • Phthisis bulbi	• Removal of the eye
Extraocular spread		• Chemotherapy • Radiation • Bone Marrow transplantation

AJCC classification (7th edition; 2009): The American Joint Committee on Cancer has newly released its classification for retinoblastoma, which includes extraocular retinoblastoma. The classification goes according the TNM system. The AJCC classification uses the suffix of c-TNM for the clinical staging described below. Pathological staging is different, which is termed as p-TNM.

Features of the tumor (T):

TX: Primary tumor cannot be assessed.

T0: No evidence of primary tumor.

T1: Tumors no more than 2/3 the volume of the eye with no vitreous or subretinal seeding.

- T1a: No tumor in either eye is greater than 3 mm in greatest dimension or located closer than 1.5 mm from the optic nerve or fovea.
- T1b: At least one tumor is greater than 3 mm in greatest dimension or located closer than 1.5 mm to the optic nerve or fovea. No retinal detachment or subretinal fluid beyond 5 mm from the base the tumor.
- T1c: At least one tumor is greater than 3 mm in largest dimension or located closer than 1.5 mm to the optic nerve or fovea, with retinal detachment or subretinal fluid beyond 5 mm from the base of the tumor.

T2: Tumors no more than 2/3 the volume of the eye with vitreous or subretinal seeding. Can have retinal detachment.

- T2a: Focal vitreous and/or subretinal seeding of fine aggregates of tumor cells are present, but no large clumps or "snowballs" of tumor cells.
- T2b: Massive vitreous and/or subretinal seeding are present, defined as diffuse clumps or "snowballs" of tumor cells.

T3: Severe intraocular disease

- T3a Tumor fills more than 2/3 of the eye.
- T3b One or more complications present, which may include tumor-associated neovascular or angle closure glaucoma, tumor extension into the anterior segment, hyphema, vitreous hemorrhage, or orbital cellulitis.

T4: Extraocular tumor detected by imaging studies.

- T4a Invasion of optic nerve.
- T4b Invasion into the orbit.
- T4c Intracranial extension not past chiasm.
- T4d Intracranial extension past chiasm.

Features of Lymph Node (N): This category of staging applies only to extra scleral extension

NX: Regional lymph nodes cannot be assessed

N0: No regional lymph node involvement

N1: Regional lymph node involvement (preauricular, cervical, submandibular)

N2: Distant lymph node involvement

Features of Metastasis (M):

MX: Metastasis cannot be assessed.

M0: No metastasis.

M1: Metastasis

- M1a Single lesion to sites other than CNS
- M1b Multiple lesions to sites other than CNS.
- M1c Prechiasmatic CNS lesion(s).
- M1d Postchiasmatic CNS lesion(s).
- M1e Leptomeningeal and/or CSF involvement.

Previously used classification systems:

1. **The Reese-Ellsworth Staging System**

This system was developed in the 1960s, when most children were being treated with external beam radiation therapy (EBRT). The terms favourable, doubtful, unfavourable, etc. in this staging system refer to the likelihood that the cancer can be effectively treated while preserving vision in the affected eye. These terms do not refer to likelihood of the child's survival. This system is currently not preferred, as it does not predict prognosis of the child or vision with the current treatment modalities.

Group 1 (Very Favourable):

a. One tumour, smaller than 4 disc diameters (DD) in size, at or behind the equator
b. Multiple tumours smaller than 4 DD in size, all at or behind the equator

Group 2 (Favourable):

a. One tumour, 4 to 10 DD in size, at or behind the equator
b. Multiple tumours, 4 to 10 DD in size, all at or behind the equator

Group 3 (Doubtful):

a. Any tumour in front of the equator
b. One tumour, larger than 10 DD, behind the equator

Group 4 (Unfavourable):

a. Multiple tumours, some larger than 10 DD
b. Any tumour extending anteriorly (toward the front of the eye) to the ora serrata (front edge of the retina)

Group 5 (Very Unfavourable):

a. Tumours involving more than half of the retina
b. Vitreous seeding (spread of tumours into the gelatinous material that fills the eye)

