ATCC

Product Sheet

Crithidia luciliae (ATCC® 30258™)

Please read this FIRST

Storage Temp.

Frozen Cultures:
-70°C for 1 week; liquid N_2 vapor for long term storage

Freeze-dried Cultures:
2-8°C

Live Cultures:
See Protocols section for handling information

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.

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: *Crithidia luciliae* (ATCC® 30258™)

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

Strain Designation: [ATCC 14765]
Deposited Name: *Crithidia luciliae* (Strickland) Wallace and Clark
Depositor: FG Wallace, T Clark
Isolation: Green bottle fly, *Phaenicia sericata*, Minneapolis, MN, 1958

Propagation

Growth Conditions
Temperature: 25°C
Culture System: Axenic

Medium
ATCC® Medium 1011: Diphasic blood agar medium
ATCC® Medium 355: Crithidia medium
ATCC® Medium 1034: Modified PYNFH medium (Available from ATCC as ATCC cat. no. 327-X)

Instructions for Complete Medium
ATCC Medium 355
(ATCC medium 1034 can also be used for cultivation and is available in a freeze-dried format from ATCC; contact sales for details.)

Protocols

Storage and Culture Initiation
Frozen ampules packed in dry ice should either be thawed immediately or stored in liquid nitrogen. If liquid nitrogen storage facilities are not available, frozen ampoules may be stored at or below -70°C for approximately one week. **Do not under any circumstance store frozen ampules at refrigerator freezer temperatures (generally -20°C).** Storage of frozen material at this temperature will result in the death of the culture.

1. To thaw a frozen ampule, place it in a 35°C water bath such that the lip of the ampule remains above the water line. Thawing time is approximately 2 to 3 minutes. Do not agitate the ampule. Do not leave ampule in water bath after thawed.
2. Immediately after thawing, aseptically transfer contents to a 16 x 125 mm screw-capped test tube containing 5 mL ATCC Medium 355.
3. Incubate upright at 25°C with caps screwed on tightly.

Culture Maintenance

1. When the culture is at or near peak density, vigorously agitate the culture.
2. Transfer approximately 0.10 mL to a fresh tube containing 5 mL of fresh ATCC medium 355.
3. Incubate upright at 25°C with caps screwed on tightly.
4. Transfer every 14 days.

Cryopreservation

Harvest and Preservation

1. Prepare a 10% (v/v) sterile DMSO solution in fresh ATCC Medium 355.
2. Transfer a culture at peak density to centrifuge tubes and centrifuge at 525 x g for 5 minutes.
3. Remove the supernatant and resuspend the cells in ATCC medium 355 to a concentration of 2 x 10^6 to 2 x 10^7 cells/mL.
4. Mix the cell preparation and the DMSO in equal portions. Thus, the final concentration will be between 10^6 and 10^7 cells/mL and 5% (v/v) DMSO.
5. Distribute the cell suspension in 0.5 mL aliquots into 1.0 mL to 2.0 mL sterile plastic screw-capped cryules (special plastic vials for cryopreservation). The time from the mixing of the cell preparation and DMSO stock solution before the freezing process is begun should be no less than 15 min and no longer than 30 min.
6. Place the vials in a controlled rate freezing unit. From room temperature cool at -1°C/min to -40°C. If the freezing unit can compensate for the heat of fusion, maintain rate at -1°C/min through the heat of fusion. At -40°C plunge into liquid nitrogen. Alternatively, place the vials in a Nalgene 1°C freezing apparatus. Place the apparatus at -80°C for 1.5 to 2 hours and then plunge ampules into liquid

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nitrogen. (The cooling rate in this apparatus is approximately -1°C/min.)

7. The frozen preparations are stored in either the vapor or liquid phase of a nitrogen freezer.
8. To establish a culture from the frozen state place an ampule in a water bath set at 35°C (2 to 3 min). Immerse the vial just sufficient to cover the frozen material. Do not agitate the vial.
9. Immediately after thawing, aseptically remove the contents of the ampule and inoculate into 5 mL of fresh ATCC medium 355 in a 16 x 125 mm screw-capped test tube. Incubate upright at 25°C with caps screwed on tightly.

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

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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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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