ATCC

Product Sheet

Parauronema acutum (ATCC® 50307™)

Please read this FIRST

Storage Temp.
Frozen: -70°C or colder
Freeze-Dried: 2°C to 8°C
Live Culture: See Protocols Section

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: *Parauronema acutum* (ATCC® 50307™)

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: 110-3 killer
Deposited Name: *Parauronema acutum* Buddenbrock
Depositor: AT Soldo, EB Small
Isolation: Not applicable

Propagation

Growth Conditions
Temperature: 25°C
Culture System: Axenic

Medium
ATCC® Medium 1651: MA medium

Protocols

Handling of Live Culture
This strain is routinely shipped as a growing culture in a glass 16 x 125 mm screw-capped test tube. The volume of the cell suspension is approximately 5 mL. When the culture arrives remove it promptly from the shipping container. **Do not store the culture at refrigeration temperatures before handling.** To assure viability, immediately loosen the test tube cap and incubate upright at 25°C for at least one hour before observing the culture. There should be numerous active trophozoites in suspension. If the numbers are low the culture may have been exposed to temperature extremes in transit. Regardless of the state of the culture, aseptically transfer a 0.5 mL aliquot to a 16 x 125 mm screw-capped test tube containing 5 mL of sterile ATCC medium 1651. Incubate the parent and daughter cultures upright with the caps on loosely at 25°C.

Culture Maintenance
1. Screw the cap on tightly and vigorously agitate the culture.
2. Aseptically transfer a 0.1-0.25 mL aliquot to 5 mL of fresh medium in a 16 x 125 mm screw-capped test tube or T-25 tissue culture flask.
3. Incubate at 25°C (test tubes are incubated upright with the cap loosened one-half turn).
4. Subculture every 3-5 days.

Cryopreservation

Harvest and Preservation
1. Harvest cells from a culture that is at or near peak density by centrifugation at 800 x g for 5 min.
2. Adjust the concentration of cells to 2 x 10^6/mL in fresh medium.
3. While cells are centrifuging prepare a 22% (v/v) solution of sterile DMSO in fresh medium.
4. Mix the cell preparation and the 22% DMSO in equal portions. Thus, the final concentration will be 10^6 cells/mL and 11% (v/v) DMSO. The time from the mixing of the cell preparation and DMSO stock solution to the beginning of the freezing process should be no less than 15 min and no greater than 60 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 should not be stored above -55°C.
8. 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.
9. Immediately after thawing, do not leave in the water bath, gently remove the contents of the ampule with a Pasteur pipette and expel slowly into a 16 x 125 mm screw-capped test tube or T-25 tissue culture flask. Incubate at room temperature (approx. 25°C) for 15 min.

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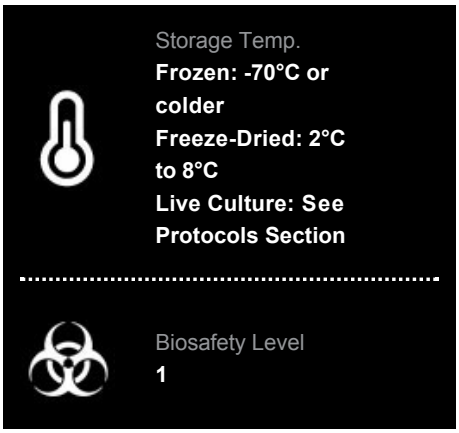

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10. At 15 min intervals add 0.25 mL of ATCC medium 1651 dropwise. Continue until the final volume is 2.0 mL.
11. Allow the culture to remain undisturbed for 15 min.
12. Add 0.5 mL of medium dropwise at 15 min intervals until the volume is 4.0 mL.
13. Allow the culture to remain undisturbed overnight.
14. On the morning of day 2 slowly add 4.0 mL of ATCC medium 1651. Allow the culture to remain undisturbed overnight.
15. When the culture is established, follow the protocol for maintenance of culture.

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.

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.

This product is sent with the condition that you are responsible for its safe storage, handling, and use. ATCC is not liable for any damages or injuries arising from receipt and/or use of this product. While reasonable effort is made to insure authenticity and reliability of strains on deposit, ATCC is not liable for damages arising from the misidentification or misrepresentation of cultures.

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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