ATCC

Product Sheet

Methanococcus vannielii (ATCC® 35089™)

Please read this FIRST

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: *Methanococcus vannielii* (ATCC® 35089™)

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

Designation: DSM 1224 [SB]
Deposited Name: *Methanococcus vannielii* Stadtman and Barker

Propagation

Medium
ATCC® Medium 2467: MS - OCM Base Medium

Growth Conditions
Temperature: 37.0°C
Atmosphere: Anaerobic; 80% H_2- 20% CO_2

Propagation Procedure
1. Sterilize the top of the Balch tube by spraying it with 70% ethanol and then flaming the top.
2. If needed, exchange the gas in the test tube for 80% H_2-20% CO_2, do not pressurize above 5 psi. If the tube is over pressurized, it will be difficult to inoculate.
3. If the medium is pink (see discussion about resazurin), add 2.0 ml of reducing agent (3% cysteine, stock solution) per 100 ml of medium. Let the medium sit at room temperature for 30 minutes (or until the resazurin becomes colorless) before inoculating.
4. Open the frozen vial and immediately place the vial under a stream of sterile anaerobic gas, to maintain anaerobic conditions. Wait for vial contents to thaw.
5. Using an anaerobic syringe (see d below), withdraw the cell suspension from the vial and transfer it to the primary Balch tube. Plate 0.1 ml of the inoculated culture onto a non-selective medium and incubate aerobically at 37ºC. Additional tubes can be inoculated by transferring 0.5 ml of the primary tube to secondary tube(s). Again, use good anaerobic technique. Once inoculated the Balch tubes should be pressurized up to 20 psi with 80% H_2-20% CO_2.
6. Growth should be detected in the broth within 48-72 hours. No growth should be detected on the aerobic plate or broth.
ANAEROBIC CONDITIONS:
a. Balch tubes (available from Bellco Glass, Vineland, NJ; are specially designed for anaerobic work and use an aluminum crimp cap to hold a rubber stopper in place. Needles can easily be inserted through the stopper, and the tubes can be pressurized up to 20 psi. Alternatively, serum vials may be used, or screw cap tubes with butyl rubber stoppers, in the latter case the stopper may be removed and the tube placed under a cannula system that dispenses sterile, oxygen free gas for addition of reducing agents or inoculation.
b. Resazurin is a commonly used redox indicator that is pink when the redox potential is above 50 mv, and colorless when the redox potential is below 110 mv. i.e. highly reducing. Most strict anaerobes require this low redox potential for optimum growth.
c. To obtain a fully reduced medium, it is necessary that the medium be anoxic and that a reducing agent be added. Common reducing agents are sodium sulfide, cysteine, dithiothreitol, and titanium citrate.

Notes

Growth may take up to one week when the frozen culture is initially inoculated into fresh medium. The broth culture appears as a smooth pellicle and growth throughout the tube.
Additional information on this culture is available on the ATCC® web site at www.atcc.org.

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

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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.

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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.

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