Automated DNA and RNA Extraction Solution on Hamilton Microlab® STAR™ Platform

Sequential isolation of DNA and RNA in two fractions from up to 10 mg tissue sample or 1 x 10^6 cells.

Kit

Mag-Bind® DNA/RNA Kit (M6932)

Sample Types

Mouse Brain, Mouse Liver, and AD293 Cells

Sample Amounts

10 mg (Brain & Liver), ~1 x 10^6 Cells

Analysis

Spectrophotometry using NanoDrop™ 2000c, RT-qPCR, qPCR & TapeStation

Materials

Mag-Bind® DNA/RNA Kit (M6932)
Homogenization Materials:

- 96-well Plate, 2 mL capacity (ThermoFisher Scientific)
- Silicone Mat for Homogenization Plate (ThermoFisher Scientific)
- 2.8 mm Ceramic Beads (Omni International)

For a complete list of system components and plastic consumables on Hamilton Microlab® STAR™, please contact us at automation@omegabiotek.com

Methods

Automated method developed by Omega Bio-tek for the Hamilton Microlab® STAR™

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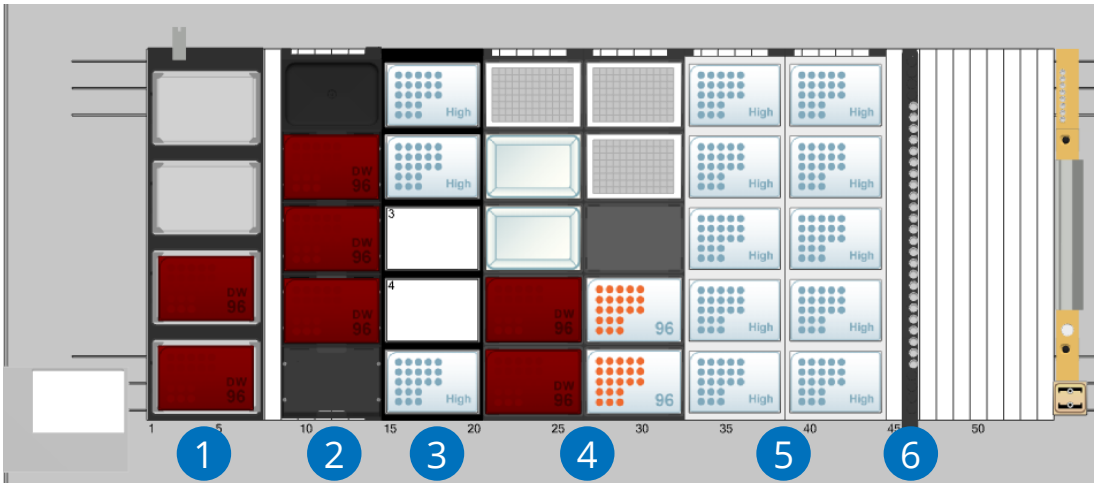
Hamilton Microlab® STAR™ Deck Layout



Component	Purpose
1. HHS Baseplate with 4x HHS units (3 mm orbital with flat bottom adaptors).	Heater/Shakers for bead resuspension and incubation steps.
2. MFX Carrier with 4x tall labware locators & liquid waste module for MPH.	For processing plate positions, magnet (Alpaqua Magnum FLX) & liquid waste module
3. Tip Isolator Carrier	For tips reused for tip mixing and liquid waste removal steps.
4. 2x DWP Stands	For processing plate positions, reagent reservoirs, and elution plate.
5. 2x Standard Tip Carriers	Tips for reagent dispensers.
6. Sample Tube Rack Carrier	Holds reagents.

Figure 1. Hamilton Microlab® STAR™ deck layout for automated DNA and RNA extraction.

Results

The Mag-Bind® DNA/RNA Kit was used to extract DNA and RNA in two fractions from 10 mg of mouse brain and liver tissue samples, in addition to ~1x10^6 AD293 cells. Extractions were carried out in triplicate, following manufacturer's instructions, as well as automated using the Hamilton Microlab® STAR™. Thermo Scientific's NanoDrop™ 2000c spectrophotometer was used to quantify the yields and purities of isolated DNA and RNA from both extraction methods. DNA and RNA yields, as well as purity, for manual and automated extraction methodologies are as shown in **Figures 2 and 3**, respectively. The yields and purity ratios of DNA and RNA extracted using the automated protocol closely resemble that of the manual protocol, validating the automation method developed on the Hamilton Microlab® STAR™. For both manual and automated methods, the absorbance ratio of A260/A280 was consistently between 1.84-1.87, indicating pure DNA, and between 2.02-2.08, indicating pure RNA.

