Designation: H-2 [DSM 13691]
Deposited Name: Geobacter hydrogenophilus Coates et al.

Medium
ATCC® Medium 1957: Geobacter medium

Growth Conditions
Temperature: 30.0°C
Atmosphere: Anaerobic

Propagation Procedure
1. Sterilize the top of the Balch tube by spraying it with 70% ethanol and then flaming the top.
2. If needed exchange the gas in the test tube for 80% N₂ 20% CO₂.
3. If the medium is pink (see discussion about resazurin) add 2.0 ml of reducing agent (3% cysteine, stock solution) per 100 ml of medium. Let the medium sit at room temperature for 10 to 20 minutes, until the resazurin becomes colorless, before inoculating.
4. After the Balch tube is ready to be inoculated let the frozen vial thaw at room temperature under a gentle stream of sterile oxygen free gas.
5. Using a 1.0 ml syringe tipped with 22 gauge needle, withdraw the entire contents of the thawed vial and transfer it to a tube of #1957 broth and incubate at 30°C for 4-5 days. Plate 0.1 ml of the inoculated culture onto a non-selective medium and incubate aerobically at 30°C.

ANAEROBIC CONDITIONS:
· 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.
· 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.
· 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.

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**Biosafety Level**

1

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**Citation of Strain**

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: *Geobacter hydrogenophilus* (ATCC® 51590™)