Comparative analysis of Bacterial Isolation Rates from Corneal Scrapes and Corneal Impression Membranes in cases of suspected Microbial Keratitis

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Background

- Microbial keratitis is a sight-threatening ophthalmic emergency.
- Delayed diagnosis and management can lead to significant visual morbidity.
- UK Annual Incidence:
  - 34.7 per 100 000 /year (Ting et al, 2021) in Nottingham
  - 6.96 per 100 000 /year (Moledina et al, 2023) in East of England
- Corneal Scrapes (CS) remains the diagnostic investigation of choice to isolate infectious microorganisms and identify sensitivities to anti-microbials.
- CS can be challenging to perform in anxious or uncooperative patients.
- An additional corneal sampling method is the use of Corneal Impression Membranes (CIM) (Kaye et al, 2016)

	CS (n=116)	CIM (n=116)	p-value
Positive Isolates	72.4%	63.8%	0.12
Commonest Isolates: *Pseudomonas aeruginosa*	20.7%	20.0%	0.87
Staphylococcus epidermidis	25.9%	24.1%	0.88
Staphylococcus aureus	7.8%	6.9%	1.00
Staphylococcus warneri	7.8%	3.4%	0.25
Mixed Organisms	28.4%	14.7%	0.02
Fungi	0%	0%	_
Acanthamoeba	0%	0%	_

Table 1: Commonest Isolates

CS=Corneal Scrape; CIM=Corneal Impression Membrane

Table 2. Comparison of the significance of the isolates cultured from Scrape and CIM samples

Specimen	Significant	Possible	Contaminant	No growth
Scrape	34.9% (44/126)	28.6% (36/126)	36.5% (46/126)	27.6% (32/116)
CIM	46.1% (42/91)	26.4% (24/91)	28.6% (26/91)	36.2% (42/116)
Significance	p=0.09	p=0.7	p=0.22	p=0.16

	Cohen's kappa	p-value
P. aeruginosa	0.89	<0.001
S. epidermidis	0.04	0.71
S. aureus	0.56	<0.001
S. warneri	0.11	0.19

Table 3: Cohen's kappa to compare the level of agreeability between corneal impression membranes and corneal scrapes in isolating micro-organisms.

Cohen's kappa Range and Strength of Agreeability:
<0.00=Poor
0.00-0.20=Slight
0.21-0.40=Fair
0.41-0.60=Moderate
0.61-0.80=Substantial
0.81-1.00=Almost Perfect

Methodology

- Aims:
  - Compare the isolation of microorganisms in paired samples of CS and CIM
  - Compares the level of agreeability in the isolation rates of bacteria using CIM and CS in patients with suspected microbial keratitis.
- Retrospective review of 116 consecutive cases which underwent both CS and CIM sampling at Moorfields Eye Hospital (City Road) during the period 26th May 2023-11th September 2023.
- Compare sensitivity and specificity of CIM compared to CS where the overall clinical interpretation of the result was used as the 'gold standard' for analysing the performance of the CIM and CS.
- Cohen's kappa to look at the level of agreeability in the isolation of specific organisms between CIM and CS.

Table 4. Performance characteristics for CS and CIM samples overall and restricted to significant pathogens only for bacterial infection.

Statistic	Overall analysis						Clinically significant isolates only					
	Clinical			Clinical			Clinical			Clinical		
Scrape	Positive	Negative	CIM	Positive	Negative		Scrape	Positive	Negative	CIM	Positive	Negative
Positive	80	46	Positive	66	26		Positive	44	82	Positive	42	41
Negative	29	37	Negative	41	59		Negative	9	57	Negative	11	98
	%	95% CI		%	95% CI	p-value		%	95% CI		%	95% CI	p-value
Sensitivity	73.4%	64.1, 81.4		61.7%	51.8, 70.9	0.07		82.4%	69.2, 91.6		79.3%	65.9, 89.2	0.68
Specificity	44.6%	33.7, 55.9		69.4%	58.5, 79	<0.01 *		41.0%	32.7, 49.7		70.5%	62.2, 77.9	<0.01 *
PPV	63.5%	58.2, 68.5		71.7%	64.1, 78.3	0.2		33.9%	29.8, 38.2		50.6%	43.4, 57.8	0.02 *
NPV	56.1%	46.3, 65.4		59.0%	52.2, 65.5	0.7		86.4%	77.2, 92.2		89.9%	83.9, 93.8	0.48
Accuracy	60.9%	53.7, 67.9		65.1%	57.9, 71.8	0.4		52.1%	44.8, 59.4		72.9%	66.1, 79.1	<0.01 *





Discussion

- CIM is a fast, efficient and safe diagnostic test in the diagnosis of microbial keratitis
- The larger surface area of the 12mm polytetrafluoroethylene disc on the cornea can potentially yield higher number of isolates compared to CS (Kaye et al, 2016).
- There is a strong level of agreeability in the identification of *P. aeruginosa* between CIM and CS, and a moderate level of agreeability in the identification of *S.aureus.*
- The strength of agreeability was slight and not statistically significant for *S. epidermidis* and *S. warneri,* although their clinical significance is often of uncertain significance.

