

Geltrex[®] LDEV-Free hESC-qualified Reduced Growth Factor Basement Membrane Matrix

Description

Geltrex[®] LDEV-Free hESC-qualified Matrix is used routinely for attachment and maintenance of human embryonic stem cells (hESCs). Each lot of Geltrex[®] LDEV-Free hESC-qualified Matrix has been function tested to provide the unique micro-environmental requirements for the growth and maintenance of pluripotency of hESCs, therefore eliminating the need for customers to test multiple lots. Geltrex[®] LDEV-Free hESC-qualified Matrix can also be used for promotion and maintenance of a differentiated phenotype in a variety of cell cultures including primary epithelial cells, endothelial cells, smooth muscle cells, and human induced pluripotent stem cells (iPSCs).

The major components of Geltrex[®] LDEV-Free hESC-qualified Matrix include laminin, collagen IV, entactin, and heparin sulfate proteoglycan which provide the foundation for three-dimensional (3D) culture studies. The extract gels at 37°C to form a reconstituted basement membrane. Basement membranes are continuous sheets of specialized extracellular matrix that form an interface between endothelial, epithelial, muscle, or neuronal cells and their adjacent stroma. In addition to its role in the physical support and compartmentalization of tissues, basement membrane influences a number of cellular functions such as proliferation, adhesion, migration, differentiation, and polarization. Basement membrane is thus implicated in biological processes such as development, tissue maintenance, regeneration, and wound repair as well as various pathological processes such as tumor growth and metastasis. Geltrex[®] LDEV-Free hESC-qualified Matrix has been employed in angiogenesis assays, neurite outgrowth assays, and tumor cell invasion assays.

| Product | Catalog No. | Amount | Storage | Shelf Life* |
|---|----------------------|--------------|----------------|-------------|
| Geltrex [®] LDEV-Free hESC-qualified Reduced Growth Factor Basement Membrane Matrix | A1413301 A1413302 | 1 mL 5 mL | -80°C to -20°C | 18 months |

* Shelf Life duration is determined from Date of Manufacture.

Product Use

For Research Use Only. Not for use in diagnostic procedures.

Important Information

- **Source:** Murine Engelbreth-Holm-Swarm (EHS) tumor, protein concentration ranges from 12–18 mg/mL. Refer to certificate of analysis for specific lot information.
- Thaw Geltrex[®] Matrix in a refrigerator at 2°C to 8°C overnight.
- When working with smaller volumes of Geltrex[®] Matrix, dispense appropriate required working volumes and store at -80°C to -20°C.
- Avoid multiple freeze/thaw cycles
- Geltrex[®] Matrix gels in 5–10 minutes above 15°C. When working from a full 5 mL vial, it is unnecessary to keep it on ice if used within 5 minutes and the environmental temperature does not exceed 25°C. Since smaller volumes warm more quickly, partial tubes and aliquots should be kept on ice to prevent premature gelling.
- Formulated without phenol red to minimize potential for estrogen-like effects.

Safety Information

Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves.

Coating Procedures

Geltrex[®] LDEV-Free hESC-qualified Matrix is tested for hESC applications. A protein concentration ≥ 9 mg/mL is used for differentiation studies of hESC's. Extract diluted below 9 mg/mL does not form a gel, and will only support the propagation and maintenance of pluripotency of hESCs when grown with media designed for feeder free propagation of hESCs.

For more information on 3D Cell Culture go to www.lifetechnologies.com/3D-cellculture.

Important: We recommend that the following procedures be performed in an aseptic environment using aseptic techniques to prevent contamination.

Thin Gel Method (non-gelling) for Propagation of hESC

1. Thaw Geltrex[®] Matrix solution. See **Important Information**.
2. Mix Geltrex[®] Matrix solution by slowly pipetting up and down; be careful not to introduce air bubbles.
3. Dilute 1 mL Geltrex[®] Matrix solution into 99 mL pre-chilled (4°C) DMEM/F-12 medium (or equivalent). Determine optimal coating concentration for your application empirically. Adjust volumes accordingly.
4. Add sufficient diluted Geltrex[®] Matrix solution to cover the entire growth surface area (e.g., 1.5 mL for 35-mm dish, 3 mL for 60-mm dish). The coated dish is stable for two weeks when wrapped with Parafilm[®] sealing film and stored at 4°C. *Do not allow coated surface to dry out. It is critical to maintain a storage temperature of 4°C to avoid premature gelling.*
5. Incubate coated plates at 37°C for a minimum of 60 minutes.
6. At time of use, *we recommend keeping plates at room temperature for one hour before aspirating.* Carefully aspirate off the supernatant above the Geltrex[®] coating and immediately plate cells in pre-equilibrated cell culture medium.

