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Gibson Assembly

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ABSTRACT

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

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.

The old kit

- 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 .
- 2 Add the following reagent to a PCR tube (4 µl).

Gibson Mix	2 µl
insert	X µl
vector	Y µl

$X+Y=2\text{ ul}$, $X : Y=3 : 1\sim 5 : 1$.

Temperature : Time= 50°C : 15-30 min

The new kit

- 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 .

4 Add the following reagent to a PCR tube (10 µl).

Gibson enzyme	1 µl
Gibson buffer	2µl
insert	X µl
vector	Y ul

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

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

Temperature : Time= 37°C : 30-60 min



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