

What science do we have to
prove our cleaning and
disinfection is effective



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Environmental cleaning and disinfection

- Cleaning and disinfection of environmental surfaces is fundamental in reducing pathogen transmission
- The risk of acquiring an MRO is increased an average of 73% if previous patient had an MRO
- HCW hands are just as likely to be contaminated from the environment than patient
- Enhanced cleaning ↓ surface contamination and ↓ nosocomial infection
- MROs can be protected in biofilms
- **CLEANING** is a PREREQUISIT to **EFFECTIVE** disinfection/sterilisation



Auditing processes

1. Visual audit or inspection
2. UV solution and fluorescent light inspection
(Mitchell -3)
3. ATP assay, A3 (ATP,ADP,AMP)
4. Culture –aerobic colony counts
5. Protein assay –instruments. In-situ based assays
good but swab sticks poor sensitivity
6. PCR and microscopy (research only)

Mitchell et al 2017 Infection, Disease and Health
2017;22: 195-202.

Visual assessment

- Mainstay of Australian facilities (Mitchell, 2017 –all 11 acute care hospitals)
- Need procedures in place eg clip board of items to look at
- If it looks dirty it is; if it looks clean it might not be
 - Whiteley (2018) showed that 68% of surfaces assessed visually as clean weren't
- Can't detect if pathogens are present

Fluorescent markers

- Shows if a surface has been wiped or not, therefore very good as a training/retraining tool
- Some correlation between removal, partial / non-removal of marker and ATP RLU
- No correlation with CFU counts
- However, in real-life hospital situations less sensitive than ATP measurements. Hung 2018 found total removal of marker equated with
 - $428.7 \pm 1,180.0$ RLU (3M benchmark 100) and 15.6 ± 77.3 CFU/100 cm².

Hung et al. Application of a fluorescent marker with quantitative bioburden methods to assess cleanliness. Infect Control Hosp Epidemiol 2018;39: 1296-300.

Portable ATP bioluminometers

- ATP occurs in food, blood, microorganisms – therefore not directly related to the number or pathogenicity of microorganisms
- Measure of contamination with biological soil

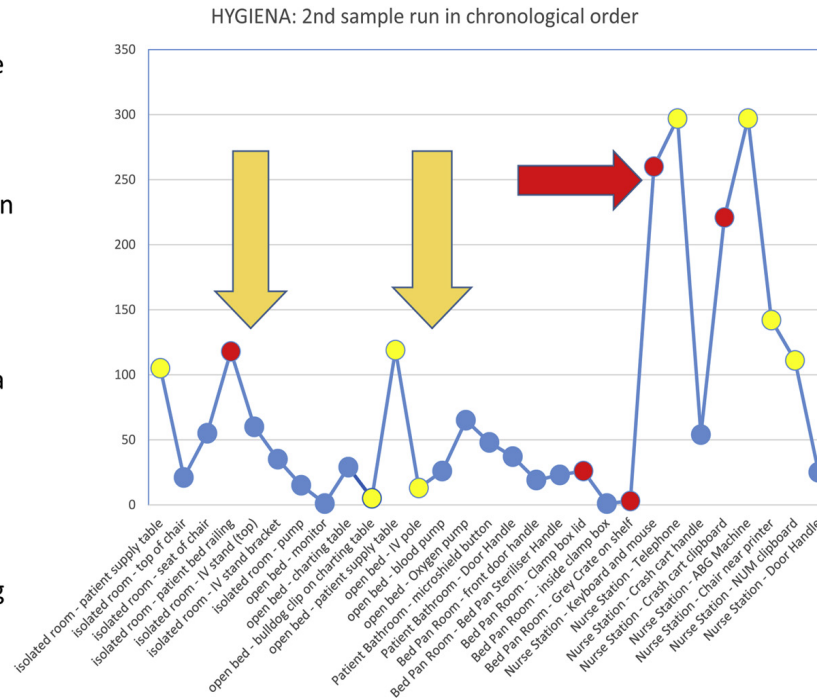


Reasonable tool for determining if a surface is clean but cant determine if surface is pathogen free

1st arrow: immediate patient zone (IPZ) sampling including around an MRSA + patient in an isolation room⁸

2nd arrow: moving away from IPZ and into non-clinical area including bed pan room (looking for VRE)²⁵

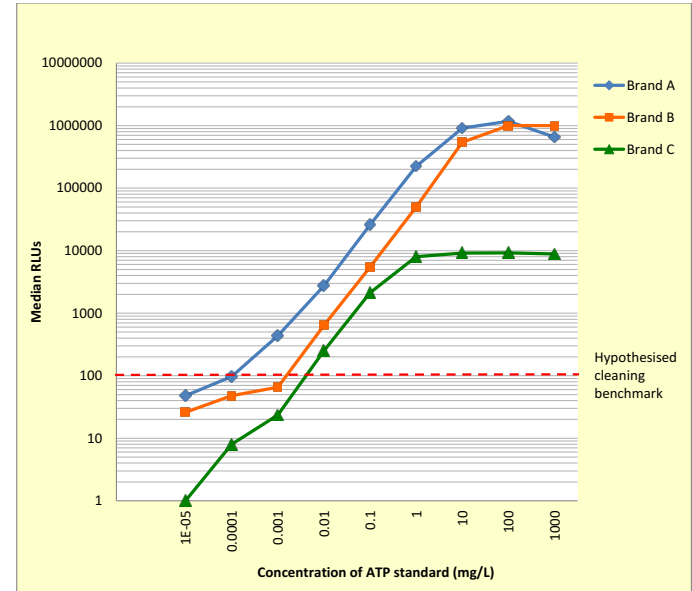
3rd (RED) arrow: moved into sampling around the nurses/clinical work station¹⁰



