DIRECTIONS FOR USE

Cyclo-Pure™
Gel Extraction Kit from Agarose Slices
(K220-50RXN)

* Fast Spin Column Format to Purify DNA (40 bp - 100 kb) from Agarose
* Compatible with Standard Agarose Gels in TAE or TBE Buffer
* High Throughput Extraction Procedure Increases Productivity

Product Description:

Cyclo-Pure is a spin column-based kit for the quick isolation of DNA fragments (40 bp - 100 kb) from standard agarose gels in TAE or TBE buffer. In just 10 minutes, extract ultra-pure DNA ready-to-use in restriction enzyme digestion, labeling, ligation, transformation, *in vitro* transcription, and sequencing protocols.

Kit Components:

The Cyclo-Pure Kit contains reagents sufficient for 50 isolations. All reagents can be stored at room temperature. Each kit includes the following components.

Component	Amount	Component	Amount
* **Binding Buffer**	40 ml	* **Cyclo-Pure Spin Columns**	50 each
* **Washing Buffer**	50 ml	* **Cyclo-Pure Collection Tubes**	50 each

Required Equipment: A micropipettor, bench-top microcentrifuge, and 1.5 - 1.7 ml microfuge tubes.

Protocol for Plasmid DNA Purification:

1. Excise the desired DNA band (≤ 200 mg) from the agarose and place it into a clean sterile micro-centrifuge tube.
2. Add 3 volumes of **Binding Buffer** to the gel slice and incubate at 55°C for 5 minutes.
3. Insert a **Spin Column** into a **Collection Tube**. When the gel slice is completely melted, apply the melted solution to the Spin Column and spin at top speed (12-14,000 x g) for 30 sec.
4. Remove the Spin Column from the Collection Tube and discard the filtrate.
5. Add 700 ul of the **Washing Solution** to the spin column and spin (at 12-14,000 x g) for 30 sec. Remove the spin column from the collection tube and discard the filtrate. Spin for an additional 2 min. (12-14,000 x g) to remove residual traces of ethanol.
6. Remove the spin column and place it into a new micro-centrifuge tube (not provided). Add 30-50 ul of preheated (60-70°C) H_2O or TE Buffer.
7. Elute the plasmid DNA by centrifugation (12-14,000 x g) for 30 sec.[1,2] Store the eluted DNA at -20°C.

1. To increase the yield of DNA by as much as 10-15%, steps 6 - 7 can be repeated.
2. For automated fluorescent sequencing, an ethanol precipitation step is recommended following elution.

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