Irradiation at 830nm Stimulates Nitric Oxide Production and Inhibits Pro-Inflammatory Cytokines in Diabetic Wounded Fibroblast Cells

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Background and Objective: Wound healing in diabetic patients remains a chief problem in the clinical setting and there is a strong need for the development of new, safe, reliable therapies. This study aimed to establish the effect of irradiating diabetic wounded fibroblast cells (WS1) in vitro on pro-inflammatory cytokines and the production of nitric oxide (NO).

Materials and Methods: Normal, wounded and diabetic wounded WS1 cells were exposed to an 830nm laser with 5 J/cm2 and incubated for a pre-determined amount of time. Changes in cellular viability, proliferation and apoptosis were evaluated by the Trypan blue assay, VisionBlueTM fluorescence assay and caspase 3/7 activity respectively. Changes in cytokines (interleukin—IL-6, IL-1b and tumour necrosis factor-alpha, TNF-a) were determined by ELISA. NO was determined spectrophotometrically and reactive oxygen species (ROS) was evaluated by immunofluorescent staining.

Results: Diabetic wounded WS1 cells showed no significant change in viability, a significant increase in proliferation at 24 and 48 hours ($P<0.001$ and $P<0.01$ respectively) and a decrease in apoptosis 24 hours postirradiation ($P<0.01$). TNF-a levels were significantly decreased at both 1 and 24 hours ($P<0.05$), while IL-1b was only decreased at 24 hours ($P<0.05$). There was no significant change in IL-6. There was an increase in ROS and NO ($P<0.01$) 15 minutes post-irradiation.

Conclusion: Results show that irradiation of diabetic wounded fibroblast cells at 830nm with 5 J/cm2 has a positive effect on wound healing in vitro. There was a decrease in pro-inflammatory cytokines (IL-1b and TNF-a) and irradiation stimulated the release of ROS and NO due to what appears to be direct photochemical processes.

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