CTAB method for DNA extraction protocol

1. Take fresh tissue in 2 ml microcentrifuge tube (MCT) and chapped them with seizure
2. Add fresh 400 µl **CTAB** solution buffer with RNAse A (the sample mass should not exceed 50 mg), vortex very well and incubate the samples at 60-65°C during 30-60 min.
3. Add 2 volume 800 µl of chloroform, vortex very well, discard the lower phase (chloroform) contains dissolved polysaccharides.
4. Spin at maximum speed in a microcentrifuge for 2 minutes, transferred the upper aqueous layer to a new 2 ml microcentrifuge tube.
5. Optionally: add 800 µl of chloroform, vortex very well for 1 minute creating an emulsion and incubate the samples at 60-65°C during 30 min. Vortex very well and discard the lower phase (chloroform).
6. Spin at maximum speed in a microcentrifuge for 5 minutes.
7. Transferred the upper aqueous layer to a new 1.5 ml microcentrifuge tube which contains of 400 µl 2-propanol (or 1 ml 96% ethanol), vortex well and centrifuge the tubes at maximum speed in a microcentrifuge for 3 minutes.
8. Discard supernatant and wash pellet by adding 1 ml 70% EtOH, vortex well. Centrifuge at 14,000 rpm for 2 min and discard ethanol.
9. The DNA pellet do not dry and dissolved immediately in 300 µl 1xTE, pH 8.0 at 55°C for 10-20 minutes.

Hexadecyltrimethyl ammonium bromide (CTAB)