

NEXTFLEX® SMALL RNA-SEQ KIT v3

DNA-SEQ · RNA-SEQ · BARCODES · METAGENOMICS · AMPLICON PANELS · TARGET CAPTURE · EPIGENETICS · SMALL RNA-SEQ

Automated Gel-Free or Low Input Small RNA Library Prep

The NEXTFLEX® small RNA-seq kit v3 uses patented and patent-pending technology to provide a reduced-bias small RNA library preparation solution for Illumina® sequencing platforms with gel-free or low-input options. Our approach to reducing ligation-associated bias involves the use of adapters with randomized bases at the ligation junctions, resulting in greatly decreased bias in comparison to standard protocols. This reduction in bias results in data that more accurately represent abundances of small RNAs in the starting material. In addition, reduction of bias allows more miRNAs to be detected with fewer total reads, increasing efficiency and reducing cost for small RNA sequencing.

The NEXTFLEX small RNA-Seq kit v3 also allows for gel-free small RNA library preparation. This is possible thanks to the dual approach used for adapter-dimer reduction. Unprecedented reduction of adapter-dimer formation allows completely gel-free small RNA library prep when starting with ≥ 200 ng of total RNA, which in turn allows for a fully automated solution on the PerkinElmer® Sciclone® G3 NGS Workstation and Zephyr® G3 NGS Workstation.

KEY FEATURES

- Completely automated protocol with inputs ≥ 200 ng total RNA
- Greater discovery/detection rates decrease sequencing cost
- Substantially reduces ligation-associated bias
- Automated on the PerkinElmer® Sciclone® G3 NGS workstation and Zephyr® G3 NGS workstation

	NEXTFLEX® Small RNA-Seq Kit v3	TruSeq® Small RNA Library Prep Kit	NEBNext® Small RNA Library Prep Kit	QIAseq® miRNA Library Prep Kit
Randomized Adapters for Reduced Bias	Yes	No	No	No
Gel-free Protocol	Yes	No	Yes	Yes
Automation Script Available	Yes	No	No	No
All Enzymes Included	Yes	No	Yes	Yes

For research use only. Not for use in diagnostic procedures.

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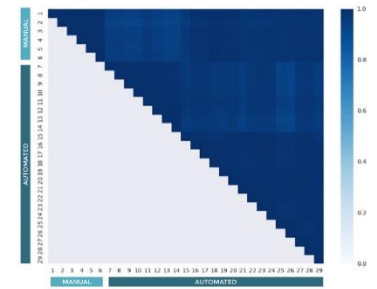
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Sequencing Metrics

	Reads Processed	Insert <15 bp (%)	Reads Passing Filter	miRBase Alignment Rate of Filtered Reads (%)	miRBase Alignment Rate of All Reads (%)	miRNA Groups Detected
Manual Average	143287.0	23.9	108982.5	61.9	47.1	376.2
Manual SD	0.0	4.0	5736.2	2.7	3.5	14.4
Automated Average	143287.0	21.8	112116.8	60.1	47.0	392.6
Automated SD	0.0	3.4	4838.9	4.9	3.3	10.7



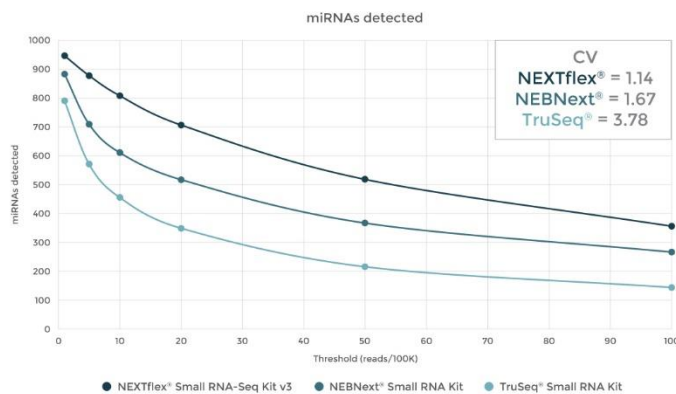
Pair-wise correlation of miRNA expression in replicate libraries created manually and on the PerkinElmer® Sciclone® NGS Workstation.

“We recommend using the BioScientific NEXTflex kit, as it detects the largest number of miRNAs, owing to its 4 N random adaptor sequence that ameliorates ligation bias.”

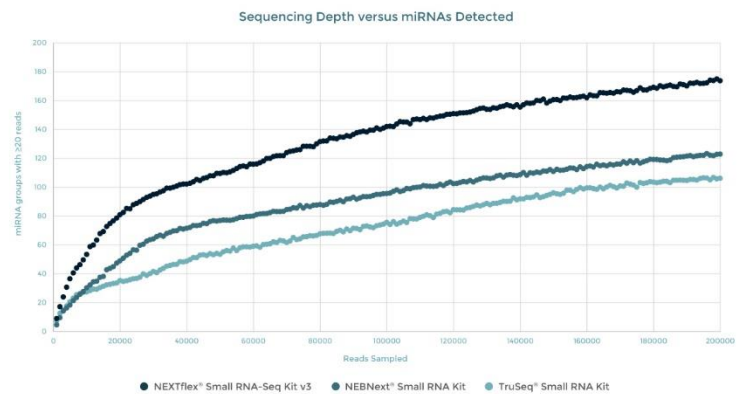
Yeri et al. Evaluation of commercially available small RNAseq library preparation kits using low input RNA. BMC Genomics. 2018. 19:331

“For the detection of human (unmodified) miRNAs the standard NEXTflex protocol performed best...”

Dard-Dascot et al. Systematic comparison of small RNA library preparation protocols for next-generation sequencing. BMC Genomics. 2018. 19:118



miRNAs detected at various thresholds - Sequencing results from small RNA libraries created in triplicate from 1 ng of Miltenyi® miRXPlore® Universal Reference, an equimolar mixture of 963 miRNAs. The number of miRNAs detected at various thresholds is shown. The inset shows the Coefficient of Variation of the 963 miRNAs in each sample.



Sequencing Depth vs. miRNAs Detected - Small RNA libraries were created in triplicate from the indicated amount of human brain total RNA. The indicated number of reads was sampled from each library and the average number of miRNA groups with ≥20 reads determined. The inset shows the number of reads required to detect 100 miRNA groups at a threshold of ≥20 reads.

ORDERING INFORMATION

Catalog #	Kit Name	Quantity
NOVA-5132-05	NEXTFLEX® Small RNA-Seq Kit v3 (8 barcodes)	8 rxns
NOVA-5132-06	NEXTFLEX® Small RNA-Seq Kit v3 (48 barcodes)	48 rxns

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