

CERTIFICATE OF ANALYSIS

PRODUCT: Corning® Matrigel® hESC-qualified Matrix, 5 ml vial
CATALOG NUMBER: 354277

LOT NUMBER: 7282005

DILUTION FACTOR: 217 uL

SOURCE: Engelbreth-Holm-Swarm (EHS) Mouse Tumor

FORMULATION: Dulbecco's Modified Eagle's Medium with 50 µg/ml gentamycin
Corning Matrigel hESC-qualified Matrix is compatible with all culture media

STORAGE: Store at -20°C. Avoid multiple freeze-thaws. Do not store in frost-free freezer. **KEEP FROZEN.**

QUALITY CONTROL:

Specification	Criteria	Result
Endotoxin	Endotoxin units (EU)/ml are measured by Limulus Amoebocyte Lysate assay.	< 1.5
hESC Qualification	Qualified for use with StemCell Technologies' mTeSR™1 medium medium. Human embryonic stem cells were grown in mTeSR™1 on Corning Matrigel hESC-qualified Matrix-coated plates for five passages and remained undifferentiated by standard morphology and surface marker expression.	PASS
Gelling	Tested for ability to gel quickly and maintain this form with culture medium for a period of 14 days at 37°C.	PASS
Biological Activity	Biological activity is determined using a neurite outgrowth assay. Chick dorsal root ganglia are plated on a 1.0 mm layer of Corning Matrigel Matrix. Tested for a positive neurite outgrowth response after 48 hours without addition of nerve growth factor.	PASS
Sterility	Tested for the presence of bacteria, fungi and mycoplasma.	NEGATIVE
MAP Test	Mouse colonies screened for Sendai, MHV, PVM, TEMV/GDVII, Ectro, Polyoma, MRV/EDIM, LCM, MCMV, M.Ad, Reo, MPV, LDEV/LDHV, MTV, Hantaan, K, RCMV, CARB	NEGATIVE
PCR Test	Tumor source tested for <i>Mycoplasma spp.</i> , <i>Helicobacter</i> , LDEV/LDHV, Sendai, MHV, PVM, MMV/MVM, MPV, Reo (1, 2, 3), MRV/EDIM, Ectro, LCM, K, MTV, Polyoma, Hantaan, Seoul, M. Ad (1, 2), MCMV, Norovirus, TMEV/GDVII, KRV, Toolan's H-1, RCV/SDA Finished goods tested for LDEV/LDHV.	NEGATIVE

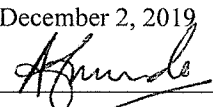
SAFETY RECOMMENDATION: Handle in accordance with good industrial hygiene and laboratory safety practices

NOTE: Human embryonic stem cell research may be restricted in your national jurisdiction. Prior to the use of this product for hESC research, please consult your applicable laws regarding such activities.

Expiration Date: December 2, 2019

Quality Assurance

Date



January 8, 2018

Discovery Labware, Inc., Two Oak Park, Bedford, MA 01730, Tel: 1.978.442.2200 (U.S.)
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GUIDELINES FOR USE

PRODUCT: Corning® Matrigel® hESC-qualified Matrix, 5 mL vial

CATALOG NUMBER: 354277

BACKGROUND: Basement membranes are continuous sheets of specialized extracellular matrix that are found at the dermal-epidermal junction, at the base of all lumen-lining epithelia throughout the digestive, respiratory, reproductive and urinary tracts and that underlie parenchyma of endocrine and exocrine glands.

Corning Matrigel hESC-qualified Matrix is a soluble basement membrane extract of the Engelbreth-Holm-Swarm (EHS) tumor that gels at room temperature to form a genuine reconstituted basement membrane.¹ The major components of Corning Matrigel hESC-qualified Matrix are laminin, collagen IV, entactin and heparan sulfate proteoglycan.²⁻³ Growth factors, collagenases, plasminogen activators and other undefined components have also been reported in Corning Matrigel hESC-qualified Matrix.⁴⁻⁵

STEM CELLS: Historically, human embryonic stem (hES) cell derivation and culturing techniques utilized serum and/or mouse embryonic fibroblast (MEF) feeder layers.⁶ An ideal environment for hES cell research consists of both a cell culture surface specifically qualified for hES cells, and a serum-free, defined medium. Corning Matrigel hESC-qualified Matrix and STEMCELL Technologies' mTeSR™1 (developed under license from the WiCell Research Institute),⁷ a high quality surface and medium combination, create the first complete environment to support feeder-independent expansion of hES cells.

