Description

CHO cell line stably expressing human Claudin-1 (NM_021101.5). Surface expression of Claudin-1 was confirmed by flow cytometry. The stable clonal cell line was selected for its high levels of Claudin-1 expression compared to the parental CHO-K1 cell line.

Background

Claudins function as major constituents of the tight junction complexes that regulate the permeability of epithelia. While some claudin family members play essential roles in the formation of impermeable barriers, others mediate the permeability to ions and small molecules. They are also involved in cell growth and differentiation. Often, several claudin family members are co-expressed and interact with each other, and this determines the overall cell permeability. Claudin-1 is required to prevent the paracellular diffusion of small molecules through tight junctions in the epidermis and is required for the normal barrier function of the skin. It is involved in normal water homeostasis and prevention excessive water loss through the skin, probably via an indirect effect on the expression levels of other proteins, since Claudin-1 itself seems to be dispensable for water barrier formation in keratinocyte tight junctions. The expression levels and localization of claudins relate to the ability of tumors to invade and to their aggressive characteristics. Claudin 1 is one of the most studied claudins in the context of cancer, but its precise roles are still unclear. Further studies into its roles will provide new cancer therapy options.

Application

- Screen and validate antibodies against Claudin-1 for drug discovery and research.
- Perform cell-based assays, such as screening for potential Claudin-1 ligands.

Materials Provided

Components	Format
2 vials of frozen cells	Each vial contains >1 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°C 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. As soon as the 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 culturing 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 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 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

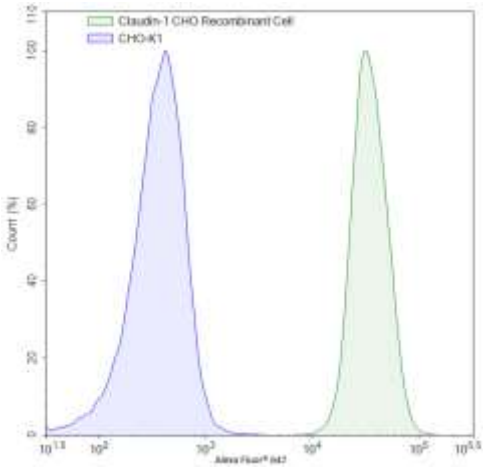

Figure 1: Expression of Claudin-1 in Claudin-1 CHO Cell Line.
Claudin-1 CHO Cell Line (Green) and control CHO-K1 cells (Blue) were stained with Human anti-Claudin-1 Alexa Fluor® 647-conjugated Antibody (R&D Systems #FAB4618R) for 30 min on ice and analyzed by flow cytometry. Y-axis represents the % cell number. X-axis indicates the intensity of Alexa Fluor® 647.

Data shown is representative. For lot-specific information, please contact BPS Bioscience, Inc. at support@bpsbioscience.com.

Sequence

> Claudin-1 (NM_021101.5).

MANAGLQLLGFILAFLGWIGAIVSTALPQWRIYSYAGDNIVTAQAMYEGLWMSCVSQSTGQIQCKVFDSLLNLSSTLQATRALM
VVGILLGVIAIFVATVGMKCMKCLEDDEVQKMRMAVIGGAIFLLAGLAILVATAWYGNRIVQEFYDPMTPVNARYEFGQALFTG
WAAASLCLLGGALLCCSCPRKTTSYPTPRPYPKPAPSSGKDYV

References

Bhat A., *et al.*, 2020, *Int J Mol Sci* 21(2):569.

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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 all further questions, please email support@bpsbioscience.com.

Related Products

Products	*Catalog #*	*Size*
Claudin-18 isoform 1 CHO Cell Line	78361	2 vials
Anti-Claudin-18 Isoform 2 Antibody, PE-Labeled	101676	25 µg/100 µg
Anti-Claudin-18 Isoform 2 Antibody, FITC-Labeled	101866	25 µg/100 µg