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## Case report

# Atypical intravitreal growth of retinoblastoma with a multi-branching configuration



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## ABSTRACT

**Purpose:** To report the clinical and histopathological findings of atypical intravitreal growth of a retinoblastoma with a multi-branching configuration.

**Observations:** A 7-month-old boy was referred to our hospital due to leukocoria in the right eye. Ophthalmic examinations identified multi-branching vessels surrounded by diaphanous tissue behind the lens in the right eye. Imaging modalities showed microphthalmos, band-shaped calcification, and cystic lesions in that eye. Because it was difficult to rule out congenital anomalies such as persistent fetal vasculature due to the atypical clinical features of retinoblastoma, we performed a biopsy using a limbal approach. An intraoperative rapid pathological examination led to the definitive diagnosis of retinoblastoma. The right eye was enucleated and postoperative adjuvant chemotherapy was administered. Immunohistochemical staining of the enucleated eyeball showed that the tumoral cells and diaphanous tumoral tissue around the vessels were positive for neuron-specific enolase and Ki-67 and partially positive for glial fibrillary acidic protein (GFAP). The vessels of the diaphanous tissues near the tumoral mass were stained by GFAP and those behind the lens were stained faintly.

**Conclusions and importance:** We described an atypical retinoblastoma of pseudo-persistent fetal vasculature with a multi-branching configuration, which expanded the clinical spectrum of retinoblastoma. Such a specific growth pattern of the embryonic tumor might occur with a combination of retinal development, retinal vasculature, and hyaloid vascular system.

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## 1. Introduction

Retinoblastoma is a major intraocular neoplasm that develops during childhood.<sup>1</sup> Various clinical phenotypes in eyes with retinoblastoma are seen including diffuse and massive tumors with vitreous seeding and retinal detachment<sup>2</sup> that are misdiagnosed as other ocular diseases such as Coat's disease, persistent fetal vasculature (PFV), vitreous hemorrhage, ocular toxocariasis, familial exudative vitreoretinopathy, retinal dysplasia, and other diseases.<sup>2</sup> In addition to clinical examinations, histopathological

examinations are important for a definitive diagnosis of retinoblastoma and to predict the disease prognosis.<sup>3</sup> The current report describes the clinical and histopathological features of an atypical retinoblastoma with a multi-branching configuration that grew intravitreally.

## 2. Case report

A 7-month-old boy born at 39 weeks gestation and weighing 3808 g was referred to our hospital with leukocoria in the right eye. He had no history of systemic diseases in his clinical course and no familial history of ocular diseases. He was esotropic with poor visual fixation in the right eye.

Slit-lamp biomicroscopy and ophthalmoscopy identified multi-branching vessels surrounded by diaphanous tissue behind the

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right lens. Slit-lamp biomicroscopy and photography using the RetCam (Massie Research Laboratories, Inc., Pleasanton, CA) showed multi-branching vessels with diaphanous tissue behind the lens (Fig. 1A, and C). Fluorescein angiography (FA) showed a strand of vessels in each branch, some of which developed abnormal branching. Hyperfluorescence was in and around the vessels without apparent fluorescein leakage (Fig. 1D). Anterior-segment optical coherence tomography (RS-3000, Nidek, Gamagori, Japan) showed multi-branching solid tissues behind the lens, some of which adhered to the posterior lens surface (Fig. 1B). The left eye was normal.

In the vitreous cavity of the right eye, B-mode ultrasonography showed strands and a total retinal detachment, computed tomography (CT) showed band-shaped calcifications and a light shadow of solid tissue (Fig. 2A), and T<sub>2</sub>-weighted magnetic resonance imaging (MRI) showed band-shaped and cystic lesions extending from the optic nerve (Fig. 2B). Slit-lamp and ultrasound biomicroscopy showed a shallow anterior chamber. Mild microphthalmos of the right eye (axial length of right eye, 17.5 mm; left eye, 19.5 mm) was estimated by ultrasonography. No orbital or systemic abnormalities were seen on the CT and MRI images.

