

# In vivo confocal microscopy in bilateral herpetic keratitis: A case report

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**PURPOSE.** *Bilateral herpetic keratitis is a rare infection of the cornea. Ocular herpes in atopic patients is more often bilateral, with more frequent recurrences. The authors report a case of bilateral herpes simplex virus (HSV) keratitis during the active phase studied by in vivo confocal microscopy.*

**METHODS.** *A 28-year-old man with 5 years history of unilateral HSV keratitis and atopic dermatitis was referred to the authors for a clinical and diagnostic evaluation.*

**RESULTS.** *The corneas showed the typical features of dendritic HSV keratitis in both eyes. Examination by in vivo confocal microscopy demonstrated similar lesions in both eyes: a distortion of the superficial and basal epithelium and the presence of irregular hyperreflective structures and dendritic particles near the epithelial cells. The subbasal nerve plexus presented a tortuous appearance with hyperreflective areas and beadlike formations along the fibers. After a week of antiviral treatment, in vivo confocal microscopy examination demonstrated an irregular epithelium with highly reflective deposits and reflective areas. A reduction of nerve fiber bundles with a large number of beadlike formations and abnormal tortuosity was also noted.*

**CONCLUSIONS.** *In vivo confocal microscopy enables a noninvasive evaluation of the ocular surface at a high magnification level. It could be useful for the early and differential diagnosis of corneal infections and when HSV keratitis recurrence is suspected. (Eur J Ophthalmol 2008; 18: 994-7)*

**KEY WORDS.** *In vivo confocal microscopy, Bilateral herpetic keratitis, HRTII*

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## INTRODUCTION

Herpes simplex virus (HSV) keratitis is the most common infectious cause of visual loss in many countries. It is characterized by a recurrent nature. The primary infection may be subclinical in most cases. In general, herpetic eye disease is characteristically unilateral. Simultaneous bilateral development of herpetic keratitis is very rare. It has been described in patients with atopy (1) and in patients with an altered immune system (2). The involvement of both eyes occurs more often in young patients.

In vivo confocal microscopy (IVCM) is becoming a useful diagnostic tool for corneal imaging.

Rosenberg et al described the corneal pathologic changes in patients with a history of HSV keratitis studied by IVCM (3).

We report a case of bilateral HSV keratitis during the active phase studied by IVCM.

## Case report

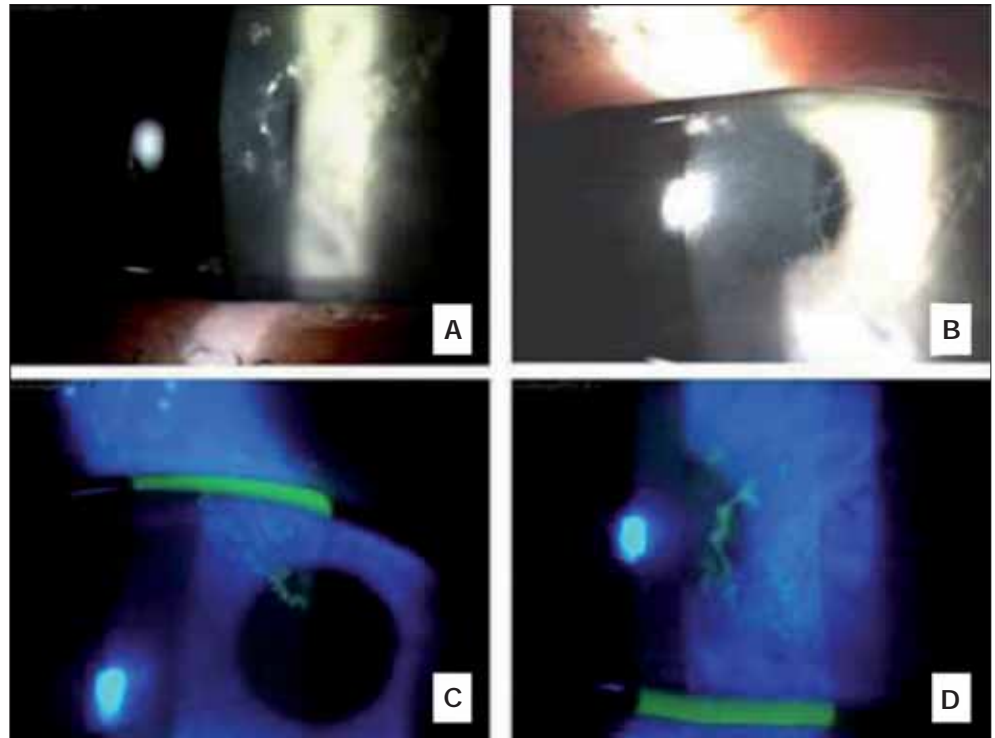
A 28-year-old man complaining of progressive pain, photophobia, and decreased vision in both eyes was referred to us for a clinical evaluation. The patient had a history of unilateral HSV keratitis in the left eye (LE) and of atopic dermatitis. He reported three recurrences in the LE in the last 2 years.

