ophthal Focus
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Dear Doctor,
Greetings from Micro Labs Limited. We are happy to bring you 'Ophthal Focus' which contains latest and interesting news in the field of Ophthalmology.

This issue contains

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Best Regards,
Medical Team, Micro Labs Limited

The influence of Polymerase Chain Reaction (PCR) on infectious uveitis diagnosis and management

Uveitis is an inflammatory condition that affects the uveal layer of the eye and can cause blindness. [1] Ocular problems such as glaucoma, cataracts, and blindness are more common in people who have uveitis. To avoid ocular problems, it's critical to diagnose infectious uveitis early on and initiate an effective antimicrobial treatment. [2] The most prevalent infectious uveitis causes are toxoplasmosis and herpes viruses. In comparison to gold standards like culturing or fundus inspection, polymerase chain reaction (PCR) has emerged as a useful diagnostic tool for infectious uveitis, demonstrating high sensitivity and quick findings. [3] Multiplex and real-time PCR are the two main types of PCR. Multiplex PCR, often known as qualitative PCR, is a technique for determining whether or not an agent is present in a sample without determining its concentration. Multiplex and real-time PCR are the two main types of PCR. Multiplex PCR, often known as qualitative PCR, is a technique for determining whether or not an agent is present in a sample without determining its concentration. [3] Real-time PCR, commonly known as quantitative PCR, is a technique for determining disease burden. Real-time PCR helps identify if the pathogen is likely a contaminant or active in the illness process by measuring the quantity present. These two forms of PCR can be used simultaneously to assist identify and confirm a causal infectious agent. [4]

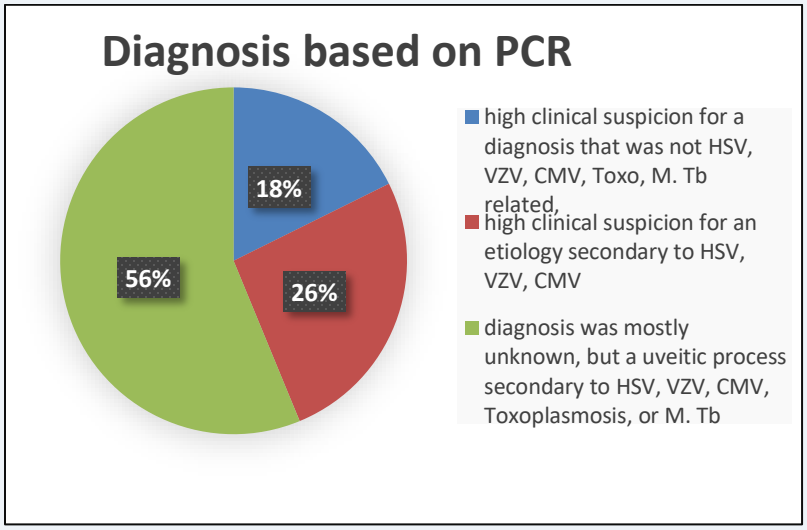
Fallon J et al. conducted a retrospective, observational study of patients with aqueous and vitreous PCR samples. Patients were deemed appropriate for inclusion if they had a sample of ocular fluid (aqueous or vitreous) sent for PCR testing. Aqueous fluid was removed from the eye via an anterior chamber paracentesis. Samples were sent to labs for Multiplex (qualitative) PCR analysis for detection of Herpes Simplex Virus (HSV), Varicella Zoster Virus (VZV), Cytomegalovirus (CMV), Toxoplasmosis gondii (Toxoplasmosis), and Mycobacterium tuberculosis (M. tb). Real-time (quantitative) PCR was used for the detection of Epstein-Barr Virus (EBV). A Chi-squared test was used to analyze the data.

- PCR testing is a well-accepted adjunct in the diagnosis of infectious uveitides
- PCR had the highest impact on diagnosis in cases where the pre-sampling diagnosis is broad.

