

Pasteurization is effective against multidrug-resistant bacteria

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Background: The emergence and rapid spread of multidrug-resistant isolates causing nosocomial infections, particularly pandrug-resistant *Acinetobacter baumannii*, pandrug-resistant *Pseudomonas aeruginosa*, methicillin-resistant *Staphylococcus aureus*, and extended-spectrum β -lactamase-producing *Enterobacteriaceae* are of great concern worldwide.

Methods: This study investigated the efficacy of pasteurization against contamination of culture tubes and plastic bags by the above 4 important multidrug-resistant bacteria. A 5-mL bacterial suspension with approximately 10^5 and 10^9 cfu/mL of each organism was inoculated into 3 sets of plastic bags and culture tubes and subjected to pasteurization in a washer at 75°C for 30 minutes.

Results: A nearly total eradication was found after pasteurization of these 4 drug-resistant pathogens.

Conclusion: Pasteurization was highly effective against drug-resistant bacteria. Strict adherence to appropriate infection control management for respiratory circuits is important for reducing the spread of drug-resistant bacteria among ventilator-assisted patients. (Am J Infect Control 2006;34:320-2.)

The emergence and rapid spread of multidrug-resistant isolates causing nosocomial infections, particularly methicillin-resistant *Staphylococcus aureus* (MRSA), vancomycin-resistant enterococci, and extended-spectrum β -lactamase (ESBL)-producing *Enterobacteriaceae* are of great concern worldwide.¹⁻⁵ In the National Taiwan University Hospital (NTUH), the rapid emergence and high prevalence of multidrug-resistant pathogens, particularly MRSA, pandrug-resistant *Acinetobacter baumannii* (PDRAB), and pandrug-resistant *Pseudomonas aeruginosa* (PDRPA) have become a severe problem.⁶⁻¹⁰ Clonal dissemination of these multidrug-resistant pathogens is frequently encountered in this hospital, particularly among patients treated in intensive care units with ventilator use. Pasteurization was recommended as the standard technique for the sterilization of the respiratory equipment (tubing and

accessories).^{11,12} However, although concerns remain about the adequacy of disinfection and sterilization of antibiotic-resistant bacteria, studies remain limited.¹³ The aim of this study was to investigate the effectiveness of pasteurization against multidrug-resistant bacteria, especially the currently emerging pandrug-resistant bacteria.⁶⁻¹⁰

METHODS

Bacteria tested in this study included PDRAB, non-PDRAB, PDRPA, non-PDRPA, MRSA, methicillin-susceptible *Staphylococcus aureus* (MSSA), ESBL-producing *Escherichia coli*, and non-ESBL-producing *E. coli*. PDRAB or PDRPA isolates were defined as resistant to all commercially available agents including penicillins, piperacillin-tazobactam, ticarcillin-clavulanate, extended-spectrum cephalosporins (ceftriaxone, ceftazidime, cefepime, and cefpirome), aminoglycosides (amikacin, tobramycin, and gentamicin), fluoroquinolones (ciprofloxacin, moxifloxacin, and levofloxacin), aztreonam, and carbapenems (imipenem, meropenem, and ertapenem); but susceptibility to tigecycline, polymyxin B, or colistin was not detected because the latter 3 agents were not available in Taiwan. Bacterial suspension with approximately 10^5 and 10^9 colony-forming units (CFU)/mL were prepared after inoculating these microorganisms into trypticase soy broth (BBL Microbiology Systems, Sparks, MD). The plastic bags with approximately 150 mL-capacity (TERUFLEX Terumo Transfer Bag; Terumo Corporation, Tokyo, Japan) and the plastic culture tubes (approximately 16 mL

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Table 1. Summary of disinfection effect of pasteurization against 8 commonly encountered bacteria in the hospital

Pathogen	Mean MPN before pasteurization (cfu/mL)	Plastic bags		Culture tubes	
		Mean MPN after pasteurization (cfu/mL)	Number of decimal reduction	Mean MPN after pasteurization (cfu/mL)	Number of decimal reduction
PDRAB	2.0×10^5	0	>5	0	>5
	1.2×10^9	0.7	$\cong 9$	0	>9
Non-PDRAB	1.8×10^5	0.7	$\cong 5$	0	>5
	1.2×10^9	0	>9	0	>9
PDRPA	1.2×10^5	0	>5	0	>5
	1.2×10^9	0	>9	1.3	$\cong 9$
Non-PDRPA	1.5×10^5	0	>5	0	>5
	1.1×10^9	0	>9	0	>9
MRSA	1.4×10^5	0	>5	0.7	$\cong 5$
	1.7×10^9	1.3	$\cong 9$	6.0	$\cong 9$
MSSA	1.6×10^5	0	>5	0	>5
	1.8×10^9	0	>9	0	>9
<i>E coli</i> , ESBL phenotype	1.6×10^5	0	>5	0	>5
	1.8×10^9	0	>9	0	>9
<i>E coli</i> , non-ESBL phenotype	1.0×10^5	0	>5	0	>5
	1.8×10^9	0	>9	0	>9