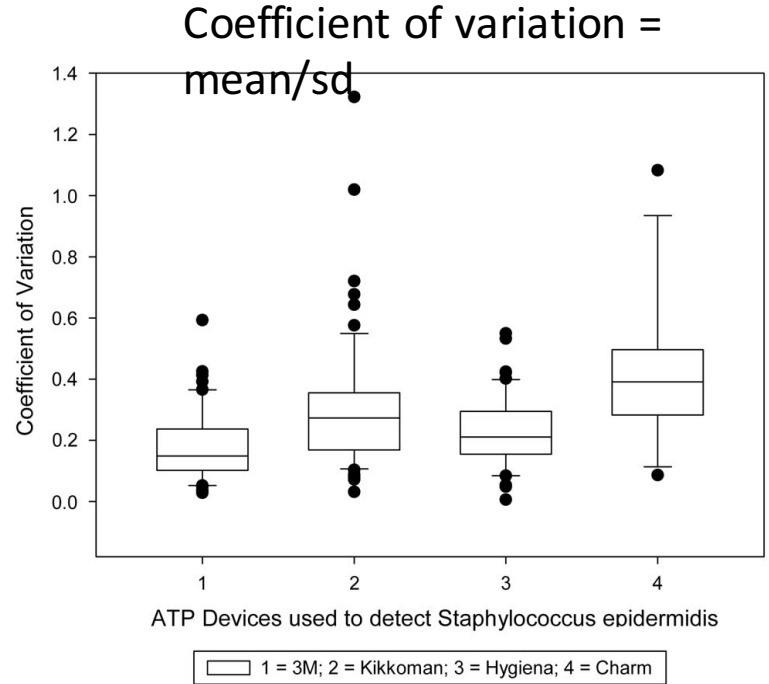
Yellow dots indicate paired data points with Hygiena, Kikkoman and Micro sampling but negative for MDRO

Red dots indicated paired sampling locations with Hygiena, Kikkoman and Micro sampling with +ve MDRO recovery.

1. ATP brands show good range and linearity over a log scale of ATP detection
2. poor standardisation between brands-100RLU
3. 100RLU hypothetical cut off – difference in amount of soiling almost 2logs (100 fold)
4. depending on the brand 0 RLU doesn't mean no ATP



1. Portable ATP bioluminometers are not so great in a narrow range
2. standard deviation can be as high as 50%



A3 test report by Kikkoman employees

- ATP degrades to ADP degrades to AMP
- LuciPac A3 Surface/Lumitester PD-30 (Kikkoman Biochemifa, Tokyo, Japan)
- A3test RLU 7.5 fold higher than ATP alone
- No measurements of variability

Bakke M et al. Evaluation of the total adenylate (ATP + ADP + AMP) test for cleaning verification in healthcare settings. *Journal of Preventive Medicine and Hygiene* 2019;60: E140-E6.

Total aerobic counts

- Advantages
 - Absolute number of aerobic, easy to culture bacteria known
 - Will pick up some pathogens eg *Staphylococcus aureus* including MRSA
 - Usually used in outbreak situations
- Disadvantages
 - Not all pathogens cultured
 - Takes 24 -48 hours for counts and identification

Comparison of methods

- Huang et al 2015. % surfaces classified as dirty:
visual inspection 12%,
aerobic culture count CFU>2.5/cm² 20%
3M ATP>50RLU 50% , common benchmark 100 RLU
- Synder 2013.
In comparison to aerobic culture counts:

	sensitivity %	specificity %
Visual	60	52
Marker	51	56
ATP	70	44

3M ATP >250 RLU as dirty –would have higher sensitivity if set at 100 RLU

Common problem with ATP and culture

Swab collection efficient at picking up planktonic bacteria but is inefficient at picking up biofilm and fixed on protein

Biofilms are in our hospital environments



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Hospital	Items	Mean Microbial Load/cm ²	Culture Positive	MDRO cultured	Biofilm positive %
Brazil	40	4.4x10 ³	26	4	100
Australia	44	5.5x10 ⁵	23	13	93
Scotland	8	2.6x10 ⁴	4	2	100
Saudi Arabia	20	3.4 x10 ⁴	13	0	70