2. **St. Jude's Children's Research Hospital Staging System**

Stage I: tumour confined to the retina:

- Occupying one quadrant (one fourth of the retina) or less
- Occupying two quadrants or less
- Occupying more than 50% of the retinal surface

Stage II: tumour confined to globe (eyeball) with vitreous seeding

- Extension to the optic nerve head
- Extension to the choroid (middle layer of the wall of the eyeball)
- Extension to the choroid and optic nerve head
- Extension to emissaries (veins near the eye)

Stage III: regional extraocular (outside of the eyeball) extension of tumour:

- Extension beyond cut ends of the optic nerve (the nerve is cut during surgery)
- Extension through the sclera (outermost layer of the wall of the eyeball) into orbital contents (tissues next to the eyeball)
- Extension to the choroid beyond cut end of the optic nerve, including subarachnoid extension (beneath the thin membrane that covers the brain)
- Extension through the sclera into orbital contents and beyond cut end of the optic nerve, including subarachnoid extension

Stage IV: distant metastases (spread):

- Extension via the optic nerve to the brain (that is, tumour nodules in the brain or tumour cells in the cerebrospinal fluid)
- Blood-borne metastases to soft tissue, bone, or viscera (internal organs)
- Bone marrow metastases

Investigations:

The diagnosis of retinoblastoma relies mostly on the clinical examination. A radiological investigation is used for confirming the diagnosis in situations where the presentation of retinoblastoma is atypical. These investigations are currently very useful to detect the extent of disease, especially to detect optic nerve involvement. They also assist in detecting a primary neuro-ectodermal tumor. These are midline tumors in the brain, which sometimes arise from the pineal gland (called a pinealoma).

Magnetic Resonance Imaging:

Retinoblastoma demonstrates as a hyperintense mass growing in the eye in T1 weighted images, and hypointense in T2-weighted images. Gadolinium can be used to demonstrate optic nerve involvement. An MRI is more sensitive than a CT scan for the detection of extraocular spread of retinoblastoma. It is especially important to attempt to avoid a CT scan in bilateral retinoblastoma to prevent additional exposure to radiation.

Computerised Tomography Scans:

A CT scan is excellent at demonstrating calcification in those cases where the diagnosis is not certain.

B-Scan Ulrasonography:

Tumors demonstrate the presence of a hyperechoic mass within the globe, which might be speckled with highly reflective spots from calcification. Calcific spots demonstrate shadowing, which extend posterior to the tumor mass.

Differential Diagnosis:

- Hereditary Conditions
    - Norrie's Disease and AR Retinal Dysplasia
    - Incontinentia Pigmenti
    - ad-FEVR
    - Juvenile Retinoschisis
- Inflammatory Conditions
    - Orbital Cellulitis
    - Toxocariasis
    - Toxoplasmosis
    - Metastatic Endophthalmitis
    - Congenital CMV Retinitis
    - Herpes Simplex Retinitis
    - Vitritis

- Tumors
    - Astrocytic Hamartoma
    - Choroidal Hemangioma
    - Combined Hamartoma
    - Meduloepithelioma
- Developmental Abnormalities
    - Persistent Hyperplastic Primary Vitreous
    - Cataract
    - Coloboma
    - Congenital Retinal Fold
    - Myelinated Nerve Fibres
    - High Myopia
    - Morning Glory Syndrome
- Others
    - Coats Disease
    - Retinopathy of Prematurity
    - Rhegmatogenous Retinal Detachment
    - Vitreous Hemorrhage
    - Leukemic Infiltration of the Iris

Management:

The management of retinoblastoma depends on:

1. **Germline status** (clinically indicated by bilateral tumors, presence of primary neuro-ecodermal tumor, presence of a family history, early age at presentation).
2. **Extent of tumor involvement** (clinical classification).

The treatment involves many modalities:

1. *Enucleation:*
   Removal of an eye with tumor is often the only treatment required for unilateral (non germline) retinoblastoma. The current care suggest we perform the surgery in such a way that we obtain a long optic nerve stump, place an orbital Implant primarily, and harvest fresh tumor at enucleation for genetic analysis. Although the practice of enucleation is not as rampant as before, one must be aware of how simple and effective this surgery is when compared to eye preservation from chemotherapy with focal therapy.

