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Product Sheet

Membrane bound H_KLK5(AA23-293) HEK-293 Cell Line

Catalog number: GM-C48090

Version 4.0.26.1021

Specifications

Description	Membrane bound H_KLK5(AA23-293) HEK-293 Cell Line is a clonal stable HEK-293 cell line that constitutively expresses the human KLK5 gene, constructed using lentiviral technology.
Quantity	5E6 Cells per vial,1 mL
Product Format	1 vial of frozen cells
Shipping	Shipped on dry ice
Storage Conditions	Liquid nitrogen immediately upon receipt
Target	Human KLK5
Gene ID/Uniprot ID	Q9Y337(AA Val 23 - Ser 293)
Host Cell	HEK-293
Recovery Medium	DMEM+10% FBS+1% P.S
Growth medium	DMEM+10% FBS+1% P.S+0.75 μg/mL Puromycin
Note	None
Freezing Medium	90% FBS+10% DMSO
Growth properties	Adherent
Growth Conditions	37°C, 5% CO_2
Mycoplasma Testing	The cell line has been screened to confirm the absence of Mycoplasma species.
Safety considerations	Biosafety Level 2
Note	It is recommended to expand the cell culture and store a minimum of 10 vials at an early passage for potential future use.

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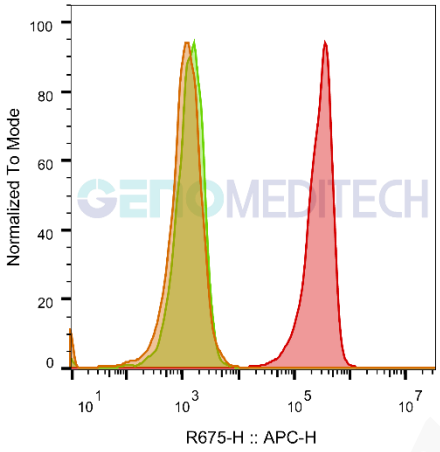
Materials

Reagent	Manufacturer/Catalogue No.
DMEM	Gibco/C11995500BT
Fetal Bovine Serum	ExCell/FSP500
Pen/Strep	Thermo/15140-122
Puromycin	Genomeditech/GM-040401
Anti-KLK5 hIgG1 Antibody (L2Ab2)	Genomeditech/GM-88920AB
Countess 3 Automated Cell Counter	ThermoFisher/A49891
NovoCyte Flow Cytometer	Agilent/NovoCyte 2060R

Data

Expression



	SampleID	Geometric Mean : R675-H
	HEK-293 anti-KLK5+APC-2nd Ab	1050
	HEK-293 H_KLK5 H_IgG+APC-2nd Ab	1289
	HEK-293 H_KLK5 anti-KLK5+APC-2nd Ab	265164

Membrane bound H_KLK5(AA23-293) HEK-293 Cell Line (Cat. GM-C48090) was determined by flow cytometry using Anti-KLK5 hIgG1 Antibody (L2Ab2) (Cat. GM-88920AB).

Cell Recovery

Recovery Medium: DMEM+10% FBS+1% P.S

To insure the highest level of viability, thaw the vial and initiate the culture as soon as possible upon receipt. If upon arrival, continued storage of the frozen culture is necessary, it should be stored in liquid nitrogen vapor phase and not at -70°C. Storage at -70°C will result in loss of viability.

a) Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the O-ring and cap out of the water. Thawing should be rapid (approximately 2 - 3 minutes).
b) Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by dipping in or spraying with 70% ethanol. All of the operations from this point on should be carried out under strict aseptic conditions.
c) Transfer the vial contents to a centrifuge tube containing 5.0 mL complete culture medium and spin at approximately 176 x g for 5 minutes. Discard supernatant.
d) Resuspend cell pellet with the recommended recovery medium. And dispense into appropriate culture dishes.
e) Incubate the culture at 37°C in a suitable incubator. A 5% CO_2 in air atmosphere is recommended if using the medium described on this product sheet.

Cell Freezing

Freezing Medium: 90% FBS+10% DMSO

a) Centrifuge at 176 x g for 3 minutes to collect cells.
b) Resuspend the cells in pre-cooled freezing medium and adjust the cell density to 5E6 cells/mL.
c) Aliquot 1 mL into each vial.
d) Place the vial in a controlled-rate freezing container and store at -80°C for at least 1 day, then transfer to liquid nitrogen as soon as possible.

Cell passage

Growth medium: DMEM+10% FBS+1% P.S+0.75 μg/mL Puromycin

For the first 1 to 2 passages post-resuscitation, use the recovery medium. Once the cells have stabilized, switch to a growth medium.

a) Subculturing is necessary when the cell density reaches 80%. It is recommended to perform subculturing at a ratio of 1:3 to 1:4 every 2-3 days. Ensure that the density does not exceed 80%, as overcrowding can lead to reduced viability due to compression.
b) Remove and discard culture medium.
c) Briefly rinse the cell layer with PBS to remove all traces of serum that contains trypsin inhibitor.
d) Add 1.0 mL of 0.25% (w/v) Trypsin-EDTA solution to dish and observe cells under an inverted microscope until cell layer is dispersed (usually within 30 to 60 seconds at 37°C).
e) Note: To avoid clumping do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach. Cells that are difficult to detach may be placed at 37°C to facilitate dispersal.
f) Add 2.0 mL of growth medium to mix well and aspirate cells by gently pipetting.
g) After centrifugation, resuspend the pellet and add appropriate aliquots of the cell suspension to new culture vessels.
h) Incubate cultures at 37°C.

Subcultivation Ratio: A subcultivation ratio of 1:3 - 1:4 is recommended

Medium Renewal: Every 2 to 3 days

Notes

a) Upon initial thawing, a higher number of dead cells is observed, which is a normal phenomenon. Significant improvement is seen after adaptation. Once the cells reach a stable state, the number of dead cells decreases after subculturing and the cell growth rate becomes stable.

b) Ensure that the cell density does not exceed 80%, as overcrowding may lead to reduced viability due to compression.

Sequence

KLK5 Q9Y337(p.23-293)

VTEHVLANNDVSCDHPSNTVPSGSNQDLGAGAGEDARSDDSSSRIINGSDCDMHTQPWQAALLLRPNQLYCGA
VLVHPQWLLTAAHCRKKVFRVRLGHYSLSPVYESGQQMFQGVKSIPHPGYSHPGHSNDLMLIKLNRRIRPTKDV
RPINVSSHCPSAGTKCLVSGWGTTKSPQVHFPKVLQCLNISVLSQKRCEDAYPRQIDDTMFCAGDKAGRDSCQG
DSGGPVVCNGSLQGLVSWGDYPCARPNRPGVYTNLCKFTKWIQETIQANS

Related Products

KLK5 KLK7	
Membrane bound H_KLK5(AA23-293) CHO-K1 Cell Line	Membrane bound H_KLK7(AA23-253) CHO-K1 Cell Line

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