

The Beneficial Effects Of A Non-Releasing Silver Containing Wound Dressings on Fibroblast Proliferation

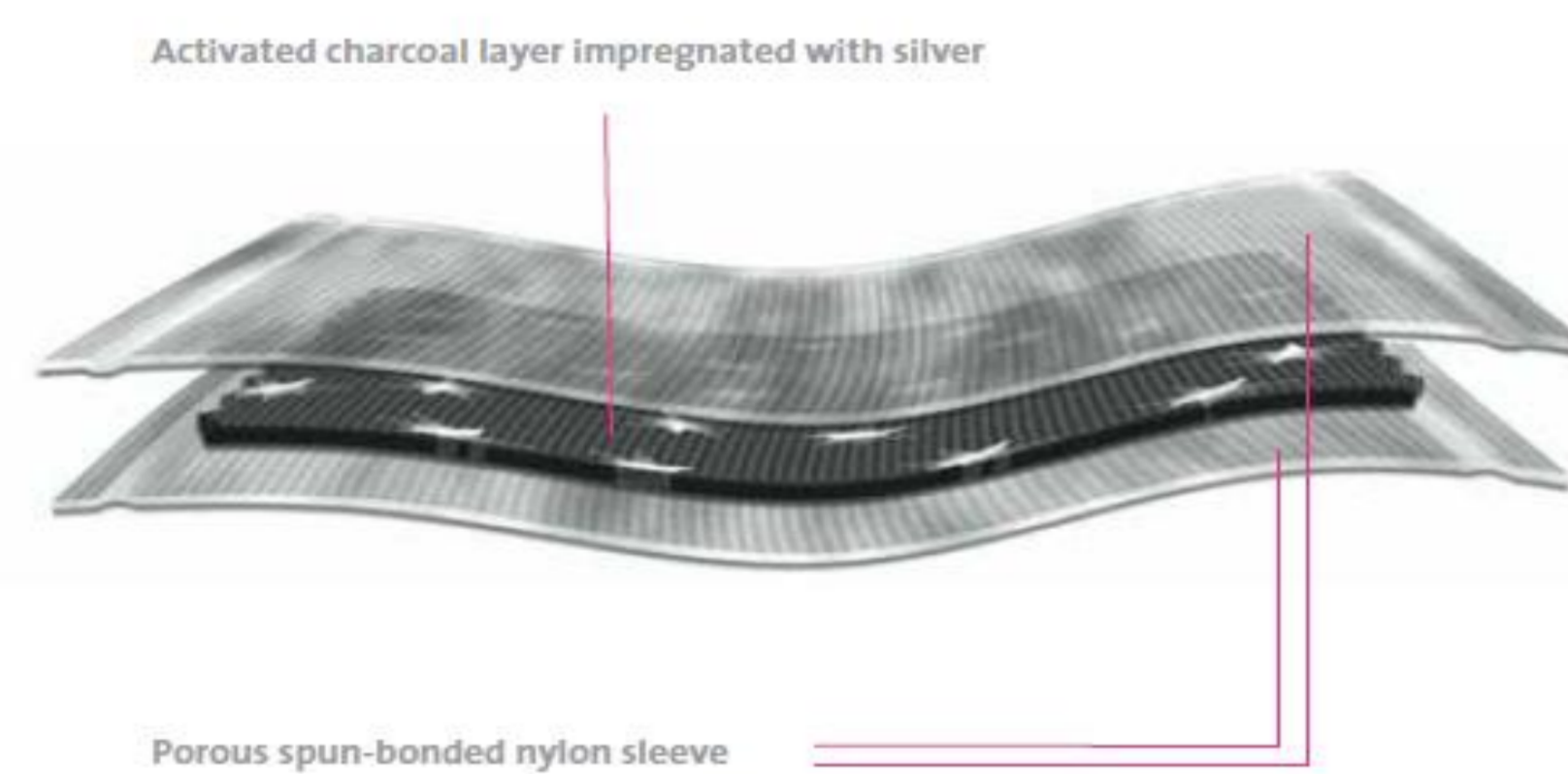
ABSTRACT

Activated charcoal cloth with silver (ACC + Silver) has been used for a number of years to help reduce bacterial load in wounds and to help prevent infection. Silver has been proven *in vitro* and clinically to be an effective treatment in the reduction of bacteria and to help prevent infection. Although a potent antimicrobial, there has been some concern associated with its potential to be cytotoxic to local cells in the wound site. Fibroblast migration and proliferation is an integral part of the wound healing process. Fibroblasts produce collagen; essential in forming the extracellular matrix¹ which forms the major constituent of repaired skin. Any wound dressing which has a detrimental effect on the fibroblast proliferation process may contribute to a delay in wound healing. Thus, having a dressing that provides the benefits of silver but not releasing silver into the wound would be advantageous.

