Mitochondrial potential changes induced by treatment with a 915 nm GaAs diode laser

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Several reports have shown that low level laser therapy (LLLT) stimulates several metabolic activity at cellular level that could enhance wound healing. Laser irradiation seems to modulate various biological process in cell culture. LLLT could affect cell proliferation, differentiation and viability; besides some recent papers suggest that the mechanism of bio-stimulation may occur through mitochondrial photoactivation.

The aim of our study was to visualize by time lapse confocal microscopy the effects of 915 nm diode laser on mitochondrial transmembrane potential ($\Delta\Psi m$) of human dermal fibroblasts. We have compared two modes of irradiation (pulsed and continuous) at different power doses: 5, 15, 45 J/cm^2. Cells were loaded with tetramethylrhodamine methyl ester (TMRM), a semi-quantitative marker of $\Delta\Psi m$. After baseline signal acquisition, cells were irradiated and real time changes of fluorescence were recorded. At time 0 the cells showed the typical mitochondrial network and a negative nucleus. Irradiation with 5J/cm^2 in pulsed mode, caused a rough 10% signal increase after 60 sec followed by a fluorescence recovery to baseline after 250 sec. Stimulation in continuous mode with the same dose induced an opposite behaviour characterized by a 5% signal decrease 120 sec after irradiation with a return to baseline after 250 sec. At higher doses, 15J/cm^2 and 45J/cm^2, both irradiation modes produced a drop of TMRM fluorescence. At 15J/cm^2 the recover after the initial, 10% signal decrease was faster in pulsed (220 sec) rather than continuously irradiated cells (400 sec). 45J/cm^2 dose caused a 20% signal decrease in both modes but once again the pulsed irradiated cells recovered faster, 260 and 550 sec respectively.

Our results suggest that low intensity (5J/cm^2), pulsed 915nm laser have a stimulatory effect on mitochondrial metabolism whereas higher doses (45 J/cm^2) irradiation, especially if repeated, could exert a toxic effect on cell, explicated by mitochondrial depolarization.

Key words
Laser, fibroblast, mitochondria, confocal microscopy