



Product Sheet

# *Methanospirillum hungatei* (ATCC® 27890™)

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: *Methanospirillum hungatei* (ATCC® 27890™)

## Description

**Designation:** JF-1 [DSM 864]

**Deposited Name:** *Methanospirillum hungatei* Ferry et al.

**Product Description:** Type strain. Genome sequenced strain.

## Propagation

### Medium

ATCC® Medium 2487: MS-OCM Base Medium with 43 mM NaCl and 5 mM sodium acetate

### Growth Conditions

**Temperature:** 37°C

**Atmosphere:** Anaerobic gas mixture, 80% H<sub>2</sub>-20% CO<sub>2</sub>; 100% nitrogen if formate is used as the substrate

### Propagation Procedure

1. Sterilize the top of the Balch tube by spraying it with 70% ethanol and then flaming the top. Formate can be added to a final concentration of 100 mM to individual tubes of Medium #2487 or to the medium before it is tubed. When formate is used as the substrate 100% nitrogen can be used as the gas.
2. If needed, exchange the gas in the test tube for 80% H<sub>2</sub>-20% CO<sub>2</sub>.
3. If the medium is pink (see discussion about resazurin) add 0.1 mL Na<sub>2</sub>S<sub>9</sub>H<sub>2</sub>O (1.5% sodium sulfide, stock solution) per 10.0 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 the frozen vial at room temperature under a gentle stream of oxygen-free gas.
5. For inoculation, use an anaerobic (see "c" below) 1.0 mL syringe tipped with 22-gauge needle, withdraw the cell suspension from the vial and transfer it to the broth. Plate 0.1 mL of the inoculated culture onto a non-selective medium and incubate aerobically at 37°C. Use 0.1 mL of the inoculated culture to inoculate a nonselective aerobic broth and an additional tube of #2487 broth. Incubate the non-selective aerobic broth tubes at 37°C. Incubate the anaerobic tube at 37°C.
6. Growth should be detected in the #2487 broth within 24 to 72 hours. There should be no growth detected on the aerobic plate or in the aerobic broth.

### ANAEROBIC CONDITIONS:

- a. 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.
- b. 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.
- c. Syringes can be made anaerobic by one of two methods.
  1. Displace the dead space in the syringe with a sterile oxygen-free gas.
  2. Displace the dead space in the syringe with a reducing agent.

## Notes

Formate: Filter-sterilize a 2 M formate solution and put in a sealed sterile Balch tube. Exchange air in the head space with 100% nitrogen. Formate can be added to a final concentration of 100 mM.

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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product is stored and handled according to the information included on this product information sheet. If the ATCC® product is a living cell or microorganism, ATCC lists the media formulation that has been found to be effective for this product. 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 product. 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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