At the moment of the visit the patient did not use topical or systemic therapy.

The best-corrected visual acuity was 40/200 in the right eye (RE) and 20/200 in the LE.

Slit-lamp microscopy showed the typical features of HSV

**Fig. 1** - Slit lamp examination of right (A, B) and left (C, D) eye. A dendritic fluorescein-positive corneal ulceration is present.



keratitis including dendritic ulcer with underlying subepithelial infiltrate, corneal edema, and conjunctival inflammation in both eyes. The lesions stained diffusely with fluorescein dye. The corneal lesions were similar in both eyes (Fig. 1, A–D). Therefore the clinical diagnosis of bilateral HSV keratitis was made.

Tear samples had been taken from both eyes to perform the polymerase chain reaction and to amplify and detect HSV DNA. This examination from both eyes was positive and confirmed diagnosis of bilateral HSV keratitis.

The central cornea and the paracentral area of both corneas were examined using the HRT II Rostock cornea module (Heidelberg Engineering GmbH, Heidelberg, Germany). Before the examination, a drop of topical anesthetic (oxybuprocaine chlorohydrate) and a drop of polyacrylic gel 0.2% (Viscotirs Gel, Medivis, Catania, Italy) was instilled into the lower conjunctival fornix.

Examination by IVCM demonstrated similar lesions in both eyes. In particular, the superficial and basal epithelium adjacent to the lesions appeared more irregular and distorted. Many irregular hyperreflective structures were also evident near the epithelial cells (Fig. 2A). The majority were circular or oval, had poorly defined edges, and ranged between 10  $\mu\text{m}$  and 50  $\mu\text{m}$  in diameter. These particles were also found inside and around large hyporefective areas. Lin-

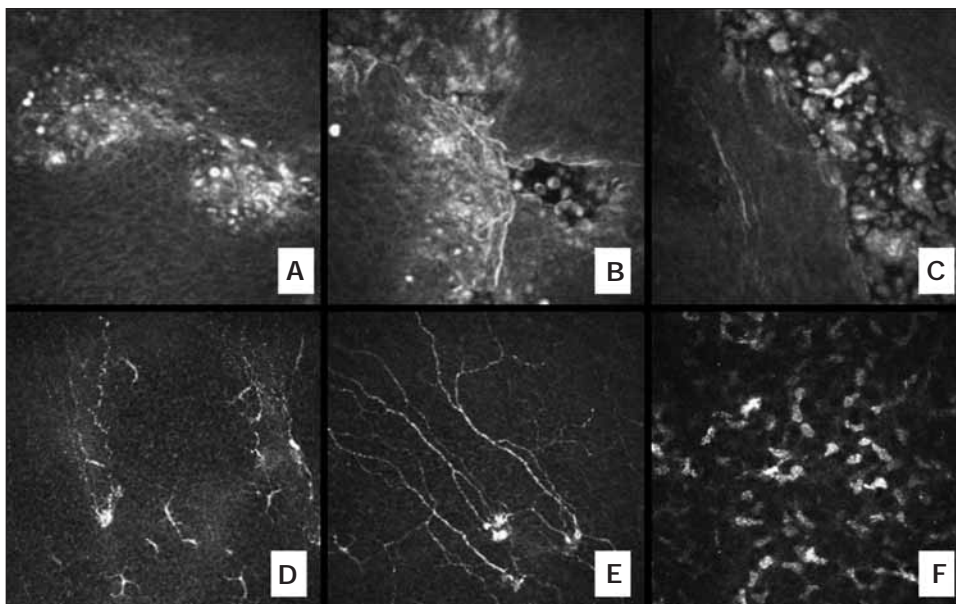
ear hyperreflective areas were also present at the level of the edges of the lesions (Fig. 2, B and C). Dendritic particles were found at the level of the basal epithelium of both eyes (Fig. 2D). The sub basal nerve plexus had a tortuous appearance with hyperreflective, irregular areas measuring approximately 50  $\mu\text{m}$  in diameter along the nerve. Moreover, some beadlike formations were observed (Fig. 2E). Many hyperreflective activated keratocytes were visible in the anterior stroma (Fig. 2F).

