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Description

Strain designation: 1351

Deposited As: *Clostridium difficile* (Hall and O'Toole) Prevot

Type strain: No

Serotype: B

Toxigenic: No

Toxin genes: *cdtB* (Binary toxin) negative; *tcdA* (Toxin A) negative; *tcdB* (Toxin B) negative

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

BSL 2

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 260: Trypticase soy agar/broth with defibrinated sheep blood

ATCC Medium 2107: Modified Reinforced Clostridial

Temperature: 37°C

Atmosphere: Anaerobic

Handling Procedures

1. Open vial according to enclosed instructions.
2. Under anaerobic conditions, withdraw 0.5 mL of #2107 broth from a single test tube (5 to 6 mL), with a Pasteur or 1.0 mL pipette. Rehydrate the entire pellet.
3. Aseptically transfer this aliquot back into the broth tube. Mix well.

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4. Use several drops of the suspension to inoculate a pre-reduced Brucella blood plate and a #260 slant. An aerobic blood plate may also be streaked to check for purity.
5. Incubate the tubes and plate under anaerobic conditions at 37°C for 24-48 hours. Incubate the purity plate aerobically at 37°C. Turbid growth is evident in the broth at 24-48 hours.

ANAEROBIC CONDITIONS:

Anaerobic conditions for transfer may be obtained by either of the following:

- Use of an anaerobic gas chamber, or
- Placement of test tubes under a gassing cannula system connected to anaerobic gas.

Anaerobic conditions for incubation may be obtained by any of the following:

- Loose screw caps on test tubes in anaerobic chamber,
- Loose screw caps on test tubes in an activated anaerobic gas pack jar, or
- Use of sterile butyl rubber stoppers on test tubes so that an anaerobic gas headspace is retained.

Notes

Anaerobe Systems Brucella Blood Plates (AS-111 or AS-141) can be used to analyze colony morphology and purity. Colonies on Brucella Blood agar are low convex, irregular, and undulate. No growth should occur on the blood plate incubated aerobically.

The absence of *tcdA* and *tcdB* genes is confirmed by PCR. The binary toxin gene *cdtB* is not amplified by PCR.

Always use freshly prepared pre-reduced media or pre-reduced media that has been previously prepared but stored under anaerobic conditions. Resazurin in the media is a color indicator for anaerobic conditions. Observance of pink color in medium before use or during incubation shows anaerobic conditions have not been met and

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oxidation has occurred. Medium should be discarded.

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: 43593 (ATCC 43593)

References

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

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