ATCC

Product Sheet

Salpingoeca punica (ATCC® 50788™)

Please read this FIRST

Storage Temp.

Frozen Cultures:

-70°C for 1 week; liquid N_2 vapor for long term storage

Freeze-dried Cultures:

2-8°C

Live Cultures:

See Protocols section for handling information

Biosafety Level

1

Intended Use

This product is intended for research use only. It is not intended for any animal or human therapeutic or diagnostic use.

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: *Salpingoeca punica* (ATCC® 50788™)

American Type Culture Collection
PO Box 1549
Manassas, VA 20108 USA
www.atcc.org

800.638.6597 or 703.365.2700
Fax: 703.365.2750
Email: Tech@atcc.org

Or contact your local distributor

Description

Strain Designation: PR1
Deposited Name: *Salpingoeca amphoridium* Clark
Depositor: TA Nerad
Isolation:

Propagation

Growth Conditions
Temperature: 25°C
Culture System: Xenic

Medium
ATCC® Medium 802: Sonneborn's Paramecium medium

Instructions for Complete Medium
ATCC Medium 802 inoculated with *Enterobacter aerogenes* (ATCC® 13048™).

Protocols

Storage and Culture Initiation
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 ampoules 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, i.e., inoculate medium with a bacteriological loop of *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.
3. Immediately after thawing, aseptically transfer the contents to a T-25 tissue culture flask containing 10 ml of bacterized ATCC Medium 802.
4. Screw the cap on tightly and incubate the tube at 25°C.

Culture Maintenance

1. Prepare bacterized ATCC medium 802.
2. Inoculate a T-25 tissue culture flask containing 10.0 mL of bacterized ATCC medium 802 with 0.1 mL from a predominantly encysted *Heteromita globosa* culture.
3. Incubate at 25°C with cap screwed on tightly.
4. For routine maintenance subculture every 10-14 d.

Cryopreservation

1. Harvest cells from a culture that is at or near peak density by centrifugation at 800 x g for 5 min.
2. Adjust the concentration of cells to 2 x 10^6 - 10^7/mL in fresh medium.
3. While cells are centrifuging prepare a 20% (v/v) solution of sterile DMSO in fresh medium.
   a. Add 2.0 mL of DMSO to an ice cold 20 x 150 mm screw-capped test tube;
   b. Place the tube on ice and allow the DMSO to solidify (~5 min) and then add 8.0 mL of ice cold medium;
   c. Invert several times to dissolve the DMSO;
   d. Allow to warm to room temperature.
4. Mix the cell preparation and the DMSO in equal portions. Thus, the final concentration will be 10^6 - 10^7 and 10% (v/v) DMSO. The time from the mixing of the cell preparation and DMSO stock solution before the freezing process is begun should be no less than 15 min and no longer than 30 min.
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 controlled rate freezing unit. From room temperature cool at -1°C/min to -40°C. If the freezing unit can compensate for the heat of fusion, maintain rate at -1°C/min through the heat of fusion. At -40°C plunge into liquid nitrogen. Alternatively, place the vials in a Nalgene 1°C freezing

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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. The frozen preparations should be stored in either the vapor or liquid phase of a nitrogen refrigerator. Frozen preparations stored below -130°C are stabile indefinitely. Those stored at temperatures above -130°C are progressively less stabile as the storage temperature is elevated. If frozen material is to be stored at temperatures between -130°C and -70°C the shelf life should be empirically tested, i.e., remove stored material at intervals to determine die-off rate.
8. To establish a culture from the frozen state place an ampule in a water bath set at 35°C. Immerse the ampule to a level just above the surface of the frozen material. Do not agitate the ampule.
9. Immediately after thawing, do not leave in the water bath, aseptically remove the contents of the ampule and inoculate a T-25 tissue culture flask containing 10 mL of bacterized ATCC medium 802.
10. Incubate with the cap screwed on tightly at 25°C.

References

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

Biosafety Level: 1

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the current publication of the *Biosafety in Microbiological and Biomedical Laboratories* from the U.S. Department of Health and Human Services Centers for Disease Control and Prevention and National Institutes for Health.

ATCC Warranty

The viability of ATCC® products is warranted for 30 days from the date of shipment, and is valid only if the product is stored and cultured according to the information included on this product information sheet. ATCC lists the media formulation that has been found to be effective for this strain. While other, unspecified media may also produce satisfactory results, a change in media or the absence of an additive from the ATCC recommended media may affect recovery, growth and/or function of this strain. If an alternative medium formulation is used, the ATCC warranty for viability is no longer valid.

Disclaimers

This product is intended for laboratory research purposes only. It is not intended for use in humans.

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Please see the enclosed Material Transfer Agreement (MTA) for further details regarding the use of this product. The MTA is also available on our Web site at www.atcc.org

Additional information on this culture is available on the ATCC web site at www.atcc.org.

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