# EchoCLEAN Organic Solvent DNA CleanUp Kit-Protocol

for single-step removal of inhibitors and a wide range of impurities from DNA solutions.

This protocol has been developed to remove impurities like inhibitors, salts, nucleotides, TRIzol™, phenol, chloroform, and other organic solvents from DNA solutions.

## Materials and equipment needed

- 80 to 110 µl of DNA sample. For DNA samples less than 80 µl, dilute with Low-TE Buffer to a minimum of 80 µl.
- Microcentrifuge with rotor for 1.5 ml and 2 ml reaction tubes.
  Important: Switch centrifuge to *relative centrifugal force, rcf* (x *g**); if this is not possible, please use formula below* to calculate the conversion of round per minute (rpm) into *rcf*.
- Vortex.
- One reaction tube (2 ml) per sample for column preparation.
- One reaction tube (1.5 ml) per sample for elution and collection of the purified DNA
- Pipets for 10 µl and 200 µl scale, corresponding pipet tips
- For fastest procedure (PROTOCOL 1): Cap Puncher (BioEcho product no. 050-001-001)

## Preparation before starting

- Set the microcentrifuge to **1000 x g.***
- Important: **Switch to relative centrifugal force, rcf (x g***, not rpm)

## Product use limitation

The EchoCLEAN Organic Solvent CleanUp Kits are for research use only. They have not been registered or authorized to be used for diagnosis, prevention, or treatment of a disease.

* Most centrifuges offer the choice between rpm and g-force (rcf); if not, calculate the rpm matching the g-force using the formula: rpm = 1,000 x $\sqrt{(\frac{g}{1.12 \times r})}$ , where r = radius of rotor in mm. and g the required g-force.
E. g., with a radius of 150 mm, the corresponding rpm to 1,000 x g is approx. 2,400 rpm.

## PROTOCOL 1: Purification **using** the Cap Puncher

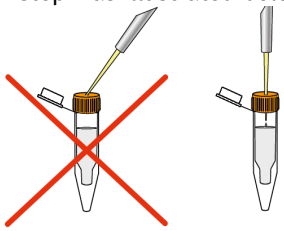
BioEcho Cap Puncher

### Column preparation

1. Vortex the **EchoCLEAN Organic Solvent Spin Column** briefly and place into a **2 ml** reaction tube.
   Let stand for about 5 min.
2. Use of the Cap Puncher (scan QR code to watch a video): Punch a hole into the column cap and lift the column together with the Cap Puncher out of the 2 ml collection tube. Snap off bottom closure of the column and detach the Cap Puncher by twisting while pulling out.
   Place the punched spin column back into the 2 ml reaction tube
3. Centrifuge for **1 min at 1000 x g*.** Discard the flow-through volume (void volume) collected in the 2 ml reaction tube.
4. Place the prepared spin column into a new **1.5 ml** reaction tube for elution of the purified DNA and place back into the rack.
   **Continue with DNA Cleanup.**

### DNA Cleanup

5. Transfer **80 – 110 µl of the DNA sample** to the prepared **EchoCLEAN Spin Column** from step 4 as illustrated below:



Insert pipet tip vertically through the hole in the column cap and pipet the sample slowly (~5 sec) into the column.

Note:

- Do not punch pipette tip into the resin bed during loading of lysate!

6. Centrifuge **1 min at 1000 x g*.** The purified genomic DNA elutes into the 1.5 ml elution tube and can be immediately applied in downstream applications.

The eluted DNA can be used immediately or stored at 4°C or for long-term storage at –20°C. For spectrophotometric analysis, use Low-TE Buffer (T) supplied with the kit as blank.

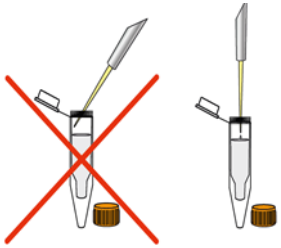
## PROTOCOL 2: Purification **without** a Cap Puncher

### Column preparation

1. Vortex the **EchoCLEAN Organic Solvent Spin Column** briefly and place into a **2 ml** reaction tube. Let stand for 5 min.
2. **Loosen** the screw cap of the spin column **half a turn** and **snap off the bottom closure.**
   **Important: Do not close the screw cap of the spin column.** The screw cap must stay loosened **half a turn** to avoid generation of a vacuum. Place the column back into the 2 ml collection tube and both into the centrifuge.
3. Centrifuge for **1 min at 1000 x g*.** Discard the flow-through volume (void volume) collected in the 2 ml reaction tube.
4. Place the prepared spin column into a new **1.5 ml** reaction tube for elution of the sample DNA and place back into the rack.
   **Continue with DNA Cleanup**.

### DNA Cleanup

5. Transfer **80 – 110 µl of the DNA sample** to the prepared **EchoCLEAN Spin Column** from step 4 as illustrated below



Open cap and pipet the sample slowly (~5 sec) onto the center of the resin bed of the prepared spin column. Close screw cap and loosen again half a turn.
**Important: Do not close the screw cap of the spin column tightly!**

Note:

- Do not punch pipette tip into the resin bed during loading of lysate!

6. Centrifuge **1 min at 1000 x g***. The purified genomic DNA elutes into the 1.5 ml elution tube and can be immediately applied in downstream applications

The eluted DNA can be used immediately or stored at 4°C or for long-term storage at –20°C. For spectrophotometric analysis, use 1x Tris Buffer (T) supplied with the kit as blank.

# EchoCLEAN Organic Solvent DNA CleanUp Kit

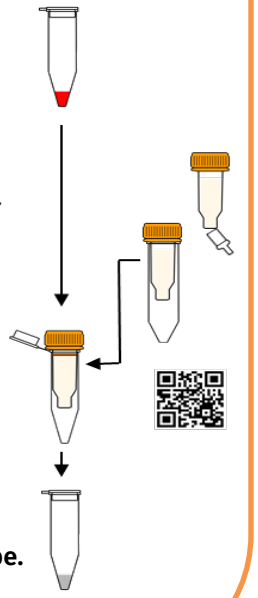
**for 1-step removal of inhibitors, TRIzol™, organic solvents and a wide range of impurities from DNA solutions**

| | | | |
|---|---|---|---|
| Product no. (rxn's) | 020-002-040-010 (10) | 020-002-040-050 (50) | 020-002-040-250 (250) |
| Kit contents | EchoCLEAN Organic Solvent DNA CleanUp Spin Columns, and Low-TE Buffer | | |

## **Quick PROTOCOL** (please read protocol first)

### Column preparation

- Vortex **EchoCLEAN spin column** and place in a **2 ml** tube
  Let stand for 10 min.
- Punch a hole in the cap with the Cap Puncher, and
  break off bottom closure (scan QR code to watch a video).
- Place spin column back into 2 ml tube.
- Centrifuge **1 min at 1000 x g*** to elute column buffer.
- Place column in a **1.5 ml** tube.



### DNA Cleanup

- Transfer **DNA sample (max. 110 µl)** by pipetting
  **slowly** through cap hole (scan QR code to watch a video).
- Centrifuge **1 min at 1,000 x g*** to elute DNA into **Elution tube.**
- Eluted DNA is ready to use.

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