



Mar 24, 2023

Version 3

DNA extraction (BOMB) V.3

DOI

<https://dx.doi.org/10.17504/protocols.io.n2bvj6mdnlk5/v3>

Tsu-Chun Hung¹, Yin-Tse Huang¹

¹KMU



Yin-Tse Huang

Kaohsiung Medical University

Create & collaborate more with a free account

Edit and publish protocols, collaborate in communities, share insights through comments, and track progress with run records.

Create free account

OPEN  ACCESS



DOI: <https://dx.doi.org/10.17504/protocols.io.n2bvj6mdnlk5/v3>

Protocol Citation: Tsu-Chun Hung, Yin-Tse Huang 2023. DNA extraction (BOMB). **protocols.io**
<https://dx.doi.org/10.17504/protocols.io.n2bvj6mdnlk5/v3> Version created by **Yin-Tse Huang**

License: This is an open access protocol distributed under the terms of the **Creative Commons Attribution License**, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited

Protocol status: Working

We use this protocol and it's working

Created: March 24, 2023



Last Modified: September 13, 2023

Protocol Integer ID: 79371

Keywords: dna extraction, bomb, dna, extraction

Abstract

DNA extraction (BOMB)



Sample Collection

3m

- 1 Add  200 μL of **1mm beads** to 2ml eppendorf tube
- 2 Add  200 μL of **0.5mm beads** to 2ml eppendorf tube
- 3 Add  867 μL Lysis master mix to 2ml eppendorf tube

Note

In 4°C fridge

Lysis master mix: **225 μL** of TE buffer + **375 μL** of lysis buffer + **267 μL** of 10M ammonium acetate

- 4 Collect  10-20 mg of **sample** to 2ml eppendorf tube

1m

Sample crush

4m

- 5 Put 2ml eppendorf tube in mixmill for sample crush, at this condition: 30 rpm/s, for 4mins
 00:04:00

4m

Centrifugation

3m

- 6 Put 2ml eppendorf tube in centrifuge for centrifugation, at this condition:
 17.0 x g, 25°C, 00:03:00

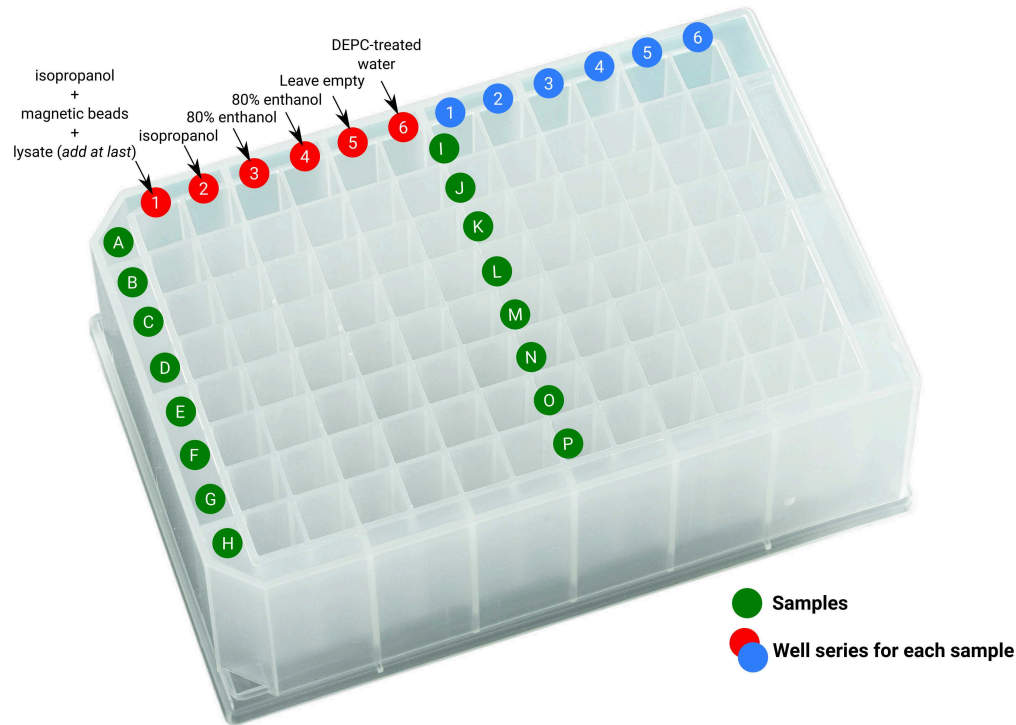
3m

DNA purification

37m 30s

- 7 Add  350 μL of **isopropanol** to the 1st well of 96 well plate

30s



7.1 Add 125 μL of **magnetic beads** (10 mg/ml) to the 1st well of 96 deep well plate

30s

Note

Vortex the bottle and pipetting before using magnetic beads; re-do vortex after adding to 3 samples to prevent set down of magnetic beads

7.2 Add 200-300 μL of the **sample (lysate)** from the 2ml centrifuged tube to the 1st well of 96 deep well plate

30s

Note

Pipetting **as much lysate as you can**, as long as it's free of any cell debris (no solids in your tip)

ADD at LAST

8 Add 400 μL of **isopropanol** to the 2nd well of 96 deep well plate

30s



- 9 Add  300 μL of **80% ethanol** to the 3rd well of 96 deep well plate 30s
- 10 Add  300 μL of **80% ethanol** to the 4th well of 96 deep well plate 30s
- 11 Add  100 μL of **DEPC-treated water** to the 6th well of 96 deep well plate 30s
- 12 Put the prepared 96 deep well plate in the automated DNA extraction machine and select the BOMB protocol 34m
- 13 After the extraction is done, collect  50-100 μL of the **eluted sample** as the DNA template for downstream experiments