

EpiAirway Technical Specifications

Tissue

<u>Kit</u>: A standard EpiAirway kit (AIR-100) 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 tissue culture substrate is chemically modified with a pore size of 0.4
 µm
- 12mm & 6.5mm Transwell tissue culture substrate is chemically modified with a pore size of 0.4 μm
- 96-well HTS plates tissue culture substrate is chemically modified with a pore size of 0.4 μm

Culture: Air-liquid interface

Histology: 3-4 cell layers – pseudostratified, mucociliary morphology and tissue structure

<u>Lot numbers</u>: 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 Room Temperature on medium-supplemented, agarose gels

Shipment day: Every Monday

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

<u>Shelf life</u>: Including time in transit, tissues may be stored at room temperature for up to 3 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 room temperature (Wednesday morning)

<u>Length of experiments</u>: Tissue cultures can be continued for THREE (3) MONTHS or more with good retention of normal morphology. Tissues must be fed every other day with 5.0 ml of maintenance medium (AIR-100-MM). Cell culture inserts are placed atop washers (EPI-WSHR) or culture stands (MEL-STND) in 6-well plates to allow us of 5.0 ml. See <u>technical reference #631</u>.

Cells

Type: Normal human tracheal/bronchial epithelial cells (NHBE);

Genetic make-up: Single donor

<u>Derived from</u>: Healthy, non-smoker (alternate donors available upon request)

Alternatives: NHBE from Asthmatic, COPD and Smoker donors

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

Medium

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

Growth factors/hormones: Epidermal growth factors and other proprietary factors

Serum: None

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

Anti-fungal agent: Amphotericin B 0.25 μg/ml

<u>pH Indicator</u>: Phenol red
<u>Other additives</u>: Proprietary

Alternatives: Phenol red-free, antibiotic-free, anti-fungal-free medium and tissues are available. Agents are

removed at least 3 days prior to shipment.

Assay medium: AIR-100-ASY

Maintenance medium: AIR-100-MM

Quality Control and Sterility

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

<u>End-use testing</u>: Transepithelial electrical resistance (TEER) of each EpiAirway lot is measured using an EVOM2 epithelial Volthommeter and Endohm electrode chamber (World Precision Instruments). A minimum TEER of 300 Ohm • cm² is required for QC release.

<u>Sterility</u>: All media used throughout the production process is checked for sterility. Maintenance medium is incubated 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

<u>Screening for pathogens</u>: 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, TEER failures will be notified by Wednesday 5 p.m.; sterility failures will be notified within 8 days of shipment)

Alternative Tissues

AIR-100-R: EpiAirway, normal rat 3D airway epithelial tissue model, 0.6cm²