ATCC

Product Sheet

ASC52telo, hTERT immortalized adipose derived Mesenchymal stem cells (ATCC® SCRC-4000™)

Please read this FIRST

Storage Temp.
liquid nitrogen vapor phase

Biosafety Level
2

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 Mesenchymal Stem Cell Basal Medium (PCS-500-030). To make the complete growth medium, add Mesenchymal Stem Cell Growth Kit (PCS-500-040) for Adipose and Umbilical-derived MSCs - Low Serum Components and G418 to the base medium as the following:
485 mL of basal medium (PCS-500-030)
10 mL of MSC supplement (2% FBS, 5 ng/mL rh FGF basic, 5 ng/mL rh FGF acidic, 5 ng/mL rh EGF)
6 mL of L-Alanyl-L-Glutamine (2.4 mM, final concentration)
1 mL of 100 mg/mL G418 (0.2 mg/mL, final concentration)

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: ASC52telo, hTERT immortalized adipose derived Mesenchymal stem cells (ATCC® SCRC-4000™)

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
Immortalization Method:
hTERT expression
Tissue: adipose tissue
Cell Type: Mesenchymal stem cells immortalized with hTERT
Gender: female
Morphology: fibroblast-like
Growth Properties: adherent
DNA Profile:
Amelogenin: X
CSF1PO: 10, 13
D13S317: 8, 12
D16S539: 10, 13
D5S818: 11, 13
D7S820: 8, 11
THO1: 7
TPOX: 8
vWA: 16

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

Recovery of Frozen Cells
To ensure the highest level of viability, thaw the vial and initiate the culture as soon as possible upon receipt. If storage of the frozen culture is necessary upon arrival, store the vial in liquid nitrogen vapor phase and **NOT** at
-70°C. Storage at -70°C will result in loss of viability.
SAFETY PRECAUTION: Always use protective gloves and clothing and wear a full face mask when handling frozen vials. Some vials leak when immersed 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.

1. Prepare a 25-cm^2 or a 75-cm^2 culture flask containing the recommended complete culture medium. Prior to the addition of the vial contents, the vessel containing the growth medium should be placed in the incubator for at least 15 minutes to allow the medium to reach its normal pH (7.0 to 7.6) and to avoid excessive alkalinity of the medium during recovery of the cells.
2. 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).
3. 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 operations from this point on should be carried out under strict aseptic conditions.
4. Transfer the vial contents to a centrifuge tube containing 9.0 ml of complete culture medium and centrifuge the cell suspension at approximately 125 xg for 5 to 7 minutes.
5. Discard the supernatant and resuspend the cells in fresh growth medium (see the batch-specific information for the recommended dilution ratio). Add this suspension to the prepared culture vessel.
6. Incubate the culture at 37°C in a suitable incubator.
7. A 5% CO_2/95% air atmosphere is recommended if using the medium described on this product sheet.

ATCC

Product Sheet

ASC52telo, hTERT immortalized adipose derived Mesenchymal stem cells (ATCC® SCRC-4000™)

Please read this FIRST

Storage Temp.
liquid nitrogen vapor phase

Biosafety Level
2

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 Mesenchymal Stem Cell Basal Medium (PCS-500-030). To make the complete growth medium, add Mesenchymal Stem Cell Growth Kit (PCS-500-040) for Adipose and Umbilical-derived MSCs - Low Serum Components and G418 to the base medium as the following:
485 mL of basal medium (PCS-500-030)
10 mL of MSC supplement (2% FBS, 5 ng/mL rh FGF basic, 5 ng/mL rh FGF acidic, 5 ng/mL rh EGF)
6 mL of L-Alanyl-L-Glutamine (2.4 mM, final concentration)
1 mL of 100 mg/mL G418 (0.2 mg/mL, final concentration)

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: ASC52telo, hTERT immortalized adipose derived Mesenchymal stem cells (ATCC® SCRC-4000™)

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

Handling Procedure for Flask Cultures

Receiving Flask Cultures
The flask was seeded with cells, incubated, and completely filled with medium at ATCC to prevent loss of cells during shipping.
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).

