SAFC Biosciences™
Accelerate Success™

Product Information

EX-CELL™ 610-HSF Serum-Free Medium for Hybridoma Cells

with L-glutamine

CATALOG NO. 14610C

Description

EX-CELL™ 610-HSF is a low-protein (11 mg/L) serum-free liquid medium. Originally developed to support the growth of hybrid cells in culture, it has been shown to support a wide range of cells including lymphoid and epithelial cells and B cell hybridomas of murine, rat and human origin. EX-CELL™ 610-HSF has been used in stationary culture systems and in large-scale bioreactors. In both types of culture, the production of cellular products, particularly monoclonal antibodies, has been shown to equal or exceed levels seen when the same cells are cultured in the presence of Fetal Bovine Serum (FBS).

Formulation

The formulation for EX-CELL™ 610-HSF is proprietary to SAFC Biosciences. For additional information please call our Technical Service department.

Precautions

Use aseptic technique when handling or supplementing this medium. This product is for research or for further manufacturing use. THIS PRODUCT IS NOT INTENDED FOR HUMAN OR THERAPEUTIC USE.

Storage

Store liquid medium at 2 to 8 C, protected from light. Do not use after the expiration date.

Indications of Deterioration

Medium should be clear and free of particulates and flocculent material. Do not use if medium is cloudy or contains precipitate. Other evidence of deterioration may include color change, pH shift or degradation of physical or performance characteristics.

Preparation Instructions

EX-CELL™ 610-HSF is formulated with L-glutamine and with sodium bicarbonate. Other supplements, such as antibiotics, can be added as sterile supplements to the sterilized solution using aseptic technique. Storage conditions and shelf life of the supplemented product may be affected by the nature of the supplements.

Methods for Use

Adaptation

1. Culture the cells to a density of 3-5 x 10^5 cells/mL in EX-CELL™ 610-HSF medium containing 5% gamma irradiated Fetal Bovine Serum (FBS) (Catalog No. 12107C). Maintain the cells through 2 passages at this concentration of serum.
2. Subculture into EX-CELL™ 610-HSF containing 2.5% serum and maintain through 2 passages with the same concentration of serum.
3. Subculture into EX-CELL™ 610-HSF containing 1% serum and maintain through 2 passages in the same serum concentration.
4. If no major growth changes in the cells are observed, pass the cells into serum-free EX-CELL™ 610-HSF.
5. Once cells are growing in EX-CELL™ 610-HSF without serum, they can be subcultured every 3 - 4 days as necessary.

Culture Techniques

Once cultures are fully adapted, the cells should be passed every 3 - 4 days at a seeding density of at least 2 x 10^5 cells/mL. An optimal seeding density should be determined by the researcher for each application and cell type. EX-CELL™ 610-HSF does not contain cholesterol. If using a cholesterol-dependent cell line, supplement with 10 - 20 µm cholesterol (3.8 - 7.7 mg/L).

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When passing the cells, carryover should not exceed 25% of the final volume. If carryover exceeds 25%, centrifugation is recommended. Cells propagated in serum-free medium are extremely fragile. Standard techniques for centrifugation must be modified to include low-speed centrifugation to prevent damage to cells that have been propagated in serum-free medium.

During adaptation, normal trypsin concentrations may be used, but incubations should be carried out at 4 C, and exposure time should be minimal. SAFC Biosciences recommends the use of a soybean trypsin inhibitor (0.1%), or sedimentation by centrifugation to remove the trypsin. Soybean trypsin inhibitor should be used with caution, as it is toxic to some cell types. Cells may also be dislodged with NO-ZYME™ (Catalog No. 59226C), a non-enzymatic dissociating agent.

Cryopreservation

Freezing:

Cells can be frozen in EX-CELL™ 610-HSF without the reintroduction of serum.

1. Choose cultures in logarithmic growth with viabilities above 90%.
2. Prepare a freezing medium consisting of 45% cold EX-CELL™ 610-HSF medium, 45% spent medium and 10% dimethyl sulfoxide (DMSO).
3. Centrifuge the cells at 200 *g* for 5 minutes. Remove the supernatant.
4. Resuspend the cells in the freezing medium at 5 x 10^6 to 1 x 10^7 cells/mL.
5. Rapidly transfer 1 - 2 mL of this suspension to sterile cryovials.
6. Place the vials at -20 C for 3 - 4 hours, then transfer to -70 C for 16 - 24 hours
7. For long-term storage, transfer the vials to liquid nitrogen vapor.

Thawing:

1. Rapidly thaw a vial of frozen cells in a 37 C water bath.
2. Transfer the cells aseptically to a centrifuge tube containing 10 mL of cold EX-CELL™ 610-HSF medium.
3. Using low-speed centrifugation, pellet the cell suspension at 200 *g* for 5 minutes and carefully decant the supernatant without disturbing the cell pellet.
4. Resuspend the cells in 5 mL of EX-CELL™ 610-HSF medium.
5. Count the cells for viability and transfer to a sterile shaker flask at a seeding density of 2-4 x 10^5 cells/mL.
6. When the culture has reached a density of 1 x 10^6 cells/mL, passage the cells using standard cell culture techniques.

Characteristics

Appearance
Clear orange-red solution

Endotoxin
≤ 10.0 EU/mL

Osmolality (as supplied)
285 - 325 mOsm/kg H_2O

pH (as supplied)
6.7 - 7.1

Sterility
No microbial growth detected

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