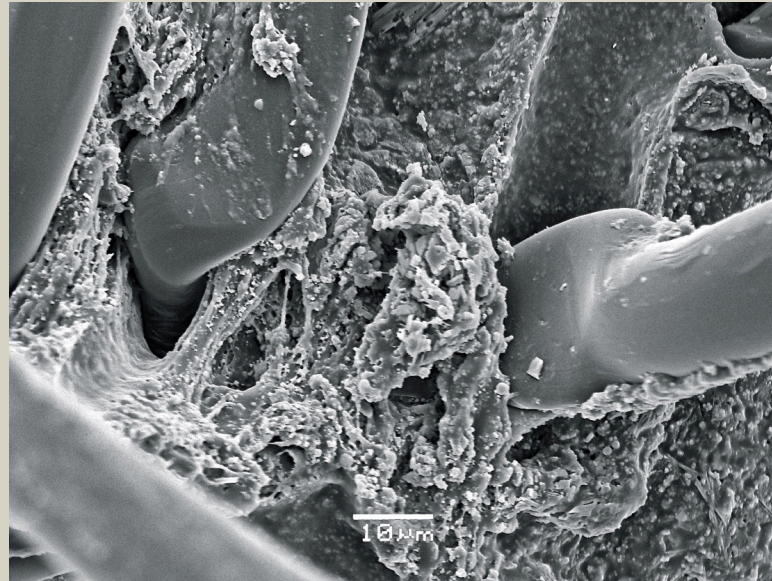
- 3 different UK hospitals,
 - DSB was found on 95% of 61 samples
 - MRSA grown from 58% of samples
-
- K. Ledwoch, S.J. Dancer, J.A. Otter, K. Kerr, D. Roposte, J.-Y. Maillard [10.1016/j.jhin.2018.06.028](https://doi.org/10.1016/j.jhin.2018.06.028)
 - Hu H, Johani K, Gosbell IB, Jacombs A, Almatroudi A, Whiteley GS, Deva AK, Jensen S and Vickery K. *J Hospital Infection* ;2015;91:35-44. DOI [10.1016/j.jhin.2015.05.016](https://doi.org/10.1016/j.jhin.2015.05.016)
 - Johani K, Abualsaud D, Costa DM, Hu H, Whiteley G, Deva A and Vickery K. *J Infect Public Health* 2017 DOI: [10.1016/j.jiph.2017.10.005](https://doi.org/10.1016/j.jiph.2017.10.005)

Where are biofilms found?

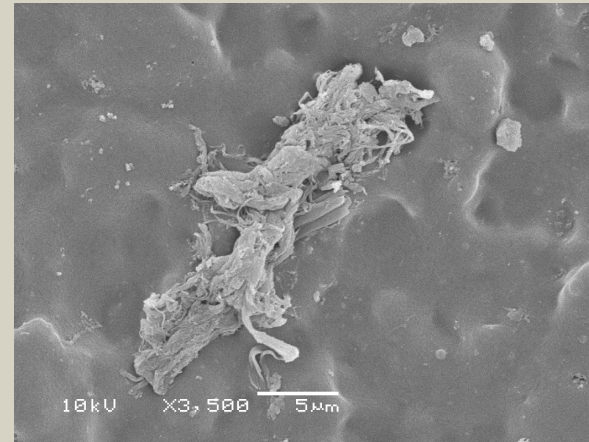
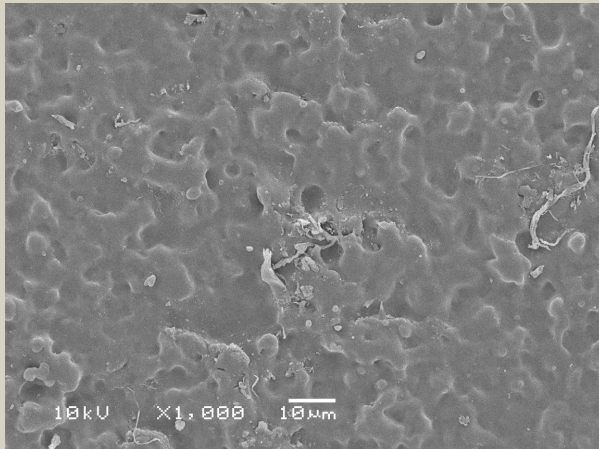


