The miRNeasy Serum/Plasma Advanced Kit (cat. no. 217204) is shipped at ambient temperature. Upon arrival store the RNeasy® UCP MinElute® spin columns at 2–8°C. Store the remaining components dry at room temperature (15–25°C). This protocol is for purification of total RNA including small RNAs from serum or plasma.

Further information

- miRNeasy Serum/Plasma Advanced Kit Handbook: [www.qiagen.com/HB-2390](http://www.qiagen.com/HB-2390)
- Safety Data Sheets: [www.qiagen.com/safety](http://www.qiagen.com/safety)
- Technical assistance: [support.qiagen.com](http://support.qiagen.com)

Notes before starting

- Buffer RWT and RPL contain guanidine salt and are therefore not compatible with disinfecting reagents containing bleach.
- If necessary, re-dissolve any precipitate in Buffer RPL or Buffer RWT by warming
- Equilibrate buffers to room temperature (15–25°C).
- All steps should be performed at room temperature (15–25°C). Work quickly.
- Add ethanol (96–100%) to Buffer RWT and Buffer RPE concentrates before use (see bottle label for volume).
- The miRNeasy Serum/Plasma Spike-In Control (cat. no. 219610) may be purchased separately. For instructions on preparing a working solution, refer to the handbook.
- Before starting, read method for preparing serum or plasma in the handbook.
1. Prepare serum or plasma or thaw frozen samples.

2. Transfer 200 μl serum or plasma into a 2 ml reaction vessel.

3. Add 60 μl Buffer RPL. Close the tube caps and vortex for > 5 s. Leave at room temperature for 3 min.

4. Add 20 μl Buffer RPP. Close the tube caps and mix vigorously by vortexing for >20 s. Incubate at room temperature for 3 min.

5. Centrifuge at 12000 x g for 3 min at room temperature to pellet the precipitate.

   **Note:** Supernatant should be clear and colorless.

6. Transfer supernatant (~230 μl) to a new reaction tube. Add 1 volume isopropanol. Mix well by vortexing. Transfer entire sample to an RNeasy UCP MinElute column. Close the lid, and centrifuge for 15 s at ≥8000 x g. Discard the flow-through.

7. Pipet 700 μl Buffer RWT to the RNeasy UCP MinElute spin column. Close the lid, and centrifuge for 15 s at ≥8000 x g. Discard the flow-through.

8. Pipet 500 μl Buffer RPE onto the RNeasy UCP MinElute spin column. Close the lid, and centrifuge for 15 s at ≥8000 x g. Discard the flow-through.

9. Add 500 μl of 80% ethanol to the RNeasy UCP MinElute spin column. Close the lid, and centrifuge for 2 min at ≥8000 x g. Discard the flow-through and the collection tube.

10. Place the RNeasy UCP MinElute spin column in a new 2 ml collection tube (supplied). Open the lid of the spin column and centrifuge at full speed for 5 min to dry the membrane. Discard the flow-through and the collection tube.

11. Place the RNeasy UCP MinElute spin column in a new 1.5 ml collection tube (supplied). Add 20 μl RNase-free water directly to the center of the spin column membrane and incubate 1 min. Close the lid, and centrifuge for 1 min at full speed to elute the RNA.

Scan QR code for handbook.