

# **BRIEF COMMUNICATION** Rapid detection of fungi and *Acanthamoeba* from corneal ulcers using a novel mobile laboratory microscope and a smartphone

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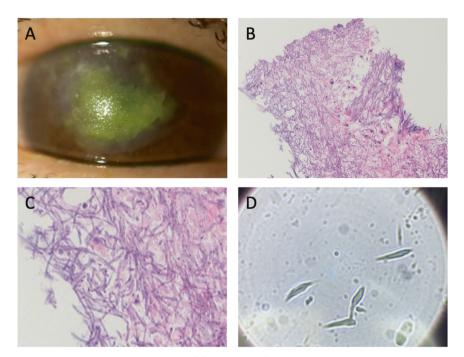
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Infectious keratitis is a devastating ocular disease that can occur in people in all ages [1–3]. Early identification of the causative microorganisms and starting adequate antimicrobial agents are crucial for good treatment results.

The mil-kin<sup>®</sup> (mil-kin, Tokyo, Japan; https://www.mil-kin.com), a mobile laboratory microscope using a smartphone, is a new device that enables us to observe bacteria, fungi, or other microorganisms without fixing and staining samples at a magnification of 1000 times. We succeeded in detecting the causative microorganisms in the corneal scrapings of two patients with infectious keratitis. The first case was a 61-year-old woman with corneal epithelitis on her right eye for over a year. She had been prescribed several antibiotic eye drops, acyclovir ointment, and low-dose steroid eye drops, but these were not completely effective. Several microbial cultures of the corneal scraping were performed, but failed to detect causative microorganisms.

In August 2021, she complained increased pain and blurred vision. A white deposit was observed on the lesion surface (Fig. 1A). A pathological examination of the removed deposit revealed clusters of filamentous fungi (Fig. 1B, C).



**Fig. 1 Clinical, pathological, and microscopic photographs of filamentous fungi taken with the mil-kin® (Case 1). A** The right cornea has a white deposit on the surface of the corneal lesion. **B** The pathology photograph of the removed white deposit contained many filamentous fungi. **C** High-magnification of the filamentous fungi. **D** Similar filamentous fungi are seen with mil-kin®.

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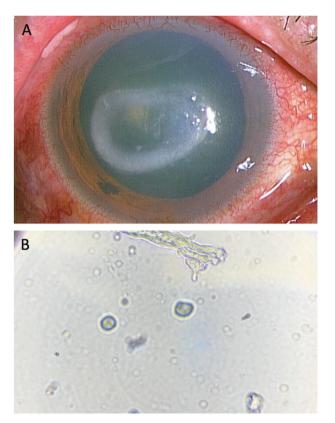


Fig. 2 Clinical photograph and Acanthamoeba cysts taken with a mil-kin<sup>®</sup> (Case 2). A The right eye has a corneal ulcer with a ring-shaped infiltration accompanied by ciliary injection. B Multiple double-walled round cysts are identified by mil-kin<sup>®</sup>.

We also examined the corneal scraping using mil-kin<sup>®</sup> (C-Type). A cover glass was placed on a small drop of distilled water (about 20–30  $\mu$ l) on a mechanical stage over an illuminator of the mil-kin<sup>®</sup>. The surface of the corneal lesion was swabbed with a wet cotton swab, which was directly stamped on a cover glass. Then, the specimen was examined using a smartphone camera at 1000× magnification (Supplementary Video). Filamentous fungi, similar to those observed in the pathological specimen, were recognized and photographed (Fig. 1D).

The second case was a 65-year-old man who had pain on his right eye for 2 months. His right cornea showed a corneal ulcer with a ring-shaped infiltration plus ciliary injection (Fig. 2A). We swabbed the ulcer with a wet cotton swab and used the specimens for microbial culture, *Acanthamoeba* DNA PCR, and mil-kin® examination. The mil-kin® observation detected many double-walled round objects, identical to *Acanthamoeba* cysts (Fig. 2B) [4]. Microbial culture and PCR also revealed *Acanthamoeba*.

Generally, corneal scraping cultures are used for reliably diagnosing the causative microorganisms of infectious keratitis. However, microbial culture requires special equipment and takes up to 1 week to obtain results. Using mil-kin<sup>®</sup>, it takes only a few

minutes from collecting samples to diagnosis. Fixing and staining the samples are not necessarily required. Photographs or short movies of the microbes can be taken simultaneously with a smartphone as a record. The simplicity and convenience of the procedure and detection of the pathological microorganisms would be useful not only for ophthalmologists but also for general practitioners.

We demonstrated the detection of fungi and *Acanthamoeba* cysts in this report. The further practices for detecting and identifying bacteria in patients with infectious keratitis are expected.

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# **AUTHOR CONTRIBUTIONS**

NK was responsible for correcting the data, screening potentially eligible studies, and writing the paper. TS helped correcting the data and interpreting results, and provided feedback on the report. ES, NM, and KN supervised the study and provided feedback on the report.

## **COMPETING INTERESTS**

The authors declare no competing interests.

# ADDITIONAL INFORMATION

Supplementary information The online version contains supplementary material available at https://doi.org/10.1038/s41433-022-02213-0.

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