It was difficult to differentiate retinoblastoma, PFV, or other congenital anomalies based on these clinical features. We performed a biopsy of the branching tissues after lensectomy via a limbal approach to reduce the risk of hematogenous metastasis of a possible malignancy (Fig. 3A). Intraoperative rapid pathological examination clarified the tissue as retinoblastoma with nuclear fission and rosettes (Fig. 3B). We enucleated the right eye. Six cycles of adjuvant chemotherapy (vincristine, etoposide, and carboplatin) were applied postoperatively to prevent metastasis. Three years after treatment the patient has had no complications.

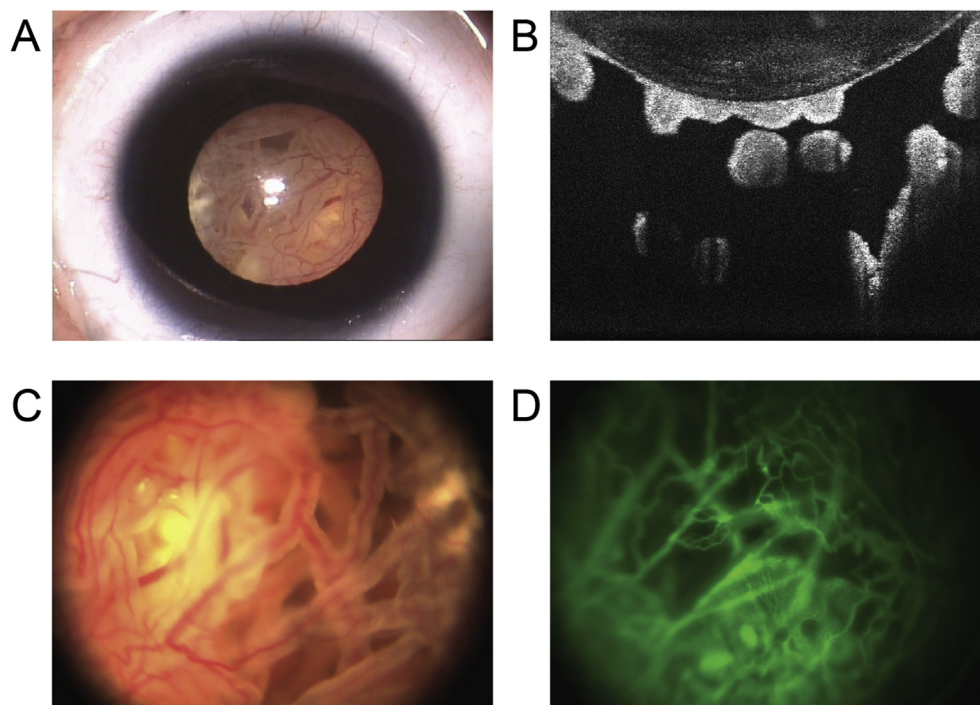
Histopathology of the enucleated eyeball showed a tumoral mass strongly positive for neuron-specific enolase (NSE) and Ki-67 immunostainings and partially positive for glial fibrillary acidic protein (GFAP) (Fig. 4B, C and D). Some retinal dysplasia was

identified, although most other retinal layers were well developed (Fig. 4A). No persistent hyaloid artery was identified in the serial sections of the optic disc. The multi-branching tissue, which filled the anterior vitreous cavity, was connected to the main body of the tumor (Fig. 5A). The tumoral cells of the branching tissue were positive for NSE and Ki-67 and partly positive for GFAP (Fig. 5B, C and D). The tumoral cells in each branch of tissue contained one central mature vessel (Fig. 6A) positive for NSE and Ki-67 (Fig. 6B and C). GFAP staining was seen around the vascular endothelium in the branching tissue near the main body of the tumoral mass (Fig. 5D), and that behind the lens was stained faintly (Fig. 6D).

