Bacterial Ocular Surveillance System (BOSS)
Sydney, Australia 2017-2018

Stephanie L. Watson, Barrie J Gatus, Maria Cabrera-Aguas, Benjamin H Armstrong, CR Robert George, Pauline Khoo, Monica M Lahra
Communicable Diseases Intelligence
ISSN: 2209-6051 Online

This journal is indexed by Index Medicus and Medline.

Creative Commons Licence - Attribution-NonCommercial-NoDerivatives CC BY-NC-ND

© 2020 Commonwealth of Australia as represented by the Department of Health

This publication is licensed under a Creative Commons Attribution-Non-Commercial NoDerivatives 4.0 International Licence from https://creativecommons.org/licenses/by-nc-nd/4.0/legalcode (Licence). You must read and understand the Licence before using any material from this publication.

Restrictions
The Licence does not cover, and there is no permission given for, use of any of the following material found in this publication (if any):

- the Commonwealth Coat of Arms (by way of information, the terms under which the Coat of Arms may be used can be found at www.itsanhonour.gov.au);
- any logos (including the Department of Health’s logo) and trademarks;
- any photographs and images;
- any signatures; and
- any material belonging to third parties.

Disclaimer
Opinions expressed in Communicable Diseases Intelligence are those of the authors and not necessarily those of the Australian Government Department of Health or the Communicable Diseases Network Australia. Data may be subject to revision.

Enquiries
Enquiries regarding any other use of this publication should be addressed to the Communication Branch, Department of Health, GPO Box 9848, Canberra ACT 2601, or via e-mail to: copyright@health.gov.au

Communicable Diseases Network Australia
Communicable Diseases Intelligence contributes to the work of the Communicable Diseases Network Australia.
http://www.health.gov.au/cdna

Communicable Diseases Intelligence (CDI) is a peer-reviewed scientific journal published by the Office of Health Protection, Department of Health. The journal aims to disseminate information on the epidemiology, surveillance, prevention and control of communicable diseases of relevance to Australia.

Editor
Tanja Farmer

Deputy Editor
Simon Petrie

Design and Production
Kasra Yousefi

Editorial Advisory Board
David Durrheim, Mark Ferson, John Kaldor, Martyn Kirk and Linda Selvey

Website
http://www.health.gov.au/cdi

Contacts
Communicable Diseases Intelligence is produced by: Health Protection Policy Branch Office of Health Protection Australian Government Department of Health GPO Box 9848, (MDP 6) CANBERRA ACT 2601

Email:
cdi.editor@health.gov.au

Submit an Article
You are invited to submit your next communicable disease related article to the Communicable Diseases Intelligence (CDI) for consideration. More information regarding CDI can be found at: http://health.gov.au/cdi.

Further enquiries should be directed to:
cdi.editor@health.gov.au.
Bacterial Ocular Surveillance System (BOSS)
Sydney, Australia 2017-2018

Stephanie L. Watson, Barrie J Gatus, Maria Cabrera-Aguas, Benjamin H Armstrong, CR Robert George, Pauline Khoo, Monica M Lahra

Abstract

This study investigated antimicrobial resistance (AMR) profiles from a cohort of patients with bacterial keratitis treated at Sydney Eye Hospital, 1 January 2017 – 31 December 2018. These AMR profiles were analysed in the context of the current Australian empiric regimens for topical therapy: ciprofloxacin/ofloxacin monotherapy versus combination therapy of cefalotin/cephazolin plus gentamicin. At our Centre, combinations of (i) chloramphenicol plus gentamicin and (ii) chloramphenicol plus ciprofloxacin are alternatively used, so were also analysed.

Three hundred and seventy-four isolates were cultured prospectively: 280/374 (75%) were gram positive, and 94/374 (25%) were gram negative. Coagulase-negative staphylococci comprised 173/374 (46%). Isolates included Staphylococcus aureus (n = 43/374) 11%; Streptococcus pneumoniae (n = 14/374) 3.7%; and Pseudomonas aeruginosa (n = 50/374) 13%.

Statistical comparison was performed. There was no significant difference between cover provided either of the current Australian recommendations: ciprofloxacin/ofloxacin vs cefalotin/cephazolin plus gentamicin (5.3% vs 4.8%, respectively; p = 0.655). However, the combination of chloramphenicol plus an anti-pseudomonal agent (ciprofloxacin/ofloxacin or gentamicin) had significantly improved cover.

Chloramphenicol plus gentamicin was superior to ciprofloxacin/ofloxacin (1.9% vs 5.3% resistance respectively; p = 0.007), and cefalotin/cephazolin plus gentamicin (1.9% vs 4.8%; p = 0.005). Chloramphenicol plus ciprofloxacin was superior to cefalotin/cephazolin monotherapy (1.3% vs 5.3%; p ≤ 0.001), and to cefalotin/cephazolin plus gentamicin (1.3% vs 4.8%; p = 0.003). Chloramphenicol plus gentamicin versus chloramphenicol plus ciprofloxacin/ofloxacin were equivalent (p = 0.48).

There was no demonstrated in vitro superiority of either the current empiric antibiotic regimens. For our setting, for bacterial keratitis, chloramphenicol in combination offered superior in vitro cover. Broadened surveillance for ocular AMR is urgently needed across jurisdictions.

Keywords: Antibiotic resistance, bacterial keratitis, corneal scrape, surveillance

Introduction

Antimicrobial resistance (AMR) is a global health threat recognised across patient populations as having a significant potential impact on treatment outcomes. In the USA, resistant infections cause about 23,000 deaths and more than two million illnesses annually with indirect societal costs of US$35 billion. Similarly, 25,000 deaths per year related to resistant infections occur in Europe. The Organisation for Economic Co-operation and Development (OECD) estimate an average of 290 deaths each year in Australia are caused by eight resistant bacteria. By 2050, approximately 10,430 people will die due to AMR and the health costs will reach $370 million in Australia. Surveillance programs of AMR are recommended by the World Health
Organization Global Action Plan to underpin disease prevention and control strategies. These strategies include evidence-based antibiotic prescribing guidelines, informed with local data, as highlighted in the medical literature. Bacterial keratitis (BK) is an ophthalmic emergency requiring immediate and effective antibiotic treatment as it can progress rapidly, causing visual impairment and, potentially, blindness. There are significant collateral costs, including a reduced quality of life for the individual, and an increased health-system burden. In the elderly, loss of an eye and blindness have been reported in 10% and 40% of patients, respectively, and in children, permanent visual loss due to amblyopia is a complication. Thus, there is a continued need to undertake AMR surveillance in order to determine the suitability of empiric antibiotic therapy for BK, given the ever-changing challenge of organisms becoming resistant to antibiotics.

