

BALANCING CELL COMPATIBILITY AND ANTIMICROBIAL EFFECTS OF 470 NM IN AN INFECTED HYPERGLYCAEMIC WOUND CELL MODEL

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Antimicrobial photobiomodulation (aPBM) with blue light (400-490 nm) is a promising non-invasive adjunct therapy for infected chronic diabetic foot ulcers (DFUs). Still, its fluence-dependent effects on human dermal fibroblasts remain poorly defined. This study investigated the fluence-dependent response of fibroblasts (BJ-5ta) cultured under three conditions: normal (N), normal wounded (NW), and hyperglycaemic wounded (HW), with or without bacterial infection. BJ-5ta fibroblasts (6×10^5 cells/mL) were co-cultured with *Staphylococcus aureus*, *Streptococcus pyogenes*, or *Pseudomonas aeruginosa* (1.5×10^3 CFU/mL) and irradiated with 470 nm blue laser light (power output 800 mW; power density 88 mW/cm²) at 5, 10, 30, 55, 100, or 120 J/cm². After 24 hours, fibroblast viability, migration, morphology, and bacterial survival were evaluated. Low fluences (5-10 J/cm²) maintained fibroblast viability at $\geq 90\%$ across all uninfected models. In contrast, higher fluences (30-120 J/cm²) caused a marked, dose-dependent decrease in fibroblast viability, with the lowest values observed at 120 J/cm² in both normal and hyperglycaemic wounded models. Infection with *S. aureus*, *S. pyogenes*, and *P. aeruginosa* increased cytotoxicity, with each species showing the greatest reduction in fibroblast viability at ≥ 55 J/cm². Fibroblast migration in the uninfected normal wounded model decreased progressively at fluences over 30 J/cm², dropping to 22-30% at 120 J/cm². In infected normal wounded models, migration declined in a species-dependent manner, with minimum values of 22-30% for *S. aureus*, 37-48% for *S. pyogenes*, and 21-27% for *P. aeruginosa* at 120 J/cm². CFU counts were significantly reduced at 5-10 J/cm² for all species. At 30-120 J/cm², *S. aureus* and *P. aeruginosa* were unaffected, whereas *S. pyogenes* exhibited a sustained, significant decrease in bacterial load. These results suggest a potential therapeutic window at 5-30 J/cm², within which fibroblast function is largely preserved while bacterial burden is reduced, supporting dose-optimised application of 470 nm aPBM in vitro.

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