

Recombinant BirA Biotin Ligase

Catalog # BBL-301

PRODUCT DESCRIPTION AND APPLICATIONS

Product Details:

Recombinant BirA (amino acids 3 - 321) with the N-therminal His-tag, MW = 36.2 kDa, expressed in an E. coli cell expression system.

E. coli biotin ligase BirA (EC 6.3.4.15) activates biotin to form biotinyl 5'-adenylate and transfers the biotin to biotin-accepting proteins and peptides. The natural substrate of BirA is the Biotin Carboxyl Carrier Protein (BCCP), requiring fusion of at least 75 residues to the target protein. However, several shorter peptide sequences have been described for BirA-mediated biotinylation such as 15 amino acids in length AviTag™ peptide (GLNDIFEAQKIEWHE). BirA site-specifically biotinylates a lysine side chain within this peptide. AviTag works at either the N or C terminus of the target protein.

Applications: In vitro biotinylation of proteins and peptides. The binding between biotin and streptavidin or avidin is one of the strongest known non-covalent biological interactions. The (strept)avidin-biotin interaction has been widely used for decades in biological research and biotechnology. Labeling of purified proteins by biotin is a powerful way to achieve protein capture, immobilization, and functionalization, as well as multimerizing or bridging molecules. Enzymatic biotinylation with BirA is more specific than chemical biotinylation which often generates heterogeneous products that may have impaired function.

The amount of BirA Ligase to add to the reaction mixture may need to be varied and optimal concentrations should be determined by the end user.

Definition of Activity: 1 Unit is the amount of enzyme that will biotinylate 1 pmol of AviTag peptide substrate in 30 minutes at 30°C in the reaction buffer containing 10 mM Tris-HCl, pH 8.0, 10 mM ATP, 10 mM MgOAc, 50 μ M d-Biotin, and 38 μ M peptide substrate.

Storage buffer: 50 mM Tris-HCl, pH 7.5, 50 mM NaCl, 3 mM DTT, and 50% Glycerol.

Concentration: 1.0 mg/mL by A280 Purity: >95% by Coomassie staining

Activity: 5,000 Units/µg

Storage is recommended at -20°C for longer periods of time.

Usage: This product is intended for laboratory research use only.