

## New England Biolabs Certificate of Analysis

**Product Name:** AlwNI  
**Catalog Number:** R0514L  
**Concentration:** 10,000 U/ml  
**Unit Definition:** One unit is defined as the amount of enzyme required to digest 1 µg of Lambda DNA in 1 hour at 37°C in a total reaction volume of 50 µl.  
**Lot Number:** 10028216  
**Expiration Date:** 11/2020  
**Storage Temperature:** -20°C  
**Storage Conditions:** 50 mM NaCl, 10 mM Tris-HCl (pH 7.4), 1 mM DTT, 0.1 mM EDTA, 50% Glycerol, 200 µg/ml BSA  
**Specification Version:** PS-R0514S/L v1.0

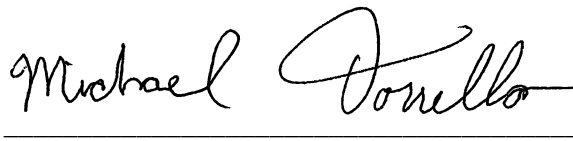
| AlwNI Component List |                       |            |                      |
|----------------------|-----------------------|------------|----------------------|
| NEB Part Number      | Component Description | Lot Number | Individual QC Result |
| R0514LVIAL           | AlwNI                 | 10028217   | Pass                 |
| B7204SVIAL           | CutSmart® Buffer      | 10021120   | Pass                 |

| Assay Name/Specification  | Lot # 10028216 |
|---|----------------|
| <b>Exonuclease Activity (Radioactivity Release)</b><br>A 50 µl reaction in CutSmart™ Buffer containing 1 µg of a mixture of single and double-stranded [ <sup>3</sup> H] E. coli DNA and a minimum of 100 units of AlwNI incubated for 4 hours at 37°C releases <0.1% of the total radioactivity.   | Pass           |
| <b>Ligation and Recutting (Terminal Integrity)</b><br>After a 20-fold over-digestion of Lambda DNA with AlwNI, >95% of the DNA fragments can be ligated with T4 DNA ligase in 16 hours at 16°C. Of these ligated fragments, >95% can be recut with AlwNI.   | Pass           |
| <b>Non-Specific DNase Activity (16 Hour)</b><br>A 50 µl reaction in CutSmart™ Buffer containing 1 µg of Lambda DNA and a minimum of 10 Units of AlwNI incubated for 16 hours at 37°C results in a DNA pattern free of detectable nuclease degradation as determined by agarose gel electrophoresis. | Pass           |
| <b>Protein Purity Assay (SDS-PAGE)</b><br>AlwNI is >95% pure as determined by SDS PAGE analysis using Coomassie Blue detection.   | Pass           |

This product has been tested and shown to be in compliance with all specifications.



Tony Spear-Alfonso  
Production Scientist  
02 Nov 2018



Michael Tonello  
Packaging Quality Control Inspector  
27 Nov 2018