Leishmania major (Yakimoff and Schokhor) Bray et al.

50132™

Description

Strain designation: MRHO/SU/59/P
Deposited As: *Leishmania major* (Yakimoff and Schokhor) Bray et al.
Type strain: No

Storage Conditions

Product format: Frozen

Intended Use

This product is intended for laboratory research use only. It is not intended for any animal or human therapeutic use, any human or animal consumption, or any diagnostic use.

BSL 2

ATCC determines the biosafety level of a material based on our risk assessment as guided by the current edition of *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, U.S. Department of Health and Human Services. It is your responsibility to understand the hazards associated with the material per your organization's policies and procedures as well as any other applicable regulations as enforced by your local or national agencies.

ATCC highly recommends that appropriate personal protective equipment is always used when handling vials. For cultures that require storage in liquid nitrogen, it is important to note that some vials may leak when submersed in liquid nitrogen and will slowly fill with liquid nitrogen. Upon thawing, the conversion of the liquid nitrogen back to its gas phase may result in the vial exploding or blowing off its cap with dangerous force creating flying debris. Unless necessary, ATCC recommends that these cultures be stored in the vapor phase of liquid nitrogen rather than submersed in liquid nitrogen.

Certificate of Analysis

For batch-specific test results, refer to the applicable certificate of analysis that can be found at www.atcc.org.

Growth Conditions

Medium:
ATCC Medium 807: Brain heart infusion blood agar
Instructions for complete medium: ATCC Medium 807
Temperature: 25°C
Culture system: Axenic

Handling Procedures

Culture maintenance:
1. When the culture has reached or is near peak density, invert tube 10 times and aseptically transfer a drop from a Pasteur pipette (0.05 ml) to another test tube containing fresh ATCC medium 807.

2. Incubate the culture vertically at 35°C with the cap screwed on tightly.

3. Transfer the culture every 3-4 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 cultures that are at or near peak density. Aseptically transfer the broth overlay to a plastic centrifuge tube and adjust the concentration of cells to 2 x 10^7/ml in fresh medium (broth overlay). If necessary, cells may be concentrated by centrifugation at 800 x g for 5 min.

2. Prepare a 10% (v/v) solution of sterile DMSO in fresh medium (broth). Cool on ice.

3. 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 solution to the start of the freezing process should be no less than 15 min and no longer than 30 min.

4. Dispense in 0.5 ml aliquots into 1.0 - 2.0 ml sterile plastic screw-capped cryules (special plastic vials for cryopreservation).

5. 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.)

6. The frozen preparations should be stored in either the vapor or liquid phase of a nitrogen refrigerator. Frozen preparations stored below -130°C are stabile indefinitely. Those stored at temperatures above -130°C are progressively less stabile as the storage temperature is elevated. Vials should not be stored above -70°C.

7. 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.

8. 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 ATCC medium 807.

9. Incubate the culture vertically at 25°C. Observe the culture daily and transfer when numerous trophozoites are observed.

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: *Leishmania major* (Yakimoff and Schokhor) Bray et al. (ATCC 50132)

References

References and other information relating to this material are available at www.atcc.org.

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Revision

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