ATCC

Product Sheet

WPE-stem (ATCC® CRL-2887™)

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 provided by Invitrogen (GIBCO) as part of a kit: Keratinocyte Serum Free Medium (K-SFM), Kit Catalog Number 17005-042. This kit is supplied with each of the two additives required to grow this cell line (bovine pituitary extract (BPE) and human recombinant epidermal growth factor (EGF). To make the complete growth medium, you will need to add the following components to the base medium:

- 0.05 mg/ml BPE - provided with the K-SFM kit
- 5 ng/ml EGF - provided with the K-SFM kit. NOTE: Do not filter complete medium.

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: WPE-stem (ATCC® CRL-2887™)

Description

Organism: *Homo sapiens*, human
Tissue: prostate, normal, peripheral zone
Disease: Papilloma
Cell Type: epithelial
Age: 54
Gender: male
Morphology: epithelial
Growth Properties: loosely adherent
DNA Profile:
Amelogenin: X,Y
CSF1PO: 13
D13S317: 8,14
D16S539: 9,11
D5S818: 12,15
D7S820: 10,11
THO1: 8
TPOX: 8,11
vWA: 14,18

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.

Note: Culture flasks should be **pre-coated** with a mixture of Mouse Collagen Type IV and Human Fibronectin (Sigma Cat. No. F-0895) 2.5µg each/ cm^2.

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 pre-coated 25 cm^2 or a 75 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 7.4).
5. Incubate the culture at 37°C in a suitable, humidified incubator. A 5% CO_2 in air atmosphere is recommended if using the medium described on this product.

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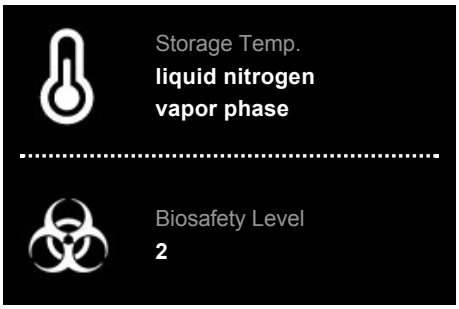

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Handling Procedure for Flask Cultures

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 mL of the shipping medium from a 25 cm^2 flask. 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.
3. **If the cells are not attached,** aseptically remove the entire contents of the flask and centrifuge at 125 x g for 5 to 7 minutes. Remove shipping medium and save. Resuspend the pelleted cells in 5 mL of this medium and add to same coated 25 cm^2 flask. Incubate at 37°C in a 5% CO_2 in air atmosphere until cells are ready to be subcultured.

Subculturing Procedure

Volumes used in this protocol are for 75 cm^2 flasks; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes. Subculture cells before they reach confluence. Do not allow cells to become confluent.

Note: Culture flasks should be pre-coated with a mixture of Mouse Collagen Type IV and Human Fibronectin (Sigma Cat. No. F-0895) 2.5 µg each/cm^2.

1. Collect spent medium containing viable, floating, single cells, and prostaspheres.
2. Briefly rinse the cell layer with Ca++/Mg++ free Dulbecco's phosphate-buffered saline (D-PBS).
3. 3.0 to 4.0 mL of 0.05% Trypsin - 0.53 mM EDTA solution, diluted 1:1 with D-PBS, and place flask in a 37°C incubator for 3 to 5 minutes. Observe cells under an inverted microscope until cell layer is dispersed (usually within 5 to 8 minutes).
   Note: To avoid clumping do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach.
4. Add 6.0 to 8.0 mL of 0.1% Soybean Trypsin Inhibitor, or 2% fetal bovine serum in D-PBS, as appropriate, add the spent medium containing floating cells, and aspirate cells by gently pipetting.
5. Transfer cell suspension to centrifuge tube and spin at approximately 125 x g for 5 to 7 minutes.
6. Discard supernatant and resuspend cells in fresh serum-free growth medium. Add appropriate aliquots of cell suspension to new pre-coated culture flasks. An inoculum of 2 X 10^3 to 4 X 10^3 viable cells/cm^2 is recommended.
7. Place culture vessels in incubators at 37°C. Cells grown under serum-free or reduced serum conditions may not attach strongly during the 24 hours after subculture and should be disturbed as little as possible during that period.

Interval: Subculture cells before they reach confluence. Do not allow cells to become confluent. Subculture when cell concentration is between 1 X 10^5 and 1.5 X 10^5 cells/cm^2.
Subcultivation Ratio: A subcultivation ratio of 1:3 to 1:5 is recommended
Medium Renewal: Every 48 hours

Cryopreservation Medium

Complete growth medium described above supplemented with 15% fetal bovine serum and 10% DMSO.
Cell culture-tested DMSO is available as ATCC® Catalog No. 4-X.

Comments

WPE-stem cells are loosely adherent and exhibit features characteristic of stem/progenitor cells present in the embryonic urogenital sinus and in adult prostatic epithelium, including p63 and ABCG2.
The cells show high expression of cytokeratin 5 and 14 and MMP-2 but low expression of cytokeratin 18.
They are androgen-independent for growth and survival.
Cells grow in soft agar with a cloning efficiency of 0.9%.
Cells are not tumorigenic in nude mice even at 6 months after injection.
WPE-stem cells were screened for CMV, HBV, HCV, HTLV 1, HTLV 2, HIV 1, HIV 2, JCV, and MoMuLV DNA sequences. Cells were also tested for 25 species of mycoplasma and *Acholeplasma laidlawii* . Cells tested negative for all of the above. (personal communication from depositor).

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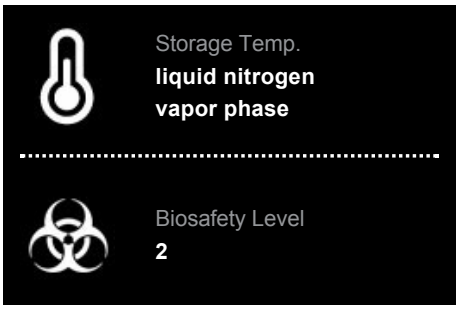

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References

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