137 cases were eligible for inclusion in this study. 21 cases were excluded, 20 of which did not meet the criteria for follow-up, at least one clinic visit following results of PCR testing, and 1 case that had an invalid toxoplasmosis PCR result. 116 cases with PCR studies were available for analysis. Only the first PCR sample sent from the patient was included leaving the final sample size with 108 cases.

Cases were classified depending on the initial diagnosis. There were 19 cases (17.6%) where there was high clinical suspicion for a diagnosis that was not HSV, VZV, CMV, Toxo, M. Tb related, examples of these include cases of presumed bacterial endophthalmitis or retinal detachments without underlying panuveitis. There were 28 cases (26%) where there is a high clinical suspicion for an etiology secondary to HSV, VZV, CMV, Toxoplasmosis or M. Tb. including 4 presumed cases of

Toxoplasmosis, 2 cases of Progressive Outer Retinal Necrosis, 7 cases of Acute Retinal Necrosis and 2 cases of CMV retinitis. The remaining 61 cases (56%) were cases where the diagnosis was mostly unknown, but a uveitic process secondary to HSV, VZV, CMV, Toxoplasmosis, or M. Tb was on the differential. Examples of unknown cases include recurrent anterior uveitis where a viral etiology was possible, cases that were acutely worsening for an unknown reason, and undifferentiated panuveitis where an infectious etiology was possible and needed to be ruled out before beginning immunosuppression.



- For the group where pre-testing diagnosis was not related to an infectious agent sampled with PCR, no cases had a diagnosis or treatment change based on the results of the PCR.
- For the group where pre-testing diagnosis was suspicious for an etiology related to PCR, 8 cases (29%) had a change in diagnosis related to PCR, and 9 cases (32%) had a change in management related to PCR result.
- In the group where diagnosis before sampling was unknown, 45 cases (74%) had a change in diagnosis based on PCR results and 20 cases (33%) had a change in management.

PCR has proven to be an asset to the diagnosis of infectious uveitides and had a more significant impact on diagnosis when pre-testing diagnosis was unknown, however, rates of treatment change were similar between those patients where pre-testing diagnosis was unknown and those patients where sampling was confirmatory in nature. A study conducted on 100 suspected cases of infectious uveitis using PCR concluded that PCR assay was an accurate technique with high sensitivity and specificity to diagnose the DNA genome in infectious uveitis. [5] A study conducted on suspected infectious uveitis using PCR and serological testing concluded that aqueous humor PCR can be a valuable diagnostic tool for confirming the infectious etiology in patients clinically diagnosed with uveitis. PCR had good predictive and diagnostic value for anterior uveitis and panuveitis compared with that for intermediate and posterior uveitis. [6]

In conclusion, PCR testing is a well-accepted adjunct in the diagnosis of infectious uveitides. Although there is no well-accepted practice pattern as to when to employ PCR testing this study

results show PCR had the highest impact on diagnosis in cases where the pre-sampling diagnosis was broad and uncertain, however even in cases where PCR was confirmatory in nature, it did still prove useful in regards to tailoring management.

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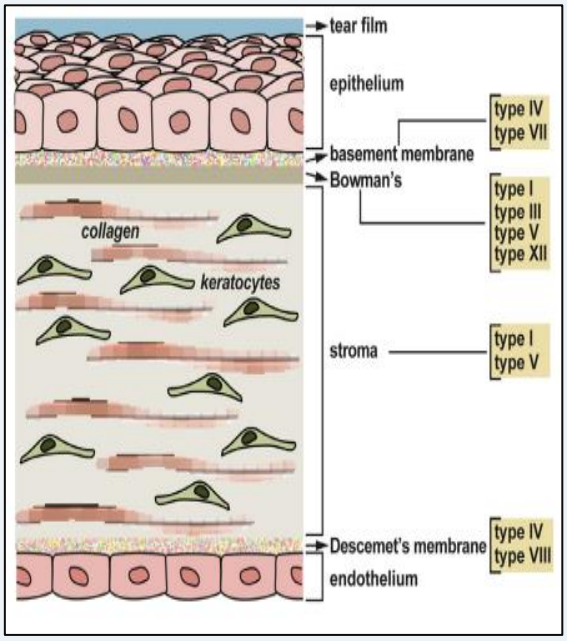
Corneal collagen as a prospective dry eye disease treatment target

