

# Product Focus

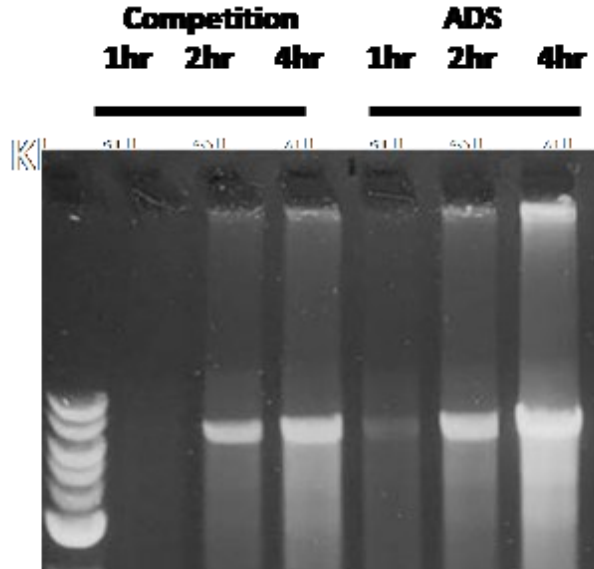
## Phi29 DNA Polymerase and RCA DNA Amplification Kit



Phi29 DNA polymerase from the *Bacillus subtilis* phage phi29 is the replicative polymerase that processes high processivity and strand-displacement activity.

The enzyme has been widely used in modern molecular biology for different applications such as amplification of plasmid templates for Sanger sequencing for amplification of plasmid templates, virus genome sequencing using next-generation sequencing, genotyping, and whole genome amplifications.

We provide Phi29 DNA polymerase and Phi29 RCA DNA amplification kit. Both products can be used to amplify DNA at 30°C for 4 to 18 hours.



Time course for phi29 DNA polymerase RCA from a single colony

### phi29 DNA Polymerase for Plasmid Template Amplification in Sanger Sequencing

Phi29 DNA polymerase can amplify DNA by the mechanism called strand displacing amplification. The enzyme can perform rolling circle amplification (RCA) on circular plasmids to make plasmid template for Sanger sequencing and make micrograms of DNA concatemers of plasmids from nanograms of DNA templates in a few hours. A small fraction of the resulting DNA can be directly used for Sanger sequencing reactions (cycle sequencing) without clean-up. This application is especially advantageous when the plasmid template amount is low, or only minute amount of culture or a single colony is available.

Use of RCA to amplify plasmid can reduce time and hands-on work compared with

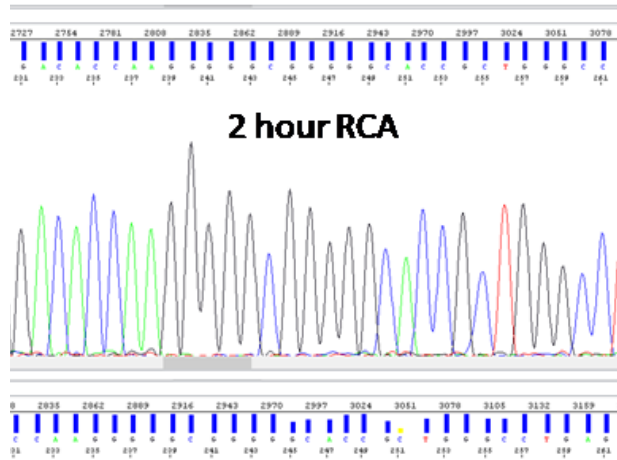
growth of cell culture and plasmid isolation and quantitation.

Here are the three major steps for RCA plasmid DNA preparation for Sanger sequencing:

- (1). Collect small amount of cell culture from bacterial stock or fresh culture, or a single colony.
- (2). Perform RCA amplification of the plasmid template at 30°C for 4 to 18 hours.
- (3). Use of a small fraction of the amplified DNA as template for Sanger sequencing reaction.

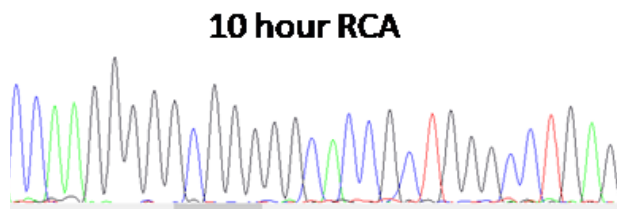
# Sanger sequencing results of a GC-rich template using RCA templates and SupreDye™ v3.1 Cycle Sequencing Kit

RCA using phi29 DNA polymerase from AdvancedSeq



2 hour RCA

4 hour RCA



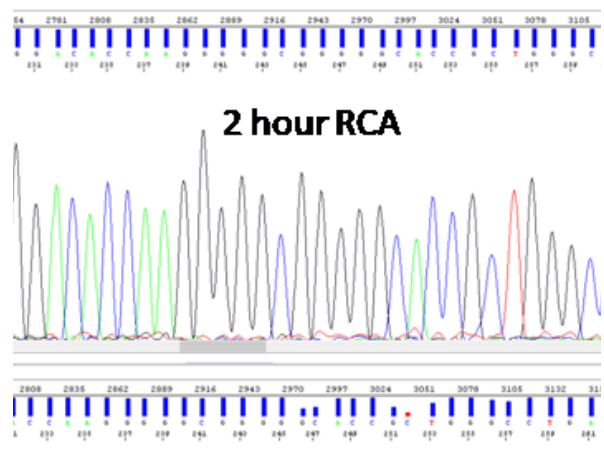
10 hour RCA

**Phi29 DNA polymerase**

Catalog number	Enzyme concentration	Unit Size
210025	10,000U/ml	250 units
210100	10,000U/ml	1000 units
210500	10,000U/ml	5,000 units

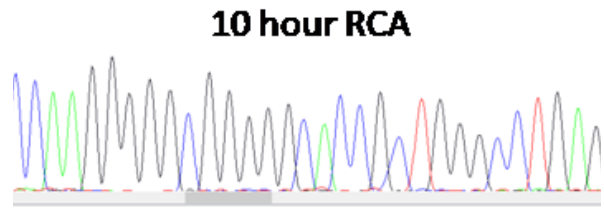
10x buffer is included

RCA using phi29 DNA polymerase from a well known vendor



2 hour RCA

4 hour RCA



10 hour RCA

**Phi29 RCA DNA Amplification Kit**

Catalog number	Enzyme concentration	Reactions
220100	10,000U/ml	100
220500	10,000U/ml	500

2x Amplification mix and lysis buffer and included