MUC16 (CA125), variant 4 (region 13785-14507) CHO Cell Line

#78848

Description

Engineered CHO cell line stably expressing human C-terminus MUC16 (CA125) transcript variant 4, from amino acid 13785 to 14507 (NM_024690.2). This portion of MUC16 contains several of the extracellular SEA modules, the transmembrane domain, and the short cytoplasmic tail of the protein. Surface expression of MUC16 was confirmed by flow cytometry. This stable clonal cell line was selected based on its high level of MUC16 expression compared to the parental CHO-K1 cell line.

Background

MUC16 (also known as Mucin16, Cell Surface Associated, and ovarian cancer marker CA125) is a protein of the mucin family. Mucins are high molecular weight, O-glycosylated proteins found on the apical surfaces of epithelia. MUC16 has been found in the cornea, the respiratory system, and the female reproductive system, where it plays a role in protecting epithelial cells from pathogen entry. MUC16 dysfunction has been associated with dry eye, cystic fibrosis and cancer, in particular ovarian cancer. MUC16 binds to mesothelin and may be involved in migration and metastasis of tumor cells. Currently MUC16 is used as biomarker for certain cancer types, in addition to being studied in the context of cell signaling. Due to its high expression in some tumors, it is investigated as a therapeutic target for antibody-based therapy as well as adaptive cell therapy.

Application

- Screen and validate antibodies against MUC16 for drug discovery and research.
- Perform binding assays to screen for potential MUC16 ligands.

Materials Provided

Components	Format
2 vials of frozen cells	Each vial contains 2 x 10^6 cells in 1 ml of Cell Freezing Medium (BPS Bioscience #79796)

Parental Cell Line

CHO-K1 cells, Chinese Hamster Ovary, epithelial-like cells, adherent

Mycoplasma Testing

The cell line has been screened to confirm the absence of Mycoplasma species.

Materials Required but Not Supplied

These materials are not supplied with the cell line but are necessary for cell culture and cellular assays. BPS Bioscience's reagents are validated and optimized for use with this cell line and are highly recommended for best results. Media components are provided in the Media Formulations section below.

Media Required for Cell Culture

Name	Ordering Information
Thaw Medium 3	BPS Bioscience #60186
Growth Medium 3J	BPS Bioscience #79974

Storage Conditions

Cells are shipped in dry ice and should immediately be thawed or stored in liquid nitrogen upon receipt. Do not use a -80°C freezer for long term storage. Contact technical support at support@bpsbioscience.com if the cells are not frozen in dry ice upon arrival.

Media Formulations

For best results, the use of validated and optimized media from BPS Bioscience is *highly recommended*. Other preparations or formulations of media may result in suboptimal performance.

Note: Thaw Media do *not* contain selective antibiotics. However, Growth Media *do* contain selective antibiotics, which are used to maintain selective pressure on the cell population expressing the gene of interest.

Cells should be grown at 37℃ with 5% CO_2. BPS Bioscience's cell lines are stable for at least 10 passages when grown under proper conditions.

Media Required for Cell Culture

Thaw Medium 3 (BPS Bioscience #60186):
F-12K medium supplemented with 10% FBS, 1% Penicillin/Streptomycin.

Growth Medium 3J (BPS Bioscience #79974):
F-12K medium supplemented with 10% FBS, 1% Penicillin/Streptomycin, plus 5 µg/ml of Puromycin.

Cell Culture Protocol

Cell Thawing

1. Swirl the vial of frozen cells for approximately 60 seconds in a 37°C water bath. Once cells are thawed (it may be slightly faster or slower than 60 seconds), quickly transfer the entire contents of the vial to a tube containing 10 ml of pre-warmed **Thaw Medium 3**.
   Leaving the cells in the water bath at 37°C for too long will result in rapid loss of viability.

2. Immediately spin down the cells at 300 *x g* for 5 minutes, remove the medium and resuspend the cells in 5 ml of pre-warmed Thaw Medium 3.

3. Transfer the resuspended cells to a T25 flask or T75 flask and incubate at 37°C in a 5% CO_2 incubator.

4. After 24 hours of culture, check for cell attachment and viability. Change medium to fresh Thaw Medium 3 and continue growing in a 5% CO_2 incubator at 37°C until the cells are ready to passage.

5. Cells should be passaged before they are fully confluent. At the first passage and subsequent passages, use **Growth Medium 3J**.

Cell Passage

1. Aspirate the medium, wash the cells with phosphate buffered saline (PBS) without Ca^{2+}/Mg^{2+}, and detach the cells from the culture vessel with 0.25% Trypsin/EDTA.

2. Once the cells have detached, add Growth Medium 3J and transfer the cell suspension to a tube.

3. Spin down cells at 300 *x g* for 5 minutes, remove the medium and resuspend the cells in Growth Medium 3J.

4. Seed into new culture vessels at the recommended sub-cultivation ratio of 1:8 to 1:10 twice or three times per week.

Cell Freezing

1. Aspirate the medium, wash the cells with PBS without Ca^{2+}/Mg^{2+}, and detach the cells from the culture vessel with 0.25% Trypsin/EDTA.