Dry Eye Disease (DED) is a tear film condition caused by a lack of tears or excessive tear evaporation, which results in damage to the interpalpebral ocular surface and ocular discomfort. [1] Tear film instability in DED both causes and is exacerbated by disruption of the corneal epithelium. A broader definition specifies disruption in tear homeostasis accompanied by ocular surface inflammation, structural damage, and neurosensory abnormalities. [2] DED is linked to instability and hyperosmolarity in the tear film. These set into motion a cycle of tissue damage involving pro-inflammatory mediators (cytokines and chemokines), matrix metalloproteinases, antigen-presenting receptors, and immune cell infiltration (lymphocytes, macrophages, and dendritic cells) that implicate (from human patient studies) a degree of autoimmunity in DED. [3]

A characteristic feature of all collagen is its triple helical structure – a set of three polypeptide chains consisting of glycine-x-y repeating sequences where x and y often (but not always) account for proline and hydroxyproline. [4] Collagen includes both fibrillar (I, II, III, V, XI) and non-fibrillar types, which themselves form different classes (for example, the basic membrane type IV and short-chain types VIII and X). Type I collagen is the most common, making up about 90% of the collagen in the human body, followed by type II-V. In a healthy eye, the subsurface membrane is renewed with basal epithelial cells as they regenerate and move from the outer edge of the cornea. The epithelial basement membrane is comprised primarily of four components: collagens, laminins, proteoglycans, and nidogens.

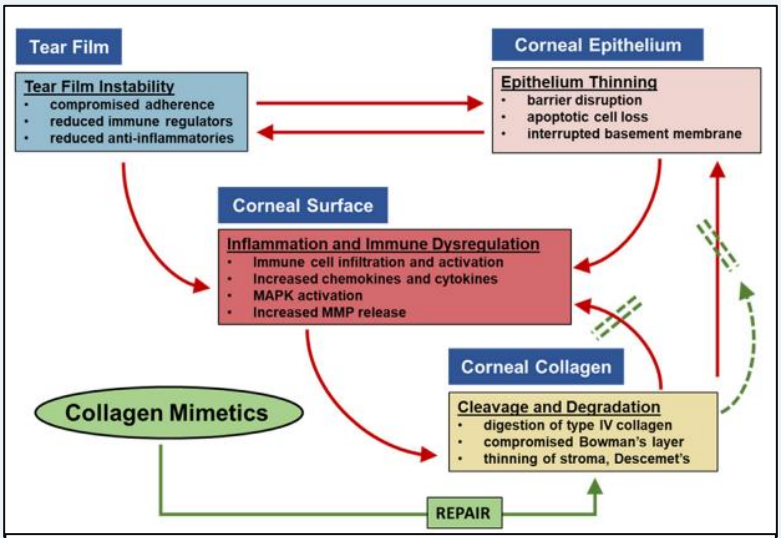
> In DED and other disorders that harm the ocular surface, mending fractured collagen in the cornea could eventually prove to be a more efficient way to break the never-ending cycle of inflammation.

In DED, apoptotic loss of conjunctival goblet cells decreases mucin biosynthesis and disrupts the protective barrier on the apical epithelium and the superficial epithelial microvilli. This reduction also depletes the tear film in critical immune regulators such as the transforming growth factor β2 (TGF-β2) that inhibits activation of dendritic cell antigen presentation on the eye surface. The lacrimal film contains several additional anti-inflammatory factors, including matrix metalloprotein tissue inhibitor (TIMP-1), a glycoprotein that neutralizes the action of MPMs in the degradation of the extracellular matrix and has anti-nociceptive effects. [5] A secondary equally pathogenic consequence of DRD is the thinning of the corneal epithelium, the degree of which is a reflection of the severity of the illness. Damage to the lower lining of the epithelial layer allows entry of inflammatory cytokines, chemokines in addition to other signaling molecules (including TGF-β) with the cornea stroma that can induce differentiation of myofibroblasts precursor cells, favoring haze, opacity, and surface irregularity. Multiple local inflammatory and immune-activating reactions result in enhanced MMP release in DED epithelial lesions caused by persistent desiccation.



