Purification of genomic DNA from water and air using Exgene™ Soil DNA mini

This protocol is especially designed for isolation of genomic DNA from environmental samples.

Preparation
- Preheat a water bath at 56℃
- Filter paper

1. Filter the water or air samples using a filter paper. After filtration, add the shredded filter paper to PowerBead™ tube. (* e.g., with scissors)
2. Add 550 µl of Buffer SL to the tube.
3. Homogenize the sample in the Precellys (or similar) equipment twice for 23 sec, each at 6,500 rpm.
   Alternatively, secure tubes horizontally on the flat or vortex with tape and vortex at maximum speed for 10 min.
4. Centrifuge at 10,000 x g for 10 min at room temperature and carefully transfer the supernatant to a 1.5 ml tube (provided).
5. Add 50 µl of Buffer RH.
6. Add 300 µl of Buffer PD and mix well by vortexing.
7. Centrifuge at 10,000 x g for 5 min at room temperature and carefully transfer the supernatant to a 2 ml tube (provided).
   Small pellet containing humic acid, cell debris, and protein can formed in the collection tube after centrifugation. Be careful not to disturb this pellet.
8. Add 900 µl of Buffer TB and mix well by vortexing.
   If Buffer TB shows signs of precipitation, pre-heat in a 56°C water bath to dissolve completely.
9. Transfer up to 700 µl of the mixture to a mini spin column.
10. Centrifuge at 10,000 x g for 30 sec at room temperature.
11. Repeat steps 9-10 two more times using the remainder of the sample.
12. Add 500 µl of Buffer NW to the mini spin column.
13. Centrifuge at 10,000 x g for 30 sec at room temperature.
    Discard the pass-through and reinsert the mini spin column back into the same tube.
14. Centrifuge at maximum speed for 1 min at room temperature to remove residual wash buffer.
15. Transfer the mini spin column to a new 1.5 ml tube (provided).
16. Residual ethanol may interfere with downstream reactions. Care must be taken at this step to eliminate the carryover of Buffer NW.
17. Add 50 µl of Buffer EB to the center of the membrane in the mini spin column.
18. Incubate for 1 min at room temperature. Centrifuge at 10,000 x g for 1 min at room temperature. Elution volume can be decreased to 30 µl for high concentration of DNA, but this will slightly decrease in overall DNA yield.
    If maximum recovery of DNA is preferred or the starting materials contain large amount of DNA, elution can be done in 200 µl of Buffer EB.

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