TEST CODE:
CT-036

Skin Corrosion Test

RECONSTRUCTED HUMAN EPIDERMIS TEST METHOD (OECD TEST GUIDELINE 431)

OVERVIEW

Skin corrosion is defined as the production of irreversible tissue damage, manifested as visible necrosis of the skin, according to the definition of the Globally Harmonized System for Classification and Labelling of Chemicals.

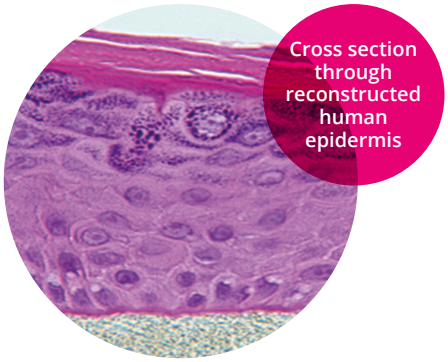
The test described here is fully accepted at a regulatory level for the hazard identification of corrosive and non-corrosive chemicals in accordance with the UN Globally Harmonized System (GHS) of Classification and Labelling. It can be used for single substances and for mixtures including finished products. It is appropriate for compliance with a range of legislation including REACH (Registration, Evaluation, Authorisation and restriction of CHemicals) and the CLP Regulation 1272/2008 (Classification, Packaging and Labelling of substances and mixtures).

The test method is based on reconstructed human epidermis (RhE), which in its overall design mimics the biochemical and physiological properties of the upper parts of the human skin. The procedure is based on the principle that corrosive chemicals are able to penetrate the skin barrier *(stratum corneum)* by diffusion or erosion, and are cytotoxic to the underlying cell layers. The test item is applied directly to the skin surface, providing a good model of "real life" exposure. Cell viability is measured by enzymatic conversion of the vital dye MTT into a blue formazan salt that is quantitatively measured after extraction from the skin tissues. Corrosive test items are identified by their ability to decrease cell viability below defined threshold levels, resulting in classification as Corrosive (UN GHS Category 1) or Non-Corrosive ("no-label").

Sub-categorisation of corrosive test items into Category 1A or 1B/C is possible.

TEST SYSTEM: RECONSTRUCTED HUMAN EPIDERMIS

Reconstructed human epidermis is a skin model composed of living human keratinocytes which have been cultured to form a multi-layered, highly differentiated epidermis. The levels of differentiation obtained are at the cutting edge of *in vitro* skin technology. The model consists of highly organized basal cells which progressively flatten out as the apical surface of the tissue is approached, analogous to the normal human *in vivo* epidermis. The model includes a functional skin barrier with an *in vivo*-like lipid profile. The profiles of key differentiation markers also mirror those seen *in vivo*. The cells are both metabolically and mitotically active, and release pro-inflammatory agents (cytokines). Reconstructed human epidermis is grown on special platforms at the air-liquid interface, allowing for direct application of test items in a way that accurately models "real life" skin exposure.



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SUMMARY OF THE TEST METHOD

- Skin models are pre-warmed in a cell culture incubator (37°C / 5% CO_2) for 60 minutes.
- The test item is applied to the surface of the skin models: triplicate models are dosed at the apical surface with 50µl (liquid) or 25mg (solid).
- Controls consist of ultrapure water (negative control) and potassium hydroxide (positive control).
- The dosed skin models are placed into a cell culture incubator for 3 minutes and 60 minutes (triplicate models for each time point).
- Test items and control substances are removed from the skin models' surface by washing.
- The viability of the skin models is assessed by MTT conversion. MTT solution is applied to the surface of the skin models and placed into a cell culture incubator for 3 hours. The blue formazan metabolite produced by viable cells is then extracted into isopropanol by incubation at room temperature for 2 hours or overnight.
- Triplicate samples of the extracted formazan solution are transferred to a microplate and the formazan product is quantified by absorbance spectrophotometry (wavelength 570nm).
- Absorbance readings of the formazan product from skin models incubated with test items are compared with those of negative controls to calculate percentage viability.
- A range of acceptance criteria must be satisfied in accordance with OECD TG431.
- If the viability is less than 50% after 3 minutes, the test item is classified as corrosive. If viability is 50% or greater after 3 minutes but less than 15% after 60 minutes, the test item is classified as corrosive. If viability is greater than 50% after 3 minutes and equal to or greater than 15% at 60 minutes, the test item is classified as non-corrosive.

TURNAROUND TIME	*6 – 8 weeks*
AMOUNT OF SAMPLE REQUIRED	*10ml (liquids) / 10g (solids). Please enquire if sample availability is limited.*
PRICE	*Our test prices are dependent on the quantity of test items. Please enquire for a quote using the contact information shown below, or the contact form on our website.*
FURTHER DOWNLOADS	OECD Test Guideline 431 XCellR8 Good Laboratory Practice (GLP) Compliance Certificate.

QUALITY STATEMENT

XCellR8 is accredited by the UK Medicines and Healthcare Products Regulatory Authority (MHRA) for the conduct of *in vitro* safety testing in compliance with Good Laboratory Practice (GLP). This means that we are able to provide our clients with test results that may be used at a regulatory level to demonstrate product safety, where the test is an approved regulatory method. The regulatory status is applicable in all global territories participating in the OECD's Mutual Acceptance of Data.

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