Materials:

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| * DNA samples with impurities (low 260/230 ratios) * Chloroform-Isoamyl alcohol (24:1) * Isopropanol (ice cold) | * 80% ethanol * 1.5 mL microcentrifuge tubes |

Protocol:

1. Obtain DNA samples, and add ¼ volume of Chloroform-Isoamyl alcohol (24:1)
2. Vortex twice to mix thoroughly.
3. Centrifuge at 5,000 rpm for 8 min.
4. Carefully remove the tubes from the centrifuge. Do not disturb or shake, as this may disrupt the layers that have formed within each tube.
   1. Inside your tubes you will see:
      1. Bottom layer: pure chloroform
      2. Middle layer: impurities (should look hazy/puffy)
      3. Top layer: cleaned supernatant
5. Transfer ONLY the supernatant into a clean tube.
6. Add equal volume of “ice-cold” isopropanol.
7. Invert the tubes a few times, and place into the -20 freezer for 10 min.
8. Spin tubes at 12,000 rpm for 15 minutes to precipitate DNA. The DNA pellet will be at the bottom of the tube.
9. Remove 80-90% of the isopropanol, roto-evaporate to remove the remainder. Keep the pellet!
10. Rinse DNA pellet in 80% ethanol. Roto-evaporate to remove the ethanol and let your pellet air dry (takes up to 45 min.).
11. Precipitate in 30 uL of AE.