Thermotoga neapolitana Jannasch et al.

49049™

Description

Strain designation: NS-E [DSM 4359]
Deposited As: *Thermotoga neapolitana* Jannasch et al.
Type strain: Yes

Storage Conditions

Product format: Freeze-dried
Storage conditions: -80°C or colder

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 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

Medium:
ATCC Medium 2507: Thermotoga Medium
Temperature: 77°C
Atmosphere: 100% N_2

Handling Procedures

1. Sterilize the top of the Balch tube (see below) by spraying it with 70% ethanol and then flaming the top.
2. If needed, exchange the gas in the test tube for 100% N_2.
3. If the medium is pink (see discussion about resazurin) add 2.0 ml – 3.0 mL of reducing agent (1.5% sodium sulfide solution) per 100 ml of medium. Let the medium sit in the dark at room temperature for at least 24 to 48 hours, until

the resazurin becomes colorless, before inoculating.

4. After the Balch tube is ready to be inoculated, let the frozen vial thaw at room temperature under a gentle stream of 100% N_2.
5. Use an anaerobic 1.0 ml syringe (see below) tipped with 22-gauge needle and withdraw the entire contents from the vial. Transfer it to the broth and incubate 77°C. Plate 0.1 ml of the inoculated culture onto a non-selective medium and incubate aerobically at 37°C. Transfer 0.1 ml of the inoculated culture to a nonselective aerobic broth and incubate at 77°C.
6. Initial growth should be detected in the broth within 24 hours. For optimal growth, remove the culture from the 77C incubator and continue incubating in the dark at room temperature. Additional tubes of #2507 broth can be inoculated by transferring 0.5 mL of the primary broth to each additional tube using good anaerobic technique. Exchange the gas in the Balch tubes again after inoculation.
7. No growth should be detected on the aerobic plate or broth.

ANAEROBIC CONDITIONS:

a. Balch tubes refer to a special type of test tube that is designed to be pressurized and are suited for anaerobic work. The Balch tubes can be purchased form Bellco Glass (www.bellcoglass.com; stock no. 2048-00150).
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

The cells are rods with a sheath or toga like structure that protrudes beyond the end of the cell.

Over pressurizing the culture by 1-2 atm. promotes growth.

Avoid using gases with hydrogen. Hydrogen has been reported to inhibit the growth of *Thermotoga*.

Growth can be detected with the increase of turbidity or microscopically at 1000X. After growth is achieved, remove the culture and place it at room temperature. If kept at high temperatures after growth, cell lysis will occur.

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: *Thermotoga neapolitana* Jannasch et al. (ATCC 49049)

References

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

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