SAFC Biosciences™
Accelerate Success™

Product Information

EX-CELL™ 405 Serum-Free Medium for Insect Cells

with L-glutamine

CATALOG NO. 14405C

Description

Baculoviruses are powerful expression systems for production of recombinant proteins. The most well known and highly investigated baculovirus vector expression system (BEVS) uses *Spodoptera frugiperda* cells (especially Sf9) as the host cell of choice for infection with recombinant *Autographa californica* nuclear polyhedrosis virus for heterologous protein expression. Recent investigations have shown that other cell lines may be better expression substrates than spodopteran cells. The BTI-TN-5B1-4 clone of *Trichoplusia ni*, commonly referred to as High Five™, is reported to be a superior host substrate for the expression of selected recombinant proteins using BEVS. This cell line portrays strong adherent characteristics, but can be adapted to suspension growth.

EX-CELL™ 405 is a serum-free liquid medium for optimal growth and viability of BTI-TN-5B1-4 (Tn5, High Five™) cells. In addition, EX-CELL™ 405 supports recombinant protein production in High Five™ cells to higher levels than reported for Sf9 cells. EX-CELL™ 405 is a complete medium. No protein supplements need to be added to this medium prior to use.

Formulation

The formulation for EX-CELL™ 405 is proprietary to SAFC Biosciences. For additional information please call our Technical Services department.

Precautions

Use aseptic technique when handling or supplementing this medium after filtration. 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 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 change or degradation of physical or performance characteristics.

Preparation Instructions

EX-CELL™ 405 is formulated with L-glutamine. Supplements, such as antibiotics, can be added to the sterilized medium using aseptic technique. Storage conditions and shelf life of the product may be affected by the nature of the supplements.

Methods for Use

Adaptation

Insect cells that have been grown in a conventional serum-supplemented medium can be readily grown in EX-CELL™ 405 with little or no adaptation. During adaptation, growth rates will usually be somewhat slower than normal expected rates.

Culture Techniques

Once fully adapted, the cells should be passed every 3 - 4 days at a seeding density of at least 2-4 x 10^5 cells/mL in shaker or spinner flasks. Seed 50 mL of cell suspension in 125 mL shaker flasks and 100 mL of cell suspension in 250 mL shaker flasks. Shaker speed should be 100 - 120 rpm and spinner speed should be 60 - 75 rpm.

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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. For successful results, care must be taken when subculturing cells. 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.

Cryopreservation

Freezing:

Cells can be frozen in EX-CELL™ 405 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™ 405 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™ 405 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™ 405 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 yellow solution

Endotoxin
≤ 20.0 EU/mL

Osmolality (as supplied)
425 - 465 mOsm/kg H_2O

pH (as supplied)
6.0 - 6.4

Sterility
No microbial growth detected

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