### 3. Discussion

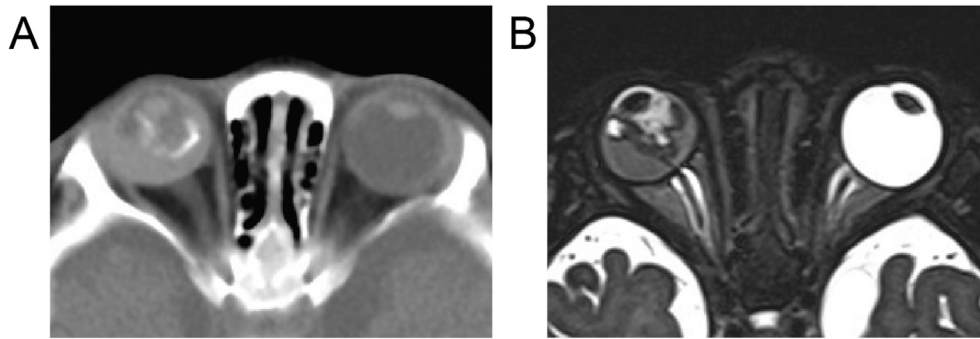
Retinoblastoma is sometimes difficult to distinguish from congenital ocular anomalies, including PFV and retinal dysplasia.<sup>2</sup> In addition, the presence of retinoblastoma and PFV in the same eye is extremely rare but has been reported in a few cases.<sup>4–7</sup> Calcification within the eyeball, which is important in suspected cases of retinoblastoma, was present in the current case; however, the calcification is not a definite diagnostic finding in retinoblastoma and also is seen in other congenital diseases such as PFV and Coat's disease.<sup>2,8</sup> The microphthalmos in the affected eye suggested PFV and other congenital anomalies. Considering the atypical clinical findings in the current case, the diagnosis of retinoblastoma was difficult to establish except by biopsy and pathological examination.

Retinal dysplasia also was seen in the current case. The clinical phenotypes between retinoblastoma and retinal dysplasia are sometimes confusing; however, the pathological characteristics differ clearly.<sup>9</sup> Retinoblastoma is a neoplasm that originates from primitive neuroretinal cells,<sup>10</sup> including cone-precursor cell,<sup>11</sup> or Muller glia,<sup>12</sup> whereas retinal dysplasia is a non-neoplastic tumor that originates from abnormal differentiation of one or more retinal

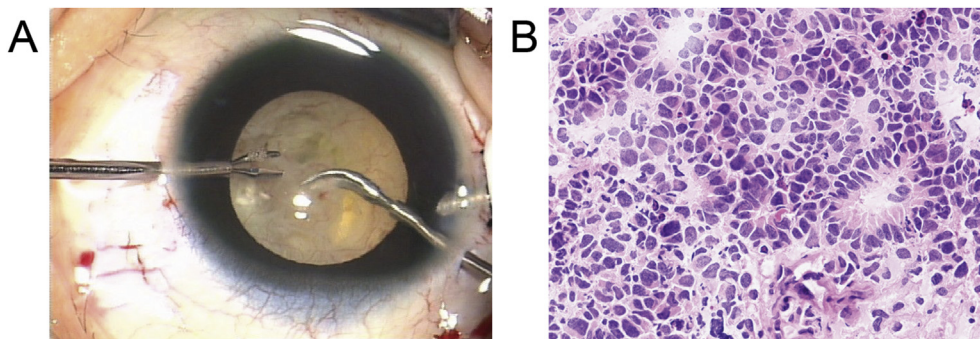


**Fig. 1.** The ophthalmic examinations of the affected right eye under general anesthesia before surgery. (A) Slit-lamp biomicroscopy. (B) Anterior-segment optical coherence tomography. (C) Photography. (D) Fluorescein angiography. The clinical findings are described in the case presentation.

