ATCC

Product Sheet

PA317 containing JR-gal (ATCC® CRL-9995™)

Please read this FIRST

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 Dulbecco's Modified Eagle's Medium, Catalog No. 30-2002. To make the complete growth medium, add the following components to the base medium: 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: PA317 containing JR-gal (ATCC® CRL-9995™)

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: *Mus musculus*, mouse
Strain: NIH/Swiss
Tissue: embryo
Cell Type: fibroblast
Age: embryo
Morphology: fibroblast
Growth Properties: adherent

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

HANDLING PROCEDURE FOR FROZEN CELLS

- Initiate culture as soon as possible upon receipt.

- Thaw by rapid agitation in 37°C water bath. Thawing should be rapid (within 40-60 seconds). As soon as the ice is melted, remove the ampule from the water bath. All of the operations from this point on should be carried out under strict aseptic conditions.

- Transfer the cell suspension and dilute it with the recommended culture medium in a culture flask (see specific batch information above for dilution ratio); incubate at 37°C with 5% CO_2 in air atmosphere. Since it is important to avoid excessive alkalinity of the medium during recovery of the cells, it is suggested that the culture medium be placed into the culture flask, tube, etc. and the pH be adjusted, as necessary, prior to the addition of the vial contents. Note that the bicarbonate content of the culture medium will determine whether an atmosphere containing CO_2 will be required.

- It is not necessary to remove the freezing additive. However, if desired, the culture medium may be changed to remove the protective freezing additive (dimethylsulfoxide) 24 hours after thawing. If it is desired that the freezing additive be removed immediately, or that a more concentrated cell suspension be obtained, centrifuge the above diluted suspension at approximately 125 x g for 10 minutes, discard the fluid and resuspend the cells with growth medium at the dilution ratio given in the specific batch information above.

FLUID RENEWAL

2-3 times weekly.

SUBCULTURE PROCEDURE
Remove medium, rinse with trypsin (0.25%) - EDTA (0.03%) solution. Add 1-2 ml additional trypsin solution and allow flasks to remain at room temperature (or incubate at 37°C) until cells detach. Add fresh culture medium, aspirate and dispense into new flasks.

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Subcultivation ratio: 1:8 to 1:12 is recommended.

Handling Procedure for Flask Cultures

HANDLING PROCEDURE FOR FLASK CULTURES (MONOLAYER)
The flask was seeded with cells (see specific batch information above for concentration), grown and completely filled with medium to prevent loss of cells in transit. Aseptically remove all of the medium (which can be saved and used as fresh medium) except for a sufficient volume to cover the floor of the flask. Incubate the culture in a flat position at 37°C in a 5% CO_2 in air atmosphere. The shipping medium contains reduced sodium bicarbonate suitable for a 5% CO_2 in air incubator. DMEM usually contains 3.7 grams of sodium bicarbonate per liter and should be incubated in a 10% CO_2 in air incubator. Sometimes in transit the cultures are handled roughly and most of the cells become detached and float in the culture medium. If this has occurred remove the entire contents of the flask and centrifuge at 300 x g for 15 minutes. Draw off the excess supernatant medium, resuspend the cells in 10-12 ml of the shipping medium and plant the entire cell suspension in a single flask of suitable size (about 25 sq. cm.).

Subculturing Procedure

Subcultivation Ratio: A subcultivation ratio of 1:10 to 1:20 is recommended
Medium Renewal: 2 to 3 times per week
Remove medium, rinse flask with fresh 0.25% trypsin, 0.02% EDTA and allow the flask to sit at room temperature (or 37C) until the cells detach. Add fresh medium, aspirate and dispense into new flasks.

Comments

This cell line was derived from PA317 (see ATCC CRL-9098) by introduction of a retrovirus vector (JR-gal). The vector is derived from the JR vector by incorporation of the E. coli beta galactosidase gene (PstI to SalI fragment of pBAG gag - beta gal fusion protein) into the BamH1 site of JR.

References

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

Biosafety Level: 1

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

ATCC® products are warranted for 30 days from the date of shipment, and this warranty is valid only if the product is stored and handled according to the information included on this product information sheet. If the ATCC® product is a living cell or microorganism, ATCC lists the media formulation that has been found to be effective for this product. 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 product. 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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the misidentification or misrepresentation of such materials.
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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