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EcoQprep™ mRNA Kit

- 50 rxn

K-3708

Name	Volume	Quantity	Name	Volume	Quantity
MRD Buffer	30 mL	1 ea	1.5mL Tube	50 ea	1 pack
MRW1 Buffer	40 mL	1 ea	One Page Protocol	-	1 ea
MRW2 Buffer	50 mL	1 ea			
MRW3 Buffer	50 mL	1 ea			
EM Buffer	25 mL	1 ea			
Magnetic Nano Bead-Oligo dT	3 mL	1 ea			

Before Start

Components	Before Use	Storage	Symbol
MRD Buffer	Before each experiment, mix MRD buffer with 1 μ L of β -mercaptoethanol per 1 mL of MRD buffer.	Do not store the prepared solution; freshly prepare it for each experiment.	Run centrifuge
All reagents	MRD Buffer, MRW1 Buffer, MRW2 Buffer, MRW3 Buffer, EM Buffer, Magnetic Nano Bead-Oligo dT	After opening, store at 2-8°C	Mix the solution as instructed Caution Heating Add the following solution

Protocol

NOTE Do not vortex after adding the solution; mix by pipetting or inverting instead.

Sample	Pre-treatment & Lysis	1 Bead Wash	2 Binding	3 1st Wash	4 2nd Wash	5 3rd Wash	6 Elution
Cultured cells	Suspension cell culture Cell pellet: $\sim 1 \times 10^6$ cells 300 xg, 5 min 300 μ L MRD with β -mercaptoethanol	50 μ L Bead to a new tube. 100 μ L MRD with β -mercaptoethanol by vortexing 1) Attach magnet	the Lysate to the tube containing bead by pipetting 1) Attach magnet	600 μ L MRW1 by pipetting 1) Attach magnet	300 μ L MRW2 by pipetting 1) Attach magnet	700 μ L MRW3 by pipetting Add to the Opposite side of the bead Do Not Vortex 1) Gently Invert 3~4X	15 - 20 μ L EM by pipetting 65°C, 2 min 1) Attach magnet
	Monolayer cell culture A) Direct harvest Discard cell culture medium 300 μ L MRD with β -mercaptoethanol B) Trypsin harvest Detach cells 300 xg, 5 min 300 μ L MRD with β -mercaptoethanol	1) Attach magnet 2) Discard sup	2) Invert 3~4X 3) Discard sup	2) Invert 3~4X 3) Discard sup	2) Invert 3~4X 3) Discard sup	2) Discard sup	2) Transfer the eluent
Plant tissue	Tissue : ~ 20 mg Grind in LN ₂ 300 μ L MRD with β -mercaptoethanol per 10 mg tissue full speed, 3 min Transfer the supernatant	3) Detach magnet	4) Detach magnet	4) Detach magnet	4) Detach magnet	3) Detach magnet	Immediately transfer the eluent to a new tube.
Animal tissue	Homogenize sample : ~ 10 mg 300 μ L MRD with β -mercaptoethanol full speed, 3 min Transfer the supernatant				300 μ L MRW2 Repeat 1) ~ 3) Do Not Detach magnet at the final step		

Ensure that no supernatant remains when discarding it, as any residual supernatant may affect the purity.