# dNEAT™ Agarose Purification Kit

PURK-AGA-050

## Description

The dNEAT™ Agarose Purification Kit provides a rapid and efficient method to extract DNA from agarose gels. It is based on the solubilisation and binding of DNA to a silica membrane in presence of chaotropic salts. The dNEAT™ minispin columns contain an exclusive membrane that binds DNA fragments previously excised from agarose gel.

## Features

- ✓ Simple procedure
- ✓ Wide spectrum of size fragments could be purified (suitable from 100 bp up)
- ✓ High Percentage of Recovery, greater than 80% on 0.7-1% agarose. Recovery is lower in more concentrated agarose gels (50-60% on 2% agarose)
- ✓ The resulting purified DNA is ready to use for all molecular biology procedures
- ✓ Suitable for any kind of agarose and gel buffer systems

## Applications

- ✓ Purification of DNA fragments (obtained by PCR or digestion with restriction enzymes) from agarose gels
- ✓ The purified DNA can be used in all molecular biology applications

## Storage

The dNEAT™ Agarose Purification Kit should be stored at room temperature (15–25°C) for up to 12 months without any reduction in performance.

## Kit Components

| | |
|---|---|
| dNEAT™ minispin columns | 50 |
| Collection tubes (2 mL) | 50 |
| QG Buffer | 60 ml |
| PE Buffer* | 11,25 ml |
| EB Buffer | 10 ml |

*Ethanol (96%-100%) [not included] must be added prior to use as indicated on the label. After ethanol has been added, mark the bottle to indicate that this step has been completed

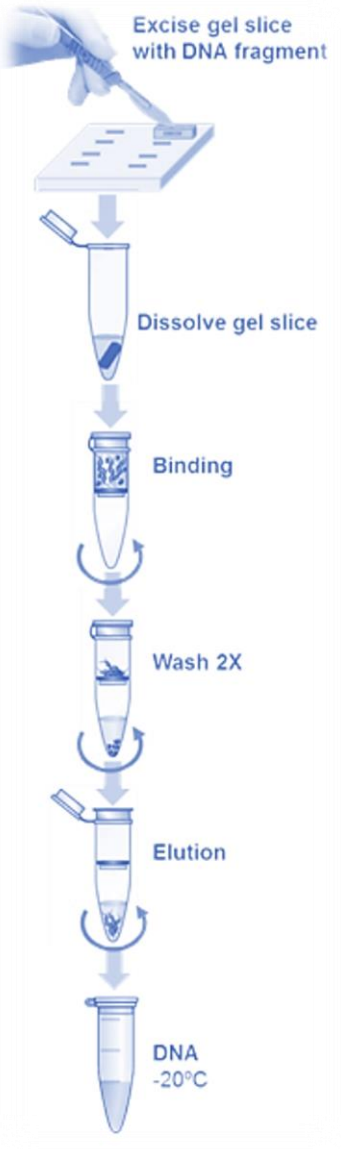
## Product use limitation

This product is developed, designed and sold exclusively for research purposes and use only. The product is not intended for diagnostics or drug development, nor is it suitable for administration to humans or animals.

## Quality Certifications

Tested in the purification of a 0.5 kb DNA fragment excised from 2% agarose gel. The purified band is analysed in agarose gel electrophoresis.

## Protocol




1. Using a clean, sharp razor blade or scalpel, excise the DNA band from the agarose gel. Remove the extra agarose to reduce the size of the gel slice. Place the gel slice in a 1.5 ml pre-weighted tube and weigh the gel slice (The maximum amount of gel slice per column is 400 mg).

2. Add 3 volumes of QG Buffer to 1 volume of gel. (For example, if the agarose gel slice is 100 mg, add 300 µl of QG buffer)
   *For gels containing more than 2% agarose, add 6 volumes de QG Buffer per mg of gel.*

3. Incubate at 50°C in a water bath for 10 min or until the gel slice has completely dissolved. During incubation, mix by vortexing or inverting the tubes every 1 minute. Make sure the gel slice is completely dissolved. For >2% gels, increase incubation time.
   *Important! For fragments <500 bp and >4 kb, add 1 volume of isopropanol to the sample and mix (For example, if the agarose gel slice is 100 mg, add 100 µl isopropanol)*

4. Label the lid of a new minispin column placed in a 2 ml collection tube. Carefully pour the mix from previous step to the spin column and centrifuge at 13000 rpm for 1 minute. For mixture volumes of more than 750 µl, load and centrifuge again using the same column.

5. Place the spin column in a new 2 ml collection tube and discard the collection tube containing the filtrate. Add 700 µl of PE buffer to the minispin column and centrifuge at 13000 rpm for 1 minute.
   *Remember! Before using it for the first time, add ethanol (96–100%) to PE Buffer as indicated on the bottle.*

6. Discard the flow-through and centrifuge again at 13000 rpm for 1 minute. This step is essential for removing any PE buffer traces.

7. Transfer the column to a clean 1.5 ml microcentrifuge tube. Add 30-50 µl of Elution Buffer (EB) or $H_2O$ (pH=7.0-8.5) to the centre of the column membrane and incubate at room temperature for at least 2 minutes to ensure the Elution Buffer is completely absorbed.* Centrifuge at 13000 rpm for 1 minute to elute and collect DNA.
   **To increase the DNA yield, warm the buffer EB/$H_2O$ to 65 °C*

8. Ultra-pure DNA is now ready to use.