Protocol for colony PCR

1.prepare the primer working solution, put Forward primer 1μl and Reverse primer 1μl into 8 μl ddH_2O

2.One kind of system(10μl):

Taq master mix: 5μl

Primer working solution: 0.1μl

DNA template: 0.1μl

ddH_2O: 4.8μl

2.Another kind of system(10μl):

Hifi enzyme: 0.1μl

Primer working solution: 0.1μl

DNA template: 0.1μl

Hifi buffer: 1μl

dNTP: 1μl

ddH_2O: 7.7μl

3.choose one kind of the two system above and mix them all together.

After preparation of the reaction mix, pick a single colony and dip in the mix. The reaction condition settings are as follows:

94℃ 30s

55℃ 30s

72℃ 1kb/min

Cylces: 30-35