

Product Data Sheet

Product Name: Ultra One Step Cloning Kit
Cat. No.: GK10033

Components

Components	20 rxns	40 rxns
2X Cloning Mix	100 µL	2 * 100 µL
pUC19 Control Plasmid, Linearized (Ampr, 50 ng/µL)	5 µL	2 * 5 µL
500 bp Control Fragment (20 ng/µL)	5 µL	2 * 5 µL

Stored at -20°C, valid for 2 years. Avoid repeated freeze-thaw cycles.

Protocol

1. Pre-PCR

1. Prepare PCR reaction mix containing Taq polymerase, dNTPs, and primers. Add 1 µL of linearized plasmid and 1 µL of control fragment to the reaction mix.

2. PCR

a. Perform PCR with 5' primer and 15-25bp control fragment.

b. Purify

5'-[15-25bp]+[PCR product]+[3']

c. Purify

5'-[15-25bp]+[PCR product]+[3']

3. Transformation

a. Transform 5' PCR product into competent cells. Add 1 µL of 15-25bp control fragment to the transformation mix.

b. Plate

Caution: Product has not been fully validated for medical applications. For research use only.
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Address: 10292 Central Ave. #205, Montclair, CA, USA

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5'-[15-25bp]+[]+[]-3'

c. []

5'-[15-25bp]+[]+[]-3'

d. []3' [15-25bp] []5' []5' [15-25bp] []3' []
[]15-25bp []

2. []PCR []

[]

[1] [15-25bp] GC [40%-60%] []

[2] []

[3] PCR [] PCR [0.1-1ng] []

3. []

[1] []

[] NanoDrop [] Onedrop [] Qubit [] PicoGreen [] A260/A280 []
[1.8-2.0] []

[2] []

a. [] 1:2 [] 0.03pmol [] 0.06pmol []
[] 20ng []

[]

b. [] 10ng [] 1:1 []

[] 10ng [] 10ng []

c. [] PCR [] DNA [] 1/5 [] 2μl []
[] ([]) []

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2. 将反应液加入含有琼脂糖的离心管中，离心后观察条带。

3. 将反应液加入含有琼脂糖的离心管中，离心后观察条带。37°C 1-3h (200-220r/min) 离心后观察条带。

注意：

1. 模板 DNA 长度 15-25bp，GC 含量 40%-60%。
2. 模板 DNA 浓度 1-5 μg，模板 DNA 浓度 > 10⁸ cfu/μg。
3. 模板 DNA 浓度 1/5。
4. 模板 DNA 浓度 > 10⁸ cfu/μg。

Background

Ultra One Step Cloning Kit 是一种基于 PCR 的克隆试剂盒，用于将 PCR 产物直接插入质粒载体中。该试剂盒包含所有必要的酶和试剂，操作简便，效率高。

Ultra One Step Cloning Kit 包含 Exnase mix 1-5，用于将 PCR 产物直接插入质粒载体中。

注意：

1. 反应温度 50°C，反应时间 5-30min。
2. 模板 DNA 浓度 1-5 μg，模板 DNA 浓度 > 10⁸ cfu/μg。
3. 模板 DNA 长度 50bp-10kb，GC 含量 90%。
4. 模板 DNA 浓度 > 10⁸ cfu/μg。

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