Lot Qualification

12 well cell culture plates are coated with each test lot of "Geltrex® LDEV-Free hESC-qualified Matrix". hESCs are grown on coated control and test plates in STEMPRO® hESC SFM and are monitored for expansion (See Figure 1). An hESC negative control is grown with retinoic acid to induce differentiation and is used as an internal PCR control (See Figure 2). After completion of the hESC Growth Assay, the test & control samples are assessed by PCR Analysis (See Figure 3).

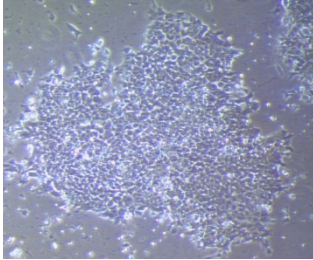


Figure 1: hESC Control

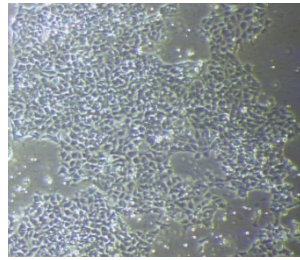


Figure 2: hESC Negative Control

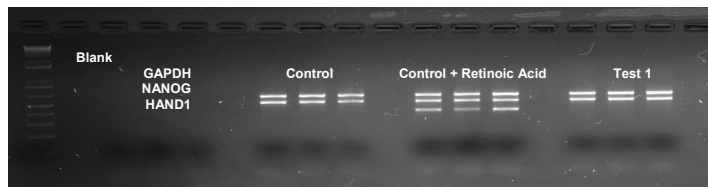










Figure 3: PCR Analysis

Related Products

| Product | Catalog No. |
|--|-------------|
| DMEM/F-12, GlutaMAX™ | 10565 |
| 2-Mercaptoethanol (1000X), liquid | 21985 |
| FGF-basic (AA 10-155) Recombinant Human | PHG0026 |
| StemPro® hESC SFM | A10007-01 |
| StemPro® MSC SFM | A10332-01 |
| StemPro® MSC SFM XenoFree | A10675-01 |
| STEMPRO® EZPassage™ Disposable Cell Passaging Tool | 23181-010 |
| STEMPRO® EZChek™ Human Tri-Lineage Multiplex PCR Kit | 23191-050 |
| TrypLE™ Select CTS™ (1X), liquid, without Phenol Red | A12589 |
| KnockOut™ DMEM (1X), liquid | 10829 |
| KnockOut™ SR XenoFree, | A10992 |
| KnockOut™ DMEM/F-12 (1X), liquid | 12660 |
| GlutaMAX™-I CTS™, 200mM (100X), liquid | A12860 |
| BG01VhOG Cells | R7799-105 |
| Human Umbilical Vein Epithelial Cells (HUVEC) | C0035C |

Explanation of Symbols and Warnings

The symbols present on the product label are explained below:

| | | | |
|--|--|--|--|
|  |  |  |  |
| Use By: | Manufacturer | Batch code | Catalog number |
|  |  |  |  |
| Temperature Limitation | Consult instructions for use | Caution, consult accompanying documents | Sterilized using aseptic processing techniques |

Limited Product Warranty

Life Technologies Corporation and/or its affiliate(s) warrant their products as set forth in the Life Technologies' General Terms and Conditions of Sale found on Life Technologies' website at www.lifetechnologies.com/termsandconditions. If you have any questions, please contact Life Technologies at www.lifetechnologies.com/support.

For additional technical information such as Safety Data Sheets (SDS), Certificates of Analysis, visit www.lifetechnologies.com/support
For further assistance, email techsupport@lifetech.com

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