



# ***Paramecium tetraurelia*** **Sonneborn**

**30977™**

## **Description**

**Strain designation:** stock d4-49

**Deposited As:** *Paramecium tetraurelia* Sonneborn

**Type strain:** No

**Genotype:** cl 1 (= clear 1)

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## **Storage Conditions**

**Product format:** Test tube

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## **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.

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## **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.

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## Certificate of Analysis

For batch-specific test results, refer to the applicable certificate of analysis that can be found at [www.atcc.org](http://www.atcc.org).

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## Growth Conditions

### Medium:

ATCC Medium 802: Sonneborn's Paramecium medium

**Instructions for complete medium:** ATCC Medium 802 inoculated with *Enterobacter aerogenes* (ATCC® 13048)

**Temperature:** 25°C

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## Handling Procedures

### Culture maintenance:

Subculture every two months to a fresh tube of bacterized medium in the following manner:

1. Transfer 0.5 ml from a growing culture to 5.0 ml of ATCC medium 802 bacterized

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with *Enterobacter aerogenes* (ATCC® 13048).

2. Add 1.0 ml of ATCC medium 802 bacterized with *Enterobacter aerogenes* ATCC 13048 twice weekly. When the tube is filled to within one inch of the top, aspirate from the bottom of the tube and reduce the volume to 5.0 ml.

3. Incubate upright at 25°C with the caps on loosely.

**Reagents for cryopreservation:**

## Cryoprotective Solution

DMSO	1.5 ml
Fresh growth medium w/o bacteria	7.5 ml
MgCl <sub>2</sub> (0.5 mM)	0.5 ml
CaCl <sub>2</sub> (0.5 mM)	0.5 ml

**Cryopreservation:** 1. Mix the components in the order listed. Before adding the MgCl<sub>2</sub> and the CaCl<sub>2</sub> allow the solution to return to room temperature. 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 200 x g for 1 min.

3. Adjust the concentration of cells to 2 x 10<sup>5</sup>/ml in fresh medium.

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.

7. Ampules are stored in either the vapor or liquid phase of a nitrogen refrigerator.

8. To establish a culture from the frozen state add 1.0 ml ATCC medium 802 to the frozen ampule and place it 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.

9. Immediately after thawing, do not leave in water bath, aseptically remove the contents of the ampule and inoculate onto the surface of an ATCC medium 919 (non-nutrient agar) plate containing an overlay of 15.0 ml of bacterized ATCC medium 802.

10. Incubate at 25°C.

11. Once the culture is established, transfer 0.5 ml to 5.0 ml of bacterized ATCC medium 802.

12. Follow the protocol for maintenance of culture.

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## Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: *Paramecium tetraurelia* Sonneborn (ATCC 30977)

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## References

References and other information relating to this material are available at [www.atcc.org](http://www.atcc.org).

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