



Product Sheet

# *Thermotoga petrophila* (ATCC® BAA-488™)

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: *Thermotoga petrophila* (ATCC® BAA-488™)

## Description

**Designation:** JCM 10881 [DSM 13995, RKU-1]

**Deposited Name:** *Thermotoga petrophila* Takahata et al.

## Propagation

### Medium

ATCC® Medium 2332: *Thermotoga petrophila* medium

### Growth Conditions

**Temperature:** 70.0°C

**Atmosphere:** Anaerobic

### 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% N<sub>2</sub>-20% CO<sub>2</sub> or 100% N<sub>2</sub>.
  3. If the medium is pink (see discussion about resazurin) add 1 drop of reducing agent (1.5% sodium sulfide stock solution) for each one or two ml of medium. Let the medium sit at room temperature for 10 to 20 minutes, until the resazurin becomes colorless, before inoculating.
  4. Open the freeze-dried vial according to the enclosed instructions. Take an anaerobic (anaerobic conditions D. below) 1.0 ml syringe tipped with 22-gauge needle and withdraw 0.5 ml of medium from the Balch tube and rehydrate the entire freeze dried pellet. Immediately place the re-hydrated vial under a stream of sterile gas, 80N<sub>2</sub>-20%CO<sub>2</sub> to maintain anaerobicity.
  5. Using the same syringe, withdraw the cell suspension from the vial and transfer it to the Balch tube. Inoculate a second pre-reduced tube of medium with 0.5 ml of the rehydrated culture. Plate 0.1 ml of the inoculated culture onto a non-selective medium and incubate aerobically at 37°C. Incubate the broth tube at 70°C.
  6. Growth should be detected in the broth within 24 to 48 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 to 2 atm. 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.
  - D. Syringes can be made anaerobic by one of two methods.
    1. Displace the dead space in the syringe with a sterile oxygen-free gas.
    2. Displace the dead space in the syringe with a reducing agent.

## Notes

Once growth has been established, the culture should be transferred every 1 or 2 days. The culture will remain viable for 1 week if stored at room temperature and maintained under anaerobic conditions.

When this freeze-dried culture is incubated at elevated temperatures (60°C or above) the cryoprotectant agent precipitates. This precipitation does not interfere with growth of the organism.

Cells appear as short rod that occur singly and in pairs. When examined under phase microscopy the sheath can sometimes be detected.

Additional information on this culture is available on the ATCC® web site at [www.atcc.org](http://www.atcc.org).

## 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.

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Department of Health and Human Services Centers for Disease Control and Prevention and National Institutes for Health.

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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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