2. Once the cells have detached, add Growth Medium 3J and count the cells.

3. Spin down the cells at 300 *x g* for 5 minutes, remove the medium and resuspend the cells in 4°C Cell Freezing Medium (BPS Bioscience #79796) at ~2 x 10^6 cells/ml.

4. Dispense 1 ml of cell suspension into each cryogenic vial. Place the vials in an insulated container for slow cooling and store at -80°C overnight.

5. Transfer the vials to liquid nitrogen the next day for long term storage.

Note: It is recommended to expand the cells and freeze at least 10 vials at an early passage for future use.

A. Validation Data

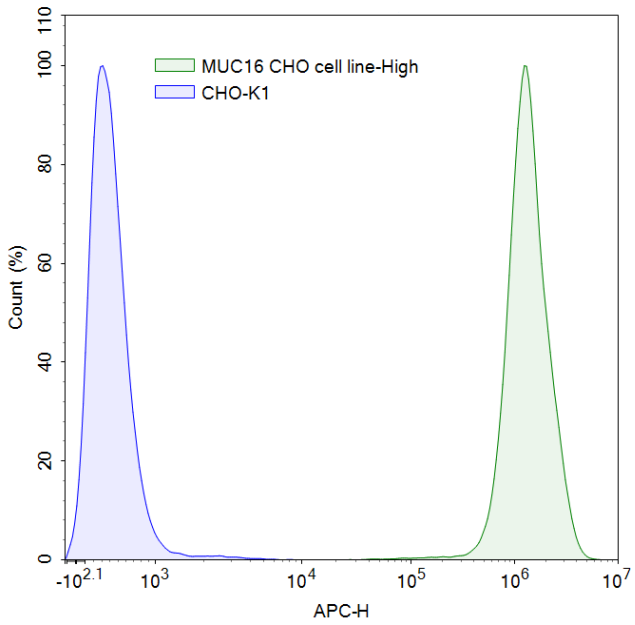

Figure 1: Flow cytometry analysis of MUC16 expression in MUC16 (CA125), variant 4 (region 13785-14507) CHO Cell Line.
MUC16 (CA125), variant 4 (region 13785-14507) CHO cells (green) and control CHO-K1 cells (blue) were stained with anti-MUC16 antibody (Thermofisher #MA1-90039) followed by Alexa Fluor® 647 anti-mouse IgG1 Antibody (Biolegend #406617). Y-axis is the % cell number. X-axis is the intensity of PE.

Sequence

MUC16 (CA125), amino acid 13785 to 14507 of transcript variant 4 (NM_024690.2) cloned into pIRESneo3.

MAVMAPRTLVLLLSGALALTQTWAASHLLILFTLNFTITNLRYEENMWPGSRKFNTTERVLQGLLRPLFKNTSVGPLYSGCRLTLLR
PEKDGEATGVDAICTHRPDPTGPGLDREQLYLELSQLTHSITELGPYTLDRDSLYVNGFTHRSSVPTTSTGVVSEEPFTLNFTINNLR
YMADMGQPGSLKFNITDNVMQHLLSPLFQRSSLGARYTGCRVIALRSVKNGAETRVDLLCTYLQPLSGPGLPIKQVFHELSQQTH
GITRLGPYSLDKDSLYLNGYNEPGPDEPPTTPKPATTFLPPLSEATTAMGYHLKTLTLNFTISNLQYSPDMGKGSATFNSTEGVLQH
LLRPLFQKSSMGPFYLGCQLISLRPEKDGAATGVDTTCTYHPDPVGPGLDIQQLYWELSQLTHGVTQLGFYVLDRDSLFINGYAPQ
NLSIRGEYQINFHIVNWNLSNPDPTSSEYITLLRDIQDKVTTLYKGSQLHDTFRFCLVTNLTMDSVLVTVKALFSSNLDPSLVEQVFL
DKTLNASFHWLGSTYQLVDIHVTEMESSVYQPTSSSSTQHFYLNFTITNLPYSQDKAQPGTTNYQRNKRNIEDALNQLFRNSSIKS
YFSDCQVSTFRSVPNRHHTGVDSLCNFSPLARRVDRVAIYEEFLRMTRNGTQLQNFTLDRSSVLVDGYSPNRNEPLTGNSDLPFW
AVILIGLAGLLGVITCLICGVLVTTRRRKKEGEYNVQQQCPGYYQSHLDLEDLQ

License Disclosure

Visit bpsbioscience.com/license for the label license and other key information about this product.

Troubleshooting Guide

Visit bpsbioscience.com/cell-line-faq for detailed troubleshooting instructions. For further questions, email support@bpsbioscience.com.

Related Products

Products	*Catalog #*	*Size*
MUC1 (CD227), FC-Fusion (IgG1) Avi-tag	100073	25 µg/100 µg
TIM-3, Avi-Tag, Biotin Labeled (Human)	79096	50 µg
TIM-3, Fc fusion (Human)	71151	25 µg/100 µg