SCOTTISH HOSPITALS INQUIRY

Bundle of document for Oral hearings commencing from 19 August 2025 in relation to the Queen Elizabeth University Hospital and the Royal Hospital for Children, Glasgow

Bundle 44 – Volume 8

Miscellaneous Documents

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Scottish Hospitals Inquiry
Request for Information from Shona Cairns and NSS Data Science Team

Introduction

1. The Inquiry is grateful to Ms Cairns and the NSS data science team for 28 May 2025 review of the HAD Report and the 20 June 2025 review of Chapter 8 of the HAD Report and Ms Cairns' statement for the Glasgow 4 hearing.

2. In her statement Ms Cairns has clarified the baseline used in the "Review of NHSGG&C paediatric haemato-oncology data" report of October 2019 ("the HPS October 2019 Review") (Bundle 7, Document 6, Page 224), but further issues have arisen in respect of that review prompted by the HAD Report and the work done by its authors and those who have commented on it since March 2025. These might help in putting the conclusions of the HAD Report and the HPS October 2019 Review in context. It would be of great assistance to the Inquiry if Ms Cairns and NSS Data Science Team could provide a short report that addresses the following questions that arise directly from the HPS October 2019 Review.

Review of Denominator Data

3. In the HPS October 2019 Review the authors validated activity data produced by NHS GGC by comparing it to activity data held by HPS. This is set out in Figure 2. The Occupied Bed Data used in the HAD Report is produced in Bundle 44, Volume 2, Document 8, Page 94 and its derivation explained in "Methodology of obtaining admissions data" (Bundle 44, Volume 1, Document 39, Page 666). This activity data overlaps with Figure 2 for a period from mid 2013 to mid 2019. An informal comparison of the table used by the HAD authors and Figure 2 suggests that the data sets may be different to some extent.

4. It would assist the Inquiry if NSS could:

    a. Produce the monthly totals of occupied bed days that underly Figure 2, in the form of a table, and

b. Compare them to the Occupied Bed Data used in the HAD Report for the overlap period.

5. Ms Cairns may wish to make observations about the significance of any differences, but these can be subject of discussion at the Glasgow 4, Part 2 hearing with Ms Cairns, the authors of the HAD Report and other witnesses.

6. A submission has been made to the Inquiry that "when Yorkhill moved to RHC there was an intended reduction in activity to allow the service to orientate itself before commencing to a full operational status – this would have been the reason for the decrease in the rates in the first 6 months". Can Ms Cairns detect any such reduction in activity in the Schiehallion Unit in Figure 2?

Rates of infection over longer periods

7. Counsel to the Inquiry has consulted with both Mr Mookerjee and Dr Drumright and would welcome the opportunity to see if any inferences can be drawn between the rates of BSI at (a) Yorkhill, (b) RHC before the decant and (c) RHC after the decant. This information is contained (to some degree) in the HPS October 2019 Review as it extends back to mid 2013.

8. It would assist the Inquiry if NSS could return to the data used to produce Figures 4 to 7 of the HPS October 2019 Review and calculate an aggregate Rate per 1,000 Total Occupied Bed Days for each figure in the review that covers three periods of time:

a. Yorkhill prior to RHC Opening on 10 June 2016,

b. RHC from 10 June 2016 to the decant to Wards 6A/4B on 28 September 2018, and

c. RHC from 28 September 2018 until the end of that data.

9. Ms Cairns may wish to make observations about the significance of any such calculated rates and whether the length of time has an impact on the weight that can be given to any such rate, but this can be subject of discussion at the

Glasgow 4, Part 2 hearing with Ms Cairns, the authors of the HAD Report and other witnesses.

Theme	Summary of Challenge/Position	Sid Mookerjee	Dr Sara Mumford	Prof Stevens, Prof Wilcox & Gaynor Evans	Dr Shona Cairns, NHS NSS
1.**HAD Report Methodology**	Infection rates alone are insufficient; must integrate environmental Sampling and epidemiological context	Explicitly challenges HAD's reliance on infection rates; calls for environmental and epidemiological data	Strongly critiques HAD for not considering environmental data and for speculative conclusions.	Agree- stress need for multifactorial analysis, not just infection rates	Agrees- emphasises need for integrated clinical, epidemiological and environmental analysis
2.**Benchmarking and Comparators**	Internal hospital comparisons are inadequate; need UK-wide benchmarking	Advocates for national benchmarking, not just local.	Explicitly states HAD failed to benchmark externally.	Agree-recommend UK-wide comparators for context	Agrees- note risk of internal only comparisons; recommends broader benchmarking
3.**Evidence of Environmental Contamination**	Strong, persistent contamination of water and (to some extent) ventilation systems with gram negative pathogens and biofilms	Cites multiple reports confirming widespread contamination	Provides detailed history and evidence of biofilm and contamination	Agree- References environmental sampling, biofilm and persistent contamination.	Agrees- Notes persistent contamination and challenges HAD's minimisation.
4.**Epidemiiological Correlation**	Infection rates of environmental organisms correlate with environmental contamination; supports causal link (Bradford Hill criteria)	Presents statistical evidence and epidemiological postulates	References same data and supports causal inference.	Agree- Cites correlation and Bradford Hill criteria	Agrees- Links infection trends to environmental data

5.**Mircoorganism Classification**	Disagreement with HADS inclusion of non-environmental/enteric organisms; need for standardised, relevant pathogen definitions.	Provides comparative tables and caveats on organism lists.	Criticises HAD for misleading classifications and inclusion/exclusion criteria.	Agree- call for standardised pathogen definitions for outbreak investigation.	Agrees- notes misclassification risk and confusion in HAD approach
6.**Outbreak Definition & IPC Practice**	Outbreaks should be identified promptly based on epidemiological signals, not delayed for definitive source or WGS confirmation.	Emphasises early action and IPC best practice	Details accepted UK IPC outbreak definitions and criticised HAD's restrictive approach	Agrees- support rapid intervention and risk-based response.	Agrees- notes importance of timely IPC response and established outbreak definitions.
7.**Complexity of Infection Sources**	Rejects HAD's "gut translocation only" theory; environmental acquisition is significant, especially in single room settings.	Argues endogenous and exogenous can co-exist	Details environmental acquisition pathways and challenges gut-only explanation	Agree- notes multiple transmission routes, including environmental	Agrees- emphasises environmental role in patient colonisation and infection
8.**Antimicrobial Usage Interpretation**	Disputes HAD's conclusions about antibiotic usage and resistance patterns; data do not support HAD's assertions.	Highlights data inconsistencies and misinterpretations.	Provides detailed prescribing data to refute HAD's claims	Agree- note HAD's misinterpretation of antimicrobial data.	Agrees- supports data driven analysis and correct interpretation.

9.**Impact of Remedial Actions**	Infection rates decreased following environmental remediation (e.g water system upgrades, ventilation improvements), supporting environmental causation	Notes infection rate drop post remediation	Cites decline in infections after remedial works	Agree- link intervention to outcome and reduction in infection rates	Agrees- note temporal association between remediation and infection reduction.
10.**Compehensive Data Review**	HAD report failed to consider all available data and prior reports, leading to incomplete or inaccurate conclusions.	Criticises HAD for selective data use	Reiterates HAD's omission of key reports and data	Agree- call for holistic data integration and review.	Agrees- notes incomplete analysis and need for comprehensive review.

Key

- Sid Mookerjee: Glasgow-4 Review (Epidemiology)
- Dr Sara Mumford: Microbiology/IPC Perspective, 16 May 2025
- Prof Stevens, Prof Wilcox & Gaynor Evans: Independent Panel, Case Note Review
- Dr Shona Cairns, NHS NSS: NHS National Services Scotland

Summary:

All four reports—Mookerjee, Mumford, Stevens/Wilcox/Evans (Independent Panel), and Cairns (NHS NSS)—are in strong agreement on the major thematic challenges to the HAD report. Their consensus covers methodological flaws, the need for robust benchmarking, strong evidence of environmental contamination, the importance of standardised definitions, and the necessity of comprehensive data review and rapid IPC response.

Omission of Mycobacterium Chelonae in the HAD report and the commentary that relates to this.

Report/Author	Did HAD Omit Mycobacterium Chelonae?	Commentary/Significance	Broader Context/Implications
Dr Sid Mookerjee	YES	Table 1 (pages 7-11) shows M. chelonae included in HPS and other expert lists but omitted in the HAD report Organisms tables	Argues that HAD's selective inclusion of organisms leads to underestimation of environmental contamination and its clinical impact.
Dr Sara Mumford	YES	Explicitly references M Chelonae as a key environmental organism identified in environmental and clinical samples (see Mumford report, Ch.6)	Emphasises that failure to include M. Chelonae distorts the true picture of water system contamination and risk to immunocompromised patients.
Prof Stevens, Prof Wilcox, Gaynor Evans (Independent Panel CNR)	YES	Their CNR and organism tables (as referenced in Mookerjee's tabulation) include M Chelonae as a relevant environmental pathogen.	State that omitting M. Chelonae results in an incomplete assessment of the water systems role in hospital – acquired infections.
Dr Shona Cairns NHS NSS	YES	NHS NSS/HPS reports (2019) specifically investigated M Chelonae outbreaks and	Argues that the omission undermines the credibility of the HAD report's conclusions

		documented its presence in water and clinical cases.	about environmental contamination.

Key Direct References

- Sid Mookerjee, Glasgow-4 Report: Table 1 and organism comparison (pp. 7–11); footnote 8 (HPS SBAR on M. chelonae).
- Dr Sara Mumford: Chapter 6, especially paras 6.13–6.14 and references to HPS and NHS NSS reports.
- Prof Stevens, Prof Wilcox & Gaynor Evans: Organism tables and commentary as referenced in Mookerjee's Table 1.
- Dr Shona Cairns, NHS NSS: NHS NSS/HPS reports on M. chelonae (see also Mookerjee, p. 7, footnote 8).

Key Points of Consensus

- All four reports independently identify the omission of Mycobacterium Chelonae by HAD
- All consider this omission a significant flaw that undermines the validity of HAD's environmental risk assessment
- All stress that Mycobacterium Chelonae was a documented, clinically relevant waterborne pathogen at QEUH/RHC recognised by national surveillance and incident management teams.
- All recommended comprehensive inclusion of such organisms for accurate outbreak investigation and infection risk evaluation.

Summary of Consensus

All four reports explicitly recognise the omission of Mycobacterium chelonae from the HAD report as a significant flaw, referencing national incident management and surveillance documentation. They agree that this omission undermines the accuracy and completeness of the HAD report's environmental risk assessment and call for comprehensive inclusion of all relevant waterborne pathogens in outbreak investigations.

Report/Author	Highlighted Paragraphs as per author
Dr Sid Mookerjee and Dr Sara Mumford	2.2...insufficient ventilation in ward 2A/2B contributed to the high levels of infection with environmental organisms, due to the failure to dilute and remove these, resulting in environmental contamination, and when looking at increases in infections this cannot be assumed to be restricted to typical airborne pathogens. As a conclusion to the preceding paragraphs in 6.1, this statement is, in our expert opinion, incomplete and fails to take into account the wider context of infection prevention mitigation which ventilation provides. 2.7..The failure to dilute contaminants in the air is, in our expert opinion, more likely to result in increased infection rates generally in immunocompromised patients than an observable increase in invasive aspergillus, only detected retrospectively with statistical analysis due to the small numbers of aspergillus infections seen in this group of patients. This is confounded further by the use of prophylactic anti-fungal drugs in patients perceived to be at risk of IA where ventilation is deemed to be inadequate. As a result, the observed incidence of invasive aspergillosis can be assumed to be an underestimate of the true risk of infection due to inadequate ventilation. 2.8... Stringent infection prevention and control and ventilation measures are put in place to prevent nosocomial (as opposed to endogenous) aspergillus infection in these patients. High quality enhanced ventilation with HEPA filtration of incoming air,

	high air changes of 12 air changes per hour (ach), and 6 Pa of positive pressure to the corridor are collectively required to minimise the risk of infection. 2.34 In summary, the evidence discussed in sections 2.23 – 2.34 does not support the authors of the HAD report in their assertion that the there were 'low absolute numbers of cases' as whilst the numbers may appear relatively small, the rates are clearly higher than would be expected in a new facility such as RHC and overall significantly higher at RHC than Yorkhill.
Prof Stevens, Prof Wilcox, Gaynor Evans (Independent Panel CNR)	We did not evaluate the involvement of the hospital ventilation systems or the airborne transmission of pathogens in the CNR but offer a few observations. In an introduction to Chapter 5 in the Case Note Review Overview Report[1] we mentioned 'chilled beam' systems (a type of radiation/convection heating, ventilation, and air conditioning system designed to heat and cool buildings) in passing only. We also cited the findings of a 2020 Independent Review,[2] which highlighted deficits in the hospital environment that included (but were not limited to) issues such as:

[1] Bundle 6, Document 38, Page 1033, Section 5.1.

[2] Bundle 27, Volume 9, Document 11, Page 145.

	the design and maintenance of the water system; lower than required air exchange in patient rooms and inadequate positive pressure protection of patient rooms; the lack of provision of particulate (HEPA) filtration in some higher risk patient areas; and uncertainties around the appropriate utilisation of chilled beams for temperature control in rooms used for immunocompromised patients[3]. We referred further to chilled beams when reviewing the built environment and its maintenance[4], describing problems with *"….maintenance of chilled beams following reports about leaks or condensation, or both, and where additional cleaning was required for control of dust".* We note that Hawkey et al. do not reference the 2020 Independent Review and nor are chilled beams mentioned.
Dr Shona Cairns NHS NSS	4.2 A description of the total number of patients and cases/episodes of infection would have assisted with interpretation to understand whether there were patients with multiple episodes and the impact this may have on the conclusion.

[3] Inkster T, Peters C, Soulsby H. Potential infection control risks associated with chilled beam technology: experience from a UK hospital. J Hosp Infect. 2020 Nov;106(3):613-616. https://doi.org/10.1016/j.jhin.2020.08.011

[4] Bundle 6, Document 38, Page 1036, Section 5.2

	4.3 Prior exposure history is particularly important when considering the links between *Aspergillus* spp. infections and the environment due to the long incubation period which can vary widely from days to months. 4.4 The *Aspergillus* spp. analyses do not include a denominator that accounts for activity… The lack of denominator in the analyses should have been acknowledged by the authors as a limitation. 4.5 Whilst a small number of cases can make epidemiological analyses more challenging, there are other methods that can be used to assess potential links to the environment. Root cause analyses can be used to assess each patient and explore the possibility of links. An important aspect of such an exercise would be consideration of a timeline that describes location information over a longer period rather than a point in time where a sample was taken. This was the approach taken in the Case Note Review. Adult Data 4.6.2 The limitations of the analyses and caveats have not been fully acknowledged in the strength of this conclusion. Paediatric Data 4.7.1 Whilst not possible to do a formal statistical test and no denominator has been used, there is period with a potentially higher number of cases that is not acknowledged by the authors. The authors have focused on the range of case numbers pre- and post-move to RHC but do not note that following the move there were 5 months between 2016 and 2020 where there were 4 or more cases identified. This is in contrast to before the move when there was only one month with 4 or more cases. Whilst it is not possible to draw a strong

	conclusion based on this data, it is worth acknowledging whilst considering the question relating to the ventilation system. 4.7.2 In Section 8.1.4 (page 130), the authors conclude that the analyses show low level variation that is expected over time. The limitations of the analyses and caveats have not been fully acknowledged in drawing this conclusion nor has the potential period where there was a higher number of cases.

National Services Scotland

Water Systems Literature review

Infection prevention and control (IPC) for safe healthcare water systems

Version 1.1

24 January 2025

ARHAI Scotland

Antimicrobial Resistance and Healthcare Associated Infection

Key information

Document title:	Literature review - Infection prevention and control (IPC) for safe healthcare water systems
Date published/issued:	24 January 2025
Date effective from:	24 January 2025
Version/issue number:	1.1
Document type:	Literature review
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Document information

Information	Description
Description:	This literature review examines the available professional literature on the infection prevention and control (IPC) aspects/impacts of the water system.
Purpose:	To inform the infection prevention and control (IPC) aspects/impacts of the water system in the National Infection Prevention and Control Manual in order to facilitate the prevention and control of healthcare associated infections/incidents in NHSScotland health and care settings.
Target Audience:	All NHS staff (including contractors and service delivery partners) involved in the prevention and control of infection in NHSScotland.
Update/review schedule:	Updated as new evidence emerges with changes made to recommendations as required. Review will be formally updated every 3 years with next review in 2027.
Cross reference:	National Infection Prevention and Control Manual
Update level:	**Practice** – The implications for practice are formulated based on a review of the available professional scientific literature on the infection prevention and control (IPC) aspects/impacts of the water systems. **Research** – The implications for research are formulated based on a review of the available professional scientific literature on the infection prevention and control (IPC) aspects/impacts of the water systems.

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Version history

This literature review will be updated in real time if any significant changes are found in the professional literature or from national guidance/policy.

Version	Date	Summary of changes
1.0	July 2024	Final for publication
1.1	January 2025	Literature review search strategy added as an appendix (appendix 5)

Approvals

Version	Date Approved	Name
1.0	July 2024	ARHAI Scotland Infection Control in the Built Environment & Decontamination (ICBED) Working Group
1.0	July 2024	ARHAI Scotland Community Infection Prevention & Control (CIPC) Working Group

Abbreviation list

Acronym	Definition
AGREE	Appraisal of Guidelines Research & Evaluation
ARHAI Scotland	Antimicrobial Resistance and Healthcare Associated Infection Scotland
BS	British Standard
BSI	British Standards Institution
CDC	Centers for Disease Control and Prevention (United States of America)
CF	Cystic Fibrosis
CFU	Colony Forming Unit
CPHM	Consultant in Public Health Medicine
CRE	Carbapenem-resistant Enterobacteriaceae
CRO	Carbapenem-resistant organism
CVC	Central Venous Catheter
ECDC	European Centers for Disease Control and Prevention
ERCP	Endoscopic Retrograde Cholangiopancreatography
HAI	Healthcare Associated Infection
HAI-SCRIBE	Healthcare Associated Infection System (for) Controlling Risk In the Built Environment
HCU	Heater Cooler Unit
HFS	Health Facilities Scotland
HIIAT	Healthcare Infection Incident Assessment Tool
HPSC	Health Protection Surveillance Centre
HP	Health Protection
HSE	Health and Safety Executive (UK)
ICBED	Infection Control in the Built Environment and Decontamination
ICD	Infection Control Doctor
ICM	Infection Control Manager
ICU	Intensive Care Unit
IMT	Incident Management Team
IPC	Infection Prevention and Control
ISO	International Organization for Standardization
NIPCM	National Infection Prevention and Control Manual
NNU	Neonatal Unit
NTM	Non-tuberculous Mycobacteria

Acronym	Definition
ORT	Outbreak Reporting Tool
PAG	Problem Assessment Group
PD	Published Document
POU	Point-Of-Use
SGHSCD	Scottish Government Health and Social Care Department
SHTM	Scottish Health Technical Memorandum
SIGN	Scottish Intercollegiate Guidelines Network
TMV	Thermostatic Mixer Valve
TVC	Total Viable Count
UKAS	United Kingdom Accreditation Service
UTI	Urinary Tract Infection
WHO	World Health Organization
WSG	Water Safety Group
WSP	Water Safety Plan

Contents

1 Objectives

The aim is to review the extant scientific literature regarding the Infection prevention and control (IPC) for safe healthcare water systems to inform evidence-based recommendations for practice.

The specific objectives of the review are to determine:

General information about water-associated organisms in healthcare settings

1. Which organisms associated with healthcare water systems are responsible for colonisation/infection of patients?
2. How do healthcare water system-associated organisms survive in the environment?
3. What are the causes/sources of environmental contamination with healthcare water system-associated organisms?
4. Which patient populations are considered as being at increased risk of colonisation/infection with a healthcare water system-associated organism?
5. What types of infection can healthcare water system-associated organisms cause?
6. What are the incubation periods of healthcare water system-associated organisms?
7. What is the period of communicability for healthcare water system-associated organisms?
8. What are the known transmission routes of healthcare water system-associated organisms?
9. Which healthcare procedures present an increased risk of transmission of healthcare water system-associated organisms?

Prevention and control of healthcare water system-associated infection

10. What are the microbiological water testing requirements at commissioning?
11. What are the responsibilities of the IPC team in regards to water safety at commissioning?
12. Is routine water testing to detect healthcare water system-associated organisms recommended?
13. What are the recommended microbiological limits for healthcare water system-associated organisms?
14. How frequently should routine water testing be conducted?
15. When should routine water testing frequency be increased?
16. Where should routine water samples be taken from (which outlets, how many samples)?
17. When should water samples from further back in the system be taken?
18. Who should water test results be reported to?
19. How should routine water test results be interpreted?
20. What are the water testing requirements following a positive test result (in the absence of clinical cases)?
21. What actions (remedial and/or clinical) should be taken following a positive test result (in the absence of clinical cases)?
22. Is routine environmental testing for healthcare water system-associated organisms recommended?
23. Are there any specific actions required if an outlet tests positive pre-flush but negative post-flush?

24. Are there any recommended methods for the removal of healthcare water system contamination?
25. What flushing regimes are recommended for healthcare settings?
26. Who should be responsible for flushing?
27. What actions can be undertaken to reduce the risk of infection/colonisation associated with direct water usage?
28. What actions can be undertaken to reduce the risk of infection/colonisation associated with indirect water usage?
29. What actions can be undertaken to facilitate the earliest possible detection and preparedness for clinical cases of water-associated colonisation or infection?

Outbreak/incident management

30. How should water-associated incidents be assessed and reported locally and nationally?
31. What are the water testing requirements during a water-associated incident/outbreak?
32. What are the environmental testing requirements when investigating healthcare water system-associated incidents/outbreaks?
33. How and by whom should water-associated incidents be investigated?
34. Should point-of-use (POU) filters be fitted in response to water-associated incidents/outbreaks?
35. When can POU filters be removed?

Organisational management

36. Whose responsibility is it to carry out any of the above actions?

2 Methodology

This targeted literature review was produced using a defined methodology as described in the National Infection Prevention and Control Manual: Development Process. The complete search strategy is provided in Appendix 5. Database searches were performed on 20 December 2022. This review screened for evidence on both non-acute (including care homes) and acute healthcare settings. However, most of the evidence was specific to acute settings. When evidence on non-acute settings is included, this is specifically highlighted. In addition to the exclusion criteria outlined in the NIPCM Development Process the following evidence was excluded from this review:

- studies without a strong epidemiological link between patient and environmental samples (not showing a link via molecular typing)
- evidence specific to dental unit waterlines (as this is covered in Literature Review and Recommendations: Management of Dental Unit Waterlines)

In total, 2808 individual pieces of evidence were retrieved using the search strategy. Details regarding the screening process are summarised in a PRISMA flowchart presented in Appendix 3 (adapted from: Moher D, Liberati A, Tetzlaff J, Altman DG, The PRISMA Group (2009). Preferred Reporting Items for Systematic Reviews and Meta-Analyses: The PRISMA Statement. PLoS Med 6(7): e1000097).

Consultation was received in addition to the ARHAI Scotland Working Groups from the following individuals:

- Tim Wafer (The Water Solutions Group)
- Dr Susanne Surman-Lee (Leegionella Ltd)
- Dennis Kelly (Pro LP Consulting Ltd)
- Dr Michael Weinbren

Engineering colleagues in Health Facilities Scotland (HFS) were consulted throughout the process.

3 Discussion

3.1 Implications for practice

3.1.1 General information about water-associated organisms in healthcare settings:

1. Which organisms associated with healthcare water systems are responsible for colonisation/infection of patients?

In total, 95 pieces of evidence were identified in relation to this research question which includes 80 outbreak studies[1-80] (graded SIGN50 level 3), eight surveillance studies (graded SIGN50 level 3),[81-88] one systematic literature review (graded SIGN50 level 2+),[89] one cohort study (SIGN50 level 3),[90] one before and after study (SIGN50 level 3),[91] two case reports (SIGN50 level 3) [92, 93] and two guidance documents (SIGN50 level 4). [94, 95]

Studies were included if the environmental source of the infection and/or colonisation incident could be linked to patient cases by molecular typing as this provides some rigour to outbreak studies. Studies were excluded if microbial typing was not performed. Molecular typing assesses the genetic material present in microorganisms, allowing comparison of the microorganisms present in clinical and environmental samples. A range of typing methods were described in the literature, and these included pulsed-field gel electrophoresis (PFGE), amplified fragment length polymorphism (AFLP), repetitive-element polymerase chain reaction (rep-PCR), random amplification of polymorphic DNA (RAPD), variable-number tandem repeat (VNTR) typing, single locus sequence typing (SLST), multi-locus sequence typing (MLST), and whole genome sequencing (WGS). It is important to note that these methods vary widely in terms of discriminatory power. Another limitation is the limited availability of typing for some microorganisms. A more general limitation of environmental investigations is that a true link between patient cases and the environment can be overlooked if there is multi-species or multi-strain involvement; this is due to the limitations of sampling technique and microbiological analysis. When isolating microorganisms from environmental samples, multiple

colonies may be present on the sampling plate, however usually only one or two colonies are selected for identification – this is due to time and financial restrictions. This sampling technique limitation is common to all environmental outbreak studies and is a challenge to the accurate interpretation of results. The epidemiological concept of time, place and person is challenging to apply to environmental outbreaks if a cluster of infection or colonisation occurs over a lengthy time period and involves multiple seemingly unrelated microbial species.[53] The retrospective nature of outbreak studies often prevents an accurate analysis of events occurring at the point of exposure. Consequently, even when an environmental source is identified, the exact transmission event that led to infection or colonisation in the patient may remain unconfirmed and this was the case for much of the evidence identified for this research question. Conducting a case-control study as part of an outbreak investigation can add rigour, however very few of the outbreak studies included these.[8, 15, 37, 60, 66, 69, 75] There is no 'standard' reporting structure for outbreak studies therefore there is inconsistency in the type of information and level of detail provided. There is likely a risk of publication bias associated with this body of evidence as not all outbreaks that occur in healthcare settings are published in scientific journals.

The most frequently reported organisms within this body of evidence were gram-negative organisms, the majority being *Pseudomonas aeruginosa* (*P. aeruginosa*) (n=33 reports) or other *Pseudomonas* species (n=4 reports),[52, 54, 67, 70] followed by Enterobacteriaceae (n=26 reports). Other gram-negative bacteria (non-Enterobacteriaceae, non-*Legionella* species) were described in 10 reports.[17, 18, 23, 36, 39, 52, 53, 66-68] These microorganisms are often described in the literature as opportunistic pathogens, meaning that while the organism may be ever-present in the environment (including water) and rarely cause harm to healthy individuals, vulnerability (for example immune impairment) in certain patient groups provides the opportunity for colonisation/infection. Guidance from the United States of America (US) Centers for Disease Control and Prevention (CDC) describes the following gram-negative bacteria as clinically important opportunistic organisms present in tap water: *Legionella* species, *P. aeruginosa*, *Burkholderia cepacia* (*B. cepacia*), *Ralstonia pikettii, Stenotrophomonas maltophilia* (*S. maltophilia*), and *Sphingomonas* species.[94]

Non-tuberculous Mycobacteria (NTM) were described in 21 reports.[1, 3, 4, 6, 7, 9, 25, 26, 31, 43, 71-75, 78, 79, 87, 93, 95, 96] Guidance from the CDC describes healthcare water as the source of patient infection and/or colonisation with the following NTM: *M. abscessus, M. avium* complex*, M. chelonae, M. fortuitum, M. marinum, M, ulcerans*, and pseudo-outbreaks involving *M. chelonae, M. fortuitium, M. gordonae, M. kansasii, M. terrae*, and *M. xenopi*.[94] A limitation of the CDC guidance is that much of the evidence referenced was published prior to the year 2000 (the exclusion criteria for this literature review) and therefore may not reflect current practice in healthcare settings.[94]

Infection with *Legionella* species was described in four reports.[2, 5, 8, 92]

One report described infection with the fungus *Fusarium solani* linked to contaminated water tanks at a hospital in Brazil where taps and drains tested positive.[77]

There were 16 reports that detailed incidences/outbreaks that occurred in the UK. These involved the following microorganisms.

- In England: Enterobacteriaceae (*Escherichia coli* (*E. coli*) and *Klebsiella* species)[61, 81] *P. aeruginosa*,[10, 11, 83, 91] *Pseudomonas fluorescens* (*P. fluorescens*),[70] *S. maltophilia*,[13] and *Mycobacterium chimaera* (*M. chimaera*).[87, 95]
- In Scotland: *Pseudomonas* species (*P. aeruginosa, P. fluorescens, P. putida*),[52] NTM (*M. mucogenicum*, *M. chelonae*, *Mycobacterium* spp.),[9, 26] Enterobacteriaceae (*E. cloacae, K. oxytoca*, *K. pneumoniae*, *Pantoea* species, *Serratia marcescens*),[52] *Chrysomonas indologenes*,[52] *Acinetobacter ursginii*,[52] *S. maltophilia*,[52] and *Cupriavidus pauculus* (*C. pauculus*).[53]
- *P. aeruginosa* was reported in both Wales,[44] and Northern Ireland.[97]

It must be noted that the volume of literature identified for each organism may not be a reflection of the true clinical or environmental risk occurring or burden experienced in healthcare settings. There is a paucity of evidence regarding spontaneous patient cases as well as evidence that sheds light on the environmental prevalence of these microorganisms in healthcare settings and whether this varies geographically. Active screening of patients in two surgical ICUs at a hospital in China, undertaken to gain

knowledge of *P. aeruginosa* colonisation/infection patterns in absence of an outbreak, revealed 9.2% (55/595) spontaneous healthcare-associated patient cases of *P. aeruginosa* colonisation/infection.[82]

During development of this literature review, stakeholders requested that the microorganisms identified in literature associated with healthcare water that cause infection and/or colonisation in patients be summarised (where possible) according to their most likely source. Sources included: microbial proliferation within the water, microbial proliferation/contamination of plumbing parts/wastewater infrastructure, contaminated water-based equipment, and patients. Further detail regarding determination of source is provided in the research question 'What are the causes/sources of environmental contamination with healthcare water system-associated organisms?'.

Microbial proliferation within the water

Twelve outbreak studies detail infection incidents where microbial proliferation within the water system preceded patient colonisation or infection.[2-9, 78, 79, 92, 93] The organisms involved included NTM and *Legionella* species. These NTMs included *M. fortuitum*, *M. mucogenicum*, *M. simiae*, *M. abscessus*, *M. chelonae,* and *M. phocaicum*. The *Legionella* species included *Legionella pneumophila* serogroup 5, serogroup 1 and *L. pneumophila* (serogroup unspecified). One of the 12 outbreak studies was a pseudo-outbreak involving NTM *M. simiae* where positive clinical samples were obtained but in the absence of clinical colonisation or infection in the patient.[4] In pseudo-outbreaks, the clinical samples are effectively contaminated by a contaminated water source, often via diagnostic equipment.

Microbial proliferation/contamination of plumbing parts/wastewater infrastructure

Sixty-three outbreak studies describe microbial proliferation/contamination of plumbing systems/infrastructure. Most of these outbreak studies (n=32) involved patient colonisation and/or infection with *P. aeruginosa,*[10, 11, 13-16, 19-22, 24, 28, 30, 32-35, 38, 44, 45, 47, 51, 52, 54, 55, 82-86, 90, 91] (two of these also involved *Pseudomonas putida*).[52, 54]

Other microorganisms included Enterobacteriaceae, detailed in 23 reports (including *Klebsiella* species (K. *pneumoniae*, *K. oxytoca*), *Enterobacter* species (*E. cloacae*,

E. aerogenes), *Citrobacter* species (*C. freudii*, *C. koseri*), *E. coli*, *Serratia marcescens, Pantoea agglomerans,* and *Raoultella planticola*),[12, 27, 29, 37, 40-42, 46, 48-50, 52, 56-65, 81] *B. cepacia*,[17, 23] *Acinetobacter* species (*A. baumanii*,[18, 36, 39, 66] *A. ursingii*),[52] *C. pauculus*,[52, 53] *Chrysomonas indologenes*,[52] *S. maltophilia*,[52] and NTMs including *M. fortuitum*, *M. mucogenicum*, *M. canariasense*, *M. chelonae*, *M. chimaera*, and *M. gordonae*.[1, 25, 26, 31, 43, 96] Four of these 63 outbreak studies were pseudo-outbreaks where positive clinical samples were obtained but in the absence of clinical colonisation or infection in the patient; the microorganisms involved were *P. aeruginosa*, *M. chimaera*, *M. gordonae* and *M. fortuitum*.[1, 31, 33, 43]

Antibiotic resistance was reported in 38 outbreak studies and one systematic literature review, all involving gram-negative microorganisms.[12, 14, 15, 18-20, 27, 29, 30, 35-42, 44, 46, 48-51, 55-62, 64-66, 76, 80, 82, 84, 89] Many of these had resistance against the beta lactam group of antibiotics including extended-spectrum beta-lactamase-producing organisms (ESBLs) and carbapenem-resistant organisms (CROs) or carbapenem-resistant Enterobacteriaceae (CRE).

All of the Enterobacteriaceae identified in these outbreak studies, as well as *P. aeruginosa*, are commensal microorganisms meaning they can be found naturally in the human intestinal tract. Therefore, it is possible that patients could introduce these bacteria into healthcare settings, where they can contaminate or 'seed' the environment and/or indirectly transmit to other patients. Further analysis of the source of environmental contamination is covered in the research question 'What are the causes/sources of environmental contamination with healthcare water system-associated organisms?'.

