

KOH Mount: A Prompt and Efficient Diagnostic Tool for Mycotic Keratitis

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ABSTRACT

Background: Mycotic keratitis (MK) is an ocular ailment occurring due to filamentous fungi or yeast infiltrating cornea. The leading threat for keratomycosis remains eye injury especially with and unrefined botanical substances. Contact lens wearing remains one of the leading causes in developed countries. Fusarium keratitis has emerged as a recent threat in the recent past. In 2020, the incidence of MK was estimated to be over 1 million cases per year. 80% of the cases were from Asia and East Asia. The MK cases are inversely interconnected to distance from the equator of planet Earth. There is increase in the possible incidence of MK worldwide. Diagnosing MK is challenging; correct diagnosis relies on reliable microscopy and culture, and by complementary apparatus such as in vivo confocal microscopy or PCR. Disappointingly these facilities are rare in availability in developing countries. The potassium hydroxide (KOH) mount is a commonly performed laboratory test to spot the presence or absence of fungal structures. Objective: This study was performed to determine the prevalence of fungal hyphae from corneal scrapings sent to the Microbiology laboratory of a tertiary care hospital, Lahore. Study Design: Retrospective observational study. Settings: Microbiology section, Pathology department, Fatimah Jinnah Medical University/ Sir Ganga Ram Hospital, Lahore Pakistan. Duration: from 2017 to 2022. Methods: Corneal scrapings were collected for fungal hyphae from Eye department and were sent to microbiology section for mycotic hyphen / hyphae on 10% KOH mount microscopy. Results: The data from an observational study showed that out of total sample of 241; 27 samples were of corneal scrapings of which 12 (44.4%) cases were found to be affirmative. Conclusion: 10% KOH mount is highly sensitive and specific, cheap and readily available method for early and quick findings and management of a mycotic keratitis.

Keywords: 10% Potassium hydroxide mount, Mycotic keratitis, Fungal hyphae.

INTRODUCTION

Opportunistic and systemic fungi are an unbridled hazard and are well calculated epidemic in equatorial hot and humid region of earth. The easily available topical steroids have added insult to the injury.¹

Visual impairment due to corneal opacity is to blame for about 1.5 to 2 million new cases on yearly basis.^{1,2} Infectious keratitis (IK) remains the foremost cause of corneal insult out of other etiological elements such as trauma, inflammation, idiopathetic and nutritional deficiency. A broad group of microorganisms, including bacteria, fungi, virus, and parasites remain the causes for IK. Eye trauma, contact lens use and post operative eye infections are well known predisposing factors for IK.2,3

Mycotic keratitis remains responsible for half of infective in South-Est-Asian countries due keratitis their environmental warmth and humidity.3,4 Agricultural active geography is adding on to the climate strains. Mycotic keratitis is clinically defying due to its diagnostic complexity, and latent complications.⁵ The frequent pathogenic fungi are of Aspergillus spp., Fusarium spp. And Candida spp. respectively followed by Fusarium spp., Alternaria, Curvularia, Helminthosporium, Bipolaris, Acremonium, Penicillium, Candida, Rhizopusspp., Paecilomyces, Rhodotorula and Mucor.5-7

The potassium hydroxide (KOH) mount is a regular, prompt, budget friendly and vital screening laboratory investigation used to confirm the fungal hyphae, buds and spores correspondingly on a daily basis of clinical practice.⁵⁻⁷

Potassium hydroxide (KOH) 10% wet mount is one of the historic and standard methods for the display of fungi in both corneal and non-corneal specimens.^{8,9} Microbial culture remains gold standard for pathogenic isolation and identification, but its availability for mycotic pathogens remains a cumbersome technique in routine and early fungal findings. There are researches that validate KOH smear being more efficient in early diagnosis than fungal cultures.¹⁰⁻¹² The KOH mount aide in swift treatment to avoid further ophthalmological complications.⁴

METHODS

An observational retrospective study of corneal scrapings sent for fungal hyphae from Ophthalmology department of Sir Ganga Ram Hospital, Lahore was conducted at Microbiology section, Pathology department, FJMU, Lahore. The data was collected from 2017 to 2022.

Infectious Keratitis (IK) is examined via slit lamp microscope. An ophthalmologist carried out corneal scrapings (CS) to be sent to our Microbiology working Laboratory for 10% KOH mount and microscopy.¹³⁻¹⁵

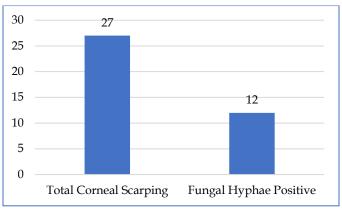
Potassium hydroxide KOH a well-known strong alkali. When specimen is mixed with 10% w/v KOH, it digests and clears the tissues which are surrounding the mycotic structures so that the fungal hyphae and conidia are visualized under a microscope. The specimen was sent to us in laboratory on a clean glass slide covered with a clean cover slip in an air tight slide box. We at our laboratory added 1 drop of 10% KOH from the lateral edge of the cover slip and waited for 15-20 minutes. After placing the prepared slide on the microscope stage, we started with a low-power (10x) and then the 40x setting (high-dry objective) for visualizing fungal structures such as hyphae or budding yeast.

