

## **Certificate of Analysis for NR-50499**

## Escherichia coli, Strain AGT01

## Catalog No. NR-50499

**Product Description:** Escherichia coli (E. coli), strain AGT01 is a flagellin (fliC) insertion mutant of the wild type strain E2348/69. Strain E2348/69 was isolated in 1969 during an outbreak of diarrhea in an infant nursery in Taunton, England.

Lot<sup>1</sup>: 2236 Manufacturing Date: 30NOV2016

TEST	SPECIFICATIONS	RESULTS
Phenotypic Analysis Cellular morphology Colony morphology <sup>2</sup> Motility <sup>3</sup> VITEK® MS (MALDI-TOF)	Gram-negative rods Report results  Non-motile E. coli	Gram-negative rods Circular, low convex, entire, smooth and cream (Figure 1)  Motile <sup>4</sup> E. coli (99.9%)
Antibiotic Susceptibility Profile <sup>5</sup> HardyDisk <sup>TM</sup> AST Disks <sup>6</sup> Chloramphenicol (30 μg) Kanamycin (30 μg) Nalidixic Acid (30 μg) Tetracyline (30 μg)	Resistant (≤ 12 mm) Susceptible (≥ 18 mm) Resistant (≤ 13 mm) Susceptible (≥ 15 mm)	Resistant (6 mm) Susceptible (25 mm) Resistant (6 mm) Susceptible (24 mm)
Genotypic Analysis Sequencing of 16S ribosomal RNA gene (~ 1480 base pairs) Riboprinter® Microbial Characterization System	≥ 99% sequence identity to <i>E. coli</i> strain E2348/69 (GenBank: NC_011601.1) <i>E. coli</i> (≥ 0.85)	99.6% sequence identity to <i>E. coli</i> strain E2348/69 (GenBank: NC_011601.1) <i>E. coli</i> (0.93)
PCR Amplification of Genetic Targets from Extracted DNA  Translocated intimin receptor (tir)  Wild type tir  Atir  S-ribosylhomocysteinase (luxS)  Wild type luxS  AluxS  Transcriptional regulator (gadX)  Wild type gadX  AgadX  Type III secretion system ATPase (escN)  Wild type escN  AescN  Flagellin (fliC)  Wild type fliC  AfliC  Plasmid pMAR2 (bfpD)	~ 1620 base pair amplicon ~ 2260 base pair amplicon ~ 1310 base pair amplicon ~ 2590 base pair amplicon ~ 1300 base pair amplicon ~ 2200 base pair amplicon ~ 710 base pair amplicon ~ 1560 base pair amplicon ~ 1560 base pair amplicon ~ 2200 base pair amplicon ~ 840 base pair amplicon	~ 1620 base pair amplicon  ~ 1310 base pair amplicon  ~ 1300 base pair amplicon  ~ 710 base pair amplicon  ~ 2200 base pair amplicon <sup>3</sup> ~ 840 base pair amplicon
Purity (post-freeze) <sup>7</sup>	Growth consistent with expected colony morphology	Growth consistent with expected colony morphology
Viability (post-freeze) <sup>2</sup>	Growth	Growth

<sup>&</sup>lt;sup>1</sup>NR-50499 was produced by inoculation of the deposited material into Tryptic Soy broth and grown for 1 day at 37°C in an aerobic atmosphere. Broth inoculum was added to Tryptic Soy agar kolles, which were grown for 1 day at 37°C in an aerobic atmosphere to produce this lot.

BEI Resources
www.beiresources.org

E-mail: contact@beiresources.org

Tel: 800-359-7370 Fax: 703-365-2898

<sup>&</sup>lt;sup>2</sup>1 day at 37°C in an aerobic atmosphere on Tryptic Soy agar

<sup>&</sup>lt;sup>3</sup>Motility test performed in Remel™ Motility Test Medium with TTC Indicator for 1 day at 37°C in an aerobic atmosphere.

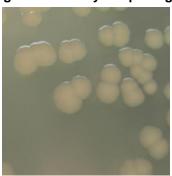


## Certificate of Analysis for NR-50499

<sup>4</sup>E. coli, strain AGT01 was deposited as non-motile due to an insertion of a chloramphenicol-resistance gene (cat) cassette into fliC. Functional inactivation of fliC was confirmed by the depositor using LB motility plates. BEI Resources confirmed the presence of the insertion by PCR, but could not recapitulate the non-motile phenotype using motility test medium with TTC indicator. The different medias used may be responsible for the differences in observed motility. For more information, please refer to Giron, J. A., et al. "The Flagella of Enteropathogenic Escherichia coli Mediate Adherence to Epithelial Cells." Mol. Microbiol. 44 (2002): 361-379. PubMed: 11972776.

<sup>5</sup>Minimum Inhibitory Concentration (MIC); MIC Interpretation Guideline: CLSI M100-S26 (2016)

Figure 1: Colony Morphology



Date: 28 AUG 2017 Signature:

**BEI Resources Authentication** 

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC®'s knowledge.

ATCC® is a trademark of the American Type Culture Collection.

You are authorized to use this product for research use only. It is not intended for human use.

BEI Resources www.beiresources.org E-mail: <a href="mailto:contact@beiresources.org">contact@beiresources.org</a>
Tel: 800-359-7370

Fax: 703-365-2898

<sup>61</sup> day at 37°C in an aerobic atmosphere on Mueller Hinton agar

Purity of this lot was assessed for 7 days at 37°C in an aerobic atmosphere with 5% CO<sub>2</sub> on Tryptic Soy agar with 5% defibrinated sheep blood.