

illustra **TempliPhi** Sequence Resolver Kit Product Specification Sheet

Introduction

Product codes

28-9035-29

28-9035-30

28-9035-31

Important

Read these instructions carefully before using the products.

Intended use

The products are intended for research use only, and shall not be used in any clinical or *in vitro* procedures for diagnostic purposes.

Safety

For use and handling of the products in a safe way, refer to the Safety Data Sheets.

Description

illustra™ TempliPhi™ Sequence Resolver Kit uses the highly processive, strand-displacing Phi29 DNA polymerase and modified nucleotides to amplify circular templates for subsequent sequencing. The formulation is designed to help resolve sequencing problems associated with repeats, sequencing stops, and compressions due to high GC content. Product amplified using the TempliPhi Sequence Resolver Kit does not form the same secondary structure artifacts and provide greatly improved results with Phred20 scores greater than 800bp.

Short Protocol

The steps outlined below describe a general protocol to amplify sequencing templates. This protocol is a starting point for optimizing the reaction in your laboratory.

Note: *The short protocol is appropriate for researchers who have optimized their TempliPhi Sequence Resolver Kit reactions. First-time users must review the Full Protocol, which can be downloaded from cytiva.com. The full protocol can also be obtained by e-mail or fax by contacting your local cytiva.com Technical Support Group.*

Heat denaturation of template in sample buffer

Step	Action
1	Mix 1 μL of template with 4 μL of sample buffer.
2	Heat to 95°C for 3 minutes.
3	Cool to 4°C on ice.
4	Proceed with the next part of the protocol.

Preparation of amplification reaction

Step	Action
1	For each amplification reaction, combine 4.5 μL of reaction buffer with 0.5 μL of enzyme.
2	Mix on ice.
3	Add this to the cooled sample.
4	Proceed with the next part of the protocol.

Incubation

Step	Action
1	Incubate the sample at 10°C for 16–18 hours.
2	Proceed with the next part of the protocol.

Post-amplification heat inactivation

Step	Action
1	Heat the sample to 65°C for 10 minutes.
2	Cool to 4°C.
3	Proceed with the next part of the protocol.

Sequencing

Step	Action
1	Use 2 µL of amplification product per 20 µL sequencing reaction.



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For use only as licensed by Qiagen GmbH. The Phi 29 DNA polymerase may not be re-sold or used except in conjunction with the other components of this kit. See US patent number 6,323,009, and equivalent patents and patent applications in other countries.

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