

## Manual

# ExTerminator

Nucleotide dye terminators removal kit for DNA cycle sequencing reaction samples.

catalog #	size
444-50	50 isolations
444-250	250 isolations

For research use only.

### Guarantee

A&A Biotechnology provides a guarantee on this product.

The company does not guarantee the correct performance of this kit in the event of:

- not adhering to the supplied protocol
- use of not recommended equipment or materials
- use of other reagents than recommended or which are not a component of the product
- use of expired or improperly stored product or its components

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## Contents

component	444-50	444-250	storage
<b>Minicolumns</b>	50 pcs	250 pcs	room temp.
<b>1.5 ml tubes</b>	50 pcs	250 pcs	room temp.
<b>WP bind/wash solution</b>	30 ml	140 ml	room temp.
<b>Mix Blue</b>	350 µl	1.5 ml	room temp.
<b>Ultrapure water</b>	8 ml	15 ml	room temp.

Binding capacity of minicolumn: up to 10 µg

## Additional equipment and reagents

### Necessary

- Microcentrifuge

# Isolation protocol

**Note:** If cycle sequencing reaction is less than 10 µl add an appropriate volume of ultrapure water to reach the final volume of 10 µl.

1. Add **5 µl** of **Mix Blue** to the cycle sequencing mixture (performed in 10-20 µl).

2. Add **100 µl** of **WP** bind/wash solution. Mix by pipetting.

3. Apply samples onto the minicolumns.

4. Centrifuge for **30 s** at **12 000-14 000 RPM**.

**Note:** light blue color of the minicolumn membrane is a result of efficient precipitation of sequencing products.

5. Apply **400 µl** of **WP** bind/wash solution onto the minicolumns.

6. Centrifuge for **2 min** at **12 000-14 000 RPM**.

7. Transfer the minicolumns to **new** 1.5 ml tubes (included).

8. Add **ultrapure water** directly onto the minicolumns resin:

- capillary sequencer - **25 µl of ultrapure water**.
- slab gel sequencer - **50 µl of ultrapure water**.

While applying water onto the minicolumn be sure that liquid is applied directly onto the resin. If some water stays on the minicolumn wall the elution will be less effective.

9. Keep for **2 min** at **room temp**.

10. Centrifuge for **1 min** at **12 000-14 000 RPM**.

11. Clear light blue appearance of the eluted samples confirms the correct isolation of cycle sequencing DNA products. Blue color of the sample does not affect the readout of the DNA sequence.

## Capillary sequencer:

The samples are ready for thermal denaturation. Thermal denaturation can be directly preceded in the tube with the minicolumn.

## Slab gel sequencer:

Dry up the samples using a vacuum dryer and dissolve in an appropriate amount of loading buffer.

12. Store the samples at **-20 °C** until later use.

# Safety information

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**DANGER**

## WP bind/wash solution

H225 Highly flammable liquid and vapor.

H319 Causes serious eye irritation.

H336 May cause drowsiness or dizziness.

P210 Keep away from heat, sparks, open flames, hot surfaces. No smoking.

P261 Avoid breathing vapors.

P305+P351+P338 If in eyes: rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.

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