

PCR AMPLIFICATION OF PHAGE STOCKS

1. Set up reactions in 0.5 ml tubes:

0.2 μg primers

5 μl **10 x SM PCR buffer**

5 μl phage stock

1 μl 10mM dNTPs

0.5 units Taq polymerase

up to 50 μl with dH_2O

2. Cover reactions with 50 μl mineral oil.

3. Amplify

1 cycle	95°C 1'	55°C 15 sec	72°C 3'
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35 cycles	95°C 15 sec	55°C 15 sec	72°C 3'
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1 cycle	95°C 15 sec	55°C 15 sec	72°C 6'
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4. Extract oil with 50 μl chloroform.

5. Load 2 μl of reaction on gel to check.

10 x SM PCR

40 mM KCl

10 mM Tris pH8.8

0.1% triton