SK-BR-3 [SKBR3]

HTB-30™

Description

SK-BR-3 [SKBR3] cells were isolated in 1970 from pleural effusion cells of a 43-year-old, White, female adenocarcinoma patient with blood type A+ who had been treated with radiation, steroids, cytoxan, and 5-fluorouracil. Use these cells in your breast cancer research.

Organism: *Homo sapiens*, human
Tissue: Breast; Mammary gland
Age: 43 years
Gender: Female
Morphology: epithelial
Growth properties: Adherent
Disease: Adenocarcinoma
Technical information: ATCC Product Experience does not have technical information on patent deposits that are not produced or characterized by ATCC. Additional information can be found in the corresponding patent available from the patent holder or with the U.S. and/or international patent office.

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 ATCC-formulated McCoy's 5a Medium Modified (ATCC 30-2007). To make the complete growth medium, add the following components to the base medium: fetal bovine serum (ATCC 30-2020) to a final concentration of 10%.

Example media preparation:

- 500 mL Mccoy's 54 (ATCC 30-2007)
- 56 mL FBS non-heat inactivated (ATCC 30-2020)

Handling Procedure:

To ensure 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 150 to 400 x *g* for 8 to 12 minutes (280 x *g* for 10 minutes).
4. Resuspend cell pellet with the recommended complete growth medium (see the lot information on Certificate of Analysis (COA) for the culture recommended dilution ratio) and dispense into a 25 cm^2 or a 75 cm^2 culture flask as recommended on the COA. The recommended seeding density for HTB-30 is 2.0 x 10^4 to 5.0 x 10^4 viable cells/cm^2. 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.

Additional notes for culturing

These cells typically recover very slowly from cryopreservation. SK-BR-3 (ATCC HTB-30) cells tend to pile on each other.

In the first week of growth, leave the HTB-30 cells undisturbed for a few days after initiation.

If there are still a lot of floaters, check viability with trypan blue. It is important to retain the floating cells by gently centrifugation and add them back to the same flask with the attached cells when replacing the medium.

Subculturing procedure:

Volumes are given for a 75 cm^2 flask. Increase or decrease the amount of dissociation medium needed proportionally for culture vessels of other sizes. Corning® T-75 flasks (catalog #430641) are recommended for subculturing this product.

Note: Do not trypsinize cells until they have reached 70-80% confluence. If cells are allowed to overgrow cells will detach.

1. Remove and discard culture medium.
2. Briefly rinse the cell layer with D-PBS (ATCC 30-2200) to remove all traces of serum which contains trypsin inhibitor.
3. Add 2.0 to 3.0 mL of Trypsin-EDTA solution (ATCC 30-2101) 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.

Subcultivation Ratio/Subculture seeding density: A subcultivation ratio of 1:2 is recommended or subculture at a seeding density of 1.5 x 10^4 to 4.0 x 10^4 viable cells/cm^2.

Note: Unless floating cells have been verified to be non-viable via a trypan blue or some other sort of viability assay, floating cells should be kept with the main culture.

Medium Renewal: 2 to 3 times per week

Culture maintenance:

HTB-30 can maintain epithelial-like and/or rounded cell morphology. Along with this, HTB-30 is historically known to generate heavy debris. Neither of these characteristics indicate the contamination or quality issue when observed in HTB-30.

Reagents for cryopreservation:

Complete growth medium supplemented with 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: SK-BR-3 [SKBR3] (ATCC HTB-30)

References

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

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Contact Information

ATCC
10801 University Boulevard
Manassas, VA 20110-2209
USA
US telephone: 800-638-6597
Worldwide telephone: +1-703-365-2700
Email: tech@atcc.org or contact your local distributor