**Zymo DNA MiniPrep Plus Extraction Protocol**

Written by B. Young

Updated: 12.13.2022 M. Studivan

All centrifugation steps are performed at **room temperature** and **15,000 x *g*** unless specified.

Prior to first use, add 1040 µl Proteinase K Storage Buffer to each Proteinase K (20 mg) tube. The final concentration of Proteinase K is ~20 mg/ml. Store at -20ºC after mixing.

1. Transfer a tissue fragment to a 2.0 mL bead tube and add 300 µL of Zymo Shield.
2. Bead-beat for 20-30 min on the vortexer or 30-60 sec on the FastPrep at 6 m/sec.
3. Centrifuge for 1 min, then transfer 300 µL of homogenate to new 2.0 mL tube.
4. Add 150 µl Solid Tissue Buffer (blue) and 10 µl Proteinase K.
5. Vortex for 10-15 sec and incubate at 55 ºC for 1 – 3 hr or until tissue solubilizes.
6. Centrifuge lysate for 1 min, then transfer 350 µL supernatant to 1.5 mL tube.
7. Add 1 volume Genomic Binding Buffer (350 µL) to the sample and vortex 10-15 sec.
8. Transfer to a Zymo-Spin IIC-XLR Column in a collection tube and centrifuge for 1 min. Transfer spin column to a **new** **collection tube**.
9. Add 400 µl DNA Pre-Washto the column and invert twice. Centrifuge for 1 min, pour out the flow-through, and dab the collection tube on a KimWipe.
10. Add 700 µL g-DNA Wash Buffer to the column and invert **only the column** twice. Centrifuge for 1 min, pour out the flow-through, and dab the collection tube on a KimWipe.
11. Add 200 µL g-DNA Wash Buffer to the filter directly (**do not invert**) and centrifuge for 1 min. Transfer spin column to a new 1.5 mL tube.
12. To elute DNA, add 50 µL of nuclease-free water heated to 60 ºC directly to the column, incubate for 5 min, and centrifuge at 19,000 x *g* for 1 min.
13. Store at -20 ºC.

**Zymo DNA Clean & Concentrator-5 Purification Protocol**

Written by B. Young

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All centrifugation steps are performed at **room temperature** and **13,500 x *g*** unless specified.

Prior to first use, add ethanol to buffer concentrates per instructions on bottles.

1. Add 2 volumes (100 µL) of DNA Binding Buffer to each sample and mix by vortexing.
2. Transfer to Zymo-Spin Columns and centrifuge for 30 sec. Pour out the flow-through and dab the collection tube on a KimWipe.
3. Add 200 µL of DNA Wash Buffer to the column and centrifuge for 30 sec. Pour out the flow-through, dab the collection tube on a KimWipe, and **repeat this step.** Transfer spin column to a new 1.5 mL tube.
4. Add 25 µL of nuclease-free water heated to 60 ºC directly to the filter and incubate for 1 min. Centrifuge at 14,000 x *g* for 1 min.
5. Store at -20 ºC.

**MiniPrep Plus Tube Prep: DNA Clean & Concentrator-5 Tube Prep:**

2.0 mL bead tubes with 300 µL Zymo Shield Zymo Spin Column w/ collection tube

2.0 mL tube for homogenate 1.5 mL catch tube

1.5 mL tube for lysate

Zymo-Spin IIC-XLR Column w/ collection tube

New collection tube

1.5 mL catch tube