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Phototransfection of mouse embryonic stem cells with plasmid DNA using femtosecond laser pulses.

Lebogang Thobakgale Sello Lebohang Manoto Saturnin Ombinda Lemboumba Malik Maaza Patience Mthunzi-Kufa

ABSTRACT

Cellular manipulation by delivery of molecules into cells has been applied extensively in tissue engineering research for medical applications. The different molecular delivery techniques used range from viral and chemical agents to physical and electrical methods. Although successful in most studies, these techniques have inherent difficulties such as toxicity, unwanted genetic mutations and low reproducibility respectively. Literature recognizes pulsed lasers at femtosecond level to be most efficient in photonic interactions with biological material. As of late, laser pulses have been used for drug and DNA delivery into cells via transient optical perforation of the cellular membrane. Thus in this study, we design and construct an optical system coupled to a femtosecond laser for the purpose of phototransfection or insertion of plasmid DNA (pDNA) into cells using lasers. We used fluorescent green protein (pGFP) to transfect mouse embryonic stem cells as our model. Secondly, we applied fluorescence imaging to view the extent of DNA delivery using this method. We also assessed the biocompatibility of our system by performing molecular assays of the cells post irradiation using adenosine triphosphate (ATP) and lactate dehydrogenase (LDH).