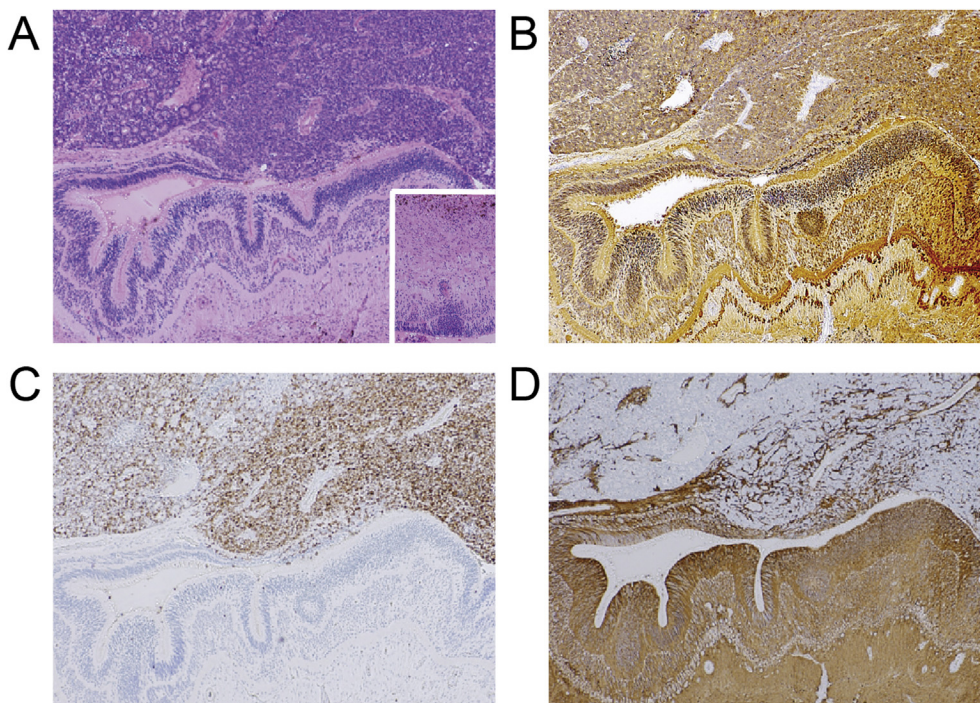




**Fig. 2. Computed tomography (CT) and magnetic resonance imaging (MRI) results of the affected right eye before surgery.** (A) A CT image shows a band-shaped calcification in the right eye. (B) A T<sub>2</sub>-weighted MRI axial image shows a band-shaped area of low density along the area of calcification in the CT image.

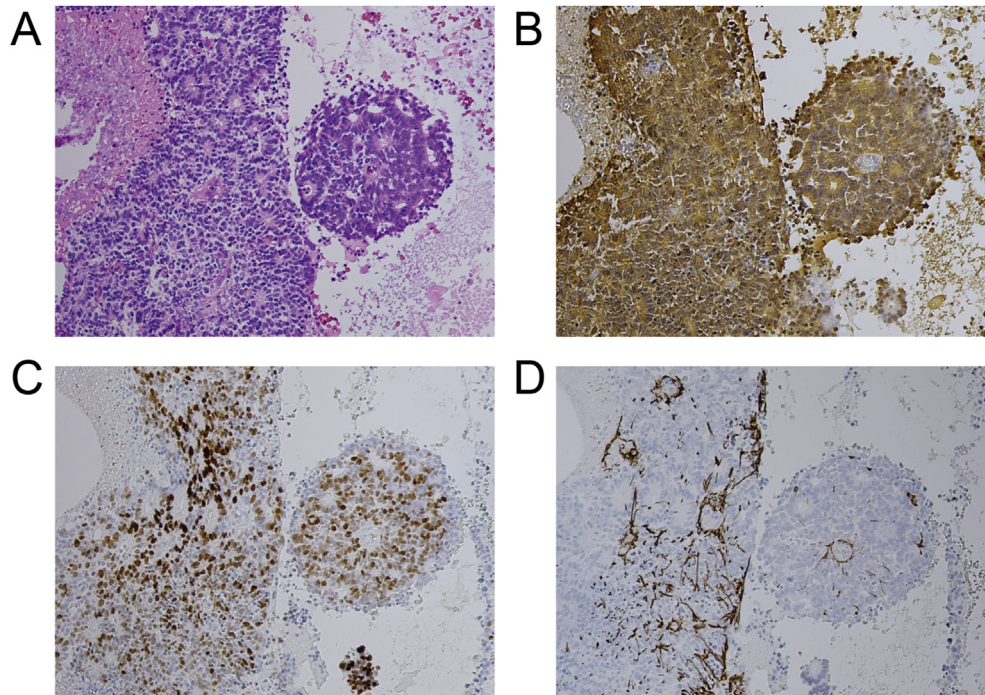


**Fig. 3. Biopsy and its pathological results during surgery.** (A) A biopsy of the diaphanous tissue behind the lens is performed after lensectomy via a limbal approach. (B) The intraoperative rapid pathological examination shows proliferation of small, round tumoral cells around the vessels with nuclear fission, calcification, and rosettes.

