

**Essential Oils for anti-cellulite applications: *In-vitro* testing of the viability, cytotoxicity, and cell permeability**

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Essential oils (EO) have been used for a very long time, particularly in the food, beverage and perfume industries. However, their use to manage cellulite, a skin condition frequently seen as an unattractive cosmetic issue, is still poorly studied. The main goal of this work was to evaluate the viability, cytotoxicity and permeability of immortalized normal human dermal fibroblasts (NHDF) when incubated for several hours with different concentrations of an EO blend composed of volatile compounds with anti-cellulite properties. An MTT colorimetric assay was used to access cell viability and cytotoxicity. The conditions chosen were: Incubations with 0.1%, 0.05%, 0.0250% and 0.0125% EO blend in culture medium for 12 and 24H. Regarding cell permeability, 2 tests were performed: TEER and Lucifer Yellow. Aliquots of culture medium were drawn at 2h, 8h, 12h and 24h as a way of perceiving the amount of essential oil that crosses the membrane during the incubation period. These aliquots were extracted and analysed for each volatile compound present in the formulation, comparing the concentrations present with a negative control. Results from the MTT assay showed that: All essential oil concentrations studied were safe for exposures equal to or less than 12H; For the 24H of incubation, the 0.1% concentration negatively affects cell viability, not being indicated for prolonged exposure; The remaining concentrations also showed a decrease in cell viability, but it does not appear as a significant value and they can still be considered safe for a prolonged exposure of 24 hours. The permeability results produced by the Teer and Lucifer Yellow assays also showed that there was no significant impairment in the fibroblast monolayer with a 24H incubation, while using a concentration of 0.0125% EO in culture medium. Trace amounts of some compounds were found in the recovered aliquots, being β -mircene and menthol the only ones present at all hours.

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