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cDNA synthesis

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1 Works for me



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dx.doi.org/10.17504/protocols.io.kqdg3948qg25/v1

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ABSTRACT

High-Capacity cDNA Reverse Transcription Kits(#4368814, Applied Biosystems - ThermoFisher)

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- 1 Prepare the 2X master mix (ul per sample):
 - 2ul RT Buffer
 - 0.8ul 25X dNTP Mix(100mM)
 - 2ul Random primers
 - 1ul Multiscribe Reverse Transcriptase

- 4.2ul Nuclease free water

- 2 Take 0.5ug of RNA in a maximum of 10ul of sample volume
- 3 Mix the 2X master mix (10ul) with the RNA samples (10ul) and mix by pipetting
- 4 Centrifuge the plates to spin down all the reagents.
- 5 Keep the samples on ice
- 6 Program the thermal cycler to 25C for 10min 37C for 120 minutes 85C for 5 minutes and 4C until finished.
- 7 Store the cDNA samples at -20C for short-term storage or -80C for long-term storage (years).