ATCC

Product Sheet

MDA-MB-435S (ATCC® HTB-129™)

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 Leibovitz's L-15 Medium, Catalog No. 30-2008. To make the complete growth medium, add the following components to the base medium:

- 0.01mg/ml bovine insulin
- 0.01mg/ml glutathione
- 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: MDA-MB-435S (ATCC® HTB-129™)

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
Tissue: previously described as: mammary gland/breast; derived from metastatic site: pleural effusion
Disease: previously described as ductal carcinoma
Cell Type: melanocyte,Melanoma
Age: 31 years
Gender: female
Morphology: spindle shaped
Growth Properties: adherent
Isoenzymes:
AK-1, 1
ES-D, 1
G6PD, B
GLO-I, 2
PGM1, 2
PGM3, 1
DNA Profile:
Amelogenin: X
CSF1PO: 11
D13S317: 12
D16S539: 13
D5S818: 12
D7S820: 8,10
THO1: 6,7
TPOX: 8,11
vWA: 16,18
Cytogenetic Analysis: modal number = 56; range = 55 to 62
The cell line is aneuploid human female (XX), with most chromosome counts in the 55 to 60 range. Normal chromosomes N6, N11, and N22 were absent, while chromosomes N7, N13, N18 and N21 were single. Most of the remainder of normal chromosomes were usually paired, but chromosome N2 was triple. Nineteen marker chromosomes were identified, with most of them formed from structural alterations of the missing copies of the normal chromosomes. Six of these markers involve regions of chromosome N7, while three are recognized as derivatives of chromosome N6. Regions of a third copy of the normal and paired chromosomes N3, N15, N17, N20 are noted in markers M1, M2, M15, and M5, respectively.

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

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

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approximately 125 x g for 5 to7 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 or a 75 cm^2 culture flask.
5. Incubate the culture at **37°C** in a suitable incubator.

The L-15 medium formulation was devised for use in a free gas exchange with atmospheric air. A CO_2 and air mixture is detrimental to cells when using this medium for cultivation

Handling Procedure for Flask Cultures

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 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 cm^2 flask. Incubate at 37°C in air atmosphere until cells are ready to be subcultured.

Subculturing Procedure

Remove medium, add fresh 0.25% trypsin - 0.53 mM EDTA, rinse and remove. Place flask at room temperature (or incubated at 37°C) for approximately 10 minutes or until the cells detach. Add fresh medium, aspirate and dispense into new flasks.

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

Medium Renewal: 2 to 3 times per week

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

This cell line was originally described as a spindle shaped variant of the parental MDA-MB-435 strain isolated in 1976 by R. Cailleau, et al. from the pleural effusion of a 31 year old female with metastatic, ductal adenocarcinoma of the breast.

However, recent studies have generated questions about the origin of the parent cell line, MDA-MB-435, and by extension HTB-129. Gene expression analysis of the cells produced microarrays in which MDA-MB-435 clustered with cell lines of melanoma origin instead of breast [PubMed ID: 10700174, PubMed ID: 15150101, PubMed ID: 15679052].

Additional studies have since corroborated a melanocyte origin of MDA-MB-435, to which ATCC has responded by pursuing its own investigation into the identity of this cell line. The cell line to which MDA-MB-435 is reported to have been cross-contaminated with is the M14 melanoma line [PubMed ID: 12354931 and PubMed ID: 17004106].

Derivatives of HTB-129 with identities in question:

M4A4, ATCC® CRL-2914
M4A4 GFP, ATCC® CRL-2915
M4A4 LM3-2 GFP, ATCC® CRL-2916
M4A4 LM3-4 CL 16 GFP, ATCC® CRL-2917
NM2C5, ATCC® CRL-2918
NM2C5 GFP, ATCC® CRL-2919

References

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

Biosafety Level: 1

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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.

Disclaimers

This product is intended for laboratory research purposes only. It is not intended for use in humans.
While ATCC uses reasonable efforts to include accurate and up-to-date information on this product sheet, ATCC makes no warranties or representations as to its accuracy. Citations from scientific literature and patents are provided for informational purposes only. ATCC does not warrant that such information has been confirmed to be accurate.
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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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