Answer – everywhere including on visually clean surfaces

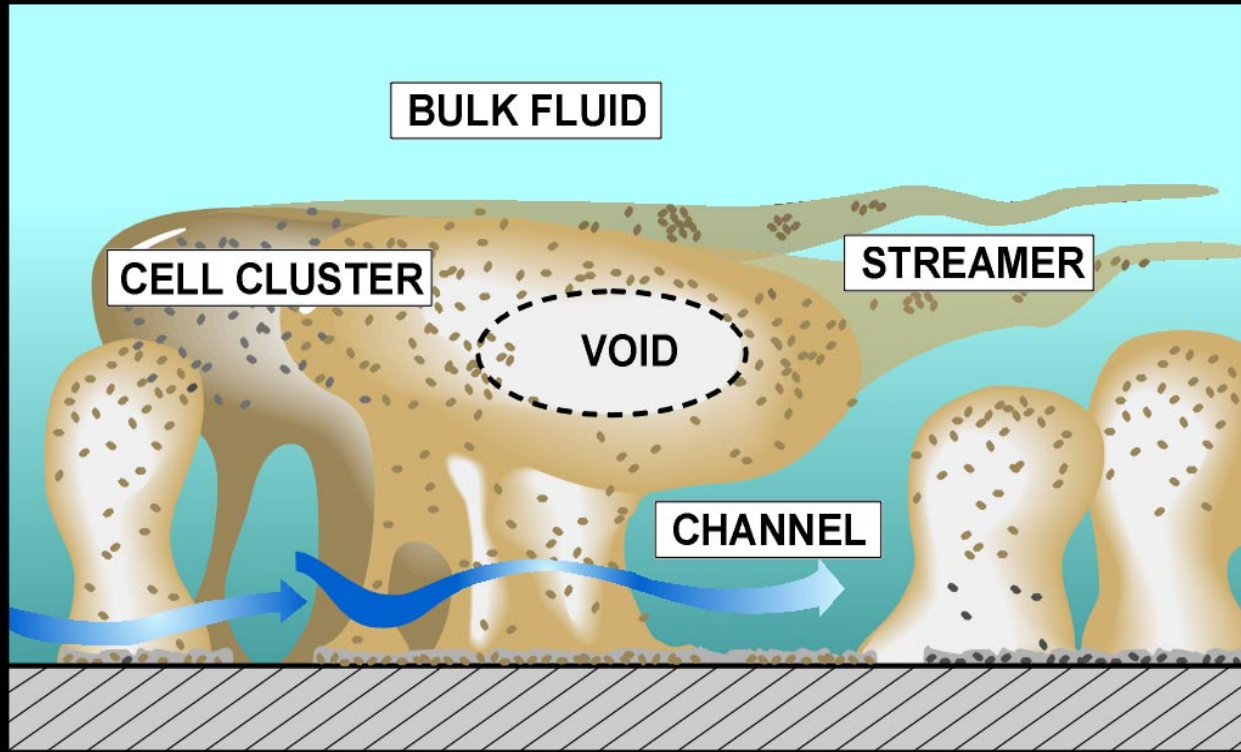
Ultrasound pillow



Mattress - MRSA, VRE, ESBL positive

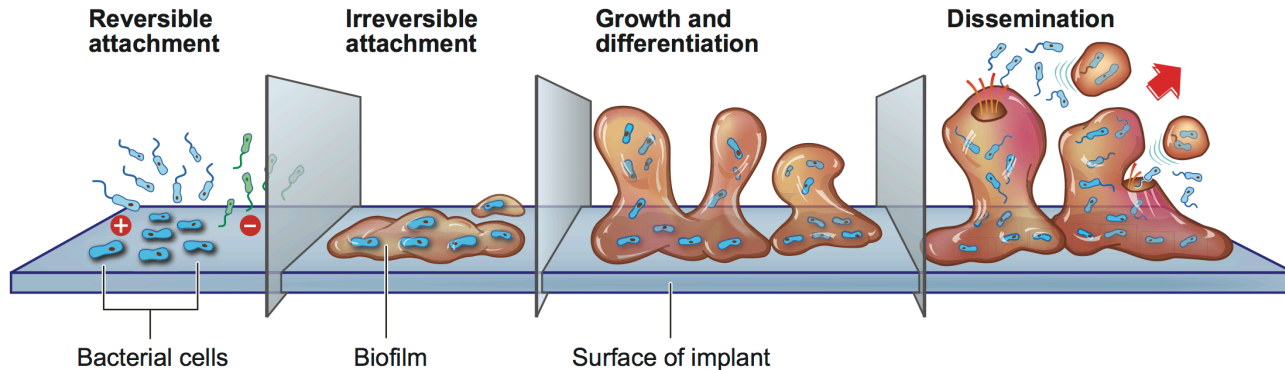


What is a biofilm?

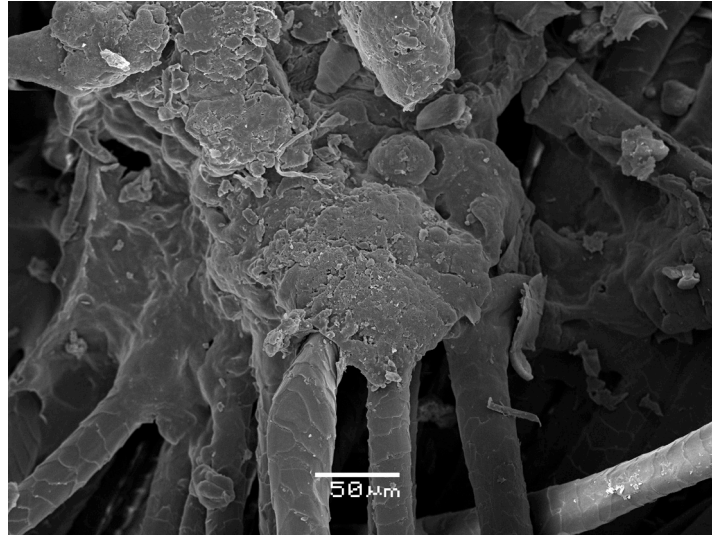


Why are biofilms important?

