WizPrep™ Plasmid DNA Mini Kit

RUO For Research Use Only

REF W70050

DESCRIPTION

The WizPrep™ Plasmid DNA Mini Kit provides a fast and simple means of isolating plasmid DNA from bacteria cells. The technique does not require any organic solvents and eliminates the need for repeated centrifugation, vacuum filtration or column separation. The kit is suitable for the isolation and purification of any plasmid, the size range for most effective purification is < 10 Kb. Good results have also been obtained with 20 Kb and bigger size plasmids although the obtained yield is reduced.

The obtained plasmid DNA is ready to use for a broad panel of downstream applications like PCR, restriction enzyme digestion, labeling, cloning, capillary sequencing.

KIT CONTENTS

Contents	100 prep	300 prep	Storage
PD1 Buffer(1)	30 ml	90 ml	Room temp.
PD2 Buffer	30 ml	90 ml	Room temp.
PD3 Buffer	40 ml	120 ml	Room temp.
Wash Buffer (concentrate)(2)	16 ml	50 ml	Room temp.
Elution Buffer	10 ml	30 ml	Room temp.
RNase A (lyophilized)(3)	3 mg	3 mg x 3	4°C
Spin Columns*	100	300	Room temp.
Instruction Manual	1	1	

(1) : Add provided RNase A to the PD1 Buffer and store at 4ºC.
(2) : Add absolute ethanol to the Wash Buffer prior to initial use (see the bottle label for volume).
(3) : The RNAse A should be stored at 4°C upon arrival.
* All Spin Columns are sterilized by electron beam.

REAGENTS AND EQUIPMENT TO BE SUPPLIED BY USER

- 96-100% ethanol (to prepare Wash Buffer)
- 1.5 mL microcentrifuge tubes
- Sterile RNase-free pipette tips
- Manual pipettors
- Centrifuge for microcentrifuge tubes
- Equipment for sample disruption and homogenization
- Personal protection equipment (lab coat, gloves, goggles

KIT SPECIFICATIONS

- Fast and easy processing using a rapid spin-column format
- High column binding capacity – up to 25 µg
- High yield of plasmid DNA – up to 20 µg from 1.5 ml of culture.

Parameter	Characteristics
Format	Silca-membrane spin column
Amount of Starting Material	0.5 - 2 ml of bacterial cultures
Typical yield	Up to 20 µg (2 ml culture)
$A_{260/280}$	1.8 - 2.1
Elution volume	50 - 100 µl
Preparation time	15 min. / 6 prep

QUALITY CONTROL ANALYSIS

In accordance with Wizbiosolutions Inc. ISO 13485-certified Quality Management System, each lot WizPrep™ Plasmid DNA Mini Kit is tested against predetermined specifications to ensure consistent product quality.

PROTOCOL

To check before start

- Before starting, add 64(200)ml of 100% ethanol to the 16(50)ml of Wash Buffer concentrate.
- Add provided RNase A to the PD1 Buffer and store at 4ºC

1. Harvesting

- Transfer 1.5 ml of cultured bacterial cells to a microcentrifuge tube. Centrifuge at 13,000 rpm for 1 min. and discard the supernatant. If more than 1.5ml of cultured bacterial cells is used, repeat the Harvesting Step.

2. Re-suspension

- Add **200µl of PD1 Buffer** (RNase A added) to the tube and resuspend the cell pellet by vortex or pipetting.

3. Lysis

- Add **200µl of PD2 Buffer** and mix gently by inverting the tube 10 times. Do not vortex to avoid shearing the genomic DNA.
- Let stand at room temperature for 2 min. or until the lysate is homologous.

4. Neutralization

- Add **300µl of PD3 Buffer** and mix immediately by inverting the tube 10 times. Do not vortex.
- Centrifuge at 13,000 rpm for 10 min.

5. Binding

- Place a Spin Column in a 2 ml Collection Tube.
- Add the supernatant from Step 4 to the Spin Column and centrifuge at 13,000 rpm for 1min.
- Discard the flow-through and place the Spin Column back in the 2 ml Collection Tube.

6. Wash

- Add **600µl of Wash Buffer** (ethanol added) into the Spin Column.
- Centrifuge at 13,000 rpm for 1min.
- Discard the flow through and place the Spin Column back in the 2 ml Collection Tube.
- Centrifuge at 13,000 rpm again for 2min. to dry the column matrix.

7. DNA Elution

- Transfer the dried Spin Column to a new micro tube.
- Add **50µl of Elution Buffer** into the center of the column matrix.
- Let stand for 3 min. or until the Elution Buffer is absorbed by the matrix.
- Centrifuge at 13,000 rpm for 1 min. to elute the DNA.

Note: To increase the final DNA yield use higher volume of Elution Buffer and increase the incubation time with the elution buffer up to 10 min.

- In order to increase the DNA concentration elute in smaller volume (the lowest recommended elution volume is 30 µl).

WizPrep™ Plasmid DNA Mini Kit

RUO For Research Use Only

REF W70050

TROUBLESHOOTING

Problem	Possiblity	Suggestions
Low yield of plasmid DNA	Bacterial cells were not lysed completely	• Following PD3 Buffer addition, break up the precipitate by inverting to ensure higher yield. • Too many bacterial cells were used.
	Incorrect Wash Buffer	• Ensure that correct volume of Ethanol was added to Wash Buffer prior to use
	Incorrect DNA Elution Step	• Ensure that Elution Buffer was added adequately and absorbed to the center of the Spin Column matrix
	Incomplete DNA Elution	• If plasmid DNA was larger than 10 kb, use pre-heated Elution Buffer (60-70℃) on Elution Step to improve the elution efficiency.
	Residual ethanol contamination	• After washing step, dry the Spin Column with additional centrifugation at full speed for 5 minutes or incubation at 60℃ for 5 minutes.
Eluted DNA does not perform well in downstream applications	RNA contamination	• Prior to using PD1 Buffer, ensure that RNase A was added. • If RNase A added PD1 Buffer is out of date, add additional RNase A. • If too many bacterial cells were used, reduce sample volume.
	Genomic DNA contamination	• Do not use overgrown bacterial culture. • During PD2 and PD3 Buffer addition, mix gently to prevent genomic DNA shearing.

SYMBOL GLOSSARY

REF	Catalogue number		Manufacturer
LOT	Batch code	RUO	Research use only
	Temperature limit		Instructions for use
	Use-by date		

ORDERING INFORMATION

Product	Cat No.	Package
WizPrep™ Plasmid DNA Mini Kit	W70050-100	100 prep
	W70050-300	300 Prep

Wizbiosolutions Inc.
#1103, 1405, 1406, A-dong, 14, Sagimakgol-ro 45beon-gil, Jungwon-gu, Seongnam-si, Gyeonggi-do, Republic of Korea
B237~B242, 14, Galmachi-ro 288beon-gil, Jungwon-gu, Seongnam-si, Gyeonggi-do, Republic of Korea

Tel : (82) 70-7603-5066
Fax: (82) 31-624-3066
e-mail: support@wizbiosolution.com
www.wizbiosolution.com