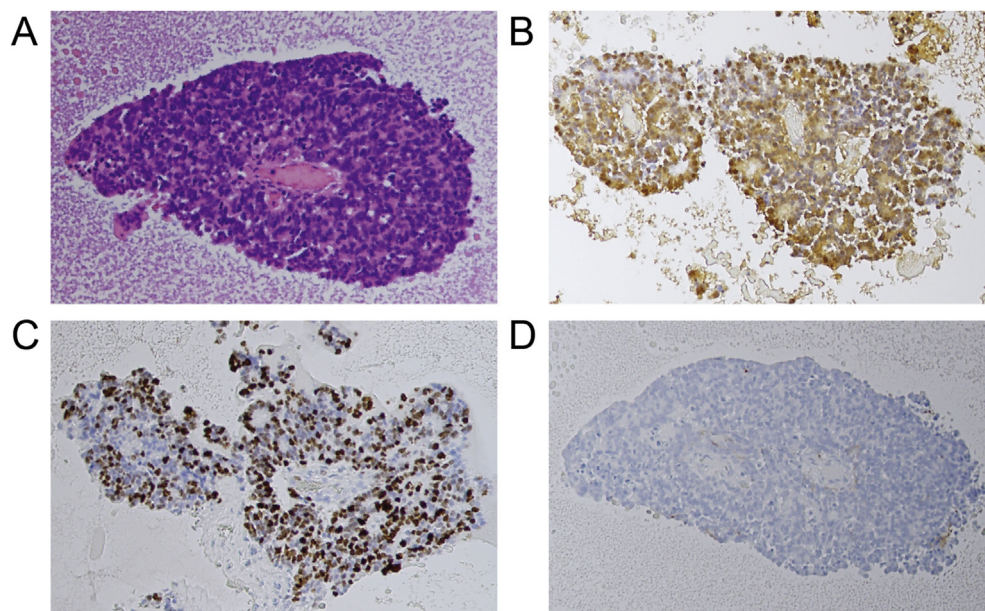


**Fig. 4. The histopathology and immunohistochemistry of the tumoral mass and adjacent retina.** Staining by (A) hematoxylin-eosin, (B) neuron specific enolase (NSE), (C) Ki-67, and (D) glial fibrillary acidic protein (GFAP). The tumoral mass (upper tissue in the section) is stained markedly by NSE and Ki-67 indicating a retinoblastoma, and stained partially by GFAP. The adjacent retina (lower tissue in the section) is stained markedly by NSE and GFAP, and some retinal dysplasia is seen in the lower right section of A.





**Fig. 5.** The histopathology and immunohistochemistry findings in the multi-branching vessels with diaphanous tissue near the main body of the tumoral mass. Staining by (A) hematoxylin-eosin, (B) neuron specific enolase (NSE), (C) Ki-67, and (D) glial fibrillary acidic protein (GFAP). The tumoral cells around the vessel show marked staining by Ki-67 and NSE. GFAP staining is seen around the vascular endothelium in the multi-branching tissue.



**Fig. 6.** The histopathology and immunohistochemistry findings in the multi-branching vessels with diaphanous tissue behind the lens. Staining by (A) hematoxylin-eosin, (B) neuron specific enolase (NSE), (C) Ki-67, and (D) glial fibrillary acidic protein (GFAP). The tumoral cells around the vessels are stained by Ki-67 and NSE. The area around the vascular endothelium is stained faintly by GFAP.

components.<sup>13</sup> The co-existence of retinoblastoma and retinal dysplasia in our case might occur occasionally or be related.

The extremely rare and remarkable finding of the current case was the multi-branching configuration in the anterior eyeball, which resulted from multi-branching vessels surrounded by tumoral cells, which was difficult to distinguish from PFV. Typical PFV is characterized by branching of the hyaloid artery covering the

posterior lens surface with a remnant stalk extending from the optic disc and the remaining hyaloid artery with proliferated fibrovascular tissue,<sup>14</sup> which were not seen in the current case. In the current case, the multi-branching vessels were surrounded by tumoral cells, some of which adhered to the posterior lens surface. FA showed little fluorescein leakage, and histopathology showed mature and well-differentiated vessels in the branching tissue.

