

***in vitro* PERCUTANEOUS ABSORPTION ASSAY**

**Theory:** The rates of absorption and passage of test chemicals into and through human skin can be modeled in the *in vitro* percutaneous absorption assay. By utilizing various skin models and test protocols, a variety of percutaneous absorption endpoints may be determined, such as the absorption and passage through the lipophilic *stratum corneum*, absorption and passage through the epidermis and dermis, and the potential for metabolism within the viable epidermis.

### Applications and Use

Skin penetration assays may be utilized to evaluate the rate of absorption or passage of individual chemicals or formulation components in personal and skin care products and pharmaceuticals to enhance product development goals, or to address product safety issues. Skin penetration assays may also be used to determine the extent of absorption of potentially toxic materials, for example, to address EPA and worker safety issues. Applications may include:

- ◆ evaluation of absorption and retention of skin care products, cosmetics, pharmaceuticals, etc.
- ◆ evaluation of passage of skin care products, cosmetics, pharmaceuticals, etc.
- ◆ evaluation of dermal delivery systems, skin penetration enhancers, formulations, lotions and vehicles
- ◆ evaluation of potential systemic absorption of toxic chemicals
- ◆ evaluation of metabolism in viable skin models
- ◆ Active ingredients and formulations may be applied to the test system at target formulation concentrations.

### Experimental Features

- ◆ A variety of skin models may be used (human cadaver-source skin, viable engineered human tissue, abattoir-derived pig skin)
- ◆ Finite or Infinite dose assays
- ◆ Bronaugh-type Flow Through chambers used
  - eliminate receptor fluid "loading" of test chemicals
  - allow collection of fractions overnight
  - skin temperatures maintained at 32°C
- ◆ Testing of radio-labeled chemicals (<sup>14</sup>C-) for accurate determinations of less than 1% of applied dose
- ◆ Barrier Integrity Qualification/Evaluation of each tissue tested using <sup>3</sup>H<sub>2</sub>O passage test
  - application of infinite dose of <sup>3</sup>H<sub>2</sub>O for 20 minutes
  - evaluation of <sup>3</sup>H<sub>2</sub>O passage over an 80-minute period
- ◆ Receptor Fluids
  - Water-soluble test materials: Hank's Balanced Salt Solution (HBSS) w/o phenol red, w/o sodium bicarbonate, with 25 mM HEPES, with 7 g/L NaCl, with 100 µg/mL gentamicin
  - Lipophilic test materials: HBSS as above with 4% bovine serum albumin
  - Custom selection of receptor fluids available
- ◆ Percutaneous Absorption is calculated as a function of **% of Applied Dose**
  - % of applied dose determined in each receptor fluid fraction / unit of permeation time
  - Independent Total Activity Controls for total applied dose
- ◆ Tape stripping of *stratum corneum* to evaluate absorption stratification in *stratum corneum*.
- ◆ Separation of epidermis and dermis to evaluate absorption stratification in full-thickness skin.
- ◆ Mass Balance Determination / Relative Recovery - >90% recovery of <sup>14</sup>C-caffeine demonstrated
  - Test material recovery determined from:
    - all receptor fluid fractions
    - un-absorbed test material (recovered by wash/rinse of tissue surfaces)
    - residual receptor fluid in receptor chamber and line
    - tape strips, epidermis and dermis
    - residues collected after tape stripping or tissue separation
    - residual test article on dosing devices (if required for creams or viscous materials)