- Dissemination/transmission
- Often difficult to detect –negative cultures
 - Sampling, low metabolic rate
- Resistant to desiccation
- Increased resistance to removal
- Decrease disinfectant efficacy

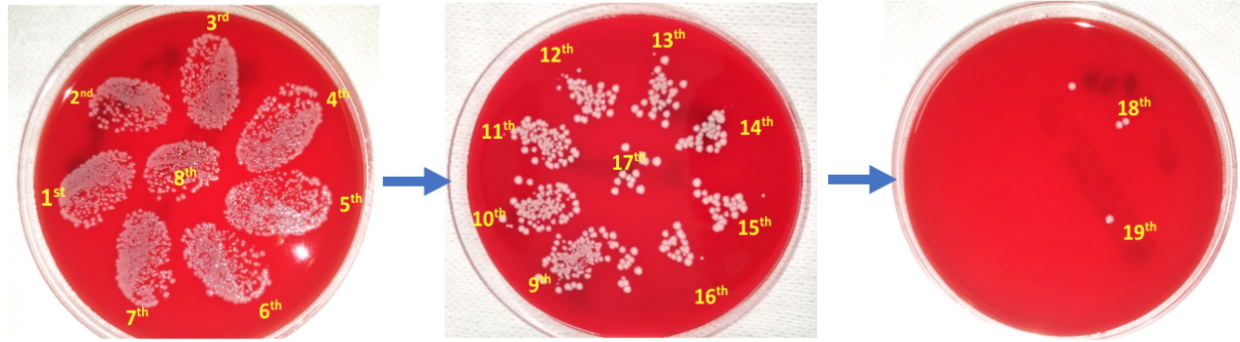


Biofilm is covered by EPS and stuck to a surface so is it transferred?



Clinical station chair
VRE positive

Frequency of transmission of bacteria

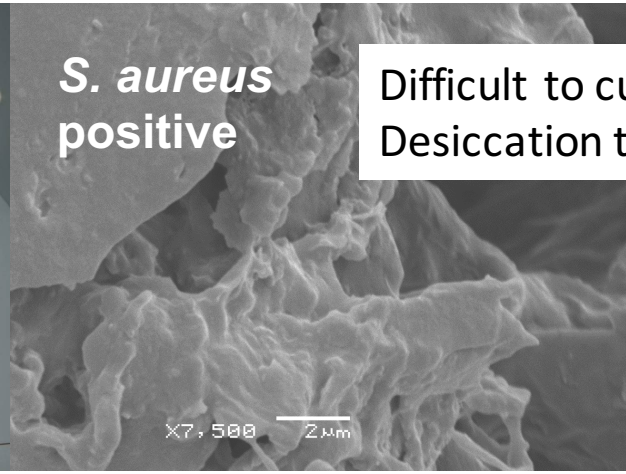


- Approximately 5-6% of the DSB is transferred to the hands following one touch
- Approx. 20% of the DSB on the hands are transferred to HBA
- 1% of the DSB is transferred from one fomite (coupon) to Hand to fomite (HBA)
- Clinical surfaces contaminated with mean of 10,000 bacteria/cm² (up to 10⁷)
- 1% = 100 bacteria. 15 *S. aureus* cause a nosocomial infection.

