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Fecal DNA extraction by bead beating V.1

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Protocol status: Working
We use this protocol and it's working

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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 µL beads
- Add 550 µL of phenol/chloroform
- Add 250 µL SDS
- Add 500 µL 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 µL 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