ATCC

Product Sheet

Brachiola algerae (ATCC® PRA-109™)

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.

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: *Brachiola algerae* (ATCC® PRA-109™)

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
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Or contact your local distributor

Description

Strain Designation: Human Muscle Isolate
Deposited Name: *Brachiola algerae*
Depositor: LM Weiss, CM Coyle
Isolation:
clinical specimen - human
Pennsylvania,
Isolation date: January 7, 2003

Propagation

Growth Conditions
Temperature: 32.0°C
Duration: RK13 cells, use Eagle's minimum essential medium (EMEM); L6E9 cells, use Dulbecco's modified Eagle's medium (DMEM)
Protocol: place spores on fresh cell monolayer
Interval: monthly

Cryopreservation

Freeze medium: culture medium, 80%; fetal bovine serum, 10%; DMSO, 10%

1. To harvest the *Brachiola* culture, detach any remaining tissue culture cells (infected and uninfected) by scraping the surface of the flask with a cell scraper.
2. Transfer the cell suspension (including parasites) to 15 ml plastic centrifuge tubes. Centrifuge at 1300 x g for 10 min.
3. Remove all but 0.5 ml of the supernatant from each tube, resuspend the cell pellets, and pool them to a single tube.
4. Pass the resulting cell suspension through a syringe equipped with a 27 gauge 1/2 in needle to break up any remaining cells. Adjust the parasite concentration to 2.0 - 4.0 x 10^7 cells/ml with fresh medium or PBS.
   NOTE: If the concentration of parasites is too low, centrifuge at 1300 x g for 10 min and resuspend in the volume of fresh medium or PBS required to yield the desired concentration.
5. Prepare a cryoprotective solution containing 20% (v/v) DMSO and 6% (v/v) HIFBS in fresh medium or PBS.
6. Mix the cell preparation and cryoprotective solution in equal portions. The final concentration will be 1.0 - 2.0 x 10^7 cells/ml, 10% DMSO, and 3% HIFBS. The time from the mixing of the cell preparation and cryoprotective solution to the start of the freezing process should be no less than 15 min. and no more than 30 min.

NOTE: To prevent culture contamination, penicillin-streptomycin solution (ATCC 30-2300) may be added to a final concentration of 50 to 100 I.U./ml penicillin and 50 to 100 µg/ml streptomycin.

7. Dispense in 0.5 ml aliquots to 1.0-2.0 ml sterile plastic screw-capped cryules (special plastic vials for cryopreservation).
8. Place vials in a controlled rate freezing unit. From room temperature cool at -1°C/min to -40°C. If freezing unit can compensate for the heat of fusion, maintain rate at -1 C/min through heat of fusion. At -40°C plunge ampules 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 nitrogen. (The cooling rate in this apparatus is approximately -1°C/min.)
9. Store frozen ampules in either the vapor or liquid phase of a nitrogen refrigerator.
10. 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.
11. Immediately after thawing, aseptically transfer contents to a T-25 tissue culture flask containing a fresh monolayer of cells (ATCC CRL-1634 or suggested alternate) and 10 ml ATCC 30-2003 with 3% (v/v) HIFBS.
12. Outgas the flask for 10 seconds with a 95% air, 5% CO_2 gas mixture.
13. Incubate in a 35-37°C CO_2 incubator with the cap screwed on tightly.

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References

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

Biosafety Level: 2

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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.

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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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