




Product Sheet


# *Tetraselmis sp.* (ATCC® 50245™)

Please read this FIRST



Storage Temp.  
**Frozen: -70°C or colder**  
**Freeze-Dried: 2°C to 8°C**  
**Live Culture: See Protocols Section**

---



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: *Tetraselmis sp.* (ATCC® 50245™)

American Type Culture Collection  
PO Box 1549  
Manassas, VA 20108 USA  
[www.atcc.org](http://www.atcc.org)

800.638.6597 or 703.365.2700  
Fax: 703.365.2750  
Email: [Tech@atcc.org](mailto:Tech@atcc.org)

Or contact your local distributor

## Description

**Strain Designation:** RG-35 [CCMP UW-473]

**Deposited Name:** *Tetraselmis sp.*

**Depositor:** RA Lewin

**Isolation:** Unknown location, 1973

## Propagation

### Growth Conditions

**Temperature:** 25°C

**Culture System:** Axenic

### Medium

ATCC® Medium 1747: Tetraselmis medium

## 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 ampules 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. To thaw a frozen ampule, place in a 35°C water bath, until thawed (2-3 min). Immerse the ampule just sufficient to cover the frozen material. Do not agitate the ampule.
2. Immediately after thawing, aseptically transfer the entire contents to a single 16 x 125 mm screw-capped test tube containing 5 mL of ATCC Medium 1747 broth. Incubate the tube on a 15° horizontal slant with the cap screwed on loosely (loosened one half turn) at 25°C under a 14 hour light (~50  $\mu$ Einsteins/m<sup>2</sup>/s irradiance)/10 hour dark cycle.

### Culture Maintenance

1. Inoculate a tube of fresh medium with 0.1 mL from a growing culture at or near peak density.
2. Incubate on a 15° horizontal slant with the cap screwed on loosely (loosened one half turn) at 25°C under a 14 hour light (~50  $\mu$ Einsteins/m<sup>2</sup>/s irradiance)/10 hour dark cycle.
3. Subculture as necessary (i.e., every 14-21 days).

## Cryopreservation

### Reagents

#### Cryoprotective Solution

DMSO, 1.5 mL

Fresh growth medium, 8.5 mL

### Harvest and Preservation

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  $2 \times 10^6$  -  $2 \times 10^7$ /mL in fresh medium.
3. While cells are centrifuging prepare a 15% (v/v) solution of sterile DMSO in fresh medium.
4. Mix the cell preparation and the 15% DMSO in equal portions. Thus, the final concentration will be  $10^6$  -  $10^7$  cells/mL and 7.5% (v/v) DMSO. The time from the mixing of the cell preparation and DMSO stock solution to the beginning of the freezing process should be no less than 5 min and no greater than 15 min.
5. Dispense in 0.5 mL aliquots into 1.0 - 2.0 mL sterile plastic screw-capped cryovials (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 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 stable indefinitely. Those stored at temperatures above -130°C are progressively less stable as the storage temperature is elevated. Vials should not be



## Product Sheet

# *Tetraselmis sp.* (ATCC®) 50245™)

### Please read this FIRST



Storage Temp.  
**Frozen: -70°C or colder**  
**Freeze-Dried: 2°C to 8°C**  
**Live Culture: See Protocols Section**

---



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: *Tetraselmis sp.* (ATCC® 50245™)

stored above -55°C.

- To establish a culture from the frozen state place an ampule in a water bath set at 35°C. Immerse the vial just to a level just above the surface of the frozen material. Do not agitate the vial.
- Immediately after thawing, do not leave in the water bath, aseptically remove the contents of the ampule and add to a centrifuge tube containing 5 mL of ATCC medium 1747. Centrifuge at 300 x g for 5 min.
- Remove most of the supernatant (=cryoprotectant, which can inhibit growth) and then resuspend the pellet. Transfer the culture to a 16 x 125 mm screw-capped test tube containing 5 mL of ATCC medium 1747.
- Incubate on a 15° horizontal slant with the cap screwed on loosely (loosened one half turn) at 25°C under a 14 hour light (~50 μEinsteins/m<sup>2</sup>/s irradiance)/10 hour dark cycle.



### References

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

While ATCC uses reasonable efforts to include accurate and up-to-date information on this product sheet, ATCC makes no warranties or representations as to its accuracy. Citations from scientific literature and patents are provided for informational purposes only. ATCC does not warrant that such information has been confirmed to be accurate.

This product is sent with the condition that you are responsible for its safe storage, handling, and use. ATCC is not liable for any damages or injuries arising from receipt and/or use of this product. While reasonable effort is made to insure authenticity and reliability of strains on deposit, ATCC is not liable for damages arising from the misidentification or misrepresentation of cultures.

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](http://www.atcc.org)

Additional information on this culture is available on the ATCC web site at [www.atcc.org](http://www.atcc.org).

© ATCC 2016. All rights reserved. ATCC is a registered trademark of the American Type Culture Collection. [12/02]

American Type Culture Collection  
PO Box 1549  
Manassas, VA 20108 USA  
[www.atcc.org](http://www.atcc.org)

800.638.6597 or 703.365.2700  
Fax: 703.365.2750  
Email: [Tech@atcc.org](mailto:Tech@atcc.org)

Or contact your local distributor