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Working

Removal of genomic DNA from RNA preparations (Thermo Scientific)- (M4455 Version)

Forked from Removal of genomic DNA from RNA preparations (Thermo Scientific)

Maximilian Dietsch[1]

[1]Heinrich-Heine Universität Düsseldorf

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M4455 - Synthetische Biologie und Biotechnologie

Sebastian Triesch
Institute for Synthetic Microbiology, HHU Düsseldorf

ABSTRACT

Removal of genomic DNA from RNA preparations

PROTOCOL STATUS

Working

We use this protocol in our group and it is working

GUIDELINES

allways waer gloves and work on ice

SAFETY WARNINGS

1

Add to an RNase free tube:	
RNA	1 µg
10X reaction buffer with $MgCl_2$	1 µl
DNase I, RNase-free	1 µl (1U)
Water	to 10 µl

2 Incubate at 37 °C for 30 min

37 °C

00:30:00

3 Add EDTA, Water and PCI and vortex thoroughly.

1 µl EDTA

80 µl Water

100 µl PCI (phenol chloroform isoamyl alcohol)

4 Centrifuge for 10 min at 10000 rpm and 4 °C

5 transfer the upper phase into a fresh tube and add 3 volumen EtOH/ 3M Natrumacetat (30:1, ph 5.2)

6 precipitate RNA over night at -20 °C

7 Centrifuge 30 min at 13000 rpm and 4 ° C

4 °C

00:30:00

8 Discard supernatant and wash pellet with 75% EtOH (do not resuspend the pellet)

9 Centrifuge 10 min at 13000 rpm and 4 °C

4 °C

00:10:00

10 go to step #8 **repeat washing step**

go to step #9 **Centrifuge**

11 Discard supernatant and dry pellet for 10 - 15 min

12 resupend pellet with 30 µl H_20