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# Extraction Method E (PRP)

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Gen-IALFirst All-tissue DNA extraction kit -This protocol provides an efficient DNA extraction and purification of fresh sample (tissue material)

DOI

[dx.doi.org/10.17504/protocols.io.inecdb](https://dx.doi.org/10.17504/protocols.io.inecdb)

<https://doi.org/10.1093/gigascience/gix053>

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<https://dx.doi.org/10.17504/protocols.io.inecdb>

protocol

Salleh FM, Ramos-Madriral J, Peñaloza F, Liu S, Mikkel-Holger SS, Riddhi PP, Martins R, Lenz D, Fickel J, Roos C, Shamsir MS, Azman MS, Burton KL, Stephen JR, Wilting A, Gilbert MTP, An expanded mammal mitogenome dataset from Southeast Asia. GigaScience 6(8). doi: [10.1093/gigascience/gix053](https://doi.org/10.1093/gigascience/gix053)



## Supporting data for "An expanded mammal mitogenome dataset from Southeast Asia"

\_\_\_\_\_ protocol ,

Jun 26, 2017

Mar 09, 2018

Jun 26, 2017  Anita Broellochs protocols.io

Jul 03, 2017  Gigascience Database GigaScience

6566

Part of collection

Supporting data for "An expanded mammal mitogenome dataset from Southeast Asia"

### MATERIALS




 Isopropanol **Contributed by users**

 Proteinease K **Contributed by users**

 Ethanol **Contributed by users**





Clean

### Extraction

- 1 Cut tissue into small pieces.
- 2 Add 500  $\mu$ l Lyse 1, 50  $\mu$ l Lyse 2 and 10  $\mu$ l Proteinase K.  
 **50  $\mu$ l Lyse 2**  
 **10  $\mu$ l Proteinase K**
- 3 Incubation in a shaker at 65°C for 12 hours.  
 **12:00:00 Incubation at 65°C**
- 4 Centrifuge for 10 min. 13000rpm.

 **00:10:00 Centrifugation**

- 5 Discard supernatant without disturbing the pellet.
  
- 6 Add 375  $\mu$ l Lyse 3  
 **375  $\mu$ l Lyse 3**
  
- 7 Vortex for 20 sec.  
 **00:00:20 Vortex**
  
- 8 Leave it for 5 min in the freezer (-20°C).  
 **00:05:00 Freezer**
  
- 9 Centrifuge 20 min. 13000rpm.  
 **00:20:00 Centrifugation**
  
- 10 Transfer the supernatant to a new tube.
  
- 11 Add 640  $\mu$ l Isopropanol.  
 **640  $\mu$ l Isopropanol**
  
- 12 Mix by inversion.
  
- 13 Centrifuge for 15 min 13000 rpm.  
 **00:15:00 Centrifugation**
  
- 14 Remove the supernatant (don't touch the pellet).

- 15 Add 150  $\mu$ l Ethanol.  
 **150  $\mu$ l Ethanol**
- 16 Centrifuge for 5 min 13000 rpm.  
 **00:05:00 Centrifugation**
- 17 Remove the Ethanol with a pipette (don't touch the pellet).
- 18 Try the pellet (37°C open lid max 5 min).  
 **00:05:00 Drying**
- 19 Re-suspend the pellet in 100  $\mu$ l ddH<sub>2</sub>O water.  
 **100  $\mu$ l ddH<sub>2</sub>O**
- 20 Prior to library construction, analyze small aliquots of each extract on Nanodrop (Thermo Fischer Scientific, Darmstadt, DE) for estimation of DNA concentration.