

# A NEW ANTI-BIOFILM HYDROFIBER® DRESSING: IN VITRO DETERMINATION OF MICROBIAL KILL RATE IN BIOFILMS



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#### Introduction

In their natural habitat, bacteria usually exist in communities encapsulated in a matrix of gelatinous extracellular polymeric substances (EPS) which is attached to a surface. This is known as biofilm.¹ Biofilm EPS is produced by the bacteria and serves as a protective niche for survival. It renders them more tolerant to the host immune response and antimicrobial therapies alike.¹²² Biofilm is increasingly recognised as being a barrier to wound healing and there is a growing evidence that biofilms are a cause of wound chronicity¹¹²,²³ – with studies demonstrating biofilm prevalence of at least 60% in chronic wounds.³ Therefore, to be clinically useful, antimicrobial therapies must be effective against biofilm.

#### Aim

This poster describes *in-vitro* studies used to measure the anti-biofilm properties of a new, custom-designed anti-biofilm Hydrofiber® dressing, **AQUACEL® Ag+**.

### Method

## Study 1

Biofilms of challenge organisms (Fig 1) were grown on sterile cotton gauzes by immersion in a bacteria-containing simulated wound fluid at 35°C for ≥ 48 hours. Gauzes were then rinsed with isotonic saline to remove any unattached bacteria. The presence of biofilm was confirmed by scanning electron microscopy (SEM). Biofilm-colonised gauzes were exposed to test dressings (Fig 2) hydrated with isotonic saline for 4, 24 and 48 hours at 35°C. Bacterial bioburden was determined by processing the gauzes in a neutralising diluent and then assaying by standard microbiological techniques.

#### Study 2

Biofilms were prepared as in study 1, then a PHMB gauze (Fig 2) was applied for a further 48 hours to eliminate any planktonic cells whilst maintaining growth of a predominately biofilm population. AQUACEL® Ag+ dressing samples were then applied hydrated with simulated wound fluid and incubated at 35°C for up to 96 hours. Quantitative analysis was performed as previously described.

Figure 1. Challenge organisms

Study	Challenge organism
1	Pseudomonas aeruginosa (NCIMB 8626)
1	Staphylococcus aureus (NCIMB 9518)
1	Candida albicans (NCPF 3179)
2	P. aeruginosa (wild type strain PA01)

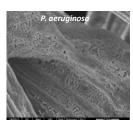
Figure 2. Test dressings

Coding	Material	Product	Manufacturer
ABHF	Anti-biofilm Hydrofiber®	AQUACEL® Ag+ (CE Marked 2013)	ConvaTec
SHF	Silver-containing Hydrofiber ®	AQUACEL® Ag	ConvaTec
HF	Hydrofiber®	AQUACEL®	ConvaTec
PHMB- Gauze	Polyhexamethylene biguanide (PHMB)- containing non-adherent gauze	Telfa™ AMD	Covidien

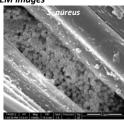
#### Results

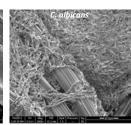
SEM confirmed the presence of biofilm on the cotton gauze substrates, including after PHMB gauze (as in Study 2; Figure 3):

Figure 3. SEM images

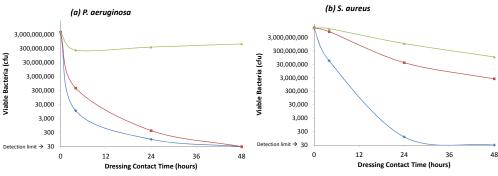


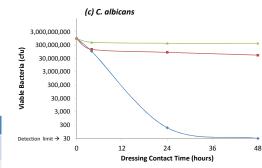






## Results (cont.)





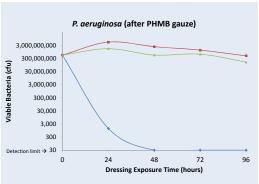


Figure 4. Study 1 − Biofilm reduction (a) P. aeruginosa; (b) S. aureus; (c) C. albicans. (♠) AQUACEL® Ag+; (■) AQUACEL® Ag; (♠) AQUACEL®

Figure 5. Study 2 – Biofilm reduction after PHMB gauze. ( $\blacklozenge$ ) AQUACEL® Ag+; ( $\blacksquare$ ) AQUACEL® Ag; ( $\triangle$ ) AQUACEL®

#### Discussion & Conclusion

Biofilm bacteria counts of potential wound pathogens fell below the limit of detection within 48 hours for the AQUACEL® Ag+ dressing. Approximately an 8 log reduction (100 million-fold) reduction in biofilm bacteria was achieved for each challenge organism for the AQUACEL® Ag+ dressing. A >3 log reduction was achieved within 4 hours for *S. aureus* and *P. aeruginosa* biofilm bacteria.

Three potential wound pathogens were shown to form dense biofilm on cotton gauze (*S. aureus, P. aeruginosa* and *C. albicans*). Biofilm forms of each potential pathogen were tolerant to two antimicrobial dressings (ionic silver and PHMB). Biofilm forms of each potential pathogen were highly-susceptible to a new anti-biofilm Hydrofiber® dressing, AQUACEL® Ag+.

#### References

- 1. Bjarnsholt T. The Role of Bacterial Biofilms in Chronic Infections. APMIS 2013; 121(Suppl. 136):1-51.
- 2. Metcalf DG, Bowler PG. Biofilm delays wound healing: A review of the evidence. Burns Trauma 2013; 1:5-12.
- 3. James GA, Swogger E, Wolcott R, et al. Biofilms in chronic wounds. Wound Rep Reg 2008; 16:37-44.