

### **Exo-Fect Exosome Transfection**

1. Resuspend the exosome collected in ultracentrifugation in 450 ul PBS
2. Combine these materials in a 1.5 ml tube: 10 ul Exo-fect solution, 20 ul mRNA solution (~100 ng/ul), 70 ul sterile 1x PBS, 50 ul exosome solution in PBS.
3. Mix the components well by inverting three times.
4. Incubate the exosome transfection solution at 37°C in a shaker for 10 minutes and then immediately place the tube on ice.
5. To stop reaction, add 30 ul of the ExoQuick-TC reagent provided in the kit to the transfected exosome sample suspension and mix by inverting 6 times. Do not vortex.
6. Place the transfected exosome sample on ice (or at 4°C) for 30 minutes.
7. Centrifuge the sample for 3 minutes at top speed.
8. Remove the supernatant and resuspend the transfected exosome pellet in 300 ul 1x PBS.
9. Add 150 ul per  $10^5$  cells (about 800 ul and 2 ml DMEM exosome-free medium in a 25 cm<sup>2</sup> flask), and incubate for 24 hours.