Contaminated water-based equipment

In 13 outbreak studies, contamination of water-based equipment was responsible for colonisation and/or infection in patients, where the following microorganisms were involved: *P. aeruginosa*,[69] *P. putida*,[67] *P. fluorescens*,[70] *Stenotrophomonas maltophilia*,[67, 68] *Serratia marcescens*,[69] ESBL *Klebsiella oxytoca*,[76, 80] *M. chimaera*,[71, 87, 95] *M. chelonae*,[72-74] and *M. fortuitum*.[75] Infection incidents in multiple countries (including England) involving *M. chimaera* were identified to be associated with water heater-cooler units used during cardiac surgery.[71, 87, 95]

In summary, below is a list of the microorganisms associated with healthcare water systems identified in the literature (those with an asterisk (*) were identified in published UK incidents and outbreaks):

- *Acinetobacter* species (spp.) (*A. baumannii, A. ursingii**)
- *Burkholderia* spp. (*B. cepacia*)
- *Chryseomonas indologenes**
- *Cupriavidus pauculus**
- Enterobacteriaceae (*C. freundii, C. koseri, E. aerogenes, E. cloacae*, E. coli*. K. pneumoniae*, K. oxytoca*, Pantoea spp.*, P. agglomerans*, S. marcescens*, R. planticola*)
- *Fusarium solani*
- *Legionella* spp. (*L. pneumophila**)
- Nontuberculous mycobacteria (NTM)* (*M. avium* complex, *M. abscessus, M. canariasense, M. chelonae*, M. chimaera*, M. fortuitum, M. gordonae, M. kansasii, M. marinum, M. mucogenicum*, M. simiae, M. phocaicum, M. terrae, M. ulcerans, M. xenopi*)
- *Pseudomonas* spp. (*P. aeruginosa**, *P. putida**, *P. fluorescens**)
- *Stenotrophomonas maltophilia**
- *Sphingomonas* spp.

2. How do healthcare water system-associated organisms survive in the environment?

In total, 14 studies were identified to answer this research question which includes eight outbreak studies,[5, 7, 13, 21, 59, 61, 68, 98] two non-systematic reviews[99, 100] and three guidance documents[94, 101, 102] that were deemed expert opinions (including one Scottish[101]) and one surveillance study.[103] In accordance with SIGN 50 methodology, nine are considered level 3 evidence (eight outbreak studies[5, 7, 13, 21, 59, 61, 68, 98] and one surveillance study[103]) and five are considered level 4 evidence (expert opinions).[94, 99-102]

Many of the organisms identified in the literature (*Legionella* species, NTMs, *Pseudomonas* species) are naturally occurring in water, including drinking water and usually posing no threat to healthy persons. If allowed to proliferate within a healthcare water system, the risk of infection increases. The CDC reports that NTM, *Pseudomonas* spp. and other gram-negative, non-fermentative bacteria have minimal nutritional requirements and can tolerate the very low nutrient levels found in disinfected healthcare water systems.[94, 99] Moreover, tolerance of high temperatures (50-55°C) may allow certain species (for example NTM, *Legionella* spp.) to survive in hot water systems if they are not maintained above 55°C.[5, 94, 99, 101] Some organisms cannot tolerate high temperatures and are more frequently associated with cold water lines and taps.[94]

Routine chlorination is standard practice in healthcare to ensure water quality. Chlorine dioxide may also be added to water as a decontamination treatment in response to contamination issues. Some organisms, particularly NTM, have a high resistance to chlorination with the ability to tolerate free chlorine concentrations of 0.05–0.2 mg/L (0.05–0.2 ppm) found at the tap.[94, 98, 99] Subsequently, chlorine resistant organisms can persist in the environment despite ongoing routine chemical treatment.[98] Disinfection can also contribute to selection for proliferation and persistence, as disinfection kills off competitors consequently selecting for those organisms that can grow on low nutrient levels resulting from disinfection.[99]

A number of outbreak studies reported low residual chlorine levels at the tap which was hypothesised as a contributory factor to the survival of organisms in the water system.[7, 103] Low residual chlorine levels may be due to insufficient flow and/or low usage at the outlet.

The major determinant for survival and persistence of healthcare water system-associated organisms is the formation of biofilms within the water distribution system, where they can form on most surfaces including pipe work, tanks, taps and filters. NTM cells, for example, have a lipid-rich hydrophobic outer membrane which promotes their attachment to particulates in the water allowing them to form biofilms in water systems.[94, 99] Biofilms create a protective environment for growth, trapping nutrients, and providing resistance to chemical disinfectants and physical removal.[94] This provides a reservoir within a water system from which organisms can continue to contaminate water systems and components, even after attempts at targeted

disinfection and/or mechanical removal. A *P. aeruginosa* outbreak at an intensive care unit (ICU) was linked to biofilms within sink traps.[21] It was shown that biofilm reactivation can occur by addition of artificial nutrient broth after removing, sealing and filling the whole sink trap with water for up to six weeks. The biofilms also resisted decontamination with hydrogen peroxide. Even without visual evidence of biofilm, an indicator of biofilm existence is the persistent contamination of a water outlet even after repeated disinfection.[59, 61, 68] This may occur when biofilm detaches/sloughs off from pipework and is then detected at the outlet.

Protozoa, unicellular eukaryotic microorganisms that are ubiquitous in various environments including water, can ingest bacteria such as *Legionella* spp. and NTMs; the ability of these bacteria to survive within protozoa protects them from the effects of biocides and other disinfection strategies.[94, 99, 100, 102]

Many water system-associated organisms have the ability to survive on wet or damp surfaces therefore allowing widespread contamination of healthcare environments in damp areas, such as sinks and drains.[13] Once the organisms are established within these environments, they may persist for a long period of time.

In conclusion, many of the organisms identified in water system-associated infection incidents and outbreaks are ubiquitous to naturally occurring water sources and naturally possess some physical and biological properties that facilitate their survival and persistence within healthcare water systems. These include the ability to survive on low nutrient levels, resistance to high water temperatures, relative resistance to disinfection, survival within protozoa and the ability to form biofilms and/or survive in biofilms within the water distribution system.

3. What are the causes/sources of environmental contamination with healthcare water system-associated organisms?

In total, 97 pieces of evidence were identified in relation to this research question which includes 81 outbreak studies,[1-51, 53-80, 93, 96] five guidance documents that were deemed expert opinion (including one Scottish[104]),[94, 95, 105, 106] nine surveillance studies,[81-87, 90, 107] one systematic review[89] and one case report.[92] In accordance with SIGN 50 methodology, one was graded SIGN50 level 2 evidence (systematic review),[89] 91 were graded level 3 evidence (81 outbreak studies,[1-51, 53-80, 93, 96, 108]

seven surveillance studies,[81-84, 86, 87, 107] two prospective studies,[85, 90] one case report[92]) and five were graded level 4 evidence.[94, 95, 104-106]

The causes of proliferation within the water system are usually related to a failure of (one or more) controls including temperature control, chemical control, water flow or pressure that allows these organisms (some of which are naturally found in water) to survive and accumulate, often within biofilms. More detail regarding survival is provided in the research question 'How do healthcare water system-associated organisms survive in the environment?'. Biofilms can act as a reservoir for a multitude of microorganisms facilitating their survival within water systems, and providing protection from control measures such as heat and chlorine.[66] Biofilm formation within water systems was reported frequently in outbreak studies.[7, 24, 26, 29, 31, 34, 37, 48, 51, 59, 61, 66, 68, 74, 76, 94, 104, 105]

There are two possible sources of contamination (entry of microorganisms to water systems) prior to proliferation; exogenous organisms that originate from within the water itself, and endogenous organisms that originate from the patient.

Patients as the source (endogenous source)

All of the Enterobacteriaceae identified in these outbreak studies, as well as *P. aeruginosa*, are commensal microorganisms meaning they can be found naturally in the human intestinal tract. Therefore, it is possible that patients could introduce these bacteria into healthcare settings, where they can contaminate or 'seed' the environment and/or indirectly transmit to other patients. In three prospective surveillance studies, attempts were made to determine the contribution of patients (endogenous source) versus environmental (exogenous) sources to transmission of *P. aeruginosa.*[82, 85, 86] A French multi-centre study compared patient samples (screened regularly from admission) with pre-flush tap water samples to determine whether patient-to-patient transmission had occurred (deemed possible when a similar strain was isolated in more than two patients hospitalised during an overlapping time period in the absence of a matched water sample).[85] Possible patient-to-patient transmission was identified for 50.5% (86/170) of patients and an exogenous origin from tap water for 17.1% (29/170) of patients. It was not possible to draw conclusions for 55 patients because the same strain types were shared by many patients and tap water samples. When isolating microorganisms from

environmental samples, multiple colonies may be present on the sampling plate, however usually only one or two colonies are selected for identification – this is due to time and financial restrictions. Due to the limitations of sampling technique and microbiological analysis of water samples, the role of water sources in transmission described in this study may have been underestimated. Further, no additional environmental sites, for example drains or other water-based equipment, were sampled. A prospective surveillance study undertaken in two Chinese intensive care units (ICUs) found that 17.6% (6/34) of colonisations/infections with *P. aeruginosa* were most likely due to patient-to-patient transmission and 50% (17/34) from endogenous flora (diagnostic clinical sample identical to rectum and/or throat sample of the same patient).[82] Analysis of environmental samples showed that 64.7% (11/17) of exogenous sourced cases were associated with contaminated sink drains; none of the strains recovered from tap water matched patient strains. This suggests that the plumbing infrastructure rather than the water was the main environmental reservoir in these settings. A third prospective surveillance study, conducted in an ICU in France, demonstrated that both patients and the tap water (pre-flush samples) were reservoirs for transmission to subsequent patients, six patients were colonised with *P. aeruginosa* on admission.[86] These three prospective surveillance studies highlight the difficulties in determining both the source of contamination and the subsequent direction of transmission (patient-to-patient, environment-to-patient, patient-to-environment) when investigating water-system associated colonisations and infections.[82, 85, 86]

Inappropriate practices and behaviours of healthcare staff, patients and visitors can increase the risk of patient seeding of the environment. Practices identified in the literature (four outbreak studies) included the disposal of organic matter (food/drinks, body fluids/waste material) and residual antibiotics into sinks (some of which were designated for hand-washing only).[58, 60, 62, 84] Introduction of organic matter into sinks provides the nutrients to maintain biofilms.

Environmental source (Exogenous source)

Analysis of the literature identified for this research question allowed categorisation of the location of environmental contamination into system-wide (deeper within the system), and/or isolated to drains and distal outlets. Separate to the water system

itself, water-based equipment can also act as an environmental reservoir for ongoing transmission.

Water system contamination

Fifteen outbreak studies detail infection incidents where widespread water system contamination was identified (rather than isolated to distal ends/outlets).[1-9, 52, 53, 78, 79, 92, 93] The organisms involved included NTM and *Legionella* species. Two of the 15 outbreak studies were pseudo-outbreaks involving NTMs where positive clinical samples were obtained but in the absence of clinical colonisation or infection in the patient.[1, 4] Generally, in pseudo-outbreaks the clinical samples are effectively contaminated by a contaminated water source, often via diagnostic equipment.[1, 4]

A number of outbreak studies involving NTMs detail the presence of the outbreak species in the incoming mains water supplied by the region.[1, 3, 109] Widespread water system contamination is often evident where multiple outlets and samples from further back in the system, as well as from non-outbreak areas of the healthcare setting, test positive.[93] Proliferation of NTMs was reported after a generator failure caused a decrease in water pressure, allowing stagnant water (created during construction work) to flow into the water system of an oncology department; this was associated with subsequent cases of *M. mucogenicum* bloodstream infections.[79] Stagnation creates the conditions for biofilm to develop on pipework and fittings. Investigation of a *Legionella pneumophila* pneumonia outbreak found the hot-water distribution system was unbalanced, with parts of the hot and cold-water pipes in close proximity without thermal protection.[92] *Legionella* was identified at inlet and outlet points, suggesting widespread contamination in the system. Further, thermostatic mixing valves at the sinks contained stagnant water where a lime deposit was found. These combined failures are likely to have supported proliferation of the Legionellae throughout the water. Health Protection Scotland (HPS) and the UK Health and Safety Executive (HSE) guidance advise that a risk of *Legionella* proliferation exists when the water temperature in all or some parts of the system is between 20-45°C.[104, 106] Improvements to water system infrastructure can have unintended consequences if knowledge of the whole system is lacking. This was one of the issues causing a *Legionella* outbreak where instantaneous water heaters were installed within a hospital water system to improve temperature control. However, the

change in the water system infrastructure resulted in an increased mixing of water flow between two adjacent departments (each served by separate water recirculation loops). [8] Poor management and unidentified dead-legs within one of these loops had created stagnant water which then flowed into the haemato-oncology department when the system was restructured. Further to these structural modifications, changes were made to operation of the copper-silver ionization water disinfection system used by the facility, which required temporarily opening bypass valves. Investigators stated this likely resulted in an influx of sediment into the system. A week after these changes, the first of 13 cases of *Legionella pneumonia* were identified.[8] Similar engineering control failures were highlighted in an outbreak involving *Mycobacterium abscessus*, where whole system contamination was suspected.[7] Mitigations included a programme of routine flushing, removal of aerators, improvements to water recirculation to ensure increased mixing, faster delivery of hot water to outlets and increased chloramine levels throughout the water system. Ineffective heat exchangers were associated with an outbreak of *Legionella pneumophila* where temperature fluctuations combined with flow rates at heat exchangers being well below the maximum designed flow rate of 230 litres per minute (leading to stagnation) preceded the infections.[5] High *Legionella* spp. counts persisted despite hyperchlorination and heat treatment, suggesting a persistent biofilm at the heat exchangers. Inadequate chlorination was associated with a *Legionella* outbreak on a haemato-oncology unit in America; although the total chlorine measured in the water (1.2 parts per million (ppm)) at the point of entry to the system was within acceptable limits, the residual chlorine levels in the cold water at distal outlets dropped to undetectable levels (<0.1 ppm).[2] This is likely to have allowed Legionellae to survive and grow. Inadequate chlorination was also highlighted as a contributing factor in a pseudo-outbreak involving NTM (*M. simiae*), where widespread contamination within the system (pipes, heat exchangers, sinks, drinking fountains, ice machines) was identified.[4]

In some cases, more complex factors related to the design and construction of water systems may contribute to the risk of contamination. Widespread water system contamination at a Scottish hospital was linked to a multi-species outbreak of bloodstream infections.[9, 52, 53] In an outbreak study of a neonatal ICU, a delay between completion of construction work and the commissioning caused water to

remain stagnant for three months and the subsequent biofilm development was the possible cause of the outbreak.[34] Poor commissioning planning is an example of inadequate management of the water system whereby a filled system does not become operational for some time leading to stagnation of water which favours biofilm development. Poor practice around construction and commissioning featured in a number of outbreak studies[8, 34, 52, 79] and is likely an underreported factor considering the age of some healthcare estate.

There is a paucity of evidence that sheds light on the environmental prevalence of these microorganisms in healthcare settings outwith outbreak situations and whether this varies geographically. An environmental surveillance study to determine the presence of *Cupriavidus* spp. in healthcare water systems across the UK found *Cupriavidus* spp. in four out of the 10 tested hospitals as well as a range of gram-negative bacteria in all 10 hospitals.[88] Another surveillance study aiming to investigate the prevalence of NTMs in hospital water supplies in Iran found that 128 (64.6%) of 198 tap water samples, taken from various departments in six hospitals, were positive for NTM species.[107] The most common strains identified were *M. gordonae* (24 isolates), followed by *M. kansasii* (18 isolates), *M. simiae* (18 isolates), *M. fortuitum* (12 isolates), and *M. chelonae* (4 isolates). However, both studies did not report patient infection/colonisation rates at the studied hospitals and thus it is not known if the *Cupriavidus* spp and NTM found in the water supply was posing a clinical risk. Whilst it is likely that there are distinct regional/geographic trends in terms of presence of NTM in public water supplies, it is unclear how geographic and environmental factors influence the prevalence of certain species and the risk of nosocomial NTM infection/colonisation.

The outbreak studies discussed above highlight the complexity of water systems and the myriad factors that can collectively increase (or decrease) the risk of naturally occurring microorganisms proliferating. In two outbreak studies, although widespread contamination of the water system was identified, the contributing factors at the local level were not reported in detail.[3, 109]

Distal outlet/drain contamination

Sixty reports describe microbial proliferation/contamination of the plumbing infrastructure mainly at distal outlets and/or drains. Most of these outbreak studies

(n=31) involved patient colonisation and/or infection with *P. aeruginosa,*[10, 11, 13-16, 19-22, 24, 28, 30, 32-35, 38, 44, 45, 47, 51, 52, 54, 55, 82-86, 90, 91] (two of these also involved *Pseudomonas putida*).[52, 54] Other microorganisms included Enterobacteriaceae, detailed in 22 reports (including *Klebsiella* species (K. *pneumoniae*, *K. oxytoca*), *Enterobacter* species (*E. cloacae*, *E. aerogenes*), *Citrobacter* species (*C. freudii*, *C. koseri*), *E. coli*, *Serratia marcescens, Pantoea agglomerans,* and *Raoultella planticola*).[12, 27, 29, 37, 40-42, 46, 48-50, 56-65, 81] Other gram-negative organisms included *B. cepacia*,[17, 23] *Acinetobacter* species (*A. baumanii*,[18, 36, 39, 66] *A. ursingii*),[52] *Chrysomonas indologenes*,[52] and *S. maltophilia*.[52] NTMs were detailed in 4 reports (*M. mucogenicum*, *M. fortuitum, M. canariasense*, *M. chelonae*, *M. chimaera)*.[25, 26, 31, 96] Two of these 60 outbreak studies were pseudo-outbreaks where positive clinical samples were obtained but in the absence of clinical colonisation or infection in the patient; the microorganisms involved were *P. aeruginosa* and *M. chimaera*.[31, 33]

From the literature, taps were identified as plumbing system reservoirs contaminated with *P. aeruginosa* in 11 reports[10, 11, 24, 30, 35, 44, 54, 83, 85, 90, 91] with NTMs (*M. mucogenicum, M. chimaera*) in 2 reports[25, 31] and *A. baumanii* in two reports.[36, 66] Two of these studies also identified positive samples from shower water.[54, 83] Sensor operated taps were specifically mentioned in four studies.[24, 25, 31, 44] An investigation of a *P. aeruginosa* outbreak on a NICU in Northern Ireland found that sensor taps had significantly greater odds of having at least one component positive for *P. aeruginosa* compared with non-sensor taps ($p < 0.05$).[24] Complex flow straighteners were present in the sensor taps (and not in the non-sensor taps), and it was unclear whether the higher rates of positivity associated with sensor taps were due to the design of the flow straighteners or another factor specific to sensor taps. Flow straighteners within taps were found to be contaminated in five studies.[24, 31, 53, 66, 78] Flow straighteners have the potential to allow build-up of organic debris and provide the structural support for biofilm development.

Showers were implicated in three outbreaks involving NTMs,[26, 96] and *P. aeruginosa*.[83]

Sink drains were reported as environmental reservoirs of Enterobacteriaceae in 18 reports[12, 27, 29, 37, 40-42, 46, 48-50, 58, 60-64, 81] and reservoirs for *P. aeruginosa* in 20 reports[13, 14, 82,15, 16, 20-22, 28, 32-34, 38, 45, 47, 51, 55, 56, 84, 86] Three of the Enterobacteriaceae outbreaks also involved contaminated shower drains,[29, 42, 48]

a fourth had contaminated toilets[37] and another had positive samples identified from patient mattresses.[49] One study involved a water dispenser that was located next to the contaminated sink.[60] Four of the *P. aeruginosa* outbreaks reported biofilm development in the pipework attached to the drain.[14, 16, 28, 32] Shower and tap water,[45] and shower drains and toilets[51] were involved in a few of these *P. aeruginosa* outbreaks. In addition to the 18 reports involving sink drains, shower drains were identified as the reservoir for Enterobacteriaceae in one outbreak,[57] and toilets in another.[59] Additional gram-negative organisms located in sink drains associated with outbreaks included *Burkholderia* species,[17] and *A. baumannii*,[18, 39] *Burkholderia cepacia* was discovered in tap water and on the inside of taps in an outbreak of multiple cases of bloodstream infection.[23]

Design features of water fittings can facilitate transmission of organisms from their environmental reservoirs to external surroundings. Two key sink design features mentioned in literature that can facilitate splashing and/or surface and hand contamination from a contaminated drain are the depth of the sink basin (shallow) and the tap positioning relative to the sink drain (direct water flow onto the drain which creates splashing).[21, 30, 62, 105] These poor design features, coupled with poor clinical practice such as storage, preparation, and handling of items (for example drug preparation) within splash zones of water outlets,[3, 10, 14, 39, 63] are risk factors for dissemination of contamination from outlets and drains.

The reuse of a cleaning brush to clean down all the sink drains in a hospital ICU, without disinfection of the brush between sinks, likely facilitated environmental spread between drains during an outbreak of carbapenem-resistant bacteria in Australia.[62]

An open floor drain was identified as the source of an outbreak of carbapenem-resistant *P. aeruginosa* in a urology ward in Spain;[19] it should be noted that open floor drains are not a common fixture in NHSScotland premises.

One report described infection with the fungus *Fusarium solani* where taps and drains tested positive at a hospital in Brazil. Although water tanks were visually dirty, sampling was not conducted to confirm whether contamination was system wide.[77]

Water-based equipment

Contaminated water-based equipment was identified in the literature as a source for ongoing transmission in 15 reports, this included cardiac water heater coolers,[71, 87, 95] automated endoscope reprocessors,[1, 67, 73] laparoscopy equipment,[74] haemodialysis wall boxes,[69] chilled water dispensers,[43, 68, 70] a tea dispenser,[65] mesotherapy equipment,[72] ice machines,[75] clothes washing machine,[76] and neonatal incubators.[80] In neonatal incubators, steam cleaning as part of cleaning was facilitating contamination of the incubators and mattresses due to residual moisture left after steaming.[80] The CDC also list equipment such as automated endoscope reprocessors and ice machines.[94] Whilst the CDC state that hydrotherapy tanks and pools can pose a risk, the evidence cited for this is all pre-2000 (mainly from the 1980s and 1990s).

In conclusion, whilst it is acknowledged that the source of contamination may be the incoming water supply (many organisms are ubiquitous in the water supply), or the patient, the causes for proliferation are commonly a failure of design and/or management of healthcare water distribution systems and associated components, fittings and equipment (such as lack of temperature, chemical control, flow or usage) that allows incoming organisms to survive and accumulate. This is compounded by multiple factors such as the inadequate design and/or management of water systems, and inadequate cleaning/decontamination protocols. Inappropriate practices and behaviours of healthcare staff, patients and visitors can present a risk of environmental seeding (transfer of patient isolates to the environment) and encourage survival of water system-associated organisms within the water system. Infection incidents involving Enterobacteriaceae and *P. aeruginosa* were predominantly linked to drains and point of use outlets. The potential involvement of patients in seeding the environment, effectively acting as the source, is difficult to determine in outbreak studies but cannot be discounted considering that these organisms are human commensals. Whilst it is challenging to determine the original source of plumbing infrastructure contamination, the evidence demonstrates that these fittings and fixtures can act as environmental reservoirs for ongoing transmission.

A major limitation of this evidence base is that most studies do not report on patient colonisation at admission (possibly because such surveillance is not routine).

Further, environmental testing beyond the tap water was often not conducted during outbreak studies, therefore limiting knowledge as to the extent of wider system contamination. Again, there is a risk of publication bias as studies that did not utilise molecular typing were excluded. The variety of discriminatory power in the different typing methods of included evidence is another limitation.

4. Which patient populations are considered as being at increased risk of colonisation/infection with a healthcare water system-associated organism?

In total, 41 pieces of evidence were identified in relation to this research question which includes 39 outbreak studies,[2, 3, 7-14, 17, 19, 23, 25, 26, 28-31, 37, 47, 50, 51, 53, 55, 58, 60, 61, 63, 64, 69, 70, 76-80, 96, 110] one English guidance document that was deemed expert opinion[111] and one cohort study.[90] In accordance with SIGN50 methodology, 39 were graded SIGN50 level 3 evidence (38 outbreak studies,[2, 3, 7-14, 17, 19, 23, 25, 26, 28-31, 37, 47, 50, 51, 53, 55, 58, 60, 61, 63, 64, 69, 70, 76-80, 96, 110] one cohort study[90]) and one was graded SIGN50 level 4 evidence (expert opinion).[111]

There are some general limitations to the evidence included within this research question. All of the studies are low quality, graded SIGN50 level 3 or level 4. Since the majority of studies (39 out of 41) are outbreak studies, there is a risk of publication bias as not all infection incidents are published in scientific journals. Moreover, outbreak studies are not controlled for and thus can only describe patient populations affected by water-associated incidents/outbreaks but not measure increased risk.

The patient populations most frequently affected by water system-associated incidents and outbreaks, and considered as high-risk patients, include haematology and oncology patients,[2, 7-10, 25, 26, 28, 37, 58, 63, 77-79] cardiac surgery patients,[7, 60, 61] bone marrow and stem cell transplant patients,[3, 7, 31, 51, 70] neonatal,[47, 80, 110] paediatric[17, 76, 77] and adult ICU patients,[12, 13, 50, 55, 60, 64, 90] transplant patients,[7, 30] and any other patients that are severely immunocompromised through disease or treatment, for example burn patients,[11, 13, 29] patients with compromised skin integrity and patients with chronic lung disease.[3, 14, 19] Patients with non-intact skin or indwelling peripheral/central venous catheters may also be at risk.[3, 23, 69, 96] Not only patients

admitted to hospital are at risk, but also patients at outpatient facilities (for example oncology clinics).[63] Clinical judgement is required to assess individual patient risk for any patient not being managed in these high-risk settings.

The approved code of practice (L8) published by the Health and Safety Executive (HSE) discusses patients at higher risk for infection with *Legionella* spp. specifically.[111] These include patients aged over 45 years, those with respiratory disease, chronic kidney disease, diabetes, heart disease, or patients with an impaired immune system.

5. What types of infection can healthcare water system-associated organisms cause?

In total, 22 pieces of evidence were identified in relation to this research question which includes 17 outbreak studies,[1, 3, 4, 7, 11, 23, 25, 26, 31, 33, 63, 67, 69, 73, 78, 79, 96] one cohort study,[90] two international guidelines (both deemed expert opinion[94, 102]) and one surveillance study.[81] In accordance with SIGN 50 methodology, 20 were graded level 3 evidence (17 outbreak studies,[1, 3, 4, 7, 11, 23, 25, 26, 31, 33, 63, 67, 69, 73, 78, 79, 96] one cohort study,[90] and two surveillance studies[81, 87]) and two were graded level 4 evidence (expert opinion).[94, 102]

A general limitation is the low quality of evidence (all 22 studies are graded SIGN50 level 3 or level 4). Due to most studies (17 out of 22) being outbreak studies, there is a risk of publication bias as not all outbreaks/infection incidents are published in scientific journals and thus some (possibly unusual or rare) types of infection could be missed. However, at least two studies were identified for each type of infection described below. Conversely, there is a possibility that rare or unusual types of infections are more likely to be published resulting in an overestimation of their risk.

Within the identified studies, the different types of infection described to be associated with water system organisms include bloodstream,[3] respiratory (pneumonia, ventilator-associated pneumonia), skin and soft tissue (including insertion site infections around any invasive device), surgical site infection (endocarditis, wound infection),[87] urinary tract infection (UTI) and disseminated disease.[7, 11, 23, 25, 26, 63, 69, 78, 79, 81, 90, 96, 102]

Pseudo-outbreaks, where positive clinical samples were identified but in the absence of clinical colonisation/infection, were described in six outbreak studies linking the contamination of patient samples to a contaminated water source.[1, 4, 31, 33, 67, 73] The CDC advises that pseudo-outbreaks of *M. chelonae*, *M. gordonae*, and *M. xenopi* have been associated with both bronchoscopy and gastrointestinal endoscopy when contaminated tap water is used to provide irrigation to the site or to rinse off the viewing tip in situ, or when the instruments are inappropriately reprocessed with tap water in the final steps.[94]

6. What are the incubation periods of healthcare water system-associated organisms?

Incubation period in the context of water-associated infection refers to the period between entry of the organisms into the body and onset of infection and depends on the microorganism involved, type of disease and patient factors.

The available evidence on this topic is very limited. One outbreak study,[71] one surveillance study[87] and two guidance documents[102, 112] categorised as expert opinions were identified in the literature search. In accordance with SIGN50 methodology, two were graded level 3 evidence (one outbreak study[71] and one surveillance study[87]) and two as level 4 evidence (two expert opinions[102, 112]).

For most water system-associated organisms, it is challenging to determine an accurate incubation period and this is in part due to the difficulty in determining the exact exposure event and source, and in confirming a link between patient cases and an environmental source. Exceptions to this are for *Legionella* spp. and *Mycobacterium chimaera.*

For legionellosis, guidance from the World Health Organization (WHO) and the European Centres for Disease Control and Prevention (ECDC) advise an incubation period of 2-10 days which can extend up to 20 days in rare cases.[102, 112] This information is informed by data from community-acquired outbreaks; there is less recent data available for healthcare-associated cases. There is a very low incidence of cases in the healthcare population in Scotland. In fact, there have been no hospital-acquired Legionnaires' disease cases in Scotland in the past ten years.[113]

Two reports specific to cardiopulmonary bypass–associated *M. chimaera* infections indicate an incubation period of between three months and five years for that specific organism and exposure scenario.[71, 87]

In summary, the evidence base regarding incubation period is limited and the precise incubation period for most healthcare water system-associated organisms is varied and unknown. As previously mentioned, incubation periods may be fairly lengthy in some cases. However, very short incubation periods of less than the typically used 48 hours cut off for defining a healthcare associated infection may also be possible (see chapter 3 'Healthcare Infection Incidents, Outbreaks and Data Exceedance' of the National Infection Prevention and Control Manual). It must be noted that patients susceptible to infection with water system-associated organisms can clinically deteriorate rapidly following exposure.

7. What is the period of communicability for healthcare water system-associated organisms?

No evidence was identified to support this research question.

8. What are the known transmission routes of healthcare water system-associated organisms?

In total, 41 studies were identified in relation to this research question which includes 34 outbreak studies,[1, 3, 7, 10, 12, 13, 15, 16, 19, 21-23, 25, 26, 29, 32, 33, 35, 57-60, 63, 67, 68, 70, 71, 76, 79, 82, 86, 96, 114, 115] two surveillance studies[83, 87] and one cohort study.[90] Four guidelines were also included, of which two were Scottish guidelines[116, 117], and all were graded as SIGN50 level 4 expert opinion.[94, 95, 116, 117] In accordance with SIGN50 methodology, 37 were graded level 3 evidence (34 outbreak studies,[1, 3, 7, 10, 12, 13, 15, 16, 19, 21-23, 25, 26, 32, 33, 35, 57-60, 63, 67, 68, 70, 71, 76, 82, 86, 96] two surveillance studies[83, 87] and one cohort study).[90]

Most studies identified in literature investigating the association between infection and water systems are outbreak studies (34 out of 41 studies) which may suggest a possible, indirect association between the water source and disease transmission. However, it remains challenging to confirm the exact mode of transmission. In many instances, an exact transmission mode from an identified environmental source or

reservoir to a patient could not be determined.[58, 82, 83] In such cases, multiple water uses present multiple possible transmission routes.

There is also a risk of publication bias within this research question as not all outbreaks/infection incidents are published in scientific journals and therefore there is the possibility that the evidence may not fully reflect what is being seen in practice. Moreover, the CDC guidelines are mostly based on studies published pre-2000 which is a limitation as it might not reflect current IPC practice.[94]

Under favourable circumstances, water system-associated organisms have the ability to either proliferate in active growth or remain for long periods in highly stable environmentally resistant forms.[94] The different modes of transmission include direct contact, indirect contact, aerosolisation and aspiration.

Direct contact can occur through ingestion of contaminated water,[7, 68, 70] or contact of contaminated water with any other portal of entry (for example surgical site wound, invasive devices, exposed or wounded skin).[3, 7, 23, 25, 26, 29, 79, 96]

Indirect transmission can occur from the hands of healthcare workers after contacting a contaminated reservoir or source. Healthcare workers hands are often hypothesised as a transmission route in outbreak studies in ICUs where patients are bedbound, but this is challenging to confirm.[15, 16, 55, 63, 86, 90, 114] Of seven *P. aeruginosa* strains isolated from HCW hands on a medical ICU, the genotype was the same as that from the last patient touched by the HCWs in six cases, and in the seventh was the same as the last tap water sample used.[86] Even where samples are taken from HCW hands, they may test positive, even when poor hand hygiene is observed.[63] At an Italian haematology unit, a contaminated soap dispenser (soap type 'Triclosan') was the source for onwards transmission via HCW hands.[114]

Indirect transmission can also occur when contaminated equipment comes into contact with patients. Examples of contaminated equipment include diagnostic equipment (bronchoscopes,[33, 67] bronchoscope automatic washing machine[1, 76]), medicine prep trays,[10] surgical equipment (arthroscope),[22] ventilator equipment (suction apparatus),[35] breast pump equipment,[32] surgical drape that was re-used despite being single-use,[19] and hydrotherapy shower mattresses.[13] For example, contamination of infusion therapy procedure trays used to carry intravenous preparations to patient rooms was identified in an outbreak study as the link between

contaminated taps and infected patients, with Hickman lines providing an entry route for organisms.[10]

Aerosolisation can occur through contaminated water generated from the process of water splashing or spraying onto or from clinical wash hand basins, drains, sinks, shower cubicles and when flushing toilets.[16, 21, 57, 59, 60, 63, 94] Aerosols can also be released from contaminated water-based equipment for example cardiopulmonary bypass machines and heater-cooler units used during cardiac surgery,[7, 71, 87, 95, 116], humidifiers within mechanical ventilators,[32, 94] as well as room air humidifiers.[94]

Finally, the aspiration of contaminated water can be another mode of transmission. Patients with nasogastric tubes and those requiring oral fluid replacement (both often intubated patients) may inhale contaminated water into the airways.[115] The 2003 CDC guidelines (expert opinion) state that aspiration is a transmission mode, however the references provided are all in relation to *Legionella* spp. and all published before the year 2000.[94]

In summary, the various transmission routes can be classed in four groups: direct contact, indirect contact (including via contaminated personnel and patients, environment, equipment, and medical products), aerosolization, and aspiration.

9. Which healthcare procedures present an increased risk of transmission of healthcare water system-associated organisms?

In total, 20 studies were identified in relation to this research question which includes 14 outbreak studies,[3, 7, 10, 13, 23, 25, 26, 33, 57, 67, 69, 71, 79, 96] two Scottish guidance documents (expert opinion),[116, 117] one English guidance document (expert opinion),[95] one international guideline (expert opinion),[94] one cohort study[90] and one surveillance study.[87] In accordance with SIGN50 methodology, 16 were graded level 3 evidence (14 outbreak studies,[3, 7, 10, 13, 23, 25, 26, 33, 67, 69, 71, 79, 96] one cohort study[90] and one surveillance study[87]) and four were graded level 4 evidence (four expert opinions[94, 95, 116, 117]).

