

50366TM

Description

Strain designation: DE-1

Deposited As: Cercomonas bodo Meyer

Type strain: No

Storage Conditions

Product format: Frozen

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.

BSL₁

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

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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

Temperature: 25°C **Atmosphere:** Anaerobic

Incubation: Grown with Enterobacter aerogenes ATCC 13048 and mixed bacteria

Handling Procedures

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. One day before thawing the ampule, prepare bacterized ATCC medium 802 or

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1773, i.e. inoculate medium with a bacteriological loop of either *Klebsiella pneumoniae* subsp. *pneumoniae* (ATCC® 700831) or *Enterobacter aerogenes* (ATCC® 13048) from a nutrient agar slant (ATCC medium 3).

- 2. 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 it is thawed.
- 3. Immediately after thawing, aseptically transfer contents to a 16 x 125 mm screw-capped test tube containing 12 ml of bacterized ATCC Medium 802 or 1173. Screw the cap on tightly and incubate on a 15° horizontal slant at 25° C.

Culture maintenance:

- 1. Prepare bacterized ATCC medium 802 or 1773 one day in advance.
- 2. When the culture is at or near peak density, rub the surface of the tube with a sterile cotton swab, and agitate the swab to dislodge the adherent cells. Invert gently 10 times to distribute cells evenly.
- 3. Transfer approximately 0.25 ml to a 16 x 125 mm screw-capped test tube containing 12 ml of freshly bacterized ATCC medium 802 or 1773.
- 4. Screw the cap on tightly and incubate on a 15° horizontal slant at 25°C.

Reagents for cryopreservation:

Cryoprotective Solution

DMSO 2.0 ml

Fresh growth medium w/o bacteria 8.0 ml

Cryopreservation: 1. Mix the components in the order listed. When the medium is added to the DMSO the solution will warm up due to chemical heat.

- 2. Harvest cells from a culture that is at or near peak density by filtration and centrifugation at $850 \times g$ for 5 min.
- 3. Adjust the concentration of cells at least $2 \times 10^6/\text{ml}$ in fresh medium.
- 4. Mix the cell preparation and the cryoprotective solution in equal portions.



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- 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. 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. Ampules are stored in either the vapor or liquid phase of a nitrogen refrigerator.
- 8. To establish a culture from the frozen state place the vial in a 35°C water bath. Immerse the vial to a level just above the surface of the frozen material. Do not agitate the vial. Immediately after thawing, do not leave in water bath, aseptically remove the contents of the ampule and inoculate into a 16 x 125 mm screw-capped test tube containing 12 ml of ATCC Medium 802 or 1173 bacterized with *Klebsiella pneumoniae* subsp. *pneumoniae* (ATCC® 700831) or *Enterobacter aerogenes* (ATCC® 13048).
- 9. Incubate at 25°C with the cap screwed on tightly.
- 10. Follow the protocol for maintenance of culture.

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: *Cercomonas bodo* Meyer (ATCC 50366)

References

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

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