



## Major Article

## Assessment of a novel antimicrobial surface disinfectant on inert surfaces in the intensive care unit environment using ATP-bioluminescence assay

Charles E. Edmiston Jr PhD<sup>a,\*</sup>, Maureen Spencer MEd<sup>b</sup>, Brian D. Lewis MD<sup>a</sup>, Peter J. Rossi MD<sup>a</sup>, Kellie R. Brown MD<sup>a</sup>, Michael Malinowski MD<sup>a</sup>, Gary R. Seabrook MD<sup>a</sup>, David Leaper DSc<sup>c</sup>

<sup>a</sup> Department of Surgery, Medical College of Wisconsin, Milwaukee, WI

<sup>b</sup> Infection Prevention Consultants, Boston, MA

<sup>c</sup> Department of Clinical Sciences, University of Huddersfield, Huddersfield, United Kingdom



## Key Words:

Adenosine triphosphate bioluminescence assay  
ABT  
Isopropyl alcohol/organofunctional silane  
RODAC plate counts  
Surface disinfection

**Background:** Cross-contamination from inanimate surfaces can play a significant role in intensive care unit (ICU)-acquired colonization and infection. This study assessed an innovative isopropyl alcohol/organofunctional silane solution (IOS) to reduce microbial contamination on inert surfaces in a medical ICU.

**Methods:** Baseline adenosine triphosphate bioluminescence testing (ABT)-measurements (N = 200) were obtained on designated inert ICU surfaces followed by IOS treatment. At 1 and 6 weeks, selective surfaces were randomized to either IOS-treated or nontreated controls for comparison using ABT (N = 400) and RODAC colony counts (N = 400). An ABT value of  $\leq 45$  relative light units (RLU) was designated as “clean,” whereas  $>45$  was assessed as “dirty.”

**Results:** Mean RLU baseline values ranged from 870.3 (computer keyboard) to 201.6 (bed table), and 97.5% of surfaces were assessed as “dirty.” At 6 weeks, the mean RLU of surfaces treated with IOS ranged from 31.7 (physician workstation) to 51.5 (telephone handpiece), whereas values on comparative control surfaces were 717.3 and 643.7, respectively ( $P < .001$ ). Some 95.5% of RODAC cultures from IOS-treated sites at 6 weeks were negative, whereas 90.5% of nontreated sites were culture-positive, yielding multiple isolates including multidrug-resistant gram-positive and gram-negative bacteria.

**Conclusions:** IOS-treated surfaces recorded significantly lower RLU and RODAC colony counts compared with controls ( $P < .001$ ). A single application of IOS resulted in a persistent antimicrobial activity on inert ICU surfaces over the 6-week study interval.

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The intensive care unit (ICU) is a significant source of patient morbidity and mortality.<sup>1</sup> In the ICU, inanimate surfaces may be contaminated by bacteria, including multidrug-resistant (MDR) isolates.<sup>2</sup> Furthermore, a recent study conducted in adult, pediatric, and neonatal ICUs has documented, using confocal scanning electron microscopy, that microbial biofilms were present on all sampled, high-touch surfaces. Culture results revealed the presence of multiple MDR hospital-acquired pathogens including, methicillin-resistant *Staphylococcus aureus* (MRSA), vancomycin-resistant enterococci, and extended-spectrum  $\beta$ -lactamase producing gram-negative pathogens.<sup>3</sup> Cross-transmission of microorganisms from inanimate surfaces can play a significant role in ICU-acquired colonization and infection. The goal of

surface decontamination is to reduce, if not eliminate, the bioburden within the environment of care. However, inert surface contamination is often widespread throughout the ICU environment, and is not limited to those areas immediately adjacent to resident patient populations.<sup>4</sup> The present study assessed the efficacy of an innovative, proprietary antimicrobial quaternary ammonium coating (isopropyl alcohol/organofunctional silane solution [IOS]), which bonds covalently to inert surfaces to reduce microbial surface contamination in a medical ICU, and the role of adenosine triphosphate bioluminescence testing (ABT) to validate efficacy.