In 2016, a keratitis antimicrobial resistance surveillance programme was established at The Sydney Eye Hospital. This report examined the types, frequency of isolation, and the antibiotic resistance profiles of bacteria isolated from corneal scapes of patients where bacterial keratitis was clinically apparent. This was the first comprehensive study of ocular AMR in Australia. The Sydney Eye Hospital, established in 1882, is the largest quaternary ophthalmic referral hospital in the southern hemisphere, providing surgical and medical care for patients with corneal, vitreo-retinal, glaucoma, oculo-plastic, uveitis, and oculo-oncology conditions.

Our report of 2016 and studies elsewhere demonstrate that gram-positive bacteria including coagulase-negative Staphylococcus spp., Staphylococcus aureus and Streptococcus pneumoniae comprise the majority of organisms isolated from the cornea of patients with bacterial keratitis. However, infection with Pseudomonas aeruginosa is a major concern, particularly in people wearing contact lenses; thus, empiric, topical antibiotic therapy must include an antibiotic effective both against commonly-isolated gram-positive bacteria and Pseudomonas aeruginosa.

The purpose of the present study was to expand and update the information given in our inaugural report of 2016; and to identify the types and prevalence of the different types of bacteria isolated from corneal scrapes of patients with bacterial keratitis at the Sydney Eye Hospital during the period 2017-2018. In addition, the antibiotic susceptibilities of organisms were examined and analysed statistically in order to assess the appropriateness of the guidelines used for the empiric antibiotic treatment of bacterial keratitis in Australia: Therapeutic Guidelines – Antibiotic, Version 16, 2019.

The current guidelines in Australia recommend empiric fluoroquinolone monotherapy (0.3% ciprofloxacin or 0.3% ofloxacin) or fortified combination therapy with 5% cephazolin plus 0.9% gentamicin. The combinations of chloramphenicol 0.5% plus gentamicin 0.9%, or chloramphenicol 0.5% plus ciprofloxacin 0.3%, are used on occasion at our Centre as empiric therapy for BK and were therefore included in the analysis. Resistance to chloramphenicol is of additional interest given the drug was made available in Australia over the counter in 2010.

Methods

We conducted a review of the microbiology results of the cohort of patients who presented with clinical bacterial keratitis to Sydney Eye Hospital during the period January 1, 2017 to December 31 2018. Corneal scrape specimens were taken in accord with local protocols from patients who had a clinical diagnosis of keratitis at presentation to the Sydney Eye Hospital. Ethics approval for this study was granted by the Sydney Local Health District Human Research Ethics Committee (approval number: 2020/ETH00457). The microbiological methods have been previously described in our inaugural report of 2016. Briefly, corneal scapes were inoculated onto agar
media or onto the same media from an enrichment medium. Identification of organisms was performed using matrix assisted laser desorption ionisation–time of flight mass spectrometry (MALDI-TOF MS, Bruker Daltonics® Germany). Antibiotic susceptibility testing was performed via the calibrated dichotomous sensitivity (CDS) agar disc diffusion method.26

A statistical comparative analysis of antibiotic resistance to the following regimens was performed: ciprofloxacin/ofloxacin monotherapy, combination therapy with cefalotin/cephazolin plus gentamicin; combination therapy with chloramphenicol plus gentamicin, and combination therapy with ciprofloxacin/ofloxacin and chloramphenicol. These comparisons were made using McNemar’s test, with data analysed using Jamovi version 1.2.19.

Results

Bacteria isolated

Three hundred and seventy-four bacterial isolates were cultured prospectively, from 297/471 inoculated plates (a 63% positive culture rate). Of these, 280/374 (75%) were gram positive, as shown in Table 1; and 94/374 (25%) were gram negative, as shown in Table 2. Coagulase-negative staphylococci were isolated most frequently in 173/374 cases (46%);

Staphylococcus aureus (n = 43/374) comprised 11%;

Streptococcus pneumoniae (n = 14/374) 3.7%; and

Pseudomonas aeruginosa (n = 50/374) 13% of the total, as shown in Table 3.

Antibiotic resistance to ciprofloxacin/ofloxacin

There were 20/374 isolates resistant to ciprofloxacin/ofloxacin, giving an overall rate of resistance, as shown in Table 4, of 5.3% with a 95% confidence interval (95% CI) of 3.1–7.6%. With regards to ciprofloxacin/ofloxacin resistance by organisms isolated: for coagulase-negative staphylococci, 10/173 (6%) were resistant; 7/43 (16%) Staphylococcus aureus were resistant; 2/12 (17%) Corynebacterium spp. were resistant, and 1/9 (11%) Serratia marcescens was resistant. No Pseudomonas aeruginosa (0/50) isolate was resistant.

Antibiotic resistance to cefalotin/cephazolin

There were 113/374 organisms resistant to cefalotin/cephazolin, giving an overall rate of resistance of 30% as shown in Table 5. For cefalotin/cephazolin, 33/173 coagulase-negative staphylococci (19%) were resistant; and for Staphylococcus aureus, 6/43 (14%) were resistant. No Streptococcus pneumoniae was resistant to cefalotin/cephazolin (0/14). Pseudomonas aeruginosa is intrinsically resistant to cefalotin/cephazolin.

Antibiotic resistance to gentamicin

There were 335 of 374 organisms tested for gentamicin susceptibility. Of these, 44/335 (13%) were resistant, as shown in Table 6. Moraxella spp. (n = 24), Corynebacterium spp. (n = 12) and Haemophilus influenzae (n = 3) were not tested, as calibrations for these organisms against gentamicin are not provided in the CDS method. Of the coagulase-negative staphylococci isolated, there were 14/173 (8.1%) that were resistant to gentamicin, as were 4/43 (9.3%) of Staphylococcus aureus. Streptococcus species are considered resistant to gentamicin monotherapy. No Pseudomonas aeruginosa isolated (0/50) was resistant to gentamicin.

Antibiotic resistance to chloramphenicol

All isolates were tested against chloramphenicol and the proportion resistant was 21% (79/374), as shown in Table 7. Of the coagulase-negative staphylococci isolates, 21/173 (12%) were resistant to chloramphenicol, as were 2/43 (5%) of the Staphylococcus aureus isolated. Of the Streptococcus pneumoniae isolated, one (1/14; 7%) was resistant to chloramphenicol. Pseudomonas aeruginosa has intrinsic resistance to chloramphenicol.
Table 1. The types, numbers (n) and percentages (%) of gram-positive organisms isolated from corneal scrapes during the period 2017–2018.

| Organism                        | n  | %  |
|---------------------------------|----|----|
| Coagulase-negative staphylococci| 173| 62 |
| *Staphylococcus aureus*         | 43 | 15 |
| *Streptococcus pneumoniae*      | 14 |  5 |
| Corynebacterium spp.            | 12 |  4 |
| *Micrococcus luteus*            | 10 |  4 |
| *Streptococcus mitis/oralis*     |  8 |  3 |
| Bacillus spp.                   |  6 |  2 |
| Rothia spp.                     |  3 |  1 |
| *Streptococcus gordonii*        |  2 |  1 |
| Other                          |  9 |  3 |
| **Total**                      | 280| 100|

a Including methicillin-resistant *Staphylococcus aureus*.

b Other: *Abiotrophia defectiva* (1), *Clostridium sporogenes* (1), *Lysinibacillus sphaericus* (1), *Propionibacterium* spp. (1), *Enterococcus faecalis* (1), *Brevibacillus borstelensis* (1), *Paenibacillus* spp. (1), *Streptococcus sanguinis* (1), *Streptococcus dysgalactiae* (1).

