



Oct 24, 2020

Gibson Assembly

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ABSTRACT

This protocol includes two different kits. Choose one to use.

DOI

dx.doi.org/10.17504/protocols.io.bnwtmfen

PROTOCOL CITATION

Zhujun Wei 2020. Gibson Assembly. **protocols.io**
<https://dx.doi.org/10.17504/protocols.io.bnwtmfen>

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CREATED

Oct 24, 2020

LAST MODIFIED

Oct 24, 2020

PROTOCOL INTEGER ID

43699

MATERIALS TEXT

The old kit: Trelief™ SoSoo Cloning Kit Ver.2
The new kit: ClonExpress®II One Step Cloning Kit

SAFETY WARNINGS

Please wear gloves for the experiment.

ABSTRACT

This protocol includes two different kits. Choose one to use.

- 1 Set up a small box with ice, put DNA and Gibson Mix into it before going into the metal bath or the Bio-rad S1000TM thermocycler cyclers.
- 2 Add the following reagent to a PCR tube (4 µl).

| | |
|------------|------|
| Gibson Mix | 2 µl |
|------------|------|

| | |
|--------|-----------|
| insert | X μ l |
| vector | Y μ l |

$X+Y=2$ μ l, $X:Y=3:1\sim5:1$.

Temperature : Time= 50°C : 15-30 min

- 3 Set up a small box with ice, put DNA and Gibson enzyme and Gibson buffer into it before going into the metal bath or the Bio-rad S1000TM thermocycler cyclers.

- 4 Add the following reagent to a PCR tube (10 μ l).

| | |
|---------------|-----------|
| Gibson enzyme | 1 μ l |
| Gibson buffer | 2 μ l |
| insert | X μ l |
| vector | Y μ l |

$X=0.02 \times \text{vector length} / \text{vector concentration}$

$Y=0.02 \times \text{insert length} / \text{insert concentration}$

Temperature : Time= 37°C : 30-60 min