

## Special protocol for saliva DNA extraction v.1212

### Prepar a water bath at 60 °C for step 6.

1. Transfer 1 ml saliva sample to a 15 ml centrifuge tube. Add 4 ml PBS (not provided) and mix well.
2. Centrifuge the tube at 1,800 x g for 5 min.
3. Carefully discard the supernatant. Resuspend the pellet in 150 µl PBS and transfer the sample to a 1.5 ml microcentrifuge tube.  
**Note: Do not disrupt the pellet.**
4. **(Optional):** If RNA-free genomic DNA is required, add 4 of 100 mg/ml RNase A to the sample and incubate at room temperature for 2 min .
5. Add 20 µl Proteinase K and **200 µl FATG2 Buffer** to the sample. **Mix thoroughly by pulse-vortexing.**
6. Incubate at 60 °C for 15 minutes to lyse the sample. **During incubation, vortex the sample for every 5 minutes.**
7. Add 200 µl ethanol (96- 100 %) to the sample. **Mix thoroughly by pulse-vortexing for 30 sec.**
8. Transfer the sample mixture (including any precipitate) carefully to FATG Column.  
Centrifuge for 1 minute **then place FATG Mini Column to a new Collection Tube.**
9. Wash FATG Mini Column with 500 µl W1 Buffer by centrifuge for 1 minute then discard the flow-through.
10. Wash FATG Mini Column with 750 µl Wash Buffer by centrifuge for 1 min then discard the flow-through.  
**- Make sure that ethanol has been added into Wash Buffer when first open.**
11. **Centrifuge for an additional 3 minutes to dry the column.**  
**Important Step!** The residual liquid can affect the quality of DNA and inhibit subsequent enzymatic reactions.
12. Place FATG Mini Column to Elution Tube.
13. Add 100 ~ 200 µl of Elution Buffer or ddH<sub>2</sub>O (pH 7.5- 9.0) to the membrane center of FATG Mini Column.  
**Stand FATG Column for 3 minutes.**  
- **Important Step!** For effective elution, make sure that the elution solution is dispensed onto the membrane center and is absorbed completely.  
- Standard volume for elution is 200 µl. If less samole to be use, reduce the elution volume (100~150 µl) to increase DNA concentration.
14. Centrifuge for 2 minutes to elute total DNA.
15. Store total DNA at 4 °C or -20 °C.