Table 2. The numbers (n) and percentages (%) of gram-negative organisms isolated from corneal scrapes during the period 2017–2018.

| Organism                   | n  | %  |
|----------------------------|----|----|
| *Pseudomonas aeruginosa*   | 50 | 53 |
| *Moraxella* spp.           | 24 | 26 |
| *Serratia marcescens*      |  9 | 10 |
| *Haemophilus influenzae*   |  3 |  3 |
| Other                      |  8 |  9 |
| **Total**                  | 94 | 100|

a Other: *Acinetobacter* spp. (1), *Achromobacter xylosidans* (1), *Enterobacter cloacae* complex (1), *Klebsiella oxytoca* (1), *Roseomonas mucosa* (2), *Citrobacter koseri* (1), *Proteus mirabilis* (1).

b Other gram-positive: *Abiotrophia defectiva* (1), *Clostridium sporogenes* (1), *Lysinibacillus sphaericus* (1), *Propionibacterium* spp. (1), *Enterococcus faecalis* (1), *Brevibacillus borstelensis* (1), *Paenibacillus* spp. (1), *Streptococcus sanguinis* (1), *Streptococcus dysgalactiae* (1).

c Other gram-negative: *Acinetobacter* spp. (1), *Achromobacter xylosidans* (1), *Enterobacter cloacae* complex (1), *Klebsiella oxytoca* (1), *Roseomonas mucosa* (2), *Citrobacter koseri* (1), *Proteus mirabilis* (1).

Table 3. Frequency (n) and percentage (%) of the total number of organisms isolated from corneal scrapes during the period 2017–2018.

| Organism                        | n  | %  |
|---------------------------------|----|----|
| Coagulase-negative staphylococci| 173| 46 |
| *Pseudomonas aeruginosa*        | 50 | 13 |
| *Staphylococcus aureus*         | 43 | 12 |
| *Moraxella* spp.                | 24 |  6 |
| *Streptococcus pneumoniae*      | 14 |  4 |
| Corynebacterium spp.            | 12 |  3 |
| *Micrococcus luteus*            | 10 |  3 |
| *Serratia marcescens*           |  9 |  2 |
| Other gram-positive              |  9 |  2 |
| *Streptococcus mitis/oralis*     |  8 |  2 |
| Other gram-negative              |  8 |  2 |
| Bacillus spp.                   |  6 |  2 |
| Rothia spp.                     |  3 |  1 |
| *Haemophilus influenzae*        |  3 |  1 |
| *Streptococcus gordonii*        |  2 |  1 |
| **Total**                      | 374| 100|

a Including methicillin-resistant *Staphylococcus aureus*.
Analysis of *in vitro* resistance to combination therapy

**Combined antibiotic resistance to cefalotin/cephazolin plus gentamicin**

There were 39 isolates (24 *Moraxella* spp., 12 *Corynebacterium* spp. and three *Haemophilus influenzae*) that were not able to be tested for gentamicin susceptibility. All *Moraxella* spp. and *Corynebacterium* spp., plus 2/3 isolates of *Haemophilus influenzae*, were susceptible to cefalotin/cephazolin, and therefore *in vitro* susceptibility to the combined regimen of cefalotin/cephazolin and gentamicin was determined according to tested susceptibility for cefalotin/cephazolin. A single isolate of *Haemophilus influenzae* was resistant to cefalotin/cephazolin, and therefore susceptibility to the combined regimen was unable to be confirmed; this isolate was included in the analysis as likely resistant to combined cefalotin/cephazolin plus gentamicin. For the staphylococci, 12/173 (7%) coagulase-negative staphylococci and 3/43 (7%) *Staphylococcus aureus* were resistant to both agents. No *Streptococcus pneumoniae* isolates (0/14) and no (0/50) *Pseudomonas aeruginosa* were resistant to both agents.

Overall, resistance to the combination of cefalotin/cephazolin plus gentamicin, where both agents were tested, or resistance to cefalotin/cephazolin when gentamicin was not tested, was 4.8% (18/374; 95% CI: 2.6–7.0%). These data are shown in Table 8.

**Combined antibiotic resistance to chloramphenicol plus gentamicin**

There were 7/374 isolates resistant to the combination of chloramphenicol plus gentamicin giving an overall rate of resistance of 1.9% (95% CI: 0.5–3.2%). Of the coagulase-negative staphylococci, 5/173 (2.9%) were resistant. All isolates of *Staphylococcus aureus* were susceptible (n = 43). A single isolate each of *Corynebacterium* spp. (1/12; 8.3%) and *Serratia marcescens* (1/9; 11%), were resistant. All isolates of *Pseudomonas aeruginosa* were susceptible (n = 50).

**Statistical analysis of differences in coverage of the antibiotic susceptibility between antibiotic combinations**

**Ciprofloxacin/ofloxacin versus cefalotin/cephazolin plus gentamicin**

As displayed in Tables 4 and 8, 20/374 isolates (5.3%) were resistant to ciprofloxacin/ofloxacin, and 18/374 (4.8%) to combined cefalotin/cephazolin plus gentamicin. A single isolate of *Haemophilus influenzae* was resistant to cefalotin/cephazolin, and unable to be tested against gentamicin given the lack of available calibrations. As *in vitro* susceptibility was unable to be confirmed, this isolate was included in the analysis as resistant to the combination of cefalotin/cephazolin and gentamicin. Across all 374 isolates tested, nine isolates were concurrently resistant to both regimens (six isolates of coagulase-negative staphylococci and three isolates of *Staphylococcus aureus*). When the difference in coverage of the two regimens was examined, there was no significant difference detected ($\chi^2 = 0.20$; degrees of freedom (df) = 1; $p = 0.655$).
Table 4. The total number of organisms isolated from corneal scrapes during the period 2017–2018 and the number (n) and percentage (%) resistant to ciprofloxacin/ofloxacin.

