

Automated RNA Extraction (MagMax Kit) Reagent Preparation



Category	Experimental Procedures
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Version	6

Labels:

Step 1 - Number of Reactions

Determine the number of deep well plates to be run on the RNA Extraction Hamilton Microlab Star.

The ONLY value that needs to be edited is the Total # of Plates

1Total # Plates (number between 1 and 4)

Step 2 - Binding Bead Mix

Vortex or invert stock magnetic beads to ensure that the bead mixture is homogenous.

Prepare the Binding Bead Mix according to the table below. Mix well by inversion, centrifuge to remove bubbles (<1,500 rpm for 1 minute), and then store at room temperature. Prepare a Binding Bead Mix trough using robotic reservoir.

You will need a total of 2 Binding Bead Mix Conical Tubes

Prepare the Binding Bead Mix in a 50 mL Centrifuge Tube as follows:

Binding Bead Mix per 50 ml Conical Tube		
Component	Volume Per Well	Total Volume in each 50 ml Conical
Binding Solution	265 µL	31.8 ml
Magnetic Beads	10 µL	1.2 ml
Total Volume	275 µL	33 ml

Step 3 - Wash Buffer

Measure out the total required Wash Buffer for the number of reactions. Prepare a Wash Buffer trough using a robotic reservoir.

Total Wash Buffer Plus Overage

68 ml

Step 4 - Elution Buffer

Measure out the total required Elution Buffer for the number of Reactions. Prepare a Elution Buffer 60 mL reservoir (select appropriate reservoir dependent on automation or manual)

Total Elution Buffer Plus Overage

8.3 ml

Step 5 - Nuclease Free Water

Measure out the total required Nuclease Free Water for the number of Reactions. Prepare a Water trough using a reservoir.

Total Water Plus Overage

87.2 ml

Step 6 - 80% Ethanol

Prepare 80% ethanol required using Ethanol, Absolute, Molecular Biology Grade, and Nuclease-free water (not DEPC-Treated) for the required number of reactions. For each 200 μ L sample input volume, prepare 750 μ L of 80% ethanol. Prepare a 80% Ethanol trough using robotic reservoir.

First Wash- Reservoir 1: 80% Ethanol Needed Plus Overage

68 ml

Second Wash- Reservoir 2: 80% Ethanol Needed Plus Overage

68 ml

Third Wash - Reservoir 3: 80% Ethanol Needed Plus Overage

88 ml

Step 7 - Proteinase K

Using the 8-channel pipettor, manually transfer 5 μ L of Proteinase K into each well (with sample, NEC, or NTC) of the aliquoted 96-well extraction plate, other than A1.

980 μ L

Note: For automation, A1 is excluded in the PCR prep step during the extraction plate transfer.

Attachments

No file attachments

*This procedure was originally created by **Lena Landaverde***

