

RNA 3'-end Labeling by Ligation

This protocol is for the RNA 3'-end Labeling by Ligation

1. Prepare the following in a single RNase-free microfuge tube:

10X ligation buffer for T4 RNA Ligase	2 μ l
10 mM ATP	1 μ l
RNA	50-100 pmol
[³²P]-pCp	50-100 pmol (equimolar amount)
T4 RNA Ligase	1 μ l (10 u)
DEPC-treated Water	to 20 μ l
Total volume	20 μl

2. Incubate at 4°C for 10-12 hours (overnight).
3. Separate labeled RNA from unincorporated label by gel filtration on Sephadex G-50.

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