



Feb 07, 2022

Reverse Transcription of Phage RNA

Frej Larsen¹¹UCPH

1



protocol .

FOOD Micro UCPH



Frej Larsen

Reverse transcription (RT) is the process of turning ssDNA into cDNA using the reverse transcriptase enzyme. This protocol uses the RT reaction to turn ssRNA and dsRNA into ssDNA and is specifically used for virome analysis.

Frej Larsen 2022. Reverse Transcription of Phage RNA. **protocols.io**
<https://protocols.io/view/reverse-transcription-of-phage-rna-b4p4qvqw>



protocol ,

Feb 04, 2022

Feb 07, 2022

57820

SuperScript IV VILO Master Mix from Invitrogen. Can be bought here:

<https://www.thermofisher.com/order/catalog/product/11756500?SID=srch-srp-11756500>

- 1 Add **16 µL** sample to a PCR tube and denature RNA by heating to **95 °C** for **00:05:00** on a ThermoCycler 5m
- 2 Centrifuge for **00:00:10** and put on ice 10s
- 3 Add **4 µL** of the 5X master mix to the PCR tube, then run the following program on a ThermoCycler: 25m

- 🔥 25 °C for ⌚ 00:10:00 (primer annealing)
- 🔥 50 °C for ⌚ 00:10:00 (reverse transcription)
- 🔥 80 °C for ⌚ 00:05:00 (enzyme inactivation)

4 Centrifuge for ⌚ 00:00:10

10s

5 Purify by bead cleaning and elute in 📄 10 µL MQ water