

50208<sup>TM</sup>

#### Description

**Strain designation: 331** 

**Deposited As:** Pseudocohnilembus marinus Thompson

Type strain: No

#### **Storage Conditions**

Product format: Test tube

#### 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<sub>1</sub>

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 1796: CRYS medium

Instructions for complete medium: ATCC Medium 1796

**Temperature:** 25°C **Culture system:** Axenic

#### Handling Procedures

#### **Culture maintenance:**

- 1. When the culture is at or near peak density, vigorously agitate the culture.
- 2. Transfer approximately 0.10 ml to a fresh tube containing 5 ml of fresh ATCC medium 1796.
- 3. Incubate upright at 25°C with caps loosened one-half turn.



#### **Cryopreservation:**

- 1. Prepare a 20% (v/v) sterile DMSO solution in fresh ATCC Medium 1796.
- 2. Transfer a culture at peak density to centrifuge tubes and spin at 230 x g for 5 minutes.
- 3. Remove the supernatant and resuspend the cells in ATCC medium 1796 to a concentration of 2 x  $10^6$  cells/ml.
- 4. Add the cryoprotectant solution, prepared in step 1, to the cell suspension in three equal aliquots every 2 minutes. The cell suspension should be at a 1:1 ratio with the cryoprotectant solution when it reaches its final volume. This will give a solution of 10% DMSO and 10<sup>6</sup> cells/ml.
- 5. Distribute the cell suspension in 0.5 ml aliquots into 1.0-2.0 ml sterile plastic screw-capped cryules (special plastic vials for cryopreservation). The time from mixing the cell preparation and the DMSO solution, before the cooling cycle begins should be no less than 15 min and no more than 30 min.
- 6. Place the vials in a controlled rate freezing unit. Use the following cooling cycle: From room temperature cool at
- -10°C/min to the heat of fusion; from the heat of fusion to
- -40°C cool at -1°C/min. At -40°C plunge into liquid nitrogen.
- 7. The frozen preparations are stored in either the vapor or liquid phase of a nitrogen freezer.
- 8. To establish a culture from the frozen state place an ampule in a water bath set at 35°C (2-3 min). Immerse the vial just sufficient to cover the frozen material. Do not agitate the vial.
- 9. Immediately after thawing, aseptically remove the contents of the ampule and inoculate into 5 ml of fresh ATCC medium 1796 in a 16 x 125 mm screw-capped test tube. Incubate upright at  $25^{\circ}$ C with caps loosened one-half turn.

#### Material Citation

If use of this material results in a scientific publication, please cite the material in the

following manner: Pseudocohnilembus marinus Thompson (ATCC 50208)

#### References

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

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#### Revision

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