

PR∆-34[™]

Description

Strain designation: CR-1 [NE 070899/1]

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



PRA-34

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 2356: Sonneborn's Paramecium medium, alkaline

Instructions for complete medium: ATCC Medium 2356

Temperature: 20°C **Culture system:** Xenic

Incubation: Grown with Klebsiella pneumoniae subsp. pneumoniae ATCC 700831

added as a food source

Handling Procedures

Culture maintenance:

- 1. When the culture has thoroughly encysted, scrap the bottom of the flask with a cell scrapper and evenly suspend cysts.
- 2. Aspetically transfer a 0.2 ml aliquot of the cyst suspension to a T-25 flask containing 10 ml of ATCC Medium 2356 bacterized with *Klebsiella pneumoniae* subsp. *pneumoniae* ATCC 700831. Incubate the flask with the cap screwed on tightly at 25°C.

PRA-34

3. Transfer the culture every 3-4 weeks as described in steps 1-2. The transfer interval will depend on the quantity of the inoculum and the quality of the medium. This should be empirically determined by examining the culture on a daily basis until the growth cycle has stabilized.

Cryopreservation:

- 1. Allow the culture to encyst.
- 2. Place the flasks on ice and allow them to equilibrate for at least 30 minutes. Gently scrap the bottom of each flask with a cell scrapper to detach adherent cysts.
- 3. Transfer the cyst suspensions to 15 ml plastic centrifuge tubes and centrifuge at 800 x g for 5 minutes. Place the concentrated cyst suspension on ice. Adjust the concentration of cells to 2×10^5 /ml in fresh cold medium.
- 4. Mix the cell preparation and the cryoprotective solution in equal portions. Keep the suspension on ice.
- 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 Nalgene 1°C freezing apparatus. Place the apparatus at -80°C for 2.0 to 2.5 hours and then plunge ampules into liquid nitrogen. (The cooling rate in this apparatus is approximately -1°C/min.)
- Ampules are stored in either the vapor or liquid phase of a nitrogen refrigerator.
- 8. To establish a culture from the frozen state refer to the instructions above.

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: *Engelmanniella* sp. (ATCC PRA-34)

PRA-34

References

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

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PRA-34

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PRA-34

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