Distribution of collagen in cornea

Increased levels of MMPs, cytokines, and chemokines, all of which have secondary pro-inflammatory effects that often cascade and magnify each other through positive feedback, are inflammatory as a result of events that disturb the corneal surface in DED. A recent review of clinical and experimental trials summarises these connections. [6] While it is widely accepted that inflammation in DED affects the integrity and function of cellular structures (conjunctival goblet cells, corneal epithelium, stromal keratocytes, and so on), inflammation may also directly challenge the integrity of the collagen substrate, which serves as the cornea's backbone and the foundation for the epithelial cell layer's integrity. MMPs generated by damaged corneal epithelium in DED could have acute surface consequences. MMP-9, as well as the stromelysins MMP-3 and MMP-10, can digest-type IV collagen, which is essential for the integrity of the epithelial basement membrane. [7] Intact collagen in its stable triple helix structure is



Corneal Collagen as an Inflammatory Mediator and Therapeutic Target

incorporated into the extracellular matrix during normal turnover, presenting multiple binding sites that act as ligands for a variety of cell surface receptors, including integrins, discoidin domain receptors, glycoprotein VI, and proteoglycan receptors. When these receptors bind, they influence other molecules and cellular pathways involved in remodeling, inflammation, and immunological control. Apart from their structural significance as scaffolding proteins, nearly all collagens activate particular receptors on the cell surface to trigger cellular signaling cascades.

Intact collagen, on the other hand, supplies ligand sites for signaling pathways implicated in inflammation, such as the leukocyte receptor complex (LRC), which is made up of a varied array of cell surface receptors produced largely by immune cells. The leukocyte-associated immunoglobulin-like receptor or LAIR-1, which is the most highly expressed gene in human Meibomian gland cells, is a member of this category. When it binds to ligand sites on intact triple-helical collagen, LAIR-1 (also known as CD305) inhibits the activation of most immune cells, including T cells, B cells, mast cells, eosinophils, basophils, and monocytes/macrophages, while decreasing proinflammatory cytokine production. The activation power of the crosslinking interaction with collagen determines the strength and efficiency of inhibitory signals in LAIR-1. Immune cells are inhibited by triple-helical collagens crosslinking LAIR-1 at glycine-proline-hydroxyproline repeats (GPO10), whereas reduced LAIR-1 or its binding sites in disturbed collagen enhance immune cell activation and lead to persistent tissue inflammation. 38 Collagen degradation results in the formation of significantly shorter triple helical proteins, as well as single-strand fragments. Collagens, by binding LAIR-1, set a threshold for immune cells entering injured tissue to be inhibited or activated based on the number of intact binding sites.

MMP-induced destruction of collagenous structures in the cornea during DED may limit the binding of important modulators of inflammation, thereby exacerbating the damage cycle and obstructing ocular surface regeneration. Type IV collagen, which makes up the epithelial basement and Descemet's membrane, is one of the highly glycosylated collagens, and its fragmentation encourages the endocytic cell-surface receptor uPARAP/Endo180 to bind to it (urokinase plasminogen activator receptor-associated protein28). According to tumor cell research, this receptor belongs to the macrophage mannose receptor family of endocytic transmembrane glycoproteins and is involved in the uptake and lysosomal destruction of collagen fragments formed by initial MMP-mediated breakage. uPARAP/Endo180 also aids fibroblast adhesion and migration along with fibrillary collagen. This mechanism is involved in the production of corneal scars, a common dry eye problem. Finally, in vitro studies using fibroblasts show that type I collagen fragments (found in the Bowman layer and stroma) promote the release and activity of IL-1, a cytokine that aids in the recruitment and motility of pro-inflammatory immune cells and has been linked to corneal damage during human DED. [7]

