
EpiDerm Technical Specifications

Tissue

Kit: A standard EpiDerm kit (EPI-200) consists of 24 tissues. (Tissue “kits” contain tissues, a small amount of culture medium, and plasticware; contact MatTek for specific kit contents)

Formats: 9mm & 22mm individual inserts and 96-well HTS plates – tissue culture substrate is chemically modified with a pore size of 0.4 μm

Culture: Air-liquid interface

Histology: 8-12 cell layers plus stratum corneum (basal, spinous, and granular layers)

Lot numbers: Tissue lots produced each week are assigned a specific lot number. A letter of the alphabet is appended to the end of the lot number to differentiate between individual kits within a given lot of tissues. All tissue kits within a lot are identical in regard to cells, medium, handling, culture conditions, etc.

Shipment: At 4°C on medium-supplemented, agarose gels

Shipment day: Every Monday. Shipment on Thursday also possible upon special request

Delivery: Tuesday morning via FedEx priority service (US). Outside US: Tuesday-Thursday depending on location

Shelf life: Including time in transit, tissues may be stored at 4°C for up to 4 days prior to use. However, extended storage periods are not recommended unless necessary. In addition, the best reproducibility will be obtained if tissues are used consistently on the same day, e.g. Tuesday afternoon or following overnight storage at 4°C (Wednesday morning)

Length of experiments: Cultures can be continued for at least 2 weeks with good retention of normal epidermal morphology. Cultures must be fed every other day with 5.0 mL of New Maintenance Medium (EPI-100-NMM-3). Cell culture inserts are placed hang top plates (HNG-TOP-12, provided) to allow the use of 5.0 mL.

Cells

Type: Normal human epidermal keratinocytes (NHEK)

Genetic make-up: Single donor

Derived from: Neonatal-foreskin tissue (NHEK)

Alternatives: NHEK from adult breast skin

Screened for: HIV, Hepatitis-B, Hepatitis-C, mycoplasma

Medium

Base medium: Dulbecco's Modified Eagle's Medium (DMEM)

Growth factors/hormones: Epidermal growth factor, insulin, hydrocortisone and other proprietary stimulators of epidermal differentiation

Serum: None

Antibiotics: Gentamicin 5 µg/ml (10% of normal gentamicin level)

Anti-fungal agent: Amphotericin B 0.25 µg/ml

pH Indicator: Phenol red

Other additives: Lipid precursors used to enhance epidermal barrier formation (proprietary)

Alternatives: Phenol red-free (EPI-200-PRF), antibiotic-free (EPI-200-ABF), anti-fungal-free (EPI-200-AFF), or hydrocortisone-free medium and tissue (EPI-200-HCF) are available. Agents are removed at least 3 days prior to shipment. Please discuss your specific media formulation and tissue needs with MatTek scientific personnel before placing your order.

Assay/Maintenance medium: Three maintenance media are available. EpiDerm maintenance medium (EPI-100-MM) is equivalent to the assay medium (EPI-100-ASY) and is used for short-term toxicological testing (≤ 24 hrs). New Maintenance Medium, EPI-100-NMM, contains Keratinocyte Growth Factor (KGF) and is used for studies up to 3 days. EPI-100-NMM-3 contains higher levels of KGF and is used for longer-term studies (> 3 days). For good retention of epidermal morphology for at least 2 weeks, the use of EPI-100-NMM-3 (5.0 mL changed every other day) is recommended

Quality Control and Sterility

Visual inspection: All tissues are visually inspected and if physical imperfections are noted, tissues are rejected for shipment

End-use testing: Tissues are exposed to 1% Triton X-100 for 4, 6, 8 and 10 hours. The time of exposure required to reduce the tissue viability (ET-50) using the MTT viability assay is determined (See MatTek EpiDerm MTT ET-50 Protocol) for each lot of tissue. ET-50's must fall within the range of the 1996 EpiDerm database of 4.77 – 8.72 hours. ET-50's in customers' labs may differ slightly from the MatTek results.

Sterility: All media used throughout the production process is checked for sterility. Maintenance medium is incubated with and without antibiotics for 1 week and checked for sterility. The agarose gel from the 24-well plate used for shipping is also incubated for 1 week and checked for any sign of contamination

Screening for pathogens: All cells are screened and are negative for HIV, hepatitis B and hepatitis C using PCR. However, no known test method can offer complete assurance that the cells are pathogen free. Thus, these products and all human derived products should be handled at BSL-2 levels (biosafety level 2) or higher as recommended in the CDC-NIH manual, "Biosafety in microbiological and biomedical laboratories," 1998. For further assistance, please contact your site Safety Officer or MatTek technical service

Notification of lot failure: If a tissue lot fails our QC or sterility testing, the customer will be notified and the tissues will be replaced without charge when appropriate. Because our QC and sterility testing is done post-shipment, notification will be made as soon as possible (Under normal circumstances, ET-50 failures will be notified by Wednesday 5 p.m.; sterility failures will be notified within 8 days of shipment)