A general limitation is the low quality of evidence (all 20 studies are graded SIGN50 level 3 or level 4). Due to the large number of studies (13 out of 20) being outbreak studies, there is a possibility of publication bias as not all outbreaks/infection incidents are published in scientific journals and thus the risk of transmission of

organisms following some healthcare procedures might be underestimated. Moreover, the international guidelines included (CDC) are limited as they are mostly based on studies published pre-2000 and therefore might not reflect current IPC practices and the associated risks.[94]

In theory, all diagnostic, treatment, and patient care procedures that involve a water source may present a risk of transmission of healthcare water system-associated organisms. Uses of water in procedures have been traced back as the potential transmission route to the patient in the literature, as follows: two outbreak reports,[33, 67] one international guideline published by the CDC[94] (SIGN50 level 4) and one other expert opinion guidance[117] provide evidence that reusable medical equipment (bronchoscopy, endoscopy) present a risk due to poor disinfection and inappropriate reprocessing of instruments with tap water.

Patient hygiene (bathing and washing) including wound care was evidenced in three outbreak reports and one cohort study.[3, 7, 57, 90] Involvement of CVCs via submersion in water was evidenced in three outbreak reports[25, 26, 96] and in CDC guidelines (SIGN50 level 4). Procedures involving CVC care including haemodialysis[10, 23] was described in two outbreak reports. Two outbreak studies conducted case-control assessments which demonstrated CVCs to be significant risk factors for infection.[69, 79] Hydrotherapy was evidenced in one outbreak report[13] and CDC guidelines (SIGN50 level 4).

Oral care and enteral tube flushes was evidenced in one outbreak report.[7]

Use of cardiac heater cooler units during surgery was evidenced in four outbreak reports and one surveillance study and two guidance documents (both SIGN50 level 4).[7, 71, 87, 95, 116]

In summary, the evidence was consistent in demonstrating that any diagnostic, treatment or patient care procedure that involves a water source (for example oral care, washing/bathing, enteral tube flushes, intravenous procedures including management, hydrotherapy, use of cardiac heater coolers during surgery) may present a risk of transmission.

3.1.2 Prevention and control of healthcare water system-associated infection:

10. What are the microbiological water testing requirements at commissioning?

In total, six pieces of evidence were identified to answer this research question which includes two guidance documents published by the British Standards Institution[118, 119] (including one code of practice[118]), two Scottish guidance documents[120, 121] (part of the Health Facilities Scotland Scottish Health Technical Memorandum (SHTM) 04-01 series on water safety), one British guidance document[122] (part of the Department of Health, Health Technical Memorandum (HTM) 04-01 series on water safety) and one Scottish incident report.[52] All six pieces were deemed to be expert opinions due to the lack of a rigorous methodology or evidence base in guidance development. In accordance with SIGN50 methodology, these six expert opinions are graded level 4 evidence.[52, 118-122] The small volume of evidence and the lack of high quality evidence is a limiting factor and makes it challenging to answer this research question based on the published scientific articles and guidance documents alone.

Water samples should be obtained as standard UK practice at water system commissioning to ensure a safe handover of a newly constructed or refurbished water system from the contractor.[52, 118, 120-122] However, there is no mention of specific microbiological water testing requirements at commissioning in the literature. It is mentioned in SHTM 04-01 (Water safety for healthcare premises, Part A) that after disinfection (which is also part of the pre-commissioning process), microbiological tests for bacteria colony counts at 37°C and coliform bacteria, including *Escherichia coli*, should be carried out to confirm that the water is of potable quality.[120]

Scottish guidelines SHTM 04-01 (Water safety for healthcare premises, Part A) and the British Standard BS 7592:2022 state that the NHS board water safety group (WSG) should agree a sampling regime and appropriate parameters prior to tender, including microbiological, depending on the intended use of the system and vulnerability of the patients.[118, 122] These guidance recommend that to confirm

effective disinfection, samples should be taken no sooner than 48 hours after disinfection and immediately prior to handover.[118, 122] The British Standards Institution PD 855468:2015 extends this period, stating that samples should be taken between two and seven days after disinfection to avoid false negative results.[119] In the case of *Legionella*, Scottish guidance indicates that a period of three days – and preferably five – should be allowed following disinfection for the system to settle prior to sampling.[121] It is recommended that sampling is undertaken by an accredited organisation independent of the contractor.[122]

Due to the lack of evidence identified here, more research and inclusion of commissioning in current guidance or the development of new guidance is needed especially regarding specific microbiological water testing requirements.

11. What are the responsibilities of the IPC team in regards to water safety at commissioning?

In total, six pieces of evidence were identified in relation to this research question which includes three Scottish guidance documents,[101, 123, 124] two British Standards[125, 126] and one Scottish incident report.[52] All six pieces were deemed to be expert opinions due to the lack of a rigorous evidence base underpinning and/or methodology in developing the guidance. In accordance with SIGN 50 methodology, these six expert opinions are graded level 4 evidence.[52, 101, 123-126] The lack of high quality evidence is a limiting factor for answering this research question.

Throughout the planning and implementation process, ongoing input from a multidisciplinary team, which includes IPC professionals, is required to ensure that infection control remains at the forefront of the design, planning, construction refurbishment and maintenance of healthcare facilities.[123] The Healthcare Associated Infection System (for) Controlling Risk In the Built Environment (HAI-SCRIBE) procedure has been developed to identify, manage and record built environment infection control risks.[124] It is a requirement for IPC teams to have involvement with the HAI-SCRIBE process.[124]

The SHTM 04-01 Water safety for healthcare premises, Part A, states that the water system should not be brought into service until the infection control team certifies that the water is of potable quality.[120] The SHTM 04-01 (Water safety for healthcare

premises, Part B) includes the responsibilities for the WSG (which includes the Infection Control Manager, the Infection Prevention and Control Doctor and the Consultant Microbiologist) for the maintenance of water quality from the point it leaves the tap.[101] They should also advise on infection control policy and this policy should be agreed with the Infection Prevention and Control Team.

IPC teams should be represented in WSGs within NHS boards who commission and develop a Water Safety Plan (WSP) as outlined in SHTM 04-01 and BS 8680 which includes a risk assessment and actions to mitigate risks.[101, 123, 125] The British Standard (BS 8580-2:2022) also mentions the input of IPC teams during the development of a risk assessment to identify the types and location of healthcare water system associated infections which could be linked to water exposure and for assessment of surveillance practices.[126]

In general, there is a lack of detail in extant guidance regarding the specific roles and responsibilities of the IPC team at commissioning, beyond certifying that the water is of potable quality.[120] There is consensus that a multidisciplinary team, which includes IPC professionals, should have ongoing input during commissioning.

12. Is routine water testing to detect healthcare water system-associated organisms recommended?

In total, 15 pieces of evidence were identified in relation to this research question which includes three guidance documents published by the British Standards Institution,[105, 118, 126] two Scottish Heath Technical Memorandums,[101, 127] six other guidance documents that were classed as expert opinion (including two derived from Scotland, three from England, one from the Republic of Ireland, one CDC guidance document and one WHO guidance document)[94, 102, 116, 117, 128-131] one outbreak study[53], Healthcare Infection Society (HIS) Working Party guidelines.[94, 132] In accordance with SIGN 50 methodology, one was graded level 3 evidence (one outbreak study)[53] and 12 were graded level 4 evidence (nine expert opinions)[101, 102, 105, 118, 126-130] The HIS guidelines included in this section were graded using the AGREE tool as ‘Recommend’.[132]

The low quality of evidence is a general limitation of the included evidence (out of the 15 studies included, 13 are level 4 and one is level 3). Most of the included guidance

documents are classed as expert opinion due to their limited methodology and/or lack of a rigorous search of evidence. The CDC guidelines included are mostly based on studies published pre-2000 and therefore might not reflect current IPC practices and the associated risks.[94]

Routine water testing is currently recommended for *P. aeruginosa* in the UK and the Republic of Ireland in augmented care areas only (for example NNUs and ICUs).[128, 130] This is because little reassurance can be gained from one-off samples which can give rise to false negative results, and may miss other contaminated water outlets. Since the clinical risks of severe disease are higher in augmented care, the benefits of routine water testing outweigh the disadvantages.

The CDC mentions that environmental surveillance involving periodic culturing of water samples for *Legionella* spp. from the hospital's potable water system can be an advantage as this is less costly than routine lab diagnostic testing for all patients who have healthcare associated pneumonia.[94] Routine sampling for the presence of *Legionella* is currently only recommended in England and Scotland when temperatures are not consistently attained or control methods other than heat are used, or where it is found to be necessary by the risk assessment (for example in high risk units and/or procedures).[101, 118, 129, 130] The World Health Organization (WHO) also recommends testing for *Legionella* in wards with high-risk patients.[102] The need for routine water testing other than *Legionella* and *P. aeruginosa* should also be based on risk assessment. Guidance on risk assessments relevant to water can be found in BS 8580-1 regarding *Legionella* and BS 8580-2 regarding *P. aeruginosa* and other healthcare water system-associated pathogens.[105, 126]

Total Viable Counts (TVC) can provide evidence on general background microorganism levels and a number of guidance documents agree that routine TVC testing can be useful to indicate deteriorating water quality.[101, 105, 127, 130] Any significant changes in monitored levels can provide an early warning of possible emerging problems. The Department of Health, Health Technical Memorandum (HTM 04-01) only recommend monitoring of TVC levels in case of taste or odour problems and when doing so, it may be used to analyse trends.[130] Although not currently mandated in Scottish guidance, the SHTM 04-01 mentions that many estates management staff continue to test for TVCs and this can be especially important in high risk units and/or where high risk procedures are undertaken.[101, 127]

As an example, regular and consistent water testing (surveillance) resulted in timely recognition of elevated TVC levels in a Scottish outbreak study which successfully minimised the clinical impact of the outbreak.[53] However, no further studies were identified in literature that routinely tested for TVC levels. Further research, for example a pilot study in an NHSScotland healthcare facility, would be valuable to create a baseline for TVC testing.

Additional routine testing of water used in patient care procedures that use water separately from the main hot and cold water distribution system (for example HCUs, endoscopy, renal dialysis) is beneficial. Scottish guidance specific to cardiac HCUs recommended fortnightly water testing as HCUs are a known reservoir of healthcare water system-associated organisms.[116] Scottish guidance specific to the management of endoscopy rinse water recommend weekly testing of endoscopy final rinse water as described in SHTM 2030.[117] The Healthcare Infection Society (HIS) Working Party have produced detailed guidelines on final rinse water where they recommend that TVC monitoring should be done weekly and testing for the presence of environmental mycobacteria and *P. aeruginosa* quarterly, which is in line with guidance from Public Health England.[131, 132] Healthcare facilities are advised by the CDC guidelines and guidance documents from England and Republic of Ireland to sample dialysis fluids monthly as well as product water used to prepare dialysate using standard microbiologic assay methods for healthcare water system-associated microorganisms.[94, 128, 131] Additionally, English and Republic of Ireland guidance recommend to sample hydrotherapy pool water weekly for *E.coli,* coliform bacteria, *P. aeruginosa* and English guidance adds quarterly testing for *Legionella* spp.[128, 131]

Republic of Ireland guidelines state that water provided by water dispensers, water coolers or filtered water unit taps and associated pipe work, which are commonly made of plastic and particularly prone to microbial biofilm contamination, should be subject to periodic microbiological testing to ensure good water quality.[128]

It is important to note that the detection of low TVCs is an indication of the overall water quality and signifies a generally unfavourable environment for bacteria, but is not necessarily an indication of the absence of water system-associated organisms (including *Legionella* spp.).[101, 105] Therefore, these results need to be interpreted carefully when they are being considered in a risk assessment.[105]

To summarise, routine water testing is recommended for *P. aeruginosa* and *Legionella* spp. in high-risk areas (for example NNUs and ICUs) to protect vulnerable patients. Guidance on risk assessments to determine if routine water testing is needed for organisms other than *P. aeruginosa* and *Legionella* spp., can be found in BS 8580-1 (*Legionella* spp.) and BS8580-2 (*P. aeruginosa* and other water healthcare water system-associated pathogens). For organisms other than *P. aeruginosa* and *Legionella* spp. it can be valuable to monitor TVC levels and analyse trends as changes in TVC levels can highlight potential issues with the water system. Routine testing of water that is separate from the main hot and cold water distribution system is recommended for HCUs, endoscopy rinse water, water used for renal dialysis and hydrotherapy pool water.

13. What are the recommended microbiological limits for healthcare water system-associated organisms?

In total, 19 pieces of evidence were identified in relation to this research question which includes seven Scottish guidance documents,[101, 104, 116, 117, 120, 127, 133] three English guidance documents,[129, 131, 134] four British Standards,[105, 119, 126, 135] one mandatory Scottish legislation,[136] one Republic of Ireland guidance document,[128] one international guidance document (WHO),[102] one UK guidelines[132] and one outbreak study.[53] In accordance with SIGN 50 methodology, one was graded level 3 evidence (one outbreak study[53]) and 16 were graded level 4 (16 expert opinions[101, 102, 104, 105, 116, 117, 119, 120, 126-129, 131, 134, 135]). The UK guidelines included in this section was graded using the AGREE tool as ‘Recommend’.[132]

Most of the included guidance documents (16 out of the 17) are classed as expert opinion due to their limited methodology and/or lack of a rigorous underlying evidence base. Moreover, the fact that 17 out of the 18 included studies are low quality (one is graded SIGN50 level 3 and 15 SIGN50 level 4) is a limitation of the evidence base for this question.

Hot and cold water systems

Twelve sources provide evidence regarding limits for hot and cold water systems.[53, 101, 102, 119, 120, 127-129, 131, 134-136] A water supply is not expected to be entirely free from healthcare water system associated organisms, but systems should be in place to

avoid favourable conditions for microbial growth.[120] TVC testing results can indicate when the system itself is deteriorating or where there is a local contamination issue.[101, 120, 135] Consequently, the microbiological limits are not defined for the hot and cold water system and monitoring over time is recommended by five evidence sources to capture any significant increase in TVC counts.[119, 127, 135, 136] The British Standards Institution mention an increase in microbiological counts in their published document (PD855468:2015) as TVC results in excess of a 2 log difference (100 times) above that found in incoming water.[119]

Elevated TVCs following frequent water testing from various points within the water system and the clinical isolation of a rare pathogen *Cupriavidus pauculus* in a sterile aseptic pharmacy unit prompted an outbreak investigation. The unit had prior agreed TVC cut-off levels of <10 cfu/ml at 37°C and, <100 cfu/ml at 22°C, but on several occasions the TVCs were reported over 300 cfu/ml which the authors interpreted as being potentially associated with the incident.[53]

In general, the mandatory Scottish Water Regulations 2014 states that water (for general use, not specific to health and care settings) must have 0 cfu/100 ml for Enterococci and Coliform bacteria (including *Escherichia coli*) to pass as potable, and must have 'no abnormal change' for TVCs in 1ml water sample at 22°C and 37°C.[136] This is also indicated in English and Republic of Ireland guidelines where microbiological limits of potable water are specified as 0 cfu/100 ml for Enterococci and Coliform bacteria (including *E. coli*) and no upper limit for other bacterial species except *P. aeruginosa.*[128, 131] For *P. aeruginosa* in all areas, the recognised microbiological limit is 0 cfu/100ml.[128, 131]

Specifically, the microbiological limits for *Legionella* spp. in the healthcare facilities' hot and cold water system are consistent in Scottish and English guidance to be no greater than 100 cfu/litre.[101, 104, 127, 129, 131, 134] It is recommended by the WHO and English guidelines, that in high-risk areas such as transplant units and ICUs, water from the outlet should be free of *Legionella* spp. (that is 0 cfu/litre water) in order to protect susceptible patients.[102, 129, 134]

High-risk procedures

In addition to water systems, UK and Republic of Ireland guidance documents recommend specific microbiological limits for healthcare procedures that present an

increased risk for contamination with healthcare water system-associated organisms (for instance HCU water, hydrotherapy water, endoscopy final rinse water, final rinse water in surgical instrument washer disinfectors and renal dialysis fluid/water).[116, 117, 128, 131-133] These limits include endotoxin levels to measure the presence of gram-negative bacteria (<0.25 EU/ml in endoscopy and surgical instrument washer disinfectors final rinse water and <0.125 EU/ml in renal dialysis fluid/water) and TVC cut-off levels (<100 cfu/100 ml in HCU waters, <50 cfu/ml in renal dialysis fluid/water, <10 cfu/ml in hydrotherapy water, <10 cfu/100 ml in endoscopy final rinse water and <1 cfu/100 ml in final rinse water in surgical instrument washer disinfectors).[116, 117, 128, 131-133] For HCU water and endoscopy final rinse water, a limit of 0 cfu/100 ml *Mycobacterium* spp. is also recommended.[116, 128, 131, 132] Specific to hydrotherapy water, a limit of <20 cfu/litre for *Legionella* spp. is recommended by English guidance (PHE) and 0 cfu/100 ml for *Staphylococcus aureus*, only as part of wider investigations, which is in agreement with the Republic of Ireland HPSC guidance .[128, 131]

To summarise, extant guidance provides the following recommended microbiological limits for all water system testing in healthcare facilities:

- Coliform bacteria (incl. *Escherichia coli*): 0 cfu/100 ml
- Enterococci: 0 cfu/100 ml
- *P. aeruginosa*: 0 cfu/100 ml
- *Legionella* spp.: <100 cfu/litre in non-high risk units and 0 cfu/litre in high risk units and procedures

Additional microbiological limits are recommended for healthcare procedures that present an increased risk:

- Heater cooler unit water
    - 0 cfu/100 ml *Mycobacterium* spp. [116, 131]
    - TVC cut-off levels of <100 cfu/100 ml [116]
- Hydrotherapy water
    - <20 cfu/litre for *Legionella* spp. [131]

  - 0 cfu/100 ml for *Staphylococcus aureus* as part of wider investigations only (local decision) [128, 131]
  - TVC cut-off levels of <10 cfu/ml [128, 131]

- Endoscopy final rinse water
  - 0 cfu/100ml for *Mycobacterium* spp.[128, 131, 132]
  - TVC cut-off levels of <10 cfu/100 ml [117, 132]
  - Endotoxin limit of <0.25 EU/ml [128, 133]
- Final rinse water in surgical instrument washer disinfectors
  - TVC cut-off levels of <1 cfu/100 ml [131]
  - Endotoxin limit of <0.25 EU/ml [131]
- Renal dialysis fluid and water
  - TVC cut-off levels of <50 cfu/ml [128, 131]
  - Endotoxin limit of <0.125 EU/ml [128, 131]

Two British Standards advise that determination of acceptable limits for other locations (beyond augmented care/high-risk units) in the healthcare facility should be based on a risk assessment.[105, 126] In general, there is a paucity of published literature to inform risk assessment requirements..

14. How frequently should routine water testing be conducted?

In total, 10 pieces of evidence were identified in relation to this research question which includes eight guidance documents that were deemed to be expert opinion (of which three were English[130, 131, 134]),[102, 128, 131, 134, 137] three British Standards[118, 119, 126] and one before-after study.[91] In accordance with SIGN50 methodology, one was graded level 3 evidence (one before and after study[91]) and nine were graded level 4 evidence (nine expert opinions[102, 118, 119, 126, 128, 130, 131, 134, 137]).

All of the included guidance documents (nine in total) are classed as expert opinion due to their limited methodology and/or lack of a rigorous underlying evidence base. Moreover, no high quality evidence is identified (out of the 10 studies, nine are level 4 and one is level 3) which is a limitation of the evidence base for this question.

Most guidance on the frequency of microbiological water testing, including for *P. aeruginosa* and *Legionella* spp., do not define a specific time frame, but recommend that it should be based on a comprehensive risk assessment and in agreement with the WSG.[118, 126, 128, 134] The WHO mentions that the frequency of testing for *Legionella* spp. depends on the status of the water system (for example variation in biocide treatment, storage or distribution temperatures).[102]

English guidance recommends testing water outlets at least every six months for *P. aeruginosa*; however, these recommendations are based on expert opinion and do not have scientific studies referenced.[130, 131] Six-monthly testing may be insufficient particularly in settings where contamination of tap outlets has been found.[91]

To determine the frequency of water testing, the growth rate of the organism needs to be considered.[119] It is also important to have a sufficient frequency for trend analysis to establish evidence-based confidence that control measures remain effective.[126] Guidance from the WHO adds that the frequency also depends on the source of the water supply.[137] Once the frequency is established, British Standards guidance advises that it should be reviewed by the WSG based on sample findings.[126]

15. When should routine water testing frequency be increased?

In total, eight pieces of evidence on this subject were identified which includes two Scottish guidance documents,[101, 127] five other guidance documents[102, 118, 128-130] and one outbreak study.[8] All seven guidance documents were deemed to be expert opinions due to the lack of a rigorous search and/or methodology in developing the guidance. In accordance with SIGN50 methodology, one was graded level 3 evidence (one outbreak study[8]) and seven were graded level 4 evidence (seven expert opinions[101, 102, 118, 127-130]). There was no primary scientific evidence identified to inform this research question.

The Scottish Health Technical Memorandum 04-01 recommends increasing the water sampling to a weekly frequency when a water system is brought back into use after a *Legionella* contamination has been resolved to closely monitor the water quality.[127] It mentions that where the results of three consecutive weekly water

system samples remain below 100 cfu/litre in non-high risk units, those identified by the WSG (for example the Authorised Person (Water), Consultant Microbiologist, authorising engineer for water) would be informed and sampling would revert to a monthly sampling frequency.[127] Once three consecutive monthly samples remain below 100 cfu/litre, the sampling can be reverted to quarterly sampling (every three months).[127]

A recent outbreak study presented implementation of changes to the water treatment strategy as the cause of a *Legionella* spp. outbreak and therefore recommended that microbiological levels should be assessed after implementing changes to the water system and/or its treatment strategy.[8]

UK guidance (including relevant British Standards), Republic of Ireland guidance and the WHO cover more situations when the routine water testing should be increased. These situations are summarised below:

- after implementing changes to the water system and/or its treatment strategy (for example contamination has been resolved and system is brought back into use)[8, 127, 130]
- during a suspected or confirmed outbreak or if surveillance identifies an increased incidence of infection[101, 118, 128]
- pre-flush trend analysis demonstrates increasing cfu/100 ml for *P. aeruginosa*[130]
- when control levels of the treatment regime (for example temperature or disinfectant concentrations) are not consistently achieved[101, 102, 129]

16. Where should routine water samples be taken from (which outlets, how many samples)?

In total, 10 pieces of evidence were identified in relation to this research question which includes four Scottish guidance documents,[101, 120, 127, 138] three documents published by the British Standards Institution,[118, 119, 135] two other English guidance documents[131, 134] and one Republic of Ireland guidance document.[128] All 10 pieces of evidence were deemed to be expert opinions due to the lack of a rigorous search and/or methodology in developing the guidance. In accordance with SIGN 50

methodology, these 10 expert opinions are graded level 4 evidence. [101, 118-120, 127, 128, 131, 134, 135, 138] The lack of high quality evidence is a general limitation for this research question.

The British Standards Institute's BS 7592:2022 on *Legionella* sampling, which is considered best practice, advises that on new sites or where there is no established sampling plan, a survey of each water system is necessary, together with preparation of an up-to-date schematic diagram of each system.[118] The results of this survey and any other available data should be used to develop a sampling plan and identify the sampling points, keeping in mind the risk factors within the water system for the growth of Legionellae (for instance temperatures between 20°C – 50°C, infrequent use, materials of construction and so on.).[118] The identified sampling points should be noted on a simple room plan or site schematic to enable later resampling.[118] BS 8554:2015 Code of Practice for the sampling and monitoring of hot and cold water services in buildings states that a sampling plan should also take into account the time water is resident in the building, residual disinfectant decay, storage capacity and residence time/water age, the effects of temperature, and other relevant factors likely to have an effect on water quality such as condition of components, which is in agreement with English guidance.[131, 135] The sampling plan should identify equipment that constitutes a significant risk of infection because it produces an aqueous aerosol (see "Which healthcare procedures present an increased risk of transmission of healthcare water system-associated organisms?"), and areas where there is the potential for ingress, stagnation and biofilm build-up.[135] The plan should include fixed sampling points that can be included in a long-term sampling plan (for instance routine testing during operational management) to enable trend analysis.[135]

The Republic of Ireland HPSC provides guidance regarding routine sampling for *Legionella* spp. and recommends that the number and types of sites that should be tested to detect *Legionella* spp. must be determined on an individual system basis because of the diversity of plumbing, heating, ventilation and air-conditioning systems in the various institutions that may be sampled.[128] Scottish and English guidance state that water samples should be taken from selected areas within the distribution system and this selection should be on the basis of risk assessments relating to system configuration or patient susceptibility.[120, 127, 134] The Scottish Health

Technical Memorandum 04-01 mentions that the following samples should be taken as a minimum:[101]

- from the cold water storage and the furthermost outlet from the tank, on every loop
- from the calorifier flow, or the closest tap to the calorifier, and the furthermost tap on the hot water service circulating system
- additional samples should be taken from the base of the calorifier where drain valves have been fitted
- additional random samples may also be considered appropriate where systems are known to be susceptible to colonisation

Examples of recommended sample sites for routine water testing are given in numerous UK guidance documents (including British Standards and Scottish guidance) and these are summarised in Table 1:

Table 1: Example sample sites for routine water sampling.[101, 118, 127, 128, 131, 135]

System	Sample points
Cold water system	• Incoming main, close to meter, where facilities exist to do so • Storage tank (inlet and outlet) • Nearest, mid-point and furthest outlet from the storage tank, on every loop • Other outlets in areas considered to represent a particular risk for example hospital waters with ‘at risk’ patients
Hot water system	• Calorifier outlet (or header cistern) or nearest tap to the calorifier outlet • Return supply or nearest outlet to the return supply • Base of calorifier where drain valves have been fitted • Furthest outlet from the calorifier (for instance sentinel outlet) • Return to calorifier • Other outlets in areas (for instance taps and showers) considered to represent a particular risk e.g. hospital wards with ‘at-risk’ patients

System	Sample points
Other systems/sources	• Water supply points with removable hoses and devices • Hydrotherapy pool water (Furthest point away from inlets/outlets and balance tank) • Renal unit waters and dialysis fluids (end of distribution loop or the last machine in a dead-end system) • Evaporative cooling systems • Additional random samples may also be considered appropriate where systems are known to be susceptible to colonisation

English guidance further expands on the requirement to include samples from sites in the hot water system that are coolest and sites in the cold water system that are likely to be warmest (thus representing sites most likely to support the growth of *Legionella* spp.).[131] Of note, this guidance and the British Standard BS 7592:2022 also mention that the first water delivered from the outlet (meaning pre-flush) should be used for routine monitoring.[118, 131]

There is limited evidence to determine the appropriate number of samples that should be taken. PD 855468:2015 recommends that samples should be sufficient in number to be fully representative of the distribution system, sub-branches, tanks and cisterns, as well as the condition to be evaluated for example efficacy of distribution of disinfectant.[119] Examples of sampling frequencies for distribution networks are provided in PD 855468:2015, where samples would be taken from each branch and at suitable intervals along the run of pipes.[119]

Republic of Ireland *Legionella* guidance references Dutch guidance which lists a recommended number of outlets to sample that is dependent on the number of outlets the building has.[128] The numbers represent 0.75% to 4% of total outlets.
A review of a Scottish hospital water testing commented that 5% of the total outlets in the hospital were sampled at commissioning.[138] There is no evidence in the literature to determine whether these percentages are sufficient or not.

Available guidance on how to take samples is provided in SHTM 04-01 - Part C, BS ISO 5667-24:2016, and BS 8680:2020.

To summarise, samples should be taken from the proximal and distal ends of each water system with a number of sampling points in between (examples of sampling points are provided in Table 1) ensuring that areas identified as 'high risk' both in terms of supporting microorganism growth (for instance cooler parts of the hot water system, warmer parts of the cold water system) and patient susceptibility (for instance high risk units), are represented. Sample points should be noted in a sampling plan to enable resampling and trend analysis. The number of samples that should be taken depend on the number of outlets of the building, but in general this number should be sufficient to represent the entire water system.

17. When should water samples from further back in the system be taken?

Very limited evidence was found in relation to this research question. In total, only one outbreak study was identified to be relevant.[14] In accordance with SIGN50 methodology, this outbreak study was graded level 3 evidence.[14]

The lack of evidence is a limiting factor and makes it challenging to answer this research question. Moreover, the outbreak study is specific to *P. aeruginosa* and thus their findings and conclusions may not be generalisable to other organisms.

When positive tests reoccur after removal of organisms from an initial reservoir, for example a tap, it could indicate a reservoir or source further down (distal) the pipes.[14] Negative pre-flush and positive post-flush samples could also indicate contamination further back in the water system. In these situations, it might be beneficial to take water samples further back in the system.

18. Who should water test results be reported to?

There is limited evidence to inform recommendations regarding the reporting of water sample test results, especially who to report to. In total, seven pieces of evidence were identified which includes three Scottish guidance documents,[101, 121, 127] two British Standards[125, 135] and two English guidance documents.[129, 134] All seven pieces of evidence were deemed to be expert opinions due to the lack of a rigorous search and/or methodology in developing the guidance. In accordance with SIGN50

methodology, these seven expert opinions were graded level 4 evidence. [101, 121, 125, 127, 129, 134, 135] The small amount of evidence and lack of high quality evidence is a limiting factor and makes it challenging to answer this research question.

UK code of practice recommend that test results need to be recorded (either written or electronic) and reported in order to assist those interpreting the results (IPC Team as discussed in "How should routine water test results be interpreted"). [125, 129, 135] However, in these documents/guidance it is not mentioned who it should be reported to. Scottish and English guidance on *Legionella* mention that the local WSG, which includes the Authorised Person (water), an infection control doctor/nurse and consultant medical microbiologist, must be informed and provided with copies of the samples in writing when water test results come back positive. [127, 134] The water safety plan WSP includes pre-determined actions to mitigate risks including when non-compliant results occur as outlined in SHTM 04-01 and BS 8680 and thus it would be appropriate to document the procedure for reporting test results (including who and how) in this plan. [101, 125]

During commissioning, it is recommended by SHTM 04-01 that the contractor supplies a full set of the water sample analysis to the site supervisor for approval before the system is put into use. [121] The water sample analysis should be cascaded by the site supervisor to other relevant stakeholders such as the WSG members.

19. How should routine water test results be interpreted?

In total, eight pieces of evidence were identified in relation to this research question which includes three British Standards, [118, 125, 135] three English guidance documents, [119, 129, 131] one guidance from the Republic of Ireland [128] and one guidance published by the WHO. [102] All eight pieces of evidence were deemed to be expert opinions due to the lack of a rigorous search and/or methodology in developing the guidance. In accordance with SIGN 50 methodology, these eight expert opinions were graded level 4 evidence. [102, 118, 119, 125, 128, 129, 131, 135]

As the growth and survival of organisms are dependent on a number of non-microbiological environmental factors, results should be interpreted alongside other quantifiable factors (known at the time of sampling) that relate to the water system, that is water temperature, pH, residual disinfectant, water softeners, water

turnover.[125, 135] These details should be recorded at the time of sampling to aide interpretation of results and reviewed along with the system's schematic diagram.[102, 125, 135] In this way, the current results can be interpreted with previous recorded results to allow trends to be visible over time. Further to this, the HSE advises that monitoring of hot and cold water systems where thermostatic mixer valves (TMVs) are fitted needs careful consideration to ensure that results are interpreted in the context of the conditions in place at the time of sampling.[129]

The British Standards Institution has recently updated their guidance on *Legionella* sampling (BS 7592:2022) to only recommend pre-flush samples as part of routine monitoring purposes.[118] Pre-flush samples taken with no at-tap disinfection or adjustment of devices or inserts obtain a reflection of the water as it is used.[118] Furthermore, when a sample of water is taken for analysis, results only reflect the quality of the sampled water and not the whole body of water. Therefore there are limitations in the conclusions that can be drawn from any single sample; multiple samples are required to provide confidence in the interpretation of the condition of the system as a whole.[135] This is particularly the case for microbiological samples where contamination can be intermittent.[135]

If contamination is detected in the pre-flush samples, differentiation between local and systemic colonisation can be achieved by collecting post-flush samples and comparing bacterial counts between pre-flush and post-flush samples.[118, 128] When a pre-flush sample is positive, but a post-flush sample is negative or has a significantly lower bacterial count, this may indicate a local colonisation of an outlet and/or associated pipework and fittings near to that outlet.[118, 128] Positive post-flush samples may be an indication that the whole water system is contaminated.[118, 128]

The British Standards Institution mention that for TVC sampling, results in excess of a 2 log difference above that found in incoming mains water may indicate that the system has deteriorated.[119] BS 8554:2015 further states that a significant increase in TVC counts could indicate failing disinfection and/or the establishment of biofilms which could in turn lead to the colonisation by other previously suppressed organisms.[135]

Regarding *Legionella* spp. testing, there are challenges in interpreting results owing to the recognised limitations associated with test methods (poor recovery of

L. pneumophila due to for example residual disinfectant, heat treatment to repress growth of other non-*Legionella* bacteria and addition of antibiotics to culture medium).[102] Consequently, the HSE advises that a negative result is no guarantee that a system is free of *Legionella* spp.[129] Additionally, no direct relationship has been established between the number of *Legionella* bacteria detected in a water system and the risk of infection, which further limits interpretation of results.[102] The HSE also advises that a positive *Legionella* test result may not always indicate a failure of controls, as *Legionella* spp. are present in almost all natural water sources.[129]

English and UK guidance recommend that advice on the interpretation of results should be sought from a microbiologist with experience of the healthcare environment and that water test results should be interpreted by a competent person.[129, 131] In practice, this would be the IPC team with input from the Infection Control Manager, the Infection Prevention and Control Doctor and the Consultant Microbiologist to ascertain there is microbiological knowledge, clinical expertise and an understanding of the installed water system.

To summarise, microbiological results should be interpreted alongside other quantifiable factors that are known at the time of sampling such as temperature and residual disinfectant and these should be recorded to allow for trend analysis. Differentiation between local and systematic colonisation can be achieved by collecting post-flush samples when pre-flush samples test positive. If post-flush samples test positive as well, it may indicate a systemic colonisation in the water supply. If the post-flush samples have significantly lower bacterial counts, it may indicate a local colonisation.

