

PRODUCT DESCRIPTION

Basement membranes are continuous sheets of specialized extracellular matrix that form an interface between endothelial, epithelial, muscle, or neuronal cells and their adjacent stroma. Basement membranes are degraded and regenerated during development and wound repair. They not only support cells and cell layers, but they also play an essential role in tissue organization that affects cell adhesion, migration, proliferation, and differentiation. Basement membranes provide major barriers to invasion by metastatic tumor cells.

INTENDED USE

Cultrex Basement Membrane Extract (BME), 5X Solution is a soluble form of basement membrane purified from Engelbreth-Holm-Swarm (EHS) tumor. This solution has been developed, produced and qualified for the Cultrex BME Cell Invasion Assays. The Cultrex BME, 5X Solution is processed to reduce matrix-associated growth factors and provide a more defined model system.

PRODUCT SPECIFICATIONS

Concentration	10 mg/mL
Source	Murine Engelbreth-Holm-Swarm (EHS) tumor.
Storage Buffer	Dulbecco's Modified Eagle's Medium containing 10 µg/mL gentamicin sulfate and no phenol red.
Stability	Product is stable for 3 months from date of manufacture. See lot specific Certificate of Analysis for expiration date.
Storage	Store at ≤ -70 °C. Product may be thawed and dispensed into working aliquots. Avoid freeze-thaw cycles.

PRECAUTION

When handling bio-hazardous materials such as human cells, safe laboratory procedures should be followed and protective clothing should be worn.

LIMITATIONS

- FOR LABORATORY RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES.
- The safety and efficacy of this product in diagnostic or other clinical uses has not been established.
- Results may vary due to variations among tissue/cells derived from different donors or sources.

MATERIAL QUALIFICATIONS

Sterility Testing:

- No mycoplasma concentration detected by PCR.
- Tested following USP <71> sterility guidelines.
- Endotoxin concentration ≤ 20 EU/mL by LAL assay.

Cell Invasion:

- Less than 10% cell invasion for MCF7 cells in response to 10% FBS.

COATING PROCEDURE

1. Refrigerator temperatures may vary; therefore thaw Cultrex BME, 5X Solution at 2-8 °C overnight on ice in a refrigerator. Cultrex™ BME, 5X Solution gels in 15-30 minutes above 15 °C; keeping the BME container and coated labware on ice will prevent gelling and extend working times.
2. Dilute 500 μ L 10X Coating Solution (R&D Systems®, Catalog # 3455-096-03) in 4.5 mL of sterile, deionized water to make 1X Coating Solution; filtration at 0.2 μ m is recommended. Store at 2-8 °C.
3. For highly invasive cells, dilute 1 mL of Cultrex BME, 5X Solution in 4 mL of 1X Coating Solution on ice immediately before coating. Less invasive cell types may require a more permissive barrier, the Cultrex BME, 5X Solution may be diluted as far as 0.1X.
4. Working on ice, prepare 5 mL of 1X BME Solution in a sterile 15 mL conical tube, cap tube, and gently invert to mix.
5. Aliquot 50 μ L of 1X BME Solution per well of Cell Invasion/Migration Plates, 96-well (R&D Systems, Catalog # 3455-096-01) or 100 μ L of 1X BME Solution per insert of Cell Invasion/Migration Plates, 24-well (R&D Systems, Catalog # 3455-024-01). Gently tap side of device a few times, and visually inspect wells for dispersion of coating. All wells should be coated except at least three migration control wells (optional). Coat for 4 hours or overnight at 37 °C in a CO₂ incubator.
6. Carefully aspirate coating solution from wells and immediately add cells.

REFERENCES

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