

WideUSE Plasmid Purification Kit

Cat. No: AN0068 (50 reactions)

Cat. No: AN0069 (100 reactions)

Description

WideUSE Plasmid Purification kit offers a rapid and convenient method for the routine isolation of high quality plasmid preparations in mini format. The kit is based in DNA ability to bind silica in the presence of high concentrations of chaotropic salts. **WideUSE** spin columns have packet a proprietary exclusive silica membranes and it binds up to 20 µg DNA.

Features

- **High yields** of up to 24 µg (*in 5% of isolations*) of DNA suitable for all molecular biology procedures.
- No phenol-chloroform extraction.
- **Ready to use** plasmid DNA.
- **Just a few minutes** procedure.
- **Mini format**

Applications

All molecular biology applications, such as:

- Digestion with restriction enzymes.
- Automated sequencing.
- PCR template.
- Bacterial transformation.
- Transfection.

Kit Components

Item	AN0068	AN0069
WideUSE minispin columns	50	100
Collection tubes (2 mL)	50	100
S-I Buffer	16 ml	16 ml
S-II Buffer	16 ml	16 ml
S-III Buffer	16 ml	16 ml
Binding Buffer	30 ml	60 ml
Washing Buffer*	8 ml	2X8 ml
Elution buffer	10 ml	10ml
RNase A	160 µl	160 µl

*Add 32 mL of 96-100% ethanol to the bottle containing 8 mL. This will give a final volume of 40 mL. The label on the bottle has a box that can be checked to indicate that ethanol has been added.

Storage

WideUSE Plasmid Purification Kits should be stored at room temperature (15–25°C) for up to 12 months without any reduction in performance.

Store the RNase A at -20°C. After addition of RNase A to Buffer S-I reagent can be stored at 4°C.

Quality Certifications

WideUSE Plasmid Purification Kit is tested for the isolation of any plasmid DNA from transformed *E.coli*.

The quality of purified DNA is analysed by:

- Ratio 260/ 280.
- Agarose gel electrophoresis.
- Digestion with restriction endonucleases.

(Continued on reverse side)

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Protocol: Plasmid DNA Purification

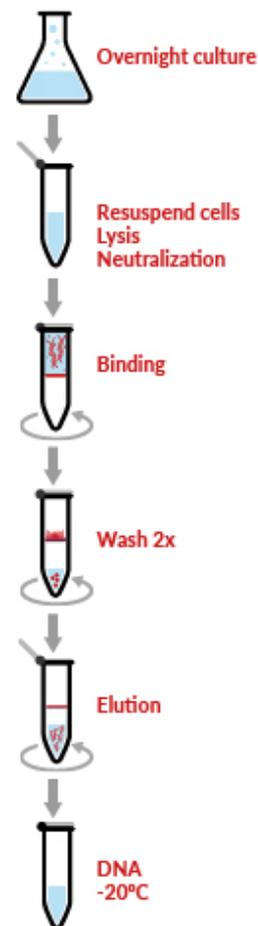
PREPARATION OF WORKING SOLUTIONS

Before starting the protocol prepare the following reagent:

- ✓ To prepare the S-I Buffer add 160 µl of RNase A.
- ✓ Add the volume ethanol (96%-100%) specified [Not included] to Washing Buffer prior to initial use. After ethanol has been added, mark the bottle to indicate that this step has been completed.

ASSAY PROCEDURE

1. Pour the culture in a 1.5 ml centrifuge tube and harvest the bacterial cells by centrifugation at 13000 rpm for 2 minutes. For low-copy plasmids should be collect 3 mL of culture and using 2 volumes of each solution to obtain good yields.
2. Resuspend the bacterial pellet in 100 µL of Buffer S-I
3. Add 100 µL of Buffer S-II, mix thoroughly by inverting the tube 6 times.
4. Add 100 µL of Buffer S-III, mix thoroughly by inverting the tube 8 times.
5. Centrifuge at 13000 rpm in a microcentrifuge for 10 min. Recover supernatant containing plasmid DNA promptly into a 1.5 ml centrifuge tube.
6. Add 500 µL of Binding Buffer, mix by inverting the tube several times. Incubate at room temperature for 5 min.
7. Apply the supernatants from step 6 to the **WideUSE spin column** by decanting or pipetting.
8. Centrifuge at 5500 rpm (9500g) for 90 seconds. Discard the flow-through.
9. Wash the **WideUSE spin column** by adding 700 µL washing Buffer and centrifuging at 5500 rpm for 90 s. Discard the flow-through.
10. Place the **WideUSE spin column** in a collection tube and add 500 µL of Isopropanol pure.
11. Centrifuge at 5500 rpm for 90 s. Discard the flow-through.
12. Again Centrifuge at 13000 rpm for 90 s. This step helps to remove traces of isopropanol.
13. Place the **WideUSE spin column** into a new, labelled 1.5 microcentrifuge tube and pipet 50-60µl Elution buffer directly into the membrane or pre-warm water. Close the cap and incubate for 1 minute at room temperature.
14. Centrifuge at 13000 rpm for 1 minute to elute DNA.



PRODUCT USE LIMITATION : This product is developed, designed and sold exclusively for research purposes and *in vitro* use only. The product was not tested for use in diagnostics or for drug development, and is not suitable for administration to humans or animals. Please refer to www.canvaxbiotech.com for the Material Safety Data Sheet of the product.

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