Small biomolecules that target ligand binding sites on collagen as potential treatments are being found as well, including areas linked to tumor growth (such DDR1 and DDR2) and inflammatory signaling. Collagen peptides have shown promise in a variety of domains for healing fractured or partially digested collage. In DED and other disorders that harm the ocular surface, mending

fractured collagen in the cornea could eventually prove to be a more efficient way to break the never-ending cycle of inflammation.

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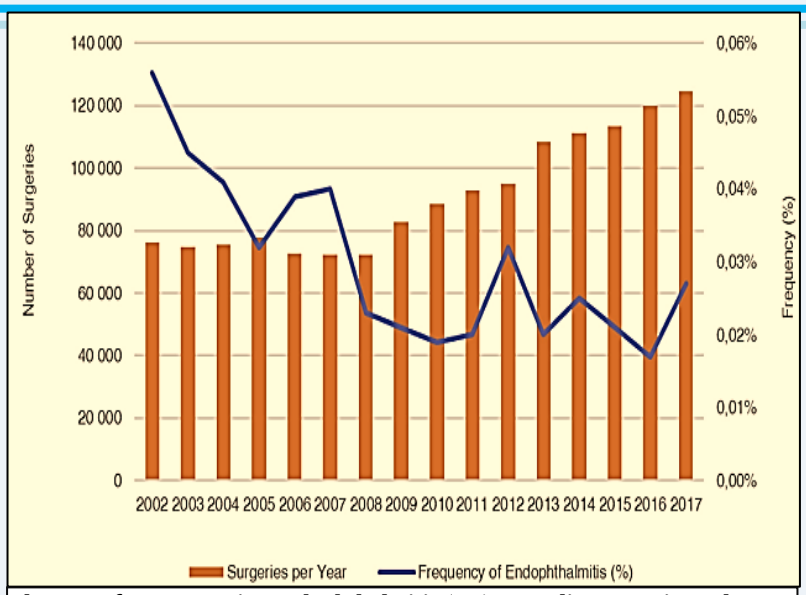
Immediate Sequential Bilateral Cataract Surgery: Postoperative Endophthalmitis

Simultaneous bilateral cataract surgery (SBCS), also known as immediately sequential cataract surgery (ISBCS) in recent years, is controversial, yet it is becoming more common in wealthy countries. [1] Among the main arguments for this approach are increased patient convenience, faster total eyesight recovery, and logistical advantages translating into cost savings. Conversely, there remains a worry of sight-threatening complications such as postoperative endophthalmitis (PE), which could occur bilaterally in the worst-case scenario. [2] The current study was a retrospective cohort registry study where patient data was collected from 14,57,172 cataract extractions, including 13,64,934 unilateral surgeries and 92,238 ISBCSs. In terms of patient characteristics, surgical approach, and capsule complication, endophthalmitis cases reported to the Swedish National Cataract Register (NCR) over 16 years (2002-2017) were compared to all control cases. Incidence and determinants of PE in ISBCS compared to unilateral operations were the main outcome measures.

In total, 422 cases of PE were found in 14,57,172 cataract extractions, resulting in a 0.029 percent overall incidence. The rate for unilateral procedures was 0.0299 percent, or 408 cases out of 13,64,934 operations, whereas the rate for ISBCS was 0.0152 percent or 14 events out of 92,238 operations. Non-use of intracameral (IC) antibiotics (ABs), capsule problem, age 85 years or more, male gender, and ocular comorbidities were determined to be independent risk variables for PE in a logistic regression model that

The study shows reduced incidence of endophthalmitis with immediately sequential cataract surgery (ISBCS).

