



شرکت تحقیقاتی تولیدی پویا ژن آزما  
POUYA GENE AZMA Co. (PGA)

## PGA Plasmid purification Kit

Catalog No. PL240-050

Quantity: 50/25 Prep

PGA Plasmid purification kit is designed to isolate up to 30  $\mu$ g of high-quality plasmid DNA from 1-5 mL bacterial cultures in 30 minutes or less. Plasmid DNA purification is simplified with Mini Column technology. Purified plasmid DNA is immediately ready for a wide variety of downstream applications such as routine screening, restriction enzyme digestion, and DNA sequencing.

### Features and Benefits

Spin column format  
Rapid purification  
Stable and consistent result  
Instant use: No need of additional materials  
30  $\mu$ g of binding capacity and high purity  
No use of organic solvents  
Ready for use in fluorescent sequencing, cloning, hybridization,  
Electroporation and other enzymatic manipulation



for more information

### Component list

	50 preps	25 preps
column	50	25
Collection tube	50	25
Buffer R - Cell Resuspension Buffer	12 ml	6
Buffer L - Cell Lysis Buffer	14 ml	7
Buffer N - Neutralization Buffer	20 ml	10
Buffer EW - Column Wash Buffer	26 ml	13
Buffer W - Column Wash Buffer	4 ml	2
Buffer EB - Elution Buffer	6 ml	3
RNase A (10 mg/ml)	50 $\mu$ l	30

**Storage:** RNase A at -20°C for one year; others at room temperature (15-25°C) for one year

### Procedure

1- Add 1-5 ml (High copy number plasmid) or up to 10 ml (Low copy number plasmid) overnight cultured bacterial suspension to a microcentrifuge tube.

- 2- Centrifuge at 13000 rpm for 1 minute. Discard the supernatant.
- 3- Add 200 µl of R buffer (premixed with Rnase A) to the cell pellet and resuspend it completely by pipetting.
- 4- Add 250 µl of L buffer, mix immediately and thoroughly by inverting the tube 4-6 times.
- 5- Add 350 µl of N buffer, mix thoroughly by inverting the tube 4-6 times. When the neutralization is complete, precipitate will form. Incubate the lysate at room temperature for 2 minutes.
- 6- Centrifuge at 13000 rpm for 10 minutes. Transfer the supernatant into a spin column.
- 7- Centrifuge at 13000 rpm for 1 minutes. Discard the flow through.
- 8- Add 500 µl of EW buffer to the column, centrifuge at 13000 rpm for 1 minute. Discard the flow through. (optional: When working with *endA*<sup>+</sup> strains, endonucleases can be efficiently removed by optional wash step with buffer EW to ensure that plasmid DNA is not degraded during storage or enzyme reactions.)
- 9- Add 500 µl of W buffer (check to ensure you have added 16 ml ethanol) to the column, centrifuge at 13000 rpm for 1 minute. Discard the flow through.
- 10- Centrifuge the empty column at 13000 rpm for 1 minute to remove residual EW completely.
- 11- Place the spin column in a clean microcentrifuge tube, add 30-100 µl of EB buffer or distilled water directly to the center of the column matrix. Incubate the column at room temperature for 1 minute. Centrifuge the column at 12000 rpm for 1 minute to elute DNA. The isolated plasmid DNA is ready to use or can be stored at -20°C.

The genotype of various *E.coli* strains

<b><i>EndA</i><sup>+</sup> strains</b>	<b><i>EndA</i><sup>-</sup> strains</b>
BL21(DE3), CJ236, HB101, JM83, JM101, JM110, LE392, MCI061, NM series, P2392 PR series, RRI, TBI, TGI, BMH71-18, ESI301, wild-type and etc.	DH1, DH20, DH21, DH5α, JM103, JM105, JM106, JM107, JM108, JM109, MM294, SK1590, SRB, XLI-Blue, XLO and etc.

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