Now includes
NEBNext UltraExpress® DNA and FS DNA Library Prep

NEBNext® for DNA Library Prep

FOR THE ILLUMINA® PLATFORM

NEW ENGLAND Biolabs®

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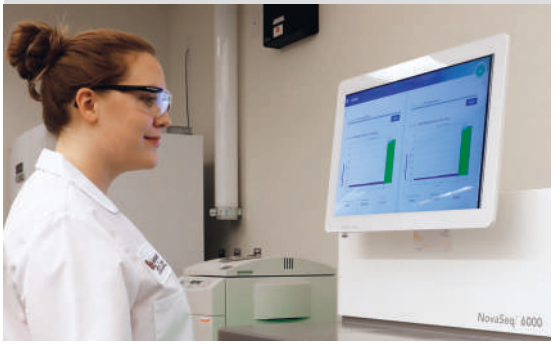
TOOLS & RESOURCES

Visit NEBNext.com to find:

- The full list of products available
- Video protocols
- Online tutorials to help with product selection, general handling tips and more
- Access to **NEBNext Selector Tool**, our online tool for help with selecting the right NEBNext product
- Over 30,000 NEBNext citations
- Product Manuals & FAQs
- **NEBNext Automation Platform Compatibility Chart**, for details on which liquid handling automation platforms have been used to automate NEBNext kits



Streamlined for speed.

NEBNext UltraExpress

Why Choose NEBNext for DNA?

There is no shortage of options available for nearly any step of any NGS workflow. We know you have choices, so below are just a few of the reasons to choose NEBNext.

High Performance and User Friendly

The NEBNext suite of products supports DNA sequencing on the Illumina® platform with sample preparation tools that streamline workflows and minimize inputs, while improving library yields and quality.

NEBNext UltraExpress is the latest generation of library prep kits, further streamlining workflows and reducing consumables waste, while maintaining performance for the most commonly used samples. Many NEBNext DNA library prep kits are driven by our Ultra II technology and are compatible with high- and low-quality samples, PCR-free and standard workflows, as well as a broad range of input amounts:

- 100 pg–1 µg (Ultra II DNA & Ultra II FS DNA)
- 250 ng–1 µg (Ultra II DNA PCR-free)
- 50 ng–500 ng (Ultra II FS DNA PCR-free)

Our growing selection of indices (barcodes) provides a wide selection of options for library multiplexing. Beyond library construction, NEBNext also enables 5mC and 5hmC analysis, target enrichment, repair of FFPE DNA, enrichment of microbiome DNA, and qPCR-based library quantitation.

Reliable and Time Tested

Since our first product release in 2009, the NEBNext brand has stood for quality you can count on. In addition to the extensive QCs performed on individual kit components, all NEBNext kits for Illumina are functionally validated by preparation of a library, followed by Illumina sequencing. Additionally, NEBNext products have been cited in over 30,000 publications.

Flexible Formats

NEBNext library prep reagents are available in multiple kit and workflow formats, for maximum convenience and flexibility.

Kits and modules

Kits are the most convenient option, as they include reagents for the entire library prep workflow. Many kits are available with SPRISelect™ beads for clean-up and size-selection steps.

With flexibility as a priority, NEBNext modules contain reagents for the individual steps in library preparation. These modules can be combined to cover the entire library prep workflow, or a subset of NEBNext modules can be combined with other reagents to enable a customized workflow for your specific needs.

An expanded range of adaptors and primers is also available, allowing for increased flexibility in multiplexing options.

Large volume & custom formats:

When your reagent needs exceed standard volumes, or you require a specialized formulation or kit, consider NEBNext's Customized Solutions options. As reagent manufacturers, we are able to provide customized components, kits and modules to meet your specific needs. We encourage consultation with the Customized Solutions team at NEB.

Please complete the NEB Custom Contact Form at **www.neb.com/CustomContactForm** to learn more.

