Designation: 335 [CCUG 1776, CIP 104763, DSM 531, ICPB 3984, JCM 11282, NCCB 82041, NCIB 11842, NRRL B-14690, NRRL B-4383, R-10-e]

Deposited Name: Hydrogenomonas eutropha Wittenberg and Repaske

Product Description: Type strain. Formerly the type strain of Wautersia eutropha. Produces poly-D-3-hydroxybutyric acid from carbon dioxide.

Medium
ATCC® Medium 3: Nutrient agar or nutrient broth

Growth Conditions
Temperature: 26°C
Atmosphere: Aerobic

Propagation Procedure
1. Open vial according to enclosed instructions.
2. From a single tube of #3 broth (5 to 6 mL), withdraw approximately 0.5 to 1.0 mL with a Pasteur or 1.0 mL pipette. Rehydrate the entire pellet.
3. Aseptically transfer this aliquot back into the broth tube. Mix well.
4. Use several drops of the suspension to inoculate a second tube of broth, a slant and/or plate.
5. Incubate all tubes and plate at 26°C for 24 hours.

Notes
Two colony types are observed on #3 plates; one small, one large. Both are opaque, circular with entire margins.
This strain may be tested for autotrophic growth in Medium #1246 (see formula below) with the gas mixture indicated:

KH$_2$PO$_4$, 2.3 g
Na$_2$HPO$_4$·2H$_2$O, 2.9 g
NH$_4$Cl, 1.0 g
MgSO$_4$·7H$_2$O, 0.5 g
NaHCO$_3$, 0.5 g
CaCl$_2$·2H$_2$O, 0.01 g
Ferric ammonium citrate, 0.05 g

Distilled water, 980.0 mL
Agar (if needed), 15.0 g

Sterilize ferric ammonium citrate (0.05 g in 20 mL H$_2$O) separately and then add to basal medium. Incubate at 30°C under a gas mixture of 60% H$_2$, 10% CO$_2$, 25% N$_2$, 5% O$_2$.

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: 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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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
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: Cupriavidus necator (ATCC® 17697™)