CREATIVE BIOMART®
Assay Kit

Midi Protein Expression Kit

Product Information

Cat

Kit-0884

Common Name

Protein

Cat.No. Kit-0884

Product Overview

Midi Protein Expression Kit is designed for in vitro transcription and translation of target DNA to protein in a single reaction. This product contains an optimized E.coli extract containing T7 RNA polymerase for transcription and ribosome for translation. An optimized Master mix provides all other required components, including amino acids, rNTPs, and appropriate salts to express high-level production of recombinant proteins. This product contains reagents for expression of protein directly from a variety of DNA templates, includingT7 promoter and terminator as well as RBS (ribosomal binding site). This product can be used to express a wide range of target proteins and has the capacity for a yield of more than 300 μg/mL of protein within 3 hours

Applications

This product can be used to express a wide range of target proteins and has the capacity for a yield of more than 300 μg/mL of protein within 3 hours.

Storage

Store kit at -20°C ~ - 70°C.

Size

1 mL x 5 reactions

Kit Components

Master mix: 1.2 mL × 2 tube;
E.coli extract: 0.28 mL × 5 tube;
DEPC DW: 1 mL × 2 tube;

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Positive Control DNA: 1 tube.

Features & Benefits

High-speed: synthesize your target protein quickly (within 3 hours) and economically
Easy-to-Use: includes all necessary components for transcription and translation
Flexible: synthesize proteins from various DNA templates.
synthesize proteins of toxic proteins
incorporate non-natural amino acids
use additives such as metal ions, detergents, cofactors, binding partners

Assay Protocol

1. Thaw components in the Midi Protein Expression kit on ice.
2. After thawing, spin-down the the Master mix and E.coli extract, and then place the tube on ice bath.
3. Prepare the reaction mixture (one reaction).
Negative: Master mix 21 µL; E.coli extract 12 µL; DEPC DW 12 µL; Total 45 µL.
Positive: Master mix 21 µL; E.coli extract 12 µL; DNA template 1µL; DEPC DW 11 µL; Total 45 µL.
Sample: Master mix 470 µL; E.coli extract 270 µL; DNA template X µL; DEPC DW (260-X) µL; Total 1000 µL.
4. Mix reaction mixture by tapping the tube gently and spin down briefly to collect the mixture at the bottom of the tube.
5. Incubate the reaction mixture at 30°C in a water bath for 3 hours.
6. After completing incubation, spin down briefly to collect the mixture at the bottom of the tube.
7. Analyze the mixture using SDS-PAGE, western blotting or bioactivity assay.

Analysis

After expression of proteins using Midi Protein Expression Kit, you can the expression of target protein via SDSPAGE
1. Prepare loading samples.
5 µL of expression sample + 10 µL of D.W. + 5 µL of 4x Loading dye (Final volume is 20 uL)
2. Denature all samples at 95°C for 5 min.
3. Load 5 µL of each samples on SDS-PAGE minigel.

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4. Run SDS-PAGE gel.
5. Perform staining with coomassie blue R-250.
   Ex) Result of positive control (28kDa)
   -condition : 12% SDS-PAGE gel, at 50 V for 20 min→at 150 V for 50 min

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