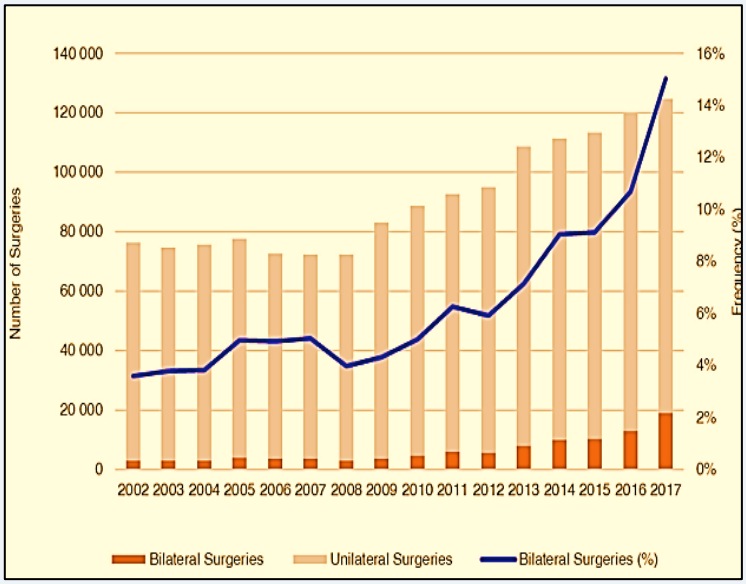
included all cataract surgeries. In ISBCS, all of these factors were less common. Despite this, ISBCS was found to be associated with a considerably decreased risk of PE in the same multivariate analysis. 5 of the 14 PE cases in the ISBCS cohort had a visual acuity (VA) of 20/200 or worse at the follow-up. One of the ISBCS patients, a 93-year-old woman, developed a bilateral infection.



The rate of postoperative endophthalmitis (PE) according to registry data

High-volume registry data are highly useful for researching rare events like PE after cataract surgery, especially after ISBCS, but are less common in many parts of the world. To get a more solid number of ISBCS procedures, the data from 2002-2010, which had already been reviewed in two studies, [3,4] was combined with data from 2011-2017. Apart from ISBCS, where 66 percent of surgeries were performed in the later period, surgical volume figures were nearly equal between eras.

A study [5] conducted reported a healthy 81-year-old woman got PE four days following her operations, which were performed as separate procedures and ended with prophylactic IC cefuroxime. The same strain of Staphylococcus epidermidis susceptible to vancomycin was grown in intraocular samples from both eyes. The patient's vision finally improved to 6/9 on both sides. Employing operational equipment from the same sterilization cycle without using chemical indication verification was a violation of the separation protocol suggested by the International Society of Bilateral Cataract Surgeons, according to correspondence regarding this case. The authors countered that the decontamination method followed industry norms, making it an improbable cause of infection. The bacterium was thought to have originated from the patient's own ocular flora. The incidence described in this study is strikingly similar to the one described in this.



Evolvement of cataract surgery volumes demonstrating numbers of unilateral and immediate sequential bilateral surgery (ISBCS)

Capsule complications is a well-deservedly feared side effect because it raises the risk of a number of vision-threatening consequences, including cystoid macular edema, retinal detachment when compared to all PE. The low rate of capsule complication seen in ISBCS could be due to a careful selection of subjects with a low risk of developing this complication. Another factor contributing to the low frequency could have been abandoning the intended ISBCS if a capsule problem developed during the first eye operation; however, the registry does not indicate whether the complication occurred in the first or second eye in ISBCS instances.

Patient selection may have contributed to the reduced incidence of endophthalmitis in ISBCS in this study. However, in the logistic regression model, performing ISBCS per se was found to be a protective factor against PE, implying that this was attributable to practice tactics not considered in this model. ISBCS is more likely to be performed by high-volume surgeons, and their expertise in doing non-traumatic surgery and creating well-sealed wounds may have had a role.

