ATCC

Product Sheet

Helicosporidium sp.
(ATCC® 50920™)

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.

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: *Helicosporidium sp.* (ATCC® 50920™)

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: Sj-1
Deposited Name: *Helicosporidium* sp.
Depositor: DG Boucias
Isolation:
black fly larvae, Simulium jonesi, Hatchet Creek, Alachua County, FL, 1998

Propagation

Growth Conditions
Temperature: 25.0°C

Medium
ATCC® Medium 28: Emmons' modification of Sabouraud's agar

Instructions for Complete Medium
ATCC Medium 28

Protocols

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 ampules 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 in a 35°C water bath, until thawed (2-3 min). Immerse the ampule just sufficient to cover the frozen material. Do not agitate the ampule.
2. Immediately after thawing, aseptically transfer the material to a 100 mm agar plate of ATCC medium 28 and evenly distribute the material over the surface of the agar with a spread bar.
3. Incubate the plate upright at 25°C. Viable cells should be apparent within 5 days.

Culture Maintenance

1. Aseptically transfer a loopful of material to a fresh plate of ATCC medium 28 and spread evenly over the surface with a spread bar.
2. Subculture every 4-6 weeks when incubated at 25C, or every 6-12 months when incubated at 18C.

Cryopreservation

1. Harvest cells from a culture that is at or near peak density. Add 2-3 ml fresh ATCC medium 28 broth to each plate and wash cells into suspension.
2. Collect cells by centrifugation at 800 x g for 5 min. Adjust the concentration of cells to 2 x 10^6 - 2 x 10^7/ml in fresh medium.
3. While cells are centrifuging prepare a 10% (v/v) solution of sterile methanol in fresh broth medium.
4. Mix the cell preparation and the 10% methanol solution in equal portions. Thus, the final concentration will be 10^6 - 10^7 cells/ml and 5% (v/v) Methanol. The time from mixing of the cell preparation and methanol stock solution to the beginning of the freezing process should be no less than 5 min and no greater than 15 min.
5. Dispense in 0.5 ml aliquots into 1.0 - 2.0 ml sterile plastic screw-capped cryules (special plastic vials for cryopreservation).
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 nitrogen. (The cooling rate in this apparatus is approximately
   -1°C/min.)
7. The frozen preparations should be stored in either the vapor or liquid phase of a nitrogen refrigerator. Frozen preparations stored below -130°C are stabile indefinitely. Those stored at temperatures above -130°C are progressively less stabile as the storage temperature is elevated. Vials can be stored between -80 and -70°C for no longer than one week.
8. To establish a culture from the frozen state place an ampule in a water bath set at 35°C. Immerse the vial to a level just above the surface of the frozen material. Do not agitate the vial.

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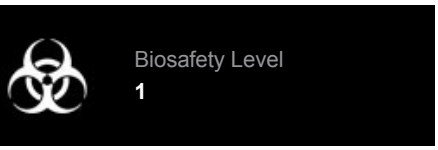

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9. Immediately after thawing, do not leave in the water bath, aseptically remove the contents of the ampule and transfer to a test tube containing 5 ml of ATCC medium 28 broth or to the surface of an ATCC medium 28 agar plate.
10. Incubate a test tube culture upright at 25°C with the cap screwed on loosely (loosened one-half turn); incubate a plate upright at 25°C. Subculture every 4-6 weeks when incubated at 25C, or every 6-12 months when incubated at 18C.

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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