PDRAB, pandrug-resistant *Acinetobacter baumannii*; PDRPA, pandrug-resistant *Pseudomonas aeruginosa*; MRSA, methicillin-resistant *Staphylococcus aureus*; MSSA, methicillin-sensitive *Staphylococcus aureus*; ESBL, extended-spectrum β -lactamase; MPN, most probable number.

capacity) (Creative Microbiologicals, Taipei, Taiwan) simulating the respiratory circuits were used in the experiments. Each plastic bag or culture tube was inoculated with 5 mL trypticase soy broth with each bacterium, and the studies were repeated twice.

The inoculated plastic bags and culture tubes were processed in the same manner as respiratory circuits of patients. A therapist-technician placed the plastic bags and culture tubes in the basket of the washer (High Level Disinfection Systems: Clear 540; ClearMedical, Bellevue, WA). The washer provided a spray cycle, a detergent cleaning cycle, a hot water spray rinse, and then the immersion in "still" water at 75°C for 30 minutes, completing the disinfection process.

A noninoculated culture tube was included in each set for disinfection as a negative control. A positive control culture tube was prepared for each organism but was not disinfected. This culture tube was left at room temperature and processed the next day to determine the viability of the organisms in the tubes. After disinfection, 1 mL suspension fluid from the culture tubes or plastic bags was inoculated onto trypticase soy agar. The original pathogens without disinfection were also inoculated. All plates were incubated 37°C overnight. Following incubation, colony counts were performed on each plate.

RESULTS

The results are summarized in Table 1. Nearly all organisms were totally eradicated after pasteurization, except for PDRAB (10^9 cfu/mL), non-PDRAB (10^5 cfu/mL),

and MRSA (10^9 cfu/mL) in plastic bags; and PDRPA (10^9 cfu/mL) and MRSA (10^5 cfu/mL and 10^9 cfu/mL) in culture tubes.

DISCUSSION

It was well-known about the importance of disinfection and sterilization for equipment of invasive procedure.¹¹⁻¹³ Because invasive procedures involve contact between a medical device or surgical instrument and a patient's sterile tissue or mucous membranes, a major risk of all such procedures is the introduction of pathogenic microbes that could lead to infection. Failure properly to disinfect or sterilize reusable medical equipment carries a risk associated with breach of the host barriers.¹¹⁻¹³ Recent reports have emphasized the importance of reprocessing of endoscopes. Although the concern about antibiotics-resistant bacteria has been mentioned, few studies about the aseptic ability of pasteurization against drug-resistant bacteria have been performed.¹¹⁻¹³ Previous studies only showed that pasteurization could eliminate common pathogens such as *S aureus* and *Klebsiella pneumoniae*, with the exception of spore-forming bacteria.¹⁴ This study illustrates that resistant pathogens in the culture tubes and plastic bags were also eliminated by this process.

This study found no differences in the effectiveness of pasteurization between drug-susceptible pathogens and drug-resistant pathogens on culture tubes and plastic bags. The decimal reduction by pasteurization with 75°C for 30 minutes was at least 9. However,

few organisms tested in this study were apparently more resistant to sterilization, particularly MRSA. The reason for this phenomenon was unclear. Because higher density of bacterial contamination in respiratory equipment might exist, further investigation using higher concentration of bacterial inoculation in respiratory equipment and accessories are needed.

This study has demonstrated that pasteurization of the culture tubes and plastic bags had comparable high effectiveness against either susceptible or drug-resistant bacteria. Although we did not directly use the respiratory equipment and accessories in this study, the results of our studies still confirm the disinfection and sterilization ability of pasteurization against drug-resistant bacteria. Strict adherence to appropriate infection control management for respiratory circuits is a crucial infection control measure for reducing the spread of drug-resistant bacteria among ventilator-assisted patients.

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