PureYield™ Plasmid Miniprep System from Promega

- Prepare Lysate
  - ◊ Add 600 µL of bacterial culture to a 1.5 mL microcentrifuge tube.
  - ◊ Add 100 µL of **Cell Lysis Buffer (Blue)**, and mix by inverting the tube 6 times.
  - ◊ Add 350 µL of cold **Neutralization Solution**, and mix thoroughly by inverting.
  - ◊ Centrifuge at maximum speed in a microcentrifuge for 3 minutes.
  - ◊ Transfer the supernatant to a **PureYield™ Minicolumn** without disturbing the cell debris pellet.
  - ◊ Place the minicolumn into a Collection Tube, and centrifuge at maximum speed in a microcentrifuge for 15 seconds.
  - ◊ Discard the flowthrough, and place the minicolumn into the same Collection Tube.
- Wash
  - ◊ Add 200 µL of **Endotoxin Removal Wash (ERB)** to the minicolumn. Centrifuge at maximum speed in a microcentrifuge for 15 seconds.
  - ◊ Add 400 µL of **Column Wash Solution (CWC)** to the minicolumn. Centrifuge at maximum speed in a microcentrifuge for 30 seconds.
- Elute
  - ◊ Transfer the minicolumn to a clean 1.5 mL microcentrifuge tube, then add 30 µL of **Elution Buffer** or nuclease-free water directly to the minicolumn matrix. Let stand for 1 minute at room temperature.
  - ◊ Centrifuge for 15 seconds to elute the plasmid DNA. Cap the microcentrifuge tube, and store eluted plasmid DNA at -20 °C.

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