1. Add 400 uL of lysis solution (20 mM NaAc 3M pH 5.2, 1 mM EDTA, SDS 0,5%, all made in DEPC water) to the filters and incubate them in the water bath at 65 C for 2 min.

2. Add lysis matrix (From [FastDNA extraction kit](http://www.mpbio.com/product.php?pid=116540600) or similar) to the lysate and 1 mL of [TRIzol®](<https://www.thermofisher.com/order/catalog/product/15596026>).

3. Bead beat for 1 min at medium speed (3.5).

4. Add internal standard.

5. Centrifuge 5 min at 14000 x g

6. Transfer the supernatant to a new eppendorf tube.

7. Add 300 uL of chloroform, mix by gentle inversion 20 times, making an emulsion. If mixed more vigorously you can get more RNA, but the sample gets more contaminated with genomic DNA.

8. Incubate at RT for 3 min.

9. Centrifuge at 13000 RPM for 15 min at 4 C.

10. Take aqueous phase, about 800 uL avoiding interphase (organic phase, pink-colored can be used for DNA extraction). Do this on ice.

11. Divide the 800 uL into two 1.5-eppendorf tubes. Add 1 mL of 100% cold ethanol and 40 uL of NaAc 3M pH 5.2 to each tube. Mix the tubes by inversion to avoid freezing the sample.

12. Incubate at -20 for 2 h or at -80 for 15 min. Centrifuge at 12500 RPM for 30 min at 4 C.

13. Wash with 70% cold ethanol. Centrifuge at 12500 RPM for 25 min at 4 C.

14. Extract as much as solvent as possible and resuspend the (probably invisible) pellet in 350 uL buffer RW1 (from [QIAGEN RNeasy® Mini kit](https://www.qiagen.com/us/shop/sample-technologies/rna/rna-preparation/rneasy-mini-kit#resources))

15. Add the resuspended RNA to the spin column from the RNeasy kit. Spin for 15 seconds at 12,000 rpm. Discard flowthrough.

16. Add 10 uL DNAse 1 to 70 uL buffer RDD. Gently invert to mix and centrifuge briefly.

17. Add DNAse mix to column (80 uL) and incubate at room temp for 15 minutes.

18. Add 350 uL of buffer RW1 to the column. Spin for 15 seconds at 12,000 rpm. Discard flowthrough.

19. Add 500 uL of buffer RP1 to the column. Spin for 15 seconds at 12,000 rpm. Discard flowthrough.

20. Add 500 uL of buffer RP1 to the column. Spin for 2 minutes at 12,000 rpm. Discard flowthrough.

21. Place column in new 1.5 mL tube. Add 30 uL of RNAse free water and centrifuge for 1 minute at 12,000 rpm.

22. Quantify using the Qubit kit for RNA.