E-ISSN: 3050-9459
Journal of Ophthalmology and Advance Research

ATHENAEUM SCIENTIFIC PUBLISHERS

Case Report

Comparative Histomorphology and Immunofluorescence of Anterior and Posterior Lens Capsular Plaques from the Same Eye

Gaurang Gajjar[1], Abhay Vasavada[2], Kaid Johar SR[1*]

[1]Department of Zoology, Biomedical Technology and Human Genetics, Gujarat University, Ahmedabad-380009, India

[2]Iladevi Cataract and IOL Research Centre, Raghudeep Eye Hospital, Ahmedabad-380052, India

***Correspondence author:** Kaid Johar SR, Department of Zoology, Biomedical Technology and Human Genetics, Gujarat University, Ahmedabad-380009, India; Email: kaidjohar@gujaratuniversity.ac.in

Citation: Gajjar G, et al. Comparative Histomorphology and Immunofluorescence of Anterior and Posterior Lens Capsular Plaques from the Same Eye. J Ophthalmol Adv Res. 2026;7(2):1-8.

https://doi.org/10.46889/JOAR.2026.7202

Received Date: 27-03-2026

Accepted Date: 28-04-2026

Published Date: 05-05-2026

Abstract

Purpose: Capsular plaques, the fibrotic lesions derived from Lens Epithelial Cells (LECs), are frequently observed with pediatric cataracts. Although anterior capsular plaques (ACP) and Posterior Capsular Plaques (PCP) are both well-known in clinical settings, there are few comparative studies examining them within the same eye. In the present study, we investigated ACP and PCP collected from the same patient with pediatric congenital cataract.

Methods: Plaque samples were obtained during cataract surgery and were processed to obtain wax sections. Sections were analyzed by Hematoxylin and Eosin (HE) staining for general morphology. Antibodies against collagen-I, collagen-IV, α-smooth muscle actin (αSMA), paired box protein 6 (Pax6), Proliferating Cell Nuclear Antigen (PCNA) and β-crystallin were used for immunofluorescence analysis. Nuclear counterstaining was done with DAPI.

Results: ACP appeared thin, weakly eosinophilic and composed of relatively few cells within an ECM-abundant, loosely organized Matrix (ECM). PCPs were thicker, more compact and densely packed with granular and fibrous ECM. In ACP, collagen-I was found to be evenly spread, whereas in PCP, it was limited to the fibrous extracellular matrix. Similarly, collagen-IV covered the entire ACP but was confined to the fibrous areas in PCP. α-SMA was expressed in nearly all cells, confirming epithelial-to-mesenchymal transition. Pax6-positive undifferentiated LECs were sparse in ACP but comparatively abundant in PCP near the capsular surface. β-crystallin localized to the posterior surface of ACP and the anterior surface of PCP, suggesting early fiber cell differentiation. PCNA revealed higher proliferative activity in PCP, correlating with greater thickness and clinical progression.

Conclusion: ACP and PCP exhibit distinct cellular and ECM profiles that may reflect divergent pathogenic tendencies. Our observations suggest that ACP may be more strongly associated with ECM remodelling, while PCP may be more influenced by cellular proliferation; however, these findings require validation in a larger cohort of samples.

Keywords: Anterior Capsular Plaque; Posterior Capsular Plaque; Epithelial–Mesenchymal Transition; Immunofluorescence; Extracellular Matrix; Pediatric Cataract

Introduction

Childhood cataracts are a significant cause of preventable blindness, with congenital cataracts accounting for a substantial proportion of cases [1-5]. Unlike adults, pediatric cataracts exhibit a substantial wound-healing response driven by the high

proliferative and migratory potential of Lens Epithelial Cells (LECs). These cells undergo an Epithelial-Mesenchymal Transition (EMT), transforming into myofibroblast-like cells that secrete Extracellular Matrix (ECM) components, leading to the formation of fibrotic plaques on the lens capsule [6,7].

