**Protocol Name:** DNA CPT v1 - Isolation of DNA from whole blood using BD Vacutainer Cell Preparation Tube with Sodium Citrate

**Notes:**
- Total RNA Isolation must be performed prior to DNA Isolation.
- Initial 5 steps below must be performed prior to DNA Isolation.
- Set constant temperature incubator to 37 °C.

1. Spin tubes containing Trizol and sample at 12,000g, room temperature, for 2 minutes to separate phases.
2. Carefully remove any remaining upper aqueous phase using a pipet.
3. Add 400ul of 100% ETOH, mix by inversion.
4. Spin at 2,000g, 4°C, for 5 minutes.
5. Aliquot supernatant equally into two separate tubes (~500ul each) for the protein extraction.

**DNA Isolation:**
1. Make 0.1M NaCitrate in 10% ETOH (wash buffer).
2. Add 1ml wash buffer to pellet, vortex lightly.
3. Incubate at room temperature for 30 minutes, mix by inversion periodically.
4. Spin at 2,000g, 4°C, for 5 minutes.
5. Remove supernatant and add 1ml of wash buffer.
6. Incubate at room temperature for 30 minutes, mix by inversion periodically.
7. Spin at 2,000g, 4°C, for 5 minutes.
8. Remove supernatant and add 1ml 75% ETOH.
9. Incubate at room temperature for 20 minutes, mix by inversion periodically.
10. Spin at 2,000g, 4°C, for 5 minutes.
11. Remove supernatant, dry down in speed vac on medium heat for ~30seconds.
12. Add 300ul 8mm NaOH, pass the pellet through the pipet tip a few times, and incubate overnight at 37 °C.
13. Pre-spin PLG tube at 14,000rpm for 20seconds. Add equal volume PCL as NaOH (300ul) to sample, vortex, transfer phenol-sample mix to PLG tube, and spin at 14,000rpm for 2 minutes. Transfer top, clear aqueous phase to new 1.5ml tube.
14. Add 0.1 volumes 3M Sodium Acetate (~30ul). Add 1ul 5mg/ml Glycogen. Add 2.5 volumes ice-cold 100% ethanol (~830ul). Incubate 1 hour at -80°C.
15. Spin at 14,000rpm, 4°C, for 20 minutes.
16. Remove and discard supernatant and add 1ml ice-cold 80% ethanol.
17. Spin at 14,000rpm, RT, for 2 minutes.
18. Remove and discard supernatant and add 1ml ice-cold 80% ethanol.
19. Spin at 14,000rpm, RT, for 2 minutes.
20. Remove and discard supernatant, let pellet air dry, and resuspend in 22ul of water.
21. Quantitate on NanoDrop, blank instrument with water. Typical yields from one 8ml CPT Tube range from 10 to 15ug of DNA.