| Organism                        | Total | Resistant | n | %  |
|---------------------------------|-------|-----------|---|----|
| Coagulase-negative staphylococci| 173   | 10        | 6 |    |
| *Pseudomonas aeruginosa*        | 50    | 0         | 0 |    |
| *Staphylococcus aureus*         | 43    | 7         | 16|    |
| *Moraxella spp.*                | 24    | 0         | 0 |    |
| *Streptococcus pneumoniae*      | 14    | 0         | 0 |    |
| *Corynebacterium spp.*          | 12    | 2         | 17|    |
| Micrococcus luteus              | 10    | 0         | 0 |    |
| *Serratia marcescens*           | 9     | 1         | 11|    |
| Other gram-positive (a)         | 9     | 0         | 0 |    |
| *Streptococcus mitis/oralis*    | 8     | 0         | 0 |    |
| Other gram-negative (b)         | 8     | 0         | 0 |    |
| *Bacillus spp.*                 | 6     | 0         | 0 |    |
| Rothia spp.                     | 3     | 0         | 0 |    |
| *Haemophilus influenzae*        | 3     | 0         | 0 |    |
| *Streptococcus gordonii*        | 2     | 0         | 0 |    |
| **Total**                       | 374   | 20        | 5.3|   |

(a) Including methicillin-resistant *Staphylococcus aureus*.
(b) Other gram-positive: *Abiotrophia defectiva* (1), *Clostridium sporogenes* (1), *Lysinibacillus sphaericus* (1), *Propionibacterium* spp. (1), *Enterococcus faecalis* (1), *Brevibacillus borstelensis* (1), *Paenibacillus* spp. (1), *Streptococcus sanguinis* (1), *Streptococcus dysgalactiae* (1).
(c) Other gram-negative: *Acinetobacter* spp. (1), *Achromobacter xylosoxidans* (1), *Enterobacter cloacae* complex (1), *Klebsiella oxytoca* (1), *Roseomonas mucosa* (2), *Citrobacter koseri* (1), *Proteus mirabilis* (1).

Table 5. The total number of organisms isolated from corneal scrapes during the period 2017–2018 and the number (n) and the percentage (%) resistant to cefalotin/cephazolin.

| Organism                        | Total | Resistant | n | (%) |
|---------------------------------|-------|-----------|---|-----|
| Coagulase-negative staphylococci| 173   | 33        | 19|     |
| *Pseudomonas aeruginosa* (IR)    | 50    | 50        | 100|    |
| *Staphylococcus aureus*         | 43    | 6         | 14|     |
| *Moraxella spp.*                | 24    | 0         | 0 |     |
| *Streptococcus pneumoniae*      | 14    | 0         | 0 |     |
| *Corynebacterium spp.*          | 12    | 0         | 0 |     |
| Micrococcus luteus              | 10    | 0         | 0 |     |
| *Serratia marcescens* (IR)      | 9     | 9         | 100|    |
| Other gram-positive (a)         | 9     | 0         | 0 |     |
| *Streptococcus mitis/oralis*    | 8     | 2         | 25|     |
| Other gram-negative (b,e)       | 8     | 8         | 100|    |
| *Bacillus spp.*                 | 6     | 4         | 66|     |
| Rothia spp.                     | 3     | 0         | 0 |     |
| *Haemophilus influenzae*        | 3     | 1         | 33|     |
| **Total**                       | 374   | 113       | 30|     |

(a) IR: Intrinsic resistance, denotes organisms intrinsically resistant to cefalotin/cephazolin.
(b) Including methicillin-resistant *Staphylococcus aureus*.
(c) Other gram-positive: *Abiotrophia defectiva* (1), *Clostridium sporogenes* (1), *Lysinibacillus sphaericus* (1), *Propionibacterium* spp. (1), *Enterococcus faecalis* (1), *Brevibacillus borstelensis* (1), *Paenibacillus* spp. (1), *Streptococcus sanguinis* (1), *Streptococcus dysgalactiae* (1).
(d) Other gram-negative: *Klebsiella oxytoca* (1), *Citrobacter koseri* (1), *Proteus mirabilis* (1) were tested and displayed acquired resistance to cefalotin/cephazolin.
(e) *Acinetobacter* spp. (1), *Achromobacter xylosoxidans* (1), *Enterobacter cloacae* complex (1), *Roseomonas mucosa* (2) are considered intrinsically resistant.
Table 6. The total number of organisms isolated from corneal scrapes and tested against gentamicin during the period 2017–2018 and the number (n) and percentage (%) resistant.

| Organism                        | Total | Resistant (n) | (%) |
|---------------------------------|-------|---------------|-----|
| Coagulase-negative staphylococci| 173   | 14            | 8   |
| Pseudomonas aeruginosa          | 50    | 0             | 0   |
| Staphylococcus aureus\(^a\)     | 43    | 4             | 9   |
| Streptococcus pneumoniae        | 14    | 14            | 100 |
| Micrococcus luteus              | 10    | 0             | 0   |
| Serratia marcescens             | 9     | 0             | 0   |
| Other gram-positive\(^b\)       | 9     | 1             | 12  |
| Streptococcus mitis/oralis group| 8     | 8             | 100 |
| Other gram-negative\(^c\)       | 8     | 1             | 12  |
| Bacillus spp.                   | 6     | 0             | 0   |
| Rothia spp.                     | 3     | 0             | 0   |
| Streptococcus gordonii          | 2     | 2             | 100 |
| **Total**                       | **335**| **44**       | **13** |

\(^a\) Including methicillin-resistant Staphylococcus aureus.

\(^b\) Other gram-positive: Abiotrophia defectiva (1), Clostridium sporogenes (1), Lysinibacillus sphaericus (1), Propionibacterium spp. (1), Enterococcus faecalis (1), Brevibacillus borstelensis (1), Paenibacillus spp. (1), Streptococcus sanguinis (1), Streptococcus dysgalactiae (1).

\(^c\) Other gram-negative: Acinetobacter spp. (1), Achromobacter xylosoxidans (1), Enterobacter cloacae complex (1), Klebsiella oxytoca (1), Roseomonas mucosa (2), Citrobacter koseri (1), Proteus mirabilis (1).

Table 7. The total number of organisms isolated from corneal scrapes during the period 2017–2018 and the number (n) and percentage (%) resistant to chloramphenicol.

| Organism                        | Total | Resistant (n) | (%) |
|---------------------------------|-------|---------------|-----|
| Coagulase-negative staphylococci| 173   | 21            | 12  |
| Pseudomonas aeruginosa (IR)\(^a\)| 50    | 50            | 100 |
| Staphylococcus aureus\(^b\)     | 43    | 2             | 5   |
| Moraxella spp.                  | 24    | 0             | 0   |
| Streptococcus pneumoniae        | 14    | 1             | 7   |
| Corynebacterium spp.            | 12    | 2             | 2   |
| Micrococcus luteus              | 10    | 0             | 0   |
| Serratia marcescens             | 9     | 1             | 11  |
| Other gram-positive\(^c\)       | 9     | 1             | 11  |
| Streptococcus mitis/oralis group| 8     | 0             | 0   |
| Other gram-negative\(^d\)       | 8     | 1             | 13  |
| Bacillus spp.                   | 6     | 0             | 0   |
| Rothia spp.                     | 3     | 0             | 0   |
| Haemophilus influenzae          | 3     | 0             | 0   |
| Streptococcus gordonii          | 2     | 0             | 0   |
| **Total**                       | **374**| **79**       | **21** |

\(^a\) IR: Intrinsic resistance.

