Nuclearia sp.

50694™

Description

Strain designation: CDC
Deposited As: *Nuclearia* sp.
Type strain: No

Storage Conditions

Product format: Frozen
Storage conditions: -80°C or colder for 1 week, vapor phase of liquid nitrogen for long-term storage

Intended Use

This product is intended for laboratory research use only. It is not intended for any animal or human therapeutic use, any human or animal consumption, or any diagnostic use.

BSL 1

ATCC determines the biosafety level of a material based on our risk assessment as guided by the current edition of *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, U.S. Department of Health and Human Services. It is your responsibility to understand the hazards associated with the material per your organization's policies and procedures as well as any other applicable regulations as enforced by your local or national agencies.

ATCC highly recommends that appropriate personal protective equipment is always

used when handling vials. For cultures that require storage in liquid nitrogen, it is important to note that some vials may leak when submersed in liquid nitrogen and will slowly fill with liquid nitrogen. Upon thawing, the conversion of the liquid nitrogen back to its gas phase may result in the vial exploding or blowing off its cap with dangerous force creating flying debris. Unless necessary, ATCC recommends that these cultures be stored in the vapor phase of liquid nitrogen rather than submersed in liquid nitrogen.

Certificate of Analysis

For batch-specific test results, refer to the applicable certificate of analysis that can be found at www.atcc.org.

Growth Conditions

Medium:
ATCC Medium 802: Sonneborn's Paramecium medium
Instructions for complete medium:
ATCC Medium 802 inoculated with *Klebsiella pneumoniae* subsp. *pneumoniae* (ATCC® 700831™) or *Enterobacter aerogenes* (ATCC® 13048™).
Temperature: 25°C
Culture system: Xenic

Handling Procedures

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 , until thawed (2-3 min). Immerse the ampule enough to cover only the frozen material. Do not agitate the ampule.
2. Immediately after thawing, aseptically transfer contents to a plastic screw-capped tube containing 5 ml ATCC medium 802 bacterized with *Klebsiella pneumoniae* subsp. *pneumoniae* (ATCC® 700831) or *Enterobacter aerogenes* (ATCC® 13048). Incubate the tube on a 15° horizontal slant at 25°C with the cap screwed on tightly.

Culture maintenance:

1. Vigorously agitate a culture at or near peak density and aseptically transfer 0.1-0.2 ml to a fresh tube of ATCC medium 802 bacterized with *Klebsiella pneumoniae* subsp. *pneumoniae* (ATCC® 700831 or *Enterobacter aerogenes* (ATCC® 13048).
2. Incubate the tube on a 15° horizontal slant at 25°C with the cap on tightly.

Cryopreservation:

1. Harvest cells from a culture that is at or near peak density by centrifugation at 600 x g for 5 min. Pool the cell pellets into a single tube.
2. Adjust the concentration of cells to 2.0 x 10^6/ml. If the concentration is too low, centrifuge at 600 x g for 5 minutes and resuspend the cell pellet with a volume of supernatant to yield the desired concentration.
3. Prepare a 20% (v/v) sterile DMSO solution in fresh medium w/o bacteria, as follows: Add the required volume of DMSO to a glass screw-capped test tube and place on ice. Allow the DMSO to solidify. Add the required volume of refrigerated ATCC medium 802 w/o bacteria. Dissolve the DMSO by inverting several times. If the DMSO solution is not prepared on ice, an exothermic reaction will occur that may precipitate certain components of the medium.
4. Mix the cell preparation and the DMSO in equal portions. Thus, the final concentration will be 10^6 and 10% (v/v) DMSO. 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 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 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.
7. The frozen preparations are stored in either the vapor or liquid phase of a nitrogen refrigerator.
8. To establish a culture from the frozen state place an ampule in a water bath set at 35°C. Immerse the vial enough to cover only the frozen material. Do not agitate the vial.
9. Immediately after thawing, do not leave in the water bath, aseptically remove the contents of the ampule and inoculate into 5.0 ml of fresh ATCC medium 802 bacterized with *Klebsiella pneumoniae* subsp. *pneumoniae* (ATCC® 700831 or *Enterobacter aerogenes* (ATCC® 13048)..
10. Incubate the tube on a 15° horizontal at 25°C with the cap screwed on tightly.

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: *Nuclearia* sp. (ATCC 50694)

References

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

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Revision

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Contact Information

ATCC
10801 University Boulevard
Manassas, VA 20110-2209
USA
US telephone: 800-638-6597
Worldwide telephone: +1-703-365-2700
Email: tech@atcc.org or contact your local distributor