NEW ENGLAND BioLabs Inc.

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New England Biolabs
Certificate of Analysis

Product Name: *SHuffle® Express Competent E. coli*
Catalog #: *C3028J*
Lot #: *0081803*
Assay Date: *03/2018*
Expiration Date: *03/2019*
Storage Temp: *-80°C*
Specification Version: *PS-C3028J v1.0*
Effective Date: *18 Sep 2017*

Assay Name/Specification (minimum release criteria)	**Lot #0081803**
Antibiotic Resistance (Nitrofurantoin) - 15 µl of untransformed SHuffle® Express 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® Express 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® Express 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® Express 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 (Chloramphenicol) - 15 µl of untransformed SHuffle® Express Competent *E. coli* streaked onto a LB or Rich Broth plate containing Chloramphenicol will not form colonies after incubation for 16 hours at 37°C.	**Pass**
Antibiotic Sensitivity (Kanamycin) - 15 µl of untransformed SHuffle® Express 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® Express 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**
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® Express 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**

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Assay Name/Specification (minimum release criteria)	**Lot #0081803**
Phage Resistance (Φ 80) - 15 µl of untransformed SHuffle® Express 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® Express 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
Derek Robinson
18 Sep 2017

Inspected by
Quiting Ren
19 Apr 2018

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