Quick Protocol

SuperPhi DNA Amplification Kit

Cat#: Phi100 or Phi1000

FOR RESEARCH USE ONLY

SuperPhi DNA Amplification Kit is intended for molecular biology use and *in vitro* use only. This product is not intended for diagnosis, prevention or treatment of a disease in human beings or animals.

Store Kit at -80°C on Receipt

Protocol Review

SuperPhi DNA Amplification Kit has capacity to detect as low as 1 pictogram of template DNA under optimal condition. 0.2~0.5 μl of saturated overnight culture or 1/10 to 1/100 of the colony (approximately 10^2 – 10^4 cells) would be enough. Please keep in mind that excess culture or colony will inhibit SuperPhi DNA Amplification reaction! Yields and kinetics may vary if crude or un-quantified samples are amplified. We recommend to consider a starting point for adapting your specific reaction.

Quick Protocol

1. Preparation of Sample Mix for Bacterial Colonies:

Transfer 4 μl of Sample Buffer into a 0.2 ml PCR tube.
Pick 1/10 to 1/100 of the colony (approximately 10^2–10^4 cells) and add to the above PCR Tube.
Heat to 95 ˚C for 3 minutes and then quickly cool to 4˚C. Keep the samples on ice until use.

Note: *Heating at higher temperature or longer time may increase the probability of releasing host genomic DNA into cell lysis to the reaction, where the host genomic DNA will compete with the desired template DNA during amplification.*

2. DNA Amplification:

Mix 5 μl of Reaction Buffer and 1 μl of SuperPhi Enzyme and add to the above PCR tube contains 4 μl of Sample Mix as showed below:

Component	Volume/reaction
Sample Mix	4 μl
Reaction Buffer	5 μl
SuperPhi Enzyme	1 μl

One can prepare the master mix of Reaction Buffer and SuperPhi Enzyme for multiple reactions. The final volume is 10 μl. Mix well and incubate at 30 ˚C for 3~18 h.

3. Inactivate SuperPhi: Inactivate the enzyme by incubating at 65 ˚C for 10 min, and then cool to 4 ˚C.

4. Perform Downstream Application: The sample can be examined by agarose gel or applied for cloning and cycle sequencing. An aliquot of the amplified DNA can be directly added into the cycle sequencing reaction without further purification. Specially, the amplified RCA products as concatemers can be used to transform *Bacillus* directly.

5. FAQ and Troubleshooting: Please contact us at info@evomicscience.com.