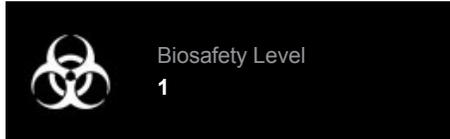




Product Sheet

# *Cercomonas vibrans* (ATCC® 50530™)

Please read this **FIRST**



## 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: *Cercomonas vibrans* (ATCC® 50530™)

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

**Deposited Name:** *Cercobodo* sp.  
**Depositor:** J Martinez-Cruz  
**Isolation:**  
dry soil, Mexico City, Mexico

## Propagation

### Growth Conditions

**Temperature:** 25.0°C

**Protocol:** This strain is shipped as a growing culture and includes *Adriamonas* sp. (food source for *Cercomonas vibrans*) and *Enterobacter aerogenes* ATCC 13048 (food source for *Adriamonas* sp.). The food will not be completely eliminated by the predator, *C. vibrans*. The *Adriamonas* sp. will eventually encyst. *C. vibrans* also produces cysts. Because those cysts are not very stable, the culture must be passaged every 7-10 days to ensure the culture's continued viability. Upon arrival of the shipment, agitate the culture, aseptically transfer the entire contents to a T-25 tissue culture flask, and incubate it at 25C. The bacterial flora present in the shipped culture will support growth of the *Adriamonas* sp., but growth can be improved by using medium that has been bacterized with *E. aerogenes* ATCC 13048 approximately 24 hours before inoculation with the *Cercomonas/Adriamonas* culture. (Other species of bacteria may work equally well, but this one has been empirically assessed.) After bacterization of the medium, agitate the culture of ATCC 50530 and aseptically transfer 0.25 ml to the new medium. Screw the cap on tightly and incubate at 25C.

### Medium

ATCC® Medium 1967: Soil freshwater cereal grass medium

### Instructions for Complete Medium

ATCC Medium 1967 inoculated with *Klebsiella pneumoniae* subsp. *pneumoniae* (ATCC® 700831) or *Enterobacter aerogenes* (ATCC® 13048).

## Protocols

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 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. Do not leave ampule in water bath after it is thawed.
2. Add the thawed contents to a T-25 flask containing 10 ml of ATCC medium 802 bacterized with *Klebsiella pneumoniae* subsp. *pneumoniae* (ATCC® 700831) or *Enterobacter aerogenes* (ATCC® 13048).
3. Incubate with the cap tightly sealed at 25°C.

### Culture Maintenance

Subculture every 7-10 days to a fresh T-25 flask of bacterized medium in the following manner:

1. Vigorously agitate the flask and aseptically transfer 0.5 ml from a growing culture to a T-25 tissue culture flask containing 10.0 ml of ATCC medium 802 bacterized with *Klebsiella pneumoniae* subsp. *pneumoniae* (ATCC® 700831) or *Enterobacter aerogenes* (ATCC® 13048).
2. Incubate flask at 25°C with the cap on tightly.

## Cryopreservation

### Cryoprotective Solution

|                                  |        |
|----------------------------------|--------|
| DMSO                             | 2.0 ml |
| Fresh growth medium w/o bacteria | 8.0 ml |

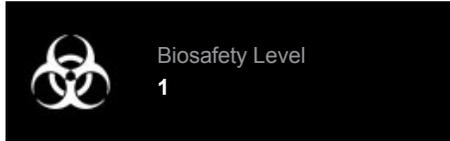
1. Mix the components in the order listed. When the medium is added to the DMSO the solution will warm up due to chemical heat.
2. Harvest cells from a culture that is at or near peak density by filtration and centrifugation at 800 x g for 5 min.



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- Adjust the concentration of cells at least  $2 \times 10^6$ /ml in fresh medium.
- Mix the cell preparation and the cryoprotective solution in equal portions.
- Dispense in 0.5 ml aliquots into 1.0 - 2.0 ml sterile plastic screw-capped cryovials (special plastic vials for cryopreservation).
- Place vials in a controlled rate freezing unit. From room temperature cool at  $-1^\circ\text{C}/\text{min}$  to  $-40^\circ\text{C}$ . If freezing unit can compensate for the heat of fusion, maintain rate at  $-1^\circ\text{C}/\text{min}$  through heat of fusion. At  $-40^\circ\text{C}$  plunge ampules into liquid nitrogen. Alternatively, place the vials in a Nalgene  $1^\circ\text{C}$  freezing apparatus. Place the apparatus at  $-80^\circ\text{C}$  for 1.5 to 2 hours and then plunge ampules into liquid nitrogen. (The cooling rate in this apparatus is approximately  $-1^\circ\text{C}/\text{min}$ .)
- Ampules are stored in either the vapor or liquid phase of a nitrogen refrigerator.
- To establish a culture from the frozen state place the vial in a  $35^\circ\text{C}$  water bath. Immerse the vial to a level just above the surface of the frozen material. Do not agitate the vial. Immediately after thawing, do not leave in water bath, aseptically remove the contents of the ampule and inoculate into a T-25 tissue culture flask containing 10 ml ATCC medium 802 bacterized with *Klebsiella pneumoniae* subsp. *pneumoniae* (ATCC® 700831) or *Enterobacter aerogenes* (ATCC® 13048).
- Incubate at  $25^\circ\text{C}$  with the cap screwed on tightly.
- Once the culture is established, vigorously agitate the flask and aseptically transfer 0.5 ml to 10.0 ml of bacterized ATCC medium 802.
- Follow the protocol for maintenance of 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

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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