The deeper stroma and the endothelium were normal.

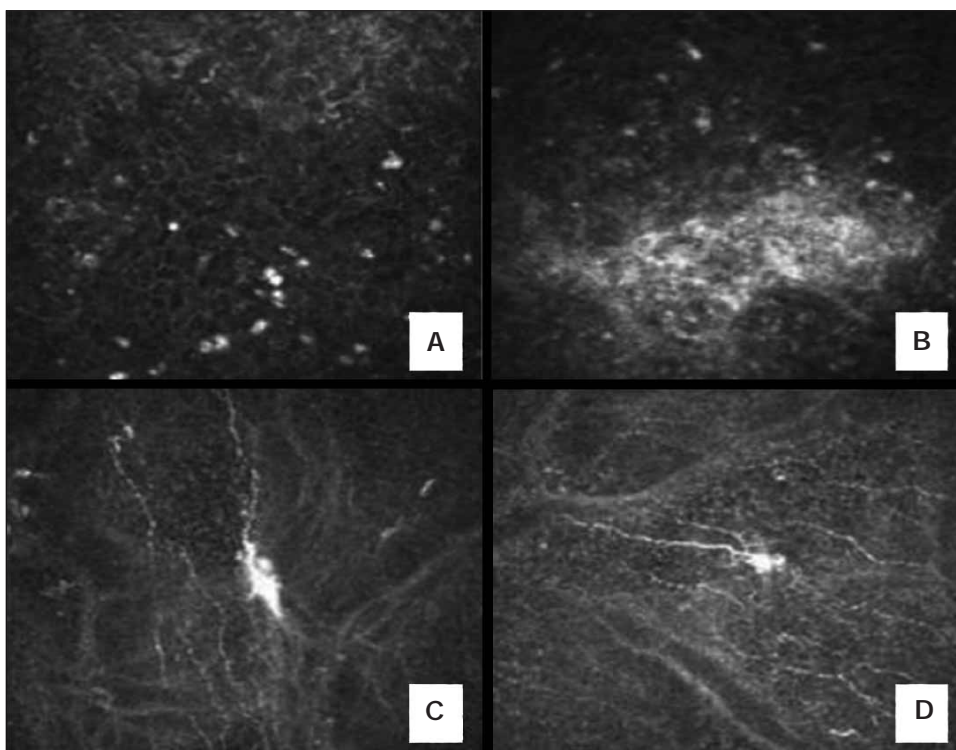
The patient was initially treated with 800 mg of acyclovir orally five times daily and with topical acyclovir ointment three times daily and tear substitutes.

After a week, biomicroscopic examination showed that the corneal epithelial defect in both corneas was completely resolved without fluorescein staining.

IVCM examination demonstrated that the surface epithelium was irregular with highly reflective deposits between the cells. In the LE an irregular reflective area was found at the level of the basal epithelium (Fig. 3, A and B). A reduction of long nerve fiber bundles with a large number of beadlike formations and abnormal tortuosity was noted. Along the nervous fibers some hyperreflective zones of wide diameter were still visible (Fig. 3, C and D).



**Fig. 2 -** *In vivo* confocal micro-scopic examination. Irregular and distorted epithelium with highly reflective deposits located between the cells (30  $\mu$ m) (A). Many highly reflective formations in various sizes and linear hyper-reflective areas were located at the level of the edges of the herpetic lesions (40  $\mu$ m) (B, C). Multiple non-epithelial cells, possibly Langerhans cells, were observed anterior to Bowman's layer (60  $\mu$ m) (D). Abnormal nerve appeared very reflective and tortuous with reflective deposits around the fibers (65  $\mu$ m) (E). Many hyperreflective keratocytes were found in the anterior stroma (90  $\mu$ m) (F).



**Fig. 3 -** *In vivo* confocal micro-scopic examination after antiviral treatment. Reflective particles (A) and hyper-reflective irregular scar tissue (B) extending from basal epithelium to the Bowman membrane was visible in left eye (50  $\mu$ m). Hyperreflective zones of wide diameter were still present along the fibers (60  $\mu$ m) (C, D).

## DISCUSSION

IVCM has been effectively applied to diagnose and describe corneal disease. It can provide details of structures of anterior segment at the cellular level.

To our knowledge, this is the first time that IVCM has been

used to investigate acutely HSV-infected corneas. This specimen was obtained from a patient with a classical dendritic ulcer clinically diagnosed as HSK. Rosenberg et al described the corneal pathologic changes by IVCM in patients with a history of HSV keratitis with intact epithelium (3).