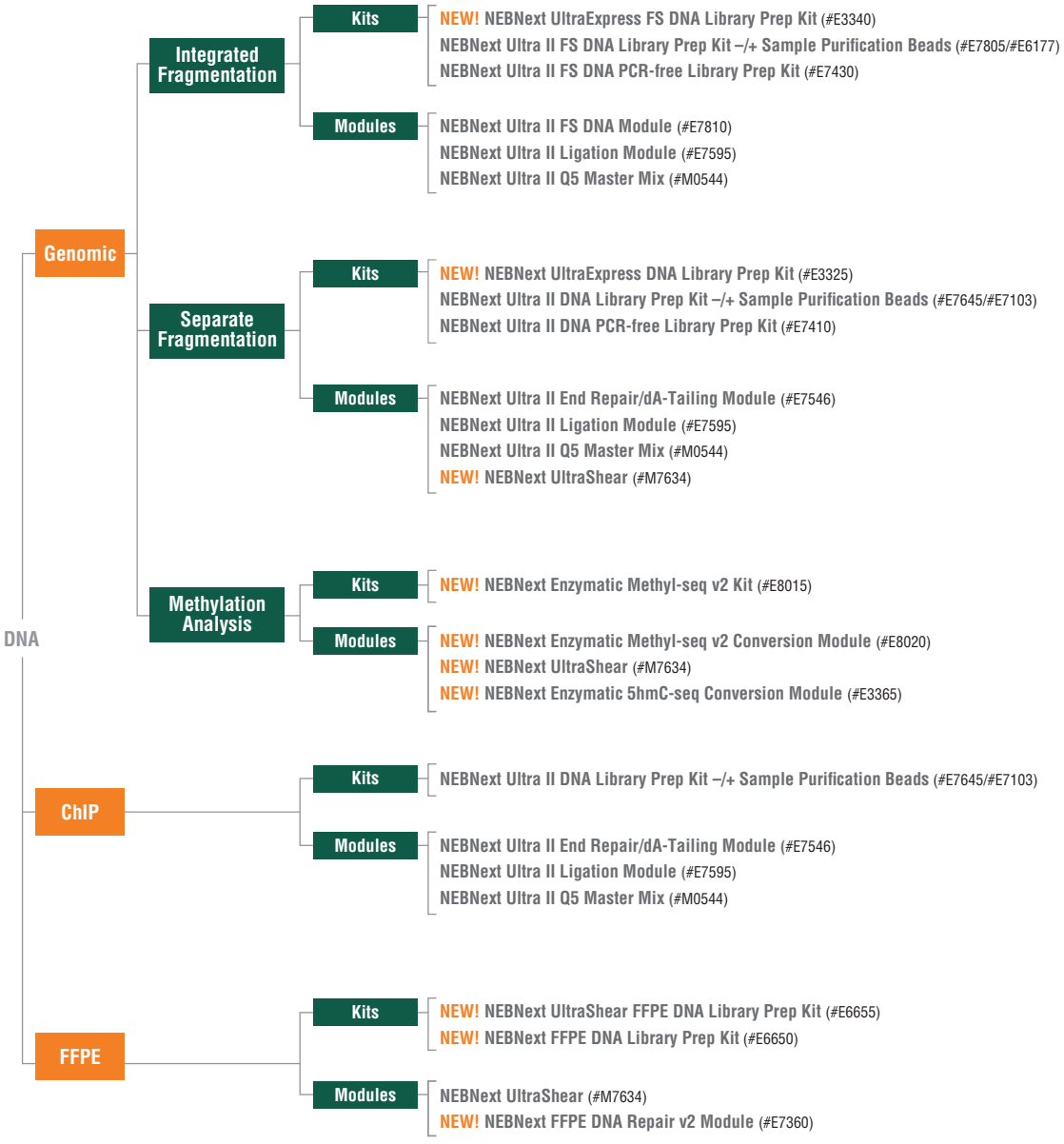
WHAT'S NEW IN NEBNEXT FOR DNA?