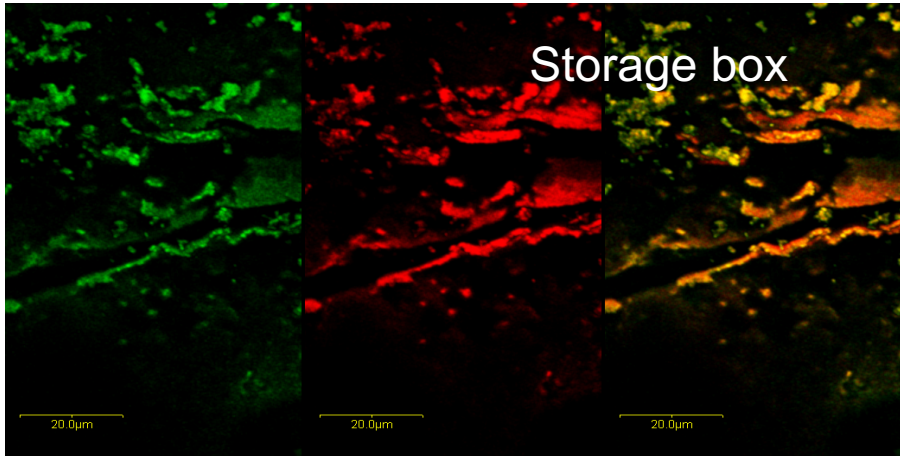


S. aureus
positive

Difficult to culture
Desiccation tolerant



X7,500 2 μm

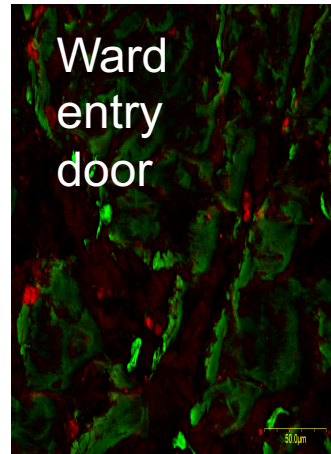


Storage box

20.0 μm

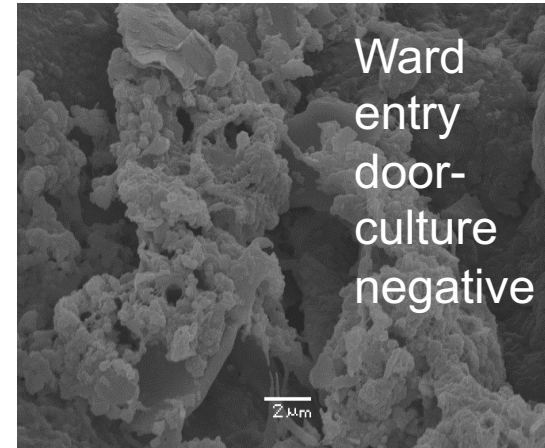
20.0 μm

20.0 μm



Ward
entry
door

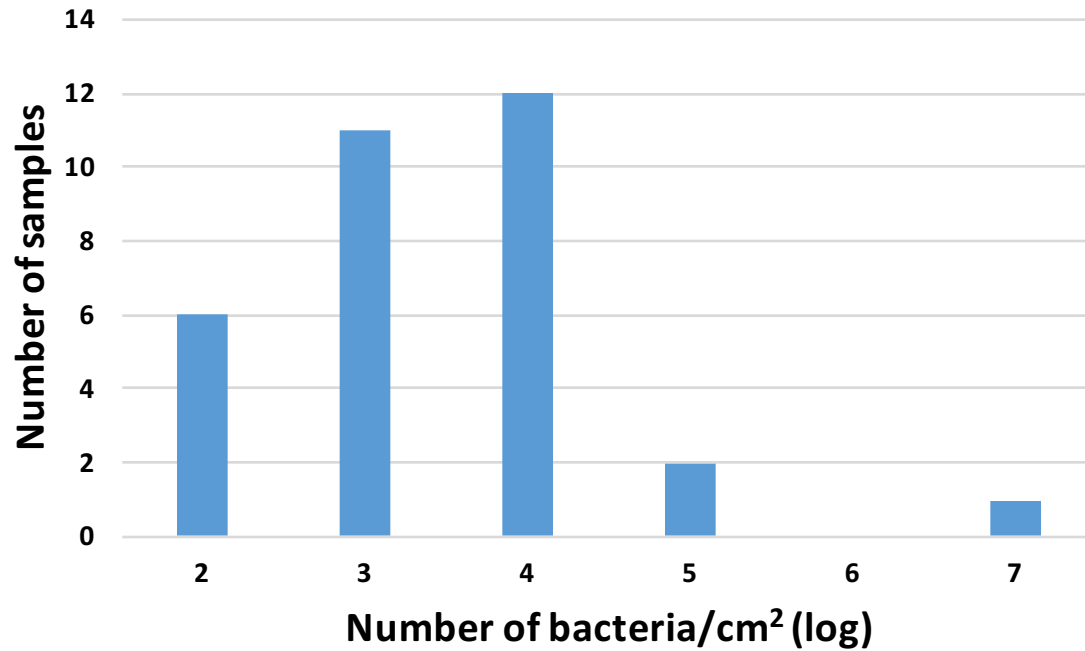
