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Antimicrobial phototherapy using blue laser light: A promising strategy to treat wound infections

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Background & Objectives: The development of novel antimicrobial strategies has become of crucial importance due to increasing resistance towards conventional antibiotics. The aim of the present study was to evaluate the antimicrobial effect of blue laser light on *Pseudomonas aeruginosa* (PAO) *in vitro* and *in vivo*.

Materials & Methods: A multiple wavelength diode laser device was employed. To evaluate the antimicrobial efficacy of blue laser (445 nm) and select the most effective protocols, multiple screening sessions have been performed employing several irradiation protocols in terms of power density and fluence on PAO grown in planktonic state and on agar plates. The protocols showing the best antimicrobial activities have been then applied on eukaryotic cell lines (human keratinocytes and fibroblasts) to evaluate possible toxicity. All the experiments have been conducted testing also a red (660 nm) and infrared (970 nm) laser wavelength. The effect of laser irradiation has been evaluated also on mature PAO biofilms grown in flow cells visualized in 3-dimensional reconstructions using laser scanning confocal microscopy. The best protocol in terms of antimicrobial efficacy and low toxicity was then employed *in vivo* on a mouse model of skin of PAO treated with blue and infrared laser (IR) with the same wound infection. The possible involvement of oxidative stress in the parameters. Only Blue laser was able to exert antimicrobial effects. mechanism of action has also been investigated. Furthermore, a blue LED light source was tested for possible differences.

Results: Among the different laser wavelengths tested with multiple protocols, only the blue ones showed a significant antimicrobial effect ($p<0,0001$). The irradiation protocol with power density 0,3W/cm2 and fluence 60J/cm2 was selected, confirming its efficacy and low toxicity *in vivo* ($p<0,01$). Oxidative stress seems to be involved in the blue light- mediated antimicrobial mechanism. Blue LED light showed a similar but slightly lower antimicrobial effect.

Conclusions: The use of antimicrobial phototherapy employing blue laser light has the potential to become a concrete treatment option for superinfected wounds.

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