20. What are the water testing requirements following a positive water test result (in the absence of clinical cases)?

In total, eight pieces of evidence were identified in relation to this research question which includes two Scottish guidance documents,[101, 120] two guidance documents published by the British Standards Institution,[118, 119] three English guidance documents,[130, 131, 134] and one guidance document from Republic of Ireland.[128] All eight pieces of evidence were deemed to be expert opinions due to the lack of a

rigorous search and/or methodology in developing the guidance. In accordance with SIGN 50 methodology, these eight expert opinions were graded level 4 evidence.[101, 118-120, 128, 131, 134]

In general, re-sampling is typically carried out after any remedial actions, as an assurance that said actions have been effective. The SHTM 04-01 part A recommends that microbiological tests for bacteria colony counts at 37°C and coliform bacteria, including *Escherichia coli*, should be carried out after disinfection to establish that the work has been satisfactorily completed.[120]

Where *P. aeruginosa* and/or coliforms are identified in individual sampling points, PD 855468:2015 recommends that the sampling point should be cleaned externally, flushed and re-sampled.[119] If non-compliant results persist, investigation and subsequent remedial actions should follow. Regarding *P. aeruginosa* in augmented care, the Department of Health, Health Technical Memorandum (HTM 04-01) guidance mentions that following a positive pre-flush and remedial actions, re-testing the outlet using pre- and post-flush sampling until three consecutive negative samples is recommended at an interval of three days, two weeks and four weeks.[130] In non-high risk areas, English and Republic of Ireland guidance recommend to resample when there is a low count of *P. aeruginosa* found (1-10 cfu in 100 ml) and when the count is above 10 cfu/100 ml, remedial actions should take place and resampling should be done after three weeks.[128, 131]

Specific to *Legionella* spp., guidance on testing requirements after a positive *Legionella* test result depend on the level of cfu/litre. If *Legionella* results are between 100 and 1000 cfu/litre in the minority of samples, re-sampling should be carried out.[101, 128, 131, 134] The reasons for this are not given. It could be to ensure that the result is not a false-positive or to determine if the count has risen further while the *Legionella* plates were being cultured which may affect the clinical risk. If *Legionella* results are >1000 cfu/litre, guidance across the UK recommend that the system should be re-sampled and remedial actions should take place.[101, 128, 131, 134] Re-sampling should be undertaken following between two and seven days after disinfection, and at frequent intervals thereafter until a satisfactory level of control has been achieved.[101]

The updated guidance on *Legionella* sampling from British Standards Institution recommends that initially only a pre-flush is necessary, but when the pre-flush sample is positive, disinfected post-flush samples should be collected to determine whether the contamination is local or systemic.[118] This has also been discussed above in "How should routine water test results be interpreted?".

In conclusion, the system/outlet should be resampled following a positive water test result and, when remedial actions have taken place to ensure the actions have been effective and thus the system is not contaminated anymore. When only a minority of sampling points test positive and contain a low level of microbial counts, resampling could ensure the results are true (that is no false-positives or false-negatives). Resampling can also confirm the extent of microbial growth that has taken place while awaiting initial results. To help determine if the contamination is local or systemic, pre- and post-flush samples should be collected.

21. What actions (remedial and/or clinical) should be taken following a positive water test result (in the absence of clinical cases)?

In total, 10 pieces of evidence were identified in relation to this research question which includes one Scottish guidance document,[101] two guidance documents published by the British Standards Institution,[119, 126] four English guidance documents,[129-131, 134] one guidance document from the Republic of Ireland[128] and two international guidelines (WHO and CDC).[94, 137] All 10 guidance documents were deemed to be expert opinions and in accordance with SIGN 50 methodology, these 10 expert opinions were graded level 4 evidence.[101, 119, 126, 128-131, 134, 137]

There are some limitations to the evidence as it is mainly low quality (all 10 are SIGN level 4). The included guidance documents that were deemed expert opinion lack a rigorous search and/or methodology while developing the guidance and often refer to the same references/guidance. The CDC guidelines are mostly based on studies published pre-2000 which is a limitation as it might not reflect current IPC practices.[94]

There is limited guidance to inform remedial/clinical actions specifically in response to water samples testing positive (or out with acceptable limits). All UK guidance mention that disinfection should be considered, but that an immediate review of

control measures and risk assessment should be carried out to identify any other remedial action required.[101, 128, 129, 131, 134] These UK guidance documents do not specify remedial actions that can be taken. The British Standard 8580-2:2022 states that the WSG advises on the remedial actions required when water systems or outlets are found to be contaminated.[126] The WHO states in their guidance that the entire distribution system, including water-using devices, point-of-use and end-of-pipe devices will need to be flushed and possibly disinfected or decontaminated.[137] They also mention that small point-of-use (POU) filters could harbour contamination and may need replacing.[137]

In general, remedial actions will be specific to the water system in question and will depend on the level and location of contamination. As microbial growth and survival are dependent on a number of environmental factors, remedial actions should be determined based on consideration of the water test results in context with the water system as a whole, for instance considering other control measures (for example temperature control, pressure control, flushing) as well as chemical and potability analysis results. The British Standards Institution briefly discusses remedial actions in PD 855468:2018 and state that where system disinfection fails to remove established biofilm, consideration should be given for continuous supplementary dosing of disinfectants or removal of affected pipes and fittings for cleaning or replacement.[119] It also advises that where *Legionella* spp. have been identified following disinfection, the system should be reassessed as defined in HSG 274 Part 2.[129] Regarding clinical actions, the CDC recommends diagnostic *Legionella* testing for all patients with health-care associated pneumonia when a water sample is found to be *Legionella* culture-positive.[94]

English guidance provides a list of engineering-based actions for when a routine sample tests positive for *P. aeruginosa* or *Legionella* spp. in a high risk unit.[130] These include checking for any system dead legs, ensuring appropriate materials have been installed, and checking that connections to TMVs are appropriate.[130]

In conclusion, remedial actions will be specific per water system and depend on the contamination level and location. Guidance advises an immediate review of control measures and risk assessment to identify remedial actions required.

22. Is routine environmental testing for healthcare water system-associated organisms recommended?

In total, five pieces of evidence were identified in relation to this research question which includes one Scottish guidance document,[116] one guidance document published by the British Standards Institution,[126] two English guidance documents[131, 134] and one international guidelines (CDC).[94] All five guidance documents were deemed to be expert opinions and in accordance with SIGN 50 methodology, these five expert opinions were graded level 4 evidence.[94, 116, 126, 131, 134]

There are some limitations to the evidence as it is low quality (all five are SIGN level 4). The included guidance documents that were deemed expert opinion lack a rigorous search and/or methodology while developing the guidance and often refer to the same references/guidance. Moreover, the CDC guidelines are mostly based on studies published pre-2000 which is a limitation as it might not reflect current IPC practices.

Routine air sampling is recommended fortnightly for cardiac HCUs as this is a known reservoir of healthcare water system-associated organisms dispersed in aerosols which can indirectly infect patients undergoing cardiac surgery. For specific testing requirements, see the NHSScotland Guidance for Decontamination and testing of Cardiac Heater Cooler Units (HCUs).[116]

English guidance and the BS code of practice state that the frequency and sites for routine environmental sampling of *Legionella* spp., *P. aeruginosa* and other healthcare water system-associated organisms in healthcare facilities should be based on a comprehensive risk assessment and should be part of an overall management strategy.[126, 134] Guidance on risk assessments relevant to healthcare water system-associated organisms can be found in BS 8580.[105, 126]

Environmental sampling can be expensive and time-consuming and therefore the CDC does not recommend routine environmental sampling except in the situations where sampling is directed by epidemiological findings and results can be applied directly to infection control decisions.[94] English guidance advises that routine sampling of environmental surfaces in healthcare environments is not usually indicated, because regulations state that cleaning of the hospital environment is essential and would prevent any type of surface contamination.[131] However, as

demonstrated in the section "What are the causes/sources of environmental contamination with healthcare water system-associated organisms?", environmental reservoirs exist for these organisms and these are not routinely cleaned (examples being sink and shower drains). In this regard, there is currently inconsistency between extant guidance (currently no recommendation for environmental testing) and the primary scientific literature. Outbreak studies demonstrate that healthcare water fittings and fixtures can be environmental reservoirs for water system-associated organisms and may persist as reservoirs if the contamination is not eliminated. Regular testing of potential environmental sources of infection such as sinks, drains, shower trays and toilet cisterns could provide very valuable information in the event of a trigger breach, cluster or outbreak. This is discussed in the section "What are the environmental testing requirements during a water-associated incident/outbreak?"

23. Are there any specific actions required if an outlet tests positive pre-flush but negative post-flush?

There is limited evidence available that specifically mention outlets that test positive pre-flush but negative post-flush. Most guidance discusses lower or reduced post-flush counts compared to pre-flush counts instead. Four pieces of evidence were identified to inform the recommendations including two guidance documents published by the British Standards Institution,[118, 119] one other English guidance document,[130] and one guidance document from the Republic of Ireland.[128] All four pieces of evidence were deemed to be expert opinions due to the lack of a rigorous search and/or methodology in developing the guidance. [118, 119, 128, 130] In accordance with SIGN 50 methodology, these four expert opinions were graded level 4 evidence.[118, 119, 128, 130] The small amount and low quality of evidence identified is a limitation of the evidence base for this question.

As mentioned previously, the British Standards Institution recommends that when pre-flush tests results are positive, disinfected post-flush samples should be collected to determine whether the contamination is local or systemic.[118] Post-flush samples that are negative or have low counts would indicate a local contamination.[130] It is therefore recommended that remedial measures should be

directed towards the tap and associated pipework and fittings near to that outlet.[128, 130]

The sampler needs to be aware of the possibility that the test can fail to detect the organisms and/or contaminated water outlets can be missed. The results are representing the taken samples and not the entire water system. When samples are taken immediately after a disinfection process, false negative results might arise.[118, 119] Biocides can continue their action after sample collection and might result in lower counts or false negative test results and therefore may not be truly representative of the safety of the system at the time of sampling.[118] To prevent this, extant guidance advises that in systems where biocides are present, sterile bottles containing suitable neutralizers should be used to stop the action of the biocide at the time of collection.[118]

24. Are there any recommended methods for the removal of healthcare water system contamination?

In total, 48 pieces of evidence were included to answer this research question which includes 41 outbreak studies,[7, 8, 12-16, 18, 20, 21, 25, 26, 28-42, 44-46, 49, 50, 54, 56, 57, 62-64, 66, 78, 86] two case reports,[92, 103] two intervention studies,[55, 91] two guidance documents,[128, 139] and one international guideline.[94] The 41 outbreak studies, two case reports and two intervention studies were graded SIGN50 level 3. Three guidance documents (SHTM 04-01 Part D[139], HPSC IPC water guidelines[128] and CDC guidelines[94]) were graded as SIGN50 level 4 expert opinion due to a lack of systematic supporting evidence.

This research question is not concerned with the routine management of healthcare water systems to maintain safe water however it is acknowledged that in response to an infection incident or outbreak, efforts may focus on improvements to routine management (for example where noncompliance or failure of routine processes has been identified). Water and environmental sampling in response to an infection incident or outbreak may indicate both that contamination is present and inform the disinfection approach taken. For further details see research questions 31 ‘What are the water testing requirements during a water-associated incident/outbreak?’ and 32. ‘What are the environmental testing requirements when investigating healthcare water system-associated incidents/outbreaks?’.

Aside from two intervention studies[55, 91] most of the evidence sources for this research question consisted of outbreak studies (n=41) where more than one method of removal was implemented often at the same time (or as a result of the first attempt having failed), and removal methods implemented at the same time as other IPC remedial measures. Publication bias is also a concern. Consequently, this body of evidence was unable to determine a superior method(s) for removing microorganisms from environmental sources. A summary of this evidence base is provided below.

Thirty-seven studies (all SIGN level 3) describe physical removal of sinks, pipes, taps and associated fittings.[7, 8, 12, 14-16, 18, 20, 21, 25, 26, 28, 30-34, 36-42, 44, 46, 49, 50, 54, 56, 57, 63, 64, 78, 91, 92, 103] Tap replacement was described in 13 studies;[15, 16, 25, 30, 44, 49, 55, 64, 78, 91, 92, 103] four of these detailed installation of taps or drains with inbuilt disinfection systems or taps that could be easily dismantled for disinfection.[16, 55, 64, 91] Sensor taps were replaced with conventional mixer taps in 3 studies[25, 44, 92] Sink replacement was described in eight studies although detail as to specific sink model and design was limited.[14, 18, 38-40, 50, 56, 63] Replacement and/or redesign of drains was described in eight studies.[12, 20, 21, 28, 32, 41, 42, 46] Features included lockable P-traps and shut-off valves; in an outbreak of *P. aeruginosa*, P-traps were changed at patient discharge whenever a patient stay exceeded 1 week.[28] Modifications to shower and/or shower drainage was described in three studies.[26, 54, 57] Six studies described multiple engineering modifications to the plumbing system.[7, 8, 26, 33, 36, 63] Two studies mentioned replacement of aerators[31, 34] and one study detailed installation of rimless toilet bowls in response to a CPE outbreak.[37] There was no trend observed with regards to the type of microorganism involved and the method of removal attempted across the studies.

Twenty-nine studies (all SIGN level 3) describe chemical disinfection either of the outlet(s) or the water system itself. Hyperchlorination of the water system was described in 8 studies.[8, 12, 32, 34, 49, 54, 92, 103] Three of these were related to *Legionella* spp. contamination.[8, 92, 103] Three were in relation to *P. aeruginosa.*[32, 34, 54] One study described an outbreak of ESBL *Enterobacter cloacae* where chlorination of the water supply was carried out.[49] One study described heat treatment alongside chlorination of the main water tank and terminal points, in an attempt to reduce transmission of *S. marcescens*.[12] The concentration and frequency of hyperchlorination varied

across these studies. Silver nitrate disinfection of the water system was part of the response to an outbreak of *P. aeruginosa*.[44]

Whole system water disinfection may be carried out in addition to local (distal) outlet disinfection when contamination is identified throughout a water system (not just isolated to the outlet).[32, 34, 44] For example, hyperchlorination of the entire water system was carried out when both tap water and environmental swabs of taps and drains were positive with *P. aeruginosa*.[34] Hyperchlorination was also carried out prior to the sampling results, then repeated once results were available. The outbreak was considered over when both water and swab samples were negative. Similarly, hyperchlorination was carried out following identification of both water and environmental contamination in response to a *P. aeruginosa* NICU outbreak.[32] Although the hyperchlorination was associated with negative post-flush *P. aeruginosa* samples, pre-flush tap water samples remained positive, indicating an ongoing contamination issue at the outlet. In another outbreak study, hyperchlorination was implemented after *P. aeruginosa* continued to be present at outlets following outlet-focused control measures (in this case replacement of shower heads).[54] This coincided with installation of POU filters therefore it was not possible to determine the impact of the hyperchlorination in isolation. In some outbreak studies, it is not clear why whole system disinfection was implemented as specific details of the water sampling protocol are not provided.[49] It may be as a precautionary measure when the extent of the system contamination is unknown. However, whole system water disinfection is not designed to resolve biofilm accumulation, therefore is unlikely to have any impact where biofilms may be present in drains.[12, 49]

Twenty-one studies described chemical disinfection of outlets and associated plumbing. This included drains in 13 studies, using various methods (pouring disinfectants down drains, steam cleaning, physical scrubbing).[13-15, 18, 21, 26, 28, 29, 41, 42, 45, 46, 62] In response to an outbreak of *A. baumannii* in an ICU, all sinks were filled with sodium hypochlorite and sink plugs pulled simultaneously to synchronise the drain flushing to occur at the same time; this was carried out daily for seven days and then weekly thereafter.[18] Before the bleaching intervention, 18 patients over 10 months were infected or colonised with *A. baumannii*. After the intervention, this decreased to 19 patients over 28 months, a statistically significant decrease

(P < 0.01). The bleaching protocol was initiated after both sink autoclaving and sink replacement failed to prevent ongoing drain colonisation. In an *K. oxytoca* outbreak, a drain flushing protocol using biguanide 1.6% (a quaternary ammonium compound) was initiated on a weekly basis and left to sit for 30 minutes (by closing valves), followed by running hot water (70°C) for 5 minutes.[41] This (plus the initial complete replacement of the entire drainage system) was associated with negative environmental samples and no further patient cases of *K. oxytoca* in the following 6 months. Bleach (undisclosed concentration) was poured into drains in an attempt to remove an environmental reservoir of *P. aeruginosa* in a burns unit.[13] Combined with other remedial measures (including active environmental surveillance), this was associated with a decreased incidence of *P. aeruginosa* recovered from clinical samples from 44.7 per 1000 admissions in 2011 to 35.6 in 2012. Acetic acid (24%) was poured weekly into sink drains and left for 30 minutes before flushing in an attempt to remove environmental reservoirs of *P. aeruginosa.*[14] This was instituted as a temporary measure whilst awaiting sink replacement but became a routine measure every three months after sinks became recolonised after replacement. Pipework further downstream from the sinks was found to be colonised. Follow up at 21 months with routine environmental sampling showed sinks remained negative.

Drain disinfection proved unsuccessful in an outbreak of *K. pneumoniae* in an ICU, where drains were vaporised with a steam cleaner, followed by pouring a combination of 0.1% sodium hypochlorite, 0.1% sodium hydroxide, and 0.1% C12-C16 alkyl-dimethyl amine oxide into drains.[42] Sink drains remained colonised when tested two months later and the drains were eventually replaced. Attempts to remove biofilm by physically cleaning deep drains (which required hospital plumbers to remove screwed down sink traps) and chemical cleaning with a chlorine-based product was unsuccessful at removing the environmental CPE reservoir in a burns unit.[29] The drain cleaning was paired with pre-and post-environmental screening. Thrice daily disinfection of sink drains using accelerated hydrogen peroxide was associated with a decreased number of infections but failed to eradicate ESBL-producing *K. oxytoca* from an ICU.[46] Drains were eventually replaced.

Similar chemical disinfection measures were described for contaminated taps.[21, 46, 86] Twice monthly chlorine disinfection (aqueous solution (4.5%)) of sodium hypochlorite injected into taps with a 60mL syringe for 15 minutes was associated with a

decrease in the positivity rate of tap water.[86] *P. aeruginosa* was found in 34 out of 180 (18.8%) samples before and in 22 of 288 (7.6%) after tap disinfection was implemented (P < 0.01).

Ten of the 45 studies described some form of heat treatment. This included whole system heat treatment in 3 studies.[12, 92, 103] Two of these described infection incidents with *Legionella* spp., where heat treatment (reheating water to 58°C, and maintaining hot water at 70°C) was combined with hyperchlorination.[92, 103] Main water tanks were heat treated (and hyperchlorinated) in response to an outbreak of *S. marcesens*.[12] Two studies described localised heat treatments facilitated through flushing (flushing with 70°C hot water for five minutes following water system disinfection;[41] flushing with 90°C hot water for five minutes at high pressure[14]). Pressurised steam was used to disinfect grates and drains at 170°C in an attempt to eliminate CRE.[62] Thermal disinfection, or pasteurisation, of taps was described in three studies;[35, 64, 91] specific methods included thermal disinfection of removable taps,[91] installation of self-heating taps,[64] and heating taps in situ.[35] Two of these studies were intervention studies, the first in a UK ICU, involving installation of detachable taps that could be decontaminated in a benchtop washer-disinfector.[91] Counts of *P. aeruginosa* at the tap were compared with control taps. A negative binomial regression model showed that *P. aeruginosa* counts from the test taps were significantly lower (P < 2 x 10-16) than those of control taps. The second intervention study took place in a Dutch ICU and involved installation of disinfection devices in sink drains that apply repeated heating (to at least 85°C) and electromechanical vibration, designed to prevent biofilm formation.[55] Colonisation in the drain with MDR *P. aeruginosa* decreased to 5.1% (P < 0.001) from 51.2%. The number of positive cultures from patients significantly decreased from 4.8 per 1000 patient-days in the pre-intervention period to 2.1 per 1000 days in the first intervention period (P < 0.001).

Six of the 45 studies used all three methods (physical removal, chemical disinfection, and heat treatment)[12, 14, 41, 64, 92, 103] and the organisms included *Legionella* spp.,[92, 103] *K. oxytoca*,[41] *P. aeruginosa*,[14] *S. marcescens*,[12] and ESBLs (*C. freundii*, *R. planticola*, *E. cloacae*).[64] Both the *Legionella* spp. incidents were single patient cases, the other studies described outbreaks (multiple patients).

Installation of point of use (POU) filters was described in nine studies alongside other disinfection methods.[7, 8, 16, 28, 32, 34, 45, 54, 92] However it must be noted that POU filters do not remove microbial contamination, they only prevent microorganisms from leaving the tap.

Three guidance documents were included which are discussed below.[94, 128, 139] The guidance documents that were graded SIGN50 level 4 expert opinion lack a rigorous search and/or methodology for developing the guidance. Moreover, the CDC guidelines are mostly based on studies published pre-2000 and thus might not reflect current IPC practices.

Guidelines published by CDC (graded SIGN50 level 4) states that 'the principal approaches to disinfection of potable systems are heat flushing using temperatures 160°F–170°F (71°–77°C), hyperchlorination, and physical cleaning of hot-water tanks.' CDC advises that potable water systems are easily recolonised and may require continuous intervention, which many of the included outbreak studies for this research question demonstrated.

Expert opinion guidance on the disinfection of domestic water systems for the control of *Legionella* spp. (graded SIGN50 level 4) published by HFS (04-01 Part D) advises that 'when considering the most suitable method of disinfection for a healthcare facility a number of parameters have to be taken into consideration, factors to be considered include the condition of estate, the health of the occupants, the quality of the public water supply, finance, and the availability of resources to implement a particular regime'.[139] Disinfection methods that are covered include heat and flush, continuous chlorination, chlorine dioxide, ultraviolet light, copper silver ionisation, silver catalysed hydrogen peroxide, and ozone and chloramines. This guidance is mainly focused on routine disinfection but does indicate where specific methods may be suitable for emergency response (for outbreaks).

Expert opinion guidance (graded SIGN50 level 4) published by the Republic of Ireland HPSC describes 'systemic continuous disinfection' (temperature control, chlorine dioxide, monochloramines, copper-silver ionisation, electrochemically activated water), 'systematic intermittent disinfection' (superheat and flush, shock hyperchlorination, shock chlorine dioxide, silver catalysed hydrogen peroxide) and 'focal continuous disinfection' (ultraviolet light, ozone) methods.[128] Focal disinfection

is described as only involving a portion of the water distribution system, 'acting at the point of application with no residual effect'. Systemic intermittent methods are advised during or following an outbreak of Legionnaires' disease or other waterborne microorganisms.

In summary, extant guidance is mainly focused on routine disinfection of water systems for the control of *Legionella*. There is limited information in guidance on emergency disinfection in response to outbreaks involving microorganisms other than *Legionella* spp., and on disinfection of distal outlets and drains. When considering the most suitable method of water system disinfection, the advantages and disadvantages should be considered – limited guidance is provided in the Scottish Health Technical Memorandum (SHTM) 04-01 part D 'Disinfection of Domestic Water Systems'.[139] In addition to heat and chemical treatment of water systems, physical cleaning may be required to help eliminate built-up of scale and sediment that can protect microorganisms from the biocidal effects of disinfection systems. For emergency disinfection of outlets in response to infection incidents and outbreaks, published outbreak literature was unable to demonstrate a superior or universal disinfection method (or bundle of methods). This is in part due to the limitations of the evidence base, as most outbreak studies fail to appropriately evaluate whether interventions were successful. It must also be acknowledged that disinfection methods work at their optimal performance within different parameters, and this variation in parameters is high across the different healthcare settings described in the literature.

25. What flushing regimes are recommended for healthcare settings?

In total, eight pieces of evidence were identified in relation to this research question which includes two Scottish guidance documents,[101, 140] two guidance documents published by the British Standards Institution,[119, 126, 129, 141] two other English guidance documents,[119, 126, 129, 141] one guidance document from the Republic of Ireland,[128] and one environmental surveillance study.[142] All included guidance documents were deemed to be expert opinions and in accordance with SIGN50 methodology, these seven expert opinions were graded level 4 evidence.[101, 119, 126,

[128, 129, 140, 141] The environmental surveillance study was graded level 3 evidence using the SIGN50 methodology.[142]

The main limitation of the evidence base included in this section is the low quality of evidence. The included guidance documents that were deemed expert opinion lack a rigorous search and/or methodology while developing the guidance, often refer to the same references and guidance or do not elaborate on their conclusions and provide no indication of a regular schedule of guidance update.

Flushing of outlets is undertaken to control the temperature and maintain a flow of water to reduce the risk of microbial growth. Gavalda et al performed an eight-year longitudinal study looking at the importance of flushing among other things to control *Legionella* spp. colonisation of the healthcare water system.[142] This study showed that daily-used taps in facilities with a horizontal design (lowest floor of the studied Spanish healthcare hospital) were significantly (p=0.009) less colonised with *Legionella* spp. compared to taps that were not used daily.[142]

There is consensus in Scottish, English and Republic of Ireland guidance that in augmented care, all outlets that are not used frequently (for instance multiple times per day) should be flushed at least daily and a record should be kept of when they were flushed.[126, 128, 140, 141] The English Health Technical Memorandum Part C, Republic of Ireland and Scottish guidance all recommend that flushing should be for a period of one minute, first thing in the morning.[128, 141] Scottish guidance adds that this should be at the maximum flow rate that does not give rise to any splashing beyond the basin to avoid wider contamination.[140]

UK guidance recommends to have a twice-weekly flushing regime in healthcare facilities outside of augmented care settings.[119, 129] Republic of Ireland guidelines mention a weekly flushing regime as a minimum and acknowledge that there may be significant variances in each healthcare facility with types of taps and showers, water pressure and contamination levels and thus recommend to perform a local risk assessment.[128] If outlets are used less than weekly, removal of the outlet could be considered.[101]

In terms of the length of flushing, English guidance mention flushing for several minutes, whereas Republic of Ireland guidance mention flushing for three minutes.[128, 129] Scottish guidance specifically mention twice-weekly flushing of WC

cisterns and opening of all outlets for three minutes in a scenario when wards or departments are temporarily closed, but add that regular flushing applies to all sporadically used outlets.[101] Republic of Ireland guidelines specify that the cold and hot water outlets need to be run separately and the timing should start once the temperature is comparable with the supply.[128]

Finally, it is worth noting that if the outlet is fitted with a POU filter, the filter should not be removed in order to flush the tap unless the manufacturer's instructions advise otherwise.[141]

In conclusion, there is consensus that daily flushing for one minute in high-risk settings is required. Different frequencies and lengths outwith high-risk settings are mentioned in the included guidance documents, varying between weekly and twice-weekly for several minutes to three minutes. Performing a risk assessment would be valuable, taking into account the local water pressure and flow rate.

26. Who should be responsible for flushing?

A limited amount of evidence is available in relation to this research question. In total, four pieces of evidence were identified which includes two Scottish guidance documents,[101, 140] one British Standard[126] and one guidance document from the Republic of Ireland.[128] All included guidance documents were deemed to be expert opinions and in accordance with SIGN 50 methodology, were graded level 4 evidence.[101, 126, 128, 140] Regarding the flushing responsibilities within Scottish health and care settings, it is appropriate to only include Scottish and UK guidance documents and thus not having wider evidence is not a limitation.

Republic of Ireland advises that the Senior Charge Nurse or Clinical Lead responsible for the area must ensure that flushing is being performed as specified.[128] To ensure this task is carried out regularly, the British Standard 8580-2:2022 mentions that flushing could be incorporated into the local cleaning schedule together with the training of all relevant staff.[126]

The WSG should have oversight and provide an assurance to the NHS board on compliance with requirements including flushing.[101, 140]

27. What actions can be undertaken to reduce the risk of infection/colonisation associated with direct water usage?

In total, 34 pieces of evidence were identified in relation to this research question. The evidence base on this subject consists of 19 outbreak studies,[3, 30, 31, 34, 37, 39, 44, 45, 50, 51, 58, 60, 62, 64, 75, 110, 143-146] four Scottish guidance documents,[101, 120, 147, 148] four other guidance documents[128, 141, 149, 150] three before and after studies,[91, 151, 152] two letters to editor (expert opinion),[153] one British Standard,[126] and one international guideline.[94] Nine guidance documents are assessed as expert opinion.[94, 101, 120, 128, 141, 147-150] According to the SIGN50 methodology, 22 were graded level 3 evidence (19 outbreak studies[3, 30, 31, 34, 37, 39, 45, 50, 51, 58, 60, 62, 64, 110, 143, 144, 146]and three before and after studies[91, 151, 152]) and 12 were graded level 4 evidence (11 expert opinions[44, 94, 101, 120, 128, 141, 147-150, 153] and the British Standard[126]).

There are some limitations to the evidence that is included in this research question. The evidence is mainly low quality (all 34 studies are either SIGN50 level 3 or level 4) and the guidance documents that were deemed expert opinion lack a rigorous evidence base and/or methodology while developing the guidance and often refer to the same references/guidance. Moreover, the CDC guidelines are mostly based on studies published pre-2000 which is a limitation as it might not reflect current IPC practices.[94] Since not all outbreaks/infection incidents are published in scientific journals and numerous of the studies included here (19 out of 34) are outbreak studies, there is a risk of publication bias.

In order to reduce the risk of infection/colonisation associated with direct water usage, actions are focused on a) reducing the risk of contamination at the source and b) breaking the chain of transmission from source to patient. Direct water usage refers to uses such as bathing, oral care and drinking, whereby transmission occurs through contact and inhalation of droplets and mists. The available evidence has been summarised in different categories below covering education, sink use and design (for example preventing splash risk), engineering controls (for example use of rimless toilet bowls and removal of unused outlets), POU filters, hand hygiene (for example use of hand rub), patient exposure to tap water (for example use of sterile water for high-risk patient care), water-free care/removal of outlets, and environmental cleaning.

Education

Republic of Ireland guidance recommends that health and care staff should be trained to be aware of the potential risk of HAI from water sources and water outlets.[128] Patients also need to be informed of the risks related to healthcare water systems, and the below relevant actions they (and their care takers) can take to reduce the risk. In response to a case of *Mycobacterium mucogenicum* in a bone marrow transplant patient, staff and patients were educated on safe showering and changes were made to ensure that shower heads were left hanging straight without loops to reduce build of up stagnant water inside the shower hose.[3] In practice, it may not be possible to avoid looped shower hoses as Scottish technical guidance advises that shower heads must not be capable of being accidentally immersed in water, come into contact with drains or other potential sources of contamination.[120]

Appropriate sink use

Three studies identified inappropriate use of clinical wash hand sinks as a risk factor for source contamination; notably none of these studies describe actions following the outbreak events to improve good practice around sink use.[60, 62, 143] Sinks in patient rooms of a Korean ICU were identified as a reservoir for carbapenemase-producing Enterobacterales (CPE); dialysis solution and patient/caregiver drinks were being emptied into handwashing sinks and may have acted as a nutritional source for biofilm.[60] This study did not detail any actions to reduce inappropriate sink use. Kotsanas et al. also identified poor clinical hand sink practice during an investigation into a CPE outbreak on an Australian ICU - residual antibiotics and clinical waste were being disposed of into hand wash sinks creating a suitable environment for biofilm growth.[62] The exact transmission route was not identified however it was hypothesised that contaminated healthcare worker hands may have transmitted infection during direct patient care for example maintenance of central venous catheters. Again, attempts to reduce inappropriate sink use were not detailed in this outbreak study. In another two studies, poor sink practice was also identified as a possible contributor to transmission (for example disposal of patient waste water, nasogastric tube waste, oral medications and/or body fluids).[50, 143] Republic of Ireland guidance advises that clinical wash hand sinks should be dedicated for the purposes of hand washing only, and that alternative sinks and

sluices should be used for other purposes.[128] Garvey et al. advises disposal of patient waste water directly into the sluice or macerator after using absorbent gel sheets.[91] HTM 04-01 Part C also advises that clinical wash hand sinks should be used solely for hand washing, and that the spout should not be touched when washing hands.[141]

Sink design

Poor sink design has been identified as a risk factor for contamination in a number of studies.[30, 58, 62] There is a lack of information in the literature detailing the most effective sink design to reduce contamination and splash risk however it is evident that sinks should not be designed such that water from the tap directly hits the drain hole. A combination of poor sink design causing severe splashing around the sink, and poor practice of staff using towels to prevent overflow, contributed to an outbreak of *Acinetobacter baumannii* in a paediatric ICU in Korea.[39] Further, sink basins should be deep enough to allow hand hygiene to be performed without making contact with the basin or taps.[30] In a French outbreak study, taps were found to be too close to the sink collar making it challenging to wash hands without touching the taps while also having a risk of splashback and this likely contributed to cross contamination.[30] In an Australian outbreak study, the sink design created splashing from the drain which likely contaminated surrounding surfaces; the sink design was not compliant with Australian regulations.[62]

The British Standard (BS) 8580-2:2022 Part 2: Risk assessments for Pseudomonas aeruginosa and other waterborne pathogens - Code of practice, advises that liquid soap dispensers should not be placed directly above the sink as it can lead to soap drips on the sink surfaces that may support bacterial growth.[126]

Engineering controls

Sink design is just one example of how infection risk could be designed out of healthcare settings. Further engineering measures to reduce risk associated with direct water usage include regular replacement of tap parts. This was implemented after a pseudo-outbreak of *Mycobacterium chimaera*, where biofilms were detected on flow straighteners of handwashing sinks in patient rooms.[31] HTM 04-01 Part C

advises that flow straighteners should be avoided altogether, as they can present a surface area for colonisation of microorganisms.[141]

Two studies describe replacement of taps in response to outbreaks. Replacing taps was found to coincide with a decrease in the acquisition of *P. aeruginosa* on an ICU; however holistic measures were implemented at the same time (new tap cleaning protocol, and improved waste water management).[91] Wolf et al. also implemented tap replacement in response to patient colonisation with ESBLs found in the sink; siphons from sinks in the ICU including hand wash sinks were replaced with self-disinfecting siphons, samples taken for the eight months following installation were all negative.[64] The authors state hand wash sinks were also used to discard patient waste water and to rinse medical instrumentation prior to decontamination – it is not clear whether there was a change to this practice following the outbreak. There was no evidence identified in the literature to support a schedule of regular tap replacement outwith outbreak management.