Among the structural alterations observed in the pediatric lens, Anterior Capsular Plaques (ACP) and Posterior Capsular Plaques (PCP) represent distinct sites involving fibrotic remodeling [8,9]. The anterior capsule is bathed in aqueous humor. In contrast, the posterior capsule lies in proximity to the vitreous, which is rich in growth factors [10]. These differences in microenvironment suggest that the extent and nature of EMT and fibrosis may vary between ACP and PCP. Most existing studies have focused on anterior or posterior capsular plaques obtained from different patients [8,9,11]. In the present study, we have examined ACP and PCP obtained from the same eye, providing a unique opportunity for comparative analysis and eliminating inter-individual variation arising from genetic background and systemic conditions. Such a comparison can reveal site-specific differences in transdifferentiation markers, fibrotic changes and ECM deposition, thereby providing insight into the aggressive nature of pediatric LECs and their role in the development of Posterior Capsule Opacification (PCO).

Case Description

The study was conducted on a pediatric patient (male, 20 weeks old) with a non-traumatic mixed cataract, with no family history of pediatric cataract. The patient underwent congenital cataract surgery at Raghudeep Eye Hospital and the surgeon collected the capsule using the PCCC method. The type of cataract and the presence of ACP and PCP were determined using a slit lamp biomicroscope and an operating microscope (under anesthesia). The sample was collected from the hospital and further processed at the Iladevi Cataract and IOL Research Centre in Ahmedabad. Approval was obtained from the Institutional Ethics Committee and the specimen was examined in accordance with their recommendations. Informed consent was obtained from the subject.

The sample was fixed in 4% paraformaldehyde-PBS for 30 minutes and processed to obtain 5-µm-thick wax sections. After fixation, the sample was washed in PBS (pH 7.4), processed through an alcohol series for dehydration, embedded in paraffin and stained with Hematoxylin and Eosin (HE). It was then examined under a light microscope (Axioskope 2; Carl Zeiss, Gottingen, Germany). Photographs were taken with a Jenoptic camera (Genoptic, Germany). The sample was processed for immunofluorescence to localize collagen types I and IV, β-crystallin, PCNA, PAX6 and α-SMA. The plaque section was first dewaxed, then hydrated with a graded alcohol series and washed in PBS. Antigen retrieval was performed by incubating the samples in sodium citrate buffer in a microwave oven, followed by incubation in sodium borohydride to quench the autofluorescence. For collagen I, sections were treated with 1 mg/mL trypsin in 0.01M NaOH for 30 minutes at 37°C and for collagen IV, they were treated with 1 mg/mL pepsin in 0.01 M HCl for 30 minutes at 37°C [8]. Furthermore, the sections were washed thoroughly in PBS and the slides were immersed in PBS containing 1% bovine serum albumin and 3% normal goat serum for 15 minutes to reduce non-specific staining of the plaques. Incubation with the primary antibody diluted in PBS containing 0.1% BSA was performed overnight at 4°C. The primary antibodies were diluted 1:100: mouse anti-α-SMA, rabbit anti-β-crystallin, mouse anti-PAX6, mouse anti–collagen type I and mouse anti–collagen type IV. The sections were then rinsed in PBST (PBS containing 0.05% Tween 20) for permeabilization and washing and incubated with the appropriate secondary antibody conjugated to Alexa Fluor 488 or Alexa Fluor 546 (1:500 dilution) in PBS/BSA. For negative controls in each immunofluorescence study, sections were incubated without the primary antibody. All sections were further counterstained with DAPI for nuclear staining, rinsed thoroughly with PBST and mounted in DABCO. The sections were observed using an epifluorescence microscope (Axioskope II; Carl Zeiss) and images were captured with a cooled CCD camera (Cohu, San Diego, CA). For each parameter, 5 sections from different parts of the samples were stained and comparable regions were selected and scanned through a camera under a 10X objective. Regions depicting specific staining patterns were photographed under a 40X objective. The size of the given area was measured using BioVis Image Analysis Software (ExperVision Lab Pvt Ltd, Mumbai, India). Staining interpretation was performed by two independent observers and any discrepancies were resolved by consensus.

Morphology

The ACP appears as a well-demarcated, white, multifocal, fibrous, opaque area attached to the inner anterior surface of the lens capsule. The PCP was thick, white, dense and opaque, attached to the posterior inner surface of the lens capsule. To assess the gross morphology of the plaques, hematoxylin and eosin (HE) staining was performed on pediatric samples that exhibited both ACP and PCP (Fig. 1).

