NCI-H1355 [H1355]

CRL-5865™

Description

NCI-H1355 [H1355] is a cell line that was isolated from the lungs of a 53-year-old, White male with stage 4 adenocarcinoma. This product can be used for cancer research.

Organism: *Homo sapiens*, human
Tissue: Lung
Age: 53 years
Gender: Male
Growth properties: Suspension
Disease: Adenocarcinoma; Stage 4

Storage Conditions

Product format: Frozen
Storage conditions: Vapor phase of liquid nitrogen

Intended Use

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

BSL 1

ATCC determines the biosafety level of a material based on our risk assessment as guided by the current edition of *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, U.S. Department of Health and Human Services. It is your responsibility to understand the hazards associated with the material per your organization's policies

and procedures as well as any other applicable regulations as enforced by your local or national agencies.

ATCC highly recommends that appropriate personal protective equipment is always used when handling vials. For cultures that require storage in liquid nitrogen, it is important to note that some vials may 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 vial exploding or blowing off its cap with dangerous force creating flying debris. Unless necessary, ATCC recommends that these cultures be stored in the vapor phase of liquid nitrogen rather than submersed in liquid nitrogen.

Certificate of Analysis

For batch-specific test results, refer to the applicable certificate of analysis that can be found at www.atcc.org.

Growth Conditions

Temperature: 37°C
Atmosphere: 95% Air, 5% CO_2

Handling Procedures

Unpacking and 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.

Complete medium:

The base medium for this cell line is DMEM:F12 (ATCC 30-2006).To make the complete growth medium, add the following components to 475 mL of the base medium: 25 mL ACL-4 20X Cocktail (see below), 7 mL Bovine Serum Albumin (Sigma A7979); Transferrin, 1 mg/mL stock- use 10 µL/1 mL culture medium (see preparation notes below); EGF, 1 µg/mL stock (see preparation notes below)- use 1 µL/1 mL culture medium.

20 X ACL-4 cocktail is made up of

- 24 mL RPMI
- 40 mL of 4mg/mL Human Insulin
- 40 mL of 5 µM Sodium Selenite
- 8 mL 10 mM Ethanolamine
- 8 mL of 10 mM Phosphorylethanolamine
- 7 uL 30 µM Triiodo-L-thyronine
- 40 mL of 100 mM Sodium Pyruvate
- 80 mL 1 M HEPES
- 80 mL 200 mM L-glutamine
- 80 mL 5 µM Hydrocortisone

To prepare a 1mg/mL **Transferrin** stock solution, aseptically combine:

- 1 vial of Transferrin containing 100 mg (sterile lyophilized) (Sigma T-1147)
- 100 mL serum free medium EMEM (ATCC 30-2003) or RPMI-1640 (ATCC 30-2001)

Sterile filter using 0.22 µm filter

Note: Transferrin is stable at 2-8°C for up to 7 days. Do not use Transferrin which has been at 2-8°C for more than 7 days.

To prepare a 1 µg/mL **EGF** stock solution, aseptically combine:

- 1 vial of 10 µg Gibco™ EGF (ThermoFisher Scientific PHG0314)
- 10 mL PBS with 0.1% BSA (150 mL Ca/Mg free PBS + 0.15 g BSA) or equivalent

To prepare 0.1% **BSA** solution

- 150 mL Ca/Mg free PBS (ATCC 30-2200)

- 0.15 g BSA (Sigma A8022)

Note: Aliquot in working volumes and store at -20°C. Avoid freeze/thaw cycles. EGF is stable for 1 year after reconstitution when prepared and stored as indicated above.

Note: Once prepared, the above ACL-4 basal media is stable when stored at 2-8°C for up to 1 month.

Because of limited stability, the following components should be added to an aliquot of the above culture medium fresh prior to seeding or performing fluid additions. Complete medium with the below components can be stored at 2.0 to 8.0°C and expires in 7 days.

Handling Procedure:

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.

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 strict aseptic conditions.
3. Transfer the vial contents to a centrifuge tube containing 9.0 mL complete culture medium. and spin at approximately 125 x g 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 a **25 cm^2** 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% CO_2 in air atmosphere is recommended if using the medium described on this product sheet.

Subculturing procedure:
Cultures can be maintained by the addition of fresh medium or by replacement of medium. Alternatively, cultures can be established by centrifugation and subsequent resuspension of the pellet. The cells grow in floating aggregates of round cell clusters.
Medium Renewal: twice weekly
Note: Add fresh medium as the cell density increases.
Reagents for cryopreservation: Complete growth medium supplemented with 10% (v/v) FBS and 5% (v/v) DMSO (ATCC 4-X)

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: NCI-H1355 [H1355] (ATCC CRL-5865)

References

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

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