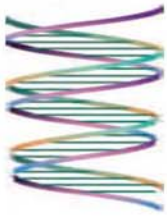

DIRECTIONS FOR USE

Cyclo-Prep™

Genomic DNA Purification Kit

Product Description:

Cyclo-Prep, Genomic DNA Purification Kit is a spin column-based kit for rapid purification of genomic DNA from up to 30 mg of animal tissue or $1x10^9$ bacterial cells. In as little as 20 minutes after sample lysis, prepare ultra-pure genomic DNA ($A_{260}/A_{280} \geq 1.8$) using your tabletop microcentrifuge. The genomic DNA isolated from Cyclo-Prep is suitable for PCR Amplification, RFLP Analysis, Southern Blotting, DNA Fingerprinting, and Bacterial-Genome Sequencing. The simple protocol provides a fast and convenient alternative to common genomic DNA purification procedures.

Kit Components:

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

* **Reagent 1**	10 ml	* **Cyclo-Prep Spin Columns**	50 each
* **Reagent 2**	10 ml	* **Collection Tubes**	50 each
* **Reagent 3**	50 ml	* **Plastic Pestles**	50 each
* **Water, Nuclease-Free**	10 ml		

Necessary Reagents and Equipment (not supplied with the kit):

Proteinase K (20 mg/ml solution for enzymatic lysis of animal tissue samples)
TE Buffer, pH 8.0 containing 1 mg/ml Lysozyme (for enzymatic lysis of bacterial samples)
95% Ethanol‡
Micropipettor
Bench-top microcentrifuge
1.5 – 1.7 ml sterile microfuge tubes.

Protocol for Genomic DNA Purification:

Tissue Preparations:

When preparing animal tissue, cut the tissue into pieces (up to 30 mg total) and place in a microcentrifuge tube. Add 180 µl of Reagent 1 and 20 µl of Proteinase K solution (20 mg/ml), vortex thoroughly and incubate at 70°C for 30 minutes. Homogenize the sample by using a pestle (provided). Continue until the tissue has been completely homogenized. Add 200 µl of Buffer 2, vortex thoroughly, and incubate at 70°C for 15 minutes. Add 200 µl of 95% ethanol‡ to the

sample and vortex thoroughly.

Bacterial Preparations:

When preparing bacterial cells, resuspend the cells (up to 2 x 10^9) in TE Buffer, pH 8.0 containing 1.0 mg/ml Lysozyme. Vortex and incubate at room temperature for 10 minutes. Add 180 µl of Buffer 2 and 20 µl of Proteinase K solution (20 mg/ml), vortex immediately, and incubate at 70°C for 30 minutes. Following the complete lysis of the bacteria, add 200 µl of 95% ethanol‡ and vortex thoroughly.

‡ Following the addition of ethanol, a white precipitate may form. Be sure to apply the entire sample, including the precipitate, to the Cyclo-Prep spin column.

For Both Tissue and Cell Lysates:

1. Apply the sample, including any precipitate that may have formed, to the Cyclo-Prep spin column placed inside a collection tube (provided). Centrifuge for 3-5 minutes at maximum speed. Discard the flow-through.
2. Add 500 µl of Reagent 3 to the column and centrifuge for 1 minute at maximum speed to wash the column. Discard the flow-through.
3. Add an additional 500 µl of Reagent 3 to the column and centrifuge for 2-3 minutes at maximum speed to dry the Cyclo-Prep membrane.
4. *Optional*: Transfer the Cyclo-Prep spin column to a clean microcentrifuge tube (not provided) and centrifuge at maximum speed for one minute. This ensures adequate drying of the Cyclo-Prep membrane.
5. Transfer the Cyclo-Prep spin column to a sterile microcentrifuge tube (not provided) and pipette 100 µl of sterile, nuclease-free water to the column. Centrifuge for 1 minute at maximum speed to elute the DNA. (This step may be repeated to increase the yield of DNA; however, this will decrease the final concentration of the DNA.) TE buffer may be used in place of water, if desired.

interchim

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