Paper #1733

Copper for Preventing Microbial Environmental Contamination

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Summary

Background
Transmission of infection involves various vehicles, including contaminated surfaces which have stimulated interest in antimicrobial materials. Copper has antimicrobial activity and its application in the clinical setting has been explored. Activity of copper against a wide range of hospital pathogens was also determined.

Methods
In vitro activity - Microorganisms were applied to copper and stainless steel and viability determined over 3 hours at room temperature following their recovery into a universal neutralising solution. Viability on the metal was also determined by direct observation using epifluorescence microscopy of propidium iodide/SYTO 9 stained cells.

Clinical assessment - A pilot study assessed the number of microorganisms on copper-containing toilet seats, grab rails, tap handles, light switches and door push plates items on a medical ward. The copper-containing items harboured fewer microorganisms than standard items on a control ward (p=0.01). The study design was adjusted to sample copper-containing and control items on the same ward. A copper-containing: toilet seat, set of tap handles and a ward entrance door push plate were sampled and compared against equivalent standard items.

Results
In vitro activity - The viability of *Staphylococcus aureus, Escherichia coli, Klebsiella pneumoniae, Acinetobacter baumanii, Enterococcus spp.* and *Candida albicans* was progressively reduced by at least 3 log 10 cycles over 3 hours on copper but not stainless steel surfaces.

Clinical assessment – All copper-containing items harboured significantly fewer microorganisms (90%-100%) than their control equivalents.

Conclusions
Copper surfaces exhibit a pronounced antimicrobial action upon a range of pathogens, reducing viability over 3 hours contact at room temperature. Antimicrobial activity was also evident over a period of several months in the clinical setting. Copper surfaces may therefore, be a valuable tool in preventing nosocomial infection.

Introduction

- 1 in 10 patients who are admitted to UK hospitals subsequently contract a healthcare-associated infection.[1]
- This equates to 300,000 cases a year, costing the National Health Service in the UK around £1 billion annually.
- Environmental contamination has been implicated in the transmission of microorganisms in healthcare settings.[2]
- This has consequently stimulated interest in antimicrobial surfaces.
- The potential use of copper surfaces to reduce survival of common pathogens in the healthcare environment has been suggested following *in vitro* investigations proving the antimicrobial activity of copper.[3]

Objectives

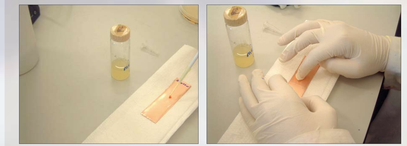
- To confirm the *in vitro* antimicrobial activity of copper against common hospital pathogens.
- To investigate the antimicrobial activity of copper in the clinical setting

In vitro evaluation of copper

Methods

- Microorganisms tested: Clinical isolates of: *E. coli* expressing extended spectrum beta-lactamase CTX M-15, Meticillin-sensitive *S. aureus* (MSSA), Meticillin-resistant *S. aureus* (EMRSA-15 and EMRSA-16), *A. baumanii, C. albicans, K. pneumoniae* and *E. faecium*.
- 0.05mL of each bacterial cell suspension (~10^9 cfu/mL) was placed on the surface of pure copper and stainless steel coupons (1cm x 1cm) and incubated for 3 hrs at room temperature.
- Viable cell counts were determined after resuspending the microorganisms from the coupon in 1mL of liquid medium (Difco™ D/E neutralising broth, BD),[4] serial dilution and standard microbiological culture.
- A further 0.01mL of bacterial suspension (EMRSA-15) (~10^9 cfu/mL) was placed on pure copper and stainless steel coupons under a glass cover slip with BacLight stain (SYTO 9/ propidium iodide, Molecular Probes) (Figure 1).
- Each coupon was viewed over time by epifluorescence microscopy (Zeiss Axioskop, Plan-NEOFLUAR 100x/1.30 objective, filter set 09) to observe the colour change of bacteria from green (live) to orange (dead).

Figure 1 - bacterial suspension with BacLight stain (SYTO 9/ propidium iodide) placed on metal coupon under a glass cover slip



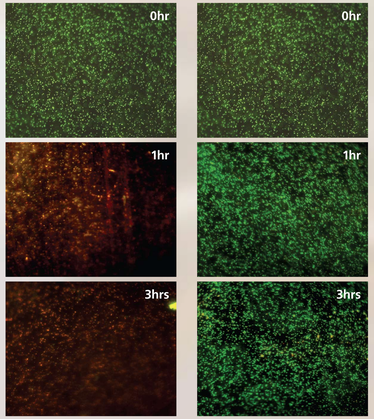
Results

- *In vitro*, the viability of *S. aureus, E. coli, K. pneumoniae, A. baumanii, E. faecium* and *C. albicans* was reduced by at least 3 log 10 cycles over 3 hrs on copper. No reduction was observed on stainless steel (table 1).
- By epifluorescence, the lethal action of copper on EMRSA -15 over 3 hours was demonstrated (figure 2). Indeed, the majority of EMRSA-15 cells were killed after 1 hr and complete kill was achieved following 3 hrs.

