



Product Sheet

# *Chlorogonium elongatum* (ATCC® 12895™)

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: *Chlorogonium elongatum* (ATCC® 12895™)

American Type Culture Collection  
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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:** 12-2/A [CCAP 12/2a, UTEX 11]  
**Deposited Name:** *Chlorogonium elongatum* Dangeard  
**Depositor:** EG Pringsheim  
**Isolation:** Freshwater, Germany, (?)



## Propagation

### Growth Conditions

**Temperature:** 20°C to 25°C  
**Culture System:** Axenic

### Medium

ATCC® Medium 5: Sporulation agar  
ATCC® Medium 351: Hutner's medium for Euglena

### Instructions for Complete Medium

**Media:** ATCC Medium 5  
**Alternate Media:** ATCC Medium 351 w 0.1% Na Acetate



## 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 sufficiently 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 5 broth (or alternatively, 5 mL ATCC Medium 351 supplemented with 0.1% Na Acetate). Allow the tube to stand upright for one to several hours (or centrifuge very lightly for 3-5 min) to allow most cells to settle to the tube base.
3. Gently remove most of the supernatant (save in a secondary tube), then resuspend the remaining cells in additional fresh medium to a total volume of 5-6 mL.
4. Incubate tubes on a 15° horizontal slant with the cap screwed on loosely (loosened one half turn) at 20-25°C under a 14 hour light (~50 µEinsteins/m<sup>2</sup>/s irradiance)/10 hour dark cycle.

**Note:** Some algae may grow poorly or not at all when recovered from the frozen state in broth media. In such cases, the algae should initially be recovered on agar media before eventually transferring to broth culture.

### Culture Maintenance

1. Inoculate a tube of fresh broth medium with 0.25 mL from a growing culture at or near peak density.
2. Incubate tubes on a 15° horizontal slant with the cap screwed on loosely (loosened one half turn) at 20-25°C under a 14 hour light (~50 µEinsteins/m<sup>2</sup>/s irradiance)/10 hour dark cycle.
3. Subculture as necessary (i.e., typically every 14-21 days in broth media).



## Cryopreservation

### Harvest and Preservation

1. Harvest cells from a culture that is at or near peak density by centrifugation at 400-500 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 DMSO in fresh medium.
4. Mix the cell preparation and the 10% DMSO in equal portions. Thus, the final concentration will be 10<sup>6</sup> - 10<sup>7</sup> cells/ml and 5% (v/v) DMSO. 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 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



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- 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.)
- 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.
  - 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 5 broth (or alternatively, 5 mL ATCC Medium 351 supplemented with 0.1% Na Acetate).
  - Gently remove most of the supernatant (save in a secondary tube), then resuspend the remaining cells in additional fresh medium to a total volume of 5-6 mL.
  - Incubate tubes on a 15° horizontal slant with the cap screwed on loosely (loosened one half turn) at 20-25°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

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### Disclaimers

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

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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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