2. Chemotherapy:
    a. *Systemic chemotherapy:*
    The various commonly used regimen involves treatment with vincristine, etoposide and carboplatin. Vincristine inhibits microtubule formation, etoposide is a eukaryotic type II topoisomerase inhibitors and carboplatin is an alkylating-like agent that causes non-cell cycle specific DNA damage. The dosages used vary in different regimens. Current practices involve the use of large doses of cyclosporine-A, which blocks P-glycoprotein induced efflux and counteracts chemotherapy resistance.
    b. Periocular chemotherapy:
    Chemotherapeutic agents such as carboplatin are injected into the sub-tenons space, which diffuse into the globe through the sclera. This might assist in the treatment of diffuse vitreous seeds. Due to the local complications associated with carboplatin injections, the use of improved agents such as carboplatin nanoparticles and topotecan.
    c. Intra-arterial chemotherapy:
    Recent advances in image guided therapy allow the ophthalmic artery to be selectively isolated for injection of melphalan. This mode of delivery of chemotherapy could be associated with complications, and needs prospective trials to understand its long term complications.
    d. Intraocular chemotherapy:
    Melphalan has been injected into the vitreous to treat vitreous seeds. Despite its slowly growing popularity in developing nations, this practice is not considered safe by many authorities as there is a risk of the tumor spreading through the site of injection.

3. Focal therapy:
    Focal therapy with laser and cryotherapy help consolidate the response obtained after chemotherapy. Very small tumors may be amicable to only focal therapy, without chemotherapy.
    a. Lasers:
    Retinal lasers with 532nm and 810nm lasers may be delivered on active tumor with an indirect laser ophthalmoscope. Discrete tumors are treated with laser burns up to two burn widths outside the margin of the tumor. The surface of the tumor might require to be treated to consolidate response. Lasers are ideal for tumors located posterior to the equator. Transpupillary Thermo-therapy may be used to treat lesions in the posterior pole, which uses a large spot size and lower energy infra-red laser continuously delivered over a longer period to treat the tumor.
    b. Cryotherapy:
    Peripheral tumors are treated with three cycles of freezing and thawing. The tumors are destroyed while thawing. Complete thawing is ensured when a gap of one minute is provided between the onset of thawing and the beginning of the next freeze.

4. Radiotherapy:
   Radiation was the primary modality of treatment until the 90s. The use of chemotherapy consolidated with focal therapy has taken over as the primary treatment of choice in those eyes not undergoing enucleation. Radiation is now used as a salvage, to treat a one-eyed patient with the disease, or to treat obvious extraocular disease. The complications of radiation include a higher incidence of primary CNS tumors, ocular surface problems and cataracts. The types of radiation used include:
   a. Standard external beam radiation: Does not target the involved area.
   b. Stereotactic radiation: Can precisely isolate the area to be treated, without the surrounding tissues being exposed to high doses of radiation.
   c. Bracytherapy: Can be used to treat localised tumor activity which is not amicable to focal therapy.

Genetic Mutation Analysis:

Determining the specific RB1 gene mutation is important in the clinical care of retinoblastoma to determine the germline status of the child, and by that the risk for the siblings and other members of the family at risk. Knowing the absence of a germline status can reduce costs of screening involved in the care of the family members at risk. In addition, this knowledge can provide us with the ability to provide pre-natal diagnosis and counselling for family planning.

Early Detection of Retinoblastoma:

Early detection has direct correlation with prognosis, and may reduce the requirement to enucleate an eye. Early detection can be encouraged by promoting leukocoria awareness amongst the community and facilitating red-reflex screening of every pre-verbal child. Any child detected to have a white reflex or a squint is advised to undergo a detailed eye examination within 48 hours by an ophthalmologist. Children found to have cataract and glaucoma should be examined with a B-scan to detect the presence of an intraocular mass. All children with retinoblastoma must be treated at a facility which has a multidisciplinary support. Early detection of the tumor in siblings must be facilitated by genetic counselling and RB1 gene mutation analysis.