DNA Yield and Quality from Brain, Liver, and Cells

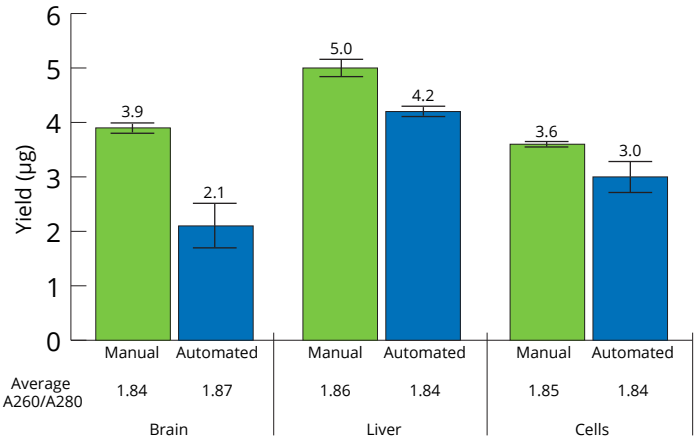


Figure 2. DNA was extracted from 10 mg brain and liver tissue, as well as 1x10^6 cells, both manually and using the Hamilton Microlab® STAR™. The average yields were comparable for liver and cultured cells between extraction methods and lower for brain samples using automated methodology. The average A260/A280 ratios were comparable between extraction methods, regardless of sample type.

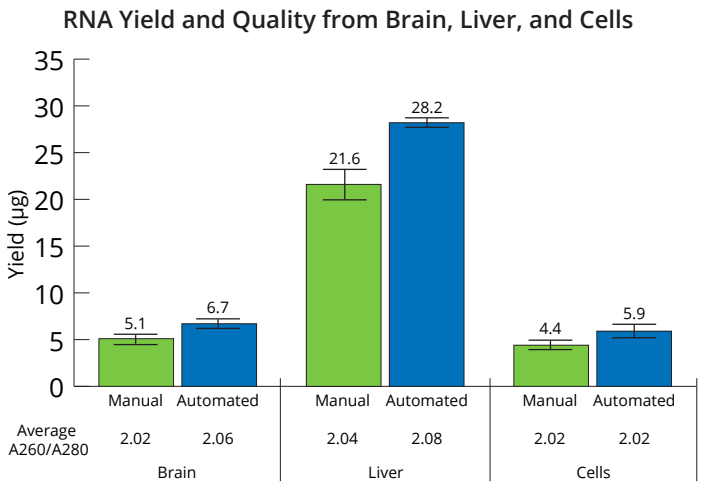


Figure 3. RNA was extracted from 10 mg brain and liver tissue, as well as 1x10^6 cells, both manually and using the Hamilton Microlab® STAR™. The average yields and A260/A280 ratios of RNA were comparable between extraction methods, regardless of sample type.

qPCR and RT-qPCR reactions were conducted with 10-fold dilutions of extracted DNA and RNA, respectively. Average ΔCts between 10-fold and 100-fold dilutions was ~3.3, indicating no inhibition and were comparable between extraction methods and sample types **(Figure 4)**.

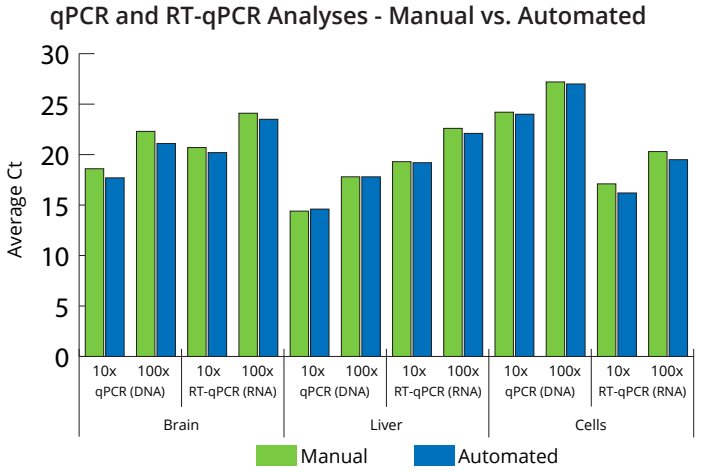


Figure 4. DNA and RNA were extracted from 10 mg liver and brain tissues, as well as 1x10^6 cells, both manually and automated on the Hamilton Microlab® STAR™. Average ΔCts between 10-fold and 100-fold dilutions were comparable between extraction methods.

