RNA Isolation for Tissue using TRIzol V.2

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Protocol status: In development

We are still developing and optimizing this protocol

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- 1 Add 1 mL TRIzol of TRIzol per 30 mg tissue of tissue and homogenize using handheld homogenizer.

- 2 Incubate at 25 °C for 00:05:00 to allow nucleoprotein complexes to dissociate.
- 3 Add 200 µL chloroform of chloroform to each ml of TRIzol carefully and vortex to mix well.
- 4 Centrifuge at max speed for 00:15:00 at 4 °C .
- 5 Carefully remove the top aqueous phase and transfer to a new Eppendorf tube. The interphase and bottom organic phase can be saved for DNA and protein respectively.
- 6 Add 500 µL 100% isopropanol of 100% isopropanol to the aqueous phase, mix by inversion and incubate at -20 °C for a minimum of 02:00:00 .
- 7 Spin down at maximum speed for 30 mins to precipitate RNA.
- 8 Remove supernatant, and add 1 mL of 75 % volume ethanol ethanol to wash pellet.

- 9 Spin down at max speed for 15 minutes and remove supernatant.
- 10 Resuspend pellet in appropriate volume of nuclease free water.