Both ACP and PCP are fibrous structures with distinct cell morphologies. The cells adjacent to the capsule exhibited an epithelial morphology, with cuboidal cells. Cells within the plaque area lost their epithelial morphology and acquired a more elongated, spindle-shaped appearance. ACP was located below the lens capsule, thin, weakly eosinophilic, ECM-abundant, with relatively few cells. PCPs were situated above the capsule, appeared thick and strongly eosinophilic and contained comparatively less abundant ECM than ACP. The ECM of ACP was non-homogeneous and less compact, while that of PCP was uniform and compact.

The ECM was either arranged in bundles or in a granular form. Cells suspended in a bundle form of ECM were spindle-shaped, while those in a granular ECM had oval nuclei. ACP and PCP consisted of large amounts of ECM and few cells. In ACP, the ECM was in bundles near the lens capsule, whereas it was granular towards the lens fibers. The bundle form of ECM was less eosinophilic, while the granular form of ECM was more eosinophilic.

Immunofluorescence Characteristics

An Immunofluorescence study was performed to determine the differentiation status of plaque cells and the nature of the ECM. Expression of proliferation, differentiation and Lens Epithelial Cell (LEC) markers in the ACP and PCP was detected by immunofluorescence using specific antibodies and nuclei were counterstained with DAPI (Fig. 2).

Collagen-I is an Extracellular Matrix (ECM) protein secreted by cells that have undergone transdifferentiation. Therefore, it was not surprising that both plaque types were positive for collagen type I. Consistent with the morphological findings, collagen-I was preferentially distributed in the fibrous ECM of both plaques, with weaker staining in the granular ECM of PCP. Hence, collagen-I was more uniformly distributed in ACP, whereas in PCP, it was parallel and evenly spaced between cells. Collagen-IV in the lens is expressed only in the lens capsule. Hence, the capsules of both plaques were clearly positive for collagen-IV. In addition to lens capsules, collagen-IV was present throughout the ECM of ACP, whereas in PCP it was restricted to the fibrous ECM. αSMA is a marker of transdifferentiation. Almost all cells of both ACP and PCP were positive for αSMA.

Pax6 is a transcription factor expressed in many ocular epithelial cells and present in undifferentiated LECs. A few cells of ACP towards the capsule and many cells of PCP towards the capsule and exterior were positive for pax6. PCNA was selected as a proliferation marker and both ACP and PCP contained many PCNA-positive cells, evenly distributed throughout the plaque. PCPs have more positive cells than ACPs. In ACP, PCNA-positive cells were predominantly located near the capsule, whereas in PCP, they were fewer in the basal region and more numerous in the upper area. β-Crystallin is a marker of LECs that starts undergoing differentiation into fibre cells. The posterior surface of ACP and the anterior surface of PCP contained cells positive for β-crystallin.