References

1. Green M, Carnt N, Apel A, Stapleton F. Queensland Microbial Keratitis Database: 2005-2015. Br J Ophthalmol. 2019 Oct;103(10):1481-1486. doi: 10.1136/bjophthalmol-2018-312881. Epub 2019 Jan 5. PMID: 30617096.
2. Ting DSJ, Ho CS, Cairns J, Elsahn A, Al-Aqaba M, Boswell T, Said DG, Dua HS. 12-year analysis of incidence, microbiological profiles and in vitro antimicrobial susceptibility of infectious keratitis: the Nottingham Infectious Keratitis Study. Br J Ophthalmol. 2021 Mar;105(3):328-333.
3. Moledina, M., Roberts, H.W., Mukherjee, A. et al. Analysis of microbial keratitis incidence, isolates and in vitro antimicrobial susceptibility in the East of England: a 6-year study. Eye 37, 2716–2722 (2023).
4. Kaye S, Tuft S, Hau S, Bowman R, Sidaginamale R, Lau C, Tole D, Daniels J, Kinsler V, Sharp S, Dart J, Ficker L, Roberts F, Tullo A, Bunya VY, Hammersmith K, Rauz S, Cabrera-Aguas M, Lahra M, Shah A, Armstrong M, Ormonde S, Edwards K, Watson S, Lim L, Larkin DF, Williams O, Kaufman C, Ghosh A, Shah DO, Zhu H, Doan T, Lietman T. Topical tobramycin therapy for corneal ulcers: pharmacokinetics and clinical outcomes. Invest Ophthalmol Vis Sci. 2014 Jun 19;55(6):3811-9. doi: 10.1167/iovs.14-14112. PMID: 24854858; PMCID: PMC4066139.
5. Kaye SB, Tuft S, Hau S, Bowman R, Sidaginamale R, Lau C, Tole D, Daniels J, Kinsler V, Sharp S, Dart J, Ficker L, Roberts F, Tullo A, Bunya VY, Hammersmith K, Rauz S, Cabrera-Aguas M, Lahra M, Shah A, Armstrong M, Ormonde S, Edwards K, Watson S, Lim L, Larkin DF, Williams O, Kaufman C, Ghosh A, Shah DO, Zhu H, Doan T, Lietman T. Topical tobramycin therapy for corneal ulcers: pharmacokinetics and clinical outcomes. Invest Ophthalmol Vis Sci. 2014 Jun 19;55(6):3811-9. doi: 10.1167/iovs.14-14112. PMID: 24854858; PMCID: PMC4066139.
6. Kaye S, Tuft S, Hau S, Bowman R, Sidaginamale R, Lau C, Tole D, Daniels J, Kinsler V, Sharp S, Dart J, Ficker L, Roberts F, Tullo A, Bunya VY, Hammersmith K, Rauz S, Cabrera-Aguas M, Lahra M, Shah A, Armstrong M, Ormonde S, Edwards K, Watson S, Lim L, Larkin DF, Williams O, Kaufman C, Ghosh A, Shah DO, Zhu H, Doan T, Lietman T. Topical tobramycin therapy for corneal ulcers: pharmacokinetics and clinical outcomes. Invest Ophthalmol Vis Sci. 2014 Jun 19;55(6):3811-9. doi: 10.1167/iovs.14-14112. PMID: 24854858; PMCID: PMC4066139.
7. Kaye S, Tuft S, Hau S, Bowman R, Sidaginamale R, Lau C, Tole D, Daniels J, Kinsler V, Sharp S, Dart J, Ficker L, Roberts F, Tullo A, Bunya VY, Hammersmith K, Rauz S, Cabrera-Aguas M, Lahra M, Shah A, Armstrong M, Ormonde S, Edwards K, Watson S, Lim L, Larkin DF, Williams O, Kaufman C, Ghosh A, Shah DO, Zhu H, Doan T, Lietman T. Topical tobramycin therapy for corneal ulcers: pharmacokinetics and clinical outcomes. Invest Ophthalmol Vis Sci. 2014 Jun 19;55(6):3811-9. doi: 10.1167/iovs.14-14112. PMID: 24854858; PMCID: PMC4066139.