Organism: Homo sapiens, human
Tissue: liver
Disease: hepatocellular carcinoma
Age: 42 years
Gender: male
Morphology: epithelial
Growth Properties: mixed adherent and floating
Virus Susceptibility:
Viral Testing: ATCC confirmed this cell line is positive for the presence of HBV viral DNA sequences via PCR.
DNA Profile:
- Amelogenin: X,Y
- CSF1PO: 13
- D13S317: 11
- D16S539: 10,14
- D5S818: 12
- D7S820: 10,11
- THO1: 7,9
- TPOX: 11
- vWA: 17,18
Cytogenetic Analysis: aneuploid; modal number = 62

SAFETY PRECAUTION

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.

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 cm² or a 75 cm² 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

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 Growth Medium

RPMI 1640 medium, 90%; heat-inactivated fetal bovine serum, 10%

Unpacking & Storage Instructions

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

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: SNU-398 (ATCC® CRL-2233™)

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
The flask was seeded with cells (see specific batch information), grown, and completely filled with medium at ATCC to prevent loss of cells during shipping.

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. Also, check to determine if the majority of cells are still attached to the bottom of the flask; during shipping the cultures are sometimes handled roughly and many of the cells often detach and become suspended in the culture medium (but are still viable).

2. If the cells are still attached, aseptically remove all but 5 to 10 mL of the shipping medium. The shipping medium can be saved for reuse. Incubate the cells at 37°C in a 5% CO₂ in air atmosphere until they are ready to be subcultured.

3. If the cells are not attached, aseptically remove the entire contents of the flask and centrifuge at 125 x g for 5 to 10 minutes. Remove shipping medium and save. Resuspend the pelleted cells in 10 mL of this medium and add to 25 cm² flask. Incubate at 37°C in a 5% CO₂ in air atmosphere until cells are ready to be subcultured.

**Subculturing Procedure**

Volumes used in this protocol are for 75 cm² flask; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes.

1. This is a cell line that grows as both attached and suspended cells. The suspended cells are viable and should not be discarded. Recover the suspended cells by gentle centrifugation (125 x g) for subculture along with the adherent cell population.

2. To dissociate adherent cells, briefly rinse the cell layer with 0.25% (w/v) Trypsin-0.53 mM EDTA solution to remove all traces of serum that contains trypsin inhibitor.

3. Add 2.0 to 3.0 mL of Trypsin-EDTA solution to flask and observe cells under an inverted microscope until cell layer is dispersed (usually within 5 to 15 minutes).

   **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.

4. Add 6.0 to 8.0 mL of complete growth medium and aspirate cells by gently pipetting.

5. Add appropriate aliquots of the cell suspension to new culture vessels.

6. Incubate cultures at 37°C

**Subculture Ratio:** 1:3 to 1:6

**Medium Renewal:** 2 to 3 times a week.

**Note:** For more information on enzymatic dissociation and subculturing of cell lines consult Chapter 10 in Culture of Animal Cells, a manual of Basic Technique by R. Ian Freshney, 3rd edition, published by Alan R. Liss, N.Y., 1994.

**Cryopreservation Medium**

Complete culture medium described above supplemented with 5% (v/v) DMSO.

Cell culture tested DMSO is available as ATCC Catalog No. 4-X.

**Comments**

Grossly, the original tumor was single nodular with perinodular extensions. Histologically, it was trabecular type.

The cultured cells are multinucleated, and maintain the production of intracytoplasmic hyaline globules as seen in the original tumor.

Hepatitis B virus (HBV) DNA was detected by Southern blot hybridization.

HBV genomic RNA was not expressed.

**References**

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

**Biosafety Level: 2**

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

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