



Best Tools for Scientists!

TOOLSharp Serum Exosome Extraction Kit

For exosome extraction from serum

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Introduction

The TOOLSharp Serum Exosome Extraction Kit is specially designed for isolating the exosome, which contains the RNA and protein secreted by various types of cells, from serum. Compared with traditional ultrahigh-speed centrifugation, the simple low-speed centrifugation used in the aforementioned kit causes less centrifugal stress on the exosomes; thus, the morphology of the exosomes remains more intact when using low-speed centrifugation than when using ultrahigh-speed centrifugation. Moreover, this product can save time, requires a small amount of input samples, and provides high isolation efficiency. The exosomes obtained using this product can be applied in various downstream applications, such as protein research, RNA analysis, and high-throughput sequencing.

Kit Contents

| Content | TTC-RF02 |
|----------------------------------|-----------------|
| Serum exosome extraction reagent | 10 mL |

Output

A total of 5–20 ng of total RNA is extracted using the following protocol: obtain 1 mL of fresh serum (or unthawed frozen serum stored at $-70\text{ }^{\circ}\text{C}$), use the serum exosome extraction reagent to isolate the exosome, and perform RNA isolation.

Storage

The TOOLSharp Serum Exosome Extraction Kit should be stored at $2\text{--}8\text{ }^{\circ}\text{C}$.

Protocol

1. Serum preparation

- (a) Hold the collected whole blood (without anticoagulants) still at room temperature for 1–2 h or at 2–8 °C overnight until the blood coagulates.
- (b) Centrifuge the blood at 4 °C and $1000\text{--}2000 \times g$ for 5–10 min. Then, gently aspirate the supernatant (serum, transparent yellow liquid at the upper layer) into a new centrifuge tube without disturbing the cell component.
- (c) This collected serum can be directly used in downstream experiments or stored at –70 °C after the splitting is packed.

2. Sample preparation

- (a) Place the serum on ice. If the sample is frozen, thaw it in a 25 °C water bath until it completely becomes liquid and then place it on ice.
- (b) Centrifuge the serum at room temperature at $2000 \times g$ for 30 min to remove cells and debris.
- (c) Gently aspirate the supernatant into a new centrifuge tube without disturbing the sediment and residual liquid, and place the supernatant on ice until use.

3. Exosome extraction

- (a) Transfer a required volume of serum into a new centrifuge tube. To this serum, add the serum exosome extraction reagent with one-fifth the volume of the serum.

For example:

| Serum volume | Reagent volume to be added |
|--------------|----------------------------|
| 1 mL | 0.2 mL |
| 2 mL | 0.4 mL |

- (b) Gently invert the solution or pipette it up and down several times to mix it well until a homogenous solution is obtained.

Note: The solution should be cloudy.

- (c) Stand the mixture at 2–8 °C for 30 min for incubation.
- (d) Centrifuge the mixture at room temperature and $10\,000 \times g$ for 5 min.
- (e) Carefully and thoroughly discard the supernatant by using a pipette to obtain the exosome contained in the sediment.
- (f) Resuspend the exosome sediment by using $1 \times$ PBS or directly apply the exosome sediment in the subsequent experiments.

Note: If necessary, store the exosome at 2–8 °C for up to 1 week or at –20 or –70 °C for a long period.

Troubleshooting

◆ What sample volume is required for a single experiment?

A serum volume of at least 0.5 mL is required because a lower serum volume will fail to extract sufficient exosome to meet the experiment demands.

◆ How can the isolated exosome be applied in downstream experiments?

The isolated exosome sediment can be resuspended using 1× PBS or the reagent used in the downstream experiment (e.g., some lysis buffer).

◆ How can the exosome sediment be resuspended?

The isolated exosome sediment can be resuspended using 1× PBS or the reagent used in the downstream experiment. If the downstream experiment does not require the exosome to remain intact, a low-speed homogenizer can be used in the resuspension procedure.

◆ Other important points

The blood component is complex. For some difficult-to-isolate samples, such as hyperlipidemia samples, the exosome extraction and subsequent treatment should be adjusted according to the actual situation.