By designing the dressing to provide antimicrobial protection while containing silver within the dressing we can prevent high concentrations of silver being released into the wound site. To demonstrate that this does not cause any detrimental effect to cell growth we examined the effect of ACC + Silver on dermal fibroblasts exposed to the dressing and the effect on cell proliferation was measured.

Furthermore, the silver in ACC + Silver has still been shown to be effective in the treatment of contaminated wounds even though it is not released into the wound. The elemental silver has been shown to be effective against microbes in lab studies and the benefits of the dressing against critically colonized wounds and non-healing wounds have been demonstrated clinically on over 12,000 patients.²

DISCUSSION



Dressings were tested that hold a layer of activated charcoal impregnated with silver, inside a spun bonded nylon sleeve.

Without releasing silver into the wound, this allows:

- 1 Binding of toxins³
- 2 Kill the bacteria⁴
- 3 Trap Odour⁵

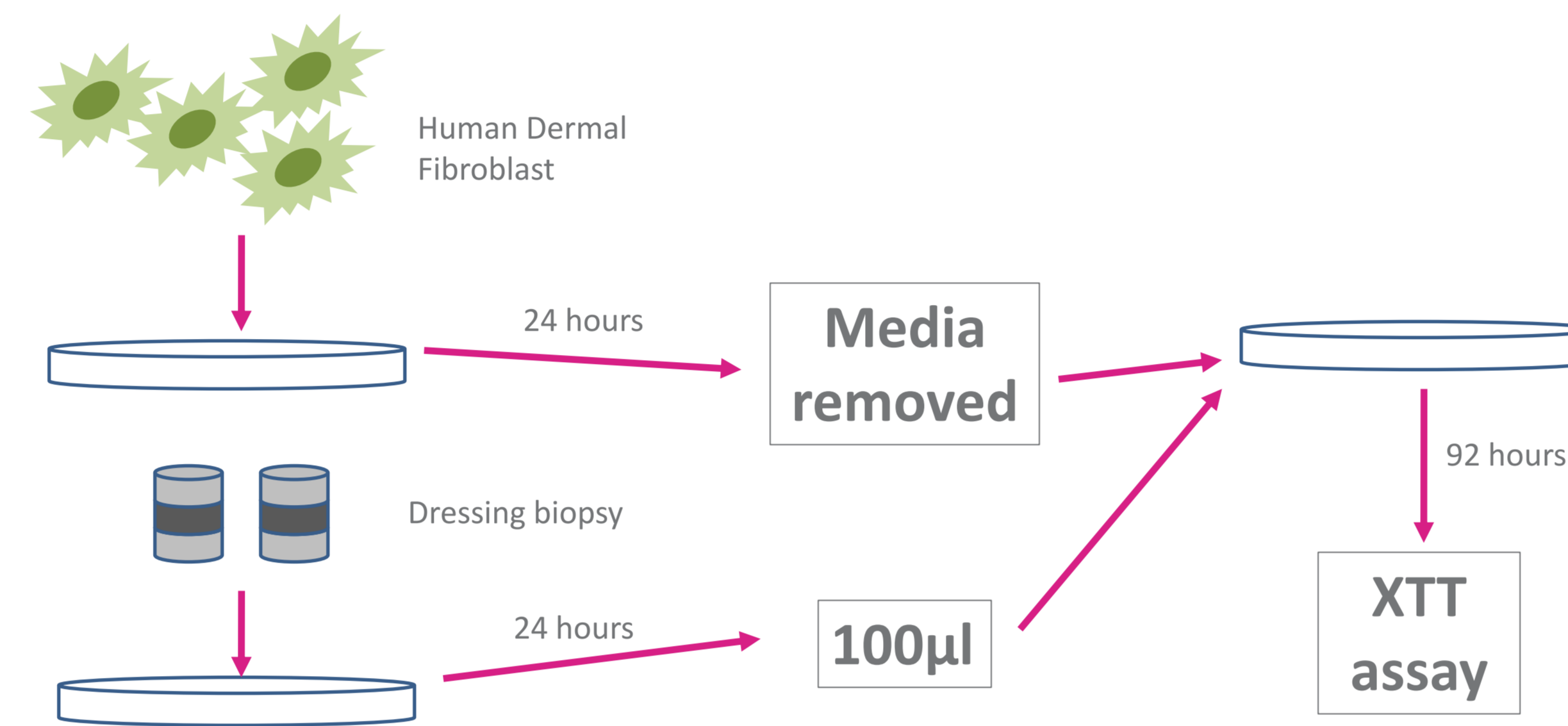
REFERENCES

1. Krishnamoorthy L, Morris HL, Harding KG. A dynamic regulator: the role of growth factors in tissue repair *Journal of Wound Care* 2001; 10: 99-1
2. Stadler et al. Survey of 12,444 patients with chronic wounds treated with Charcoal dressing (ACTISORB®). *Akt Dermatol* 2002; 28: 351-354
3. Russel AD et al. Antimicrobial activity and action of silver. *Progress in Medicinal Chemistry* 31, 351-370. Elsevier Service 1994
4. Price PE et al. *J Wound Care*, 9(2):93-95
5. Kramer A et al. Antibacterial activity and endotoxin binding capacity of ACTISORB® Silver 220 J *Hospital Infection*.2003;53:L511-514.

OBJECTIVE To establish the effect of the silver in Activated Charcoal cloth (ACC) on the proliferation of dermal fibroblasts, known to be important to wound healing.

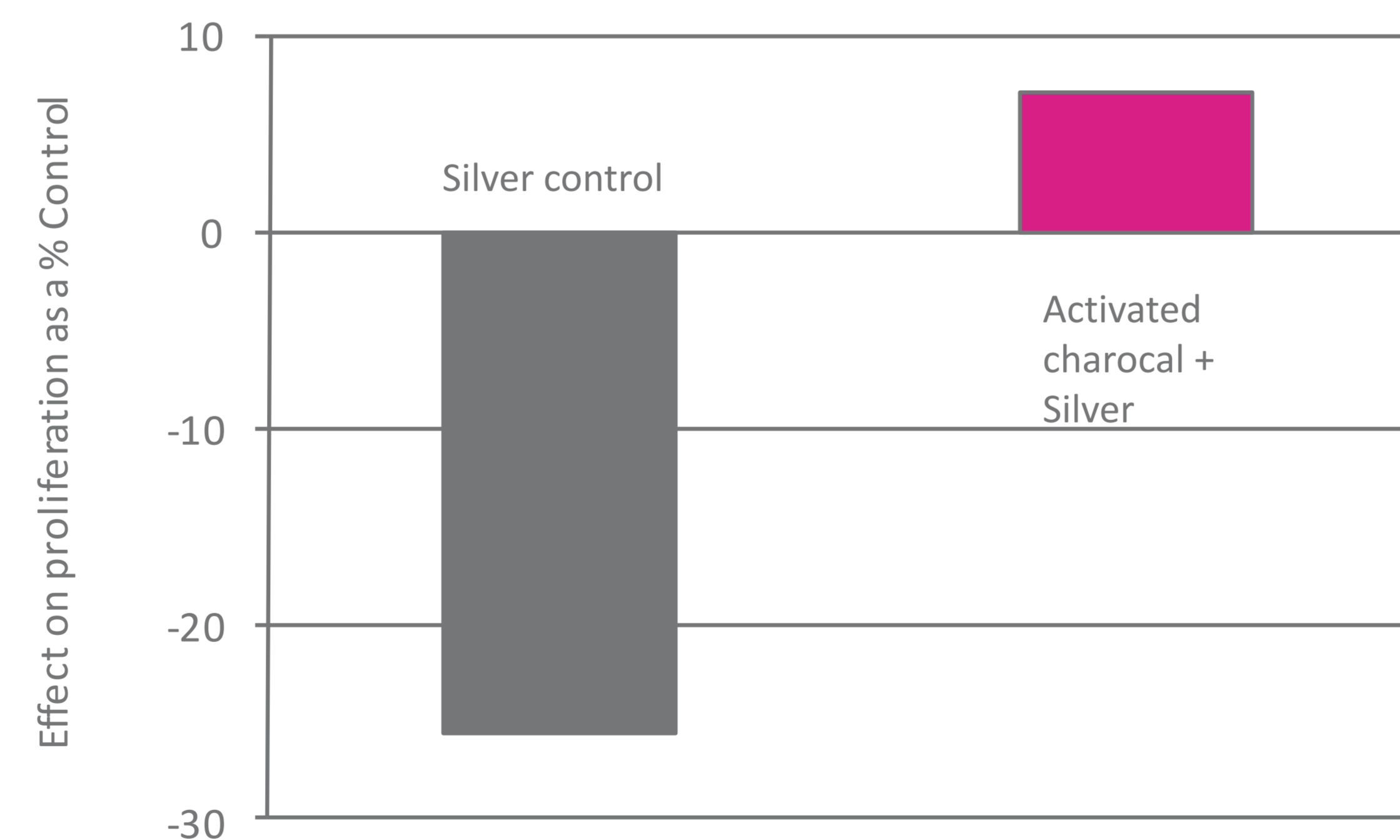
METHOD: Cell Proliferation using XTT assay

