



Product Sheet

# *Ministeria vibrans* (ATCC® 50519™)

## Please read this FIRST



## 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: *Ministeria vibrans* (ATCC® 50519™)

## Description

**Deposited Name:** *Ministeria vibrans* Tong

**Depositor:** S Tong

**Isolation:**

seawater, Atlantic Ocean, Southampton, England, 1992

## Propagation

### Growth Conditions

**Temperature:** 25.0°C

**Protocol:** ATCCNO: 50519 SPEC: The strain is routinely distributed as a frozen stabulate. See the general instructions for thawing and storage of frozen material before proceeding. Thaw the ampule and aseptically transfer the material to a T-25 tissue culture flask containing 10 ml of ATCC medium 1525. The bacterial flora that is present in the shipped culture will support growth. Growth may be improved by using bacterized ATCC medium 1525 with a single species of bacteria. Bacterization is accomplished by inoculating the medium with *Enterobacter aerogenes* ATCC 13048 approximately 24 hours before use. Other species of bacteria may work equally well but this must be empirically assessed. The culture is routinely passaged every 14 days. This strain does not form a cyst stage and can be lost if it is not transferred regularly. To subculture, agitate and aseptically transfer 0.25 ml to 10 ml of freshly bacterized medium in a T-25 flask. Screw the cap on tightly and incubate at 25°C.

### Medium

ATCC® Medium 1525: Seawater 802 medium

### Instructions for Complete Medium

ATCC Medium 1525 inoculated with *Klebsiella pneumoniae* subsp. *pneumoniae* (ATCC® 700831).

### Culture Maintenance

Subculture every two weeks to a fresh T-25 flask of bacterized medium in the following manner:

1. Vigorously agitate the flask and aseptically transfer 0.5 ml from a growing culture to a T-25 tissue culture flask containing 10.0 ml of ATCC medium 1525 bacterized with *Klebsiella pneumoniae* subsp. *pneumoniae* (ATCC® 700831)
2. Incubate flask at 25°C with the cap on tightly.

## Cryopreservation

Cryoprotective Solution

DMSO	2.0 ml
Fresh growth medium w/o bacteria	8.0 ml

1. Mix the components in the order listed. When the medium is added to the DMSO the solution will warm up due to chemical heat.
2. Harvest cells from a culture that is at or near peak density by filtration and centrifugation at 800 x g for 5 min.
3. Adjust the concentration of cells at least  $2 \times 10^6$ /ml in fresh medium.
4. Mix the cell preparation and the cryoprotective solution in equal portions.
5. Dispense in 0.5 ml aliquots into 1.0 - 2.0 ml sterile plastic screw-capped cryules (special plastic vials for cryopreservation).
6. Place vials in a controlled rate freezing unit. From room temperature cool at -1°C/min to -40°C. If freezing unit can compensate for the heat of fusion, maintain rate at -1 C/min through heat of fusion. At -40°C plunge ampules into liquid nitrogen. Alternatively, place the vials in a Nalgene 1°C freezing apparatus. Place the apparatus at -80°C for 1.5 to 2 hours and then plunge ampules into liquid nitrogen. (The cooling rate in this apparatus is approximately -1°C/min.)
7. Ampules are stored in either the vapor or liquid phase of a nitrogen refrigerator.
8. To establish a culture from the frozen state place the vial in a 35°C water bath. Immerse the vial to a level just above the surface of the frozen material. Do not agitate the vial. Immediately after thawing, do not leave in water bath, aseptically remove the contents of the ampule and inoculate into a T-25 tissue culture flask containing 10 ml ATCC medium 1525 bacterized with *Klebsiella pneumoniae* subsp. *pneumoniae* (ATCC® 700831).
9. Incubate at 25°C with the cap screwed on tightly.
10. Once the culture is established, vigorously agitate the flask and aseptically transfer 0.5 ml to 10.0 ml of

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bacterized ATCC medium 1525.

11. Follow the protocol for maintenance of culture.

## 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.

## ATCC Warranty

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

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

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