### METHODS

#### Study environment

The assessment of a novel, proprietary quaternary ammonium bonding solution (IOS: MicroCare XLP, Parasol Medical, LLC, Buffalo

\* Address correspondence to Charles E. Edmiston, Jr, PhD, Department of Surgery, Medical College of Wisconsin, 8701 Watertown Plank Rd, Milwaukee, WI 53226.

E-mail address: [edmiston@mcw.edu](mailto:edmiston@mcw.edu) (C.E. Edmiston).

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Grove, IL) was conducted in a 26-bed ICU at a tertiary 505-bed health care facility. The daily patient census during the first assessment period (week 1) ranged from 88%–100%, whereas the daily census during the second assessment period (week 6) was 100%. Routine housekeeping practices including postdischarge terminal room cleaning, floor and surface cleaning were not altered during the period of study.

#### Baseline assessment: ABT

Over a 2-week baseline period, 200 separate measurements of surface bioburden contamination were assessed using ABT (Getinge SafeStep Handheld Luminometer, Getinge USA, Rochester, NY). The 4 individual high-touch surfaces selected for study included: telephone handpieces (4), computer keyboards (4), physician/nursing workstations (4), and door handles (4). Two additional patient items were sampled for ABT: individual blood pressure cuffs (10), and patient bed tables (10). The individual surfaces were sampled Monday through Friday (mid-morning) for 2 consecutive weeks.

The testing protocol involved sampling a 2 cm<sup>2</sup> surface area by rubbing a test swab (SafeStep Test Swabs, Getinge USA, Rochester, NY) back and forth using a rotating motion for 15 seconds. All samples were tested within 60 seconds of collection. A value of  $\leq 45$  relative light units (RLUs) indicated a surface with little or no bioburden (designated as clean), whereas a value  $\geq 46$  RLU was designated as dirty. Samples for ABT studies were tested within 60 seconds of collection.

#### ABT and RODAC plate sampling of IOS-treated and control surfaces

Following baseline assessment, designated ICU surfaces were randomly assigned 1:1 to either control or surfaces treated with the antimicrobial/disinfectant coating (ie, IOS). The IOS was applied to each test surface using a soft microfiber cloth covered sponge and allowed to dry. RLU values were obtained from the randomized IOS-bonded and nonbonded surfaces during week 1 and week 6 postapplication. A combined total of 400 ABT readings were obtained in weeks 1 and 6: 20 ABT assessments were made each day (Monday through Friday) of both control and IOS-bonded surfaces. Comparative RODAC (BD Diagnostics, Sparks, MD) plate cultures were obtained at 1 and 6 weeks postapplication from both control and IOS-bonded sites. A combined total of 400 RODAC plate cultures were obtained in weeks 1 and 6: 20 cultures were taken each day (Monday through Friday) from the selected antimicrobial coated (ie, IOS)/noncoated surfaces. All plates were incubated for 24–48 hours at 35°C followed by enumeration of the colony forming units (CFUs) under high magnification. Selective colonies were picked from the surface of the RODAC plates, and microbial identification and in vitro antimicrobial testing was conducted by standard methodology.<sup>5</sup>

#### Statistical analysis

RLU and RODAC colony count data were analyzed by analysis of variance and 2-sided t test to assess the difference in RLU values and recovery of CFUs between IOS-treated (antimicrobial/disinfectant) and nontreated control surfaces at the  $P < .05$  level of significance. Statistical analysis was conducted using the Minitab Statistical Program release 15 (Minitab, State College PA).

