



Product Sheet

# *Trypanosoma lewisi* (ATCC® 30147™)

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: *Trypanosoma lewisi* (ATCC® 30147™)

American Type Culture Collection  
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## Description

**Strain Designation:** New Orleans-67  
**Deposited Name:** *Trypanosoma lewisi* (Kent) Laveran and Mesnil  
**Depositor:** RG Yaeger  
**Isolation:**  
rat, *Rattus norvegicus*, New Orleans, 1967

## Propagation

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

**Medium**  
ATCC® Medium 1029: LIT medium

## Instructions for Complete Medium

ATCC medium 1029 supplemented with an additional 5-10 µg/mL hemin  
Note: This strain grows poorly without the additional hemin in the growth medium

## 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 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 contents to a T-25 tissue culture flask containing 10 mL ATCC medium 1029 supplemented with an additional 5-10 µg/mL hemin. Incubate at 25°C with the cap screwed on tightly.

## Culture Maintenance

1. Vigorously agitate a culture at or near peak density and aseptically transfer 0.1-0.2 mL to a fresh flask containing 10 mL ATCC medium 1029 supplemented with an additional 5-10 µg/mL hemin.
2. Incubate at 25°C with the cap screwed on tightly.
3. Subculture every 10-14 days as described in step 1. The transfer interval will depend on the quantity of the inoculum and the quality of the medium. This should be empirically determined by examining the culture on a daily basis until the growth cycle has stabilized.

## Cryopreservation

1. Harvest cells from a culture which is at or near peak density by centrifugation at 1,300 g for 5 min.
2. Adjust concentration of cells to  $2 \times 10^7$ /mL in fresh medium.
3. While cells are centrifuging prepare a 10% (v/v) solution of sterile DMSO in fresh medium (broth). The DMSO solution when first prepared will warm up due to chemical heat. The solution should be allowed to return to room temperature prior to use.
4. Mix the cell preparation and the DMSO solution in equal portions. The final concentration will be  $10^7$  cells/mL and 5% (v/v) DMSO. The time from the mixing of the cell preparation and DMSO stock solution before the freezing process is begun should be no more 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 ampules 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. Store in either the vapor or liquid phase of a nitrogen refrigerator.
8. 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 thawed.



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9. Immediately after thawing, do not leave in the water bath, aseptically transfer the contents of the ampule into a fresh flask containing 10 mL ATCC medium 1029 supplemented with an additional 5-10 µg/mL hemin.
10. Incubate at 25°C with the cap screwed on tightly.
11. Maintain as described above.



### 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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