



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

## PGA Plasmid purification kit

Catalog No. PL240-050

Quantity: 50 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  
Compatible with endA+ strains  
No use of organic solvents  
Ready for use in fluorescent sequencing, cloning, hybridization, electroporation and other enzymatic manipulation

### Component list

column	50
Collection tube	50
Buffer R - Cell Resuspension Buffer	10 ml
Buffer L - Cell Lysis Buffer	12.5 ml
Buffer N - Neutralization Buffer	17.5
Buffer W - Column Wash Buffer	25 ml
Buffer EB - Elution Buffer	5 ml
RNase A (20 mg/ml)	150 $\mu$ l
Protocol Handbook	1

**Storage :** RNase A at -20oC for one year; others at room temperature (15-25oC) 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  $\mu$ l of R buffer (premixed with Rnase A) to the cell pellet and resuspend it completely by pipetting.
- 4- Add 250  $\mu$ l of L buffer , mix immediately and thoroughly by inverting the tube 4-6 times.
- 5- Add 350  $\mu$ l 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 temprature for 2 minutes.
- 6- Centrifuge at 13000 rpm for 5 minutes. Transfer the supernatant into a spin column.
- 7- Centrifuge at 13000 rpm for 1 minutes. Discard the flow through.
- 8- Add 650  $\mu$ l of W buffer (check to ensure you have added ethanol) to the column , centrifuge at 13000 rpm for 1 minut. Discard the flow through.
- 9- Centrifuge the empty column at 13000 rpm for 1 minut to remove residual EW completely.
- 10- Place the spin column in a clean microcentifuge tube, add 30-100  $\mu$ l of E buffer or distilled water directly to the center of the column matrix. Incubate the column at room temprature 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.

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