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Persistent contamination of heater—cooler units for extracorporeal circulation cured by chlorhexidine—alcohol in water tanks

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S U M M A R Y

Recently, surgical site infections due to non-tuberculous mycobacteria (NTM) have been linked to heater—cooler unit contamination. The European Centre for Disease Prevention and Control and manufacturers now recommend the use of hydrogen peroxide in filtered water to fill heater—cooler unit tanks. After implementation of these measures in our hospital, heater—cooler units became heavily contaminated by opportunistic waterborne pathogens such as *Pseudomonas aeruginosa* and *Stenotrophomonas maltophilia*. No NTM were detected but fast-growing resistant bacteria could impair their detection. The efficiency of hydrogen peroxide and chlorhexidine—alcohol was compared *in situ*. Chlorhexidine—alcohol treatment stopped waterborne pathogen contamination and NTM were not cultured whereas their detection efficiency was probably improved.

Keywords:

Heater—cooler units
Non-tuberculous mycobacteria
Stenotrophomonas maltophilia
Pseudomonas aeruginosa
Chlorhexidine—alcohol

Introduction

Heater—cooler units (HCUs) used for extracorporeal circulation in cardiac surgery are a recognized bacterial reservoir

for waterborne pathogens such as pseudomonas and legionella [1]. However, the transmission from HCUs and their involvement in surgical site infections (SSIs) have been demonstrated only recently, with worldwide clustered cases of cardiac SSI caused by *Mycobacterium chimaera*, a non-tuberculous mycobacterium (NTM) originating from HCUs [1,2]. This outbreak led to the provision of a rapid risk assessment by the European Centre for Disease Prevention and Control (ECDC) in April 2015 that recommended the implementation of disinfection practices for the microbiological control of HCUs [3,4].

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Heater-cooler unit manufacturers subsequently recommended adding medical-grade hydrogen peroxide in the filtered water used for filling the HCU tanks; and this unstable solution was to be changed weekly [4]. Additionally, a weekly disinfection of the tank using biocides from a list provided by the manufacturer was recommended. Hydrogen peroxide and other recommended biocides were chosen for their activity against NTM. After implementation of this protocol in our hospital, water in HCU tanks became heavily contaminated by fast-growing waterborne pathogens such as *Pseudomonas aeruginosa* and *Stenotrophomonas maltophilia*.

This study aimed to evaluate disinfection protocols by microbiological sampling of water from HCU tanks in our hospital. For this, we compared, *in situ*, the efficiency of the recommended hydrogen peroxide disinfection method with another protocol using chlorhexidine-alcohol at 0.1% final concentration.

Methods

The cardiac surgery ward of the University Hospital of Montpellier used four 3T HCUs (LivaNova, London, UK). The manufacturer recommended the weekly replacement of the hydrogen peroxide, and a full decontamination every two weeks, but this time-consuming protocol quickly turned into an in-house degraded protocol named herein 'hydrogen peroxide protocol': water tanks were decanted every two weeks and filled with filtered (Aquasafe filter; Pall Corporation, Port Washington, New York, USA) tap water containing 100 mL of medical-grade 3% hydrogen peroxide, with the addition of 50 mL of 3% hydrogen peroxide every week [4]. A full HCU decontamination using Puristeril® 340 (Fresenius Medical Care, Bad Homburg, Germany) was performed once a month. A new protocol, named 'chlorhexidine-alcohol protocol', subsequently replaced hydrogen peroxide with 0.5% chlorhexidine-alcohol, with the same frequencies of replacement and decontamination as during 'hydrogen peroxide protocol'.

The water tanks of the four HCUs were sampled twice a month, prior to disinfection. Half a litre of water was aseptically sampled in a sterile bottle containing sodium thiosulphate (20 mg/L final concentration) (Gosselin, Borre, France). Water was filtered through 0.45 µm-pore-size cellulose sterile filters (Sartorius, Göttingen, Germany) using a vacuum filter apparatus. Filters were deposited on to plate count agar (PCA) (bioMérieux, Marcy l'Etoile, France) to enumerate the total mesophilic bacteria, and on to cefrimide and MacConkey agar plates (bioMérieux) for the detection of *Pseudomonas aeruginosa* and Enterobacteriaceae, respectively. Media were incubated for 72 h at 30°C. To detect NTM, 50 mL of water was decontaminated using 4% NaOH for 15 min. After neutralization using pH 6.8 phosphate buffer, the suspension was centrifuged at 3000 g for 30 min. Pellet (500 µL) was plated on to blood agar medium and Middlebrook Agar 7H11 medium (bioMérieux). The decontamination step could be avoided if the water sample was free from fast-growing mesophilic bacteria. Media were incubated for 10 days at 30°C.

All colony morphotypes were identified by matrix-assisted laser desorption/ionization time-of-flight mass spectrometry, using Microflex and MALDI Biotyper database (Bruker, Billerica, MA, USA).

