

## Chondroitin Sulfate DNA Extraction Kit Instructions

### Composition

Chondroitin Sulfate DNA Extraction Kit	50 Preps
Cat. No.	7901050
Spin Columns	50
2 ml Collection Tubes	50
Buffer S	30 ml
Buffer P3	30 ml
Buffer WB (concentrate)	12 ml
Buffer TE	5 ml
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### Storage

1. All the reagents and components can be stored for up to 2 years at room temperature (15~25°C). For longer storage, it is recommended to keep at 2~8°C.
2. The product stored at 2~8°C should be restored to room temperature before use.

### Technical Support

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### Introduction

This kit uses the principle of column purification of nucleic acids, which is suitable for extraction and recovery of up to 12 µg of high-purity DNA (70 bp~10 kb) from chondroitin sulfate, with a recovery efficiency of 75~90%, and the purified DNA can be directly used in PCR detection experiments.

### Equipment and Reagents to Be Supplied by User

1. Absolute ethanol.
2. 1.5 ml tubes, pipettes, and tips.
3. Disposable gloves, tissues and protective equipment.
4. Microcentrifuge(s) (with rotor for 1.5 ml and 2 ml centrifuge tubes).

### Preparation Before Use

1. If the centrifuge has a refrigeration function, set the temperature to 25°C.
2. Add absolute ethanol to Buffer WB according to the instructions on the label of the reagent bottle and tick the box on the label to mark "Ethanol Added".
3. 3 M sodium acetate (pH 5.0) may be required.

## Protocol

**1. Weigh 100~200 mg chondroitin sulfate, add 2 times volume of Buffer S, vortex until all the chondroitin sulfate dissolved.**

\* For example, if 150 mg of chondroitin sulfate is weighed, 300  $\mu$ l Buffer S will be added to this conversion to 150  $\mu$ l volume of chondroitin sulfate

\* If there is an insoluble pellet, centrifuge at 12,000 rpm for 30 sec and take the supernatant to step 2).

**2. Add 500  $\mu$ l Buffer P3 to a clean 1.5 ml centrifuge tube, transfer 100  $\mu$ l chondroitin sulfate solution from step 1 to the 1.5 ml centrifuge tube and vortex to mix well.**

\* Chondroitin sulfate solution can be very viscous, if chondroitin sulfate solution adheres to the tip, the tip can be pipetted several times in Buffer P3 in the centrifuge tube to fully dissolve chondroitin sulfate in Buffer P3.

\* The dye added to Buffer P3 is an indication of pH, if the solution turns purplish red after adding a sample to Buffer P3, the sample is too alkaline, and approximately 10  $\mu$ l 3 M sodium acetate (pH 5.0) should be added to restore the solution to its original orange-yellow color, otherwise it will affect the binding of DNA to the Spin Column.

**3. Transfer the mixture to a Spin Column (the Spin Column is placed in a 2 ml Collection Tube), close the lid, and centrifuge at 12,000 rpm for 30 sec.**

**4. Discard the filtrate, put the Spin Column back into the 2 ml Collection Tube, add 700  $\mu$ l Buffer WB to the Spin Column, close the lid, and centrifuge at 12000 rpm for 30 sec.**

\* The filtrate does not need to be completely discarded, if you want to avoid the contamination of the centrifuge by the filtrate adhered to the mouth of the Collection Tube, you can slap the 2 ml Collection Tube upside down on a paper towel.

\* Ensure that Absolute ethanol has been added to Buffer WB.

**5. Discard the filtrate, put the Spin Column back into the 2 ml Collection Tube. Centrifuge at 14,000 rpm for 1 min.**

\* If the centrifuge does not reach 14,000 rpm, centrifuge at full speed for 2 min.

\* This step of high-speed spin dry is to remove the residual ethanol in Buffer WB.

**6. Discard the 2 ml Collection Tube, put the Spin Column in a clean 1.5 ml centrifuge tube, add 30~50  $\mu$ l Buffer TE to the Spin Column center, close the lid, incubate at room temperature for 1 min, and centrifuge at 12000 rpm for 30 sec to elute the DNA.**

\* If the centrifuge does not have a leak-proof lid, it is recommended to change the conditions for eluting DNA to 8000 rpm for 1 min to prevent the 1.5 ml centrifuge tube lid from falling off and damaging the centrifuge.

\* DNA can also be eluted with deionized water but ensure that the pH of the deionized water used is 7.0~8.5, otherwise the elution efficiency of the DNA will be affected.

**7. Discard the Spin Column, and the eluted DNA can be immediately used for various molecular biology experiments or stored at -20°C for later use.**