

Phototoxicity

Background Information



'The *in vitro* 3T3 NRU phototoxicity test is based on a comparison of the cytotoxicity of a chemical when tested in the presence and in the absence of exposure to a non-cytotoxic dose of simulated solar light.'

¹OECD Guideline for the Testing of Chemicals 432: *In Vitro* 3T3 NRU Phototoxicity Test; April 2004

- Phototoxicity is defined as a 'toxic response from a substance applied to the body which is either elicited or increased (apparent at lower dose levels) after subsequent exposure to light, or that is induced by skin irradiation after systemic administration of a substance'¹.
- The assay is appropriate for pharmaceuticals, chemicals or cosmetics/personal care products which absorb in the wavelength range of 290-700nm, and are applied to, or can reach, the skin or eyes.
- Cyprotex's phototoxicity service is a non-GLP screening assay performed using Balb/c 3T3 cells. The cells are incubated with increasing concentrations of the test article in the presence and absence of a non-toxic dose of UVA irradiation. Cytotoxicity is assessed via Neutral Red uptake.

Protocol

Model Available

Balb/c 3T3 mouse fibroblast cell line

Number of Replicates/Concentrations

6 replicates at 8 concentrations of test article

Exposure Conditions

Condition 1: 3T3 cells kept in dark chamber
Condition 2: 3T3 cells exposed to calibrated non-toxic dose of UVA

Assay Controls

Chlorpromazine (Positive control)
Vehicle
Background

Endpoints

Neutral Red uptake (3T3 cells)

Data Delivery

Photo irritation factor (PIF)
Mean photo effect (MPE)
Phototoxic category

‘A positive result in the 3T3 NRU-PT should not be regarded as indicative of a likely clinical phototoxic risk, but rather a flag for follow-up assessment.’²

Figure 1

Graph illustrating neutral red uptake data for the positive control compound, chlorpromazine, in the presence and absence of UVA irradiation using 3T3 cells.

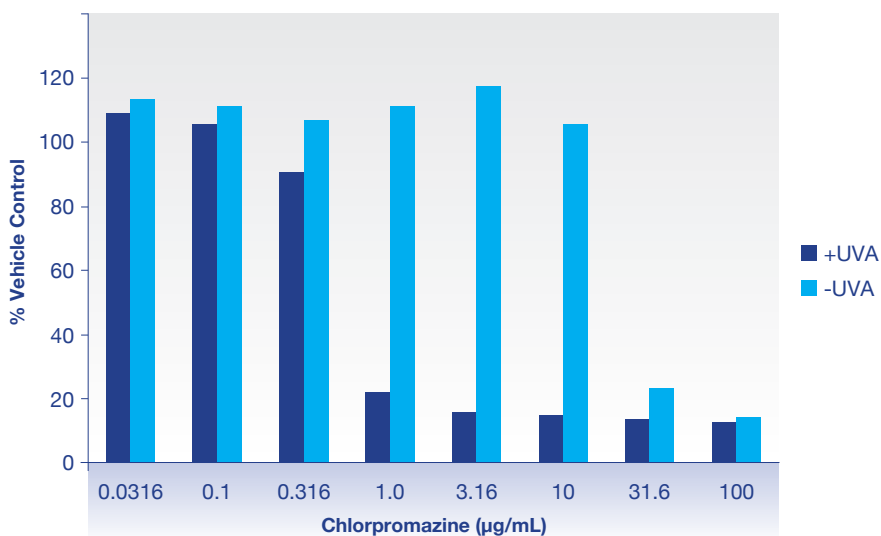


Table 1

Interpretation of phototoxic potential using the *in vitro* 3T3 Neutral Red Uptake test.

Phototoxic Potential Categorisation	Data from <i>in vitro</i> 3T3 Neutral Red Uptake test
No phototoxicity	PIF < 2 or MPE < 0.1
Probable phototoxicity	PIF between 2 and 5 or MPE between 0.1 and 0.15
Phototoxicity	PIF > 5 or MPE > 0.15

PIF = photo-irritation factor
MPE = mean photo effect

References

¹ OECD Guideline for the Testing of Chemicals 432: *In Vitro* 3T3 NRU Phototoxicity Test; April 2004
² ICH Harmonised Tripartite Guideline: Photosafety Evaluation of Pharmaceuticals S10; Nov 2013