



***Thermoanaerobacter brockii* subsp. *brockii* (Zeikus et al.) Lee et al.**

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Description

Thermoanaerobacter brockii subsp. *brockii* is a thermophilic bacterial type strain that was isolated from a hot spring in Yellowstone National Park. This strain is part of the National Park Service (NPS) Special Collection.

Strain designation: [ATCC 33075, DSM 1457, HTD 4]

Deposited As: *Thermoanaerobium brockii* Zeikus et al.

Type strain: Yes

Patent depository: This material was deposited with the ATCC Patent Depository to fulfill U.S. or international patent requirements. This material may not have been produced or characterized by ATCC. As an International Depository Authority (IDA) for patent deposits, ATCC is required to complete viability testing only at time of initial deposit of patent material. Patent deposits are made available on behalf of the Depositor when the pertinent U.S. or international patent is issued, but material may not be used to infringe the patent claims.

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Storage Conditions

Product format: Freeze-dried

Storage conditions: 2°C to 8°C

Intended Use

This product is intended for laboratory research use only. It is not intended for any

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animal or human therapeutic use, any human or animal consumption, or any diagnostic use.

BSL 1

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

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Medium:

ATCC Medium 1465: Basal thermophile medium

Temperature: 60°C**Atmosphere:** Anaerobic

Handling Procedures

1. Sterilize the top of the Balch tube by spraying it with 70% ethanol and then flame the top.
2. If needed exchange the gas in the test tube for 80% N₂ 20% CO₂.
3. If the medium is pink (see discussion about resazurin) add 0.1 ml Na₂S·9H₂O (1.5% sodium sulfide, stock solution) per 5 - 10 ml of medium. Let the medium sit at room temperature for 10 to 20 minutes until the resazurin becomes colorless before inoculating.
4. When the Balch tube is ready to inoculate, thaw the frozen vial at room temperature under a gentle stream of oxygen-free gas.
5. For inoculation, use an anaerobic (see c below) 1.0 ml syringe tipped with 22-gauge needle, withdraw the cell suspension from the vial and transfer it to the broth. Plate 0.1 ml of the inoculated culture onto a non-selective medium and incubate aerobically at 37°C. Use 0.1 ml of the inoculated culture to inoculate a nonselective aerobic broth and an additional tube of #1465 broth. Incubate the non-selective aerobic broth tubes at 37°C. Incubate the #1465 broth culture at 60°C.
6. Growth should be detected in the #1465 broth within 24 to 48 hours. There should be no growth detected on the aerobic plate or in the aerobic broth.

ANAEROBIC CONDITIONS:

1. 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.
2. 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.
3. Syringes can be made anaerobic by displacing the dead space in the syringe with a sterile gas

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Notes

Cells are non-motile rods that occur singly, in pairs and chains. Cells within the same chain may be of different lengths. Colonies on #1465 are creamy and rounded, with irregular edges.

Additional information on this culture is available on the ATCC web site at www.atcc.org.

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: *Thermoanaerobacter brockii* subsp. *brockii* (Zeikus et al.) Lee et al. (ATCC 53556)

References

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

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Contact Information

ATCC

10801 University Boulevard

Manassas, VA 20110-2209

USA

US telephone: 800-638-6597

Worldwide telephone: +1-703-365-2700

Email: tech@atcc.org or contact your local distributor