These findings suggested two possibilities for the rare tumoral growth. First, the tumoral cells proliferated with a feeding vessel that originated from the retinal vessels. Second, the tumoral cells grew along the ectopic vessels that might indicate a relationship of PFV or ectopic intravitreal growth of retinal vessels. Immunohistochemistry suggested that GFAP-positive astrocytes were present around the vessels in the branches of the tumor in the pathway from the tumoral mass to the posterior lens surface. Astrocytes migrate and develop along with retinal vessels and also are present around the trunk of the hyaloid artery as an outer sheath of Bergmeister papilla that extends into the posterior half of the vitreous cavity during the developmental stage. In the current case, because histopathology failed to identify a trunk of hyaloid artery that extended from the optic disc, it was difficult to determine if the vessels in the branches of the tumor originated from abnormal retinal vessels or PFV.

We report an atypical retinoblastoma of pseudo-PFV with a multi-branching configuration, which expanded the clinical spectrum of retinoblastoma. Such a specific growth pattern of the embryonic tumor might occur with a combination of retinal development, retinal vasculature, and hyaloid vascular system.

#### 4. Patient consent

The patient's legal guardian provided written informed consent for publication of this case report and any accompanying images.

#### Acknowledgements and disclosures

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##### Conflict of interest

The authors declare that there is no conflict of interests regarding this paper.

#### Authorship

All authors attest that they meet the current ICMJE criteria for Authorship.

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#### References

1. Sang DN, Albert DM. Retinoblastoma: clinical and histopathologic features. *Hum Pathol.* 1982;13:133–147.
2. Shields CL, Schoenberg E, Kocher K, Shukla SY, Kaliki S, Shields JA. Lesions simulating retinoblastoma (pseudoretinoblastoma) in 604 cases: results based on age at presentation. *Ophthalmology.* 2013;120:311–316.
3. Chawla B, Sharma S, Sen S, et al. Correlation between clinical features, magnetic resonance imaging, and histopathologic findings in retinoblastoma: a prospective study. *Ophthalmology.* 2012;119:850–856.
4. Irvine AR, Albert DM, Sang DN. Retinal neoplasia and dysplasia. II. Retinoblastoma occurring with persistence and hyperplasia of the primary vitreous. *Investigative Ophthalmol Vis Sci.* 1977;16:403–407.
5. Morgan KS, McLean IW. Retinoblastoma and persistent hyperplastic vitreous occurring in the same patient. *Ophthalmology.* 1981;88:1087–1089.
6. Liang JC, Augsburger JJ, Shields JA. Diffuse infiltrating retinoblastoma associated with persistent primary vitreous. *J Pediatr Ophthalmol strabismus.* 1985;22:31–33.
7. Jakobiec FA, Rai R, Rashid A, Kanoff J, Mukai S. Dystrophic hyaloid artery remnants and other abnormalities in a buphthalmic eye with retinoblastoma. *Surv Ophthalmol.* 2014;59:636–642.
8. Levy J, Frenkel S, Baras M, Neufeld M, Pe'er J. Calcification in retinoblastoma: histopathologic findings and statistical analysis of 302 cases. *Br J Ophthalmol.* 2011;95:1145–1150.
9. Biswas S, Paul PC, Chakraborty J, Chakrabarti S. Unilateral retinal dysplasia clinically mimicking retinoblastoma. *Online J Pediatr Ophthalmol strabismus.* 2010;47:e1–3.
10. Kyritsis AP, Tsokos M, Triche TJ, Chader GJ. Retinoblastoma—origin from a primitive neuroectodermal cell? *Nature.* 1984;307:471–473.
11. Xu XL, Singh HP, Wang L, et al. Rb suppresses human cone-precursor-derived retinoblastoma tumours. *Nature.* 2014;514:385–388.
12. Pajovic S, Corson TW, Spencer C, et al. The TAg-RB murine retinoblastoma cell of origin has immunohistochemical features of differentiated Muller glia with progenitor properties. *Investigative Ophthalmol Vis Sci.* 2011;52:7618–7624.
13. Godel V, Nemet P, Lazar M. Retinal dysplasia. *Documenta Ophthalmol Adv Ophthalmol.* 1981;51:277–288.
14. Shastri BS. Persistent hyperplastic primary vitreous: congenital malformation of the eye. *Clin Exp Ophthalmol.* 2009;37:884–890.