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المؤتمر السنوي الدولي للمجعية المصرية  
INTERNATIONAL CONGRESS OF THE  
EGYPTIAN OPHTHALMOLOGICAL SOCIETY

# EOS 2023




## Evaluation of the Efficacy of **High Fluence PACK-CXL** in Fusarium Keratitis in Rabbits

**By:**  
**RAMY AWAD ELBASSIOUNY**  
MD, PHD, MRCS-Ed

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**No conflicts of interest to declare**

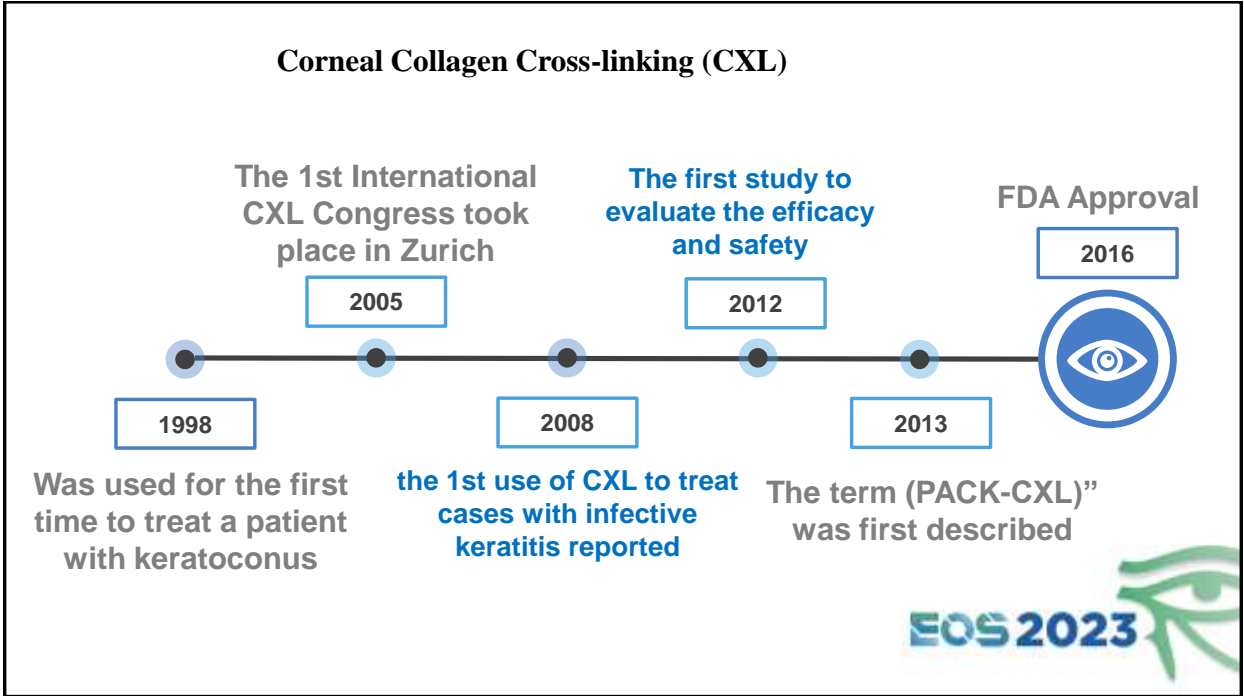


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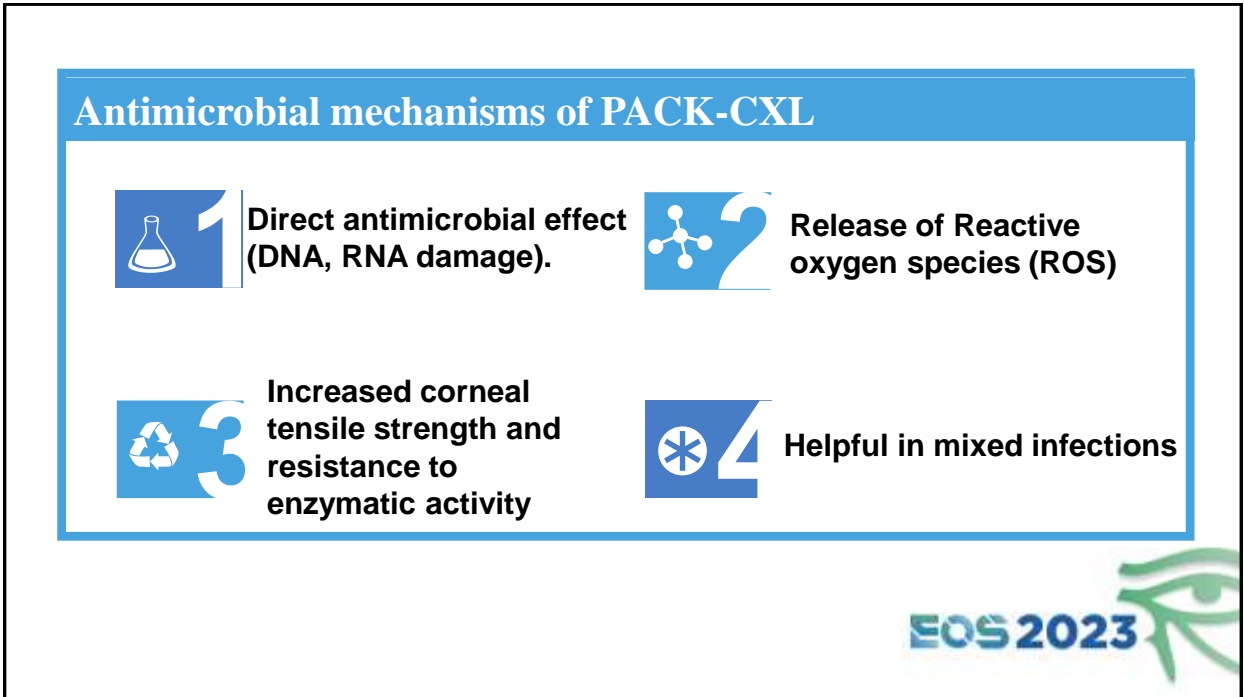
**Introduction**



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**PACK-CXL was performed same way as Dresden protocol with some modifications:**

UV beam should involve the lesion even in peripheral

Manual removal of the epithelium along with debridement

Riboflavin is used without any viscosity agent

Fluorescein should be avoided during the procedure

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## **Aim of the work**


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**Investigate the efficacy of high fluence CXL and  
to compare it with topical voriconazole  
treatment in fungal keratitis models  
experimentally induced in rabbit eyes**



