

Version No. FDL1720

FineQuick Rapid Mini Plasmid Extraction Kit

(Spin Column)

Cat. No.: D905

Kit Content:

Content	D905 (50 preps)
RNase A (10 mg/ml)	150 µl
Buffer BL	30 ml
Buffer P1	15 ml
Buffer P2	15 ml
Buffer P5	20 ml
Buffer DWQ	10 ml
Buffer EB	15 ml
DNA Pure Spin Columns	50 pcs
2 ml Collection Tubes	50 pcs

Storage:

FineQuick Rapid Mini Plasmid Kit can be stored at room temperature (15-25°C) for up to 12 months. Any significant reduction in performance and quality will not happen during storage period.

This Kit Special for Scientific Research.

Introduction

FineQuick Rapid Mini Plasmid Extraction Kit uses the optimized solution (Buffer P5 and Buffer DWQ) and unique silica membrane technology, which can specifically purify high-quality plasmid DNA within 8 minutes. This kit is suitable for plasmid DNA extraction from 1-4 ml from overnight culture of E. coli in LB (Luria-Bertani) medium. The purified plasmid DNA is applicable to a variety of molecular operations, including restriction enzyme digestion, PCR, sequencing, ligation, transformation, library screening, in vitro translation, transfection of conventional passage cells, etc..

Yield:

Plasmid Type	Bacterial Cells Yield	Plasmid Volume	Plasmid
Low Copy	1-4 ml	3-10 µg	pBR322, pACYC, pSC101,PET
High Copy	1-4 ml	6-24 µg	pTZ, pUC, pBS, pGM-T

Important Notes

1. Add RNase A solution to Buffer P1 before use, mix well and store at 2-8°C.
2. Check whether there is salt precipitation in Buffer P2 and Buffer P5, if any, re-dissolve the precipitation by incubation at 37°C.
3. Avoid to touch Buffer P2 and P5 directly by skin, and immediately tighten the lid after use.
4. All the centrifugation steps are finished at 12,000 rpm (~13,400× g) at room temperature (15-25°C).
5. The yield and quality of isolated plasmid DNA depend on the cell strain, cell culture condition, lysis of cells, copy number of plasmids, the stability of plasmid and the type of antibiotics.
6. Add ethanol (96-100%) to Buffer DWQ indicated on bottle label before use.

Protocol: (Please read the precautions carefully before use)

1. Column equilibration: Add 500 µl of Buffer BL to DNAPure Spin Column (the adsorption column is placed in a collection tube), centrifuge at 12,000 rpm (~13,400×g) for 1 min, and discard the filtrate. Place back the adsorption column into the collection tube. (The best performance of the equilibrated adsorption column is maintained for one day)
2. Harvest 1-4 ml bacterial cells in a microcentrifuge tube by spinning at 12,000 rpm (~13,400× g) for 1 min at room temperature (15-25°C), then discard all traces of supernatant.

3. Resuspend the bacterial cell pellet in 150 μ l Buffer P1 by pipetting or vortex (Confirm that RNase A is added to Buffer P1).

Note: Cell clumps indicate incomplete lysis, which will result in lower yield and purity.

4. Add 150 μ l Buffer P2 and mix gently by inverting 8-10 times.

Note: Gently mix by inverting the tube. Vigorous vortex will cause genomic DNA fragmentation. The solution will become viscous and slightly clear. To avoid the plasmid rapture, the lysis time does not exceed 5 min. If not clear, it is probably due to incomplete lysis, please reduce the cells density by inceasing P1, P2 and P5 in proportion.

5. Add 350 μ l Buffer P5 and immediately mix it by gently inverting 8~10 times. White flocculent precipitation will appear in the solution.

6. Centrifuge at 12,000 rpm (\sim 13,400 \times g) for 2 min, transfer the supernatant into Spin Column CP3 (the column is place in a collection tube), and avoid to pipet the white pellet.

Note: Please centrifuge again if a lot of white precipitate can be seen in the supernatant.

7. Centrifuge at 12,000 rpm (\sim 13,400 \times g) for 30 sec. Discard the flow-through and put back the Spin Column CP3 into the Collection Tube.

8. Add 300 μ l Buffer DWQ (confirm that ethanol (96-100%) is added to Buffer DWQ) and centrifuge at 12,000 rpm (\sim 13,400 \times g) for 30 sec. Discard the flow-through, and put back the Spin Colum CP3 into the Collection Tube.

9. Repeat step 8 once.

10. Centrifuge at 12,000 rpm (\sim 13,400 \times g) for 1 min to remove residual liquor from the Spin Column CP3.

11. Place the Spin Column CP3 in a clean 1.5 ml microcentrifuge tube. To elute DNA, add 50-100 μ l Buffer TB to the center area of the silica membrane in Spin Column CP3, and centrifuge at 12,000 rpm (\sim 13,400 \times g) for 30 sec.

Note: The volume of elution buffer should be no less than 60 μ l, otherwise it may affect recovery efficiency. The pH value of elution buffer should be kept between 7.5 and 8.0, if not, elution efficiency will decline.



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