## RESULTS

Table 1 documents the mean RLU values determined during baseline testing. RLU values obtained during baseline exhibited variable levels of bioburden on select inert surfaces. The highest mean baseline RLU was noted as 870.3 from the surface of a desktop computer

**Table 1**

Mean baseline RLUs and range for samples taken from ICU sites (N = 200)

ICU sampled surfaces*	(N) <sup>†</sup>	Mean RLU (range)
Telephone handpieces	(40)	570.3 (197.7–732.9)
Computer keyboards	(40)	870.3 (856.7–1,996.8)
Physician workstations	(40)	234.6 (152.3–325.2)
Patient room door handles	(40)	257.5 (84.9–401.6)
Outer surface of blood pressure cuffs	(20)	215.8 (44.6–345.1)
Patient bed tables	(20)	201.6 (37.8–327.5)

ICU, intensive care unit; RLU, relative light unit.

\*Six high-touch surfaces sampled over 2 consecutive weeks.

<sup>†</sup>Total number of adenosine triphosphate bioluminescence testing assessments per ICU sample site.

keyboard, whereas the lowest mean value 201.6 RLU was recorded on the top surface of a patient's bed table. Overall, 97.5% of surfaces were assessed as "dirty" based on luminometer cut point.

The week 1 and week 6 mean RLU and RODAC plate CFU values for control and surfaces treated with antimicrobial/disinfectant bonding solution are reported in Table 2. In week 1, the mean RLU values for IOS-treated surfaces ranged from 13.8 (physician/nursing workstations) to 81.7 (computer keyboard); the comparative mean RLU values for nontreated controls were 400.6 and 322.7, respectively. The highest mean RLU recorded in week 1 was noted on the telephone handpieces (743.2), whereas the mean microbial recovery from these surfaces was 79.1 CFUs. The comparative RLUs/CFUs from IOS-treated telephone handpieces was 63.1 and 2.1, respectively. At 6 weeks postapplication, the highest RLU was recorded on physician/nursing workstations (717.3); the respective RLUs and RODAC colony counts from IOS-treated workstations were 31.7 and 0 CFUs. Overall, at 6 weeks post-IOS application, 3 selective ICU surfaces (patient room door handles, blood pressure cuffs, and physician workstations) all recorded RODAC cultures of 0 CFU; respective control surface CFUs were recorded as 43.7, 11.1, and 35.4 CFUs, respectively. IOS treatment resulted in a significant reduction in the mean RLU and RODAC microbial recovery (ie, CFU) at 1 and 6 weeks postapplication ( $P < .001$ ).

Ninety-six percent of RODAC cultures taken from surfaces treated with IOS were culture-negative, whereas 90.5% of all RODAC cultures

**Table 2**

Mean RLUs/RCCs values for IOS-treated and nontreated control surfaces at 1 and 6 weeks postapplication\*

Samples surfaces	RLUs (N = 400) / RCCs (N = 400)	
	Week 1	Week 6
<b>Telephone handpieces</b>		
Control surfaces	743.2 / 79.1	643.7 / 50.6
Treated surfaces	63.1 / 2.1	51.5 / 1.1
<b>Computer keyboards</b>		
Control surfaces	322.7 / 118.8	515.9 / 88.7
Treated surfaces	81.7 / 0	49.4 / 2.3
<b>Physician/nursing workstations</b>		
Control surfaces	400.6 / 81.5	717.3 / 35.4
Treated surfaces	13.8 / 0	31.7 / 0
<b>Patient room door handles</b>		
Control surfaces	227.8 / 29.7	404.1 / 43.7
Treated surfaces	79.7 / 3.1	50.8 / 0
<b>Blood pressure cuffs</b>		
Control surfaces	67.3 / 14.8	92.6 / 11.1
Treated surfaces	50.9 / 1.1	35.7 / 0
<b>Patient bed tables</b>		
Control surfaces	203.9 / 21.8	418.8 / 47.7
Treated surfaces	59.8 / 2.5	48.9 / 1.6

IOS, isopropyl alcohol/organofunctional silane solution; RCC, RODAC colony counts; RLU, relative light units.

