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Optimizing Photobiomodulation Parameters for Tenogenic Differentiation

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Tendons are frequently damaged by acute injuries, such as sports injuries or chronic overuse and age-related degeneration. Native tendon healing is lengthy and ineffective due to the tissue's inherently low cellularity, limited vascularization, and low metabolic activity of resident tenocytes. Natural healing is often accompanied by fibrosis, adhesion formation and re-injury is common. Current treatment options focus on symptom management and gradually strengthening the tissue over time. Mesenchymal stem cell (MSC) therapy offers a promising alternative due to the ability of MSCs to proliferate, differentiate into tenocytes and produce ECM (extracellular matrix) components to facilitate tendon repair. Photobiomodulation (PBM), uses specific light wavelengths to stimulate intracellular chromophores and activate various cellular functions. PBM has demonstrated potential in enhancing stem cell viability and proliferation, as well as tenogenic differentiation and ECM production. Despite this, there is a lack of standardized PBM parameters (wavelength and fluency) for tenogenic differentiation, hindering reproducibility, cross-study comparisons and translation into clinical trials. This study aimed to evaluate the potential of PBM to enhance tenogenic differentiation and to determine the optimal PBM parameters. Adipose-derived mesenchymal stem cells (ADMSCs) were irradiated using 525 nm, 825 nm, and a combination of both wavelengths at fluences of 5 and 10 J/cm^2. Prior to PBM treatment, the stem cell nature of the ADMSCs was confirmed via immunofluorescent detection of CD44, CD90, and CD166. Post-differentiation assessments included morphological analysis (May-Grünwald-Giemsa staining), cytotoxicity, and proliferation assays. Tenogenic differentiation was evaluated via gene expression (Scleraxis, Tenomodulin, Collagen I, Tenascin-C, and Biglycan) and immunofluorescence staining (Scleraxis, Tenomodulin, and Collagen). The results confirmed the stemness of the ADMSCs and showed that tenogenic differentiation, particularly when combined with PBM, enhanced cell viability, proliferation, and expression of tenogenic markers, without significant morphological changes. These findings highlight PBM as a promising adjunct to improve tenogenic outcomes.

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