The interaction between Neurons and Skin cells *in vitro*

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Background: Pain is often the first signal of tissue damage. We examined whether neurons were able to enhance skin wound healing *in vitro*.

Method: Sensory neurons were isolated from mice Dorsal Root Ganglia (DRG), keratinocytes and fibroblasts were isolated from human skin. Several culture media were tested to determine the optimum growing conditions for skin and nerve co-culture. Cell viability was assessed using MTT assays or a numeric scoring system. Skin cells and neurons were co-cultured and the skin cells were wounded in a scratch assay. The closure of the "wound" was assessed at 0hr, 24hr and 48hrs.

Results: Neuron growth was examined in 8 different media for 7 days. Apart from their standard medium Neuralbasal A plus B27 (NBA) (score 2.8 ± 0.372) they survived best in Pereira medium (score 2.1 ± 0.418). Primary fibroblasts were assessed in 6 different media. After 6 days, there was 19.5% viability in NBA and 42.1% viability in Pereira medium compared to 100% in standard Fibroblast medium (DMEM + 10% serum). Primary keratinocytes were assessed in 6 different media. After 7 days, there was 0.2% viability in NBA and 80.2% in Pereira compared to 100% in standard keratinocyte media (Serum Free Media) but no growth in NBA. Fibroblast scratch assays performed in NBA showed no significant improvement in healing with the presence of neurons. Keratinocyte scratch assays are currently being performed in Pereira medium.

Conclusion: We have successfully isolated neurons and can grow them in culture with skin cells. Initial results with mouse neurons showed no improvement in human skin cell regrowth. Studies are continuing to characterize the isolated nerve cells as well as if the nerves require a damage stimulus in order to stimulate regrowth.

Key Words

Skin cells, fibroblasts, keratinocytes, Dorsal Root ganglia neurons, Scratch assay.

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