Echinamoeba thermarum Baumgartner et al.

PRA-13™

Description

Strain designation: OSB1
Deposited As: *Echinamoeba thermarum* Baumgartner et al.
Type strain: Yes

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.

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 2338: Echinamoeba broth
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ATCC Medium 2339: Echinamoeba agar
Instructions for complete medium: ATCC Medium 2338 inoculated with *Rhodobacter* sp. (ATCC® BAA-867)
Temperature: 50°C
Incubation: Grown with Rhodobacter sp., strain OSrt, ATCC BAA-867

Handling Procedures

Culture maintenance:
Subculture every week to fresh bacterized medium in the following manner:

1. For a plate culture, transfer cells with an inoculating loop to a fresh plate of previously-bacterized agar medium from a growing culture at or near peak density. For a broth culture, inoculate a fresh flask of previously-bacterized broth medium with 0.5 ml from a growing culture at or near peak density.

2. Incubate at 50°C. Keep the cap tightly sealed in the case of a flask culture.

Reagents for cryopreservation:
Cryoprotective Solution

DMSO	1.5 ml
Fresh growth medium w/o bacteria	8.5 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 800 x g for 5 min.

3. Adjust the concentration of cells at least 2 x 10^6/ml in freshmedium.

4. Mix the cell preparation and the cryoprotective solution in equal portions.

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 T-25 tissue culture flask containing 10 ml bacterized ATCC medium 2338. Screw the cap on tightly and

incubate the culture at 50°C. Alternatively, add the thawed contents of the ampule to the surface of a previously-bacterized Petri plate of ATCC medium 2338 agar. Wrap the plate culture with parafilm and incubate upright at 50°C.

9. 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: *Echinamoeba thermarum* Baumgartner et al. (ATCC PRA-13)

References

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

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