In the name of God

DNA extraction kit from tissues and blood (For research only CN: KPG-DNKct)

Component of DNKct kit for extraction of DNA:

- DNK solution (CN: KPG-DNK), 2 vial 25 mL
- Precipitation (CN: KPG-PS), 1 vial 25 mL
- Washing buffer (CN: KPG-WB), 1 vial 25 mL
- DNase free water (CN: KPG-DW), 1 vial 3 mL
- High absorbent column (50 Preps)

Other materials needed: Samplers, Centrifuge, 1.5 mL tubes, Tissue homogenizer

Samples: This kit was designed to extract DNA from blood samples, cell culture, and homogenized tissues. To extract the DNA from the various sources add the following volume:

1. Blood/Serum/Plasma: 200 µL.
2. Cell culture: Up to 1×107 cells
3. Homogenized tissues: 30-50 mg (The tissues need to be homogenized previously)

Tissue preparation

Before using the kit, you need homogenize the tissue samples. Harvested tissues should be used freshly or stored at very low temperature as quickly as possible. Grinding in mortal and pestle under liquid nitrogen is a good method for homogenizing the sample, but alternative methods, such as a homogenizer or a bead-beater, can be employed in case by case for efficient disruption. Shaking or vortex during incubation for lysis may greatly accelerate the efficiency of lysis, resulting in reduced time for complete lysis. Note that the freshness and the particle size of disrupted sample is the **key** for good result and that the frozen sample should be kept on ice until use.

Experimental procedure

- Shake the DNK solution bottle vigorously. Add 1000 µL of DNK solution for tissues and 500 µL of DNK solution for blood and cell cultures to a new tube and then add appropriate amount of the samples to the tube. Vigorously vortex the tube and incubate for 10 minutes in the room temperature.
- Centrifuge the tube at 8000 rpm for 1 min.
- Transfer the supernatant to a new tube and add 500 µL of precipitation, then **gently** mix the tube component and incubate for 3 minutes at -10 to -20ºC (Freezer).
- **Gently** mix the tube component again and transfer the combination of the watery supernatant/precipitation into a column as much as possible. Incubate the column in room temperature for 2 minutes.
- Discard the filtered solution in the collection tube and centrifuge the tube at 10000 rpm for 20 seconds.
- Discard the solution in the tube and add 500 µL of washing solution into the column (Shake immediately the washing buffer just before using) and centrifuge the tube at 10000 rpm for 20 seconds.
- Transfer the column to a new 1.5 mL tubes and then add 40 µL pre-heated (60ºC) DNase free water on the column and incubate for 1 minute.
- Centrifuge the tube at 10000 rpm for 1 minute. The pellet contains the DNA.
- Transfer the pellet to the column and repeat the centrifuge.

Note: To increase the yield of DNA, you can add lower amounts of DNase free water or multiply the volume. For example, add 500 µL DNK to 250 µL blood and ETC. However, you need to purchase additional DNK, Solution A and Precipitation to multiply the volume.