\(^b\) Including methicillin-resistant Staphylococcus aureus.

\(^c\) Other gram-positive: Abiotrophia defectiva (1), Clostridium sporogenes (1), Lysinibacillus sphaericus (1), Propionibacterium spp. (1), Enterococcus faecalis (1), Brevibacillus borstelensis (1), Paenibacillus spp. (1), Streptococcus sanguinis (1), Streptococcus dysgalactiae (1).

\(^d\) Other gram-negative: Acinetobacter spp. (1), Achromobacter xylosoxidans (1), Enterobacter cloacae complex (1), Klebsiella oxytoca (1), Roseomonas mucosa (2), Citrobacter koseri (1), Proteus mirabilis (1).
Chloramphenicol plus gentamicin versus cefalotin/cephazolin plus gentamicin

Of the isolates tested, 18/374 (4.8%) were resistant to combined cefalotin/cephazolin plus gentamicin, and 7/374 isolates (1.9%) were resistant to chloramphenicol plus gentamicin, as shown in Tables 8 and 9. A single isolate of *Haemophilus influenzae* was resistant to cefalotin/cephazolin, and unable to be tested against gentamicin. As *in vitro* susceptibility was unable to be confirmed, this isolate was included in the analysis as resistant to the combination of cefalotin/cephazolin and gentamicin. Five isolates (all coagulase-negative *Staphylococcus* spp.) were resistant to all three agents. Statistically, the combination of chloramphenicol plus gentamicin had significantly better coverage across all isolates than the combination of cefalotin/cephazolin plus gentamicin ($\chi^2 = 8.07; df = 1; p = 0.005$).

Ciprofloxacin/ofloxacin versus chloramphenicol plus gentamicin

As shown in Tables 4 and 9, 20/374 isolates (5.3%) were resistant to ciprofloxacin/ofloxacin, and 7/374 (1.9%) to combined chloramphenicol plus gentamicin. Of these, only 2/374 isolates were concurrently resistant to both regimens (both coagulase-negative *Staphylococcus* spp.). Statistically, the combination of chloramphenicol plus gentamicin had significantly better coverage across all isolates than ciprofloxacin/ofloxacin ($\chi^2 = 7.35; df = 1; p = 0.007$).

Ciprofloxacin/ofloxacin monotherapy versus ciprofloxacin/ofloxacin plus chloramphenicol

As displayed in Tables 4 and 10, the addition of chloramphenicol to ciprofloxacin/ofloxacin reduced *in vitro* predicted treatment failure from 20/374 (5.3%) to 5/374 isolates (1.3%). *In vitro* resistance to the combination of chloramphenicol and ciprofloxacin/ofloxacin was detected in three isolates of coagulase-negative *Staphylococcus* spp., one *Serratia marcescens* and one *Corynebacterium* spp. The combination of ciprofloxacin/ofloxacin and chloramphenicol offered statistically significantly improved cover over ciprofloxacin/ofloxacin monotherapy ($\chi^2 = 16.00; df = 1; p \leq 0.001$).

Cefalotin/cephazolin plus gentamicin versus ciprofloxacin/ofloxacin plus chloramphenicol

As displayed in Tables 8 and 10, 18/374 (4.8%) isolates were resistant to combined cefalotin/cephazolin plus gentamicin, and 5/374 (1.3%) to combined chloramphenicol plus ciprofloxacin/ofloxacin. Of these, only 2/374 isolates were concurrently resistant to both regimens (both coagulase-negative *Staphylococcus* spp.). The combination of ciprofloxacin/ofloxacin and chloramphenicol offered statistically significantly improved cover over cephazolin/cefalotin plus gentamicin ($\chi^2 = 8.89; df = 1; p = 0.003$).

Chloramphenicol plus gentamicin versus ciprofloxacin/ofloxacin plus chloramphenicol

As displayed in Tables 9 and 10, 7/374 (1.9%) isolates were resistant to chloramphenicol plus gentamicin, and 5/374 (1.3%) to combined chloramphenicol plus ciprofloxacin/ofloxacin. Of these, only 2/374 isolates were concurrently resistant to both regimens (both coagulase-negative *Staphylococcus* spp.). When the difference in coverage of these two regimens was examined, there was no significant difference detected ($\chi^2 = 0.50; df = 1; p = 0.480$).
Table 8. Combined resistance to cefalotin/cephazolin plus gentamicin: the total number of organisms isolated from corneal scrapes during the period 2017–2018 and the number (n) and percentage (%) resistant to the combination of cefalotin/cephazolin plus gentamicin.

| Organism                             | Total | Resistant (n) | (%) |
|--------------------------------------|-------|---------------|-----|
| Coagulase-negative staphylococci     | 173   | 12            | 7   |
| Pseudomonas aeruginosa               | 50    | 0             | 0   |
| Staphylococcus aureus<sup>a</sup>    | 43    | 3             | 7   |
| Moraxella spp.<sup>b</sup>           | 24    | 0             | 0   |
| Streptococcus pneumoniae             | 14    | 0             | 0   |
| Corynebacterium spp.<sup>b</sup>    | 12    | 0             | 0   |
| Micrococcus luteus                   | 10    | 0             | 0   |
| Serratia marcescens                  | 9     | 0             | 0   |
| Other gram-positive<sup>c</sup>      | 9     | 0             | 0   |
| Streptococcus mitis/oralis group     | 8     | 2             | 25  |
| Other gram-negative<sup>d</sup>      | 8     | 0             | 0   |
| Bacillus spp.                        | 6     | 0             | 0   |
| Rothia spp.                          | 3     | 0             | 0   |
| Haemophilus influenzae<sup>e</sup>   | 3     | 1             | 33  |
| Streptococcus gordonii               | 2     | 0             | 0   |
| **Total**                            | **374**| **18**        | **4.8** |

<sup>a</sup> Including methicillin-resistant Staphylococcus aureus.

<sup>b</sup> Moraxella spp., Corynebacterium spp. and Haemophilus influenzae are not calibrated for gentamicin susceptibility testing by the CDS method. Isolates were therefore analysed as either sensitive or resistant on the basis of their susceptibility to cefalotin/cephazolin.

<sup>c</sup> Other gram-positive: Abiotrophia defectiva (1), Clostridium sporogenes (1), Lysinibacillus sphaericus (1), Propionibacterium spp. (1), Enterococcus faecalis (1), Brevibacillus borstelensis (1), Paenibacillus spp. (1), Streptococcus sanguinis (1), Streptococcus dysgalactiae (1).