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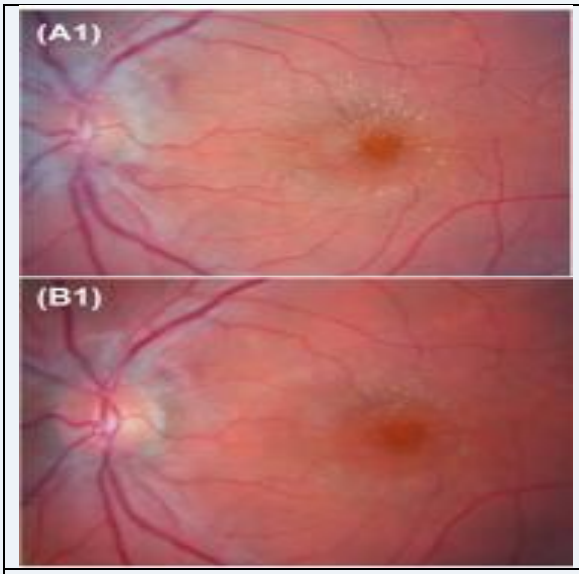
Isolated primary herpes-simplex virus neuroretinitis in an immunocompetent adult: a case report.

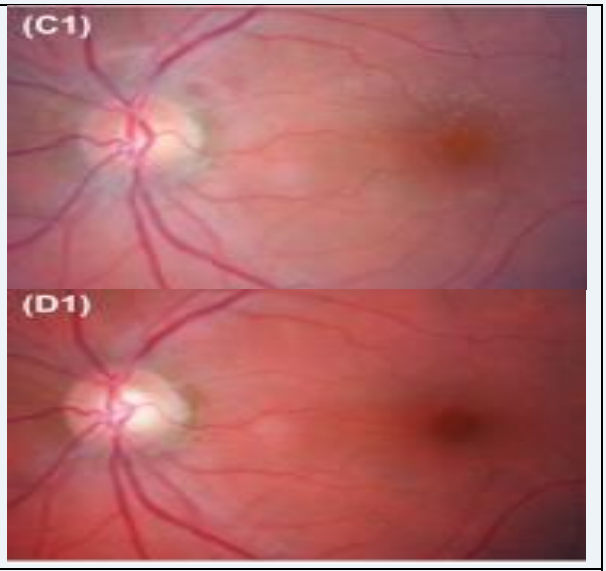
Herpes simplex virus (HSV-1 and HSV-2) are common viruses that can cause conjunctivitis, keratitis, and other less common ocular conditions such as acute retinal necrosis syndrome or neuroretinitis. [1] A case of isolated unilateral neuroretinitis as a symptom of a primary HSV infection in an immunocompetent adult without encephalitis or other clinical signs was given in this report.

A 60-year-old immunocompetent female reported scotoma and painless central vision loss in her left eye. She had no substantial medical or surgical history. Her medical history indicated that she had cataract surgery in both eyes a year earlier and had suffered a

myocardial infarction 9 months prior to her appointment. In the right eye, the best-corrected visual acuity (BCVA) was 20/20, whereas, in the left eye, it was 20/200. There were no typical herpetic skin lesions found on a general inspection. Optic disc edema and juxtapapillary hemorrhages were discovered during an ocular examination in the left eye. There were no additional indicators of intraocular inflammation, and the right eye had no distinguishing traits.



Color retinography images showing a macular star (A1) and its progressive reduction (B1 at 2weeks



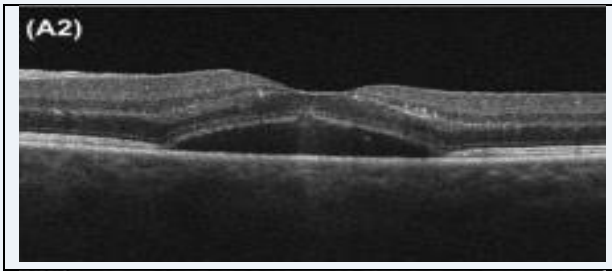
C1 at 1month, and D1 at 3month.

