

## User Bulletin

### DNA Advanced™ DNA Clean Up and Concentrator System

DNA Advanced™ DNA Clean Up and Concentrator System provides a simple and fast method to clean up and concentrate DNA (range from 100bp to 23kb) from samples, such as **Oragene™ DNA**, enzymatic reactions, PCR reaction, and salts without phenol/chloroform extraction. This system is based on binding of up to 50µg DNA to silica-based membranes in chaotrophic salts with average recoveries of 60 to 95 % of >10 kb DNA fragments.

#### Downstream Application

- \* Sequencing & PCR
- \* Restriction digestion & enzymatic reaction
- \* Ligation
- \* Labeling & hybridization

#### Product Contents

Cat. No	DC1001	DC1002
Preps	50	250
PX Buffer	30	150
WN Buffer	6	30
WS Buffer	6	30
Elution Buffer	5	25
DC Column	50	250
Collection Tube	50	250
Protocol	1	1

*All buffers need to be mixed well before use.*

## Shipping & Storage

The sample of DNA Advanced™ DNA Clean Up and Concentrator System should be shipped and stored at ambient temperature for up to 6 months.

If precipitate form by freezing temperature on any buffer, warm up at 37°C to redissolve.

## Protocol

- ❖ **Please read the following notes before starting the procedure.**

## Important Notes

- Add 24 ml for DC1001 or 120 ml for DC1002 of 98 ~ 100 % ethanol to WN Buffer bottle when first open.
- Add 24 ml for DC1001 or 120 ml for DC1002 of 98 ~ 100 % ethanol to WS Buffer bottle when first open.
- Buffers provided in this system contain irritants. Appropriate safety apparels such as gloves and lab coat should be worn.
- All procedures should be done at room temperature (20 ~ 25 °C).
- All centrifugation steps are done at 10,000 x g or 13,000rpm in a microcentrifuge.
- For long-term storage of the eluted DNA, TE buffer should be used for elution. Since EDTA in TE may affect downstream applications, Elution Buffer (provided) or ddH<sub>2</sub>O (pH 7.0 ~ 8.5) is preferred for elution.

## Viogene's unique design – EasyLid™

The EasyLid™ is designed to prevent contamination during the procedure.

### Tips for EasyLid™ –

Twist the arm of the cap and pull the cap to break the EasyLid™.



### A. DNA Fragment Isolation Protocol for Spin Method:

- 1. Pipet 100 to 500  $\mu$ l of sample or DNA solution to a new 1.5 ml centrifuge tube. Add equal volume of PX Buffer and mix well.**
- 2. Add all the mixture from step 1 into DC column. And preheat 10 mM Tris-HCl (pH 9.0), ddH<sub>2</sub>O, or TE buffer (provided by user) at 70 °C (500  $\mu$ l/prep) for DNA elution.**

Load no more than 0.7 ml mixture into the column each time.
- 3. Centrifuge for 30-60 seconds. Discard the flow-through.**
- 4. Wash the column with 0.5 ml WN Buffer by centrifuging for 30-60 seconds. Discard the flow-through.**

Make sure that ethanol has been added into WN Buffer bottle when first open.
- 5. Wash the column with 0.5 ml WS Buffer by centrifuging for 30-60 seconds. Discard the flow-through.**

Make sure that ethanol has been added into WS Buffer bottle when first open.

- 6. Centrifuge the column at full speed<sup>+</sup> for another **3 minutes or more** to remove residual ethanol.**

It is important to remove ethanol residue, residual ethanol may inhibit subsequent enzymatic reactions.

- 7. The 1<sup>st</sup> elution. Place the column onto a new 1.5 ml centrifuge tube. Add 10-50  $\mu$ l of Elution Buffer (provided) onto the **center of the membrane**.**

For effective elution, make sure that the elution solution is dispensed onto the center of the membrane and is completely absorbed.

- 8. Stand the column for 2 - 3 minutes and centrifuge at full speed<sup>+</sup> for 1 - 2 minutes to elute DNA.**
- 9. The second elution. Add another 10-50  $\mu$ l of Elution Buffer onto the column and centrifuge at full speed<sup>+</sup> for 1 - 2 minutes to elute DNA, the second elution will elute less concentrated of residual DNA, if one need more concentrated DNA, should use the 1<sup>st</sup> eluted DNA.**

- 10. Store DNA at 4°C or –20°C**

<sup>+</sup> The full speed may depend on the top speed of the microcentrifuge.

## B. DNA fragment Isolation Protocol for Vacuum Method:

- 1. Pipet 100 to 500  $\mu$ l of sample or DNA solution to a new 1.5 ml centrifuge tube. Add equal volume of PX Buffer and mix well.**
- 2. Add all the mixture from step 1 into DC column. And preheat 10 mM Tris-HCl (pH 9.0), ddH<sub>2</sub>O, or TE buffer (provided by user) at 70 °C (500  $\mu$ l/prep) for DNA elution. Apply the vacuum to pull all the liquid into the manifold.**
- 3. Apply vacuum to draw all the liquid into the manifold.**
- 4. Wash the column once with 0.5 ml WN Buffer by re-applying vacuum to draw all the liquid.**  
Make sure that ethanol has been added into WN Buffer bottle when first open.
- 5. Wash the column once with 0.5 ml WS Buffer by re-applying vacuum to draw all the liquid.**  
Make sure that ethanol has been added into WS Buffer bottle when first open.
- 6. Place the column onto a Collection Tube. Centrifuge the column at full speed<sup>+</sup> for another **3 minutes or more** to remove residual ethanol.**  
It is important to remove ethanol residue, residual ethanol may inhibit subsequent enzymatic reactions.

- 7. The 1<sup>st</sup> elution. Place the column onto a new 1.5 ml centrifuge tube. Add 10-50  $\mu$ l of Elution Buffer (provided) onto the **center of the membrane**.**  
For effective elution, make sure that the elution solution is dispensed onto the center of the membrane and is completely absorbed.
- 8. Stand the column for 2 - 3 minutes and centrifuge at full speed<sup>+</sup> for 1 - 2 minutes to elute DNA.**
- 9. The second elution. Add another 10-50  $\mu$ l of Elution Buffer onto the column and centrifuge at full speed<sup>+</sup> for 1 - 2 minutes to elute DNA, the second elution will elute less concentrated of residual DNA, if one need more concentrated DNA, should use the 1<sup>st</sup> eluted DNA.**
- 10. Store DNA at 4°C or -20°C.**

<sup>+</sup> The full speed depends on the top speed of the microcentrifuge.