ATCC

Product Sheet

# *Sorogena stoianovitchae* (ATCC® 50031™)

**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: *Sorogena stoianovitchae* (ATCC® 50031™)

American Type Culture Collection
PO Box 1549
Manassas, VA 20108 USA
www.atcc.org

800.638.6597 or 703.365.2700
Fax: 703.365.2750
Email: Tech@atcc.org

*Or contact your local distributor*

## Description

**Strain Designation:** PNG 76-73
**Deposited Name:** *Sorogena stoianovitchae* Bradbury and Olive
**Depositor:** RL Blanton
**Isolation:** Old aborted fruits and stalks of *Ficus botryocarpa*, Papua New Guinea, 1976

## Notes

This strain feeds on *Colpoda* sp. (both organisms are included in the culture). This is a xenic culture that contains bacteria.

## Propagation

**Growth Conditions**
**Temperature:** 20°C to 25°C

**Medium**
ATCC® Medium 1330: Sorogena medium

## Protocols

**Handling of Live Culture**
This strain is routinely shipped as a growing culture in a glass 16 x 125 mm screw-capped test tube. The volume of the cell suspension is approximately 5-6 mL. When the culture arrives remove it promptly from the shipping container. **Do not store the culture at refrigeration temperatures before handling.** To assure viability, immediately loosen the test tube cap and incubate upright at 25°C for at least one hour before observing the culture. There should be a number of attached *Sorogena* sorocarps near the fluid meniscus. If the numbers are low the culture may have been exposed to temperature extremes in transit. Regardless of the state of the culture, suspend attached sorocarps by placing the tube on ice for 10 min, then rub the inside surface of the tube with a sterile inoculating loop or cotton swab to detach cells. Aseptically transfer the entire contents of the tube to a petri plate or T-25 tissue culture flask containing a bed of agar medium with 6-8 mL additional liquid overlay (ATCC medium 1330). Incubate the culture at 20-25°C under a 14 hour light (~50 µEinsteins/$m^2$/s irradiance)/10 hour dark cycle.

**Culture Maintenance**

1. When the *Sorogena* have sufficiently reduced the number of prey *Colpoda* in the culture, their ability to aggregate and form sorocarps is diminished, and both predator and prey form cysts. To revive an encysted culture, the liquid overlay should be aseptically removed and replaced with fresh medium, which will stimulate new bacterial growth and encourage excystment of *Colpoda* and then *Sorogena*.
2. Incubate the culture at 20-25°C under a 14 hour light (~50 µEinsteins/$m^2$/s irradiance)/10 hour dark cycle. Active trophozoites (ciliates) of both *Colpoda* and *Sorogena* should be observed within 2-3 days. *Sorogena* sorocarps (cysts) should form just above the air-liquid interface in 5-7 days.
3. To encourage faster proliferation of *Sorogena*, additional *Colpoda* sp. may be added to the culture from a separate culture of the prey organism kept in parallel (i.e., ATCC® 30920™ or similar, not provided).
4. The *Sorogena* may be passaged to a new petri plate or T-25 flask by gently rubbing the agar surface with a spread bar to dislodge attached sorocarps and/or cysts, then transferring 0.5 to 2 mL to a fresh petri plate or T-25 flask containing a bed of agar medium and 10-15 mL total liquid overlay (ATCC medium 1330).
5. Incubate the culture at 20-25°C under a 14 hour light (~50 µEinsteins/$m^2$/s irradiance)/10 hour dark cycle. Active trophozoites (ciliates) of both *Colpoda* and *Sorogena* should be observed within 2-3 days. *Sorogena* sorocarps (cysts) should form just above the air-liquid interface in 5-7 days. Several cycles of growth and encystment may be required in order for the *Sorogena* culture to produce fruiting bodies.

## Cryopreservation

**Reagents**
Cryoprotective Solution
DMSO, 1.5 mL

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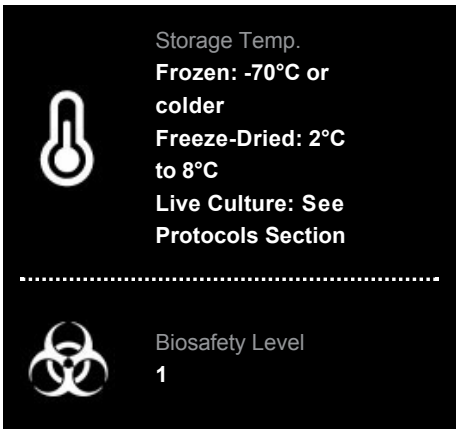


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Fresh growth medium w/o bacteria, 8.5 mL

**Harvest and Preservation**

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 *Sorogena* and *Colpoda* cysts from a culture that has recently passed peak density by centrifugation at 1000 x g for 5 min.
3. Adjust the concentration of cells to at least 2 x $10^4$/mL in fresh medium.
4. Mix the cell preparation and the cryoprotective solution in equal portions by adding the cryoprotective solution to the cell suspension in 3 equal aliquots at 2 min. intervals.
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 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.)
7. Ampules are stored in either the vapor or liquid phase of a nitrogen refrigerator.
8. To establish a culture from the frozen state place the vial in a 35°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 transfer to a petri plate or T-25 tissue culture flask containing a bed of agar medium and 10-15 mL total liquid overlay (ATCC medium 1330).
9. Optionally, aseptically transfer 0.2-0.5 mL from a growing culture of *Colpoda* sp. to the petri plate or T-25 flask (See section on Culture Maintenance).
10. Incubate the culture at 20-25°C under a 14 hour light (~50 µEinsteins/$m^2$/s irradiance)/10 hour dark cycle. Active trophozoites (ciliates) of both *Colpoda* and *Sorogena* should be observed within 2-3 days. Once the culture is established, follow the protocol for maintenance of 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

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.

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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.