The initial approach was made in an ischemia situation because of its properties. The patient's history, supplementary testing, and medical evaluations were used to rule out both arteritic and non-arteritic anterior ischemic optic neuropathy (AION). The absence of extraocular clinical symptoms and a normal erythrocyte sedimentation rate ruled out arteritic AION. The likelihood of a non-arteritic AION was explored, and patients were closely monitored. The Goldmann visual field assessment two weeks later indicated no abnormalities. Optic disc edema, submacular fluid, and the formation of a macular star were discovered on the fundus of the left eye. A serious retinal detachment was discovered on macular optical coherence tomography (OCT).

The presence of a macular star in the fundus aided in the development of a novel differential diagnosis that includes hypertensive or radiation-induced retinopathy, as well as secondary or idiopathic neuroretinitis. There was no previous history of arterial hypertension or ionizing radiation exposure. The neuroretinitis course was

The presence of HSV IgM in the absence of other laboratory results may be sufficient evidence to begin HSV therapy, In immunocompetent individuals with a macular star as a solitary lesion

thoroughly investigated, with clinical and biochemical studies were out. Contact hazards were extensively investigated, including eating habits, recent travel, sexual experience, and animal encounters. A thorough physical check was conducted to look for rashes or inoculation spots, but the results were unimpressive.



Optical coherence tomography images showing serous retinal detachment (A2), and its progressive resolution (B2) at 2weeks

C2 at 1month, and D2 at 3month

In terms of laboratory testing, Bartonella, Epstein-Barr virus, Rickettsia, Lyme disease, histoplasmosis, syphilis, chlamydia, human immunodeficiency virus, toxoplasmosis, brucellosis, viral hepatitis B and C, HSV-1, and HSV-2 were all conducted. The only clinical anomaly was a positive immunoglobulin M (IgM) for HSV-1, which permitted etiological identification in the absence of any other systemic clinical characteristic. After starting a therapy schedule of 1 g of valacyclovir every 8 hours, we saw a clinical improvement after 2 weeks, with a decrease in symptoms and gradual clearance of fundus indications.

After 48 hours of antiviral therapy, a 6-week oral tapering prednisone regimen was started, and valacyclovir was reduced to 1 g per day until the oral prednisone was stopped (length of antiviral treatment of 10weeks). The therapy was well tolerated, and there were no side effects. Over the course of three months, the patient healed with no signs of intraocular inflammation and a BCVA of 20/25.

When more prevalent etiologies like Bartonella henselae have been ruled out, uncommon pathogens should be explored. In immunocompromised people, infectious neuroretinitis is rather prevalent. In a primary HSV infection, this report described an uncommon instance of neuroretinitis in an immunocompetent patient with no additional systemic symptoms or indications. When a patient exhibits typical HSV infection symptoms, a clinical diagnosis may be sufficient. Biological diagnostics, such as blood testing for particular antibodies or

nucleic acid amplification tests, might be effective in an immunocompetent patient with unusual signs like neuroretinitis. A PCR test can be very helpful in confirming an HSV diagnosis. It's worth noting that the PCR test on ocular fluids has been shown to be less reliable in immunocompetent individuals than in immunosuppressed patients. [2]

In immunocompetent individuals, a negative PCR result cannot confidently rule out infection, whereas a positive result can clearly confirm it. Unfortunately, we were unable to run a PCR test on our patient during the acute stage, therefore we had to make the diagnosis based on positive IgM titers and negative IgG titers for HSV-1. This was further corroborated by the positive antiviral treatment response, which was similar to that seen in previously reported instances in immunocompetent individuals. [3]

In immunocompetent individuals with a macular star as a solitary lesion, the presence of HSV IgM in the absence of other laboratory results may be sufficient evidence to begin HSV therapy after ruling out alternative non-infectious reasons such as arterial hypertension or ionizing radiation exposure. Even if there are no other usual symptoms or indications that may imply the disease, rare infectious agents in immunocompetent patients must be investigated in the differential diagnosis of neuroretinitis.

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