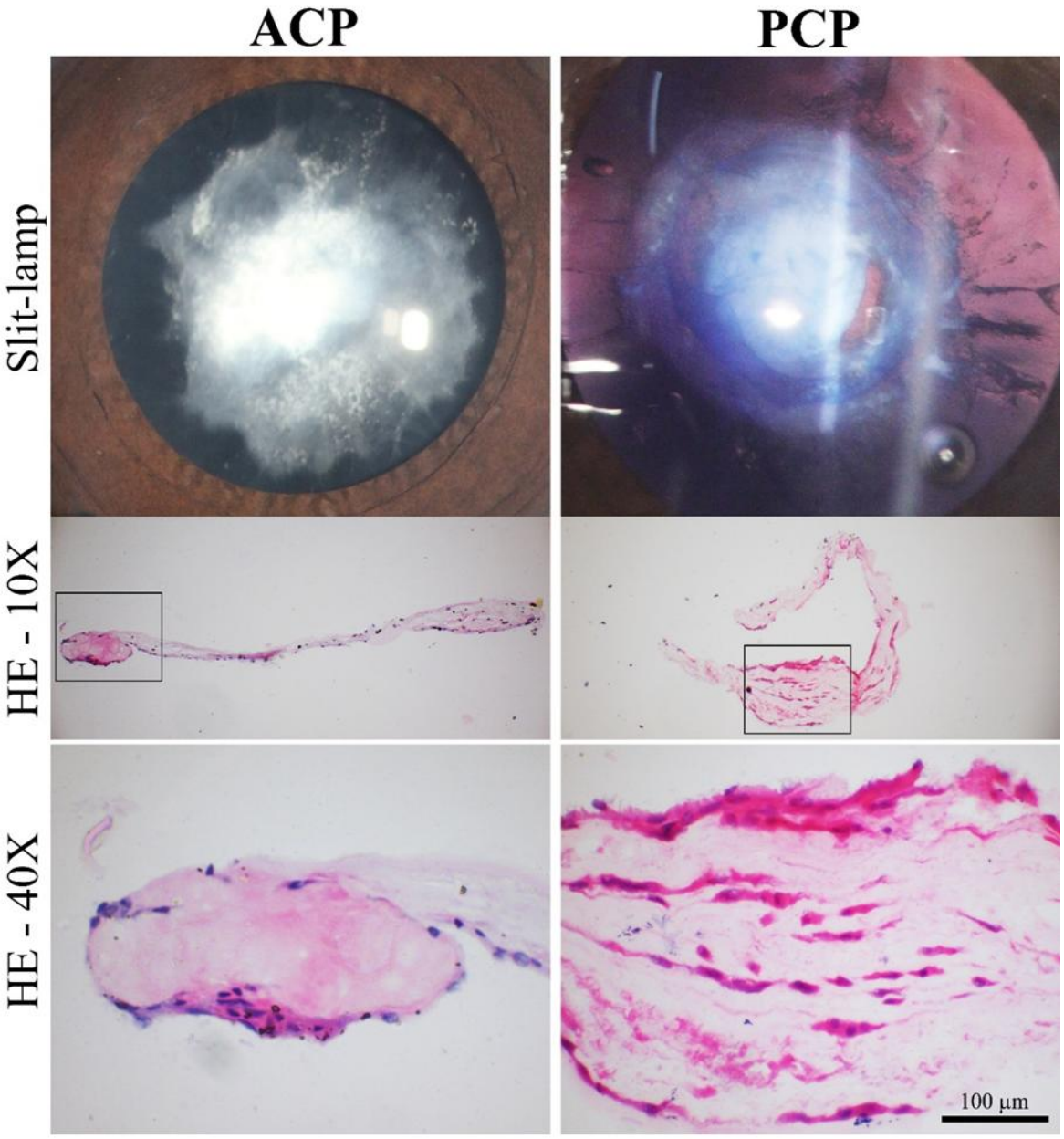

Figure 1: Slit lamp photographs of ACP and PCP and light microscope images of histological sections of the ACP and PCP. Anterior capsular plaque in the eye before the extraction of the cataract (Left side image) and posterior capsular plaque in the eye after the extraction of a cataract (Right side image). Hematoxylin-eosin (HE) stained sections of ACP and PCP. A square box in a 10X picture, further magnified under 40X. ACP and PCP consisted of large amounts of extracellular matrix (ECM) and few cells. In ACP, the ECM was in the form of bundles, whereas in PCP, the ECM near the lens capsule was in the form of bundles and towards the lens fibers, it was granular. The bundle form of ECM was less eosinophilic, while the granular form of ECM was more eosinophilic. Cells suspended in a bundle form of ECM were spindle-shaped, while those in a granular ECM had oval nuclei. Bar = 100 µm.

