ATCC

Product Sheet

Clostridium difficile (ATCC® 43255-FZ™)

Please read this FIRST

Storage Temp.
Frozen: -80°C or colder
Freeze-Dried: 2°C to 8°C
Live Culture: See Propagation Section

Biosafety Level
2

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: *Clostridium difficile* (ATCC® 43255-FZ™)

American Type Culture Collection
PO Box 1549
Manassas, VA 20108 USA
www.atcc.org

800.638.6597 or 703.365.2700
Fax: 703.365.2750
Email: Tech@atcc.org

Or contact your local distributor

Description

Designation: VPI 10463
Deposited Name: *Clostridium difficile* (Hall and O'Toole) Prevot
Genotype: Toxinotype: 0
Product Description: Used as toxinotype 0 reference strain. Ribotype 087.

Propagation

Medium
ATCC® Medium 5608:
ATCC® Medium 2107: Modified Reinforced Clostridial Agar/Broth Medium (pre-reduced)

Growth Conditions
Temperature: 37°C
Atmosphere: Anaerobic gas mixture, 80% N_2-10% CO_2-10% H_2 or 80% N_2-20% CO_2

Propagation Procedure

1. Thaw the frozen vial under anaerobic conditions.
2. Under anaerobic conditions, transfer the entire contents of the thawed vial into a single tube (5 - 6 mL) of #5608 or #2107 broth. Mix well.
3. Additional tubes of broth may be inoculated with 0.5 mL each from the suspension and 0.1 mL may be inoculated onto a slant. Streak several blood plates for colony morphology and purity.
4. Incubate the tubes and plate under anaerobic conditions at 37°C. Incubate the purity plate aerobically at 37°C. Within 24-48 hours, growth should be evident by turbidity in the broth and by colonies on the anaerobic agar surfaces. No growth should occur on agar plates incubated aerobically.

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

Colonies on anaerobic blood plates may appear erose, glistening, flat, and gray. Cells are Gram-positive straight rods. Agar growth takes between 24 to 48 hours.
Initial growth in #2107 may take between 24 to 48 hours, but after consecutive transfers, growth reaches peak viability and density between 6 and 12 hours. Better growth is observed on #5608.
The presence of both *tcdA* and *tcdB* genes have been confirmed by PCR. Toxin B production has been confirmed by cytotoxicity assay using Vero cells (Cell line # CCL-81). The binary toxin gene *cdtB* is not amplified by PCR
NOTE: Vial should be stored at -80C or lower until ready to be used. Storage in vapor phase liquid nitrogen is preferred for long term storage.
Additional information on this culture is available on the ATCC® web site at www.atcc.org.

References

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

Biosafety Level: 2

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

ATCC Warranty

The viability of ATCC® products is warranted for 30 days from the date of shipment, and is valid only if the

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product is stored and cultured according to the information included on this product information sheet. ATCC lists the media formulation that has been found to be effective for this strain. While other, unspecified media may also produce satisfactory results, a change in media or the absence of an additive from the ATCC recommended media may affect recovery, growth and/or function of this strain. If an alternative medium formulation is used, the ATCC warranty for viability is no longer valid.

Disclaimers

This product is intended for laboratory research purposes only. It is not intended for use in humans.

While ATCC uses reasonable efforts to include accurate and up-to-date information on this product sheet, ATCC makes no warranties or representations as to its accuracy. Citations from scientific literature and patents are provided for informational purposes only. ATCC does not warrant that such information has been confirmed to be accurate.

This product is sent with the condition that you are responsible for its safe storage, handling, and use. ATCC is not liable for any damages or injuries arising from receipt and/or use of this product. While reasonable effort is made to insure authenticity and reliability of strains on deposit, ATCC is not liable for damages arising from the misidentification or misrepresentation of cultures.

Please see the enclosed Material Transfer Agreement (MTA) for further details regarding the use of this product. The MTA is also available on our Web site at www.atcc.org

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

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