The protocol is derived from NEB T3010. The major difference for plants is the addition of a tissue lyser and separation for lysate from tissue in step 3/4.

**Lysis buffer + Proteinase K mix.**

200 uL tissue lysis buffer + 10 uL proteinase K per sample. Make extra for pipetting error.

N = 200 \* N = uL Lysis buffer. For N = 25, take 5000 uL lysis buffer.

10 \* N = uL proteinase K. For N = 25, take 250 uL proteinase K

**Protocol**

1. Set heat block to 56 C & water bath to 60 C. Make lysis buffer/proteinase K master mix & incubate in 60 C until warm.
2. Lyse tissue (10 mg dry tissue) with bead mill @ F:20 for 12 seconds.
3. Add 210 uL lysis buffer mix and incubate at 56 C for at least 1.5 hours (up to 3) in the orbital shaker @ 150 rpm.
4. Centrifuge for 3 minutes at >12,000 rcf to separate plant tissue from extraction buffer.
5. Transfer supernatant to a clean, labelled tube without taking tissue. Use 10 uL pipette.   
   Spin down supernatant for 1 min at >12000 rcf to pellet.
6. Repeat step 4, then proceed to step 6.
7. Add 3 uL RNase A per sample. Vortex to thoroughly mix. Incubate at 56 C for 5 min.
8. Add 400 uL gDNA binding buffer per sample and pulse vortex for 5 to 10 seconds.
9. Transfer ~600 uL lysate to gDNA purification column. Avoid transferring tissue or foam. Avoid touching the upper column area with lysate, tips, or fingers. Spin column once.
10. Centrifuge first for 3 minutes at 1000 rcf to bind DNA to screen.
11. Centrifuge for 1 min. at >12,000 rcf to clear membrane. Discard flow through and collection tube.
12. Label final tube with tube id# and ind\_code.
13. Transfer column to a new collection tube and add 500 uL gDNA wash buffer. Invert to mix wash. Centrifuge for 1 min. at >12,000 rcf, discard flow through. Tap collection tube and column on kimwipe to remove residual ethanol.
14. Repeat wash step 10. Discard flow through and collection tube. Clean column of ethanol if necessary.
15. Place column in a labelled 1.5 mL tube. Add 100 uL of warm (60 C) elution buffer. Centrifuge for 1 min. at max speed.