<sup>d</sup> Other gram-negative: Acinetobacter spp. (1), Achromobacter xylosoxidans (1), Enterobacter cloacae complex (1), Klebsiella oxytoca (1), Roseomonas mucosa (2), Citrobacter koseri (1), Proteus mirabilis (1).

Table 9. The total number of organisms isolated from corneal scrapes during the period 2017–2018 and the number (n) and percentage (%) resistant to the combination of chloramphenicol plus gentamicin.

| Organism                             | Total | Resistant (n) | (%) |
|--------------------------------------|-------|---------------|-----|
| Coagulase-negative staphylococci     | 173   | 5             | 3   |
| Pseudomonas aeruginosa               | 50    | 0             | 0   |
| Staphylococcus aureus<sup>a</sup>    | 43    | 0             | 0   |
| Moraxella spp.<sup>b</sup>           | 24    | 0             | 0   |
| Streptococcus pneumoniae             | 14    | 1             | 7   |
| Corynebacterium spp.<sup>b</sup>    | 12    | 0             | 0   |
| Micrococcus luteus                   | 10    | 0             | 0   |
| Serratia marcescens                  | 9     | 0             | 0   |
| Other gram-positive<sup>c</sup>      | 9     | 1             | 11  |
| Streptococcus mitis/oralis group     | 8     | 0             | 0   |
| Other gram-negative<sup>d</sup>      | 8     | 0             | 0   |
| Bacillus spp.                        | 6     | 0             | 0   |
| Rothia spp.                          | 3     | 0             | 0   |
| Haemophilus influenzae<sup>e</sup>   | 3     | 1             | 33  |
| Streptococcus gordonii               | 2     | 0             | 0   |
| **Total**                            | **374**| **7**         | **2** |

<sup>a</sup> Including methicillin-resistant Staphylococcus aureus.

<sup>b</sup> Moraxella spp., Corynebacterium spp. and Haemophilus influenzae are not calibrated for gentamicin susceptibility testing by the CDS method. Isolates were therefore analysed as either sensitive or resistant on the basis of their susceptibility to chloramphenicol.

<sup>c</sup> Other gram-positive: Abiotrophia defectiva (1), Clostridium sporogenes (1), Lysinibacillus sphaericus (1), Propionibacterium spp. (1), Enterococcus faecalis (1), Brevibacillus borstelensis (1), Paenibacillus spp. (1), Streptococcus sanguinis (1), Streptococcus dysgalactiae (1).

<sup>d</sup> Other gram-negative: Acinetobacter spp. (1), Achromobacter xylosoxidans (1), Enterobacter cloacae complex (1), Klebsiella oxytoca (1), Roseomonas mucosa (2), Citrobacter koseri (1), Proteus mirabilis (1).
Table 10. Combined resistance to chloramphenicol plus ciprofloxacin/ofloxacin: the total number of organisms isolated from corneal scrapes during the period 2017–2018 and the number (n) and percentage (%) resistant to the combination of chloramphenicol plus ciprofloxacin

| Organism                             | Total | Resistant (n) | (%) |
|--------------------------------------|-------|---------------|-----|
| Coagulase-negative staphylococci     | 173   | 3             | 2   |
| Pseudomonas aeruginosa               | 50    | 0             | 0   |
| Staphylococcus aureus<sup>a</sup>    | 43    | 0             | 0   |
| Moraxella spp.                       | 24    | 0             | 0   |
| Streptococcus pneumoniae             | 14    | 0             | 0   |
| Corynebacterium spp.                 | 12    | 1             | 8   |
| Micrococcus luteus                   | 10    | 0             | 0   |
| Serratia marcescens                  | 9     | 1             | 11  |
| Other gram-positive<sup>b</sup>      | 9     | 0             | 0   |
| Streptococcus mitis/oralis group     | 8     | 0             | 0   |
| Other gram-negative<sup>c</sup>      | 8     | 0             | 0   |
| Bacillus spp.                        | 6     | 0             | 0   |
| Rothia spp.                          | 3     | 0             | 0   |
| Haemophilus influenzae               | 3     | 0             | 0   |
| Streptococcus gordonii               | 2     | 0             | 0   |
| **Total**                            | **374** | **5**        | **1** |

<sup>a</sup> Including methicillin-resistant Staphylococcus aureus.

<sup>b</sup> Other gram-positive: Abiotrophia defectiva (1), Clostridium sporogenes (1), Lysinibacillus sphaericus (1), Propionibacterium spp. (1), Enterococcus faecalis (1), Brevibacillus borstelensis (1), Paenibacillus spp. (1), Streptococcus sanguinis (1), Streptococcus dysgalactiae (1).

<sup>c</sup> Other gram-negative: Acinetobacter spp. (1), Achromobacter xylosidans (1), Enterobacter cloacae complex (1), Klebsiella oxytoca (1), Roseomonas mucosa (2), Citrobacter koseri (1), Proteus mirabilis (1).

Discussion

The use of combination therapy in the empirical management of BK is to offer the broadest spectrum of antibacterial cover, including both gram-positive and -negative organisms. To optimise clinical outcomes, empiric therapeutic regimens for infections should be informed, where possible, with local and quality assured AMR data. For the empiric treatment of clinical bacterial keratitis, there is a lack of recent and longitudinal AMR data to inform guidelines. In 2016, an international survey of corneal specialists sought to determine the clinical practice patterns for the empiric treatment of bacterial keratitis. In 2016 an international survey of corneal specialists identified regional variations in practice influenced by drug availability, toxicity, broad spectrum coverage and resistance.

In our study, 16% of *Staphylococcus aureus* isolates (7/43) were resistant to ciprofloxacin, higher than the rate reported in the Australian community in 2017 for blood isolates (6.2%), and for isolates from other sources (4.5%). All *Streptococcus pneumoniae* isolates were susceptible to cefalotin and ciprofloxacin, indicating lower resistance than the background rates in isolates from other sites reported from the Australian community in 2017 (3.9% for blood culture isolates). All *Pseudomonas aeruginosa* isolates were susceptible to ciprofloxacin and gentamicin. This differs from the AMR rates in the Australian community in 2017, where the AMR rate for ciprofloxacin was 6.4% and for gentamicin 5.7%. We note, however, that the numbers of isolates in our study were small relative to the national data set and from a single site, so differences might be expected.

