Product sheet

NCI-H446 Cells | 305049

General information

Description	This cell line was established in 1982 by D. Carney, A.F. Gazdar and associates from the pleural fluid of a patient with small cell cancer of the lung. The original tumor morphology was not characteristic of small cell lung cancer. The cell line is a variant small cell lung cancer in biochemistry and morphology, and expresses neuron specific enolase as well as the brain isoenzyme of creatine kinase. None of L-DOPA decarboxylase, bombesin, vasopressin, oxytocin or gastrin releasing peptide has been detected in the cell line. This cell line exhibits a 20-fold higher degree of c-myc DNA amplification and a 15-fold higher degree of c-myc RNA. The cell line was originally propagated in serum free RPMI 1640 medium supplemented with 10 nM of hydrocortisone, 5 microgram/mL of insulin, 10 microgram/mL of transferrin, 10 nM of 17-beta-estradiol, and 30 nM of sodium selenite. Transplantable tumors with non-typical small cell lung cancer histology can be formed by the cells.
Organism	Human
Tissue	Lung
Disease	Lung small cell carcinoma
Metastatic site	Pleural Effusion
Synonyms	NCI-H446, H-446, NCI-446, NCIH446

Characteristics

Age	61 years
Gender	Male
Ethnicity	European
Morphology	Epithelial-Like
Growth properties	Adherent/suspension

Identifiers / Biosafety / Citation

Citation	H446 (Cytion catalog number 305049)
Biosafety level	1

Expression / Mutation

Tumorigenic	Yes, in nude mice (The cells form transplantable tumors with non-typical SCLC histology).

Handling

Culture Medium	RPMI 1640, w: 2.1 mM stable Glutamine, w: 2.0 g/L NaHCO3 (Cytion article number 820700a)
Medium supplements	Supplement the medium with 10% FBS
Passaging solution	Accutase
Subculturing	Collect suspension cells in a 15 ml tube and carefully rinse the adherent cells using PBS without calcium and magnesium (3-5 ml PBS for T25, 5-10ml for T75 cell culture flasks). Add Accutase (1-2ml per T25, 2.5ml per T75 cell culture flask), the cell sheet must be covered completely. Incubate at ambient temperature for 10 minutes, then centrifuge the cells growing in suspension and the adherent cells together. Carefully resuspend the cells and dispense into new flasks which contain fresh medium.
Split ratio	1: 3 to 1: 4
Fluid renewal	2 to 3 times per week
Freeze medium	CM-1 (Cytion catalog number 800100) or CM-ACF (Cytion catalog number 806100)
Handling of cryopreserved cultures	NCI-H446 cells are shipped in a deep-frozen state on dry ice. Upon receipt, confirm that the vial remains frozen. For storage, place the cryovial immediately at temperatures below -150 degrees. If you plan to culture the cells immediately, swiftly thaw the vial by shaking it in a 37 degrees water bath with clean water and an antimicrobial agent for 40-60 seconds. Remove the vial once a small ice clump persists, ensuring it remains cold. Proceed with all subsequent steps under aseptic conditions. In a sterile flow hood, disinfect the cryovial with 70% ethanol. Then, gently open the vial and transfer the cell suspension into a 15 ml centrifuge tube pre-filled with 8 ml of room temperature culture medium. Gently mix the cells. For cell separation, centrifuge at 300 x g for 3 minutes and dispose of the supernatant. Skipping centrifugation is optional, although any residual freezing medium should be removed after 24 hours. Resuspend the pellet gently in 10 ml of fresh culture medium and divide between two T25 culture flasks. Follow the subculture protocol for subsequent steps.
Handling of proliferating cultures	One or two cell culture flasks come filled with cell culture medium. Collect the entire medium in a 50 ml centrifuge tube. Spin down the collected medium at 300 x g for 3 minutes to collect the cells which may have detached during transit. If a cell pellet is visible, resuspend the cells in 5 ml of cell culture medium and transfer to a T25 cell culture flask. Carefully add 5 ml of cell culture medium to each T25 cell culture flask. Examine cell morphology and confluency using a microscope. Finally, incubate the flasks at 37 degrees Celsius for at least 24 hours.

Quality control / Genetic profile / HLA

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Sterility	Mycoplasma contamination is rigorously excluded using both PCR-based assays and luminescence-based mycoplasma detection methods. To ensure there is no bacterial, fungal, or yeast contamination, cell cultures are subjected to daily visual inspections.

STR profile

Amelogenin: x
CSF1PO: 13
D13S317: 8
D16S539: 12
D5S818: 11
D7S820: 10,11
TH01: 8,9.3
TPOX: 9,11
vWA: 18,19
D3S1358: 17
D21S11: 28
D18S51: 12,13
Penta E: 9,1
Penta D: 12,13
D8S1179: 13,15
FGA: 22
D1S1656: 14,16.3
D6S1043: 11
D2S1338: 18,2
D12S391: 17,18
D19S433: 13,14