

The effects of silver-based wound dressings on protease activity and cell proliferation

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OBJECTIVE

To evaluate the effect of two silver-based antimicrobial dressings on the activity of elastase in-vitro, commonly found to be elevated in non-healing wounds. To assess the impact of the silver-based dressings on the proliferation of fibroblasts, known to be important in the normal wound repair process.

TEST DRESSINGS

- 1) A silver impregnated activated charcoal dressing (SIAC) - ACTISORB[®] Silver 220 Dressing, Johnson and Johnson Wound Management Worldwide, a division of ETHICON, INC.
 - 2) A rayon/polyester core laminated between layers of silver coated polyethylene mesh (ACT) - Acticoat[®] (with SILCRYST[™] nanocrystals), Antimicrobial Barrier Dressing, Smith & Nephew.
 - 3) SOF-WICK[®] - gauze control, Johnson and Johnson Wound Management Worldwide, a division of ETHICON, INC.
 - 4) ORC/Collagen - PROMOGRAN[®] Dressing, Johnson and Johnson Wound Management Worldwide, a division of ETHICON, INC.
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INTRODUCTION

Biochemical Balance in Wounds

Increasing evidence from research indicates that there is a delicate biochemical balance which exists within a normal wound healing environment. This balance can however be disturbed or disrupted by numerous factors, including, critical colonization by bacteria, or increased host inflammatory response, which may in turn result in impaired wound healing.

One such biochemical imbalance that can occur in non-healing wounds is elevated levels of proteolytic enzymes¹. Although proteolytic enzymes are important in the normal wound healing process elevated levels can lead to excessive local destruction of tissue components and growth factors increasing the probability of non-healing¹. One such proteolytic enzyme which has been shown to have elevated activity in non-healing wounds is elastase².

As discussed, critical colonization or infection can play an important role in causing biochemical imbalance in non-healing wounds, therefore a dressing which reduces these elevated levels may be beneficial in re-balancing the biochemical environment.

The Role of Fibroblasts

An integral part of the wound healing process is the migration and proliferation of fibroblasts. Fibroblasts produce a variety of substances including glycosaminoglycans and collagen which are essential in forming the extracellular matrix³, which eventually forms the major constituent of repaired skin.

Any wound dressing which has a detrimental effect on the fibroblast proliferation process may contribute to delayed wound healing.

METHODOLOGY (1)

Elastase Activity Assay

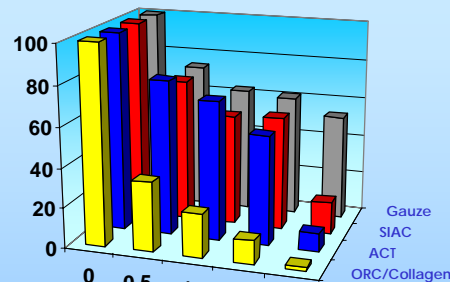
- 1) Punch biopsy (6mm) of dressings taken
- 2) Biopsies pre-wet with phosphate buffered saline
- 3) Dressing biopsies transferred to elastase solution
- 4) Samples (5µl) of solution (dressing biopsy + elastase) removed at 0, 0.5, 1, 2, and 24 hrs
- 5) Samples assayed for residual protease activity by adding samples to elastase substrate with attached fluorescent marker (methoxysuccinyl-ala-pro-val-7-amino-4-methylcoumarin). Any residual elastase reacted with the substrate and the fluorescent marker was released and detected by fluorimeter at 455nm.
- 6) Controls containing gauze or no dressing were also tested

METHODOLOGY (2)

Fibroblast Proliferation (Human Dermal)

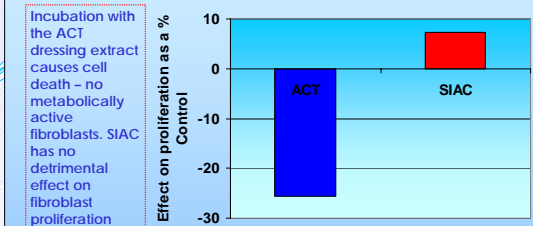
- Punch biopsies (6mm) of the dressings were taken
- Dressing biopsies were incubated in serum free DMEM for 24 hours at 37°C.
- Samples (100µl) of solution were taken and added to fibroblast cultures.
- Fibroblast cultures were incubated for 72 hours at 37°C.
- After incubation, a pre-mixed solution of the tetrazolium salt XTT and an activation reagent was added to the fibroblast cultures.
- The absorbance of the solutions at 450nm is read immediately and at 0.5, 1, 2, 2.5, 3, and 5hrs.

Figure 1. Activity of Protease over 24 hours in the presence of test dressing⁴



Both SIAC and ACT produced significant decreases in elastase activity compared to gauze and the negative control. ORC/Collagen (PROMOGRAN dressing) was included in the test as a positive control. This produced the largest decreases in elastase activity in this experiment.

Figure 2. Effect of DMEM extracts of test dressings on fibroblast proliferation versus control



The control is DMEM solution. Negative values in figure 2 indicate decreased viable cells compared to the control. The high negative value for the ACT extract indicated no viable cells after incubation.

REFERENCES

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CONCLUSIONS

After 24 hours exposure to elastase solution in-vitro, both test dressings were found to result in a significant reduction in protease activity when compared to a gauze control.

When a solution extract from each dressing was added to fibroblast cultures only ACTISORB Silver 220 dressing was found to have no detrimental effects on fibroblast proliferation.

These results suggest that silver-based dressings may help reduce protease activity although only ACTISORB Silver 220 dressing was found to have no detrimental effect on fibroblast proliferation in this in-vitro study.