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Abstract

3D Cell Culture Models for the Study of New Anti-Inflammatory Drugs: Toxicity and Cell Viability †

Juliano C. T. T. Almeida [1,*], Luan B. V. Costa [1], Vinícius Q. F. Silva [1], Maria da G. da Silva [1], Andreanne G. Vasconcelos [1,2] and José Roberto de S. A. Leite [1,2]

[1] Applied Immunology and Morphology Research Centre (NuPMIA), Morphology Area, Faculty of Medicine, University of Brasília (UnB), Brasília 70910-900, Brazil; luanbvazcosta@gmail.com (L.B.V.C.); viniciusquirino32@hotmail.com.br (V.Q.F.S.); gloriosa@unb.com (M.d.G.d.S.); andreannegv@gmail.com (A.G.V.); jrsaleite@gmail.com (J.R.d.S.A.L.)
[2] People&Science Pesquisa Desenvolvimento e Inovação Ltda, Centro de Apoio ao Desenvolvimento Tecnológico (CDT), University of Brasília (UnB), Brasília 70910-000, Brazil
* Correspondence: julianotupina@gmail.com
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Introduction: In Brazil, pressure ulcer wounds have a significant impact on public health. However, many studies on new drugs for this issue rely on 2D cell culture models, which do not accurately reproduce the in vivo environment. 3D cell models provide greater similarity to living tissues, allowing a better assessment of cell viability. This study proposes to validate this culture model through a cytotoxicity analysis, including the experimental drug Notlex. **Methodology**: The study employed several protocols, using UV radiation and 70% alcohol for cabinet sterilization. Keratinocytes maintenance was performed twice a week, mainly with trypsin and DMEM. Spheroids were obtained by combining agarose film with the cells in U-bottom wells under plate agitation. For cytotoxicity assessment, cells were exposed to different concentrations of the drug, along with cell and death controls. Readings were taken by using Luciferase and a luminometer. **Results:** An additional experiment evaluated *Escherichia coli* sensitivity. Findings showed that the MIC of the drug was 800 times lower than that applied in humans. Consequently, cytotoxicity tests were conducted with higher concentrations, and it was observed that, from 25× to 3200× lower concentrations, the drug demonstrated viability similar to the positive control, indicating low toxicity. **Conclusions:** The project validated a 3D keratinocyte spheroid cell culture model for evaluating new drugs, particularly for pressure ulcers. The results provided a better understanding of cell behavior in both 2D and 3D models, in addition to incorporating an assay to determine the MIC/MBC of the drug against a common wound-related bacterium.

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