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Working

Fecal DNA extraction by bead beating









Version 1

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ABSTRACT

From Surana Lab protocols

This protocol is suitable for extracting DNA from either human or mouse feces. Best results will be obtained with 10-60 mg of starting material

- 1 **For each 2ml screw cap tube,**
 - Add  400 μ l beads
 - Add  550 μ l of phenol/chloroform
 - Add  250 μ l SDS
 - Add  500 μ l of PB buffer found in the Qiaquick PCR purification kit
- 2 Bead beat on Precellys, setting 2
- 3 Spin down the tubes for  00:05:00 at 4000 RPM in microcentrifuge
- 4 Proceed to PCR purification kit – for the purification of up to 10 ug PCR products
- 5 Label & Place a QIAquick column in a 2ml collection tube (provided)
- 6 Apply the aqueous top layer of your sample onto the column and centrifuge for  00:01:00 , 17,900g (13,000 RPM), at room temperature.
- 7 After the spin, dump the contents of the collection tube.
- 8 Wash the column by adding  750 μ l of Buffer PE and repeat the spin at the same conditions for  00:01:00 .

- 9 Repeat steps 7 and 8
- 10 After the second wash and spin, and after dumping the contents of the collection tube, Spin the column and collection tube one last time to remove residual wash buffer for 🕒 00:01:30
- 11 Place each QIAquick column in a clean 🧴 1.5 ml microcentrifuge tube.
- 12 To elute DNA, add 🧴 50 µl Buffer EB (provided in kit) OR water (pH of 7.0-8.5) to the center of the Qiaquick membrane and allow the tube to sit for 🕒 00:02:00
- 13 Centrifuge the column for 🕒 00:01:30 17,900g (13,000 RPM), at room temperature
- 14 Quantify with the Qubit dsDNA BR Assay kit. Alternatively, a nanodrop suffices



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