In the United States, the Antibiotic Resistance Monitoring in Ocular microorganisms (ARMOR) surveillance studies report trends in antibiotic resistance of organisms isolated from ocular infections, providing data to assist health care practitioners in making informed choices regarding the treatment of ocular infections.
with ophthalmic antibiotics. However, they do not provide recommendations for therapeutic combinations.\textsuperscript{21}

A study from the United Kingdom, published in 2017, reported an analysis of AMR trends in isolates causing microbial keratitis at a tertiary hospital. In this study, similar empiric alternatives to our Centre were investigated, where ofloxacin was the first-line empiric therapy, and the second line cefuroxime plus gentamicin. The key findings included that ofloxacin remained an effective first line therapy; nonetheless, dual therapy offered broader coverage for clinical isolates in that setting. However, alternative, combination empiric antibiotic therapies were not investigated. There were no definitive recommendations for treatment and there was no further analysis of alternate antibiotic regimens in that study.\textsuperscript{19}

In 2014, a systematic review and meta-analysis using Cochrane methodology was undertaken to evaluate the efficacy of topical antibiotics in the management of bacterial keratitis. This review identified that, despite numerous clinical trials, there remained a lack of consensus as to which topical antibiotics, and which regimen (i.e., monotherapy or combination therapy) provided superior outcomes. Whilst there was no evidence of difference in comparative efficacy between fluoroquinolone monotherapy and aminoglycoside–cephalosporin combination treatment options in the management of BK, there were differences in safety profiles. When compared with aminoglycoside–cephalosporin combination therapy, fluoroquinolones had benefits in terms of reduced ocular discomfort and chemical conjunctivitis, but an increased risk of white corneal precipitates with the use of ciprofloxacin drops.\textsuperscript{28}

These studies indicate that empiric recommendations for bacterial keratitis vary with regards to fluoroquinolone monotherapy versus combination therapy, without clear superiority demonstrated in clinical studies. The prime recommendation for empiric topical antibiotic in Australia, according to Therapeutic Guidelines – Antibiotic is with 0.3% ciprofloxacin or 0.3% ofloxacin.\textsuperscript{23} In this study we compared AMR profiles from clinical isolates from patients with presumed bacterial keratitis covered with fluoroquinolone monotherapy (0.3% ciprofloxacin or 0.3% ofloxacin) or fortified combination therapy (5% cephazolin plus 0.9% gentamicin; chloramphenicol 0.5% plus gentamicin 0.9%; or 0.5% chloramphenicol plus 0.3% ciprofloxacin or ofloxacin).

In the present study, as with the findings from elsewhere,\textsuperscript{19,21} gram-positive bacteria (coagulase-negative staphylococci, \textit{Staphylococcus aureus} and \textit{Streptococcus pneumoniae}) were the most commonly-isolated organisms from the cornea and accounted for 75% of isolates. Our detection of \textit{Pseudomonas aeruginosa} in 13% of the total of 374 bacteria isolated is important, as it provides evidence that empiric topical antibiotic therapy must include an effective antipseudomonal agent rather than narrower approaches that target only commonly-isolated gram-positive bacteria.

Our findings support the current recommendations in Australia, \textit{Therapeutic Guidelines – Antibiotic, Version 16, 2019,}\textsuperscript{23} for the empiric, topical antibiotic therapy of bacterial keratitis. In our setting, there was greater \textit{in vitro} antibiotic coverage with combinations including chloramphenicol and an anti-pseudomonal agent (either gentamicin or a fluoroquinolone). The combination of chloramphenicol 0.5% plus gentamicin 0.9% was statistically better than the regimens of either 0.3% ciprofloxacin/0.3% ofloxacin (\(p = 0.007\)) or fortified 5% cephazolin plus 0.9% gentamicin (\(p = 0.005\)). The combination of 0.5% chloramphenicol plus 0.3% ciprofloxacin/0.3% ofloxacin was also superior to the regimens of 0.3% ciprofloxacin/0.3% ofloxacin monotherapy (\(p \leq 0.001\)) and 5% cephazolin plus 0.9% gentamicin (\(p = 0.003\)).

Whilst our data represents a limited sample size, it demonstrates an overall fluoroquinolone resistance of 5.3% (95% CI: 3.1–7.6%); rates for cefalotin/cefazolin plus gentamicin were similar, with an overall rates of resistance of 4.8% (95% CI: 2.6–7.0%) while the combinations of
chloramphenicol plus gentamicin resistance was 1.9% (95% CI: 0.5–3.2%), and chloramphenicol plus ciprofloxacin/ofloxacin was 1.3% (95% CI: 0.2–2.5%).

In 2020, the lack of monitoring of AMR in ophthalmic practice seems unwise in this era of otherwise well-placed caution. It is known that susceptibility patterns change according to climate and geographical region, and can fluctuate over time. A coordinated national program is urgently needed across Australia to provide wider-scale information on AMR in bacterial keratitis.

Acknowledgements

Professor Stephanie Watson is supported by the Sydney Medical School Foundation. The Sydney Eye Hospital Foundation supported the study. We thank Ryanbi Pratama BSc (Hons) for the data extractions for this study.

Conflict of interest

None declared. All authors contributed to this work.

Funding source

The Sydney Eye Hospital Foundation provided funding for this work. Professor Watson was supported by an NHMRC Career Development Fellowship (APP1050524) and is supported by a Sydney Medical School Foundation Fellowship.

Author details

Prof Stephanie L. Watson PhD FRANZCO
A/Prof Barrie J Gatus MRCP DTM&H FRCPA MD
Dr Maria Cabrera-Aguas PhD MIPH MBBS
Dr Benjamin H Armstrong MBBS BBioMedSci
Dr CR Robert George PhD FRCPA
Ms Pauline Khoo BSc (Hons)
Prof Monica M Lahra PhD FRCPA

1. The University of Sydney, Save Sight Institute, Discipline of Ophthalmology, Faculty of Medicine and Health, Sydney, NSW, Australia
2. Sydney Eye Hospital, Sydney, NSW, Australia
3. WHOCC for STI and AMR, NSW Health Pathology Microbiology, The Prince of Wales Hospital, Randwick, NSW, Australia
4. School of Medical Sciences, University of New South Wales, NSW, Australia
5. NSW Health Pathology Microbiology John Hunter Hospital, NSW, Australia

Corresponding author

Dr Maria Cabrera-Aguas
The University of Sydney, Save Sight Institute.
Level 1, South Block, Sydney Hospital
8 Macquarie Street, Sydney NSW 2000
Phone: + 61 431 737 428
Email: maria.cabreraaguas@sydney.edu.au
References

1. Australian Government. Antimicrobial resistance: Responding to the threat of antimicrobial resistance. Australia’s first national antimicrobial resistance strategy 2015–2019. [Internet.] Canberra: Australian Government; 30 June 2015. Available from: https://www.amr.gov.au/resources/national-amr-strategy.

2. Annunziato G. Strategies to overcome antimicrobial resistance (AMR) making use of non-essential target inhibitors: a review. Int J Mol Sci. 2019;20(23):5844.

3. Australian Commission on Safety and Quality in Health Care (ACSQHC). AURA 2019: third Australian report on antimicrobial use and resistance in human health. [Internet.] Sydney: ACSQHC; 2019. Available from: https://www.safetyandquality.gov.au/our-work/antimicrobial-resistance/antimicrobial-use-and-resistance-australia-surveillance-system-aura/aura-2019.