In acute epithelial disease, dendritic ulceration has been con-

sidered a pathognomonic finding that often leads to the diagnosis.

Using IVCN, we found at the epithelial level the presence of a distortion of the superficial and basal cells around the lesion. Moreover, many hyperreflective particles of irregular shape and dimension were present inside and around the lesions. These particles could be the numerous cells involved in the process of an immunologic response against the herpes virus. In fact, many studies have shown that the disease is the result of a cocktail of inflammatory cells consisting of polymorphonuclear leukocytes, macrophages, and T cells (4). Thiel et al described through impression cytology of herpetic simplex keratitis in rabbits that corneal lesions consisted mainly of round epithelial cells, inflammatory cells, ballooning cells, multinucleated giant cells, and various inclusion bodies. In the recovery phase, the number of detached cells and infiltrated inflammatory cells decreased (5).

The dendritic structures described by impression cytology are also involved in the cornea's immune response. In our case report, the patient during the active phase showed small highly reflective dendritic structures in or under the basal epithelium and between the nerve fiber bundles in both eyes. For their size and shape we have proposed that these particles could represent Langerhans' cells (LCs) (6). The number of these cells after a week clearly is reduced. Even if LCs are not totally specific for viral disease, the visualization of these cells could be helpful for the clinician when deciding between a diagnosis of herpetic or bacterial keratitis (7).

It is known that the alteration in corneal innervation produces neurotrophic keratitis. In our case report, the fibers of the subbasal nervous plexus were found tortuous and irregular. Moreover along the fibers many hyperreflective areas of various dimensions were present. These areas could be the demonstration of high metabolic activity of nerve for the particular tropism of the virus. After a week beadlike formations were visible along the nerves. The higher number of beads and the higher tortuosity are indices of an important metabolic activity, directed to repair the alterations at the epithelial level (8).

The association of atopy and herpetic infection is well known. Ocular herpes in atopic patients often is bilateral with more frequent recurrences. The reactivation has been reported to be more frequent and often more serious (2). Wilhelmus et al reported that bilateral herpetic disease presents a higher proportion of subsequent complications compared with unilateral herpetic eye disease (1). Because the high prevalence of atopy was noted in patients with bilateral herpetic keratitis, a specific and rapid laboratory diagnosis of HSV keratitis is es-

sential for the initiation of specific antiviral therapy. IVCN could be an *in vivo*, noninvasive, useful tool to detect very early recurrence.

In conclusion, IVCN provides a new tool for corneal examination that may be used in addition to slit-lamp biomicroscopy. Further studies will verify the clinical utility of IVCN in the early diagnosis of bilateral HSV keratitis and to reveal if it might play a role in determining the success of early treatments.

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## REFERENCES

1. Wilhelmus KR, Falcon MG, Jones BR. Bilateral herpetic keratitis. *Br J Ophthalmol* 1981; 65: 385-7.
2. Pepose JS. External ocular herpes virus infections in immunodeficiency. *Curr Eye Res* 1991; 10: 87-93.
3. Rosenberg ME, Tervo TM, Muller LJ, et al. *In vivo* confocal microscopy after herpes keratitis. *Cornea* 2002; 21: 265-9.
4. Maertzdorf J, Verjans GM, Remeijer L, van der Kooi A, Osterhaus AD. Restricted T cell receptor beta-chain variable region protein use by cornea-derived CD4+ and CD8+ herpes simplex virus specific T cells in patients with herpetic stromal keratitis. *J Infect Dis* 2003; 187: 550-8.
5. Thiel MA, Bossart W, Bernauer W. Improved impression cytology techniques for the Immunopathological diagnosis of superficial viral infections. *Br J Ophthalmol* 1995; 79: 473-5.
6. Mastropasqua L, Nubile M, Lanzini M, et al. Epithelial dendritic cell distribution in normal and inflamed human cornea: *in vivo* confocal microscopy study. *Am J Ophthalmol* 2006; 142: 736-44.
7. Hazlett LD, Moon MM, Dawisha S, et al. Age alters ADPase positive dendritic (Langerhans) cell response to *P. aeruginosa* ocular challenge. *Curr Eye Res* 1986; 5: 343-55.
8. Benitez del Castillo JM, Wasfy MA, Fernandez C, et al. An *in vivo* confocal masked study on corneal epithelium and subbasal nerves in patients with dry eye. *Invest Ophthalmol Vis Sci* 2004; 45: 3030-5.

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