**The effect of photobiomodulation associated with disodium dexamethasone phosphate on the expression of inflammatory cytokines and nitric oxide in activated M1 profile macrophages.**

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Macrophages polarized to the M1 profile play a crucial role in the inflammatory process, being essential in the body's immune response to pathogens, tissue injuries, and other forms of cellular stress, modulating, through their products, the different phases of cellular repair. Many anti-inflammatory drugs have been used for the treatment of inflammatory processes, with one of the most common in clinical practice being the disodium dexamethasone phosphate (FD-Dexa). Despite being very effective, FD-Dexa can cause numerous side effects. The risk of adverse effects, some of which may be potentially severe, increases with higher doses and longer treatment durations. In the literature, photobiomodulation (PBM) has demonstrated its ability to attenuate the inflammatory process through various mechanisms, including the modulation of macrophage activity and secretory products, inducing polarization to an anti-inflammatory phenotype (M2), which is associated with inflammation control and tissue repair promotion. Thus, both isolated PBM and its combination with corticosteroids show potential effects on the modulation of inflammatory cytokine expression by M1 macrophages. The objective of this study is to evaluate the effects of PBM, with or without the addition of disodium dexamethasone phosphate (FD-Dexa), on the secretion of inflammatory cytokines and nitric oxide synthesis in M1-polarized macrophages. For this study, J774 macrophages will be used and evaluated in the following groups: (1) Control Group - J774 cells without any treatment; (2) M1 Group - J774 activated to the M1 phenotype; (3) M1 + PBM Group J774 activated to the M1 phenotype and submitted to PBM; (4) M1 + FD-Dexa 2µM Group including the Dexa treatment; (5) M1 + FD-Dexa 4µM Group; (6) M1 + PBM + FD-Dexa 2µM Group and (7) M1 + PBM + FD-Dexa 4µM Group. Duplicate analyses will be conducted for all experimental groups. The J774 macrophages will be activated to the M1 phenotype using Dulbecco's Modified Eagle Medium (DMEM) supplemented with 10% fetal bovine serum (FBS) and 1% antibiotic-antimycotic solution, LPS 1µg/mL and 0.2µg/mL interferon-gamma (IFN-Ɣ). They will be incubated for 2 hours at 37°C in a humidified atmosphere with 5% CO2. The PBM groups will be irradiated using an Aluminum Gallium Arsenide laser (780 nm, 70 mW, 17.5J/cm2, 1J) and FD-Dexa 2µM and 4µM will be added to the corresponding groups, and then the cells of all experimental groups will be incubated at 37°C, 5% CO2 for 24 and 48 hours. At the end of the incubation period, culture medium will be collected, total protein quantified using a Nano spectrophotometer (Nanodrop), and then analysis of IL-6, IL-1β, and TNF-α synthesis will be performed using ELISA technique and the nitric oxide (NO) synthesis using the Griess method. Experiments will be conducted in triplicate, and data will be subjected to statistical analysis.

Key words: J774 macrophages, dexamethasone, inflammation, photobiomodulation, nitric oxide, cytokines.

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