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# **Materials**



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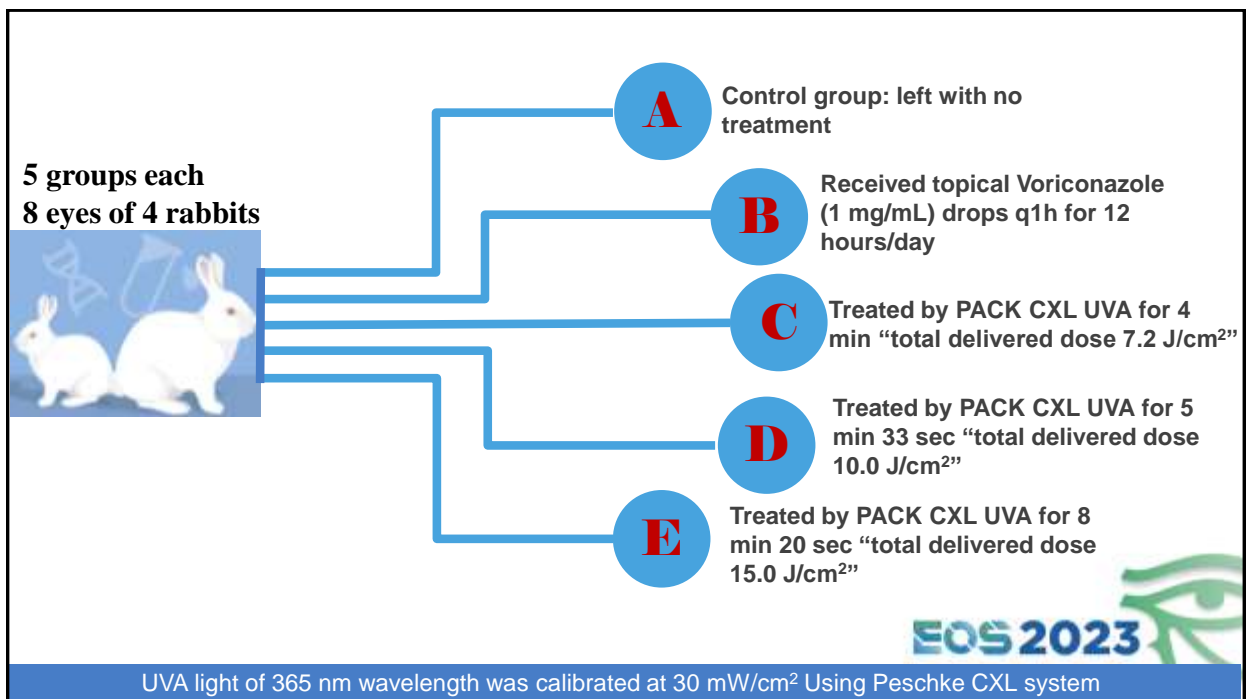
- 20 adult male New Zealand white rabbits.
- Weight:  $2.75 \pm 0.25$  Kg.
- Age at the beginning of the study: 17 weeks (Equivalent to an adult human 16 years old).
- Both eyes were included in the study



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# Methods



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- **Interventional Analytical study**



- Conducted in animal lab of Medical Research Institute, Alexandria University
- in collaboration with



Ophthalmology



Microbiology



Pathology



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### A) Isolation of Fungal sp:

- **Fusarium solani**
- **Isolated from patient with corneal ulcer**



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### B) Preparation of fusarium suspensions:

- **Fungal suspensions with a concentration of  $10^5$  CFU/ml were prepared and kept in sterile cryo vials.**



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### C) *Fusarium* keratitis induction in rabbits

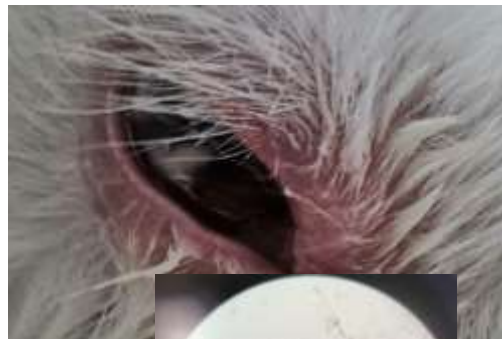


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### D) Confirmation of infection and initiation of treatment

On the 4th day:

- Fungal infection was confirmed.
- Group (A) received no treatment.
- Group (B) received topical Voriconazole (1 mg/mL) drops every hour.

