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RNA isolation for tissue V.3

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Protocol status: Working
We use this protocol and it's working

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1 Homogenization in TriZol
Add 1ml 1 mL Trizol per 30 mg tissue and homogenize using handheld homogenizer.

2 Incubate at RT for 00:05:00 to allow nucleoprotein complexes to dissociate.

3 Add 200 µL Chloroform carefully, and vortex to mix well.

4 Spin down at max speed in a chilled centrifuge for 00:15:00 .

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 To the aqueous phase, add 500 µL of 100% isopropanol, mix by inversion and incubate at -20 °C for a minimum for 02:00:00 .

7 Spin down at max speed for 00:30:00 to precipitate RNA.

8 Remove supernatant, and add 1 mL 75% Ethanol to wash the pellet.

9 Spin down at max speed for 00:15:00 and remove supernatant.

10 Resuspend pellet in appropriate volume of nuclease free water.