- **NEW! NEBNext UltraExpress DNA and FS DNA** workflows for streamlined simplicity
- Optimize library prep from FFPE-treated DNA with **NEBNext UltraShear FFPE DNA Library Prep Kit**
- Analyze the methylome with **NEBNext Enzymatic Methyl-seq v2 (EM-seq)** and **NEBNext Enzymatic 5hmC-seq (E5hmC-seq)**
- Adapt to lower volumes with 5 sets of **LV Unique Dual Indices** and **NEBNext LV Unique Dual Index Primers**

Visit **NEBNextSelector.neb.com** to try the **NEBNext Selector Tool**, our online tool for help with selecting the NEBNext product that fits your needs

Illumina® DNA Product Selection Chart

Use the following chart to determine the best NEBNext products for your Illumina DNA prep needs.
For the most up-to-date product and pricing information, visit NEBNext.com.



Multiplex Oligos

Single Index	Dual Index	Unique Dual Index	LV Unique Dual Index	Unique Dual Index UMIs
Set 1 (#E7335), Set 2 (#E7500), Set 3 (#E7710), Set 4 (#E7730), 96 Index Primers (#E6609)	Set 1(#E7600), Set 2 (#E7780)	Set 1 (#E6440), Set 2 (#E6442), Set 3 (#E6444), Set 4 (#E6446), Set 5 (#E6448), Primer Pairs for EM-seq (#E7140)	Set 1(#E3400), Set 2 (#E3402), Set 3 (#E3404), Set 4 (#E3406) Set 5 (#E3408), Set 2A (#E3390), Set 2B (#E3392)	DNA Set 1(#E7395), DNA Set 2 (#E7874), DNA Set 3 (#E7876), DNA Set 4 (#E7878)

NEBNext Selector

For help selecting products, try our online product selection tool at **NEBNextSelector.neb.com**

Tips for working with DNA

DNA Sample Input Guidelines

Integrity of DNA

- The quality of the input material directly affects the quality of the library. Absorbance measurements can be used as an indication of DNA purity. Ideally, the ratio of the absorbance at 260 to 280 nm should be between 1.8–2.0. However, measurements can be affected by the presence of RNA or small nucleic acid fragments. A DNA Integrity Number (DIN) can be determined using the Agilent® TapeStation® or similar instrumentation, and qPCR-based methods can also provide a measurement of DNA integrity.

Quantitation of Input DNA

- It is important to quantify accurately the DNA sample prior to library construction. Fluorescence-based detection which utilizes dsDNA-specific dyes, such as the Qubit® from Thermo Fisher Scientific®, is more accurate than UV spectrometer based measurements, as the presence of RNA or other contaminants can result in overestimation of the amount of the DNA sample by the latter. This can result in use of non-optimal adaptor dilutions and numbers of PCR cycles, compromising library prep efficiency.

Bead-based cleanups and size selection

- Be sure to vortex the beads well just before use. They should form a uniform suspension. When beads have not been used for several weeks, plan for extra time for bead vortexing and agitation.
- Do not over-dry the beads. Beads should still be dark brown and glossy when eluting. Over-drying can make resuspension difficult and reduce yield.
- Take care not to remove beads after separation.
- Remove all of the supernatant after the bind step. Incomplete supernatant removal can cause leftover adaptor dimer or PCR primers to remain in the libraries.
- When adding beads to the sample, aspirate slowly, remove any droplets from the outside of the tip and make sure to dispense the full volume into the sample.

Indices/Barcodes

- When using a subset of the indices supplied in a kit, or using indices from more than one kit, it is important to optimize the combination of indices used, in order to ensure balanced sequencing reads. We provide recommendations for NEBNext index combinations in the manuals for NEBNext Oligos products.
- For index primers provided in vials, open only one vial at a time, to minimize the risk of contamination.
- Be sure to change pipette tips for each index primer.
- For 96-well plate formats, NEBNext index primers are provided in single-use plates with pierceable foil lids. To avoid risk of contamination, do not pipette from a well more than once.
- For help choosing the right indices/barcodes, visit **www.neb.com/oligos.**

NEBNext Magnetic Separation Rack

NGS library prep workflows include magnetic bead-based purification and size-selection steps, and it is important for library yield and quality that bead separation be highly efficient and fast.