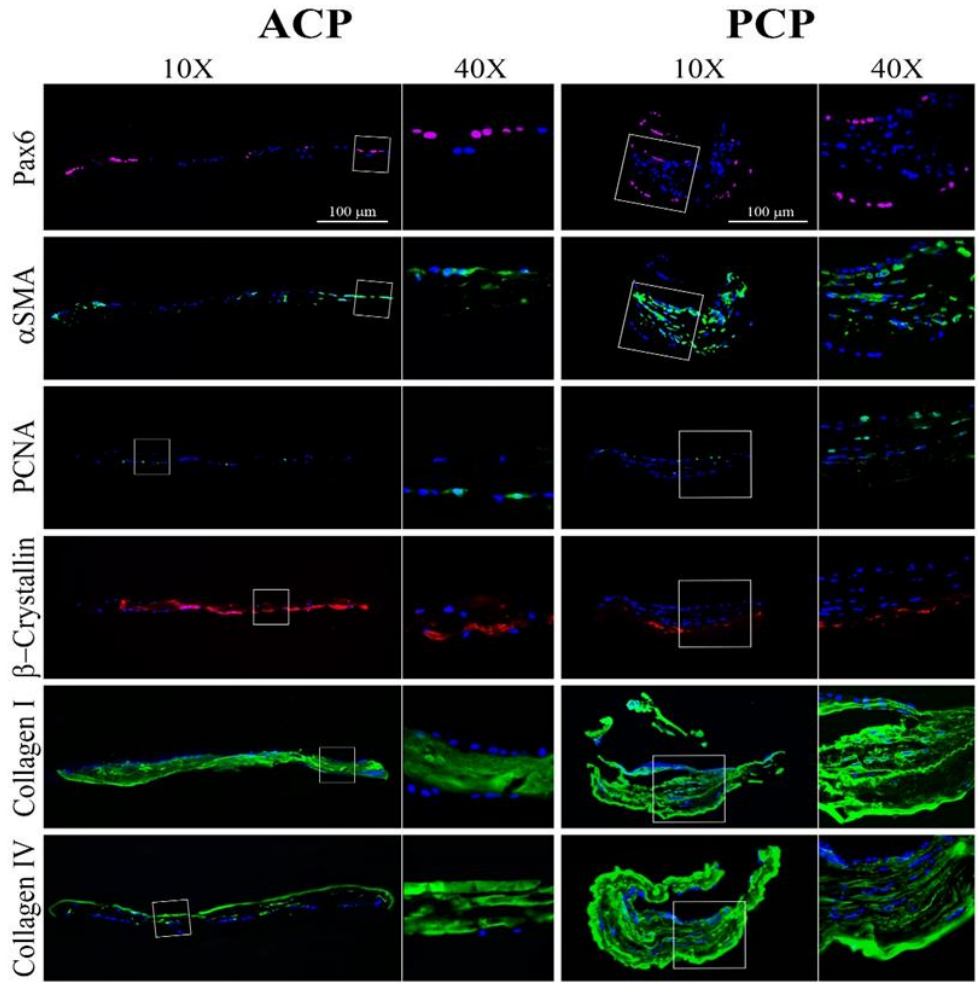

Figure 2: Immunofluorescence images of sections of the ACP and PCP. Expression of proliferation, differentiation and lens epithelial cells-related markers in the pediatric ACP and PCP. The expression of the marker protein was detected by immunofluorescence with specific antibodies and the nuclei were counterstained with DAPI. A square box in a 10X picture, further magnified under 40X. Pax6-positive undifferentiated LECs were few in ACP but more abundant in PCP near the capsule and surface. Most cells in both plaques were αSMA-positive, confirming transdifferentiation. PCNA staining indicated higher proliferative activity in PCP, with positive cells mainly in the upper region. β-Crystallin positivity at the posterior surface of ACP and the anterior surface of PCP indicated partial differentiation into fiber-like cells. Collagen-I was present in the fibrous bundles of both plaques, with uniform distribution in ACP and parallel arrangement in PCP, whereas the granular ECM of PCP showed weak staining. Collagen-IV was strongly expressed in the capsule of both plaques and throughout the ECM of ACP, but was restricted to fibrous regions in PCP. Bar = 100 μm.

Discussion

In the present study, ACP and PCP were characterised with respect to their morphology, ECM organization and expression of markers associated with epithelial cell differentiation, proliferation and transdifferentiation. It is important to note at the outset that this is a single-case observational study involving one pediatric patient and all findings should therefore be interpreted as preliminary and hypothesis-generating rather than definitive. While the paired collection of ACP and PCP from the same eye offers a unique and controlled comparative opportunity, the inherent limitation of a single case precludes generalization, statistical inference or mechanistic conclusions. These observations are intended to inform the design of future studies with larger cohorts.

Histomorphology examination indicates that ACP appeared beneath the lens capsule, thin, weakly eosinophilic and composed of relatively few cells embedded within an ECM-abundant, loosely organised matrix. In contrast, PCP were localized above the capsule, appeared thicker and more eosinophilic and contained more compact ECM. These morphological variations suggest a possible tendency toward greater ECM dominance in ACP and greater cellular proliferation in PCP; however, these observations