Remodelling of sanitary and water supply systems such as installing newly designed shower drains with covers and replacing standard toilet bowls with rimless bowls was highlighted as a control measure in a German outbreak study.[51] Rimless toilet bowls were also used as a measure in a CPE outbreak study; whilst a direct link from the toilets to patients could not be confirmed, the rimless bowls facilitated easier cleaning and therefore reduced the risk of a reservoir within the toilet.[37] Both UK Health Building Note 00-10 and Scottish Health Technical Memorandum 64 advise that hospital toilets should be rimless.[148, 149] HBN 00-10 states that toilet seats should not have a cover however if covers are to be considered, consultation should take place with the infection control team at the planning stage; toilet covers are typically not recommended for independent wheelchair and assisted toilets, as they prevent the use of the backrest.[149] There is no clear consensus in literature regarding aerosolisation/splash risk from flushed toilets and whether covers should be installed as standard; further research is required in this area. Practical issues for consideration include the need for regular cleaning/decontamination of toilet covers and associated parts (for example hinges). There may also be a requirement for anti-ligature fixtures and fittings.[154]

Engineering controls to reduce the risk of contamination of water systems and thus subsequent onward transmission are covered in more detail in technical guidance published by Health Facilities Scotland.[120]

Point-of-use filters

Point-of-use (POU) filters are frequently installed during outbreak management as a short-term control measure (see "Should point-of-use (POU) filters be fitted in response to water-associated incidents/outbreaks?"). However, they are sometimes used as a long-term solution to reduce transmission risk, outside of outbreaks. Kinsey et al. installed POU filters to reduce the risk of transmission from contaminated tap water. In their NNU outbreak, patients had higher odds of having received care in a room with no POU filters installed on the sink tap during the seven days before positive *P. aeruginosa* culture (eOR, 37.55; 95% CI, 7.16–∞).[34] All 31 case patients were in rooms without POU filters during the seven days before positive *P. aeruginosa* culture, compared with 14 (45%) control patients. Further, HTM 04-01 part C advises that unless water testing has shown absence of *P. aeruginosa* in augmented care units, water (and ice) should either be sterile or should be supplied through a POU filter which suggests long-term use of POU filters for certain water uses.[141]

Hand hygiene

Use of hand rub after hand washing has been implemented during outbreaks.[34, 44] If there are ongoing concerns around water safety it could be used as a long-term approach to reducing risk of transmission from tap water. However, hand washing rather than hand rub is required for outbreaks/incidents of spore-forming infectious agents (for example *Clostridioides difficile*) and gastrointestinal (GI) viruses (for example norovirus) therefore a full switch to hand rub should be assessed on a case-by-case basis, taking into consideration this additional risk.

Minimise patient exposure to tap water

Avoidance of tap water is frequently employed as a control measure during infection incidents until the contamination and/or transmission route can be removed. There is less evidence to inform tap water avoidance for prevention of infection incidents/outbreaks. There is inconsistency in current guidance regarding the use of

tap water for specific patient groups. Republic of Ireland guidance advises that tap water may be used for washing adult and paediatric augmented care patients, provided there are no current incidents suggesting water system contamination.[128] HTM 04-01 Part C advises that only sterile water or water supplied through a point of use filter should be used for augmented care patients, unless the water has been shown to be free of *P. aeruginosa*.[141] The CDC advises that tap water should be avoided for immunocompromised patients but this is specific to *Legionella* spp. only.[94] For neonatal units, HPSC Ireland advise that sterile water or saline should be used for washing non-intact or fragile skin of neonates, including nappy changes. Tap water can be used for bathing other high risk infants with intact skin and that do not require placement in a humidified incubator.[128]

Republic of Ireland guidance advises that if a powdered infant formula feed is required, it should be prepared using boiled potable water in accordance with manufacturer's instructions.[128] There is consensus in UK and Republic of Ireland guidance that tap water should not be used in neonatal units for the process of defrosting frozen breast milk.[128, 141] Frozen breast milk may be defrosted using a warming device, defrosted in a fridge, or defrosted at room temperature and any excess discarded. Milk must never be warmed by placing the container in tap water, unless the water has first been boiled.[128]

HTM 04-01 part C further advises that single use cleaning wipes should be considered for patient hygiene on augmented care units; the guidance also advises that use of water for wet shaving and washing of patients should be reconsidered but there are no details provided for alternatives.[141] The use of cleaning wipes is not without risk; factory-level contamination with *P. aeruginosa* was responsible for more than 300 patient cases country-wide in Norway in 2022.[144]

In response to a case of *Mycobacterium mucogenicum* in a bone marrow transplant patient, staff and patients were educated on safe showering to reduce CVC contamination (disconnecting IV catheters prior to bathing, covering connections with waterproof materials).[3]

In an US outbreak study, the ice machine was found to be the likely source for colonisation of 40 patients at a HIV ward.[75] There is inconsistency in UK and Republic of Ireland guidance about the use of ice making machines in high-risk

units.[128, 141] In two outbreak studies, ice used for treatment purposes (bronchoalveolar lavage, bronchoscopy) was linked to pseudo-outbreaks in immunocompromised patients.[145, 146] Where ice is needed for treatment purposes, HTM 04-01 Part C advises it should be made using water obtained through a microbiological POU filter or boiling water in sterile ice trays or ice bags.[141] HBN 00-09 'Infection Control in the Built Environment' advises that ice for consumption by immunocompromised patients should be made by putting drinking water into single-use ice-making bags and into a conventional freezer.[150] Republic of Ireland HPSC guidance advises that an automatic dispenser should be used and the use of open chest freezer storage compartments should be avoided.[128]

Water-free care/removal of outlets

Complete unit-wide avoidance of tap water for patient care combined with or without removal of sinks as a preventative measure has been documented in three studies.[110, 151, 152] Hopman et al. introduced 'water-free' patient care in a Dutch ICU in response to an outbreak of extended-spectrum β-lactamase (ESBL)-producing *Enterobacter cloacae*.[152] All patient care related activities that take place in the patient room and that would normally involve the use of tap water were adapted to a 'water-free' alternative. Sinks were removed from patient rooms and hand washing was replaced with wipes followed by hand rub. Hair was washed with a rinse-free shampoo cap, bathing was carried out with moistened disposable wash gloves. Dental care and medicine preparation used bottled water. The overall gram-negative bacteria (GNB) colonisation rate significantly dropped from 26.3 to 21.6 GNB/1000 ICU admission days (colonisation rate ratio 0.82; 95% CI 0.67–0.99; P = 0.02). This effect was more pronounced in patients with a longer length of stay. The ICUs had to purchase mobile handwash sinks to use in the event of a *C. difficile* outbreak, acknowledging the low efficacy of hand rub against spore-forming bacteria. In an American hospital, strict tap water avoidance where sinks remained in place across 4 ICUs was associated with a significant decrease in respiratory acquisition of NTM.[151] The prevalence of positive environmental biofilm cultures for NTM was not significantly different over the study period, which provides support for the observed decrease in clinical acquisition being as a direct result of the intervention. Sterile water was used instead of tap water for routine care activities such as oral care, rinsing of suction catheters, and enteral tube irrigation. Patients were restricted from

showering; bathing was performed with waterless bath products or sterile water. Ice use was avoided and not provided for consumption or patient care activities.
A similar intervention was implemented in an attempt to control an Enterobacterales outbreak in an Australian NICU; initially six of the eight sinks in the unit were decommissioned and all bathing was replaced with sterile water or disposable wipes, and handwashing replaced with hand rub.[110] Prior to the outbreak, sinks were used for hand hygiene and to fill and empty baths for washing the less unwell infants. These interventions coincided with a decrease in cases of gram-negative colonisations/infections which was sustained. Following this, all sinks were removed (only unit entry and exit sinks remained), and the unit continues to have a minimal water use strategy in place (correspondence with author). HTM 04-01 part C recommends the permanent removal of existing outlets (for example showers and sinks) in settings where they are not being used.[141] Scottish guidance recommends that the most effective management of showers will be achieved by the removal of unnecessary ones; no further detail is provided regarding removal of other outlet types.[101] In a letter to the editor, an audit of shower utilisation across four wards (general medical ward, mixed medical speciality ward, mixed general acute ward, rehab ward) within two Scottish hospitals demonstrated an average daily non-use of showers of 86%.[153] The letter does not state the percentage of showers that were unused, but does indicate that the requirement for showers is low in these ward types. The authors argue for a move away from ensuite provision in all rooms, to provision of pre-determined shower facilities per ward (located out with patient rooms) in a bid to reduce the risk of plumbing system contamination and its associated burdens.

Further research, for example a pilot study of water-free (or reduced water) care in a Scottish setting, would be beneficial. Considerations for water-free care include the potential unintended harms resulting from disposal of wipes (for example blocked drains), use of mobile hand wash sinks, risk of infection from extrinsically contaminated water-free patient hygiene products, as well as consideration of patient dignity and comfort associated with water-free practices.

Environmental cleaning

HPSC Ireland advise that clinical wash hand sinks should be routinely cleaned in a manner that minimises the risk of contamination of the tap from organisms in the basin trap/drain.[128] HTM 04-01 Part C also advises this, stating that cleaning staff should be adequately trained to be aware of infection risks related to improper cleaning.[141] As identified in "What are the causes/sources of environmental contamination with healthcare water system-associated organisms?" transmission can occur due to splashing from contaminated outlets therefore patient care equipment and patient care items should not be stored on or near sinks or other outlets. Kotsanas et al. highlighted improper sink drain cleaning whereby the same brush had been used to clean all the sink drains on the ICU, without disinfecting the brush between drains – this can facilitate spread and seeding of drains across a ward.[62] In another outbreak study, it was found that contamination of surface cleaning equipment (for example cloths, mops and cleaning solutions) contributed to an outbreak of *P. aeruginosa*.[45] The NHS Scotland National Cleaning Specification provides instructions for cleaning sinks, wash hand basins and baths; it advises that new clean disposable cloths are used for separately cleaning the tap and the basin, however there are no instructions for cleaning taps fitted with POU filters.[147] The Cleaning Specification does not provide any information for cleaning staff regarding the risk of cross-contamination associated with cleaning.

This review identified the need to develop an effective method for drain decontamination for elimination of biofilm. Methods to disinfect drains such as pouring disinfectants down the drain, applying steam, or replacement of sink plumbing have had very limited success in eliminating biofilm from outlets.[62] Decontamination of contaminated outlets is covered in "Are there any recommended methods for the removal of healthcare water system contamination?".

There is existing guidance in place to reduce the risk of contamination associated with dental unit waterlines.

28. What actions can be undertaken to reduce the risk of infection/colonisation associated with indirect water usage?

In total, 15 pieces of evidence were identified in relation to this research question which includes eight guidance documents that were deemed expert opinion (including two Scottish[116, 117] and one British standard[126]), [94, 95, 116, 117, 126, 128, 141, 155] six outbreak studies[1, 54, 73, 76, 80, 86, 144] and one case-control study.[156] According to the SIGN 50 methodology, one was graded level 2 evidence (case-control study),[156] six were graded level 3 evidence (six outbreak studies),[1, 54, 73, 76, 80, 86] and eight were graded level 4 evidence (seven expert opinions).[94, 95, 116, 117, 126, 128, 141, 155]

There are some limitations to the evidence that is included in this research question. The evidence is mainly low quality (14 out of the 15 studies are either level 3 or level 4) and the guidance documents that were deemed expert opinion lack a rigorous search and/or methodology while developing the guidance. Additionally, there are a few outbreak studies included which creates a potential risk of publication bias as not all outbreaks/infection incidents are published in scientific journals.

Indirect water usage refers to coming into contact with water through other means, for example via contaminated equipment, contaminated environment or water-based equipment and thus transmission occurs through indirect contact. The available evidence is summarised within different categories below including clinical control measures (for example use of sterile water), management of the care environment (for example splash risk, the risk of refillable spray bottles), management of patient equipment (for example endoscopes, HCUs, incubators) and decontamination processes.

Clinical control measures

Contaminated mains water and associated outlets have been highlighted as sources for contamination in multiple outbreak studies (See "What are the known transmission routes of water system-associated organisms in healthcare settings?"). For all patient groups, Republic of Ireland guidance advise that only sterile water must be used when water is required for administering any medication or treatment requiring water for example intravenous medications, nebulisers.[128]

A closed system must be used for infants that require therapeutic cooling; sterile water must be used in the system.[128]

Management of the care environment

Splash risk from contaminated outlets has been highlighted as a risk factor in multiple outbreak studies (See "What are the known transmission routes of water system-associated organisms in healthcare settings?"). UK guidance advises that preparation areas for aseptic procedures and drug preparation and any associated sterile equipment should not be located where they are at risk of splashing or contamination from water outlets.[141] Further, it is advised that all surfaces on which aseptic procedures are to be performed are decontaminated prior to commencing a procedure. The British Standards Institute also advises that items of equipment and drug and food preparation areas should be in a location that splash contamination cannot occur.[126] It may be appropriate to consider removal of sinks altogether in these areas where this is possible, to eliminate the risk altogether.

Preparation of refillable disinfectant-detergent spray bottles with contaminated tap water was linked to a *P. aeruginosa* and *P. putida* outbreak in a paediatric haemato-oncology unit, the disinfectant-detergent spray was being used to clean perfusion bottles and the laminar flow hood prior to preparation of parenteral perfusions.[54] The refillable spray bottles were replaced with ready-to-use bottles and no further cases were detected in the subsequent two-year period. CDC guidance (graded SIGN50 level 4) advises against the use of refillable fluid containers (for example spray bottles used for cleaning).[128] The risk with refillable spray bottles is the difficulty to effectively decontaminate and dry all parts prior to reuse, therefore they may act as a reservoir for contamination. There was no published literature describing infection incidents/outbreaks in non-acute settings linked to reusable spray bottles. However, non-acute settings typically do not have surveillance systems in place and are reliant on case ascertainment to detect water system-associated incidents.

Management of patient equipment

As outlined in "What are the known transmission routes of water system-associated organisms in healthcare settings?", indirect transmission can occur from

contaminated patient equipment (for instance surgical devices, nebuliser cups, suction apparatus for ventilated patients), diagnostic equipment (for example bronchoscopy, ERCP), aspiration tubes for neonates, patient feeding items including containers for nutrition solutions, tube feeding equipment and milk bottles. HPSC Ireland specifically recommends that medical equipment and patient care equipment should not be placed in, or washed in, clinical wash hand basins.[128] Healthcare equipment (non-invasive) should be cleaned, decontaminated, dried and stored in accordance with local policy and based on manufacturer's instructions.[128] The NIPCM provides guidance for management of patient care equipment in Chapter 1.

Republic of Ireland guidance and UKHSA guidance[155] regarding the management of humidified incubators states that sterile water should be used for humidified incubators and the reservoir and water should be changed daily.[128] Reusable water reservoirs of humidified incubators must be sterilised between uses in a central decontamination unit. Republic of Ireland provides advice regarding maintenance of non-humidified incubators stating they must be completely dismantled, cleaned, decontaminated and thoroughly dried before being used again as per locally agreed procedure; sterile water is not required for this process.[128] UK guidance advises that detergent wipes are suitable for cleaning incubators.[141] An outbreak of *Klebsiella pneumoniae* in a French NNU identified contaminated incubators and incubator mattresses. Steam cleaning of the mattresses resulted in residual moisture which is likely to have supported ongoing contamination.[80] The decontamination policy for mattresses switched to chemical disinfection without steam cleaning which coincided with no further cases detected but low level contamination of incubators and mattresses persisted.

Republic of Ireland guidance advises that sterile water must be used for humidifiers in ventilator circuits and continuous positive airway pressure (CPAP) units.[128]

Decontamination processes

Endoscopes (bronchoscopes, duodenoscopes) and automatic endoscope reprocessors (AERs) have been involved in a number of outbreaks and incidents in healthcare settings.[1, 73, 86, 156] Nine patients were identified in a pseudo-outbreak of *Mycobacterium fortuitum* following contamination of bronchoscopes with contaminated mains water.[1] The bronchoscope washer disinfector did not have a

terminal filter, and the water supplying the machine was not filtered. It is essential to ensure that endoscopes are adequately decontaminated to avoid transmission of infectious agents to patients. Further guidance is provided in “NHSScotland Guidance for the interpretation and clinical management of endoscopy final rinse water”.[117]

Cardiac Heater Cooler Units (HCUs) are a known potential reservoir of healthcare water system-associated organisms that can indirectly infect patients undergoing cardiac surgery[95] and for this reason, specific guidance on operational procedures covering decontamination of heater cooler units used during cardiac surgeries, microbiological testing and associated actions based on water and air results is available - see NHSScotland Guidance for Decontamination and testing of Cardiac Heater Cooler Units (HCUs).[116]

Washing neonatal clothing in a domestic washing machine led to transmission of *Klebsiella oxytoca* to new-borns; the washing machine parts (for example detergent drawer, rubber sealant) were found to be contaminated and may have provided the conditions for biofilm growth.[76] Specific details regarding the laundering of healthcare linen is covered in the NIPCM safe management of linen literature review and National Guidance for Safe Management of Linen in NHSScotland Health and Care Environments - For laundry services/distribution.

29. What actions can be undertaken to facilitate the earliest possible detection and preparedness for clinical cases of water-associated colonisation or infection?

There is limited evidence available regarding actions that can be undertaken to facilitate earliest possible detection and preparedness for clinical cases of water-associated colonisation or infection. In total, six pieces of evidence were identified which includes one Scottish guidance document,[127] one outbreak study,[47] one surveillance study,[82] one British Standard[126] and two other guidance documents.[128, 131] All guidance documents were deemed to be expert opinions due to the lack of a rigorous search and/or methodology in developing the guidance. In accordance with SIGN 50 methodology, two were graded level 3 evidence (one outbreak study,[47] one

surveillance study[82]) and the four expert opinions were graded level 4 evidence.[126-128, 131]

English and Scottish guidance agree that significant changes in monitored microbiological levels of water test results can provide an early identification of water contamination.[126, 127, 131] Besides microbiological surveillance of water test results, surveillance of clinical samples could also aid in the early detection of colonisation/infection of water-associated organisms.[47] In absence of an outbreak, Zhou et al prospectively monitored patients for *P. aeruginosa* colonisation/infection in two surgical ICUs by active screening (also environmental screening when *P. aeruginosa* was isolated from clinical samples) to gain knowledge of the sources and patterns of *P. aeruginosa* colonisation/infection.[82] This can be beneficial for designing optimal infection prevention and control strategies. Republic of Ireland guidelines mention that the IPC team should have an active surveillance programme in place in each healthcare facility to detect alert organisms, clusters of infection, outbreaks, unexpected antimicrobial resistance mechanisms and unexpected infections.[128] Patient isolates of gram-negative bacteria (for example *P. aeruginosa*, *Acinetobacter* spp., *Stenotrophomonas maltophilia* and *Serratia marcescens*) from high-risk units and isolates of *Legionella* spp. from all care settings should be monitored as alert organisms as per appendix 13 of the NIPCM.[128] As detailed in the section "Which organisms associated with healthcare water systems are responsible for colonisation/infection of patients?", there are many organisms associated with healthcare water systems that can pose a clinical risk.

Being alert to the possibility that immunocompromised patients (see "Which patient populations are considered as being at increased risk of colonisation/infection with a healthcare water system-associated organism?") are at increased risk of colonisation/infection could also contribute to the early detection and preparedness for clinical cases. Good communication processes between IPC teams, laboratories, estates and facilities teams and WSG will help ensure earliest possible identification and preparedness for clinical cases of water associated colonisation or infection.

3.1.3 Outbreak/incident management

30. How should water-associated incidents be assessed and reported locally and nationally?

Very limited evidence was found in relation to this research question. In total, two pieces of evidence were included which consists of one Scottish guidance document that was deemed to be expert opinion [157] and an independent report from Northern Ireland, both graded SIGN50 level 4.[158]

The “Management of Public Health Incidents: Guidance on the Roles and Responsibilities of NHS Led Incident Management Teams” which is applicable to Scotland advises that following detection/recognition of an incident, the IPC team or HPT team should undertake an initial risk assessment. Limited detail of this risk assessment is provided in the guidance.

Following a series of *P. aeruginosa* outbreaks at four neonatal units in Northern Ireland, an independent report concluded that single cases of *P. aeruginosa* should be assessed in neonatal intensive care and high dependency units and possible causes investigated.[158] The independent report, although specific to neonatal cases of *P. aeruginosa*, recommended a nationally agreed approach to reporting of infection incidents.

At the time of writing, an assessment tool to undertake a risk assessment is provided within Chapter 3 of the NIPCM, in the form of the Healthcare Infection Incident Assessment (HIIAT) tool. The guidance in Chapter 3 of the NIPCM is in line with the “Management of Public Health Incidents: Guidance on the Roles and Responsibilities of NHS Led Incident Management Teams”[157] and is informed by a systematic literature review on healthcare infection incidents and outbreaks in Scotland. The HIIAT tool supports assessment of the impact of a healthcare infection incident/outbreak on patients, services and public health. It also indicates the national communication and reporting that is required based on the risk assessment.

31. What are the water testing requirements during a water-associated incident/outbreak?

There is limited guidance available regarding water testing requirements during a water-associated incident/outbreak. Three guidance documents were identified to inform recommendations on this subject which includes one Scottish guidance document,[101] one British Standard[118] and one guidance document from the Republic of Ireland.[128] All were deemed to be expert opinions due to the lack of a rigorous search and/or methodology in developing the guidance and in accordance with SIGN50 methodology, these three expert opinions were graded level 4 evidence.[101, 118, 128]

In general, the principles are similar to those described previously for routine water testing and are also covered in relevant guidance for instance BS 7592:2022 Sampling for Legionella bacteria in water systems – Code of practice, BS EN ISO 19458:2006 Water quality – Sampling for microbiological analysis and Scottish Health Technical Memorandum 04-01: Water safety for healthcare premises: Part C: TVC Testing Protocol. It is recommended by Scottish guidance, Republic of Ireland guidance and relevant British Standards to increase routine water testing during a suspected or confirmed outbreak or if surveillance identifies an increased incidence of infection.[101, 118, 128]

An overall investigation plan, which includes sampling, should be drawn up by the outbreak investigation team to identify and prioritize potential sources taking account of the geographical distribution of the infected cases.[118, 128] If the cases are clustered to a specific area, initial efforts should be concentrated on potential sources within that area.[118] Scottish guidance mentions that the outbreak investigation team may request that water samples are taken before any emergency disinfection is undertaken.[101]

Republic of Ireland guidelines mention that in the event of a suspected outbreak, additional testing by swabbing water outlets to obtain strains for typing may provide a means of assessing a water outlet, but this does not replace water sampling (see "What are the environmental testing requirements during a water-associated incident/outbreak?").[128]

It is also recommended to use pre-flush samples as these represent the water the patient would have been exposed to.[118] If contamination is detected in the pre-flush samples, differentiation between local and systemic colonisation can be achieved by collecting post-flush samples and comparing bacterial counts between pre-flush and post-flush samples.[118, 128]

32. What are the environmental testing requirements when investigating healthcare water system-associated incidents/outbreaks?

In total, 21 pieces of evidence were identified in relation to this research question which includes 14 outbreak studies,[16, 19, 34, 42, 48, 50, 57, 61-63, 66, 69, 70, 76] five guidance documents categorised as expert opinion (including one Scottish,[104] one English[131], one from the Republic of Ireland,[128] one international guideline[94] and two British Standards[118, 126]) and one surveillance study.[81] In accordance with SIGN 50 methodology, 15 were graded level 3 evidence (14 outbreak studies,[16, 19, 34, 50, 57, 61, 63, 66, 69, 70, 76] one surveillance study[81]) and six were graded level 4 evidence (six expert opinions[104, 118, 126, 128, 131]).

There are some limitations to the evidence that is included in this research question. The evidence is mainly low quality (all 21 studies are either level 3 or level 4) and the guidance documents that were deemed expert opinion lack a rigorous search and/or methodology while developing the guidance and often refer to the same references/guidance. Moreover, the CDC guidelines are mostly based on studies published pre-2000 which is a limitation as it might not reflect current IPC practices.[94] Additionally, there are numerous outbreak studies included (14 out of the 21 studies) which creates a potential risk of publication bias as not all outbreaks or infection incidents are published in scientific journals and therefore there is the possibility that the evidence may not fully reflect what is being seen in practice.

Water system-associated organisms are commonly found in moist areas and have been associated with environmental reservoirs in hospitals.[126, 128] As mentioned previously in "What are the known transmission routes of healthcare water system-associated organisms in healthcare settings?", transmission of healthcare water system-associated organisms can occur via direct and indirect contact as well as

aerosolisation. Therefore, removing contaminated environmental reservoirs could limit the exposure and transmission to vulnerable patients thus preventing prolonged outbreaks.[128]

Environmental sampling is advised when investigating the source of hospital acquired cases and/or outbreaks.[94, 104, 118, 131] An overall investigation plan should be drawn up by the outbreak investigation team to identify and prioritize potential sources taking account of the geographical distribution of the infected cases.[118, 126, 128] Where several infected people have visited one particular location or received one particular procedure, this area and/or the used equipment should be the focus of initial investigations. The number of samples to collect is therefore difficult to assess in advance as this is dependent on the nature and size of the outbreak and thus the available epidemiological information should continually be reassessed and updated.[118]

Where possible, it is recommended to compare patient isolates with the environmental samples to investigate the potential source of the infection and modes of transmission of the organism(s).[94, 104, 126, 128] To do so, the CDC advises to analyse the samples to species level at a minimum but beyond if possible. The British Standards Institution mentions in BS 8580-2:2022 that molecular typing is crucial to understand if transmission has occurred and to support interventions to prevent further transmission.[104, 126] However, if typing results do not match, it does not exclude the water system as a source of infection as environmental outbreaks can be polymicrobial and/or polyclonal.[126] It is not uncommon for environmental sampling to reveal multi-organism contamination of an outlet (for example a sink drain) suggestive of a biofilm which may only be microbiologically linked to a patient by a single organism isolate.[81] This highlights the complex nature of biofilms which can pose challenges to the interpretation of environmental sampling during outbreak studies. Picking and typing of several isolates from cultures increases the likelihood of detecting the relevant hazard.

Several outbreak studies reported successful containment of an outbreak after environmental sampling revealed the environmental source(s).[16, 19, 34, 42, 48, 57, 61, 63, 66, 69, 70, 76] Exposed sources linked to nosocomial cases included among others a washing machine, faucet aerator, sinks, chilled water dispenser, dialysis station wall boxes, siphons and drains. Although outbreak studies are generally considered as

low quality evidence, here they demonstrate the positive effect of environmental sampling in revealing the source and contributing to control the outbreak. However, environmental sampling can be challenging, for example due to resource limitations, and may not always provide clear evidence regarding the source or transmission mode in an incident/outbreak and thus typing, including whole genome sequencing, should be used to include but not exclude a source.

The species responsible for colonisation/infection in the patient may be an indicator for where to direct environmental sampling. As evidenced in the research question 'What are the causes/sources of environmental contamination with healthcare water system-associated organisms?', Enterobacteriaceae and *Pseudomonas* species are typically identified at the distal outlet/within drainage systems therefore environmental sampling (swabbing) of sink and shower drains and associated pipework may assist in identifying an environmental reservoir. In an outbreak involving CRE *Klebsiella pneumoniae*, five patients who had no overlap in hospital stay but were admitted to the same single room, were found to have the same outbreak strain which was genetically matched to the organism found in the sink and shower drain of the room.[42] In another report, the genotypic diversity of patient isolates in an outbreak involving 43 patients was an indicator that a single common source was not responsible. Environmental sampling revealed that a third of the sink drains in the haematology ward harboured ESBL-producing *E. cloacae*; all water samples were negative.[48] Similarly, a cluster of CRE outbreaks occurring over a 30 month period at an ICU were linked to sink drains where *S. marcescens* was recovered persistently, even after six attempts to decontaminate the drains of eight of the 11 central sinks in the ICU.[62] Tap spout and water cultures were negative for CRE. All patient cases were negative on admission.

In addition to source investigation, guidance from PHE and CDC recommend that environmental sampling can be used to verify the impact of cleaning procedures or other infection-control measures.[94, 131] However, incorporating this into business-as-usual could have significant logistical and cost implications.

To summarise, environmental sampling is advised during a water-associated incident/outbreak. To help with the identification and prioritising of sources, an investigation plan should be developed; the species responsible for patient

colonisation/infection may direct sampling efforts to focus on specific locations (for example drains and associated pipework for Enterobacteriaceae). Molecular typing can be a valuable tool to compare patient and environmental isolates and ideally several isolates should be analysed to increase the change of identification. Due to the challenges associated with molecular typing, it should only be used to support confirmation of a source and not to exclude it.

33. How and by whom should water-associated incidents be investigated?

In total, two pieces of evidence were identified and all three are guidance documents categorised as expert opinion.[128, 157] Chapter 3 of the NIPCM "Healthcare Infection Incidents, Outbreaks and Data Exceedance" outlines how a healthcare incident or outbreak should be investigated and is in line with the "Management of Public Health Incidents: Guidance on the Roles and Responsibilities of NHS Led Incident Management Teams".[157] Detailed information on how to conduct the investigation, as well as relevant templates, checklists and other tools are available in the NIPCM Chapter 3. The NIPCM literature review regarding healthcare infection incidents and outbreaks in Scotland (Version 2.0, June 2022) covers the investigation and management of healthcare infection incidents and outbreaks in hospital/acute settings.

It is the responsibility of the NHS board to establish whether an IMT is necessary to further investigate a healthcare infection incident.[157] The IMT is a multi-disciplinary, multi-agency group with responsibility for investigating and managing the incident and its membership will vary depending on the nature of the incident, but will normally include a NHS board Chair, HP team representatives, IPC team representatives, other relevant clinical staff, a communications officer and administrative support.[157] It is essential that Estates and Facilities staff are included in IMTs for incidents involving the built environment. In a healthcare setting, the IMT can be chaired by suitably experienced staff, for example the consultant in public health medicine (CPHM) or the Infection Control Doctor (ICD), depending on the circumstances and this should be agreed in advance and documented in the incident/outbreak plan.[157] As part of the IMT, a case definition(s) must be established. In addition, ongoing data relating to epidemiological and microbiological

investigations should be received and discussed, as well as any necessary control measures. [128, 157]

Specific to water associated incidents, investigations should also consider how water is used in the clinical areas where the patient has been cared for, how water was used by the patient and healthcare workers, the history of invasive device use, including antibiotic administrations and how drugs, particularly IV drugs, are prepared in the clinical area.[128]

34. Should point-of-use (POU) filters be fitted in response to water-associated incidents/outbreaks?

In total, 15 pieces of evidence were identified in relation to this research question which includes nine guidance documents that were deemed expert opinion (including three Scottish,[101, 120, 159] three English,[129, 130, 141] one British Standard[126] and one from Republic of Ireland[128]), six outbreak studies,[2, 8, 34, 51, 54, 77] and one before and after study.[160] In accordance with SIGN 50 methodology, seven were graded level 3 evidence (six outbreak studies,[2, 8, 34, 51, 54, 77] one before and after study[160]) and eight were graded level 4 evidence (eight expert opinions[101, 120, 126, 128-130, 141, 159]).

All studies included to answer this research question are low quality evidence, either level 3 or level 4. The guidance documents included are all deemed expert opinion (level 4) due to the lack of a rigorous search and/or methodology while developing the guidance and often refer to the same references and guidance. Moreover, by including outbreak studies (6 out of 15) there is a potential risk of publication bias as not all outbreaks/infection incidents are published in scientific journals.

Point-of-use (POU) filters are primarily recommended as a temporary control measure while awaiting a permanent safe engineering solution, although long-term use may be needed in some situations when there is no effective alternative.[126, 128-130] The installation and use of POU filters, including procedures for fitting, changing and cleaning filters should be agreed by the WSG and documented.[126, 130, 159] It is recommended to record the start date and lifespan of the POU filters installed and to replace them according to the manufacturer's recommendations.[129] The English and Scottish Health Technical Memorandums on operational management add to this that changing POU filters should be done at least once monthly and SHTM 04-01

part A mentions that the frequency depends on the usage of the outlets.[101, 120, 130] However, for certain POU filters manufacturer's instructions could recommend a frequency exceeding 30 days in which case the changing frequency might be extended.

A POU filter is defined in English guidance as a filter with a maximal pore size of 0.2 µm applied at the outlet, which removes bacteria from the water flow.[129, 130, 141] Filters that are effective barriers of healthcare water system-associated organisms range in size from 0.2 µm to 0.65 µm.[128] Most guidance, including two UK codes of practice, recommend filters with a pore size no greater than 0.2 µm,[126, 128-130, 141] but the Scottish Health Technical Memorandum recommends a pore size of 0.1 µm or less.[120] Scottish guidance warns that filters do not eradicate the organism, but prevent discharge to the environment from the filtered outlet only and that it may be possible for the organisms to multiply and regressively 'seed' other parts of the distribution system when retaining the organism within the pipework.[120]

Installation of POU filters should be subject to risk assessment and domestic staff and ward staff should be aware of the associated risks of installation, cleaning and removal of POU filters.[126] These risks include poor flow from filters which increases the likelihood of removal and a false sense of security which could result in reduced compliance with other prevention measures such as regular flushing, hand hygiene and safe discarding of contaminated fluids.[126] Cross-contamination is also a serious risk and can occur due to poor fitting allowing leakage around the fitting or by removing, changing or cleaning POU filters when retained organisms get released and resulting in re-seeding of the environment.[126, 141] Therefore, POU filters should not be re-attached once removed.[126] Appropriate training of staff is necessary and manufacturer's instructions should be followed at all times.[101, 130]

The use of POU filters has mostly been mentioned in the literature as part of a bundled approach of IPC measures and therefore it is difficult to determine which measure was responsible for the impact.[2, 8, 51, 54] However, a few outbreak studies reported specifically that the use of POU filters resulted in the control of the outbreak.[34, 77] In a neonatal ICU, POU filters provided a short-term solution during a *P. aeruginosa* outbreak attributed to hospital tap water. This study included a case-control element and showed that patients receiving care in a room without POU filters installed had significantly higher odds to be infected with *P. aeruginosa*.[34] In

another outbreak study of invasive fusariosis in a children's cancer hospital, multiple control measures were tried before POU filters were installed 1 year after the first case and the outbreak was finally controlled.[77] Moreover, a before-and-after study looked at the intervention of POU filters and compared the infection rate before and after the installation of the POU filters.[160] POU filters were shown to be effective and a significant reduction (56%, P<0.0003) of chronically endemic *P. aeruginosa* infections was measured on a surgical ICU.[160]

In summary, POU filters can be used in response to water-associated incidents/outbreaks. However, guidance suggests that their risks needs to be considered in a risk assessment before installation and the WSG should agree on their installation and use.

