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# **User's Manual and Instructions**

## MagSeg mRNA Purification Kit

Catalog No.: K2012008

#### Introduction

MagSeq mRNA Purification Kit is specifically designed for mRNA isolation from Total RNA by magnetic beads.

#### **Features**

- Quick mRNA purification from Total RNA in less than an hour
- mRNA for next generation RNA sequencing or cDNA synthesis

#### Description

MagSeq mRNA Purification Kit contains reagents, including oligo (dT) magnetic beads, which is enough for 8 mRNA isolation. The kit's set up is perfectly compatible for next generation sequencing instrument, such as Illumina's HySeq, using Flow Cells with 8 channels.

#### **Kit Contents**

MagSeq mRNA Purification Kit	Part No.	Amount	Storage
MagSeq Oligo (dT) Solution	K2012008-1	900 µl	4°C
Bead Washing Solution	K2012008-2	1 ml x 7	4°C
Resuspension Solution	K2012008-3	500 µl	4°C
DEPC Water	K2012008-4	1 ml	4°C

#### Protocol

# mRNA purification (vortex beads vigorously in each step to re-suspend beads and wash contaminates off)

- 1. Dilute 1~20 µg total RNA with DEPC Water to 50 µL in a 1.5 mL RNase free non-sticky tube
- 2. Heat at 70°C for 2 minutes to disrupt the secondary structures, and place on ice
- 3. Aliquot 50 µL of MagSeq Oligo(dT) Solution
- 4. Shake at RT for 5 minutes
- 5. Place the tubes on the magnetic stand for at least two minutes and remove the supernatant
- 6. Wash the beads twice with 200 µL of Bead Washing Solution. Vortex well to re-suspend beads. Place the tubes on the magnetic stand for at least two minutes and remove the supernatant
- 7. Add 50 µL of Resuspension Solution to the beads, heat at 70°C for 2 minutes to elute mRNA from the beads
- 8. Meanwhile, aliquot 50 µL MagSeq Oligo(dT) Solution to a new tube
- After heating the bead and mRNA at 70°C for 2 minutes, immediately put it on the magnet stand for at least two minutes and then transfer the supernatant (mRNA) to the tube from step 8; shake RT for 5 minutes
- 10. Place the tubes on the magnetic stand for at least two minutes and remove the supernatant
- 11. Wash the beads twice with 200 µL of Bead Washing Solution. Vortex well to re-suspend beads. Place the tubes on the magnetic stand for at least two minutes and remove the supernatant.
- 12. Add 20 µL of DEPC Water to the beads, then heat at 70°C for 2 minutes to elute mRNA from the beads. Immediately put on the magnet stand for two minutes and transfer the supernatant (mRNA) to a fresh 200µL thin wall PCR tube, and there should be ~19µL of mRNA solution.
- 13. Determine mRNA isolation by UV, and use appropriate amount for sequencing or cDNA synthesis.