are derived from a single case and should be interpreted with caution as preliminary findings. The observation that cells near the capsule retained a cuboidal morphology, while cells within the plaques were spindle-shaped, indicates transdifferentiation or Epithelial-to-Mesenchymal Transition (EMT), a process previously implicated in lens fibrosis and secondary cataract formation [8,12]. The immunofluorescence profile in this case is consistent with a potentially more advanced stage of EMT in PCP compared to ACP, though this remains speculative without quantitative molecular validation. The posterior capsule lies adjacent to the vitreous body, which is reported to be rich in TGF-β and other cytokines that may promote EMT; however, a direct causal link between vitreous-derived factors and the PCP phenotype observed here cannot be established from the present data [10]. In contrast, the anterior capsule is primarily influenced by the aqueous humor, which provides a comparatively weaker stimulus for transdifferentiation and fibrosis [13]. However, most earlier reports compared anterior and posterior capsule changes in different eyes or different patient groups, making our paired analysis from the eye a more controlled assessment of site-specific differences, clarifying the pivotal role of transdifferentiation or EMT in driving the fibrotic remodelling processes at these distinct capsular sites [8,14].

Histochemical staining highlighted significant differences in ECM arrangement. These differences indicate distinct biochemical compositions of the ECM in ACP and PCP [14,15]. Such heterogeneity in the ECM underlies differences in plaque rigidity, transparency and clinical behavior. Immunofluorescence revealed differential distribution of collagen-I and collagen-IV between ACP and PCP. Collagen-IV, generally confined to the capsule, was detected throughout the ECM of ACP but restricted to fibrous ECM regions in PCP. These findings suggest that collagen remodeling occurs in a plaque-specific manner [14-16]. The near-universal α-SMA positivity across both plaque types is consistent with widespread myofibroblastic transdifferentiation, a hallmark of lens fibrosis previously reported in pediatric capsular plaques. However, differences emerged in the expression of Pax6 and β-crystallin. Pax6-positive cells were sparse in ACP but relatively abundant in PCP, suggesting, in this case, relatively greater Pax6 positivity in PCP, which may indicate retention of undifferentiated LECs, though this inference requires confirmation in larger samples [6]. In addition, β-crystallin expression at the posterior surface of ACP and the anterior surface of PCP suggests that cells at these interfaces initiate differentiation toward lens fiber cells. The proliferation marker PCNA was expressed in both ACP and PCP, though at different levels and with different distributions. ACP contained fewer PCNA-positive cells localized near the capsule, whereas the higher PCNA positivity in the upper regions of PCP suggests active cell expansion away from the capsular surface, potentially driving the greater thickness and clinical progression characteristic of posterior plaques [6]. In the sample, we did not detect any evidence of tunica vasculosa lentis. Collectively, these observations highlight a complex spectrum of cellular states within plaques, ranging from undifferentiated epithelial cells to trans-differentiated myofibroblasts and partially differentiated fiber-like cells.

Conclusion

The observed aggressive nature of posterior capsule fibrosis in this study supports the common surgical practice of performing a primary posterior capsulotomy with anterior vitrectomy in pediatric cataract cases to prevent visual axis obscuration. The findings of this study offer insights into site-specific fibrosis and the modulation of transdifferentiation or EMT by local microenvironments. Furthermore, the effects of distinct microenvironments and the differential behavior of anterior and posterior capsules highlight the need for localized pharmacological strategies, such as TGF-β inhibitors or other anti-fibrotic agents, to reduce the risk of capsule opacification in children. However, being a single case report, the findings cannot be generalized without caution. The absence of quantitative molecular data limits the ability to conclusively link these differences to specific signaling pathways. Future studies with larger pediatric cohorts and comprehensive immunofluorescence profiling of EMT and ECM markers are necessary to validate these observations and strengthen the mechanistic understanding.

Although both ACP and PCP originate from LECs, they exhibit distinct morphological and molecular features, suggesting divergent biological mechanisms underlying their formation and progression. In this single case, ACP formation appeared to be associated with more pronounced ECM deposition and diffuse collagen remodeling, whereas PCP showed comparatively greater cellular proliferation, retention of undifferentiated cells and a more compact ECM organization. These observations are preliminary and should be regarded as hypothesis-generating rather than mechanistically conclusive. Both processes involve EMT as a central mechanism, as evidenced by widespread α-SMA expression; however, the balance between proliferation, differentiation and ECM remodeling differs markedly between plaque types.

Conflict of Interest
The authors declared no potential conflicts of interest with respect to the research, authorship and/or publication of this article.

Funding Statement
This research did not receive any specific grant from funding agencies in the public, commercial or non-profit sectors.