\*P value for IOS-treated surfaces compared with untreated controls at 1 and 6 weeks =  $P \leq .001$ ; RODAC plate count P values for IOS-treated surfaces compared with controls at 1 and 6 weeks =  $P \leq .001$ .

obtained from nontreated sites were culture-positive, yielding multiple isolates including MDR gram-positive and gram-negative bacteria (Table 3).

## DISCUSSION

The active ingredients in MicrobeCare XLP (MicrobeCare, Buffalo Grove, IL) includes 70% isopropyl alcohol and a quaternary ammonium compound (QAC), 2% (3-trihydroxysilyl) propyl dimethyl octadecyl ammonium chloride. Isopropyl alcohol is rapidly bactericidal against vegetative gram-positive and gram-negative bacteria, it is also tuberculocidal, fungicidal, and virucidal against enveloped viruses. Alcohols in general denature proteins by breaking the hydrogen bonds that link oppositely charged hydrogen and oxygen atoms on different parts of the chain-like molecules. The alcohols also have a dehydrating effect, and can dissolve lipids (fats and oils), leading to damage of the bacterial cell membranes. Chemically, QACs are

**Table 3**

Microbial recovery from IOS-treated and nontreated control surfaces at 1 and 6 weeks postapplication

IOS-treated sites (N)*	Recovery of selective genera/spp <sup>†</sup>
Telephone (4/40)	<i>Corynebacterium</i> spp <i>Micrococcus</i> spp <i>Bacillus</i> spp
Computer keyboard (2/40)	<i>Staphylococcus aureus</i> <i>Staphylococcus epidermidis</i> <i>Corynebacterium</i> spp <i>Staphylococcus hominis</i> <i>Bacillus</i> spp <i>Micrococcus</i> spp
Patient bed table (3/40)	<i>Micrococcus</i> spp <i>Staphylococcus hominis</i> <i>Staphylococcus capitis</i>
<b>Nontreated Sites</b>	<b>Recovery of Selective Genera/spp</b>
Telephone (36/40)	<i>Staphylococcus warneri</i> <i>Staphylococcus epidermidis</i> <i>Staphylococcus aureus</i> (MRSA) <i>Micrococcus</i> spp <i>Bacillus</i> spp <b><i>Escherichia coli</i> (ESBL)</b>
Computer keyboard (38/40)	<i>Staphylococcus epidermidis</i> <i>Corynebacterium</i> spp. <i>Enterobacter aerogenes</i> <i>Staphylococcus aureus</i> <i>Candida albicans</i>
Physician workstation (39/40)	<i>Staphylococcus epidermidis</i> <i>Staphylococcus aureus</i> (MRSA) <i>Staphylococcus aureus</i> <i>Escherichia coli</i> <i>Klebsiella pneumoniae</i> <i>Bacillus</i> spp
Patient room door handle (33/40)	<i>Staphylococcus epidermidis</i> <i>Staphylococcus aureus</i> (MRSA) <i>Corynebacterium</i> spp <i>Micrococcus</i> spp <i>Bacillus</i> spp <i>Enterococcus faecium</i> <i>Escherichia coli</i> (ESBL)
Blood pressure cuff (17/20)	<i>Micrococcus</i> spp <i>Staphylococcus epidermidis</i> <i>Enterococcus faecalis</i>
Patient bed table (18/20)	<i>Micrococcus</i> spp <i>Bacillus</i> spp <i>Staphylococcus epidermidis</i> <i>Staphylococcus aureus</i> <i>Klebsiella</i> spp (ESBL) <i>Enterococcus faecium</i>

ESBL, extended-spectrum  $\beta$ -lactamase; ICU, intensive care unit; IOS, isopropyl alcohol/organofunctional silane solution; MRSA, methicillin-resistant *Staphylococcus aureus*.

\*Number of sampled sites yielding positive RODAC cultures/total number of samples.

