



Product Sheet

NCI-H498 [H498] (ATCC®) CCL-254™

Please read this FIRST



Storage Temp.
liquid nitrogen
vapor phase



Biosafety Level
1

Intended Use

This product is intended for research use only. It is not intended for any animal or human therapeutic or diagnostic use.

Complete Growth Medium

The base medium for this cell line is ATCC-formulated RPMI-1640 Medium, Catalog No. 30-2001. To make the complete growth medium, add the following components to the base medium: fetal bovine serum to a final concentration of 10%.

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: NCI-H498 [H498] (ATCC® CCL-254™)

American Type Culture Collection
PO Box 1549
Manassas, VA 20108 USA
www.atcc.org

800.638.6597 or 703.365.2700
Fax: 703.365.2750
Email: Tech@atcc.org

Or contact your local distributor

Description

Organism: *Homo sapiens*, human

Disease: colorectal adenocarcinoma

Age: 56 years

Gender: male

Morphology: epithelial

Growth Properties: suspension, with some loosely adherent cells

Isoenzymes:

AK-1, 1

ES-D, 1

G6PD, B

GLO-I, 2

Me-2, 1

PGM1, 1

PGM3, 1-2

DNA Profile:

Amelogenin: X

CSF1PO: 12

D13S317: 9,11

D16S539: 9,11

D5S818: 13

D7S820: 8,11

THO1: 7

TPOX: 8

vWA: 15

Cytogenetic Analysis: modal number = 51; range = 47 to 54.

This is a hyperdiploid human cell line. The modal chromosome number is 51, occurring in 42% of the cells. The rate of higher ploidy is 5%. The 2q+ is the only marker chromosome detected and common to all cells. There are four copies for N7; 3 copies each for N8, N13 and N22, and a single copy for the X.

Batch-Specific Information

Refer to the Certificate of Analysis for batch-specific test results.

SAFETY PRECAUTION

ATCC highly recommends that protective gloves and clothing always be used and a full face mask always be worn when handling frozen vials. It is important to note that some vials leak when submersed in liquid nitrogen and will slowly fill with liquid nitrogen. Upon thawing, the conversion of the liquid nitrogen back to its gas phase may result in the vessel exploding or blowing off its cap with dangerous force creating flying debris.

Unpacking & Storage Instructions

1. Check all containers for leakage or breakage.
2. Remove the frozen cells from the dry ice packaging and immediately place the cells at a temperature below -130°C, preferably in liquid nitrogen vapor, until ready for use.

Handling Procedure for Frozen Cells

Handling Procedure for Frozen Cells

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.

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1. 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 minutes).
2. 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



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strict aseptic conditions.

3. Transfer the vial contents to a centrifuge tube containing 9.0 ml complete culture medium. and spin at approximately 125 xg for 5 to 7 minutes.
4. Resuspend cell pellet with the recommended complete medium (see the specific batch information for the culture recommended dilution ratio). and dispense into new culture flask. It is important to avoid excessive alkalinity of the medium during recovery of the cells. It is suggested that, prior to the addition of the vial contents, the culture vessel containing the complete growth medium be placed into the incubator for at least 15 minutes to allow the medium to reach its normal pH (7.0 to 7.6).
5. Incubate the culture at **37°C** in a suitable incubator. A 5% CO₂ in air atmosphere is recommended if using the medium described on this product sheet.



Handling Procedure for Flask Cultures

Handling Procedure for Flask Cultures

1. Upon receipt visually examine the culture for macroscopic evidence of any microbial contamination. Using an inverted microscope (preferably equipped with phase-contrast optics), carefully check for any evidence of microbial contamination
2. Incubate the flask in an upright position for several hours at 37°C. After the temperature has equilibrated, aseptically remove the entire contents of the flask and centrifuge at 125 xg for 5 to 10 minutes. Remove shipping medium and save for reuse. Resuspend the cell pellet in 8 ml of this medium.
3. Incubate the culture, horizontally, at 37°C in a 5% CO₂ in air atmosphere. Maintain the cell density of the culture as suggested under the subculture procedure.



Subculturing Procedure

Protocol: Cultures can be maintained by addition or replacement of fresh medium. Alternatively, the cells may be collected by centrifugation and then resuspended at 2 to 4 X 10⁴(4) viable cells/ml. When the cell concentration becomes heavy with large clumps, aspirate well and and dilute approximately 1:10. If the clumps are allowed to get too large, most of the cells will die.

Subcultivation Ratio: A subcultivation ratio of 1:3 to 1:6 is recommended

Medium Renewal: Every 2 to 3 days



Cryopreservation Medium

Cryoprotectant Medium

Complete growth medium described above supplemented with 10% (v/v) DMSO. Cell culture tested DMSO is available as ATCC Catalog No. 4-X.



Comments

This line was derived from a metastasis in the peritoneum obtained from a patient prior to therapy.

The cells form glandular structures and secrete mucin.

The cells are positive for Dopa decarboxylase, CA19-9 and TAG-72 antigens and CEA.

Although the patient from which this cell was derived was blood type A, the cell line does not react with anti type B antibody and has very low to undetectable levels of alpha-N-acetylgalactosaminyltransferase activity.



References

References and other information relating to this product are available online at www.atcc.org.



Biosafety Level: 1

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the current publication of the *Biosafety in Microbiological and Biomedical Laboratories* from the U.S. Department of Health and Human Services Centers for Disease Control and Prevention and National Institutes for Health.

ATCC Warranty


The viability of ATCC® products is warranted for 30 days from the date of shipment, and is valid only if the product is stored and cultured according to the information included on this product information sheet. ATCC lists the media formulation that has been found to be effective for this strain. While other, unspecified media may also produce satisfactory results, a change in media or the absence of an additive from the ATCC recommended media may affect recovery, growth and/or function of this strain. If an alternative medium formulation is used, the ATCC warranty for viability is no longer valid.




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Disclaimers

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Please see the enclosed Material Transfer Agreement (MTA) for further details regarding the use of this product. The MTA is also available on our Web site at www.atcc.org

Additional information on this culture is available on the ATCC web site at www.atcc.org.

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