50.0 μm



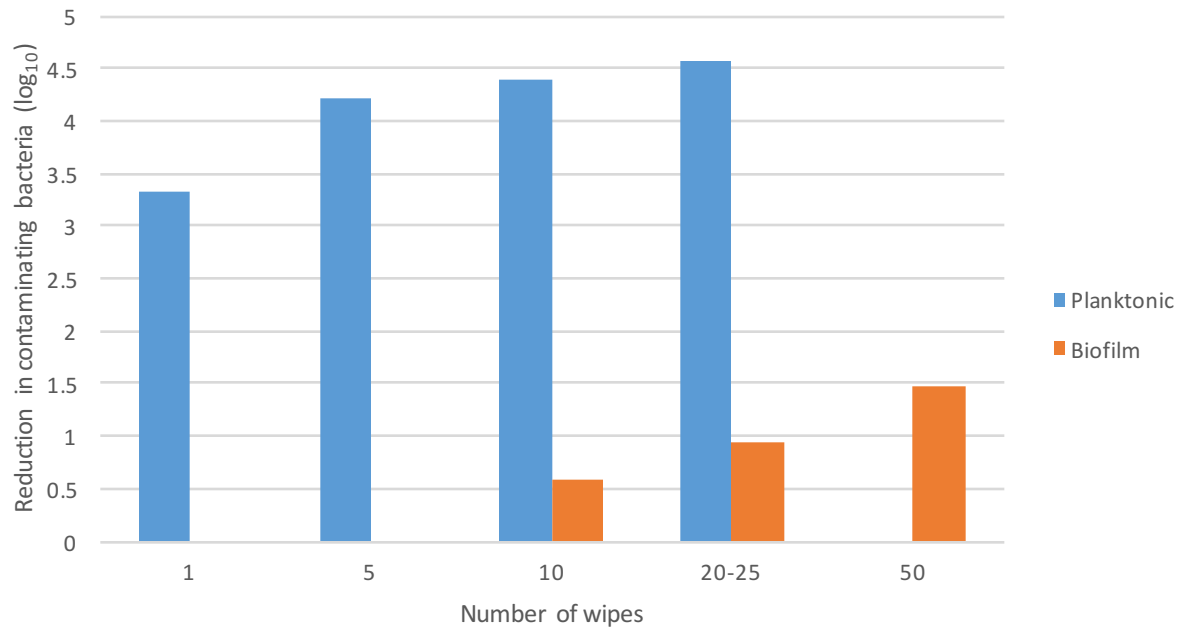
Ward
entry
door-
culture
negative

2 μm

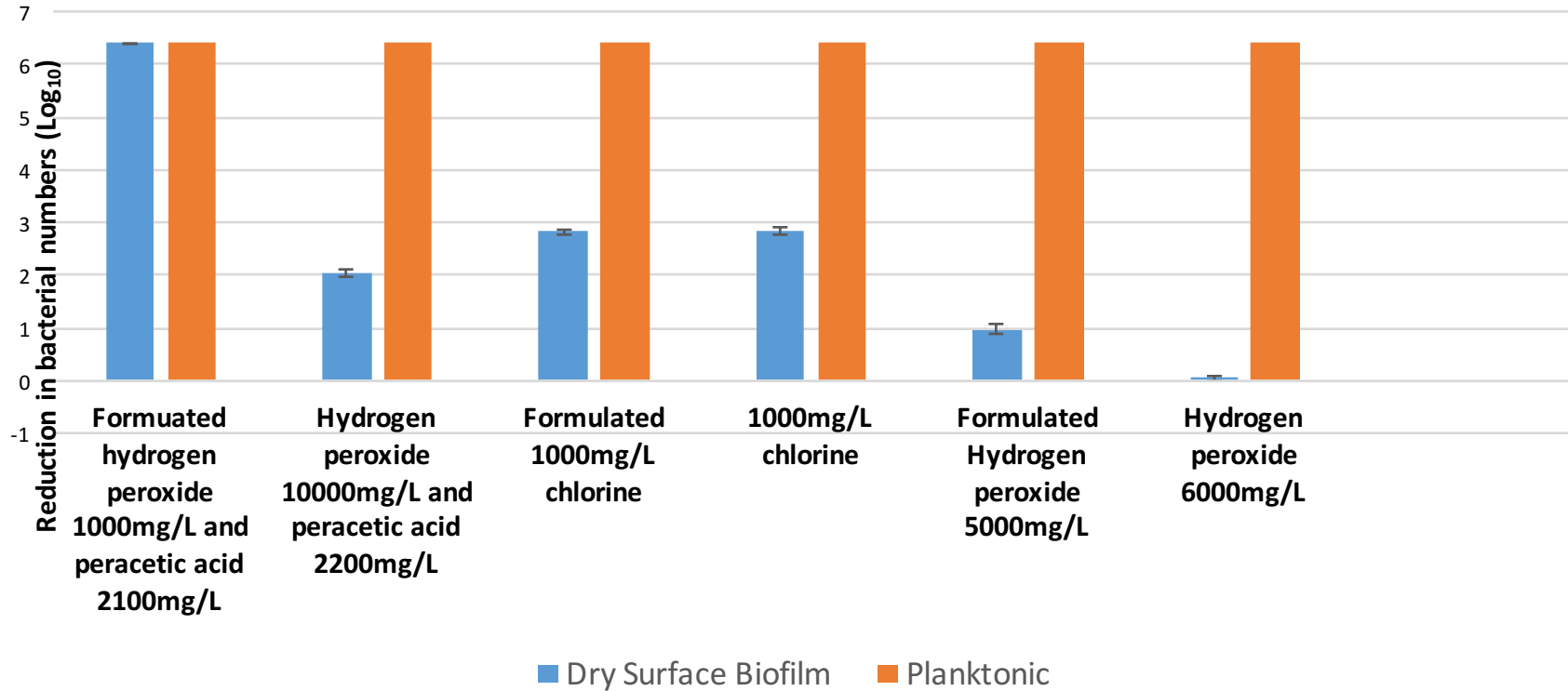
Number of bacteria per cm² (n=32) by PCR



Hard to physically remove

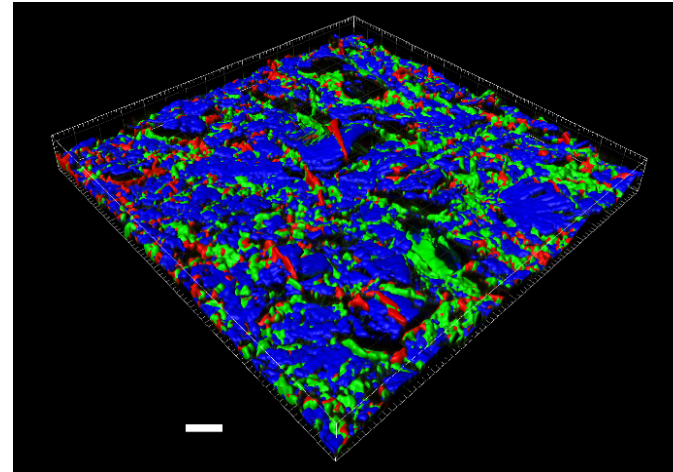
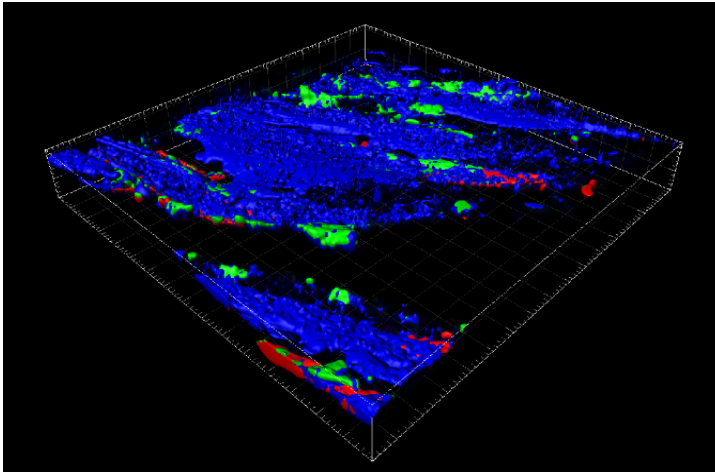


