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Qiagen DNeasy 96 blood & tissue protocol for bee guts

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1 Works for me Share

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Jocelynz

ABSTRACT

Tissue protocol for bee guts.

DOI

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PROTOCOL CITATION

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1

52931

Day 1	1h 6m 22s

1 UV sterilize supplies for 2 96 well plates worth of extractions: 1 50mL centrifuge tube, 1 15mL centrifuge tube, 1 box 1,000uL pipette tips, 2 boxes 200uL pipette tips, 24 elution microtube strip caps, steel beads, zirconia beads, and 1 reagent reservoir

- 3 Add **⊒200 µL** ATL + Proteinase K solution to each sample
- 4 Add $\sim 100 \, \mu L$ zirconia beads and 2 steel beads to each sample
- 5 Place plates in tissue lyser and run at □30 Hz for ⊙00:02:30, then rotate plates 180 degrees and lyse at □30 Hz for an additional ⊙00:02:30

TissueLyser II
Bead Mill
QIAGEN 85300

TissueLyser Adapter Set 2 x 96
Adapter set

Qiagen 69984

protocols.io

2

6 Centrifuge on "short" for © 00:00:07 then shake by hand for © 00:00:15

22s

Eppendorf™ 5810R Centrifuge Centrifuge Eppendorf 02-262-8187 ←

1h

7 Incubate § 56 °C for © 01:00:00 with weight on top of plate

Note: remove from incubator carefully with weight on top so that caps do not pop off

1m

8 Place plates in tissue lyser and run at $\square 30$ Hz for $\lozenge 00:01:00$

9 Incubate § 56 °C overnight with weight on top of plate

Day 2 53m 44s

- 10 UV sterilize supplies: 4 reagent reservoirs, 24 elution microtube strip caps (clear), 2 boxes 1,000uL tips (plus some extra), 6 boxes 200uL tips (plus some extra), 2 s-blocks, 2 elution microtube plates (dark blue) and 24 strip caps (white), and 6 96 well microplates (for aliquotting final product). Label 6 foil microplate lids for aliquots
- 11 Carefully remove plates from incubator, leaving the weight on top, and allow to sit at room temperature for a few minutes before carefully removing weight to ensure strip caps do not pop off
- 12 Shake plates by hand for © 00:00:15 , rotating plates halfway through

15s

- 14 Add \Box 410 μ L ALE to each sample
- Seal with new caps and shake by hand for **© 00:00:15**, rotating plates halfway through
- 16 Centrifuge on "short" for © 00:00:07

7s

- 17 Place DNeasy filter plates on top of s-blocks
- 18 Transfer lysate to DNeasy filter plate and seal with AirPore tape
- 19 Centrifuge **3486 x g, 00:13:00** . Discard flowthrough

13m

20 ⊠Buffer

Add **□500 µL** AW1 **Qiagen Catalog #19081** tape

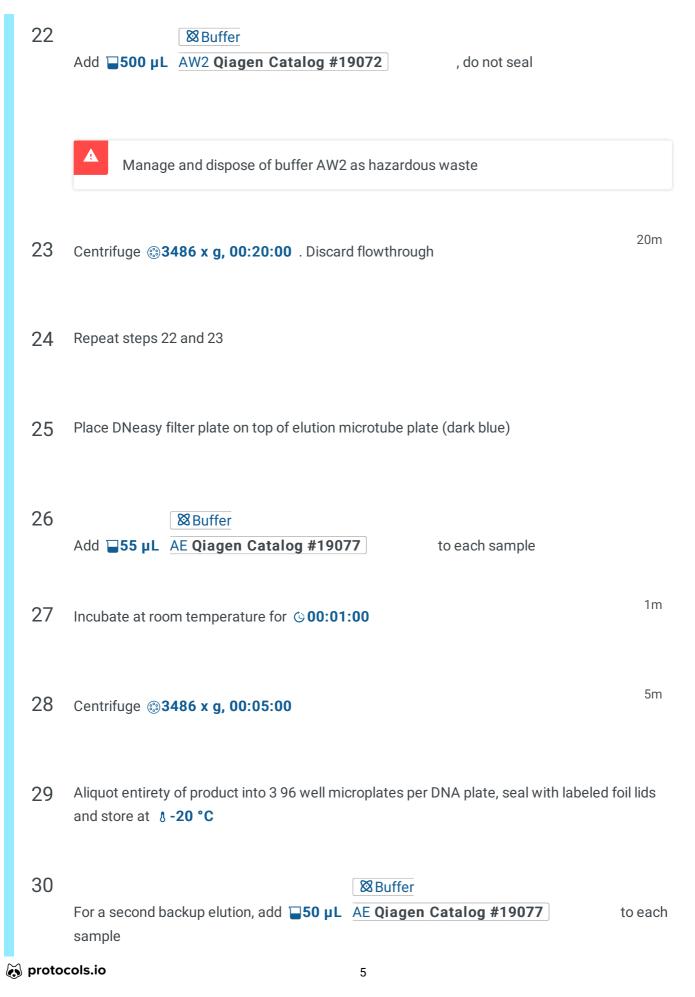
to each sample, seal with AirPore

A

Buffer AW1 contains guanidine salt - do not mix with bleach

21 Centrifuge **3486** x g, 00:08:00 . Discard flowthrough

8m



\sim 4		
31	Incubate at room temperature for	6 00.01.00
o i	incupate at room temperature for	(900.01.00

1m

32 Centrifuge **3486** x g, 00:05:00

5m

33 Seal with white strip caps

34 Store at 8 -20 °C