35. When can POU filters be removed?

In total, six pieces of evidence were identified in relation to this research question which includes two Scottish guidance documents,[101, 159] one British Standard,[126] one guidance document from the Republic of Ireland[128] and two other UK guidance documents.[129, 130] All included guidance documents were deemed to be expert opinion and in accordance with SIGN 50 methodology, these six expert opinions were graded level 4 evidence.[101, 126, 128-130, 159] The lack of high quality evidence is a limiting factor for answering this research question.

Since extant guidance recommends that POU filters are fitted in response to water associated outbreaks while awaiting a permanent solution, this suggests that removal is only appropriate when this permanent solution has been installed.[126, 128-130] Guidance do not specifically mention when exactly the POU filter can be removed, only that its removal would be a clinical decision and that amendments to plumbing and taps should have been made. The British Standard 8580-2:2022 recommend that pre-determined criteria for when filters can be removed should be in place and Scottish guidance recommend that the WSG will have to confirm that they are satisfied that the affected outlet and pipework can be removed or disinfected without compromising the rest of the water.[126, 159]

It is important to note that where POU filters are no longer required, the outlet and associated pipework should be cleaned and disinfected to remove any accumulated

debris before the system is returned to service. Manufacturer's instructions should be followed at all times.[101, 130]

3.1.4 Organisational Management:

36. Whose responsibility is it to carry out any of the above actions?

In total, five pieces of evidence were identified including two Scottish guidance documents,[101, 159] one British Standard,[126] one guidance document from the Republic of Ireland[128] and one English guidance document.[131] All included guidance documents were deemed to be expert opinion and in accordance with SIGN50 methodology, these five expert opinions were graded level 4 evidence. [101, 126, 128, 131, 159] Regarding the responsibilities within Scottish health and care settings, it is appropriate to only include Scottish/UK guidance documents and thus not having wider evidence is not a limitation.

Full description on roles and responsibilities within NHSScotland regarding water safety for healthcare premises can be found in SHTM 04-01, Part B: Operational management. Within NHS boards, SHTM 04-01 and British Standards Institute guidance recommend that a multidisciplinary team (Water Safety Group) needs to be appointed to carry out risk assessments and develop a water safety plan (WSP) to manage the identified risks associated with water.[101, 126] WSGs will be led and chaired, as a minimum, by the Responsible Person (Water) who will ensure that responsibility is taken for microbiological hazards and are identified by appropriate Group members.[101] They will assess risks, identify and monitor control measures and develop incident protocols.[101] The WSG should be a sub-group of and report to the Chair of the hospital Infection Control Committee and ensure a coordinated approach exists between Infection Prevention and Control Teams, clinical staff and Estates & Facilities on all water issues including the communication of positive environmental and water test results to the appropriate staff and teams.[101] There should be a clear line of responsibility to the Chief Executive Officer (CEO).[101]

The WSG should have oversight regarding flushing responsibilities and agree frequencies for each area – operationally this may be the Senior Charge Nurse, Clinical Lead, Domestic staff or Estates and Facilities (See "Who should be responsible for flushing?").

Scottish guidance advises that the WSG (which includes the Infection Control Manager, the Infection Prevention and Control Doctor and the Consultant Microbiologist) should advise on infection control policy (in agreement with the IPC team) and are responsible for the maintenance of water quality from the point it leaves the tap.[101]

English and Republic of Ireland guidance mention that sampling should be undertaken by appropriately trained staff to minimise contamination whereas testing should be carried out by a laboratory that is UKAS-accredited to perform the specified test.[128, 131] More information on the organisational structure for NHS boards for the management and control of risk from potential exposure to *Legionella* spp., *Pseudomonas* spp. and other similar harmful bacteria can be found in SHTM 04-01 part G: Operational procedures and Exemplar Written Scheme.[159]

3.2 Implications for research

This systematic literature review has identified gaps in literature in various subjects regarding IPC related aspects/impacts of the healthcare water system. More research and/or separate pieces of work are required to ascertain: safe use of drains in terms of IPC, safe commissioning, incubation periods for water system-associated organisms, infectious doses of organisms, design features that limit transmission of organisms from their source and reservoir to external surroundings, water testing methodologies and techniques, methodology and interpretation of environmental sampling tests. Moreover, dental units were not extensively mentioned in this review as this is covered by a separate piece of work "Literature Review and Recommendations: Management of Dental Unit Waterlines" which covers the management (including decontamination) of dental unit waterlines for the prevention of HAI in general dental practices, community dental clinics and dental hospitals waterlines.

Much of the evidence base in this literature review is composed of outbreak investigation studies where infection control strategies are bundled together making it difficult to determine which intervention(s) was/were responsible for successfully ending the outbreak. More research on single interventions would be beneficial to strengthen the evidence base. This includes water-free care and within this review it

has been highlighted that a pilot study undertaken in a Scottish ICU or NNU to trial water-free care with removal of sinks/outlets would be beneficial. More primary research for non-acute settings is also required. Overall, improved publication of outbreak studies and their management and resolutions would increase the tools for controlling and mitigating risk of water system-associated infections.

Limited robust literature was identified by this review regarding the incubation period and the period of communicability of water system-associated organisms, although there is acknowledgement that transmission can take place as long as the source is present. The incubation period is difficult to determine without the knowledge of the source and time of exposure. Investigations should be carried out to examine the potential incubation period and the linkage to organisms.

It has been mentioned by experts that the interpretation of TVC test results is difficult and that more research is needed to inform recommendations. A pilot study in an NHS health board would be useful to create a baseline for TVC monitoring. Within this pilot study, regular TVC samples should be collected, interpreted and compared with wider microbiological testing results to establish any link between results. This would help in understanding the value of TVCs and could lead into a new suite of guidance that includes among others the testing frequency, number of samples needed, sample locations and microbiological limits. There is no clear consensus in literature regarding aerosolization or splash risk from flushed toilets and whether covers should be installed as standard. Further research is required in this area as toilet covers and associated parts (for example hinges) can give rise to other contamination issues.

Moreover, there is a clear gap in literature regarding accredited testing and microbiological water testing requirements at commissioning. Limited UKAS accredited tests are available for environmental organisms. Regarding testing requirements at commissioning, guidance states that water samples are obtained as standard practice at water system commissioning to ensure a safe handover of the water system from the contractor. However, the water testing requirements and appropriate microbiological parameters are not specified. This must be agreed prior to tender, but advice on the sampling regime and microbiological parameters would be very valuable. Therefore, more research and inclusion of commissioning in

current guidance or the development of new guidance is needed especially regarding specific microbiological water testing requirements.

In a non-systematic review, it was discussed that many of the sinks involved in outbreaks do not adhere to the recommended standards despite the fact that sink design is the primary driver behind sink-related healthcare water system-associated infections. This could be due to the age of healthcare facilities and the high expense of retrofitting.[161] However, it does reflect the overlooked importance of IPC in healthcare design and this can be addressed in further research and guidance into design elements of healthcare facilities. Education on the importance of water management is also essential for awareness and understanding compliance of healthcare staff including clinicians, which was highlighted in an outbreak study on Legionnaires' disease.[2]

It is also important to note that there is a range of other environmental organisms that cause a risk to vulnerable patients in addition to water system-associated organisms as their optimum environment are warm damp areas. These organisms may not be directly found in water but can be found in moist areas such as showers (walls, basins), external parts of taps or shower outlets and near leaks or splashes. Opportunistic fungi such as *Aspergillus* spp. have been reported to be present in environmental water sampling.[162] The primary mode of nosocomial transmission is thought to be via airborne spores, but moulds can reside in water sources within the hospital and aerosolise after water activities.[163, 164] For example, *Aspergillus fumigatus* was recovered from a shower wall in a patient's room that matched the clinical *A. fumigatus* strain of the occupying patient.[165] Conversely, water system-associated organisms such as *P. aeruginosa* can be transmitted through the air which was shown by an environmental surveillance study in a UK cystic fibrosis (CF) centre revealing that 80% of the air samples inside the patient's room were positive for the endemic strain and *P. aeruginosa* was still detected in the air for one to three hours after patients' discharge.[166] More research is required to determine whether these air samples are viable and whether there is an associated exposure risk.

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Appendices

Appendix 1: Specific standards relating to water systems

This appendix provides a non-exhaustive list of standards pertaining to water systems. The standards listed represent the most recent versions available at the time of writing. Please note, however, standards are subject to amendments and the most recent versions should always be sourced and used in practice.

Standard	Title	Description	Publication date
BS EN 806	Specifications for installations inside buildings conveying water for human consumption	This European Standard specifies requirements gives recommendations on the design, installation, alteration, testing, maintenance and operation of potable water installations within buildings and, for certain purposes, pipework outside buildings but within the premises. This standard consists of 5 parts as follows: Part 1: General Part 2: Design Part 3: Pipe sizing — Simplified method Part 4: Installation Part 5: Operation and maintenance	2000 - 2015
BS 7592:2022	Sampling for Legionella bacteria in water systems – Code of practice	This standard gives recommendations and guidance on the sampling of water	February 2022

Standard	Title	Description	Publication date
		and related materials for the investigation of the presence of organisms of the genus *Legionella*. The standard is applicable to the selection of sampling sites and the methods of sampling for the purposes of routine monitoring, validation, commissioning, investigating a problem, or outbreak investigation.	
BS 8554:2015	Code of practice for the sampling and monitoring of hot and cold water services in buildings	This British Standard gives guidance and recommendations for investigative and planned collection of hot and cold water samples during the life of a building, including sampling locations and the selection of laboratory or on-site testing for those samples.	September 2015
BS 8558:2015	Guide to the design, installation, testing and maintenance of services supplying water for domestic use within buildings and their curtilages – Complementary guidance to BS EN 806	This standard provides complementary guidance to BS EN 806. It is a guide to the design, installation, alteration, testing, operation and maintenance of services supplying water for domestic use within buildings and their curtilages. BS EN 806 does not cover underground pipework, but this British Standard gives guidance on underground pipework within the curtilage of a building.	September 2015

Standard	Title	Description	Publication date
BS 8580-1:2019	Water quality – Risk assessments for Legionella control – Code of practice	This standard gives recommendations and guidance on Legionella risk assessment relevant to water systems.	January 2019
BS 8580-2:2022	Water quality Part 2: Risk assessments for *P.* aeruginosa and other waterborne pathogens — Code of practice	This standard gives recommendations and guidance on how to carry out risk assessments for *P.* aeruginosa (PA) and other waterborne pathogens whose natural habitat is within constructed water systems and the aqueous environment (autochthonous) rather than those being present as a result of a contamination event. It includes those pathogens that can colonize and grow within water systems and the associated environment, with the exception of Legionella which is covered in BS 8580-1:2019.	January 2022
BS 8680:2020	Water quality — Water safety plans — Code of practice	This standard gives recommendations and guidance for the development of a water safety plan (WSP) for water systems which can pose a risk to those exposed, either from the water itself, aerosols derived from it or the surrounding environment.	May 2020

Standard	Title	Description	Publication date
PD 855468:2015	Guide to the flushing and disinfection of services supplying water for domestic use within buildings and their curtilages	This Published Document, not to be regarded as a British Standard, provides guidance on cleaning, flushing and disinfection of services supplying water for domestic purposes within buildings and their curtilages.	September 2015
BS ISO 5667-24:2016	Water quality — Sampling — Part 24: Guidance on the auditing of water quality sampling	This standard provides an audit protocol to monitor conformity with declared, or assumed, practices in all areas of water quality sampling. It is applicable to the audit of sampling activities from the development of a sampling manual through to the delivery of samples to the laboratory.	April 2016

Legend:
BS = British Standards produced by the British Standards Institution (www.bsigroup.co.uk)

EN = European Standards (European Norm) produced by the European Committee for Standardisation (www.cen.eu)

ISO = International Standards produced by the International Standards Organization (www.iso.org)

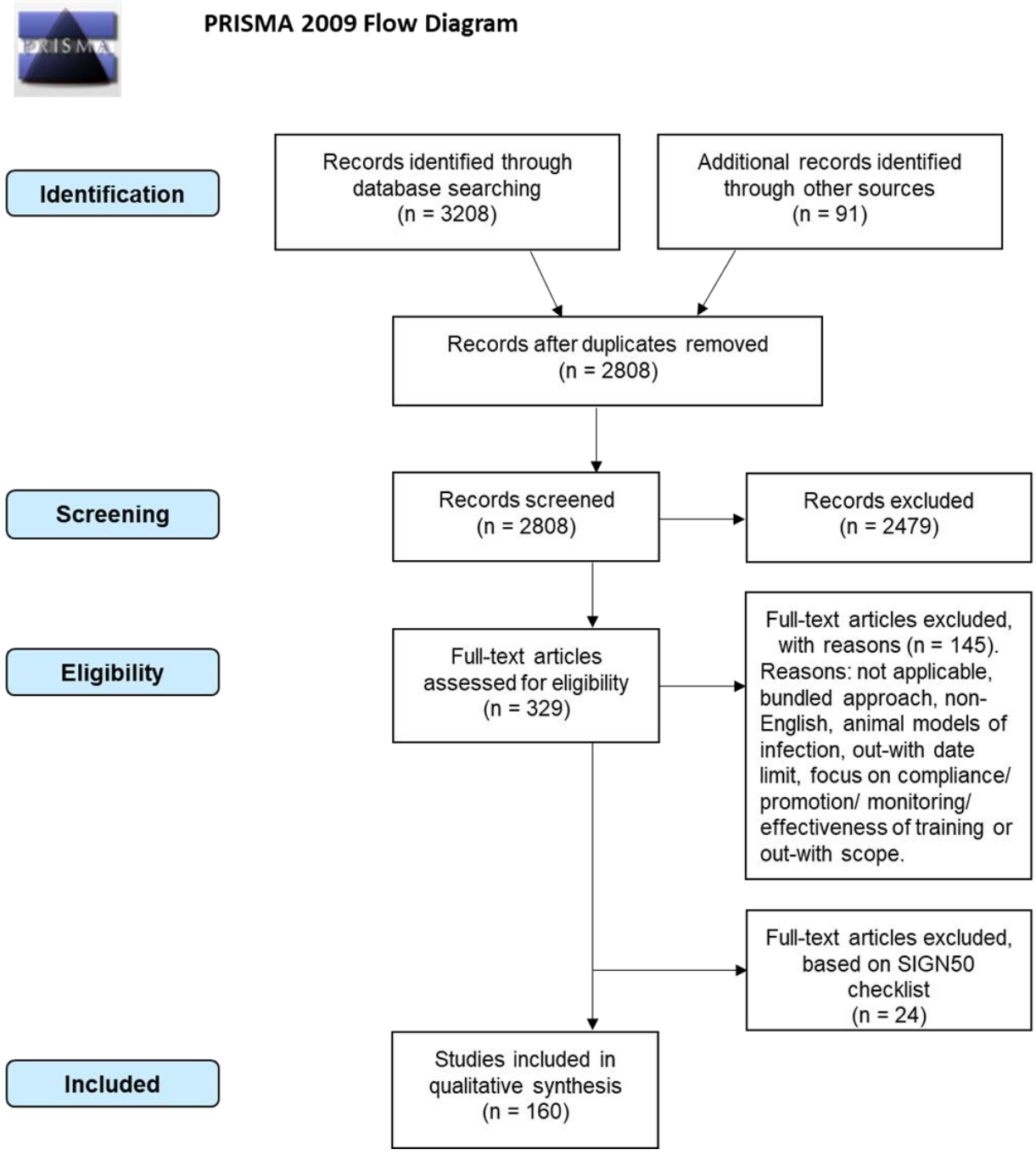
EN standards are gradually being replaced by ISO standards – when these are adopted in the UK they are prefixed with BS (e.g. BS EN; BS EN; BS EN ISO). This is usually to accommodate UK legislative or technical differences or to allow for the inclusion of a UK annex or foreword.

Appendix 2: Levels of Evidence

Grade	Description
1++	High quality meta analyses, systematic reviews of RCTs, or RCTs with a very low risk of bias
1+	Well conducted meta analyses, systematic reviews of RCTs, or RCTs with a low risk of bias
1-	Meta analyses, systematic reviews of RCTs, or RCTs with a high risk of bias
2++	High quality systematic reviews of case-control or cohort studies. High quality case-control or cohort studies with a very low risk of confounding, bias, or chance and a high probability that the relationship is causal
2+	Well conducted case control or cohort studies with a low risk of confounding, bias, or chance and a moderate probability that the relationship is causal
2-	Case control or cohort studies with a high risk of confounding, bias, or chance and a significant risk that the relationship is not causal
3	Non-analytic studies, for example case reports, case series
4	Expert opinion

Appendix 3: PRISMA Flow Diagram

For more information, visit http://www.prisma-statement.org/



Appendix 4: Excluded studies

The following studies were excluded during critical appraisal based on their limitations according to the SIGN50 checklist:

- Turner C, Mosby D, Partridge D, et al. A patient sink tap facilitating carbapenemase-producing enterobacterales transmission. *Journal of Hospital Infection* 2020; 104: 511-512. Letter.
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- Cuttelod M, Senn L, Terletskiy V, et al. Molecular epidemiology of *Pseudomonas aeruginosa* in intensive care units over a 10-year period (1998-2007). *Clinical Microbiology and Infection* 2011; 17: 57-62.
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- Gillespie TA, Johnson PRE, Notman AW, et al. Eradication of a resistant *Pseudomonas aeruginosa* strain after a cluster of infections in a hematology/oncology unit. *Clinical Microbiology and Infection* 2000; 6: 125-130.
- Perkins KM, Reddy SC, Fagan R, et al. Investigation of healthcare infection risks from water-related organisms: Summary of CDC consultations, 2014-2017. *Infection Control & Hospital Epidemiology* 2019; 40: 621-626.
- Jeanvoine A, Meunier A, Puja H, et al. Contamination of a hospital plumbing system by persister cells of a copper-tolerant high-risk clone of *Pseudomonas aeruginosa. Water Research* 2019; 157: 579-586.
- Shepherd MJ, Moore G, Wand ME, et al. *Pseudomonas aeruginosa* adapts to octenidine in the laboratory and a simulated clinical setting, leading to increased tolerance to chlorhexidine and other biocides. *Journal of Hospital Infection* 2018; 100: e23-e29.
- Breathnach AS, Cubbon MD, Karunaharan RN, et al. Multidrug-resistant *Pseudomonas aeruginosa* outbreaks in two hospitals: association with contaminated hospital waste-water systems. *Journal of Hospital Infection* 2012; 82: 19-24.
- Crivaro V, Di Popolo A, Caprio A, et al. *Pseudomonas aeruginosa* in a neonatal intensive care unit: molecular epidemiology and infection control measures. *BMC Infectious Diseases* 2009; 9: 70.
- Fanci R, Bartolozzi B, Sergi S, et al. Molecular epidemiological investigation of an outbreak of *Pseudomonas aeruginosa* infection in an SCT unit. *Bone Marrow Transplantation* 2009; 43: 335-338. Research Support, Non-U.S. Gov't.
- Livni G, Yaniv I, Samra Z, et al. Outbreak of *Mycobacterium mucogenicum* bacteraemia due to contaminated water supply in a paediatric haematology-oncology department. *Journal of Hospital Infection* 2008; 70: 253-258. DOI: 10.1016/j.jhin.2008.07.016.

- Baker AW, Stout JE, Anderson DJ, et al. Tap water avoidance decreases rates of hospital-onset pulmonary nontuberculous mycobacteria. *Clin Infect Dis* 2021; 73: 524-527. 2020/08/24. DOI: 10.1093/cid/ciaa1237.
- Sasahara T, Ogawa M, Fujimura I, et al. Efficacy and effectiveness of showerheads attached with point-of-use (POU) filter capsules in preventing waterborne diseases in a Japanese hospital. *Biocontrol science* 2020; 25: 223-230. DOI: https://dx.doi.org/10.4265/bio.25.223.
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- Backman L, Dumigan DG, Oleksiw M, et al. A cluster of gram-negative bloodstream infections in Connecticut hemodialysis patients associated with contaminated wall boxes and prime buckets. *American Journal of Infection Control* 2022. DOI: https://dx.doi.org/10.1016/j.ajic.2022.08.007.
- Pulusu CP, Manivannan B, Raman SS, et al. Localized outbreaks of *Pseudomonas aeruginosa* belonging to international high-risk clones in a south Indian hospital. *Journal of medical microbiology* 2022; 71. DOI: https://dx.doi.org/10.1099/jmm.0.001500.
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Appendix 5: Literature review search strategy

MEDLINE search 2000 to current

1. exp water/
2. water system*.mp.
3. (tap* or faucet*).mp.
4. (basin* or sink*).mp.
5. drain*.mp.
6. shower*.mp.
7. 1 or 2 or 3 or 4 or 5 or 6
8. Disease Outbreaks/
9. outbreak*.mp.
10. Waterborne Diseases/
11. nosocomial infection*.mp.
12. exp Disease Transmission, Infectious/
13. 8 or 9 or 10 or 11 or 12
14. exp Hospitals/
15. exp Health Facilities/
16. healthcare facilit*.mp.
17. healthcare setting*.mp.
18. test*.mp.
19. flush*.mp.
20. 14 or 15 or 16 or 17 or 18 or 19
21. 7 and 13 and 20

Limit 21 to English language and humans

EMBASE search 2000 to current

1. exp water/
2. water system*.mp.
3. exp Water Supply/
4. (tap* or faucet*).mp.
5. (basin* or sink*).mp.
6. drain/
7. drain*.mp.
8. shower*.mp.
9. 1 or 2 or 3 or 4 or 5 or 6 or 7 or 8
10. outbreak*.mp.
11. exp Water Borne Disease/
12. exp Disease Transmission/
13. nosocomial infection*.mp.
14. 10 or 11 or 12 or 13
15. exp Hospital/
16. exp Health Care Facility/
17. healthcare setting*.mp.
18. test*.mp.
19. flush*.mp.
20. 15 or 16 or 17 or 18 or 19
21. 9 and 14 and 20

Limit 21 to human and English language

CINAHL search 2000 to current

S18	S7 AND S13 AND S16
S17	S7 AND S13 AND S16
S16	S14 OR S15
S15	healthcare setting*
S14	(MH "Health Facilities+")
S13	S8 OR S9 OR S10 OR S11 OR S12
S12	waterborne infection*
S11	nosocomial infection
S10	(MH "Cross Infection")
S9	(MH "Disease Transmission+")
S8	outbreak*
S7	S1 OR S2 OR S3 OR S4 OR S5 OR S6
S6	shower*
S5	drain*
S4	basin* OR sink*
S3	tap* OR faucet*
S2	"water system*"
S1	(MH "Water")

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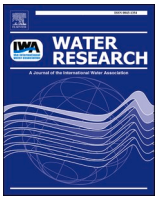

Reversing and controlling microbial proliferation in the water system of a high-risk hospital ward after extended closure and reconstruction

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Total viable count
Opportunistic premise plumbing pathogens
Point-of-use filters
Faucet
Sink

ABSTRACT

Opportunistic premise plumbing pathogens occur naturally in water but can pose a health risk to hospital patients who are more vulnerable due to illness or treatment. Ward closure periods can lead to microbial proliferation within water systems, posing a challenge to hospital estates and infection control teams. Following the 3-year closure of our paediatric haemato-oncology ward, water testing showed high total viable counts (TVCs) in over 20 % of samples and elevated counts of numerous Gram negative bacterial species (GNBs) in 73 % of samples, despite daily flushing and continuous chlorine dioxide dosing. We aimed to determine the extent of microbial proliferation, measure the impact of three sequential interventions (system disinfections with chlorine, with silver stabilised hydrogen peroxide, and then tap replacement), and assess the long-term performance of this water system. By sampling systematically across spatial and temporal scales, and using a range of microbiological tests (TVCs, *Legionella* spp., *Pseudomonas* spp., Gram negative bacteria, atypical mycobacteria and fungi), we showed that microbial proliferation was confined to the closed ward. Chlorine treatment had no significant impact on TVCs, but both silver stabilised hydrogen peroxide and tap replacement resulted in significant decreases ($p < 0.01$). Similarly, the three Gram negative species that were enriched following the reconstruction period (*Cupriavidus pauculus, Sphingomonas paucimobilis*, and *Acidovorax temperans*) were less impacted by chlorine than by the other interventions. Following these interventions, fewer than 1 % of samples exceeded our strict local TVC threshold of 10 CFU/ml and GNBs were detected in 7 % of samples. Since the ward reopened to patients in 2022, there has been no return of the high microbiological counts observed immediately after reconstruction. Gram negative bacteria have been detected only sporadically, and the taxa found in samples collected through 0.2 μm point-of-use filters shifted towards species associated with humans. Our systematic approach was successful in returning this hospital water system to a safe state, and once microbial proliferation within the system itself was rectified, further positive results were likely attributable to the interactions of users with the outlets. Distinguishing between possible sources of microbial counts in water is crucial to selecting the most suitable interventions and helping ensure provision of safe water to patients.

1. Introduction

Engineered water systems harbour diverse microorganisms, including bacteria and fungi. While most are harmless, some can occasionally cause disease and have been called 'opportunistic premise plumbing pathogens' (OPPPs) (Falkinham et al., 2015). Under certain conditions, notably in areas of stagnation and where temperature control is poor, OPPPs can proliferate both in the planktonic phase (Nisar

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et al., 2023) and by forming biofilms (Health Facilities Scotland, 2014a). The risk posed by OPPPs is heightened in hospitals, which are often within buildings that were expanded and retrofitted at various times and therefore have complex water systems, and where people who are particularly vulnerable to infection by OPPPs due to underlying disease and/or invasive medical treatment spend a considerable amount of time. The combination of complex water systems and high-risk patient populations can result in outbreaks caused by OPPPs such as *Legionella pneumophila, Pseudomonas aeruginosa*, other Gram negative bacteria, and nontuberculous (i.e. atypical) mycobacteria (Decker and Palmore, 2013; Kanamori et al., 2016). Controlling the proliferation of OPPPs is therefore especially important in hospitals. In the UK, this responsibility is increasingly shared among multidisciplinary teams that include water engineers, facilities managers, microbiologists, and specialists in infection prevention and control.

Multiple layers of control are often deployed in hospitals to minimise microbial proliferation and biofilm formation. These can include on-site filtration and chemical dosing, systematic flushing to reduce areas of stagnation, temperature control and monitoring, scheduled replacement of system components, and regular microbiological testing. However, periods of closure and reconstruction bring specific risks: some of the regular controls may be suspended, the work itself can introduce contaminants, and water movement is greatly reduced compared with occupied wards. As such, specific guidance exists for managing hospital water systems during closure periods. For example, the Scottish Health Technical Memorandum (SHTM) 04-01 (Water Safety for Healthcare Premises) specifies that during temporary closure of wards/departments, a flushing procedure must be implemented for the hot and cold water services (with all outlets flushed for 3 minutes on a twice-weekly cycle), or if this is not feasible, then a pre-occupation disinfection procedure may be carried out as recommended for new installations (Health Facilities Scotland, 2014a, 2015). Similar recommendations are found in English (Department of Health, 2016) and Irish guidance (HPSC Scientific Advisory Committee, 2015). More general guidance (not specific to healthcare) from the UK recommends that when parts of a building are temporarily taken out of use, that they then be recommissioned as though they were new, i.e. thoroughly flushed, cleaned and disinfected (Health and Safety Executive, 2014). Similarly, the U.S. Centers for Disease Control and Prevention (2024) and the U.S. Environmental Protection Agency (2020) provide guidance on managing water systems during periods of shutdown and on reopening buildings after prolonged closure, with both emphasising the importance of flushing and cleaning.

All such guidance was followed or exceeded during the 3-year closure and reconstruction of the paediatric haemato-oncology ward in the Royal Hospital for Children, part of the Queen Elizabeth University Hospital in Glasgow, UK. Despite this, when the works were completed in September 2021, microbiological testing across the ward showed total viable counts (TVCs) that exceeded our local thresholds for high-risk clinical areas, which in the absence of recommended thresholds in applicable guidance, our health board has set to 10 CFU/ml for both 22 °C and 37 °C incubation temperatures. Furthermore, testing specifically for Gram negative bacteria detected high counts of numerous species, whereas our local threshold for this test is zero CFU/100 ml (i.e. any count is deemed out of specification).

Given the evidence of microbial proliferation in this water system during the period of closure despite continuous chlorine dioxide dosing across the entire building and flushing of the system more frequently than recommended in the SHTM, and given the vulnerability of the patient population that would be occupying the ward once it reopened, we aimed to determine 1) the full extent of microbiological proliferation in the system and how it compared with nearby wards that had not been closed during this period, 2) the impact of specific interventions carried out to reduce the microbial load of the system, namely chemical treatments with chlorine (as sodium hypochlorite) and silver stabilised hydrogen peroxide, as well as replacement of taps, and 3) the long-term performance of the system once the microbial counts had been brought back within acceptable limits. We addressed these aims using a systematic approach to water testing, to implementing and assessing key interventions, and to long-term monitoring once the ward reopened to patients.

2. Methods

2.1. Background and site description

The Queen Elizabeth University Hospital (QEUH) campus, part of the UK National Health Service Greater Glasgow and Clyde health board (NHS GGC), includes an adult hospital with 1109 beds and the attached Royal Hospital for Children (RHC) with 256 beds. The model throughout these buildings, which opened in 2015, is for single patient rooms with ensuite facilities, including showers. Water is supplied to both buildings via a basement tank system that includes ultra-filtration of incoming mains water. On-site chlorine dioxide supplementation was added in December 2018, with a target concentration of 0.1 to 0.5 mg/l at the outlets as recommended in applicable guidance (Health and Safety Executive, 2014; Health Facilities Scotland, 2014a). ClO_2 levels are routinely monitored across both buildings to ensure these concentrations are maintained at the outlets.

In 2018, concerns were raised about Gram negative bacterial infections in the RHC paediatric haemato-oncology ward and possible links to the hospital water system. An initial water sampling sweep in the ward showed that 75 of 98 outlets (76.5 %) that were tested specifically for *Cupriavidus pauculus* were positive (Inkster et al., 2021), though the methods used at that time were subsequently shown to misidentify other organisms as *C. pauculus* (Inkster et al., 2022). The ward was closed in September 2018, with patients decanted to two wards in the adult hospital building. Apart from a brief period in early 2019 when it was reopened as a general ward (not for high-risk patients), it remained closed until 2021 to allow extensive reconstruction of its ventilation and water systems. This included running hot flow and return services as close as practicable to the outlets (rather than terminating in the ceiling), changing taps and wash hand basins, replacing cistern toilets with direct flush models connected directly to the cold lines, reconfiguring some rooms to suit ward operations, which resulted in the removal of two toilets, a bath, and a wash hand basin, and removing eight trough sinks from anterooms, as these were deemed surplus to requirement and risked becoming little-used outlets. The final configuration of the ward, which has both an inpatient and a day unit, includes 25 patient rooms with ensuite facilities, consulting and treatment rooms, clean and dirty utility rooms, communal spaces, and facilities for staff and visitors. The ward now has 116 water outlets, including clinical and ensuite wash hand basins, showers, clean and dirty utility sinks, and communal area sinks, plus 32 toilets and 3 sluices.

Throughout this period, all applicable guidance on managing water systems during closures was followed, notably that available in the Scottish Health Technical Memorandum (SHTM) 04–01 (Health Facilities Scotland, 2014a, 2015) and from the UK Health and Safety Executive HSG 274 Part 2 (Health and Safety Executive, 2014). In several respects, measures adopted during this period exceeded what is recommended: flushing of all outlets was carried out daily from October 2020 (rather than 1–2 times per week as recommended), and ClO_2 concentrations were monitored throughout the rest of the building to ensure that adequate levels were being maintained, i.e. that the ClO_2 being drawn through outlets was in the target concentration range of 0.1 to 0.5 mg/l. Reconstruction works were completed in September 2021.

2.2. Water sampling

Post-reconstruction water testing began with a large sampling sweep in September 2021 (Fig. 1). From October 2021, sampling was carried out systematically across the paediatric haemato-oncology ward four

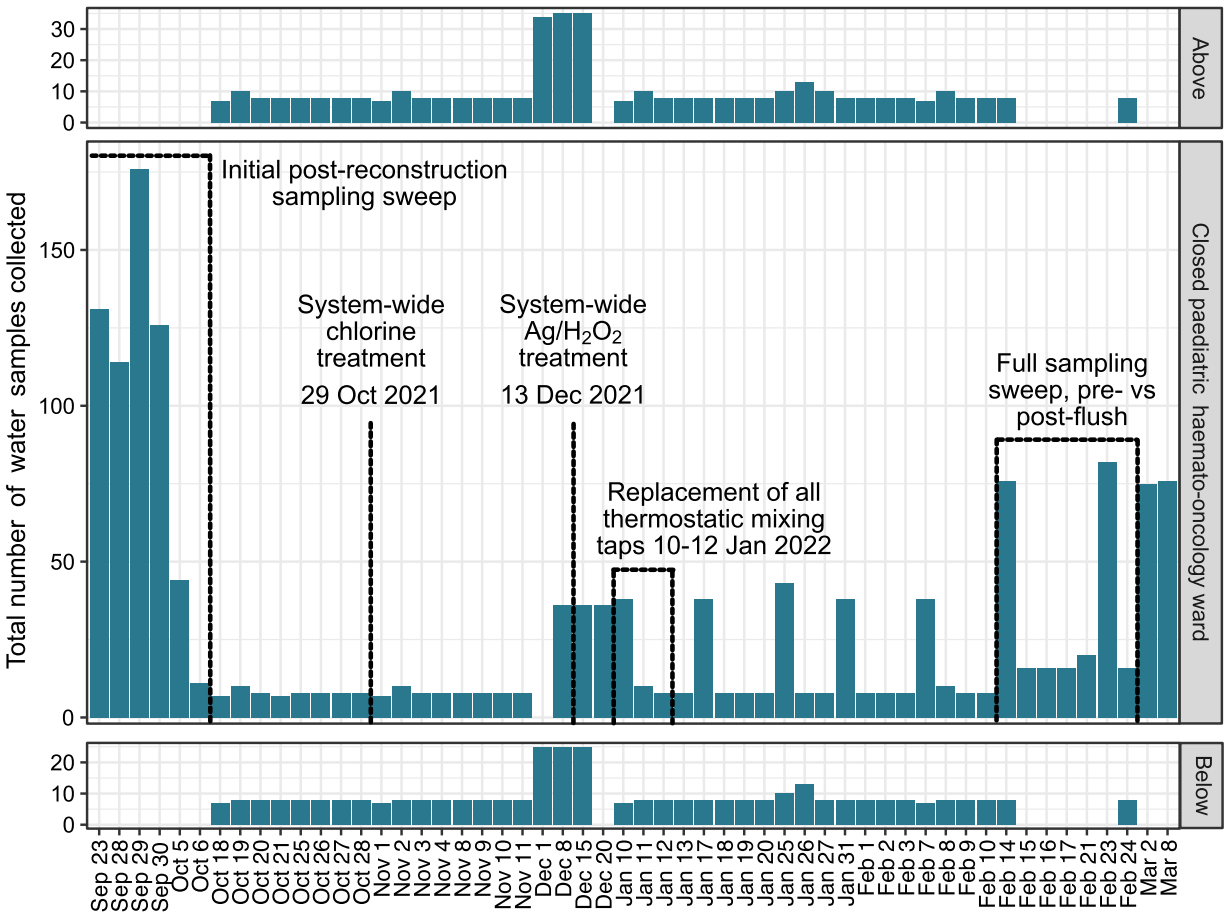

Fig. 1. Timeline of water sampling numbers and water system interventions in the closed paediatric haemato-oncology ward versus on the floors above and below, during the recommissioning period (23 Sept 2021 to 8 Mar 2022). Water samples underwent testing for total viable counts, coliforms, *E. coli, Pseudomonas* spp., Gram negative bacteria, and fungi throughout the entire recommissioning period. Testing for *Legionella* spp. and atypical mycobacteria was carried out during the initial post-reconstruction sampling sweep.

days per week, and in parallel, samples were collected from the floors directly below and above to allow comparison with wards that had remained open and to rule out problems with the wider hospital water system. Water to all these floors is supplied via the same two risers, which feed horizontal loops on each floor.