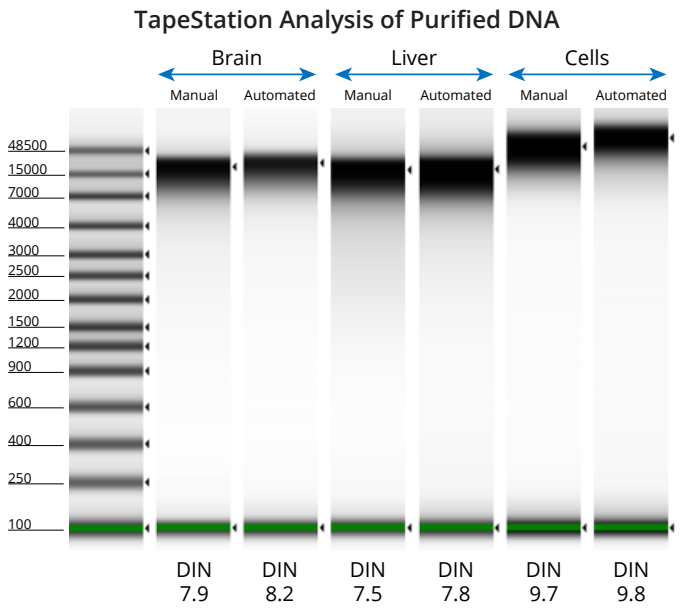


Figure 5. DNA was extracted from brain, liver, and cell samples using the Hamilton Microlab® STAR™. The Mag-Bind® DNA/RNA Kit extracted DNA > 48 kb from cultured cell samples.

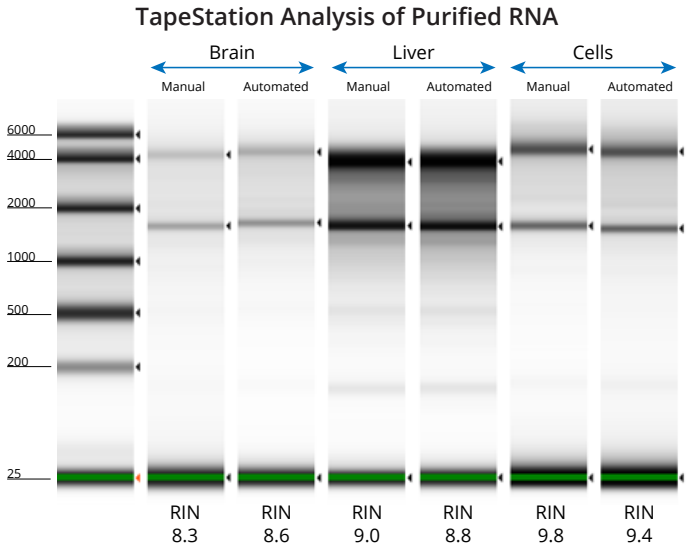


Figure 6. RNA was extracted from brain, liver, and cell samples using the Hamilton Microlab® STAR™. RNA extracted from cultured cells was of high integrity.

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Both manual and automated methodologies resulted in high-quality DNA and RNA, as illustrated by high DIN (DNA Integrity) and RIN (RNA Integrity) scores when analyzed on TapeStation® (**Figures 5 and 6**, respectively). DIN scores ranging from 7.8-9.7 and RIN scores ranging from 8.3-9.8, depending on the sample type, indicate high integrity of nucleic acids, underscoring their applicability in various downstream applications, including Next Generation Sequencing (NGS). In addition, this workflow also extracted high molecular weight DNA > 48 kb from cultured cell samples (**Figure 5**).

Conclusions

From the data presented here, it can be seen that the Mag-Bind® DNA/RNA Kit (M6932) is capable of extracting DNA and RNA in two fractions from the same sample. High yields of high-quality DNA and RNA were extracted from each sample type using the Hamilton Microlab® STAR™, providing users with a high throughput, automated solution for integrated insights into the genome (using DNA) and transcriptome (using RNA) in a single workflow.

Product Information

Description	Product No.	Size
Mag-Bind® DNA/RNA Kit	M6932-01	1 x 96
	M6932-02	4 x 96

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