Product Sheet

Hyperthermus butylicus Zillig et al. 700455™

Description

Strain designation: DSMZ 5456 **Deposited As:** *Hyperthermus butylicus* Zillig et al. **Type strain:** Yes

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 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.

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 80% H_2 and 20% $CO_2.$ Do not overpressurize at this time.

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 10 to 20 minutes - until the resazurin becomes colorless - before inoculating.

4. When the Balch tube is ready to inoculate, thaw contents of the vial in a 37°C water bath with gentle agitation.

5. For inoculation, use an anaerobic 1.0 ml syringe (*see below*) tipped with 22 gauge needle. Withdraw entire contents of the thawed vial and transfer to the Balch tube containing *Hyperthermus butylicus* broth.

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6. Plate 0.1 ml of the inoculated culture onto a non-selective medium and incubate aerobically at 37°C. Additional tubes of prepared anaerobic broth may be inoculated from the primary broth tube. After inoculation, pressurize the tubes to 1 bar overpressure with 80% H_2 and 20% CO_2 gas mixture. Incubate the inoculated tubes at 90 to 100°C.

7. Growth should be detected in the broth. There should be no growth detected on the aerobic plate.

ANAEROBIC CONDITIONS:

a. Balch tube refers to a special type of test tube that is designed to be pressurized and is suited for anaerobic work. The Balch test 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

Notes

Additional information on this culture is available on the ATCC[®] web site at <u>www.atcc.org</u>.

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: *Hyperthermus butylicus* Zillig et al. (ATCC 700455)



700455

References

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

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