T4 DNA Ligase Reaction Version 2

New England Biolabs

Abstract

This is the reaction for the "Ligation Protocol with T4 DNA Ligase"

Citation: New England Biolabs T4 DNA Ligase Reaction. **protocols.io**
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Protocol

Step 1.

2 µl of T4 DNA Ligase Buffer (10X)

AMOUNT
2 µl Additional info:

REAGENTS
T4 DNA Ligase M0202 by New England Biolabs

Step 2.

Vector DNA (4 kb) 50 ng (0.020 pmol)

AMOUNT
50 ng Additional info:

NOTES
Ben Claywell 17 Jul 2015
Use NEBioCalculator to determine concentration
Low Sin Yee 23 Jul 2015
recommended vector concentration 50ng=0.05ug
my linearized vector concentration= 5ug/ml=0.005ug/ul
1ul vector=0.005ug
how many ul of vector to make up 0.05ug?
(0.05ugx1ul)/0.005ug=10ul
thus, 10ul of vector should be added into ligation reaction.

Step 3.

Insert DNA (1 kb) 37.5 ng (0.060 pmol)

AMOUNT
38 ng Additional info:

NOTES
Ben Claywell 17 Jul 2015
Use NEBioCalculator to determine concentration

Step 4.
Nuclease-free water to 20 µl

Step 5.
T4 DNA Ligase, 1 µl

AMOUNT
1 µl Additional info:

REAGENTS
T4 DNA Ligase - 20,000 units M0202S by New England Biolabs

NOTES
mehrdad alirezaei 17 Jul 2015
T4 DNA Ligase - 100,000 units
Catalog #: M0202M