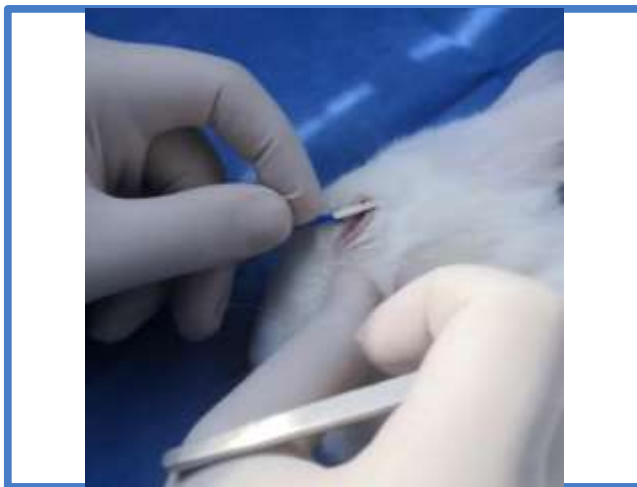


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**E) Surgical procedure (PACK CXL):**



**E) Surgical procedure (PACK CXL):**



## F) Clinical evaluation:

Modified scale of Schreiber et al. (2003)

	Grade (0)	Grade (1)	Grade (2)	Grade (3)
Conjunctival hyperemia	No hyperemia	Mild	Moderate	sever
Corneal clouding	No edema	Mild	2 quadrant	Total edema
Corneal vascularization	Clear	Up to 2 mm	> 2 mm	To the center
Degree of infiltrate	Measured in millimetres			
Hypopion degree	Measured in millimetres			

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## G) Microbiology evaluation:



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## H) Histopathology evaluation:



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## H) Histopathology evaluation:

	None	Low	Moderate	Intense
Inflammation (Inflammatory cells)	No cells	<25% of corneal thickness	25% - 50% of corneal thickness	>50% of corneal thickness
Edema	No edema	<25% of corneal thickness	25% - 50% of corneal thickness	>50% of corneal thickness
Hyphae density	No hyphae seen	hyphae seen but no mass formation	mass formation seen < 25% of corneal thickness	mass formation seen > 25% of corneal thickness