Inclusion criteria were of corneal scrapings. Exclusion criterion was non corneal scrapings i.e., skin, hair and nail.

RESULTS

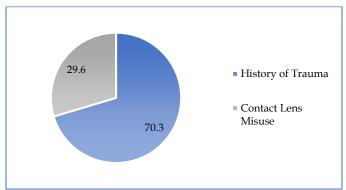
A total of 241 fungal scarping samples were received for microscopy via 10% KOH mount. Out of total sample of 241; 27 samples were of corneal scrapings of which 12 (44.4%) cases were found to be positive for fungal hyphae on 10% KOH mount microscopy. (Fig: 01)

Figure 1: 27 Corneal scarping samples received for microscopy via 10% KOH mount out of which 12 samples were fungal hyphae positive



The history of the received 27 corneal scrapings showed 70.3% cases were of eye trauma and 29.6% were of misuse of contact lenses. (Fig:02)

Figure 2: Percentage of history of trauma and contact lens misuse amongst positive cases



The positive cases further showed 18 male patients and 09 female patients. (Fig: 03)

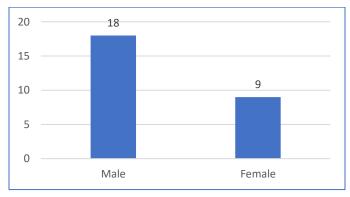


Figure 3: Showing male and female comparison among the positive cases

DISCUSSION

10% potassium hydroxide (KOH) wet mount, Gramstain, Giemsa, calcofluor white remain the usual techniques for concluding mycotic keratitis.9 10% KOH mount is a brisk and economical and one of the most regularly performed method for detection of fungi with a sensitivity of 61–94% and specificity of 91–97%. Timely diagnosis and management of mycotic keratitis is a challenge. Recent advancements in the detection methodology such as in vivo confocal microscopy and the PCR can be useful in quick and correct diagnosis.¹⁶

Mycotic keratitis is an opportunistic eye infection can lead to blindness.¹ It is difficult and challenging to examine and collect its specimen especially in paediatric population. Fungal keratitis faces a challenge in its early and accurate diagnosis by regular mycological techniques.¹⁷⁻¹⁹

A male predominance (n= 18, 66.6%) was recorded in our study, which is similar to the study conducted in South Korea and India.²⁰⁻²² The high propotion of male patients could most likely be due to their bigger chances of ocular trauma, which is one of the main predisposing reasons for MK. Mycotic keratitis time and again occurs with previous predisposition. Corneal trauma inclines the patient to that serious infection in many studies and accounts for 40%-60% of patients with MK.5,23-26 Other studies stated that contact lens wearing displayed the mostly prevalent predisposing factor.^{22,27,28} In our study eye trauma was 70% whereas contact lens misusage was 29.7%, which is in accordance with a study published from Egypt.¹⁶ They stated trauma (63.6%) a leading factor for MK followed by other predisposing factors. The direct microscopic examination of corneal scrapping material in 10% potassium hydroxide (KOH) is one of the easily available and simple techniques in detecting fungal elements.²³ Our study detected 44.4% of, fungal hyphae with 10% KOH mount microscopy.

Sharma and co-workers have stated a high specifity of 91% and 61% sensitivity. Moreover, Chowdhary A and Singh K investigating 3298 eyes stated that KOH sensitivity is 62% and specificity 97%.29 These studies and their authors have strongly recommended 10% KOH mount microscopy for quick, efficient and highly specific detection of fungal hyphae in keratomycosis.³⁰ Though Fungal culture is the gold standard in diagnosing MK besides the microscopic examination but the simple inexpensive and highly sensitive and specific test remains 10% KOH mount. False negative cultures due to the not enough specimens or specimen from an out of place site is recorded in a different study.³¹ In one of the studies where 70 corneal scrapings samples 49 (70%) samples were PCR-positive included 15 (43%) samples from culture-negative samples.1

CONCLUSION

This study concluded that it is vital to identify the precise etiology of corneal ulcer in order to save the eye from complication.10% KOH is highly sensitive and specific, cheap and readily available method for early and quick diagnosis and treatment. Other quick methods to diagnose fungal keratitis include (other than KOH Mount): Slit Lamp Microscopy, IVCM (In Vivo Confocal Microscopy) Fungal Flourescent Staining and Molecular Diagnosis. Their utilization as an everyday diagnostic device is hard due scarcity of resources and indigent health care budget at public sector hospitals.

LIMITATIONS

Unavailability of special fungal staining and fungal cultures due to our limited resources at public sector hospital.

SUGGESTIONS / RECOMMENDATIONS

In future we suggest to take our study ahead with fungal staining and culture with 10% KOH.

CONFLICT OF INTEREST / DISCLOSURE

The authors declare no conflict of interest.

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