<sup>†</sup>Selective microbial genera/spp recovered from sampled study surfaces in the ICU.

organically substituted ammonium compounds with known bactericidal, virucidal, and fungicidal activity.<sup>5</sup> The bactericidal action of these compounds has been attributed to several incompletely characterized mechanisms including denaturation of essential cell proteins and disruption of the cell membrane. It has recently been suggested that immobilized QACs cause cell death by inducing lethally strong attractive forces between bacteria and the QAC-coated surface.<sup>7</sup> The use of reactive silanes functionalized with antimicrobial agents has been demonstrated to create an insert surface that is resistant to microbial growth including subsequent biofilm formation.<sup>8</sup>

The study found that although selective IOS-coated surfaces yielded RLU values >45, 96% of these surfaces were found to be culture-negative. The ICU areas of highest bioburden included computer keyboards, telephone handpieces, patient door handles, and physician workstations. Nontreated ICU sampled sites yielded multiple gram-positive and gram-negative microbial isolates, including MDR strains such as MRSA and extended-spectrum  $\beta$ -lactamase gram-negative pathogens. A single application of IOS was effective in reducing microbial surface contamination on all inert ICU test surfaces.

A recent publication has suggested that selective antimicrobial organosilane (ie, IOS) compounds may not prevent microbial surface contamination over a prolonged period of time.<sup>9</sup> However, a study by Lewis et al<sup>10</sup> published in 2015 found that a single application of IOS was effective at providing a persistent disinfectant activity, minimizing microbial contamination in an operating room environment where terminal cleaning may be inadequate or have limited effectiveness. A recent multisite evaluation of environmental cleanliness of high-touch surfaces in the ICU demonstrated large variations in cleaning/disinfection protocols and product selection. The authors found that staff cleaning compliance using rapid methods such as relative surface marker assessment or adenosine triphosphate (ATP) bioluminescence with staff feedback was effective in assuring optimal disinfection of high-touch surfaces in the ICU.<sup>11</sup> The authors suggested that total aerobic culture counts on high-touch surfaces provided limited value for routine monitoring of cleanliness. In the present analysis, the mean RLU and microbial recovery in the IOS-bonded sites demonstrated a significant reduction compared with the nontreated cohort surfaces. Of note, RLU values at 6 weeks in the IOS treatment group exceeded the 45 RLU threshold in 23% of high-touch sampled surfaces (Table 2). However, RODAC plate counts revealed only 8 of 200 culture-positive sites (4.0%) yielding a mean microbial recovery of approximately 2.0 CFUs, compared with 181 nontreated surfaces (90.5%) yielding a mean microbial recovery of 48.7 CFUs. ATP bioluminescence technology has been noted to be an effective strategy for quantifying surface bioburden within the environment of care, especially the operating room environment.<sup>10,12</sup> However, the detection of ATP is not limited to viable bacterial cells but can include other sources of bioburden such as shed squamous epithelial cells or other organic (food or plant) residues.

## CONCLUSIONS

A single application of the novel IOS minimized microbial surface contamination over a 6-week period of study. Unlike a traditional disinfectant, the immobilized QAC provided a persistent antimicrobial activity. The results from this investigation mirrors a report published in 2014, documenting the activity of a quaternary ammonium organosilane compound that when bound to inert surfaces produces a persistent antimicrobial efficacy.<sup>13</sup> The author documented an antimicrobial activity against multiple health care-associated pathogens including MRSA, vancomycin-resistant enterococci, and carbapenem-resistant Enterobacteriaceae. The present investigation suggests that the current proprietary solution provides a significant reduction/suppression of selective health care-associated pathogens on inert clinical surfaces compared with nontreated control surfaces. Inert surfaces in the ICU

are often contaminated by gram-positive and gram-negative health care-associated pathogens, many of which express multidrug resistance. The application of this innovative proprietary antimicrobial technology (ie, IOS) to mitigate the risk of cross-contamination in vulnerable patient populations warrants further studies, validating its persistent efficacy within selective health care settings.

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