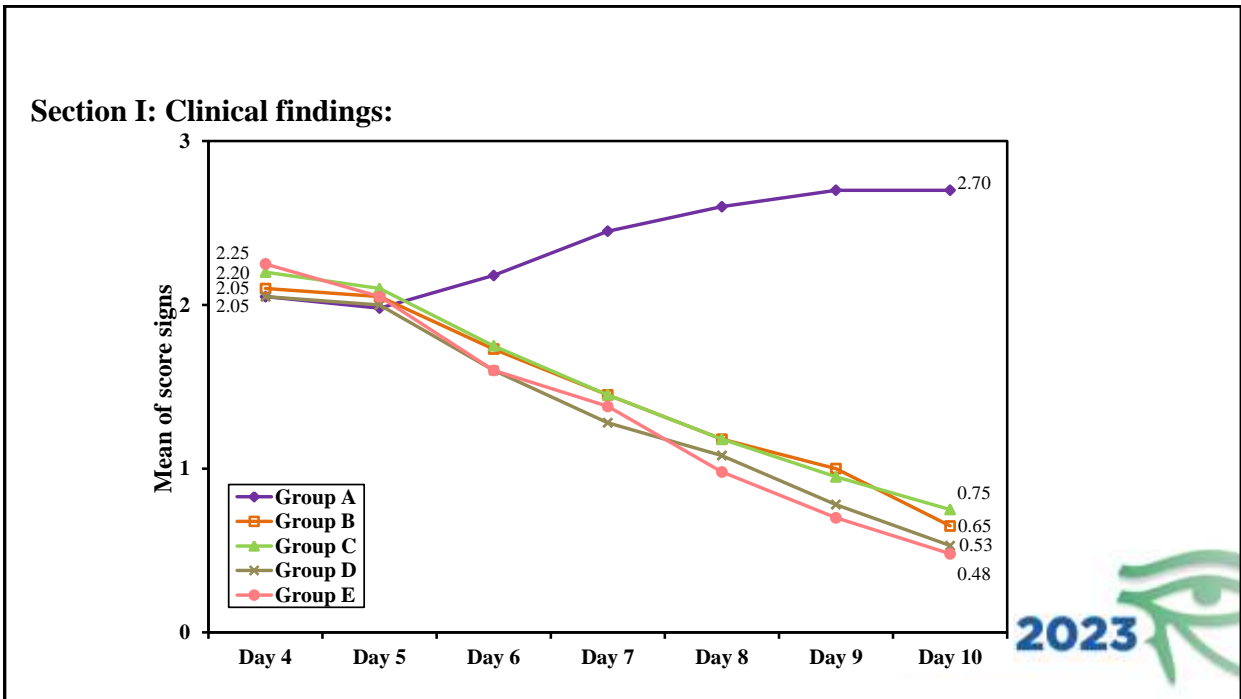


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# Results



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## Section II: Microbiological findings:

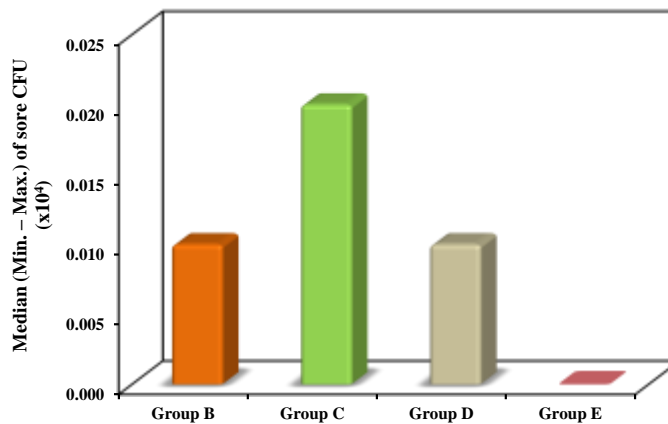
Comparison between the different studied groups according to number of (CFU/ml) of Fusarium

CFU	Group A (n = 7)	Group B (n = 7)	Group C (n = 7)	Group D (n = 7)	Group E (n = 7)	P
Min. – Max.	$1 \times 10^2 - 8 \times 10^7$	$0 - 2 \times 10^3$	$0 - 2 \times 10^7$	$0 - 2 \times 10^3$	$0 - 2 \times 10^3$	
Median (IQR)	$4 \times 10^4$ ( $2 \times 10^3 - 12 \times 10^4$ )	$1 \times 10^2$ ( $1 \times 10^2 - 6 \times 10^2$ )	$2 \times 10^2$ ( $1 \times 10^2 - 1.1 \times 10^3$ )	$1 \times 10^2$ ( $0 - 6 \times 10^2$ )	0 ( $0 - 1.1 \times 10^3$ )	0.033*
$p_0$		0.017*	0.051	0.009*	0.004*	
Sig. bet. grps.		$p_1=0.660, p_2=0.821, p_3=0.640, p_4=0.505, p_5=0.364, p_6=0.810$				

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## Section II: Microbiological findings:



Comparison between the studied groups according to mean number of Colony-forming unit (CFU/ml) of Fusarium

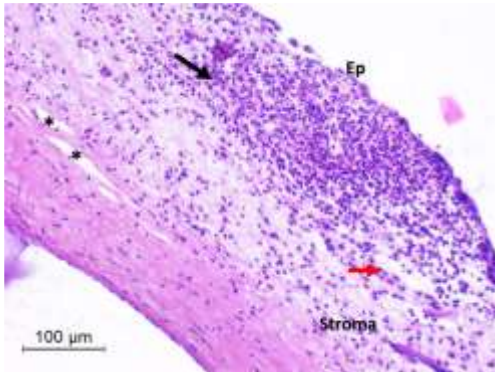
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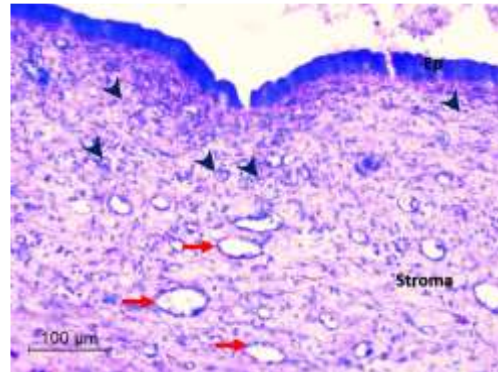