Decreased disinfectant efficacy



Mechanisms of resistance: EPS – extracellular polymeric substances or “slime”

- EPS 85-90% of the biofilm
- Composed of polysaccharides, proteins, DNA
- Hydrated biofilm 90% water – protects against desiccation
- Has been shown to play a major role in disinfectant tolerance by decreasing disinfectant penetration and inactivation of disinfectant.

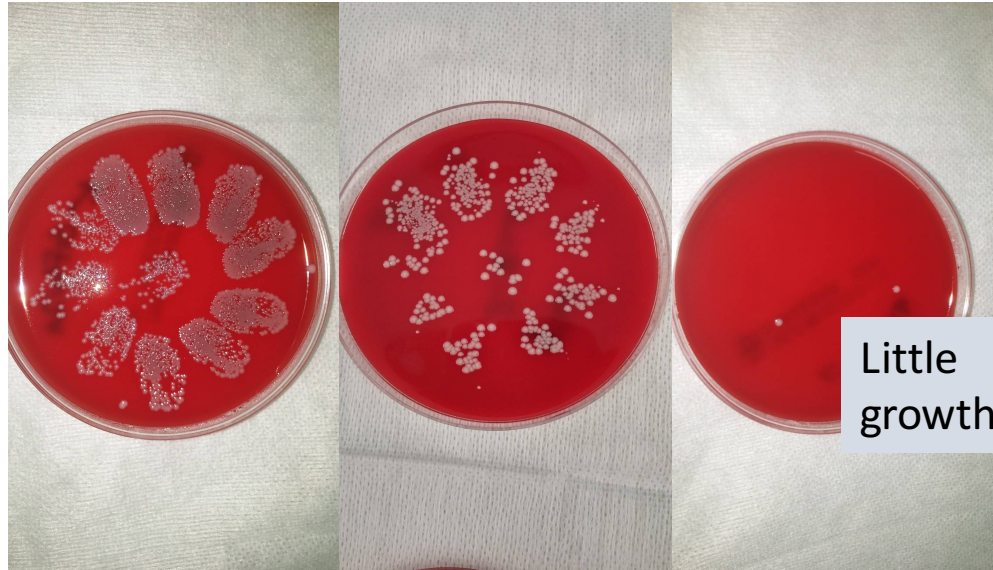


Infection control implications

- Biofilm are present on dry surfaces of the ICU
- Multi-species including aerobic, facultative anaerobic and anaerobic organisms
- Composed of organisms from the skin, gut and environment
- Can contain and protect pathogens including MROs, DHBV
- Bacteria can be transmitted from biofilms to other surfaces
- Any soil, including biofilms can decrease the efficacy of disinfection and sterilisation
- Biofilms have increased tolerance to disinfectants.
- This may be one of the mechanisms by which MROs persist within the hospital environment and contribute to

HAI

Should we give up? **NO, NO, NO**



Control transmission

**Post chlorine
disinfection**

What to do now

- Ensure that all surfaces are cleaned- Monitoring
 - Visual inspection – check sheets
 - Removal of fluorescent dyes
 - ATP
 - Microbiological monitoring

What to do now – improve cleaning

Increased resistance to biocides is lost if the biofilm is dispersed

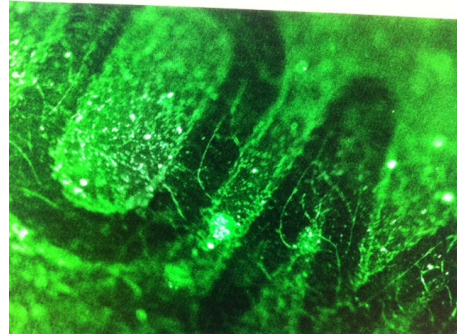
- Dentistry – scale teeth
- Industry treatment of biofilm contamination requires some sort of physical removal prior to chemical disinfection
- We cant scrape hospital surfaces but do we need to return to removing biofilm physically instead of relying on only chemistry ie use more elbow grease?

What to do now – IMPROVE CLEANING

- Increased training (+ increased pay) for cleaners –
 - Poorly trained,
 - Low paid
 - High turn over
- Increase number of cleaners
- one surface, one direction, one wipe
 - Which wipe to use where
 - Order of cleaning
- Remove gloves/wash hands between areas
- Ensure surfaces are cleanable
- Validate cleaning
- Ensure you are using a product that is designed for the job

The future

- Improved furnishings –
 - inhibit bacterial attachment
 - Kill bacteria – heavy metals eg copper
- Dispersal of bacteria by quorum sensing molecules
- Improved killing with new combined biofilm dispersal agent and disinfectant – combination approach
- Anti-biofilm surface coatings for furnishings, equipment and implants-
 - eg copper.



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