All 116 water outlets on the ward were included in the sampling programme. The majority ($n = 81$ outlets) are thermostatic mixer taps (Armitage Shanks Markwik 21+) used for handwashing, as these are found in most rooms including treatment rooms, patient bedrooms, patient ensuite facilities, and staff areas. Additional outlet types include separate hot and cold taps installed in utility sinks and in the staff kitchen ($n = 7$) plus a contour mixer tap in a single communal toilet ($n = 1$), and showers found in ensuite facilities for patients and parents ($n = 27$). There are also sampling points on the two risers supplying water to the ward, and these were sampled regularly.

Samples were collected by DMA Canyon Ltd, the water hygiene company contracted by NHS GGC. DMA Canyon use industry standard methods for water sample collection, with protocols specific to the type of outlet and microbiological test requested. Where possible, DMA's protocols aligned with those described in the Scottish Health Technical Memoranda (SHTM) and met the requirements set out in relevant Health and Safety Executive documents specific to Legionella control. However, due to the practicalities of collecting large numbers of samples within a short period of time and for multiple microbiological tests, specific methods statements were produced that reflect the complexities of working in a real-world setting. Most samples collected from September 2021 onwards were 'first flush' rather than true pre-flush samples, as the latter would have required outlets to be placed out of use for several hours prior to collecting samples for each microbiological test – though guidance is vague on how many hours out of use are needed before a sample is considered pre-flush, with statements such as 'at least 2 hours or preferably longer' (Health Protection Scotland, 2018) and 'up to several hours' (British Standards Institution, 2022). In any case, ensuring outlets were not used for several hours prior to sampling was not feasible on a day-to-day basis given staffing pressures, laboratory timings, and flushing schedules. However, in February 2022, a full sweep of true pre-flush and post-flush sampling was carried out, with outlets placed out of use for several hours (at least two) prior to collection of pre-flush samples. No point-of-use (POU) filters were in place during the recommissioning period so samples collected up to early March 2022 were unfiltered. Temperature and ClO_2 concentration were measured at the time of sampling.

Systematic water testing has continued since the ward reopened to patients on 9 March 2022. One-quarter of all outlets are sampled weekly on a rotational basis so that all outlets are tested each month (approximately 140 samples per month, Figure S1). Most samples are collected through 0.2 μm POU filters (Pall) that were fitted to all outlets just prior to reopening, but unfiltered samples are also collected from set locations across the ward at the start of each month to check for proliferation behind the filters (with a new 0.2 μm POU filter then being fitted to those outlets).

2.3. Microbiological tests

All microbiological testing was carried out in the NHS GGC Environmental Laboratory, which is accredited by UKAS (the United Kingdom Accreditation Service) to ISO/IEC 17025:2017 for analysis of potable, endoscopy, and renal waters (Table 1). Water samples collected during the paediatric haemato-oncology ward recommissioning period underwent the following UKAS-accredited tests, using industry standard protocols: total viable counts (TVC) at 22 and 37 °C, coliforms, *Escherichia coli, Pseudomonas* spp., and *Legionella* spp. In addition, the Environmental Laboratory carried out testing specifically for atypical mycobacterial species (AMS), Gram negative bacteria (GNB), and fungi, using protocols that are not yet UKAS accredited for potable waters. AMS testing was carried out during the initial sampling sweep, then implemented routinely after the ward reopened to patients, with one-quarter of outlets tested each month on a rotating basis so that all outlets are tested within each four-month period. Fungal testing occurred throughout the recommissioning period, but since reopening in March 2022, it has been limited to *ad hoc* requests. *Legionella* spp. testing was carried out during the initial sampling sweep only, and since the ward reopened to patients, monthly *Legionella* spp. testing has focused on a small number of sentinel outlets as part of a sweep of samples from across the adult and children's hospitals. Detection of any *Legionella* spp. is exceedingly rare in these buildings, with only 3 samples out of over 9000 from 2021–2024 testing positive for *Legionella* species, not *L. pneumophila* (none of these three positives was from the paediatric haemato-oncology ward nor from any other high-risk clinical area).

TVC tests reported counts per 1 ml, *Legionella* spp. tests reported counts per L, and all other tests reported counts per 100 ml with a reporting ceiling of 100 CFU/100 ml (counts higher than this were recorded as '>100 CFU/100 ml'). The *Legionella* spp. test differentiated between *L. pneumophila* serogroup 1, *L. pneumophila* serogroups 2–14, and other *Legionella* species. The bespoke GNB and fungal tests reported named species based on identification by MALDI-TOF MS (Vitek MS v3, Biomérieux), with the standard Knowledge Base database V3.2, though fungi were often reported to phenotypic groupings (e.g. dematiaceous hyphomycetes) rather than to species level. The *Pseudomonas* spp. test also reported to species level where possible.

2.4. Interventions

Three key interventions were carried out sequentially during the recommissioning period to reduce microbial counts (Fig. 1): i) a system-wide disinfection of the ward's water system on 29 October 2021, using sodium hypochlorite to 30 ppm effective free chlorine for a minimum contact period of 2 hours (pH 7.3), ii) a second system-wide disinfection on 13 December 2021, with silver stabilised hydrogen peroxide at 2000 ppm H_2O_2 for a minimum contact period of one hour, and iii) replacement of all thermostatic mixing taps ($n = 81$) with new taps of the same model (disinfected using 70 % isopropyl alcohol prior to installation), 10–12 January 2022. Both chemical treatments encompassed disinfection of the ward's hot and cold water systems from the risers to all outlets, including the hot water return circuit back to the risers. The chlorine treatment dose was typical of superchlorination levels used to disinfect new pipework, whereas the silver stabilised hydrogen peroxide dose was typical of a 'shock' disinfection treatment used to recover an unfavourable microbiological situation.

In addition to these three interventions that occurred on specific dates, other interventions were implemented over the recommissioning period. Flushing was further increased to reduce stagnation and more closely mimic the water usage of an occupied ward, with approximately 2000 litres of water being moved through the ward's water system during each daily flushing round. Cleaning by domestic staff was intensified, so that by December 2021, the unoccupied ward was being cleaned to clinical standards.

2.5. Data analysis

Microbiological results were reported using established data-sharing protocols. Briefly, the NHS GGC Environmental Laboratory provided test results to DMA Canyon, who collated these with the sampling metadata and disseminated to key NHS GGC personnel in the Water Safety Technical Group, which included representation from Estates, Capital Planning, Microbiology, and Infection Prevention and Control.

Data visualisation and statistical analyses were carried out in R version 4.1.0 (R Core Team, 2022), using tools from the Tidyverse package (Wickham et al., 2019), including ggplot2 (Wickham, 2016). Analyses included comparing the paediatric haemato-oncology ward with floors below and above, measuring the impact of specific

Table 1
Microbiological tests carried out on water samples from the reconstructed paediatric haemato-oncology ward during the recommissioning period and since the ward reopened to patients.

Test	Target organism(s)	Sample volume and processing	Growth conditions	Reported results	Local threshold for high-risk areas
UKAS-accredited tests					
Total viable counts at 22 °C (TVC22) and 37 °C (TVC37)	Heterotrophic bacteria	1 ml, pour plate method	Yeast extract agar, 22 °C for 72 h or 37 °C for 48h	CFU/ml	10 CFU/ml
Coliforms, *E.coli*	Coliforms and *E. coli*	100 ml, filtration method	Chromogenic coliform agar, 37 °C for 18h	Coliforms CFU/100 ml and *E.coli* CFU/100 ml	Any count
Pseudomonas species	*Pseudomonas aeruginosa* and other *Pseudomonas* species	100 ml, filtration method	*Pseudomonas* agar, 37 °C for 48h	CFU/100 ml of all individual *Pseudomonas* species detected, including *P.aeruginosa*	Any count
Legionella spp.	*Legionella pneumophila* and other *Legionella* species	1 L concentrated by filtration to 5 ml, acid/heat treated, 0.1 ml inoculum onto plates	GVPC agar for initial plating, 36 °C for minimum of 10 days, subculture onto BCYE(+), BCYE(-), and CBA for confirmation	CFU/L of *Legionella pneumophila* serogroup 1, L.*pneumophila* serogroups 2–14, and/or *Legionella* species	Any count
Bespoke tests (not on UKAS scope for potable waters)					
Gram negative bacteria (GNB)	*Cupriavidus* species and other Gram negative bacteria	100 ml, filtration method	Tryptic soy agar, 37 °C for 48h	Species names based on MALDI-TOF MS, CFU/100 ml for each detected species	Any count
Atypical mycobacterial species (AMS)	Nontuberculous mycobacteria	100 ml, filtration method	Middlebrooks agar, 35 °C for 42 days	CFU/100 ml	Any count
Fungi: moulds (SAB22), Fungi: yeasts (SAB30)	Fungi (moulds and yeasts)	100 ml, filtration method	Sabouraud agar, 22 °C and 30 °C for 7 days	CFU/100 ml, plus fungal species (where possible) or grouping (e.g. dematiaceous hyphomycetes)	10 CFU/ 100 ml

interventions, comparing the unoccupied paediatric haemato-oncology ward with the ward in the adult hospital where that patient cohort was being cared for during the reconstruction period, and assessing the long-term stability of the system. Microbial count data were assessed using negative binomial generalised linear models as implemented in the MASS package (Venables and Ripley, 2002), and zero inflation was examined using tools from the performance package (Lüdecke et al., 2021). Temperature and ClO_2 data were analysed using linear modelling. Semi-automated scripts were written in R to allow near-real-time visualisation of results and trends for dissemination to the Water Safety Technical Group in weekly meetings.

3. Results

3.1. Results from initial post-reconstruction sampling (September 2021)

Extensive post-reconstruction water testing was carried out across the paediatric haematology-oncology ward from 23 September to 6 October 2021. No *Legionella* spp. ($n = 186$ samples), coliforms ($n = 230$ samples) or *E.coli* ($n = 230$ samples) were detected. Five samples out of 228 (2.2 %) exceeded our local threshold of 10 CFU/100 ml on one or both fungal tests (SAB22 and SAB30), in line with the background rate of detection across the whole hospital (Table 2, Table S1).

However, this sampling sweep detected *Pseudomonas* species in 16 of 229 samples (7 %), from twelve different outlets. Furthermore, it revealed TVCs, Gram negative bacteria (GNB) counts, and atypical mycobacterial species (AMS) counts exceeding our local thresholds from outlets across the ward (Table 2, Table S1), with over 26 % of outlets exceeding the TVC22 threshold, 21.6 % exceeding the AMS threshold, and 56 % (65 out of 116 outlets) exceeding the GNB threshold. GNB tests, in particular, had a high sample positivity rate (169 of 231 samples, i.e. 73.2 %), as well as high abundances in those positive samples, with 90 samples (53.3 % of positives, or 39.0 % overall) having over 100 CFU/100 ml, the reporting ceiling for this test. Fourteen bacterial species were detected by GNB testing, the most common being *Cupriavidus pauculus* (63 samples), *Blastomonas ursincola* (58 samples), *Sphingomonas paucimobilis* (46 samples), *Acidovorax temperans* (18 samples), and *Acidovorax delafieldii* (12 samples).

3.2. Comparison with floors above and below (October 2021)

From 18 October onwards, testing was expanded to include the floors below and above the paediatric haemato-oncology ward. This confirmed that the refurbished ward had significantly higher TVC results than floors below and above (negative binomial regression, TVC22: $\chi2(2, n = 192) = 1002.7$, $p < 0.0001$ and TVC37: $\chi2(2, n = 192) = 645.45$, $p < 0.0001$, Fig. 2A and 2B).

Temperature and ClO_2 levels in these samples were comparable among all floors, with no significant differences (linear regression ANOVA, temperature: $F(2186) = 1.3608$, $p = 0.259$, ClO_2: $F(2189) = 0.1831$, $p = 0.8328$, Fig. 2C and 2D). Temperature data showed clear delineation of cold, mixed (i.e. through a thermostatic mixing valve),

Table 2
Water test results from the paediatric haemato-oncology ward, which was closed for reconstruction and had been largely unoccupied since September 2018 apart from a brief time in early 2019. Three distinct periods are shown: i) initial post-reconstruction testing sweep (23 September to 6 October 2021), ii) after recommissioning but before the ward reopened to patients (13 January to 8 March 2022), and iii) since reopening (9 March 2022 to 23 April 2024). Point-of-use (POU) filters (0.2 μm) were fitted to all 116 outlets just prior to reopening, but unfiltered samples are regularly collected from 14 representative outlets prior to a new filter being fitted.

Test[1]	POU-filtered (0.2 μm) or unfiltered outlet	Initial testing sweep 23 Sept-6 Oct 2021		Pre-opening, after recommissioning 13 Jan-8 Mar 2022		Post-opening 9 Mar 2022–23 Apr 2024	
		Samples out of spec[2]	Outlets out of spec[2]	Samples out of spec[2]	Outlets out of spec[2]	Samples out of spec[2]	Outlets out of spec[2]
Total viable counts at 22 °C (TVC22)	Unfiltered outlet	47/231 (20.3 %)	31/116 (26.7 %)	1/648 (0.15 %)	1/116 (0.86 %)	1/169 (0.59 %)	1/14* (7.1 %)
	POU-filtered outlet					8/3316 (0.24 %)	8/116 (6.9 %)
Total viable counts at 37 °C (TVC37)	Unfiltered outlet	35/231 (15.2 %)	24/116 (20.7 %)	3/648 (0.46 %)	2/116 (1.7 %)	7/169 (4.1 %)	2/14* (14.3 %)
	POU-filtered outlet					14/3316 (0.42 %)	13/116 (11.2 %)
Coliforms and *E. coli*	Unfiltered outlet	0/230 (0 %)	0/116 (0 %)	0/648 (0 %)	0/116 (0 %)	0/167 (0 %)	0/14 (0 %)
	POU-filtered outlet					0/3251 (0 %)	0/116 (0 %)
Pseudomonas species	Unfiltered outlet	16/229 (7.0 %)	12/116 (10.3 %)	0/648 (0 %)	0/116 (0 %)	0/169 (0 %)	0/14 (0 %)
	POU-filtered outlet					4/3262 (0.12 %)	4/116 (3.4 %)
Gram negative bacteria	Unfiltered outlet	169/231 (73.2 %)	65/116 (56.0 %)	47/648 (7.3 %)	14/116(12.1 %)	3/124 (2.4 %)	2/14* (14.3 %)
	POU-filtered outlet					13/3357 (0.39 %)	9/116 (7.8 %)
Atypical mycobacterial species	Unfiltered outlet	38/141 (27.0 %)	25/116 (21.6 %)			34/124 (27.4 %)	3/14* (21.4 %)
	POU-filtered outlet					4/615 (0.65 %)	4/116 (3.4 %)
Legionella spp.	Unfiltered outlet	0/186 (0 %)	0/116 (0 %)				
Fungi: moulds (SAB22)	Unfiltered outlet	5/227 (2.2 %)	5/116 (4.3 %)	1/634 (0.16 %)	1/116 (0.86 %)		
Fungi: yeasts (SAB30)	Unfiltered outlet	4/228 (1.8 %)	4/116 (3.4 %)	0/632 (0 %)	0/116 (0 %)		

[1] TVC = total viable count (at 22 and 37 °C). Sample volume varies by test: CFU/L for Legionella, CFU/ml for TVC22 and TVC37, CFU/100 ml for all other tests.
[2] Thresholds for out of specification results were set locally and vary by test: 10 CFU/ml for both TVC tests, 10 CFU/100 ml for both fungal tests, and any count on the other tests.
* Unfiltered TVC, GNB and AMS out of spec results were all from the same two outlets (a domestic services room and a utility sink in a communal area), apart from a third outlet in a different domestic services room that tested positive once for AMS at 1 CFU/100 ml.

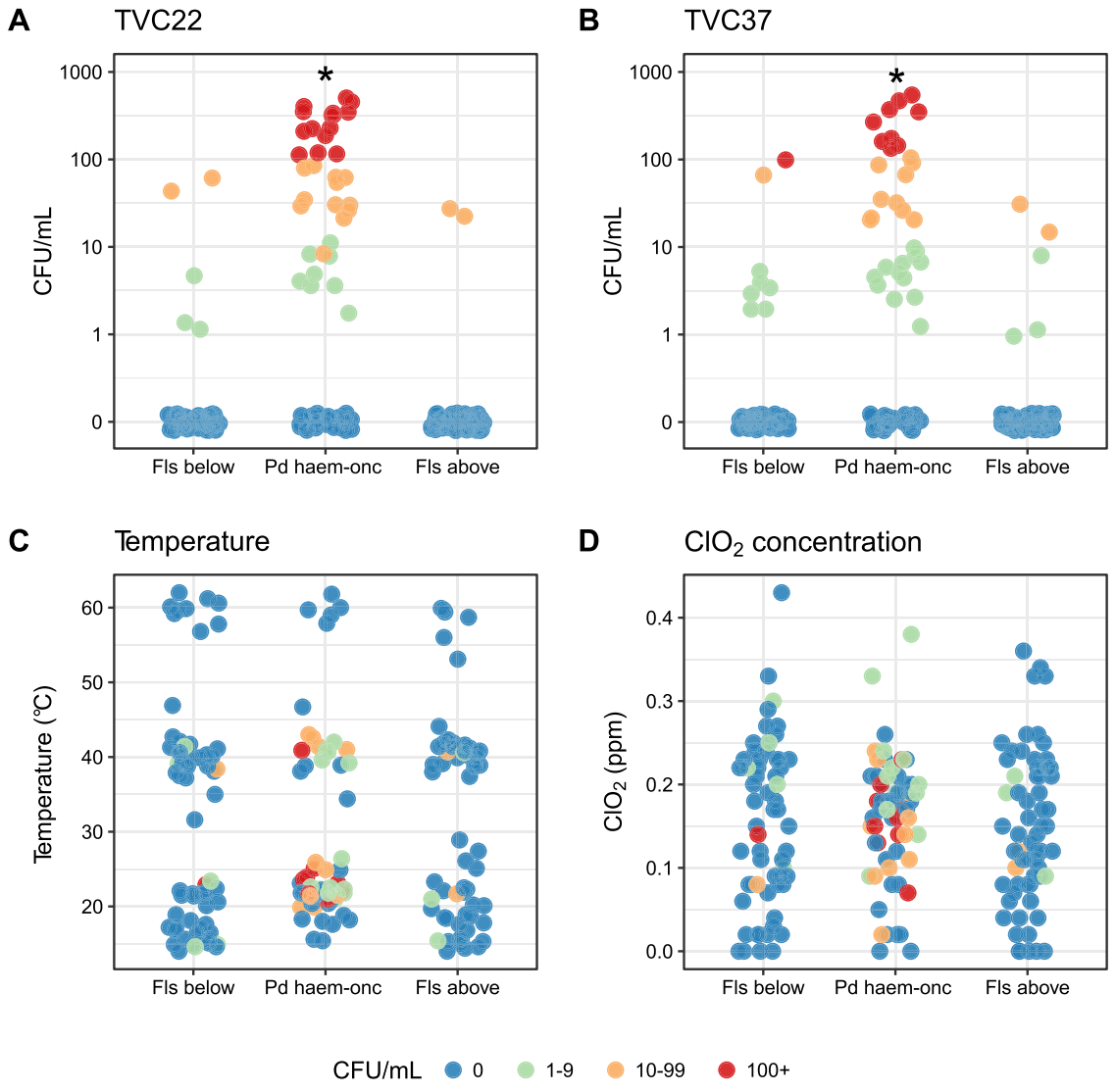

Fig. 2. Water testing results from reconstructed paediatric haemato-oncolgy ward ($n = 64$) compared with floors below ($n = 63$) and above ($n = 65$). Samples were collected 18–28 October 2021 prior to the first chemical treatment. Panels show total viable counts at 22 °C (A) and 37 °C (B), in colony-forming units (CFU) per ml, (C) sample temperature at time of collection, and (D) chlorine dioxide concentration in ppm. Points in panels (C) and (D) are coloured by the TVC37 result. Asterisks show where paediatric haemato-oncology samples were significantly different from the other floors ($p < 0.001$).

and hot water samples. All mixed and hot samples reached their expected temperature, indicating that the temperature-based controls in the system were working. ClO_2 concentrations at the tap averaged 0.15 ppm, ranging from <0.02 (the lower limit of detection) to 0.43 ppm. In the paediatric haemato-oncology ward, samples with elevated TVCs were from cold and mixed outlets, and spanned the full range of observed ClO_2 concentrations. Taking into account the influence of location (i.e. whether from the closed paediatric haemato-oncology ward or from the floors below/above), ClO_2 concentration at the time of sampling did not have a significant effect on TVC37 ($\chi2(1, n = 192) = 1.54$, $p = 0.214$), though it did have a significant effect on TVC22 ($\chi2(1, n = 192) = 4.76$, $p = 0.029$), with higher ClO_2 concentrations resulting in lower TVC22 counts.

Testing specifically for Gram negative bacteria over this period showed that three species, *Cupriavidus pauculus, Sphingomonas paucimobilis* and *Acidovorax temperans*, appeared enriched in samples from the unoccupied paediatric haemato-oncology ward compared with the occupied floors below and above (Fig. 3). Not only were they detected in more samples (31 of 64, compared with 7 of 63 from the floors below and none of the 65 samples from the floors above), but their counts per sample exceeded the reporting ceiling (>100CFU/100 ml) in all but two instances. Two other bacterial species, *Blastomonas ursincola* and *Acidovorax delafieldii*, were also frequently detected. However, contemporaneous sampling on floors below and above showed that these taxa are common across all sampled areas, though the counts of *B. ursincola* were all at the reporting ceiling in the closed paediatric haemato-oncology ward but more often lower on the other floors.

3.3. Impacts of specific interventions

Throughout the recommissioning period, flushing and cleaning were increased to more closely mimic an occupied ward, and routine ClO_2 supplementation of the entire hospital's water supply was maintained. In addition, three specific interventions were carried out sequentially to address the continuing detection of high bacterial counts: a chlorine treatment (sodium hypochlorite) on 29 October 2021, a silver stabilised hydrogen peroxide treatment on 13 December 2021, and finally, replacement of all thermostatic mixing taps on January 10–12. The impacts of these three consecutive interventions on TVC22 and TVC37 are shown in Fig. 4.

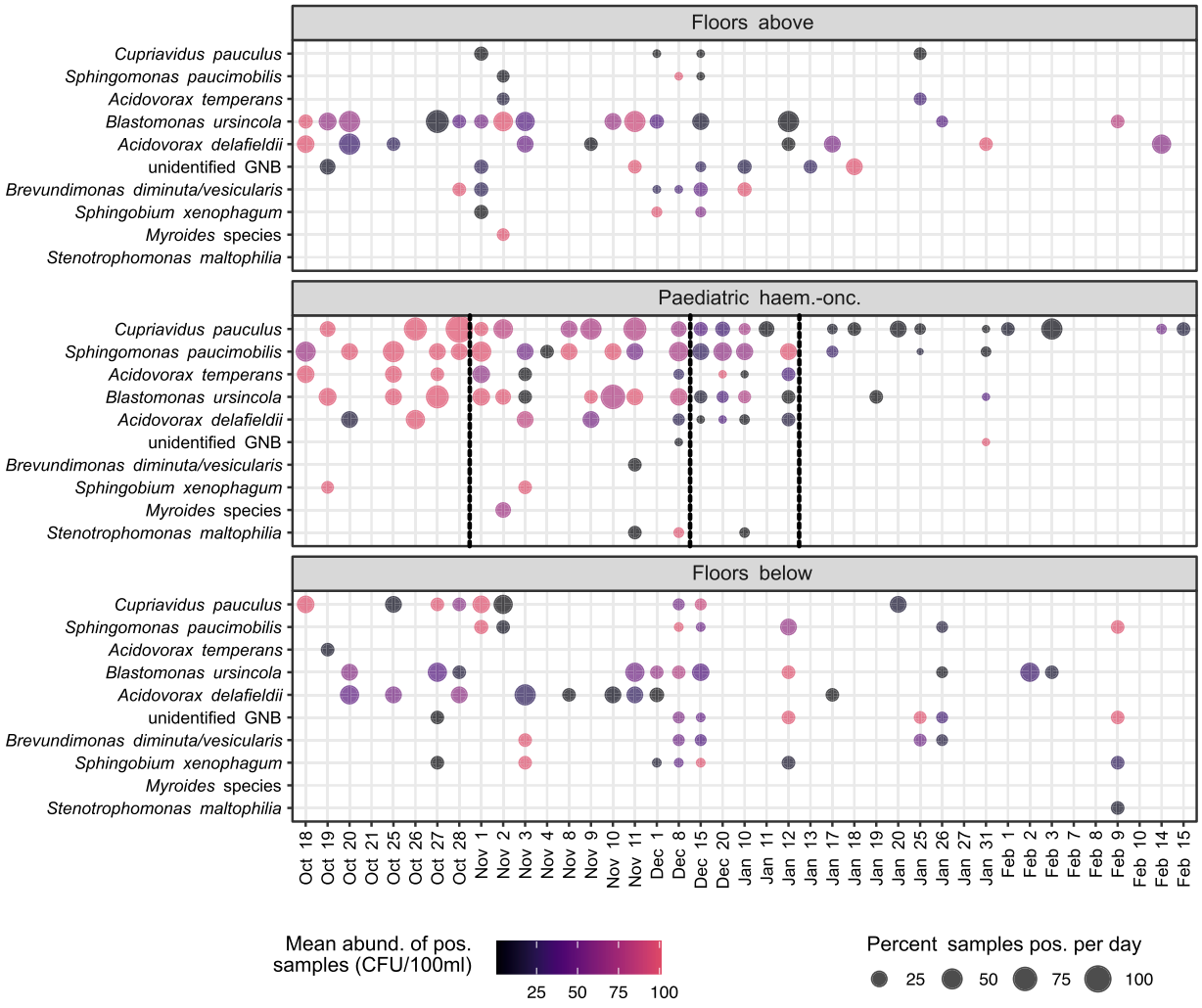

Fig. 3. Main Gram negative bacteria detected over the pre-opening period (18 Oct 2021 to 15 Feb 2022) in unfiltered water samples from the reconstructed paediatric haemato-oncology ward and from the floors immediately below and above. Dates of the three interventions (chlorine treatment, silver stabilised hydrogen peroxide treatment, tap replacement) are shown with black lines. Symbol size indicates the percent of samples collected that day that tested positive for the listed organism, and colour indicates the mean count (in CFU/100 ml) of the positive samples. For clarity, only taxa detected in five or more samples are shown here. The full list of taxa detected in the paediatric haemato-oncology ward over the study period is shown in Figure S3.

After the first chemical treatment with chlorine on 29 October 2021, there was no significant change in TVCs when comparing counts in samples collected in the period preceding treatment (2–28 October) to those in samples collected following treatment (30 October to 12 December) (negative binomial regression, TVC22: $\chi2(1, n = 213) = 0.00034$, $p = 0.985$ and TVC37: $\chi2(1, n = 213) = 1.210$, $p = 0.271$, Fig. 4A). There was also no clear decrease in *Cupriavidus pauculus* or *Sphingomonas paucimobilis*, though *Acidovorax temperans* decreased in prevalence and abundance after this treatment (Fig. 3).

The second chemical treatment, silver stabilised hydrogen peroxide on 13 December 2021, resulted in a significant decrease in TVCs when comparing counts in the period preceding treatment (8 November to 12 December) with those in the period following treatment (14 December to 9 January) (negative binomial regression, TVC22: $\chi2(1, n = 140) = 7.955$, $p = 0.0048$ and TVC37: $\chi2(1, n = 140) = 11.58$, $p = 0.00067$, Fig. 4B). Before this treatment, 42.6 % of samples (29 out of 68) exceeded our local TVC threshold of 10 CFU/ml on one or both TVC tests, whereas after treatment, this dropped to 13.9 % (10 out of 72 samples). Prevalence and abundance of *Cupriavidus pauculus* and *Acidovorax temperans* decreased after this treatment, though *Sphingomonas paucimobilis* appeared unaffected (Fig. 3).

The final intervention, replacing all 81 thermostatic mixing taps on the ward on 10–12 January 2022, resulted in significantly lower counts on both TVC tests when comparing samples collected through old versus new taps (negative binomial regression, TVC22: $\chi2(1, n = 196) = 66.95$, $p < 0.0001$ and TVC37: $\chi2(1, n = 196) = 63.06$, $p < 0.0001$, Fig. 4C), with none of the 110 samples subsequently taken from new taps up to 28 January 2022 exceeding our local threshold of 10 CFU/ml on either test. *Acidovorax temperans* was not detected at all after the taps were replaced. *Sphingomonas paucimobilis* was detected in only 3 out of 110 samples, all with counts below 15 CFU/100 ml, and *Cupriavidus pauculus* was detected in 5 out of 110 samples, with a maximum abundance of 6 CFU/100 ml (versus being detected in 11 out of 86 samples from the period immediately prior to tap replacement, with 9 of these exceeding 10 CFU/100 ml and 3 reaching the reporting ceiling of >100 CFU/100 ml). Detection of other Gram negative taxa that were previously common across all sampled floors also decreased after tap replacement (*Blastomonas ursincola, Acidovorax delafieldii*, unidentified GNB), but these continued to be detected on the floors below and above (Fig. 3).

The cumulative impact of these three interventions on TVCs, along with additional measures that were put in place over this period, notably increased flushing and cleaning to clinical standards, is shown in Fig. 5 and Table 2. From 13 January 2022, after all taps had been replaced, until just before opening (8 March 2022), TVCs in the paediatric

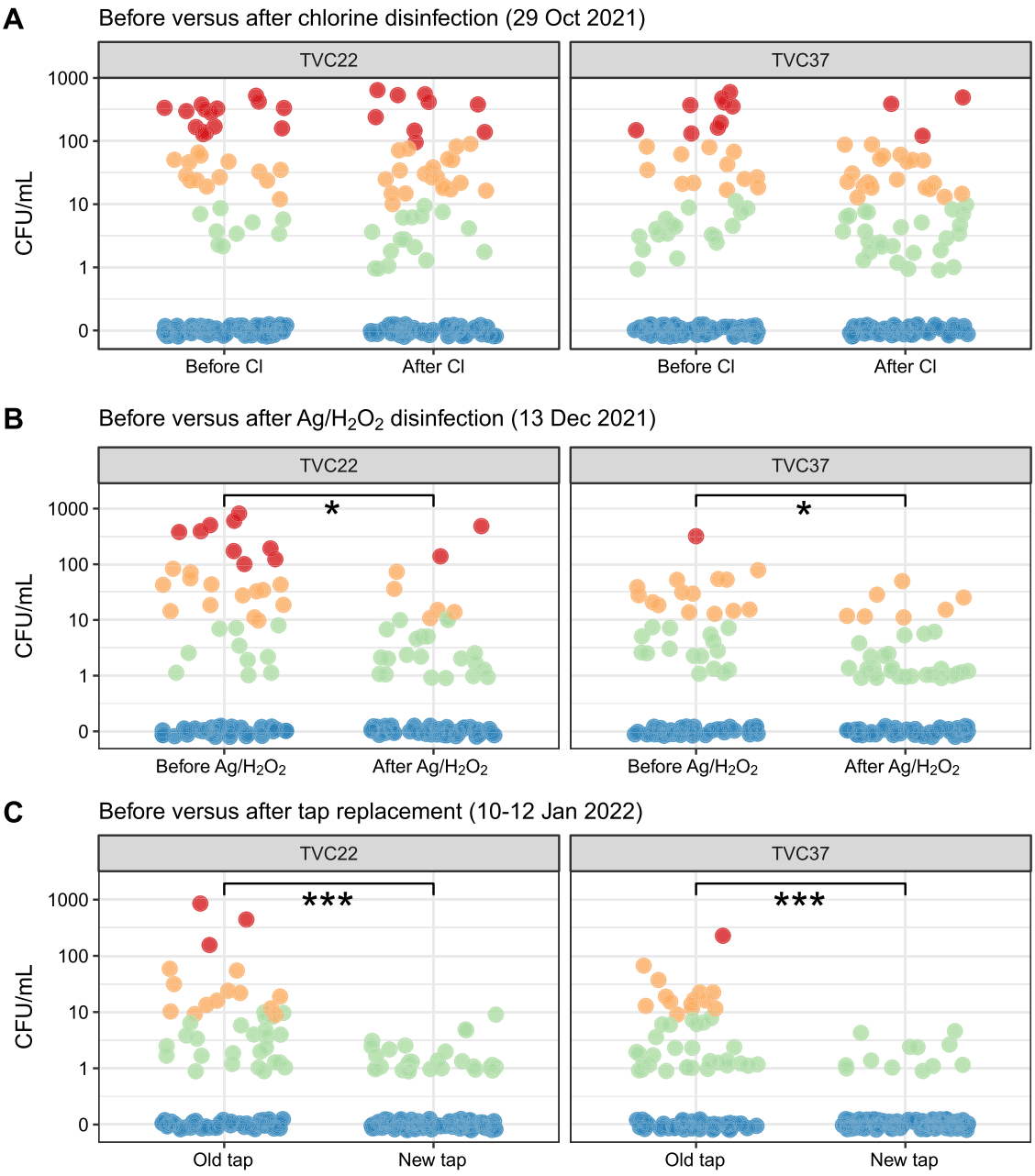

Fig. 4. Impacts of specific interventions on TVC22 and TVC37 results in the reconstructed paediatric haemato-oncology ward. (A) Before (n = 112) and after (n = 101) chlorine treatment, with samples collected 2 Oct to 12 Dec 2021, (B) before (n = 68) and after (n = 72) silver stabilised hydrogen peroxide treatment, with samples collected 8 Nov 2021 to 9 Jan 2022, and (C) before (n = 86) and after (n = 110) replacement of all thermostatic mixing taps on the ward (81 of 116 outlets), with samples collected 14 Dec 2021 to 28 Jan 2022. Only samples collected through old and new thermostatic mixing taps are included in the last comparison. As the interventions were carried out sequentially, the post-treatment group of one intervention becomes the pre-treatment group of the next so there is some overlap across panels. See Supplemental Methods for details on sample inclusion. Significant differences are indicated by asterisks (negative binomial GLM, * $p < 0.01$, *** $p < 0.0001$).

haemato-oncology ward were significantly lower than on the occupied floors below and above (negative binomial regression, TVC22: $\chi2(2, n = 952) = 113.5$, $p < 0.0001$ and TVC37: $\chi2(2, n = 952) = 86.643$, $p < 0.0001$). Furthermore, these TVCs, all from unfiltered samples, were significantly lower than those from filtered samples collected in the adult hospital ward where the paediatric haemato-oncology patients were being cared for during the reconstruction period (negative binomial regression, TVC22: $\chi2(1, n = 1040) = 63.51$, $p < 0.0001$ and TVC37: $\chi2(1, n = 1040) = 48.18$, $p < 0.0001$), despite this occupied ward having 0.2 μm POU filters installed on all outlets (Figure S2A).

