



Product Sheet

Haematococcus lacustris (ATCC® 30402™)

Please read this FIRST

Storage Temp.
Frozen Cultures:
-70°C for 1 week;
liquid N₂ 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: *Haematococcus lacustris* (ATCC® 30402™)

American Type Culture Collection
PO Box 1549
Manassas, VA 20108 USA
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800.638.6597 or 703.365.2700
Fax: 703.365.2750
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Or contact your local distributor

Description

Strain Designation: UTEX 16 [CCAP-34/1b]
Deposited Name: *Haematococcus lacustris* (Girod-Chantrons) Rostafinski
Depositor: UTEX
Isolation: Rreshwater, Czechoslovakia, (?)

Propagation

Growth Conditions
Temperature: 25°C
Culture System: Axenic

Medium
ATCC® Medium 847: Algal proteose agar

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 847 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 (~50-100 μEinsteins/m²/s irradiance)/10 hour dark cycle. Alternatively, add the entire thawed contents to the surface of a 20 x 100 mm Petri plate containing 20 mL of ATCC medium 847 agar. Wrap the plate culture with parafilm and incubate upright under the same light/dark cycle as specified for a test tube culture.

Culture Maintenance

1. For a broth culture, inoculate a tube of fresh broth medium with 0.1 ml from a growing culture at or near peak density. For a plate culture, transfer cells with an inoculating loop to a plate of fresh agar medium from a growing culture at or near peak density.
2. Incubate a test tube on a 15° horizontal slant with the cap screwed on loosely (loosened one half turn) at 25°C under a 14 hour light (~50-100 μEinsteins/m²/s irradiance)/10 hour dark cycle. Wrap a plate culture with parafilm and incubate upright under the same light/dark cycle as specified for a test tube culture.

Cryopreservation

Harvest and Preservation

1. Harvest cells from a culture which is at or near peak density by adding 3.0 mL fresh ATCC medium 847 broth to the slant and washing cells into suspension.
2. Adjust the concentration of cells to 4 x 10⁶/mL with fresh broth medium, then dilute to half this concentration by adding an equal amount of a 10% (v/v) sterile methanol solution in fresh ATCC medium 847 broth.
3. Dispense in 0.5 mL aliquots into 1.0 - 2.0 mL sterile plastic screw-capped cryovials (special plastic vials for cryopreservation). The time from mixing of the cell preparation and the methanol solution, before the cooling cycle begins, should be no greater than 15 min.
4. 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. 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.)
5. 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 can be stored



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between -80 and -70°C for no longer than one week.

6. To establish a culture from the frozen state place an ampule in a water bath set at 35°C. Immerse the vial to a level just above the surface of the frozen material. Do not agitate the vial.
7. 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 847 without agar. Centrifuge at 300 x g for 5 min.
8. Remove most of the supernatant (=methanol, which can inhibit growth) and then resuspend the pellet. Transfer the culture to a fresh tube of ATCC medium 847 and incubate on a 15° horizontal slant at 50-100 μEinsteins/m²/s irradiance at 25°C with the cap loosened one half turn. Maintain under a 14/10 h light-dark photoperiod. Alternatively, add the entire thawed contents to the surface of a 20 x 100 mm Petri plate containing 20 mL of ATCC medium 847 agar. Wrap the plate culture with parafilm and incubate upright under the same light/dark cycle as specified for a test tube culture.



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

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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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