Optimize Serum-Free Embryonic Stem Cell Culture with GIBCO® Media

- Improve growth and maintenance of undifferentiated ES cells
- Eliminate drawbacks of FBS

Product	Optimized For	Applications
KnockOut™ D-MEM	Murine and human embryonic	Growth and maintenance of undifferentiated ES cells
KnockOut [™] Serum Replacement	stem (ES) cells	for production of transgenic mice. Growth and maintenance of both human and murine ES cells used in differentiation studies.

Serum-Free Media

KnockOut[™] D-MEM

KnockOut[™] D-MEM is a special basal medium designed specifically to improve the morphology of ES cells. The osmolality of KnockOut[™] D-MEM has been reduced to better mimic the natural environment of embryonic tissue.

When used together, KnockOut[™] D-MEM and KnockOut[™] SR significantly reduce ES cell differentiation as compared to ES Cell Qualified FBS and traditional D-MEM (*figure 1*).

ES Cell Differentiation Comparison



Figure 1. Undifferentiated ES Cell Growth. Murine D3 ES cells were cultured at low density in D-MEM or KnockOut[™] D-MEM supplemented with ES Cell Qualified FBS or KnockOut[™] SR, and traditional supplements. No LIF (Leukocyte Inhibitory Factor) was used. After 7 days, colonies were fixed and stained for alkaline phosphatase, a marker for undifferentiated ES cells. Undifferentiated colonies were scored based on morphology and staining characteristics.

KnockOut[™] Serum Replacement Eliminates Use of FBS

KnockOut[™] Serum Replacement (KnockOut[™] SR) is a serum-free formulation designed to directly replace FBS in ES cell culture and provide the highest level of performance and ease of use. KnockOut[™] SR can replace FBS for growth and maintenance of undifferentiated ES cells, ES cell cryopreservation, blastocyst injection, electroporation/cationic lipid transfection, drug selection, isolation of new ES cell lines, embryoid body formation, and *in vitro* differentiation studies.

Custom Production and Packaging

When you need a unique formulation or special packaging, our Custom Product Services team can modify GIBCO[®] catalog media formulations and packaging to meet your particular requirements.

For information, call 1-800-955-6288, Ext. 46966.

References

KnockOut[™] Serum Replacement

- Assady, S., Maor, G., Amit, M., Itskovitz-Eldor, J., Skorecki, K., and Tzukerman, M. (2001) Insulin Production by Human Embryonic Stem Cells. *Diabetes* **50**, 1691.
- Goldsborough, M., Tilkins, M.L., et al. (1998) FOCUS® 20, 8.
- Hancock, C.R., Wetherington, J.P., Lambert, N.A., and Condie, B.G. (2000) Neuronal Differentiation of Cryopreserved Neural Progenitor Cells Derived from Mouse Embryonic Stem Cells. *Biochemical and Biophysical Research Communications* 271, 418.
- Schuldiner, M., Itskovitz-Eldor, J., and Benvenisty, N. (2003) Selective Ablation of Human Embryonic Stem Cells Expressing a "Suicide" Gene. Stem Cells 21, 257.
- Tzuckerman, M., Shachaf, C., Ravel, Y., Braunstein, I., Cohen-Barak, O., Yalon-Hacohen, M., and Skorecki, K.L. (2000) Identification of a Novel Transcription Factor Binding Element Involved in the Regulation by Differentiation of the Human Telomerase (hTERT) Promoter. *Mol. Biol. Cell.* 11 (12), 4381.

Ordering Information

Description	Catalog No.	Size	
- KnockOut™ D-MEM	10829-018	500 ml	
nockOut™ Serum Replacement	10828-028	500 ml	
Growth Factors			
Basic Fibroblast Growth Factor (bFGF), Human, Recombinant	13256-029	10 µg	
Epidermal Growth Factor (EGF), Human, Recombinant	13247-051	100 µg	
Insulin-Like Growth Factor-I (IGF-I), Human, Recombinant	13245-063	10 µg	
Nutritional Supplements			
GlutaMAX™-I Supplement Stable form of L-glutamine.	35050-061	100 ml	
L-Glutamine-200 mM (100X), liquid	25030-081	100 ml	
L-Glutamine, powder	21051-024	100 g	
Selective Antibiotics			
Geneticin [®] Selective Antibiotic, liquid	10131-035	20 ml	
50 mg/ml.	10131-027	100 ml	
Wash Buffer			
Dulbecco's Phosphate-Buffered Saline (1X), liquid Contains no calcium or magnesium.	14190-144	500 ml	
Cell Dissociation			
Trypsin-EDTA (1X), liquid 0.05% Trypsin, 0.53 mM EDTA•4Na.	25300-054	100 ml	
Trypsin-EDTA (1X), liquid 0.25% Trypsin, 1 mM EDTA•4Na.	25200-056	100 ml	
Trypsin Inhibitor, soybean Inactivates trypsin in serum-free cell media.	17075-029	1 g	
Additional Trypsin options	See the	See the GIBCO [®] Catalog.	
Transfection			
Lipofectamine™ 2000 Reagent	11668-027	0.75 ml	
Transfects adherent 293 cells.	11668-019	1.5 ml	
_ipofectamine™ 2000 CD An animal-origin-free, chemically-defined version of Lipofectamine [™] 2000.	12566-014	1 ml	
Related Products			
CO ₂ -Independent Medium (1X), liquid For handling mouse embryos in atmospheric conditions.	18045-088	500 ml	
MEM Non-Essential Amino Acids Solution 10 mM (100X), liquid	11140-050 11140-076	100 ml 20 × 100 ml	
2-Mercaptoethanol (1,000X), liquid Helps prevent oxidation.	21985-023	50 ml	



www.invitrogen.com



Corporate Headquarters: Invitrogen Corporation • 1600 Faraday Avenue • Carlsbad, California 92008 U.S.A. Tel: 1 760 603 7200 • Tel (Toll Free): 1 800 955 6288 • Toll Free Fax: 1 800 331 2286 • E-mail: tech_service@invitrogen.com European Headquarters: Invitrogen Ltd • 3 Fountain Drive • Inchinnan Business Park • Paisley PA4 9RF, UK Tel: + 44(0) 141 814 6100 • Fax: + 44(0) 141 814 6260 • E-mail: eurotech@invitrogen.com

These products are for research use, and where appropriate, as raw material components in further cell culture manufacturing applications. They are not intended for human or animal diagnostic, therapeutic, or other clinical uses, unless otherwise stated.[©] 2003 Invitrogen Corporation PAS03-062MS Part No. 332-032443F



ANIMAX FRED RIGIN