### Section III: Pathological evaluation:

H & E



PAS



**Histologic sections of corneal button of group A:**

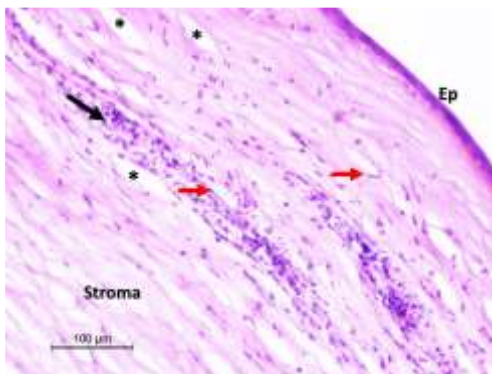
Group A (untreated) showing epithelial ulceration, dense subepithelial collection of mixed inflammatory cells (black arrow), stromal edema (\*), newly formed vascular spaces (red arrows) and numerous fungal hyphae (arrowhead) in PAS-stained sections.

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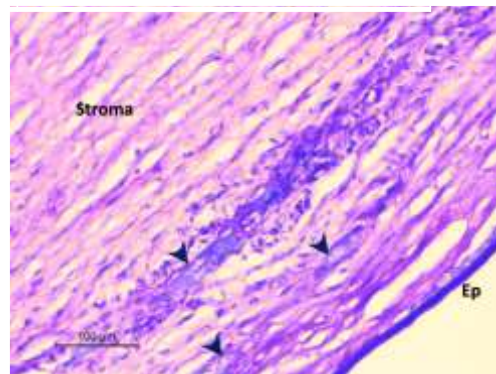
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### Section III: Pathological evaluation:

H & E



PAS



**Histologic sections of corneal button of group B:**

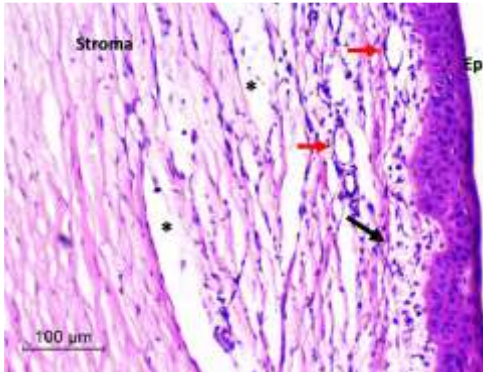
Group B (Voriconazole treated group) showing intact epithelial lining. Stroma shows moderate inflammatory infiltrate in the anterior two thirds (arrow). Edema (\*) and neovascularization (red arrows) are still seen. PAS stain shows fungal hyphae (arrowheads)

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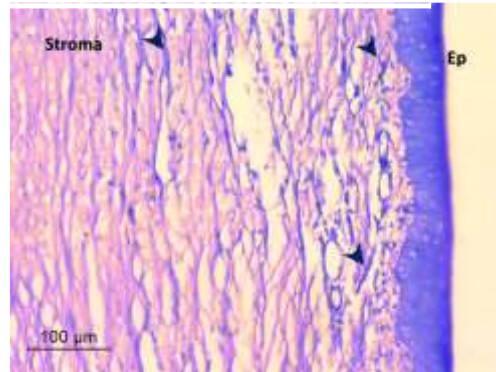
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### Section III: Pathological evaluation:

#### H & E



#### PAS



#### Histologic sections of corneal button of group C:

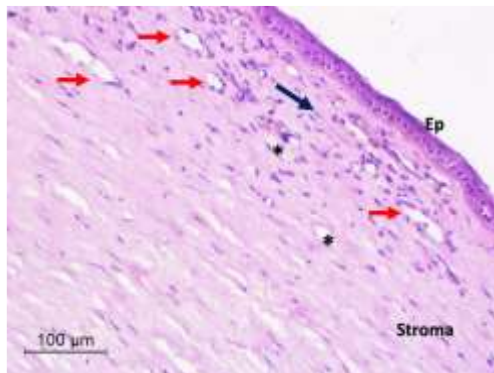
Group C showing hyperplasia of surface epithelium, moderate inflammatory infiltrate (black arrow). Corneal edema (\*) and small vascular spaces (red arrows) are still noted. PAS stain shows fungal hyphae (arrowhead)

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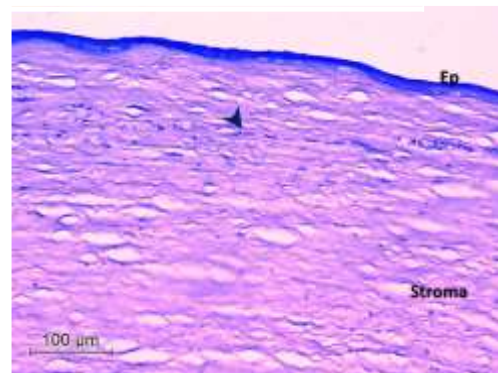
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### Section III: Pathological evaluation:

#### H & E



#### PAS



#### Histologic sections of corneal button of group D:

Group D sections reveal focal inflammatory infiltrate (black arrow) and few vascular spaces (red arrow) and mild edema (\*). Rare hyphae (arrowhead) are seen in PAS stain

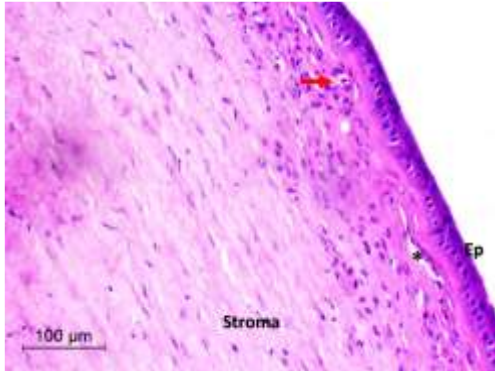
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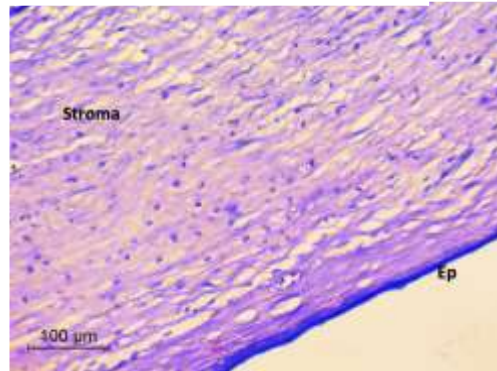


### Section III: Pathological evaluation:

#### H & E



#### PAS



#### Histologic sections of corneal button of group E:

Group E showing intact epithelial lining. The corneal stroma shows few dispersed inflammatory cells under the surface epithelium and focal edema (\*). Focal neovascularization (score 0) is seen. No fungal hyphae could be detected by PAS stain.



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## Conclusion



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## High fluence PACK-CXL

- Decreases inflammatory signs, accelerates healing and decreases the incidence of corneal perforation and intraocular inflammation.
- Increasing the total fluence of UVA is associated with better outcomes regarding clinical improvement and pathological features of inflammation.

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**Farhad Hafezi**

Apr 7 · 🌐



Awesome evidence that high-fluence PACK-CXL "does the job".

Previous experimental and clinical studies used too low of a fluence on infectious keratitis.: this would be as if you tried to operate on a rock hard cataract with very low phaco settings [00J]....

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