Genomic DNA Preparation from Plant Material

1. Picked one leaf from each plant (approximately 1 inch long or two smaller leaves). Put leaf into an appropriately labeled 1.2 mL microdilution stip-tube (USA Scientific; Cat # 1212-8000) or 1.5 mL microcentrifuge tube.
2. Grind plant tissue well with a stick into a powder, in liquid nitrogen.
3. Remove tubes from liquid nitrogen to warm up before adding extraction buffer.
4. Add 400 uL extraction buffer (see recipe at bottom of page).
5. Stir the sample with the stick and transfer by pouring the sample into a new labeled 1.5 mL tube (not necessary if starting from 1.5 mL microcentrifuge tube.
6. Vortex each tube really well for 30 seconds.
7. Spin tubes at max speed for 5 minutes.
8. Transfer supernatant (~200 uL) to a new tube using a pipet. Do not pour off and avoid the leaf debris.
9. Vortex briefly (~5 seconds) and spin tubes again at max speed for 5 minutes.
10. Transfer supernatant (~180 uL) to a new tube using a pipet (do not pour off).
11. Add an equal amount (180 uL) of isopropanol and vortex well (10 seconds).
12. Incubate at room temperature for 10 minutes.
13. Spin tubes at max speed for 10 minutes.
14. Carefully remove supernatant with a pipette (do not pour off).
15. Wash pellet with 1 mL 80% ethanol. Vortex each sample for ~15 seconds. (this can be a stopping point if necessary: place DNA under EtOH in -20°C overnight and resume the following day)
16. Spin tubes for 2 minutes at max speed and pour off EtOH.
17. Spin again 1 minute and carefully remove all traces of ethanol with a pipette.
18. Air dry tubes for 1 hour. Leave a paper towel over the top.
19. Add 100uL of H2O and resuspend pellet by pipet and vortexing (disrupt the pellet as much as possible).
20. Spin max speed 5 minutes; transfer supernatant (this is the gDNA) to new tube
21. Store gDNA at -20°C.

Extraction Buffer Recipe:

REAGENT	[STOCK]	per/500 mL	per/100 mL
200 mM Tris, pH 7.5	1.0 M	100 mL	20 mL
250 mM NaCl	5.0 M	25 mL	5 mL
25 mM EDTA	0.5 M	25 mL	5 mL
0.5 % SDS	10%	25 mL	5 mL
Water		325 mL	65 mL