

## TransZol

Cat. No. ET101

Storage: at 2-8°C in dark for one year

### Description

*TransZol* is a ready-to-use reagent for the isolation of total RNA from cells and tissues. *TransZol* combines phenol and guanidine thiocyanate in a mono-phase solution to inhibit RNase. After lysis and centrifugation, RNA remains in the aqueous phase and others in the interphase or organic phase. RNA is precipitated by addition of isopropanol.

- Isolate RNA from a variety of species: animal, plant, yeast, bacteria and virus.
- The whole procedure can be completed in one hour.
- Simultaneous isolation of RNA, DNA and protein from the same sample.
- Pink solution for easy visualizing different phases.
- Unique dissolving solution for long-term RNA storage.

### Procedures

Reagents provided by customers: chloroform, isopropanol, 75% ethanol (prepared with RNase-free water) and RNase-free water

#### 1. Homogenization

##### a. Adherent cells

- Wash culture dish once with 1×PBS
- Detach cells with cell spatula. Add 1 ml of *TransZol* to per 10 cm<sup>3</sup> culture dish. Pipetting up and down to lysis the cells.
- Transfer lysate to a microcentrifuge tube.
- Incubate at room temperature for 5 minutes.

##### b. Suspension cells

- Transfer suspension cells to a microcentrifuge tube. Centrifuge the sample at 8,000× g for 2 minutes at 2-8°C, discard the supernatant.
- Add 1 ml of *TransZol* to per 10<sup>7</sup> cells.
- Pipetting up and down until no visible precipitates are present in lysate.
- Incubate at room temperature for 5 minutes.

##### c. Animal tissue and plant materials

- After weighing, quickly transfer the frozen sample into mortar with liquid nitrogen. Grind thoroughly to a powder. Use more liquid nitrogen if needed. Incomplete grind can affect RNA yield and quality.
- Transfer the tissue powder to a microcentrifuge tube. Add 1 ml of *TransZol* to per 50-100 mg tissue. Homogenize tissue samples with a homogenizer and repetitively pipette up and down.
- Incubate at room temperature for 5 minutes.

#### 2. Add 0.2 ml of chloroform for per ml *TransZol* used. Shake the tube vigorously by hand for 30 seconds. Incubate at room temperature for 3 minutes.

3. Centrifuge the sample at  $10,000\times g$  for 15 minutes at  $2-8^{\circ}\text{C}$ . The mixture separates into a lower pink organic phase, an interphase, and a colorless upper aqueous phase which contains the RNA. The volume of the aqueous upper phase is around 60% volume of *TransZol* reagent.
4. Transfer the colorless, upper phase containing the RNA to a fresh RNase-free tube. Add 0.5 ml of isopropanol for per ml *TransZol* used. Mix thoroughly by inverting tube. Incubate at room temperature for 10 minutes.
5. Centrifuge the sample at  $10,000\times g$  for 10 minutes at  $2-8^{\circ}\text{C}$ . Discard the supernatant. Colloidal precipitate can be seen at the wall and the bottom of the tube.
6. Add 1 ml of 75% ethanol (prepared with RNase-free water), vortexing vigorously (add at least 1 ml of 75% ethanol for 1 ml *TransZol* used).
7. Centrifuge the sample at  $7,500\times g$  for 5 minutes at  $2-8^{\circ}\text{C}$ .
8. Discard the supernatant. Air-dry the RNA pellet (about 5 minutes).
9. RNA pellet is dissolved in 50-100  $\mu\text{l}$  of dissolving solution.
10. Incubate at  $55-60^{\circ}\text{C}$  for 10 minutes. For long-term storage, store the purified RNA at  $-70^{\circ}\text{C}$ .

#### Note

It is important to mix well after adding chloroform to ensure extraction performance.

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