NAUTIAGENE

Product Information

NautiaZ Plasmid DNA Extraction Mini Kit

(100/300 prep)

Cat. No. : NPZ-S100/NPZ-S300

Sample : 1-4 ml of bacterial cultures

Yield : Up to 50 µg

NPZ-S100	NautiaZ Plasmid DNA Extraction Mini Kit (100 prep)
NPZ-S300	NautiaZ Plasmid DNA Extraction Mini Kit (300 prep)
NPZ-M020	NautiaZ Plasmid DNA Extraction Midi Kit (20 prep)
NPZ-L010	NautiaZ Plasmid DNA Extraction Maxi Kit (10 prep)
NPZ-L020	NautiaZ Plasmid DNA Extraction Maxi Kit (20 prep)

Contents

	NPZ-S100T	NPZ-S100	NPZ-S300
PS1 Buffer*	1 ml	25 ml	65 ml
PS2 Buffer	1 ml	25 ml	65 ml
PS3 Buffer	1.5 ml	35 ml	95 ml
W1 Buffer	2 ml	45 ml	125 ml
W2 Buffer**	300 µl X2	15 ml	25 ml x2
Elution Buffer	1 ml	10 ml	30 ml
RNase A (50mg/ml)	Added	50 µl	150 ul
PS Column	4 pcs	100 pcs	300 pcs
Collection Tube	4 pcs	100 pcs	300 pcs
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*Add provided RNase A to PS1 Buffer and store at 4°C.

**Add 60 ml / 100 ml ethanol (96-100%) to W2 Buffer prior to the initial use.

Buffer Preparation

- Add ethanol (96-100%) to the Wash Solution prior to first use

	NPZ-S100T	NPZ-S100	NPZ-S300
PS1 Buffer RNase A	- -	25 ml 50 µl	60 ml 150 µl
W2 Buffer ethanol (96 ~ 100%)	300 µl X2 1.2 ml X2	15 ml 60ml	25 ml 100ml

Important Notes

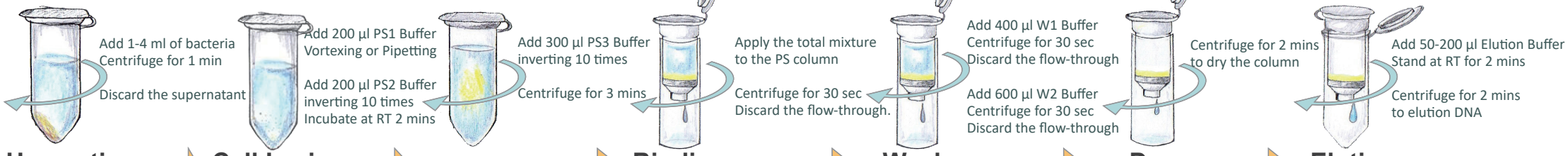
1. Buffers provided in this system contain irritants. Wear gloves and lab coat when handling these buffers.
2. Check PS2 Buffer before use. If any precipitation formed, warm the buffer in a 37°C waterbath to dissolve.
3. To avoid acidification of PS2 Buffer from CO_2 in the air, close the bottle immediately after use.
4. All centrifuge steps are done at full speed (14,000 x g) in a microcentrifuge.

Growth of Bacterial Cultures

1. Pick a single colony from a freshly streaked selective plate to inoculate 1-4 mL of LB medium supplemented with the appropriate selection antibiotic. Incubate for 16-18 hours at 37°C while shaking at 200-250 rpm. Use a tube or flask with a volume of at least 4 times the culture volume.
2. Harvest the bacterial culture by centrifugation at 8000 rpm (6800 × g) in a microcentrifuge for 2 min at room temperature. Decant the supernatant and remove all remaining medium.

Storage

RNase A should be store at 4°C , the other Buffer and Columns store at room temperature.



PURIFICATION PROTOCOLS

STEP	PROCEDURE
1 Harvest-ing	Transfer **1-4 ml of well-grown bacteria culture** to a microcentrifuge tube(*not provided*). Descend the bacteria by centrifuging at 14,000 x g for 1 min and discard the supernatant completely.
2 Resuspen-sion	**Add 200 µl PS1 Buffer (RNase A added)** to the pellet and resuspend the cells completely by pipetting or vortexing. Note: • Make sure that RNase A has been added into PS1 Buffer whenfirst use. • No cell pellet should be visible after resuspension of the cells.
3 Lysis	**Add 200 µl PS2 Buffer** and mix gently by inverting the tube 10 times to lyse the cells and incubate at room temperature for 2 mins until the lysate is homologous. Note • **Do not vortex**, vortex may shear genomic DNA. • Do not proceed this step over 5 min.
4 Neutral-ization	**Add 300 µl PS3 Buffer** and invert the tube 10 times immediately but gently. Note • **Do not vortex**, vortex may shear genomic DNA. • Invert immediately after adding PS3 Buffer will avoid asymmetric precipitation.
	Centrifuge at 14,000 x g for 3 mins. During centrifuging, place a PS Column in a Collection Tube.
5 DNA Binding	Transfer the supernatant carefully to PS Column. Centrifuge at 14,000 x g for 30 seconds then discard the flow-through. Place the PS column back into the Collection Tube. Note • Do not transfer any white pellet into the column.
6 Wash	**Add 400 µl W1 Buffer** to PS Column. Centrifuge at 14,000 x g for 30 seconds then discard the flow-through. Place the PS column back into the Collection Tube.
	Add 600 µl W2 Buffer to PS Column. Centrifuge at 14,000 x g for 30 seconds then discard the flow-through. Place the PS column back into the Collection Tube. Note • Make sure that ethanol (96-100 %) has been added into Wash uffer when first open.
7 Dry column	Centrifuge at 14,000 x g for an additional **2 min** to dry the column. Note • Important step ! This step will remove the residual liquid completely that will inhibit subsequent enzymatic reaction.
8 DNA Elution	Place PS Column to a new 1.5 ml microcentrifuge tube (not provided).
	Add 50 – 200 µl of Elution Buffer or ddH_2O to the membrane center of PS Column. Stand the column for 2 mins. Note: For effective elution, make sure that the elution solution is dispensed on the membrane center and is absorbed completely. If plasmid DNA is larger than 10 kb, use preheated 70°C Elution Buffer to improve the elution efficiency.
	Centrifuge at 14,000 x g for 2 mins to elute plasmid DNA.
9 Pure DNA	Store plasmid DNA at 4 °C or -20 °C.