- Under sterile conditions, 6mm punch biopsies of dressings were taken
- These are added to serum free DMEM and incubated for 24 hours at 37°C and 5% CO₂ (prepared at various concentrations and then diluted to match to reduce the possibility that charcoal cloth had adsorbed nutrients from the media during incubation).
- 100µl of media from each of these samples was added to wells of human dermal fibroblasts at a cell seeded density of 2.5 x 10⁴ cells/ml previously incubated for 24 hours.
- Extract samples and controls of serum free DMEM and 10% FBS in DMEM were incubated for 92 hours.
- The proliferation of the cells was quantified using a XTT assay.



RESULTS: Cell Proliferation using XTT assay

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



•The control is DMEM solution. Negative values indicate decreased viable cells compared to the silver control (Acticoat).

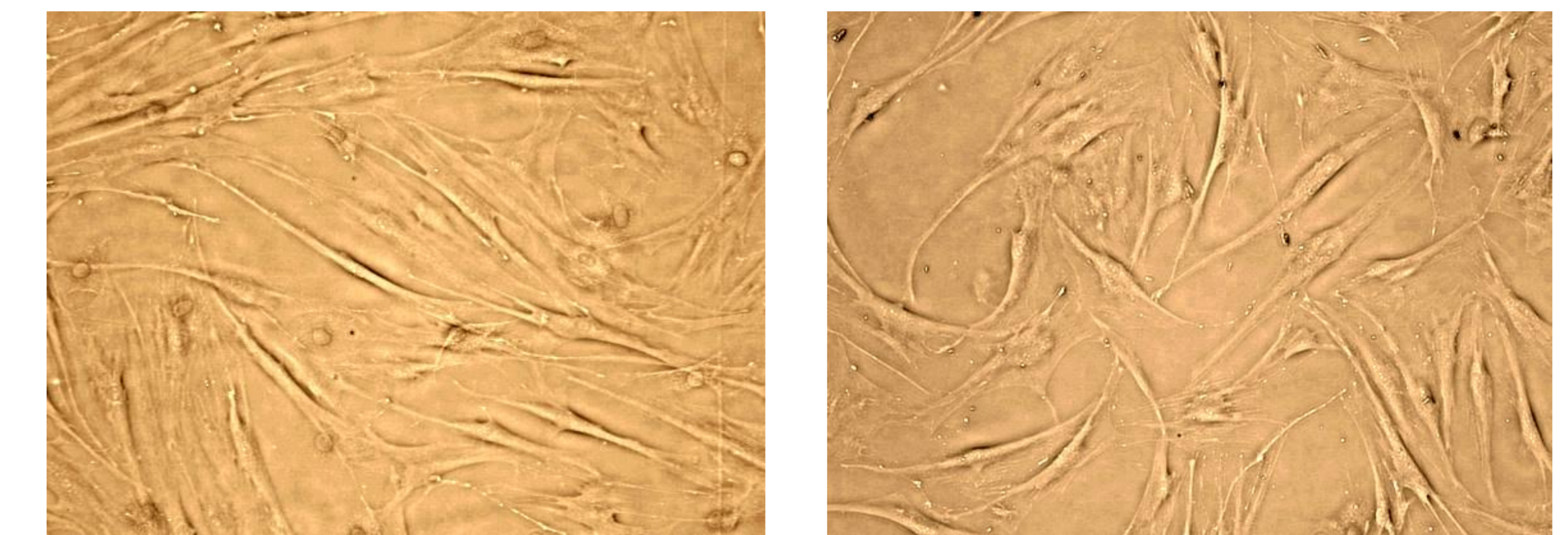
•The high negative value for the silver control extract indicated no viable cells after incubation