Results

Table I shows the bacterial inoculum in HCU tanks water and the presence of waterborne pathogens, NTM, and fungi, according to the period of the decontamination protocol. The evolution of the bacterial contamination in the water tank is also displayed in Figure 1. Prior to March 2017, the four HCUs were contaminated by mean bacterial inoculum varying from 7.10×10^1 to 7.4×10^3 colony-forming units (cfu) per 100 mL; and contamination was slightly lower for HCU1 and 2 compared to HCU3 and 4 (Table I, Figure 1). In each HCU, the bacterial inoculum varied widely from 0 to 5.10^4 cfu/100 mL according to the sample and independent to the date of sampling.

Waterborne opportunistic pathogens such as *P. aeruginosa* and *S. maltophilia* were detected as well as other nosocomial pathogens (*Enterobacter cloacae*, *Enterococcus faecalis*, and *Klebsiella pneumoniae*) but no NTM were isolated (Table I). It was noteworthy that *P. aeruginosa* and *S. maltophilia* partially resisted the in-laboratory decontamination step and probably impaired the detection of NTM. Several fungi were also retrieved in the four HCUs: *Aspergillus niger*, *Aspergillus fumigatus*, and *Fusarium* spp. (Table I).

After March 2017, the chlorhexidine-alcohol protocol was tested for filling HCU1 and HCU2 tanks. The hydrogen peroxide protocol continuing for HCU3 and HCU4 were deemed as controls. The use of chlorhexidine-alcohol did not impair the function of HCUs but perfusionists observed the formation of scale deposit in tubing, requiring their monthly replacement. The two control HCUs were still highly contaminated, mainly by *S. maltophilia* and filamentous fungi (Table I, Figure 1). NTM were not detected but again the microbiological analysis for their detection was complicated by *S. maltophilia*, which resisted the pre-analytical in-laboratory decontamination step. During October 2017, the chlorhexidine-alcohol protocol was extended to HCUs 3 and 4, permitting suppression of the waterborne pathogens, without the detection of NTM. Hence, the chlorhexidine-alcohol protocol eliminated all growing bacteria, including *S. maltophilia* and fungi. NTM were not detected during the whole study period.

Discussion

Prior to April 2015, microbiological control of HCU tank water was not regularly performed in the Hospital University of Montpellier. After an environmental investigation around a cardiac SSI case caused by *Mycobacterium wolinskyi* during which *Mycobacterium chelonae* was isolated from an HCU, microbial contamination was regularly analysed as was also recommended by the ECDC rapid risk assessment [5]. During this period, the bacterial load reached 10^4 cfu per 100 mL and *S. maltophilia* seems to have been gradually selected. Despite wide variations in load, the persistence of bacterial contamination suggested that bacterial growth and biofilm formation were not impaired by the hydrogen peroxide protocol. By contrast, the chlorhexidine-alcohol protocol eliminated bacterial contamination and did not select NTM as was feared, due to their intrinsic tolerance to chlorhexidine.

Due to the worldwide outbreak of *M. chimaera*, NTM are considered as a risk factor for cardiac surgery SSIs when present in HCUs. The risk associated with other micro-organisms colonizing HCU tanks appears to be lower. In our hospital, a

Table I
Follow-up of microbiological contamination of heater–cooler units (HCUs)