Table 1 – log reductions in viable count of clinical isolates following 3 hrs exposure to copper and stainless steel.

Microorganism	Log_{10} reduction (cfu/mL) on copper	Log_{10} reduction (cfu/mL) on stainless steel
E. coli (ESBL)	>5	0
S. aureus (MSSA)	>5	0
EMRSA 15	3.8	0
EMRSA 16	4.5	0
E. faecium	3.7	0
C. albicans	>5	0
K. pneumoniae	>5	0
A. baumanii	4.1	0

Figure 2 – Killing of EMRSA-15 on a copper and stainless steel surface at room temperature (green = live cells, orange = dead cells)



Clinical evaluation of copper

Methods

Clinical protocol

- A pilot study assessed the number of microorganisms on copper-containing toilet seats, grab rails, tap handles, light switches and door push plates items on a busy medical ward. The copper-containing items harboured fewer microorganisms than standard items on a control ward (p=0.01).
- The study design was adjusted so that copper-containing and control items on the same ward were sampled.
- A copper-containing: toilet seat (CuOF composite, approx 70% Cu), set of tap handles (CuZn, 60% Cu) and a ward entrance door push plate (CuZn, 70% Cu) (figure 3) were sampled and compared against equivalent items with plastic, chrome and aluminium surfaces, respectively.

Sampling protocol

- Items were sampled 1 day every week for 10 weeks at 7am and 5pm to determine the numbers of microorganisms present following quiet and busy time periods, respectively.
- Five weeks into sampling, the copper-containing and non-copper containing items were swapped over to exclude the possibility of preferential use of either particular item based on location.
- Each item was sampled in duplicate at each time point, selecting adjacent areas so that no area was sampled twice in any one day.
- For each toilet seat and door push plate sample, a sterile nasopharyngeal swab moistened in sterile 0.9% (w/v) saline was firmly applied 15 times horizontally and 15 times vertically over a 5cm x 5cm area using a sterile plastic template.
- The same methodology was applied to the tap handles over a 5cm x 2cm area.
- Each swab was immediately transferred to 3mL sterile neutralising broth (Difco™ D/E neutralising broth, Difco, BD).

Microbiological methods

- The neutralising broth containing the swabs were vortexed for 1 minute prior to dilution and inoculation onto a range of microbiological culture media agar plates.
- Following aerobic and anaerobic incubation at 37°C for 48 hours, the total cfu were enumerated and identified by standard microbiological techniques.
- The mean total cfu count for each duplicate set was entered into non-parametric statistical analysis.

Figure 3 – copper-containing door plate, toilet seat and taps

Results

- All copper-containing items harboured between 90% and 100% fewer microorganisms (median values) than their control equivalents at both 7am and 5pm (table 2).
- This reached statistical significance using both paired- and non-paired non-parametric statistical analysis with one exception.
- This copper-containing hot tap handle sampled at 5pm harboured 100% fewer microorganisms (median value) than the equivalent control. This reached significance in non-paired analysis.

Table 2 – median cfu counts on copper-containing items compared to controls in the clinical setting

Item	Median cfu count per control area (range)	Median cfu count per copper area (range)	Median copper cfu count as % of control cfu count (range)	Wilcoxon p value	Mann-Whitney p value
Top of toilet seat (7 am)	2190 (225-6660)	5 (0-960)	6 (0-33)	0.002	<0.0001
Top of toilet seat (5 pm)	1613 (705-6360)	30 (0-585)	2 (0-15)	0.002	<0.0001
Underside of toilet seat (7 am)	270 (0-2535)	0 (0-105)	2 (0-129)	0.02	0.007
Underside of toilet seat (5 pm)	38 (0-3045)	0 (0-105)	0 (0-220)	0.03	0.02
Push plate (7 am)	45 (0-195)	0 (0-6)	0 (0-100)	0.004	0.0002
Push plate (5 pm)	15 (0-84)	0 (0-30)	0 (0-100)	0.02	0.009
Hot tap handle (7 am)	66 (0-5040)	0 (0-30)	10 (0-*)	0.02	0.02
Hot tap handle (5 pm)	30 (0-360)	0 (0-390)	0 (0-2700)	0.2	0.02
Cold tap handle (7 am)	75 (0-870)	0 (0-30)	0 (0-100)	0.02	0.005
Cold tap handle (5 pm)	45 (0-510)	0 (0-30)	0 (0-100)	0.02	0.005

'*' indicates where a positive cfu count was present on copper surfaces compared to a zero count on standard.

Conclusions

- **These results demonstrate that copper offers the potential to significantly reduce the numbers of microorganisms both *in vitro* and in the clinical environment.**
- **The use of copper in combination with optimal infection prevention strategies may consequently reduce the risk of patients acquiring infections in hospital and other healthcare environments.**

References

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Acknowledgements

We acknowledge the following:

- Copper Development for a education grant to support this research
- Herck Metal Europe BV, Avilion Ltd and Aalco Ltd for supplying the copper toilet seat, taps and push plates, respectively
- Peter Nightingale for statistical advice

G2008-H1560 Designed and Printed by Medical Illustration (the graphics department for UHB) ☎0121 204 1594