〈原 著〉

# 培養細胞系を用いた化粧品油性原料の安全性評価

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# Safety Evaluation of Oil Ingredients for Cosmetic Using Cell Culture System

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#### Abstract

Concerning 42 oil ingredients for cosmetic on the market (13 hydrocarbons and 29 ester oils), in vitro cytotoxicity tests for safety evaluation were carried out with cultured epithelial cells derived from human skin (JTC-17).

The cells were exposed to the test substances which were dispersed in the cultured medium using ultrasonic emulsion method, and the cell viability was estimated by Neutral Red dye uptake test. Comparison was made between the results of in vitro test and primary irritancy tests on rabbit skin. Of 42 oil ingredients by in vitro, 13 ingredients were found to be toxic to the cultured cells, namely, a low viscous liquid paraffin, a pristane, 6 mono-ester oils, 3 di-ester oils, and 2 tri-ester oils. They were also primary irritants to rabbit skin.

Furthermore, from the results of in vitro tests of 29 oil ingredients used as reagents (6 hydrocarbons and 23 ester oils), it was found that most of the oil ingredients which were cytotoxic in vitro were low in molecular weight and had unsaturated bond in molecular formula. These findings were consistent with the results of in vivo studies of primary irritancy test on rabbit skin.

Though not all of the primary skin irritants show toxicity to the cultured cells (for example, isopropyl palmitate), the in vitro cytotoxicity test here seems to be simple and useful as a first screening test for primary skin irritants in oil ingredients for cosmetic and also will serve as an animals experiment, resulting in reduction in numbers of laboratory animals.

Key words: Cytotoxicity test, Primary skin irritancy test, Oil ingredients for cosmetic,
Alternative method

## 要 旨

市販の化粧品油性原料42品目(炭化水素13品目,エステル油29品目)について、ヒト皮膚由来の上皮性培養細胞(JTC-17)に対するin vitro 細胞毒性試験を実施した。細胞毒性の評価には、Neutral Red の Dye uptake test を用いた。検体は、予め培地中に超音波を用いて乳化させ、これを細胞に接触させた。そして培養細胞への毒性とウサギ皮膚一次刺激性との相関性について検討した。その結果、13品目(極低粘度流動パラフィン1品目、プリスタン、モノエステル6品目、ジエステル3品目、そしてトリエステル6品目、ジエステル3品目、そしてトリエステル2品目)に細胞毒性反応が認められた。これらの検体は、またウサギ皮膚一次刺激性試験においても明かに刺激性を有していた。

そこで更に、純度の高い試薬グレードの検体29品目 (炭化水素 6 品目、エステル油23品目)を用いて in vitro 試験を行った結果、細胞毒性を示す検体の多くは 分子量が低いこと、また分子内に不飽和二重結合を有 していることが示された。これらの知見は、併せて実 施したウサギ皮膚一次刺激性試験の結果と一致した。

ウサギ皮膚一次刺激性の高い検体が全て細胞毒性を示すとは限らないが(例、isopropyl palmitate)、今回の試験法は、化粧品油性原料中の皮膚一次刺激性物質の first screening test として簡便且つ有用であり、一つの代替法として、毒性試験に用いる動物数の削減にも役立つと考えた。

## 1. 緒 言

培養細胞を用いた化学物質の in vitro 細胞毒性試験