Period/protocol	HCU	Water tanks	Mean inoculum in cfu/100 mL (range)	NTM	Pathogenic bacteria (% of positive samples)	Fungi (% of positive samples)
February 2016 to March 2017						
Hydrogen peroxide protocol	HCU 1	Cardioplegy	7×10 ¹ (0–10 ⁴) (<i>N</i> = 20)	None	<i>Stenotrophomonas maltophilia</i> (45%) <i>Pseudomonas aeruginosa</i> (5%)	<i>Aspergillus niger</i> (5%) Fusarium (30%) Penicillium (10%) Yeast (10%)
		Patient	1.1×10 ² (0–5.10 ⁴) (<i>N</i> = 20)	None	<i>S. maltophilia</i> (50%) <i>Enterococcus faecalis</i> (5%)	Fusarium (20%) Penicillium (15%)
	HCU 2	Cardioplegy	8×10 ² (0–5.10 ⁴) (<i>N</i> = 19)	None	<i>S. maltophilia</i> (47%) <i>P. aeruginosa</i> (5%) <i>Enterobacter cloacae</i> (5%)	Fusarium (32%)
		Patient	8×10 ³ (0–5.10 ⁴) (<i>N</i> = 19)	None	<i>S. maltophilia</i> (42%) <i>P. aeruginosa</i> (5%) <i>Burkholderia cenocepacia</i> (5%)	Fusarium (32%) Penicillium (11%) Yeast (5%)
	HCU 3	Cardioplegy	5.4×10 ³ (0–10 ⁴) (<i>N</i> = 19)	None	<i>S. maltophilia</i> (74%)	Fusarium (42%) Penicillium (11%) Yeast (11%)
		Patient	6×10 ³ (40–10 ⁴) (<i>N</i> = 19)	None	<i>S. maltophilia</i> (68%)	<i>Aspergillus fumigatus</i> (5%) Fusarium (37%) Penicillium (11%)
	HCU 4	Cardioplegy	6.3×10 ³ (0–10 ⁴) (<i>N</i> = 13)	None	<i>S. maltophilia</i> (77%)	Fusarium (23%) Penicillium (8%) Yeast (15%)
		Patient	7.4×10 ³ (0–10 ⁴) (<i>N</i> = 13)	None	<i>S. maltophilia</i> (69%) <i>Klebsiella pneumoniae</i> (8%)	Fusarium (23%) Yeast (8%)
April to September 2017						
Chlorhexidine—alcohol protocol	HCU 1	Cardioplegy	<1 (<i>N</i> = 6)	None	None	None
		Patient	<1 (<i>N</i> = 6)	None	None	None
	HCU 2	Cardioplegy	<1 (<i>N</i> = 6)	None	None	None
		Patient	<1 (<i>N</i> = 6)	None	None	None
Hydrogen peroxide protocol	HCU 3	Cardioplegy	10 ⁴ (10 ⁴) (<i>N</i> = 7)	None	<i>S. maltophilia</i> (100%)	None
		Patient	10 ⁴ (10 ⁴) (<i>N</i> = 7)	None	<i>S. maltophilia</i> (86%)	None
	HCU 4	Cardioplegy	10 ⁴ (10 ⁴) (<i>N</i> = 8)	None	<i>S. maltophilia</i> (88%)	Fusarium (17%) Penicillium (17%)
		Patient	10 ⁴ (10 ⁴) (<i>N</i> = 8)	None	<i>S. maltophilia</i> (75%) <i>K. pneumoniae</i> (12%)	Penicillium (17%)
October 2017						
Chlorhexidine—alcohol protocol	HCU1	Cardioplegy	<1 (<i>N</i> = 3)	None	None	None
		Patient	<1 (<i>N</i> = 3)	None	None	None
	HCU2	Cardioplegy	<1 (<i>N</i> = 3)	None	None	None
		Patient	<1 (<i>N</i> = 3)	None	None	None
	HCU3	Cardioplegy	<1 (<i>N</i> = 3)	None	None	None
		Patient	<1 (<i>N</i> = 3)	None	None	None
	HCU4	Cardioplegy	<1 (<i>N</i> = 3)	None	None	None
		Patient	<1 (<i>N</i> = 3)	None	None	None

cfu, colony-forming units; NTM, non-tuberculous mycobacteria; *N*, number of samples.

retrospective review of patient case notes showed that no cardiac SSIs caused by *P. aeruginosa* or *S. maltophilia* occurred in this period (data not shown). Cardiac surgery SSI cases caused by *S. maltophilia*, *P. aeruginosa* or mould reported in the literature have not been linked to HCU contamination; but this potential source of infection was not specifically investigated in these reports [6,7]. Importantly, reservoirs of multidrug-resistant, bacterial pathogens are not acceptable within a surgical theatre and should be eliminated by a suitable protocol. Moreover, the absence or low load of rapidly growing bacteria

avoids mycobacterial culture media contamination, allowing the detection of slow-growing NTM. In this situation, the in-laboratory decontamination of the sample that can affect NTM viability is not required as described for NTM detection in reprocessed bronchoscopes [8,9]. In fact, we did not find NTM in our study and we cannot exclude that the decontamination step used to eradicate rapidly growing bacteria was also decontaminating the water samples of mycobacteria [10].

In practice, the manufacturer’s recommendation for HCU tank decontamination, i.e. the hydrogen peroxide protocol,

Figure 1. Evolution curves of water tank contamination of heater-cooler units (HCUs). cfu, colony-forming units.

is time-consuming and deviations can occur, due to the workload within cardiac surgery teams. Most reports indicate that change of hydrogen peroxide solution is performed less frequently than recommended [1]. In our hospital, the hydrogen peroxide protocol was followed as scrupulously as possible, considering its cumbersomeness; yet we failed to obtain acceptable microbiological quality for water in HCUs. Other authors have tried successive close cycles of decontamination to improve the decontamination and have concluded that the microbial load in the HCU had not been totally removed, and that the high frequency of decontamination may have led to damage to copper components [11].

In our hospital, the infection control team, in agreement with the cardiac surgery staff and HCU manufacturer, now recommend the use of chlorhexidine-alcohol for HCU decontamination. The tap water used for filling tanks is submitted for antimicrobial filtering and its quality is assessed four times a year. The water in HCU tanks is sampled twice a month, and is subjected to microbiological analysis, including specific search of NTM, with and without in-laboratory decontamination of the sample, thereby ensuring better detection of NTM. The main aspect to monitor while using this new protocol of disinfection is scale formation in the tubing, which needs replacing as soon as scale deposit is observed.

Conflict of interest statement

None declared.

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