

Skin corrosion protocol SkinEthic (validated)

Model: SkinEthic™ RHE

DESCRIPTION

Culture insert equilibration period: incubate for 3 h up to 24 h (37°C)

 \downarrow

Transfer tissues to fresh maintenance medium



Treatment: At least 2 tissues per exposure time: \circ Liquids and viscous: 40 μ L \pm 3 μ L (80 μ L/cm²) using nylon mesh

 \circ Solids: 20 μ L ± 2 μ L H₂O + 20 ± 3 mg (40 mg/cm²)

○ Waxy/sticky: 20 ± 3 mg (40 mg/cm²) using nylon mesh



Treatment Period: Incubate for 3 min and 60 min at RT



Rinse 20 times with a constant soft stream of 1x PBS-



Incubate tissues in 1 mg/mL MTT solution for 3 h \pm 15 min (37° C)



Extraction: Immerse the inserts in 1.5 mL isopropanol (extraction from top and bottom of insert)



Extract formazan overnight without shaking at RT or for 120 min with shaking (~120 rpm) at RT



Perforate the insert and homogenize formazan extract



Read OD with microplate spectrophotometer at 570 nm

PREDICTION MODEL

<i>In vitro</i> Result	Classification (<i>In vivo</i> Prediction)
Mean tissue viability < 50% after 3 min exposure	Corrosive: Optional Sub-category 1A
Mean tissue viability ≥ 50% after 3 min exposure AND < 15% after 60 min exposure	Corrosive: Optional combination of Sub-categories 1B-and-1C
Mean tissue viability ≥ 50% after 3 min exposure AND ≥ 15% after 60 min exposure	Non-corrosive