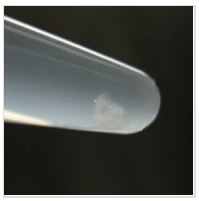

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# DNA Extraction from Yeast

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Yeast ORFans CURE

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ABSTRACT

This is a "quick and dirty" way to get some genomic DNA out of yeast cells. Not pure enough for many things, but should be fine for a PCR template.

This protocol is adapted from

Blount BA, Driessen MR, Ellis T (2016). GC Preps: Fast and Easy Extraction of Stable Yeast Genomic DNA.. Scientific reports. https://doi.org/10.1038/srep26863

who in turn adapted it from

Lõoke M, Kristjuhan K, Kristjuhan A (2011). Extraction of genomic DNA from yeasts for PCR-based applications.. BioTechniques. https://doi.org/10.2144/000113672

PROTOCOL CITATION

Brian Teague 2022. DNA Extraction from Yeast. **protocols.io** https://protocols.io/view/dna-extraction-from-yeast-cfagtibw

KEYWORDS

dna, extraction, saccharomyces, pcr, lioac, sds

IMAGE ATTRIBUTION

By gskx via Flickr. https://www.flickr.com/photos/gskx/89462961

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GUIDELINES

The centrifugation steps all specify **21000 x g** . If your microcentrifuge doesn't go this high, spin at the fastest speed available.

MATERIALS TEXT

**Equipment**

- Dry bath at 72°C
- Dry bath at 42°C (optional)

**Materials**

- Sterile water

  Lithium Acetate Dihydrate **Sigma**
- **Aldrich Catalog #L4158** Step 1 solution,

  **1 Molarity (M)**

  Sodium dodecyl sulfate **Sigma**
- **Aldrich Catalog #436143-25G** Step 1 solution,

  **10 Mass / % volume**
- Ethanol (100%, Molecular Biology Grade) **Fisher Scientific Catalog #BP2818500** In 2 steps
- Ethanol (100%, Molecular Biology Grade) **Fisher Scientific Catalog #BP2818500** In 2 steps

  solution, **70 % (v/v)**
- TE Buffer **Contributed by users** Step 10

SAFETY WARNINGS

Both lithium acetate and SDS are irritants, particularly in the eyes. Wear appropriate PPE, including safety glasses, lab coats and gloves.

SDS is particularly gnarly if it's inhaled. If you're making a solution from powdered SDS, use a dust mask and/or weigh it out in a hood.

1 Make **100 µL** of yeast lysis solution by mixing the following in a 1.7 ml microcentrifuge tube:

- **70 µL** H2O

  Lithium Acetate Dihydrate **Sigma**
- **20 µL** **Aldrich Catalog #L4158** solution,

[M]1 Molarity (M)

Sodium dodecyl sulfate **Sigma**

- **10 µL** **Aldrich Catalog #436143-25G** solution, **[M]10 Mass / % volume**

2 Choose a yeast colony to analyze and circle it on the bottom of the petri dish. A large one is best.

3 Using a micropipette tip, scrape some of the colony off and resuspend it in the lysis solution. Vortex vigorously until the colony is mixed completely into the lysis solution.

> **THIS IS NOT A CASE WHERE MORE IS BETTER**. Your solution should be slightly cloudy. If it is quite "thick", then try again.

4 Incubate at **70 °C** for **00:05:00** 5m

5 Add **300 µL** of Ethanol (100%, Molecular Biology Grade) **Fisher Scientific Catalog #BP2818500** and vortex briefly. Centrifuge **21000 x g, 00:03:00** . 3m

> Make sure the centrifuge is balanced!

6 Using a P-1000 micropipettor, carefully aspirate the supernatant and discard as biological waste. **Do not disturb the pellet.**

7 Add **500 µL** of **[M]70 % (v/v)** 3m

Ethanol (100%, Molecular Biology Grade) **Fisher Scientific Catalog #BP2818500** to the microcentrifuge tube. Centrifuge **21000 x g, 00:03:00** .

Make sure the centrifuge is balanced!

8 Using a P-1000 micropipettor, carefully aspirate the supernatant and discard as biological waste. **Do not disturb the pellet. Try to get as much of the ethanol off as you can.**

9 Let the pellet dry by leaving it in a **42 °C** dry bath for **00:15:00** . Leave the cap open. 15m

If you don't have a dry bath, just leave the tube (cap open) at room temperature. Extend the time to **00:30:00**

10 Add **100 µL** of TE Buffer **Contributed by users** and vortex to resuspend the pellet.

11 Centrifuge **21000 x g, 00:00:30** to collect the cellular debris at the bottom. The DNA remains suspended in the supernatant. 30s

12 Label the tube and store at **-20 °C** , or proceed directly to PCR.

13 If you need to use this sample again: 30s

- Thaw the sample completely.
- Vortex briefly to resuspend everything.
- Centrifuge **21000 x g, 00:00:30** to (re)collect the cellular debris at the bottom of the tube.