NEW ENGLAND BioLabs Inc.

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New England Biolabs
Certificate of Analysis

Product Name: *SHuffle® T7 Express lysY Competent E. coli*
Catalog #: *C3030J*
Lot #: *0071703*
Assay Date: *03/2017*
Expiration Date: *03/2018*
Storage Temp: *-80°C*
Specification Version: *PS-C3030J v1.0*
Effective Date: *06 Apr 2017*

Assay Name/Specification (minimum release criteria)	**Lot #0071703**
Antibiotic Resistance (Chloramphenicol) - 15 µl of untransformed Shuffle® T7 Express lysY Competent *E. coli* streaked onto a LB or Rich Broth plate containing Chloramphenicol will form colonies after incubation for 16 hours at 37°C.	**Pass**
Antibiotic Resistance (Nitrofurantoin) - 15 µl of untransformed Shuffle® T7 Express lysY Competent *E. coli* streaked onto a LB or Rich Broth plate containing Nitrofurantoin will form colonies after incubation for 16 hours at 37°C.	**Pass**
Antibiotic Resistance (Spectinomycin) - 15 µl of untransformed Shuffle® T7 Express lysY Competent *E. coli* streaked onto a LB or Rich Broth plate containing Spectinomycin will form colonies after incubation for 16 hours at 37°C.	**Pass**
Antibiotic Resistance (Streptomycin) - 15 µl of untransformed Shuffle® T7 Express lysY Competent *E. coli* streaked onto a LB or Rich Broth plate containing Streptomycin will form colonies after incubation for 16 hours at 37°C.	**Pass**
Antibiotic Sensitivity (Ampicillin) - 15 µl of untransformed Shuffle® T7 Express lysY Competent *E. coli* streaked onto a LB or Rich Broth plate containing Ampicillin will not form colonies after incubation for 16 hours at 37°C.	**Pass**
Antibiotic Sensitivity (Kanamycin) - 15 µl of untransformed Shuffle® T7 Express lysY Competent *E. coli* streaked onto a LB or Rich Broth plate containing Kanamycin will not form colonies after incubation for 16 hours at 37°C.	**Pass**
Antibiotic Sensitivity (Tetracycline) - 15 µl of untransformed Shuffle® T7 Express lysY Competent *E. coli* streaked onto a LB or Rich Broth plate containing Tetracycline will not form colonies after incubation for 16 hours at 37°C.	**Pass**

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Functional Testing (Disulfide Bond Formation) - The nuclease NucA requires disulfide bonds for its stability. When expressed at 37°C in *E. coli*, NucA is toxic to cells only in its oxidized disulfide-bonded state. Transformation of SHuffle® T7 Express lysY Competent *E. coli* using 100 pg of plasmid that expresses a MBP-NucA fusion results in < 1% of the colonies when compared to a control transformation of its wild type parent strain NEB 10-beta.	**Pass**
Phage Resistance (Φ 80) - 15 µl of untransformed Shuffle® T7 Express lysY Competent *E. coli* streaked onto a Rich Broth plate does not support plaque formation by phage Φ 80 after incubation for 16 hours at 37°C.	**Pass**
Transformation Efficiency - 50 µl of SHuffle® T7 Express lysY Competent *E. coli* cells were transformed with 100 pg of pUC19 DNA using the transformation protocol provided. Incubation overnight on LB-Ampicillin plates at 37°C resulted in >1 x 10e7 cfu/µg of DNA.	**Pass**

Authorized by
Quiting Ren
06 Apr 2017

Inspected by

ISO 9001 Registered Quality Management
ISO 14001 Registered Environmental Management
ISO 13485 Registered Medical Devices