4. Organisation for Economic Co-operation and Development (OECD). Stemming the Superbug Tide: Just A Few Dollars More. Paris: OECD; 7 November 2018. [Accessed on 9 November 2020.] Available from: http://www.oecd.org/health/stemming-the-superbug-tide-9789264307599-en.htm.

5. World Health Organization (WHO). Global Action Plan on Antimicrobial Resistance. [Internet.] Geneva: WHO; 2015. Available from: https://www.who.int/antimicrobial-resistance/global-action-plan/en/.

6. Shalchi Z, Gurbaxani A, Baker M, Nash J. Antibiotic resistance in microbial keratitis: ten-year experience of corneal scrapes in the United Kingdom. Ophthalmology. 2011;118(11):2161–5.

7. Centers for Disease Control and Prevention (CDC). Antibiotic resistance threats in the United States, 2013. Washington DC: United States Department of Heath and Human Services, CDC; 2013.

8. WHO. Antimicrobial resistance: global report on surveillance, 2014. [Internet.] Geneva: WHO; 2014. Available from: https://www.who.int/antimicrobial-resistance/publications/surveillancereport/en/.

9. UK AMR Strategy High Level Steering Group. DH UK 5 Year Antimicrobial Resistance (AMR) Strategy 2013-2018. Annual progress report, 2015. London: United Kingdom Government Public and International Health Directorate, Health Protection and Emergency Response Division; 2016. Available from: https://assets.publishing.service.gov.uk/government/uploads/system/uploads/attachment_data/file/553496/2nd_UK_AMR_annual_report.pdf.

10. Whitcher JP, Srinivasan M, Upadhyay MP. Corneal blindness: a global perspective. Bull World Health Organ. 2001;79(3):214–21.

11. Robaei D, Watson S. Corneal blindness: a global problem. Clin Exp Ophthalmol. 2014;42(3):213–4.

12. Allan BD, Dart JK. Strategies for the management of microbial keratitis. Br J Ophthalmol. 1995;79(8):777–86.

13. Khoo P, Cabrera-Aguas M, Robaei D, Lahra MM, Watson S. Microbial keratitis and ocular surface disease: a 5-year study of the microbiology, risk factors and clinical outcomes in Sydney, Australia. Curr Eye Res. 2019;44(11):1195–202.

14. Li Y, Hong J, Wei A, Wang X, Chen Y, Cui X et al. Vision-related quality of life in patients with infectious keratitis. Optom Vis Sci. 2014;91(3):278–83.

15. Keay L, Edwards K, Naduvilath T, Taylor HR, Snibson GR, Forde K et al. Microbial keratitis: predisposing factors and morbidity. Ophthalmology. 2006;113(1):109–16.
16. Butler TKH, Spencer NA, Chan CCK, Singh Gilhotra J, McClellan K. Infective keratitis in older patients: a 4 year review, 1998–2002. *Br J Ophthalmol*. 2005;89(5):591–6.

17. Hong J, Chen J, Sun X, Deng S, Chen L, Gong L et al. Paediatric bacterial keratitis cases in Shanghai: microbiological profile, antibiotic susceptibility and visual outcomes. *Eye (Lond)*. 2012;26(12):1571–8.

18. Watson S, Cabrera-Aguas M, Khoo P, Pratama R, Gatus BJ, Gulholm T et al. Keratitis antimicrobial resistance surveillance program, Sydney, Australia: 2016 annual report. *Clin Exp Ophthalmol*. 2019;47(1):20–5.

19. Tan SZ, Walkden A, Au L, Fullwood C, Hamilton A, Qamruddin A et al. Twelve-year analysis of microbial keratitis trends at a UK tertiary hospital. *Eye (Lond)*. 2017;31(8):1229–36.

20. Leibovitch I, Lai TF, Senarathe L, Hsuan J, Selva D. Infectious keratitis in South Australia: emerging resistance to cephazolin. *Eur J Ophthalmol*. 2005;15(1):23–6.

21. Asbell PA, Sanfilippo CM, Pillar CM, DeCory HH, Sahm DF, Morris TW. Antibiotic resistance among ocular pathogens in the United States: five-year results from the antibiotic resistance monitoring in ocular microorganisms (armor) surveillance study. *JAMA Ophthalmol*. 2015;133(12):1445–54.

22. Green M, Carnt N, Apel A, Stapleton F. Queensland Microbial Keratitis Database: 2005–2015. *Br J Ophthalmol*. 2019;103(10):1481–6.

23. Therapeutic Guidelines Limited. Keratitis. Bacterial keratitis. [Internet.] Melbourne: Therapeutic Guidelines Limited; April 2019. Available from: https://tgldcdp.tg.org.au/view Topic?topicfile=keratitis#toc_d1e71.

24. Robaei D, Naunton M, Watson S. Seeing red: over-the-counter chloramphenicol. *Clin Exp Ophthalmol*. 2015;43(2):99–100.

25. Khoo P, Cabrera-Aguas M, Hollhum R, Watson S. Cornea scraping guidelines for microbial keratitis. *Clin Exp Ophthalmol*. 2017;45 (Suppl 1):84.

26. Bell SM, Pham JN, Rafferty DL, Allerton JK. Antibiotic susceptibility testing by the CDS method: a manual for medical and veterinary laboratories. (Eighth edition, 2016.) Kogarah: St George Hospital, Department of Microbiology (SEALS); 2016. Available from: http://cdtest.net.

27. Austin A, Schallhorn J, Geske M, Mannis M, Lietman T, Rose-Nussbaumer J. Empirical treatment of bacterial keratitis: an international survey of corneal specialists. *BMJ Open Ophthalmol*. 2017;2(1):e000047.

28. McDonald EM, Ram FSF, Patel DV, McGhee CNJ. Topical antibiotics for the management of bacterial keratitis: an evidence-based review of high quality randomised controlled trials. *Br J Ophthalmol*. 2014;98(11):1470–7.

29. Asbell PA, Colby KA, Deng S, McDonnell P, Meisler DM, Raizman MB et al. Ocular TRUST: nationwide antimicrobial susceptibility patterns in ocular isolates. *Am J Ophthalmol*. 2008;145(6):951–8.

30. Asbell PA, Pandit RT, Sanfilippo CM. Antibiotic resistance rates by geographic region among ocular pathogens collected during the ARMOR surveillance study. *Ophthalmol Ther*. 2018;7(2):417–29.

31. Blanco N, Perencevich E, Li SS, Morgan DJ, Pines L, Johnson JK et al. Effect of meteorological factors and geographic location on methicillin-resistant *Staphylococcus aureus* and vancomycin-resistant enterococci colonization in the US. *PLoS One*. 2017;12(5):e0178254.