Corning Matrigel hESC-qualified Matrix is an optimized surface for your stem cell research. It has been qualified as mTeSR1-compatible by STEMCELL Technologies, eliminating the need for time-consuming screening, while providing the reproducibility and consistency essential for your hES cell research. When coupled with a variety of culture media, Corning Matrigel hESC-qualified Matrix has been widely accepted as an alternative substrate for the culture of hES cells as well as human induced pluripotent stem (iPS) cells.⁸⁻¹¹ The mTeSR1 formulation is defined and serum-free, and has been designed to maintain and expand hES cells in an undifferentiated state when used with Corning Matrigel hESC-qualified Matrix as a substrate. It does not require any further addition of growth factors or supplements.

The mTeSR1 formulation and Corning Matrigel hESC-qualified Matrix have been shown to be a successful combination for culturing different hES cell lines for up to 20 passages.¹² Cells maintained in mTeSR1 express high levels of pluripotency markers such as Oct-3/4 and SSEA-3, and pluripotency of cells maintained in mTeSR1 has also been demonstrated by the ability of these cells to differentiate into all three germ layers in the teratoma assay.^{7,13}

SOURCE: Engelbreth-Holm-Swarm (EHS) Mouse Tumor

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- FORMULATION:** Dulbecco's Modified Eagle's Medium with 50 µg/mL gentamycin.
Corning® Matrigel® hESC-qualified Matrix is compatible with all culture media.
- STORAGE:** Stable when stored at -20°C. Store aliquots in either the -20°C or -70°C freezer until ready for use. Freeze thaws should be minimized by aliquotting into one time use aliquots. **DO NOT STORE IN FROST-FREE FREEZER. KEEP FROZEN.**
- EXPIRATION DATE:** The expiration date for Corning Matrigel hESC-qualified Matrix is lot specific and can be found on the product Certificate of Analysis.
- CAUTION:** It is extremely important that Corning Matrigel hESC-qualified Matrix and all cultureware or media coming in contact with Corning Matrigel hESC-qualified Matrix should be pre-chilled/ice-cold since Corning Matrigel hESC-qualified Matrix will start to gel above 10°C.
- RECONSTITUTION AND USE:** Color variations may occur in frozen or thawed vials of Corning Matrigel hESC-qualified Matrix, ranging from straw yellow to dark red due to the interaction of carbon dioxide with the bicarbonate buffer and phenol red. This is normal, does not affect product efficacy, and will disappear upon equilibration with 5% CO₂.
- Thaw Corning Matrigel hESC-qualified Matrix by submerging the vial in ice in a 4°C refrigerator, in the back, overnight. Once Corning Matrigel hESC-qualified Matrix is thawed swirl vial to ensure that material is evenly dispersed. Spray top of vial with 70% ethanol and air dry. Keep product on ice and handle using sterile technique. Dispense material into appropriate aliquots in pre-cooled tubes, switching tips whenever Corning Matrigel hESC-qualified Matrix is clogging the tip and/or causing the pipet to measure inaccurately and refreeze immediately. Gelled Corning Matrigel hESC-qualified Matrix may be re-liquified if placed at 4°C in ice for 24-48 hours.
- DILUTION FACTOR:** The dilution is calculated for each lot based on the protein concentration. To use with STEMCELL Technologies' mTeSR™1 medium, prepare aliquots according to the dilution factor provided on the Certificate of Analysis. The volume of the aliquots is typically between 270-350 µL.
- To Use:** Add one aliquot of Corning Matrigel hESC-qualified Matrix to 25 mL of DMEM/F-12 to coat four 6-well plates (1 mL/well) or three 100 mm dishes (8 mL/dish). Incubate the cultureware at room temperature (15-25°C) for at least 1 hour before use. Aspirate the remaining liquid from cultureware just before use. Ensure that the tip of the pipet does not scratch the coated surface. Plates/dishes are now ready to use.

NOTE: For more details on specific applications of Corning Matrigel matrix visit support page at www.corning.com/lifesciences for technical bulletins/application notes, protocols, and frequently asked questions.

SAFETY RECOMMENDATION: Handle in accordance with good industrial hygiene and laboratory safety practices

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REFERENCES:

1. Kleinman HK, et al, Basement membrane complexes with biological activity, *Biochemistry*, **25**:312 (1986).
2. Kleinman HK, et al, Isolation and characterization of type IV procollagen, laminin, and heparan sulfate proteoglycan from the EHS sarcoma, *Biochemistry*, **21**:6188 (1982).
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7. Ludwig TE, et al, Feeder-independent culture of human embryonic stem cells, *Nat Methods*, **3**(8):637 (2006).
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12. Ludwig TE, et al, Derivation of human embryonic stem cells in defined conditions, *Nat Biotechnol*, **24**:185 (2006).
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California Proposition 65 Notice

WARNING: This product contains a chemical known to the state of California to cause cancer.

Component: **Chloroform**

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RELATED PRODUCT: mTeSR™1 Maintenance Medium for Human Embryonic Stem Cells 500 mL (1 kit) Cat. No. **05850**. Please visit www.stemcell.com for more information.

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