3.4. Additional tests and results

Throughout the recommissioning period (September 2021 to early March 2022), no sample tested positive for coliforms or *E. coli*, and no *Legionella* spp. were detected in the initial sampling sweep in September 2021. *Pseudomonas* spp. were not detected after 11 November 2021. In the months prior to reopening (13 January to early March 2022), only one fungal test out of 634 (0.16 %) exceeded our local threshold of 10 CFU/100 ml (Table 2), compared with around 2 % of samples tested in the initial September 2021 sampling round.

In mid-February 2022, true pre-flush (n = 120) as well as post-flush (n = 120) samples were collected from all outlets on the ward

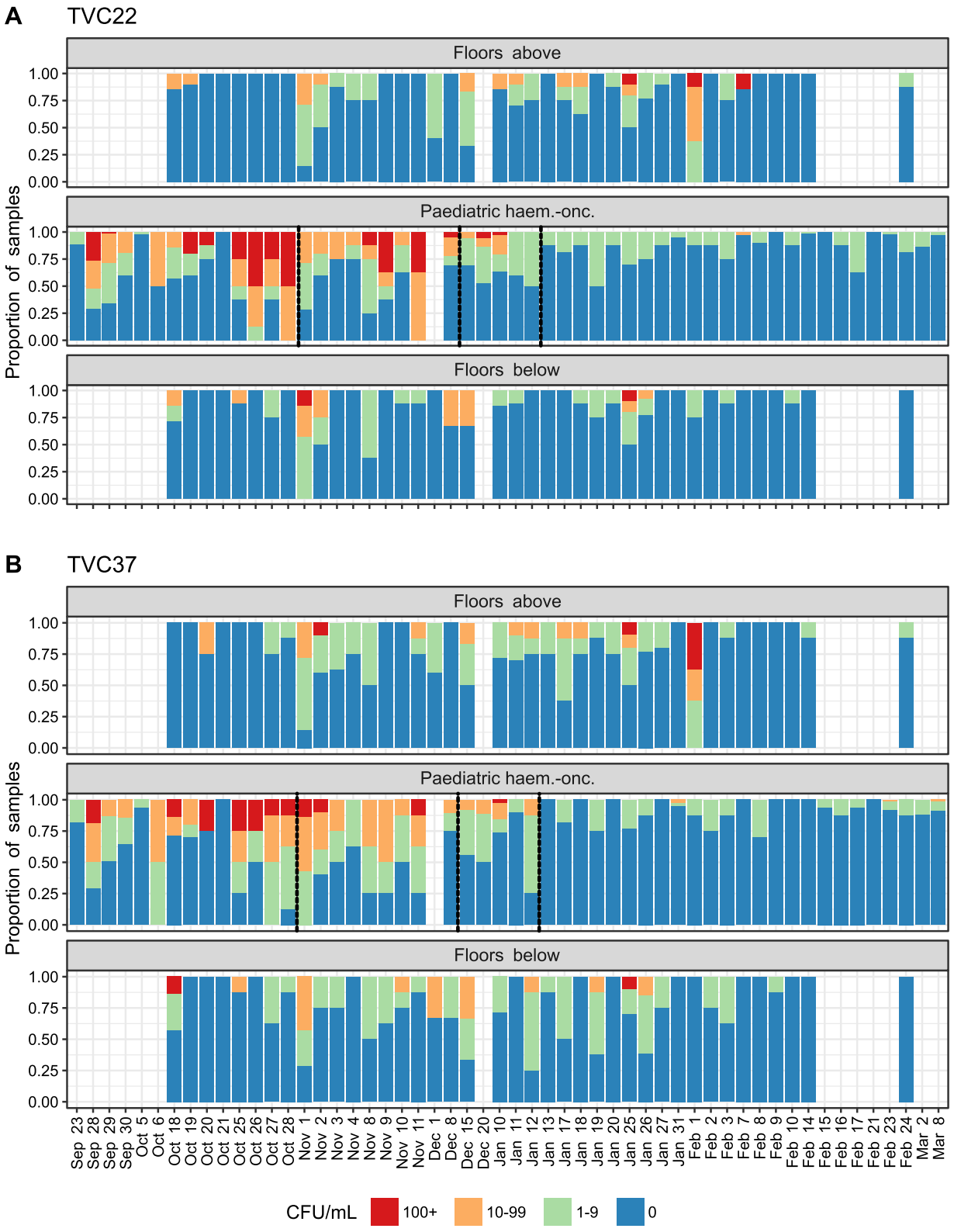

Fig. 5. TVC results for the entire pre-opening period, 23 Sept 2021 to 8 Mar 2022, showing the cumulative impact of the three interventions and other measures taken to reduce water microbial counts in the reconstructed paediatric haemato-oncology ward to the levels seen on floors above and below. Timings of the three interventions (chlorine treatment, silver stabilised hydrogen peroxide treatment, thermostatic mixing tap replacement) are shown with black lines. Blanks indicate days when no samples were collected from that floor.

(Figure S2B). Most outlets had zero counts in both pre-flush samples (TVC22: 114/120 samples, TVC37: 115/120 samples) and post-flush samples (TVC22: 113/120 samples, TVC37: 113/120 samples), and in the rare instances where counts were detected, they were generally 1 or 2 CFU/ml. Only one pre-flush sample (0.83 % of pre-flush samples, or 0.42 % overall) exceeded the 10 CFU/ml local threshold on TVC37, and none exceeded this threshold on TVC22.

3.5. Long-term stability

The ward was reopened to patients on 9 March 2022, with 0.2 μm POU filters fitted to all outlets just prior to occupation. Routine, systematic water testing continues to the present day (Figure S1), and two years of post-opening data, including over 3000 samples, shows no return of the high TVC and Gram negative bacterial counts observed in the immediate post-reconstruction period (Fig. 6, Table 2).

In general, there is no evidence of microbial proliferation behind the 0.2 μm POU filters, as unfiltered samples had low or no counts in most cases (Fig. 6, Table 2). Only two outlets repeatedly tested positive on one or more tests (TVC, GNB, AMS) when samples were collected without POU filters: a utility sink in one of the domestic services rooms, and a utility sink in a communal social space. These two outlets accounted for all but one of the out-of-specification results with unfiltered samples. Samples taken from these two outlets but with their 0.2 μm POU filters in place had no detectable GNB or AMS, and only one instance of elevated TVCs (in the domestic services room).

Samples taken through 0.2 μm POU filters had a low positivity rate overall, <1 %, on all microbiological tests (Table 2). While the unfiltered positive results were almost exclusively from two outlets, positive results from filtered outlets occurred sporadically across the ward, with outlets rarely testing positive more than once (e.g. eight filtered samples from eight different outlets were out of specification on TVC22, 14 filtered samples from 13 different outlets were out of specification on TVC37, and 13 filtered samples from 9 different outlets had detectable GNB – Table 2). Additional measures triggered whenever a filtered sample exceeded our local thresholds included sampling without the filter and then sampling through a new filter, and in every case, these subsequent samples were within our strict thresholds.

The range of GNB species detected changed once 0.2 μm POU filters were fitted and the ward was reopened to patients (Fig. 7, Figure S3), and detection was sporadic across both time and space. While *Blastomonas ursincola, Cupriavidus pauculus, Sphingomonas paucimobilis*, and

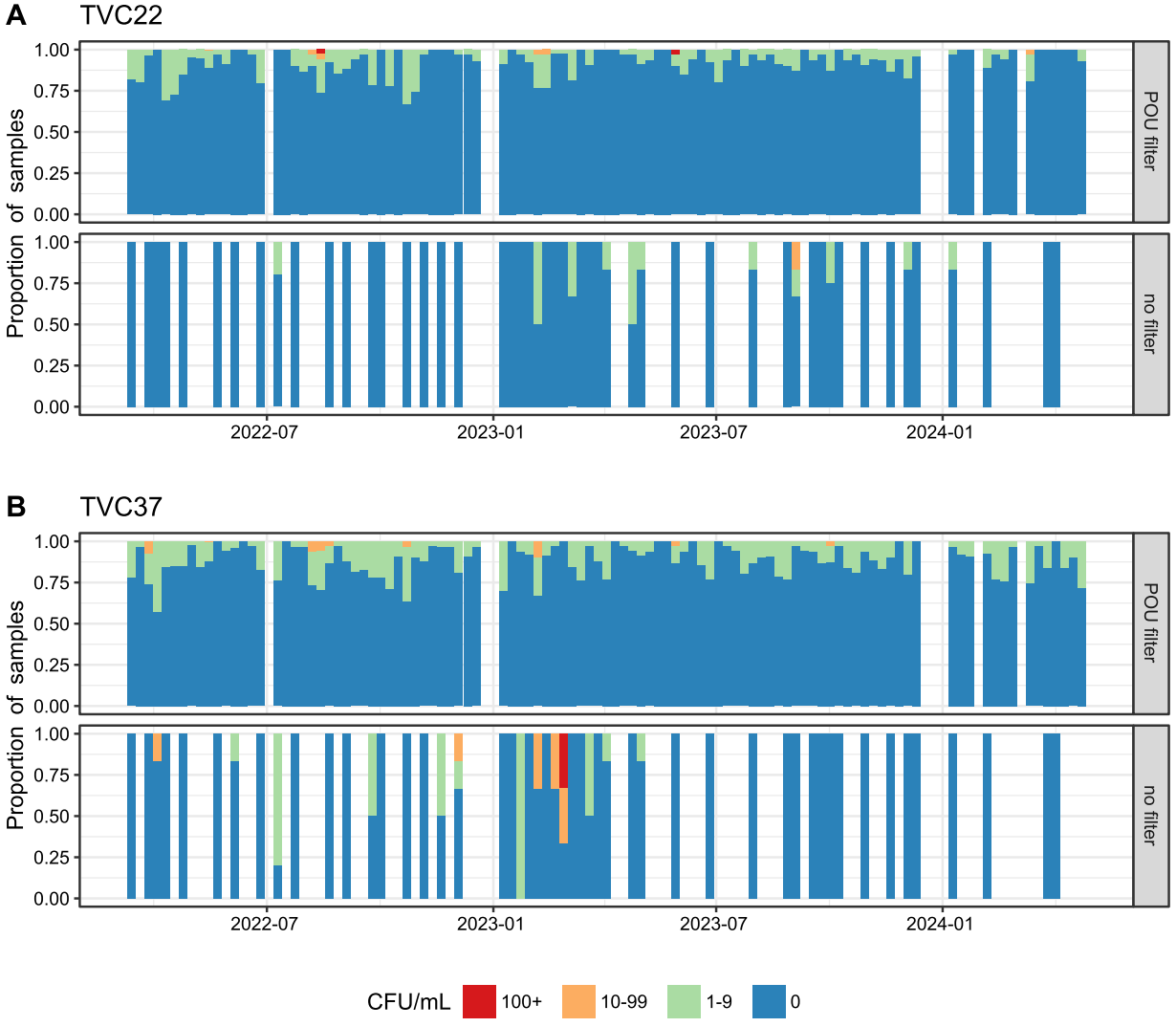

Fig. 6. TVC results for the entire post-opening period in the reconstructed paediatric haemato-oncology ward, 9 Mar 2022 to 23 Apr 2024, grouped by week. Water samples were routinely collected through 0.2 μm point-of-use filters installed on all outlets in the ward, with a subset collected without filters in place. Approximately 30–35 samples were collected per week on a rotating basis (see Supplemental Information for weekly sample numbers). Blanks indicate weeks when no samples were collected.

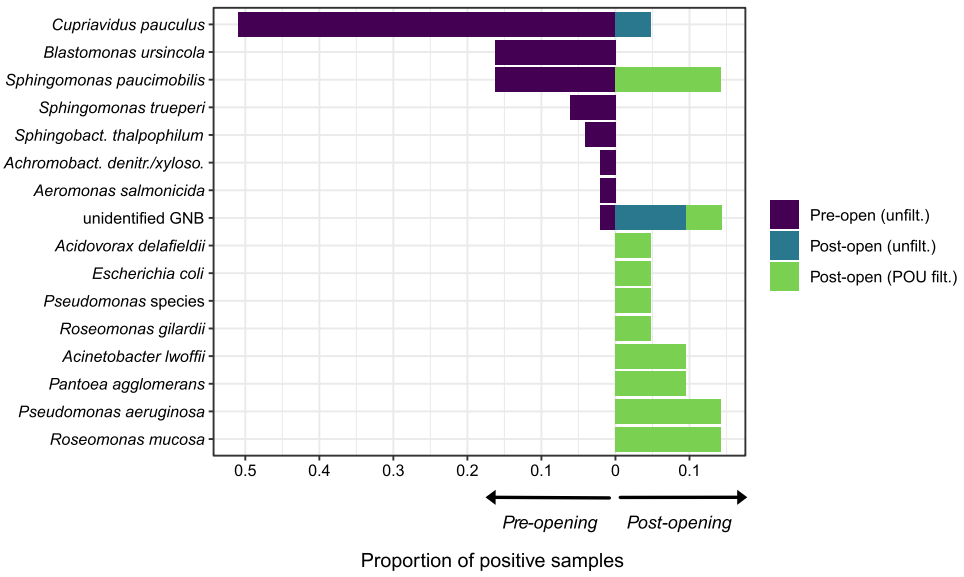

Fig. 7. Taxonomic profile of bacterial species reported from Gram negative and *Pseudomonas* spp. tests in the pre-opening period versus the post-opening period after POU filters were fitted and patients returned to the ward. Pre-opening period, 13 Jan to 8 Mar 2022: 49 organisms detected from 47 unfiltered water samples, out of 648 samples tested for both GNB and *Pseudomonas* spp. Post-opening period, 9 Mar 2022 to 23 Apr 2024: 21 organisms detected from 3 unfiltered and 17 filtered samples, out of 124 unfiltered GNB tests, 3357 filtered GNB tests, 169 unfiltered *Pseudomonas* spp. tests and 3262 filtered *Pseudomonas* spp. tests. Proportions post-opening are calculated using the total number of positives (inclusive of unfiltered and filtered).

Acidovorax spp. were the dominant species in unfiltered samples from the recommissioning period, of the three unfiltered samples that tested positive for GNB since the ward reopened (out of 124 unfiltered samples tested), only one had *Cupriavidus pauculus*, while the other two had unidentified GNB. With samples collected through 0.2 μm POU filters, the thirteen that tested positive (out of 3357 samples) encompassed seven named species plus unidentified GNB (Fig. 7). *Sphingomonas paucimobilis* was detected in three samples, with one of these also reporting *Acidovorax delafieldii*, but the remaining GNB-positive samples reported taxa that were rarely or never detected prior to the ward reopening, notably *Acinetobacter lwoffii, E. coli, Pantoea agglomerans, Roseomonas gilardii*, and *Roseomonas mucosa*. Four filtered samples also had detectable *Pseudomonas* species (three with *P. aeruginosa*, one with *Pseudomonas* sp.). No *Pseudomonas* spp. had been detected in any unfiltered sample since mid-November 2021, prior to the second disinfection and subsequent tap replacement.

4. Discussion

In any engineered water distribution system, periods of decreased or interrupted water use can lead to proliferation of microbial flora, especially in systems that might have pre-existing biofilm. When the Royal Hospital for Children's paediatric haemato-oncology ward was closed for several years to allow extensive reconstruction work, all relevant guidance on maintaining a water system during closure periods was followed, and in many respects, exceeded. In particular, considerable effort was expended on daily flushing of all 116 outlets to replenish disinfectant levels throughout the system, to maintain hot and cold temperatures, and to prevent stagnation, which is well known to cause deterioration of water quality in buildings (Proctor et al., 2020; Ye et al., 2022).

Nonetheless, post-reconstruction water testing showed some microbial proliferation across the ward. It is not clear what could have been done differently during the reconstruction stage to prevent this, as we did not have capacity to carry out flushing of 116 outlets across a building site more than once per day. Pre-existing biofilm could have exacerbated proliferation issues, and this ward was investigated for microbial problems prior to the closure, but the reported detection rate of Gram negative bacteria in water samples at the time, 76.5 % of outlets on the ward and approximately 30 % of samples from across the hospital (Inkster et al., 2021), is comparable to the positivity rate found at other UK hospitals when the same bespoke Gram negative bacterial test was used (Inkster et al., 2022), suggesting that other sites may have a similar baseline bacterial load and that closures could have similar repercussions. Our experience suggests that for extended ward closures (i.e. on the scale of years), some microbial proliferation is to be anticipated, and an intensive recommissioning period may be required to return the water system to an acceptable state. In our case, recommissioning the water system so that it consistently met our strict local microbiological thresholds for high-risk areas took over five months and delayed the ward reopening to patients.

4.1. Multidisciplinary approach

In our health board, management of hospital water systems and associated risks is overseen by a multidisciplinary Water Safety Group, as recommended in applicable guidance (Health Facilities Scotland, 2014a). In addition, a specific sub-group, the Water Safety Technical Group (WSTG), was convened to carry out the recommissioning of the water system in the reconstructed paediatric haemato-oncology ward. The WSTG was a multidisciplinary team that included key personnel from Estates and Facilities, Infection Prevention and Control, Microbiology, Capital Planning, as well as an Authorising Engineer for water, the head of our external water hygiene contractor, independent water safety advisors, and a healthcare scientist, who set up semi-automated, near-real-time visualisation and analysis of water testing results and trends for dissemination to the project team in weekly meetings. Involving experts from multiple disciplines and ensuring effective communication of results across these disciplines are two key tenets of the holistic approach to water-associated pathogen management advocated by Proctor et al. (2022), and we found both to be essential in resolving the situation we were facing.

4.2. Delineating the problem: microbiological tests

Although we relied only on culture-based testing, which underestimates the true extent of microbial diversity in water compared with molecular approaches (Scaturro et al., 2023; Wang et al., 2017), the

suite of tests that we carried out during the recommissioning period proved helpful in delineating the problem. Targeting multiple microorganisms simultaneously is a third tenet of the recommended holistic approach to water management (Proctor et al., 2022), and our experience supports this approach when dealing with microbial proliferation concerns in high-risk hospital wards. *Legionella* spp. testing alone would not have identified any microbial proliferation, as none were detected during the initial sampling sweep. *Pseudomonas* spp. testing would have underestimated the extent of the issue, and we might have focused on the relatively small number of outlets that had tested positive in the initial post-reconstruction sweep rather than working across the entire ward. Testing for atypical mycobacterial species showed that microbial proliferation had occurred across a relatively large number of outlets, but the extended incubation required for this test (42 days) limited its use in assessing interventions during the recommissioning period.

TVC testing (i.e. heterotrophic plate counts) proved to be a good general indicator, and since it is a well-established method with standardised, internationally validated protocols (Bartram et al., 2003; International Organization for Standardization, 1999; Lipps et al., 2023; Standing Committee of Analysts, 2020), it should be readily available to health boards looking to carry out water testing. GNB (Gram negative bacteria) testing also suggested widespread microbial proliferation in our system, with 73.2 % of samples from the initial post-reconstruction sweep testing positive. However, this is a bespoke method set up specifically for our hospital site in 2018, and in the absence of any established baselines or interpretive criteria, our local threshold was set to zero at that time (i.e. any GNB count per 100 ml is deemed out of specification). Given the larger sample volume (100 ml) compared with standard TVC tests (1 ml) and the diversity of normal, expected water flora, this threshold may be too strict and is somewhat at odds with our local high-risk TVC thresholds of 10 CFU/ml (which are also unusually strict), since many organisms detected in TVC tests will be Gram negative bacteria. Indeed, when this bespoke GNB test was trialed in ten other UK hospitals, the sample positivity rate was 63 % (i.e. 99 water samples out of 157 had detectable GNB), yielding 31 different named bacterial species (Inkster et al., 2022). In our experience, GNB testing was most informative when used similarly to TVC testing, i.e. not to interpret individual samples in isolation according to an arbitrary threshold but to assess spatial and temporal trends, both in total GNB counts and also in the bacterial species detected. However, this required data analysis and environmental microbiology expertise that might not be available to all boards. While we continue to carry out GNB testing routinely in this high-risk ward to provide reassurance, we would hesitate to recommend this type of testing more broadly given the complexities of interpreting the results in the absence of baseline data and the difficulty in translating these results into an assessment of clinical risk.

4.3. Delineating the problem: location, temperature, and ClO2

Tap water is not sterile, so some counts are expected in non-selective microbiological tests. Only analysis of spatial and temporal trends can distinguish true microbial proliferation from sporadic detection and background variability. By sampling intensively not only across the reconstructed ward but also across the floors below and above, which were fed from the same risers, we confirmed that the microbial proliferation was localised to the reconstructed ward and was present across it. Ruling out problems with temperature controls and chlorine dioxide distribution was also crucial. Temperature and chlorine dioxide profiles were similar across all floors. While ClO_2 concentration, an indicator of whether water is being drawn through outlets or is allowed to stagnate, did significantly influence TVC22 counts, it had no significant effect on TVC37 results. Short-term stagnation and proliferation within outlets between scheduled flushing events could therefore not entirely explain the counts we were seeing, since variability in TVC37 results was not explained by ClO_2 concentration.

4.4. Addressing the problem: interventions

Once we had established that this closed ward had higher TVCs and some more abundant Gram negative taxa compared with the other floors, we carried out two ward-wide chemical treatments. The first treatment, with chlorine (sodium hypochlorite), did not significantly reduce TVCs nor did it have a clear impact on the Gram negative bacterial taxa detected, apart from a reduction in the prevalence and abundance of *Acidovorax temperans*. These hospital buildings (QEUH Adults and RHC) have had on-site ClO_2 supplementation since late 2018, which might have selected for disinfectant-resistant organisms and would explain the poor effectiveness of this treatment. The taxa that were enriched in the closed ward, *Cupriavidus pauculus, Sphingomonas paucimobilis*, and *Acidovorax temperans*, are frequently listed in studies of disinfection-resistant bacteria (Jia et al., 2019, 2015; Khan et al., 2016; Luo et al., 2021). *Cupriavidus* and *Sphingomonas* are genera that have been recovered from the water system on the International Space Station, despite intensive, multi-step purification systems (Yang et al., 2021). Their ability to withstand disinfection is partly due to their propensity to form biofilms (Gulati and Ghosh, 2017; Zhu et al., 2020), and these would have grown more readily during the extended closure period when water use was lower than in an occupied ward. The dosing level of this first treatment, typical of superchlorination used to disinfect new pipework, might not have been high enough to impact on these taxa.

In contrast, the second chemical treatment, with silver stabilised hydrogen peroxide and dosed at a level more typical of 'shock' treatment, significantly reduced TVCs and decreased the abundance and prevalence of *Cupriavidus pauculus* and *Acidovorax temperans*, suggesting that the resistance of these species to chlorine and chlorine dioxide does not necessarily carry over to other chemicals, or that any such resistance can be overcome with higher dosing. However, TVCs were still above our local high-risk threshold of 10 CFU/ml in almost 14 % of samples, and *Sphingomonas paucimobilis* continued to be detected at relatively high abundance and prevalence, suggesting it was more resistant to this treatment than the other species.

Our final intervention was to replace all the thermostatic mixing taps on the ward, which account for the majority of outlets (81/116), with new taps of the same model, and this had the most pronounced impact on TVC and GNB results. Neither *Acidovorax temperans* nor *A. delafieldii* were detected in the subsequent two months, and *Sphingomonas paucimobilis* was detected only rarely. *Cupriavidus pauculus* continued to be detected sporadically but at much lower abundance. The effect of tap replacement illustrates how the large surface area and complexity of components inside thermostatic mixing taps are conducive to biofilm formation, and how difficult it is to eradicate such a biofilm once it is established. Preventing the initial formation of biofilm in taps is key; as such, to minimise the risk of new taps being rapidly recolonised, we opted to replace the taps towards the end of our recommissioning period once the proliferation in the wider system had been addressed.

While the last two interventions, and particularly the thermostatic mixing tap replacement, appear to have been more successful than the initial chlorine treatment, we cannot rule out a cumulative effect since these interventions were carried out sequentially. The silver stabilised hydrogen peroxide treatment did not appear to be sufficient to eradicate counts arising from localised proliferation within the taps (though perhaps a longer post-treatment period would have been required to be sure of this), but conversely, tap replacement alone is unlikely to have been sufficient for long-term control of microbial proliferation, as the new taps would have been recolonised more quickly. Furthermore, the influence of other measures, notably flushing and ward cleaning, could not easily be quantified, as they were implemented and increased gradually over the recommissioning period. However, these are also likely to have contributed to the overall reduction in microbial counts.

4.5. Measuring outcomes: sampling approaches and benchmarks

Guidance on managing hospital water systems recommends taking both pre- and post-flush samples when investigating issues of *Legionella* or *Pseudomonas aeruginosa* proliferation (Department of Health, 2016; Health Facilities Scotland, 2014b; Health Protection Scotland, 2018), with outlets being placed out of use for several hours prior to collection of the pre-flush sample. While this guidance may be implementable for small, targeted investigations, we found that it was not compatible with the scale of our recommissioning work, the number of samples to be collected within a fixed period with available staff, the requirement to collect separate samples from the same outlet for *Legionella* spp. and other microbiological tests, and the increased flushing regime in place across the ward. Instead, we opted for a more pragmatic approach and relied on 'first-flush' samples without a strict requirement that the outlets be placed out of use for hours prior to sample collection. These proved adequate to monitor trends during the recommissioning period, and the final sweep of true pre-flush sampling at the end of this period showed no additional microbiological burden that was being missed by first-flush sampling.

Once the tap replacement was completed in January 2022, the system was monitored for a further seven weeks while the ward remained closed. During this period, extensive flushing was being carried out to more closely approach water use in an occupied ward, cleaning was being undertaken to clinical standards, and systematic water sampling continued, but the ward was otherwise unoccupied. Results from this period showed that once the microbial proliferation within the water system itself had been addressed, TVCs in this empty ward, with unfiltered samples, were significantly lower than on the occupied floors above and below. Interestingly, they were also significantly lower than in the ward where the paediatric haemato-oncology patients had been relocated during the closure period, despite this temporary ward having 0.2 μm point-of-use (POU) filters in place on all outlets. Benchmarking against this temporary ward as well as against the floors below and above provided reassurance that the water system in our closed ward had been returned to a safe state.

4.6. Point-of-use filters and the source of microbial counts

In our experience and as reported by others, occasional, sporadic detection of TVCs in water samples collected through 0.2 μm POU filters, as seen in the temporary ward where patients had been relocated, is not entirely unexpected in occupied wards (Barna et al., 2014; Florentin et al., 2016). We have a series of actions that are automatically triggered when this occurs, including sampling the outlet without a filter and then through a new filter, and have yet to detect an instance of filter failure or leaking due to faulty installation. These sporadic counts are most likely due to retrograde contamination, i.e. deposition of microbes onto the outer filter casing from the air, from splashing, from contact with hands, etc. In short, microbial counts in 0.2 μm POU-filtered samples appear to come not from the water itself, but from the interactions of people with the periphery of the water system.

This conclusion is further supported by the trends observed in the reconstructed paediatric haemato-oncology ward since it reopened to patients in early March 2022. The rare samples collected through 0.2 μm POU filters that exceeded our local TVC thresholds or that tested positive for *Pseudomonas* spp. or other GNBs were sporadic and spread across the ward, a pattern suggestive of occasional retrograde contamination of outlets. There was also a clear shift in the taxonomic profile of detected bacterial species in filtered samples. During the pre-opening period prior to filter installation and reoccupation, when GNBs were detected, they belonged to bacterial taxa that are natural inhabitants of water: *Cupriavidus pauculus, Blastomonas ursincola, Sphingomonas* spp., *Sphingobacterium thalpophilum*. Most of these have not been detected at all since the ward reopened, and *C. pauculus* was found only once, in an unfiltered sample. *Sphingomonas paucimobilis* is the only taxon prevalent in the pre-opening period that has continued to be detected repeatedly after installation of POU filters, albeit sporadically (in only three filtered samples out of 3357, from three different locations, 6–9 months apart). This could be due to its reported ability to pass through 0.2 μm filters (Ryan and Adley, 2010). Apart from *S. paucimobilis*, the small number of filtered samples that have tested positive for GNBs or *Pseudomonas* spp. since the ward reopened to patients have yielded bacterial species that were rare or absent from the unfiltered samples collected in the months prior to reoccupation. This included taxa that can be found in diverse environments but are also known to occur as human commensals or in human faeces, such as *Pantoea agglomerans, Pseudomonas* spp., *Roseomonas* spp. and *Acinetobacter lwoffii* (Patil and Chopade, 2001; Romano-Bertrand et al., 2016). *E. coli*, a gut commensal, was also detected for the first time in the post-reconstruction period, in a single filtered sample. The range of taxa detected and their absence from unfiltered samples collected prior to the ward reopening strongly suggest a human source. A similar trend was reported after point-of-use filters were fitted to showers in a Japanese hospital, with detected bacteria shifting from water-associated taxa (genera *Mycobacterium, Pseudomonas, Stenotrophomonas*, and *Sphingomonas*) to human-associated taxa (*Staphylococcus* spp.) (Sasahara et al., 2020).

There has been an increasing trend to classify any microbial taxon detected in or around healthcare water systems as 'waterborne pathogens' regardless of their likely source. This definition is even sometimes extended to enteric bacteria with limited ability to multiply outside the mammalian gut. Such an oversimplification is unhelpful, as the term 'waterborne' implies that these taxa are being carried within the water system and are coming out of the outlets. Our data showed that it was possible to distinguish between taxa that were truly waterborne, i.e. those detected when the ward was empty and before 0.2 μm POU filters were fitted, from taxa that were likely from a human source and deposited on the outside of the outlets, for example via direct hand contact or water splashing during handwashing. Once the proliferation of waterborne microorganisms within this water system was rectified, further counts were more likely to have come from human interactions around the periphery of the system. Put simply, just because a microorganism is detected in a water sample does not mean it has come from the water. Conflating true waterborne organisms with those from human sources risks diverting limited resources to unsuitable and costly interventions focused on the water distribution system itself rather than on its users.

5. Conclusions

Extended closure and reconstruction of our paediatric haemato-oncology ward resulted in microbial proliferation within its water system, despite all applicable water management guidance having been followed or exceeded, particularly with regards to flushing. A multidisciplinary, systematic approach proved successful in returning the water system to a safe state. Of three sequential interventions, namely chlorine treatment, silver stabilised hydrogen peroxide treatment, and tap replacement, we showed that the latter two were more impactful than chlorine alone, though a cumulative effect cannot be ruled out. The total microbial load as well as the specific Gram negative bacterial taxa detected shifted during the recommissioning period, and it was possible to distinguish between microbial counts arising from within the water system and those that are more likely to be from interactions of people around water outlets. Monitoring of the ward's water system since it reopened to patients over two years ago has shown no return of the high microbial counts observed in the immediate post-reconstruction period, ensuring continued provision of safe water to highly vulnerable patients.

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CRediT authorship contribution statement

Dominique L. Chaput: Writing – review & editing, Writing – original draft, Visualization, Methodology, Formal analysis, Data curation, Conceptualization. **Kerr Clarkson:** Writing – review & editing, Methodology, Investigation, Conceptualization. **Linda Bagrade:** Writing – review & editing, Methodology, Conceptualization. **Aleksandra Marek:** Writing – review & editing, Conceptualization. **Dennis Kelly:** Writing – review & editing, Methodology, Conceptualization. **David Watson:** Writing – review & editing, Methodology, Investigation, Conceptualization. **Tom Steele:** Writing – review & editing, Supervision, Conceptualization. **Alistair Leanord:** Writing – review & editing, Supervision, Methodology, Conceptualization.

Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:

David Watson reports a relationship with DMA Canyon Ltd that includes: board membership. Dennis Kelly reports a relationship with Pro Lp Consulting Ltd that includes: board membership. Submitted oral and/or written evidence to the Scottish Hospitals Inquiry, which is investigating the construction of the Queen Elizabeth University Hospital, Glasgow - DLC, KC, DK, DW, AL, TS If there are other authors, they declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.watres.2025.123748.

Data availability

Data will be made available on request.

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Bundle of documents for Oral hearings commencing from 19 August 2025 in relation to the Queen Elizabeth University Hospital and the Royal Hospital for Children, Glasgow

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