New England Biolabs
Product Specification

Product Name: EnGen® Spy dCas9 (SNAP-tag®)
Catalog #: M0652S
Concentration: 1 µM
Unit Definition: N/A
Shelf Life: 24 months
Storage Temp: -20°C
Storage Conditions: 10 mM Tris-HCl, 300 mM NaCl, 1 mM DTT, 0.1 mM EDTA, 50 % Glycerol, (pH 7.4 @ 25°C)
Specification Version: PS-M0652S v1.0
Effective Date: 09 Aug 2017

Assay Name/Specification (minimum release criteria)

Endonuclease Activity (Nicking) - A 50 µl reaction in NEBuffer 3.1 containing 1 µg of supercoiled PhiX174 DNA and a minimum of 1 pmol of EnGen® Spy dCas9 (SNAP-tag®) incubated for 4 hours at 37°C results in <10% conversion to the nicked form as determined by agarose gel electrophoresis.

Exonuclease Activity (Radioactivity Release) - A 50 µl reaction in NEBuffer 3.1 containing 1 µg of a mixture of single and double-stranded [³H] E. coli DNA and a minimum of 1 pmol of EnGen® Spy dCas9 (SNAP-tag®) incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.

Functional Testing (EnGen® Spy dCas9 (SNAP-tag®), Gel Shift Assay) - A 20 µl reaction in 1X NEBuffer 3.1 containing 20 nM 100 bp FAM labeled double stranded target DNA, 20 nM TAMRA-labeled off target DNA, 100 nM sgRNA and 100 nM EnGen® Spy dCas9 (SNAP-tag®) incubated for 15 minutes at 37°C results in ≥90% binding of the substrate DNA as determined by electrophoretic mobility shift assay.

Non-Specific DNase Activity (16 Hour) - A 50 µl reaction in NEBuffer 3.1 containing 1 µg of Lambda DNA and a minimum of 1 pmol of EnGen® Spy dCas9 (SNAP-tag®) incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis.

Protein Purity Assay (SDS-PAGE) - EnGen® Spy dCas9 (SNAP-tag®) is ≥95% pure as determined by SDS-PAGE analysis using Coomassie Blue detection.

RNase Activity (Extended Digestion) - A 10 µl reaction in NEBuffer 4 containing 40 ng of a 300 base single-stranded RNA and a minimum of 1 pmol of EnGen® Spy dCas9 (SNAP-tag®) is incubated at 37°C. After incubation for 16 hours, >90% of the substrate RNA remains intact as determined by gel electrophoresis using fluorescent detection.

Derek Robinson
Director of Quality Control

Date 09 Aug 2017