ATCC

Product Sheet

Diplonema sp. 4 (ATCC® 50232™)

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: *Diplonema sp. 4* (ATCC® 50232™)

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: IVS
Deposited Name: *Diplonema* sp. 4
Depositor: TA Nerad
Isolation:
algal scrapings from rocks near shore, Solomon's Island, MD, 1986

Propagation

Growth Conditions
Temperature: 25.0°C
Duration: axenic

Medium
ATCC® Medium 1728: Enriched Isonema medium

Culture Maintenance
1. Vigorously agitate a culture at or near peak density and aseptically transfer 0.1-0.2 ml to 10 ml of fresh ATCC medium 1728 in a T-25 tissue culture flask.
2. Incubate in a 25°C incubator with the caps screwed on tightly.

Cryopreservation

1. Harvest the culture by agitating the contents of each flask. Transfer the cell suspensions to 15 ml plastic centrifuge tubes.
2. Spin the cell suspensions at approximately 500 x g for 5 min.
3. Pool the cell pellets and adjust the concentration to 2.0 - 4.0 x 10^7 cells/ml with a fresh ATCC medium 1728.
   *If the concentration is too low centrifuge at 500 x g for 5 min and resuspend in the volume of ATCC medium 1728 required to yield the desired concentration.
4. Mix the cell preparation and 20% (v/v) DMSO in equal portions. The final concentration will be 1.0 - 2.0 x 10^7 cells/ml and 10% DMSO. 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.
5. Dispense in 0.5 ml aliquots to 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 2.5 to 3 hours and then plunge ampules into liquid nitrogen. (The cooling rate in this apparatus is approximately -1°C/min.)
7. Store in either the vapor or liquid phase of a nitrogen refrigerator.
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, aseptically transfer contents to a T-25 tissue culture flask containing 10 ml ATCC medium 1728.
10. Incubate in a 25°C CO_2 incubator with the 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.

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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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