•Incubation with the silver control dressing extract causes cell death – no metabolically active fibroblasts. ACC + Silver has no detrimental effect on fibroblast proliferation

METHOD: Visual Examination of Cell Proliferation using Light Microscopy

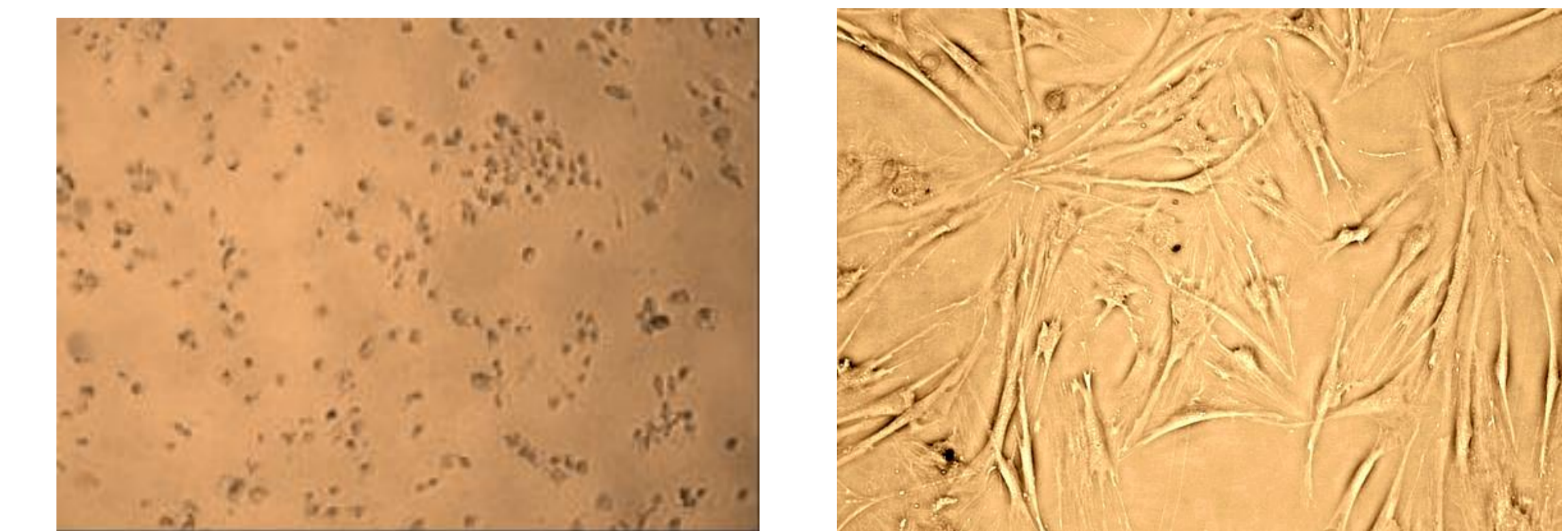
- Under sterile conditions, 6mm punch biopsies of dressings were taken
- These are added to serum free DMEM and incubated for 24 hours at 37°C and 5% CO₂.
- 400µl of media from each of these samples was added to wells of human dermal fibroblasts at a cell seeded density of 2.5 x 10⁴ cells/ml previously incubated for 24 hours.
- Extract samples and controls were incubated for 92 hours.
- Wells were washed and 200µl serum free media and 200µl 0.4% trypan blue solution added for 2 minutes.
- This solution was removed and wells examined under a Nikon Elipse TE 2000-U microscope.

RESULTS: Visual Examination of Cell Proliferation using Light Microscopy



Control

Charcoal Cloth Without Silver



Silver control (1mM)

Charcoal Cloth With Silver

Cells stained with 0.4% Trypan Blue solution showed no differences in cell morphology, no cell lysis, cell death or Trypan Blue absorbed by cells in either the control, the Charcoal Cloth without Silver or the Charcoal Cloth with Silver, supporting that activated charcoal + silver cloth is not detrimental to the proliferation of human dermal fibroblasts.

CONCLUSIONS

•The silver present in the Activated charcoal + silver wound dressing does not have any detrimental effect on cell proliferation *in-vitro* when compared to non silver ACC or the serum free negative control.

•Visual examination of the cells showed that neither ACC + Silver nor ACC was detrimental to the proliferation and morphology of fibroblasts.

•ACC + Silver provides the benefits of silver without causing any harm to host cells and does not prevent fibroblast proliferation which is essential for wound repair.