



Product Sheet

# *Nitrobacter sp.* (ATCC® 51919™)

Please read this **FIRST**



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

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Biosafety Level  
**1**

## 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: *Nitrobacter sp.* (ATCC® 51919™)

American Type Culture Collection  
PO Box 1549  
Manassas, VA 20108 USA  
[www.atcc.org](http://www.atcc.org)

800.638.6597 or 703.365.2700  
Fax: 703.365.2750  
Email: [Tech@atcc.org](mailto:Tech@atcc.org)

Or contact your local distributor

## Description

**Designation:** N-6; T 52A

**Deposited Name:** *Nitrobacter sp.*

## Propagation

### Medium

ATCC® Medium 480: Nitrobacter medium 203

### Growth Conditions

**Temperature:** 26°C

**Atmosphere:** Aerobic

### Propagation Procedure

1. Allow vial to thaw. Transfer the entire contents into 5.0 mL of #480 broth contained in a large test tube (20 X 150 mm) or a small flask.
2. Incubate tube at 26°C, static, in the dark, and in a slanted position to increase surface area.
3. Monitor culture growth daily by assaying for the disappearance of NO<sub>2</sub> in the culture medium: Withdraw 0.1 mL from the culture and place in a small test tube. Add one drop of Nitrate Reagent A followed by one drop of Nitrate Reagent B, mixing after each addition. Pink color development occurs within 10 minutes and is directly related to amount of NO<sub>2</sub> present. Initial tests will probably be dark magenta reflecting the 3 mM NO<sub>2</sub> concentration of medium #480; but as culture grows and NO<sub>2</sub> is oxidized, color will become magenta, pink and finally clear. This process can take up to 3 weeks depending on the viability of the stock.
4. When all NO<sub>2</sub> has been oxidized, inoculate a 250 mL flask containing 100 mL of medium #480 with the 5.0 mL tube culture. Wrap the flask in foil to protect from light and incubate at 26°C with gentle shaking. Monitor NO<sub>2</sub> concentration daily.
5. When NO<sub>2</sub> is again depleted in the culture medium, either transfer the culture to fresh media or feed the culture by adding additional NO<sub>2</sub>. A 10% inoculum is recommended for transfers. To feed cultures, aseptically add sterile NaNO<sub>2</sub> solution, returning NO<sub>2</sub> concentrations to approximately 10 mM (3.5 mL of 2% NaNO<sub>2</sub> per 100 mL culture). Cells should be harvested or subcultured to fresh media when cultures can utilize 10 mM NO<sub>2</sub> in less than 24 hours and cell numbers are greater than 10 cells per field at 1000X magnification.
6. Cells can be harvested by centrifugation. For longterm storage, 1.0 mL aliquots of concentrated cells can be frozen with glycerol as the cryoprotectant at a final concentration of 10%. Frozen vial should be stored at 70° C or below for best preservation.

## Notes

This item will not produce turbidity in broth. Growth can be detected by testing for the depletion of nitrite from the broth.

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

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function of this product. If an alternative medium formulation is used, the ATCC warranty for viability is no longer valid.

## Disclaimers

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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](http://www.atcc.org)

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

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