Universal Plus mRNA-Seq

with NuQuant[®]

PRODUCT SHEET



From sample to quantified libraries – a complete solution for mRNA-Seq

The Universal Plus mRNA-Seq kit with NuQuant is a novel. streamlined library preparation solution for mRNA sequencing on Illumina sequencers. The kit's integrated NuQuant technology - a proprietary library quantification method for efficient and accurate quantification of next generation sequencing (NGS) libraries eliminates the need for timeconsuming and inaccurate methods such as qPCR, fluorometry or microfluidic electrophoresis, allowing RNA-

Seq libraries to be constructed and quantified in a single day.

Why use Universal Plus mRNA-Seq with NuQuant?

Unique Dual Indexes (UDI) and Molecular Tags:

up to 384 pre-plated UDI adaptors with molecular tags are provided with the kit.

Broad dynamic input range: inputs as low as 10 ng enable use of previously inaccessible samples.

DimerFree® adaptor ligation technology: for fewer adaptor dimers and more mapped reads without adaptor dilution **AnyDeplete**[®]: for optional targeted depletion after library construction

- Maximizes informative reads without manipulation of starting RNA
- No minimum input requirements for depletion
- Standard target depletion for human and fetal globin, or complete customization available

NuQuant library quantification: providing fast and easy determination of library molarity in minutes.



Figure 1: Universal Plus mRNA-Seq with NuQuant library preparation kits provide end-to-end solutions from total RNA to quantified strand-specific libraries ready for sequencing (including library QC).



Figure 2: Universal Plus mRNA-Seq with NuQuant library prep kit generates consistent, high quality data with input ranges of 10 ng to 1 µg. Robust gene expression and even gene coverage was observed across a 100X fold difference in sample input.



Figure 3: Libraries were generated with Universal Plus mRNA-Seq with NuQuant and competitor kits starting with 100 ng of total RNA. The Universal Plus libraries provided similar metrics compared to a kit from Competitor I and better performance compared to a kit from Competitor K. Libraries generated with a kit from Competitor K, demonstrated lower exon coverage and higher percent of rRNA and other (intron, intergenic, and mitochondrial) reads.

The Universal Plus mRNA-Seq with NuQuant library prep kit can be used to generate RNA-Seq libraries from sample inputs of 10 ng to 1 µg. Figure 2 highlights the consistent performance across a broad input range.

Figure 3 shows that the performance of the Universal Plus mRNA-Seq with NuQuant kit is comparable or superior to competitor kits. An external comparison between the Universal Plus mRNA-Seq with NuQuant kit and the a kit from Competitor I demonstrated that Universal Plus libraries had better 5' transcript coverage (Figure 4), with a similar FPKM concordance and gene detection despite a 50-fold lower input.



Figure 4: The Universal Plus mRNA-Seq with NuQuant kit shows better representation of the 5' transcript ends compared to libraries constructed with Competitor I. The Universal Plus mRNA-Seq kit provides consistent 5' transcript coverage across a broad input range, even when starting with 10 ng of total RNA. Even with 500 ng of the same RNA input, the data generated with the Competitor I kit had poor 5' end coverage of the transcript.



Figure 5: Universal Plus mRNA-Seq with NuQuant libraries generated with 10 ng of total RNA were compared to Competitor I libraries generated with 500 ng total RNA. FPKM analysis showed high concordance between the data despite the 50-fold difference in input (left figure). Additionally, both data sets detected a similar number of genes (right figure). These figures demonstrate that the Universal Plus mRNA-Seq with NuQuant kit can produce high quality data even when starting with just 10 ng of total RNA.

The Universal Plus mRNA-Seq with NuQuant kit offers the flexibility to add a customizable transcript depletion step with AnyDeplete. This provides more biological information by increasing the informative data from each mRNA sequencing experiment. The kit can be integrated with fetal and adult globin AnyDeplete to increase the number of informative reads from your whole blood sequencing data or customized to your sample type. The kit's integrated NuQuant library quantification accurately measures molar library concentration without the need for separate fragment size analysis. This eliminates the need for other library QC methods, such as qPCR or fluorometric mass measurements.



Sample	Number of RefSeq Genes with FPKM>1
Without AnyDeplete	8326
Adult Globin AnyDeplete	8975
Fetal + Adult Globin AnyDeplete	9388

Figure 6: Addition of fetal + adult globin AnyDeplete to the Universal Plus mRNA-Seq with NuQuant library preparation protocol leads to a significant increase in the number of informative reads, allowing detection of more genes with the same sequencing depth.



Figure 7: Comparison of library quantification methods. NuQuant is integrated into the kit and provides simple fluorescence-based determination of molar library concentration in minutes, without additional cost.

Key Benefits

Ordering information

- Flexible multiplexing with up to 384 UDI adaptors and molecular tags
- Broad dynamic input range, from 1 μg to as low as 10 ng, enables Illumina sequencing of previously inaccessible samples
- Fewer bead purification steps ensures efficient and streamlined library preparation for any sample type
- Even 5' to 3' transcript coverage
- Integrated target depletion with AnyDeplete (Adult and Fetal globin depletion available)
- Save time and costs on library QC with NuQuant
- Automation solutions available for Tecan and other platforms

Product Name	Part no.	No. of reactions	Barcodes
Universal Plus mRNA-Seq with NuQuant	0520-24	24	1-24
	0520-A01	96	1-96
	0520B-A01	96	97-192
	0520C-A01	96	193-288
	0520D-A01	96	289-384
Universal Plus mRNA-Seq with NuQuant, Human Globin AnyDeplete	0521-24	24	1-24
	0521-A01	96	1-96
	0521B-A01	96	97-192
	0521C-A01	96	193-288
	0521D-A01	96	289-384
Universal Plus mRNA-Seq with NuQuant, custom AnyDeplete*	0522-A01	96	1-96
	0522B-A01	96	97-192
	0522C-A01	96	193-288
	0522D-A01	96	289-384

*Contact your local Tecan sales representative for more information.

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