Acknowledgement
The authors have no acknowledgments to declare.

Data Availability Statement
The data supporting the findings of this study are available from the corresponding author upon reasonable request.

Ethical Statement
The project did not meet the definition of human subject research under the purview of the IRB according to federal regulations and therefore was exempt.

Informed Consent Statement
Consent to publish this case report was obtained from the patient. This report does not contain any personal information that could identify the patient.

Authors' Contributions
Gaurang Gajjar: Writing – original draft, Investigation, Data curation, Conceptualization. Abhay Vasavada: Writing – review and editing, Supervision and resources. Kaid Johar SR: Writing – original draft, Investigation, Data curation, Conceptualization.

References
1. Johar SR, Savalia NK, Vasavada AR, Gupta PD. Epidemiology based etiological study of pediatric cataract in western India. Indian J Med Sci. 2004;58(3):115-21.
2. Sheeladevi S, Lawrenson JG, Fielder AR, Suttle CM. Global prevalence of childhood cataract: A systematic review. Eye (Lond). 2016;30(9):1160-9.
3. Tariq MA, Uddin QS, Ahmed B, Sheikh S, Ali U, Mohiuddin A. Prevalence of pediatric cataract in Asia: A systematic review and meta-analysis. J Curr Ophthalmol. 2022;34(2):148-59.
4. Dubucs C, Caillet A, Fremont F, Delteil L, N'Go V, et al. Prevalence of congenital ocular anomalies in 15 countries of Europe: Results from the Medikeye study. Birth Defects Res. 2024;116(11):e2414.
5. Anthony C, Ogieuhi I, Ajekiigbe V. Pediatric cataract in Nigeria: A scoping review of management, treatment outcomes and challenges. Egypt Pediatr Assoc Gaz. 2025;73:107.
6. Wernecke L, Keckeis S, Reichhart N, Strauß O, Salchow DJ. Epithelial-mesenchymal transdifferentiation in pediatric lens epithelial cells. Invest Ophthalmol Vis Sci. 2018;59(15):5785-94.
7. Wei Z, Gordon P, Hao C, Huangfu J, Fan E, Zhang X, et al. Aged lens epithelial cells suppress proliferation and epithelial-mesenchymal transition: Relevance for posterior capsule opacification. Cells. 2022;11(13):2001.
8. Johar K, Vasavada AR, Tatsumi K, Dholakia S, Nihalani B, Rao SS. Anterior capsular plaque in congenital cataract: Occurrence, morphology, immunofluorescence and ultrastructure. Invest Ophthalmol Vis Sci. 2007;48(9):4209-14.
9. Biró Z, Kereskai L, Tsorbatzoglou A, Vasavada AR, Berta A. Histological examination of primary posterior capsule plaques. J Cataract Refract Surg. 2007;33(3):439-42.
10. Wang Q, McAvoy JW, Lovicu FJ. Growth factor signaling in vitreous humor-induced lens fiber differentiation. Invest Ophthalmol Vis Sci. 2010;51(7):3599-610.
11. Özer F, İnanç İ, Kızıltunç PB, Atilla H, Can B. Histopathological findings of anterior lens capsule in pediatric cataract. Indian J Ophthalmol. 2024;72(12):1741-4.
12. Raj SM, Vasavada AR, Johar SR, Vasavada VA, Vasavada VA. Postoperative capsular opacification: A review. Int J Biomed Sci. 2007;3(4):237-50.
13. Ecker SM, Hines JC, Pfahler SM, Glaser BM. Aqueous cytokine and growth factor levels do not reliably reflect those levels

found in the vitreous. Mol Vis. 2011;17:2856-63.
14. Van Looveren J, Van Gerwen V, Schildermans K, Laukens K, Baggerman G, Tassignon MJ. Proteomic analysis of posterior capsular plaques in congenital unilateral cataract. Acta Ophthalmol. 2018;96(8):e963-9.
15. Azuma N, Hara T, Hara T. Extracellular matrix of opacified anterior capsule after endocapsular cataract surgery. Graefes Arch Clin Exp Ophthalmol. 1998;236(7):531-6.
16. Peng Q, Hennig A, Vasavada AR, Apple DJ. Posterior capsular plaque: A common feature of cataract surgery in the developing world. Am J Ophthalmol. 1998;125(5):621-6.