

# VISUALIZING THE DEATH OF WOUND MICROORGANISMS WITHIN A SILVER ALGINATE DRESSING

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#### INTRODUCTION

RESTORE Calcium Alginate Dressing with Silver is now widely used in the treatment of infected chronic wounds, combining the broad spectrum antimicrobial action of silver with the exudate management ability of alginate technology.

#### **AIMS & OBJECTIVES**

This study used confocal laser scanning microscopy (CLSM) and live/dead staining, to visualize the viability of wound microorganisms sequestered into a silver alginate wound dressing (RESTORE Calcium Alginate Dressing with Silver). The antimicrobial efficacy of RESTORE Calcium Alginate Dressing with Silver was compared with that of a non-antimicrobial Alginate dressing, by assessing the proportion of live/dead microorganisms over a 16 hour time course.

#### **MATERIALS & METHODS**

Reference strains of the frequently encountered wound pathogens Pseudomonas aeruginosa (ATCC 15692), Staphylococcus aureus (NCTC 8325) and Candida albicans (ATCC 90028) were cultured in Mueller Hinton broth until mid-log phase. Suspensions of these microorganisms (approx. 108 cfu/mL) were stained with the Baclight live/ dead viability kit (Invitrogen), and 100 µL of the preparations were used to hydrate the control alginate and the RESTORE Calcium Alginate Dressing with Silver and placed on glass slides. Fibers were then monitored by rapid CLSM with images taken at 20 minute intervals for up to 16 hours. The relative proportions at different time points of live and dead cells (appearing green and red, respectively) was assessed using image analysis software (Leica).

#### RESULTS

When the dressings were hydrated with stained microbial culture the alginate fibers swelled quickly, causing immobilization of the microorganisms in the gel-like spaces between the fibers. With the silver-free dressing, all microbial species quickly formed aggregates of viable cells. In comparison, when the same organisms were added to RESTORE Calcium Alginate Dressing with Silver they progressively appeared red (i.e. dead) within the first hour.

**Figure 1** shows that both *Pseudomonas aeruginosa* and *Staphylococcus aureus* were rapidly killed within the RESTORE Calcium Alginate Dressing with Silver. In the case of *P. aeruginosa*, total kill was evident within 16 hours, while for *S. aureus* 4 hours was sufficient time for a total cidal effect to be observed. Over the same period the control alginate dressing did not exhibit an antimicrobial effect.

The broad spectrum antimicrobial effect of silver was evident when *C. albicans* was exposed to the dressing fibers. This fungal organism was also killed over a 16 hour exposure period (**Figure 2**) with no equivalent fungicidal effect observed in the control.

#### CONCLUSIONS

The ability of the RESTORE Calcium Alginate Dressing with Silver to quickly sequester and kill potentially pathogenic microorganisms, as demonstrated in this study, highlights the value of this dressing in reducing wound bioburden, thus potentially generating an environment supportive to the healing of infected wounds.

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## **RESULTS (continued)**

Figure 1. CLSM and Live/dead staining of wound bacterial pathogens in test dressings



### Live bacteria appear green, dead bacteria appear red

Figure 2. CLSM and Live/dead staining of *Candida albicans* in test dressings









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