



Product Sheet

# *Chlorella ellipsoidea* (ATCC® 30404™)

Please read this FIRST

Storage Temp.  
**Frozen Cultures:**  
-70°C for 1 week;  
liquid N<sub>2</sub> 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: *Chlorella ellipsoidea* (ATCC® 30404™)

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800.638.6597 or 703.365.2700  
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Or contact your local distributor

## Description

**Strain Designation:** UTEX 20 [CCAP-211/1a]  
**Deposited Name:** *Chlorella saccharophila* var. *ellipsoidea*  
**Depositor:** UTEX  
**Isolation:** Freshwater, (?)

## Propagation

**Growth Conditions**  
**Temperature:** 25°C  
**Culture System:** Axenic

**Medium**  
ATCC® Medium 847: Algal proteose agar

## Protocols

### Storage and Culture Initiation

This strain is distributed as a freeze-dried preparation. See the general procedures for opening a freeze-dried vial.

1. To rehydrate an ampule, aseptically add 0.5 mL of ice-cold medium 847 broth containing 12% sucrose to the freeze-dried inner shell vial.
2. Once completely rehydrated, 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  $\mu$ Einsteins/m<sup>2</sup>/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, screw the cap on tightly and vigorously agitate the culture. Aseptically transfer a 0.1 mL aliquot to 5 mL of fresh medium in a 16 x 125 mm screw-capped test tube. For a plate culture, transfer cells to a fresh plate with an inoculating loop.
2. Loosen the cap on a test tube culture and incubate on a 15° horizontal slant at 25°C under a 14 hour light (~50  $\mu$ Einsteins/m<sup>2</sup>/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
3. Subculture every 14-21 days.

## Cryopreservation

### 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 x 10<sup>6</sup> - 2 x 10<sup>7</sup>/mL in fresh medium.
3. While cells are centrifuging prepare a 10% (v/v) solution of sterile methanol in fresh medium.
4. Mix the cell preparation and the 10% methanol in equal portions. Thus, the final concentration will be 10<sup>6</sup> - 10<sup>7</sup> cells/mL and 5% (v/v) Methanol. The time from the mixing of the cell preparation and methanol 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 stored above -55°C.



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- 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 inoculate a 16 x 125 mm screw-capped test tube containing 5 mL of ATCC medium 847 broth or to the surface of ATCC medium 847 agar plate (20 x 100 mm Petri plate containing 20 mL of ATCC medium 847 agar).
- Incubate the culture on a 15° horizontal slant at 25°C with the cap screwed on loosely (loosened one-half turn) and incubate under a 14 hour light (~50 μEinsteins/m<sup>2</sup>/s irradiance)/10 hour dark cycle. Alternatively, add the entire contents of the thawed ampule to the surface of a 20 x 100 mm Petri plate containing 20 mL of ATCC medium 847 agar. Spread the material evenly over the plate with a sterile cooled spread bar. Incubate in the same manner as the test tube culture.



### 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

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### Disclaimers

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

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