1. **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% CO_2 in air atmosphere until they are ready to be subcultured.
2. **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 a 5% CO_2 in air atmosphere until cells are ready to be subcultured.

Population Doubling Capacity

Longevity: >25 PDLs post-thaw

Subculturing Procedure

Protocol:
1. Passage immortalized adipose-derived MSCs when the culture has reached approximately 80% confluence.
2. Warm both the Trypsin-EDTA for Primary Cells (ATCC PCS-999-003) and the Trypsin Neutralizing Solution (ATCC PCS-999-004) to room temperature prior to dissociation. Warm the complete growth medium to 37°C prior to use with the cells.
3. For each flask, carefully aspirate the spent media without disturbing the monolayer.
4. Rinse the cell layer one time with 3 to 5 mL D-PBS (ATCC 30-2200) to remove residual medium.
5. Add prewarmed trypsin-EDTA solution (1 to 2 mL for every 25 cm^2) to each flask.
6. Gently rock each flask to ensure complete coverage of the trypsin-EDTA solution over the cells, and then aspirate the excess fluid off of the monolayer.
7. Observe the cells under the microscope. When the cells pull away from each other and round up (typically within 3 to 5 minutes), remove the flask from the microscope and gently tap it from several sides to promote detachment of the cells from the flask surface.
8. When the majority of cells appear to have detached, quickly add an equal volume of the Trypsin Neutralizing Solution (ATCC PCS-999-004) to each flask. Gently pipette or swirl the culture to ensure all of the trypsin-EDTA solution has been neutralized.
9. Transfer the dissociated cells to a sterile centrifuge tube and set aside while processing any remaining cells in the culture flask.
10. Add 3 to 5 mL D-PBS (ATCC 30-2200) to the tissue culture flask to collect any additional cells that might have been left behind.
11. Transfer the cell/D-PBS suspension to the centrifuge tube containing the trypsin-EDTA-dissociated cells.
12. Repeat steps 10 and 11 as needed until all cells have been collected from the flask.
13. Centrifuge the cells at 270 x g for 5 minutes.
14. Aspirate neutralized dissociation solution from the cell pellet and resuspend the cells in 2 to 8 mL fresh, prewarmed, complete growth medium.
15. Count the cells and seed new culture flasks at a density of 5,000 viable cells per cm^2.
16. Place newly seeded flasks in a 37°C, 5% CO2 incubator for at least 24 to 48 hours before processing the cells further.

Cell seeding density: 5,000 viable cells per cm^2
Medium renewal: every 2 to 3 days

Cryopreservation Medium

ATCC

Product Sheet

ASC52telo, hTERT immortalized adipose derived Mesenchymal stem cells (ATCC® SCRC-4000™)

Please read this FIRST

Storage Temp.
liquid nitrogen vapor phase

Biosafety Level
2

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 Mesenchymal Stem Cell Basal Medium (PCS-500-030). To make the complete growth medium, add Mesenchymal Stem Cell Growth Kit (PCS-500-040) for Adipose and Umbilical-derived MSCs - Low Serum Components and G418 to the base medium as the following:
485 mL of basal medium (PCS-500-030)
10 mL of MSC supplement (2% FBS, 5 ng/mL rh FGF basic, 5 ng/mL rh FGF acidic, 5 ng/mL rh EGF)
6 mL of L-Alanyl-L-Glutamine (2.4 mM, final concentration)
1 mL of 100 mg/mL G418 (0.2 mg/mL, final concentration)

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: ASC52telo, hTERT immortalized adipose derived Mesenchymal stem cells (ATCC® SCRC-4000™)

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

Cryopreservation Medium
DMEM: F12 Medium, 80%; FBS, 10%; DMSO, 10%
Store in liquid nitrogen vapor. Avoid immersing vials into liquid nitrogen.

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.

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.
This product is sent with the condition that you are responsible for its safe storage, handling, and use. ATCC is not liable for any damages or injuries arising from receipt and/or use of this product. While reasonable effort is made to insure authenticity and reliability of strains on deposit, ATCC is not liable for damages arising from the misidentification or misrepresentation of cultures.
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.
© ATCC 2014. All rights reserved. ATCC is a registered trademark of the American Type Culture Collection. [06/25]