The NEBNext Magnetic Separation Rack was designed for this application and contains rare earth Neodymium Iron Boron (NdFeB) magnets, the most powerful commercially available magnets, in an anodized aluminium rack. The rack holds 24 0.2-ml tubes, and is compatible with single tubes or strip tubes.

ADVANTAGES

- Fast separations in purification and size-selection steps in next generation sequencing workflows
- 24-tube capacity

NEBNEXT MAGNETIC SEPARATION RACK

Choose the right kit for your needs
NEBNext UltraExpress DNA/FS DNA or NEBNext Ultra II DNA/FS DNA

Choosing the right NEBNext kit for your DNA library prep doesn't need to be complicated!
By answering just a few questions, you'll be able to confidently select the right kit for your needs.

1 What's your sample type/quality?

Whether you're working with an established model organism with a well-annotated reference genome or you're breaking new ground with a non-model organism, either NEBNext DNA library prep kit is an excellent place to start. Both have been rigorously analyzed with a range of samples from different organisms, but we encourage the end-user to confirm whether a kit works adequately well on any sample types that aren't explicitly identified in the supporting data.

DNA sample quality, typically judged by sample integrity, can range from 1 to 10 DIN. Low quality samples can make it difficult to produce high quality libraries. Libraries prepped from samples that were stored for a long time, inadequately preserved, or exposed to challenging environments may exhibit hallmark signs of damage. For example, DNA exposed to FFPE treatment may have several characteristic forms of DNA damage (e.g., C-T deamination, nicks, gaps, and oxidation) and for which, NEB recommends the NEBNext UltraShear FFPE DNA Library Prep Kit. Also, while libraries exposed to the caustic chemicals and high temperatures of sodium bisulfite treatment routinely exhibit fragmentation, sample loss, and bias (for which, NEB recommends NEBNext Enzymatic Methyl-seq and/or NEBNext Enzymatic 5hmC-seq). For all other samples of sub-standard DNA quality, consider choosing NEBNext Ultra II DNA or FS DNA library prep.

2 How much sample can you spare?

The answer to this question is critical to your choice between NEBNext UltraExpress and NEBNext Ultra II. If your samples are within a 10–200 ng input range, we'd strongly recommend choosing NEBNext UltraExpress due to its speed and more streamlined workflow. Spending less time on library prep means you can spend more time on answering your underlying questions. If, instead, your samples are available only in low amounts, or of low or unknown quality (see above), NEBNext Ultra II offers a workflow that works with inputs as low as 100 pg, stretching your samples further.

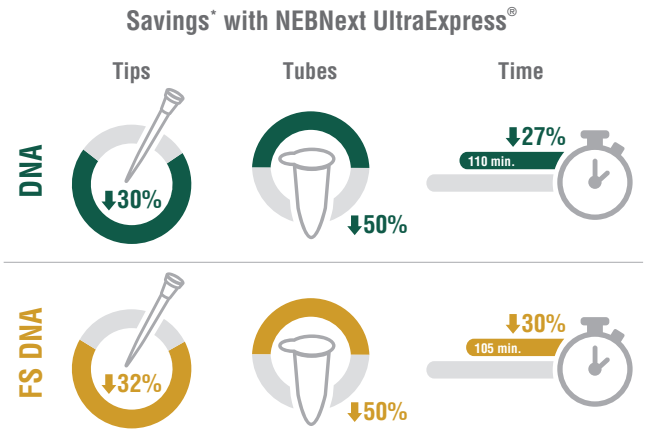
3 How important is speed?

NEBNext UltraExpress was intentionally developed to further streamline the library prep workflow, beyond the already-streamlined workflow of NEBNext Ultra II. NEBNext UltraExpress DNA and FS DNA library prep workflows make quick work of DNA library prep from either fragmented or intact DNA, taking you from input DNA to library in under two hours.

The NEBNext Ultra II DNA/FS DNA workflows are longer than that of NEBNext UltraExpress, but it is the recommended option for samples of low quality or that are available only in low amounts.

4 Do you prefer mechanical or enzymatic shearing?

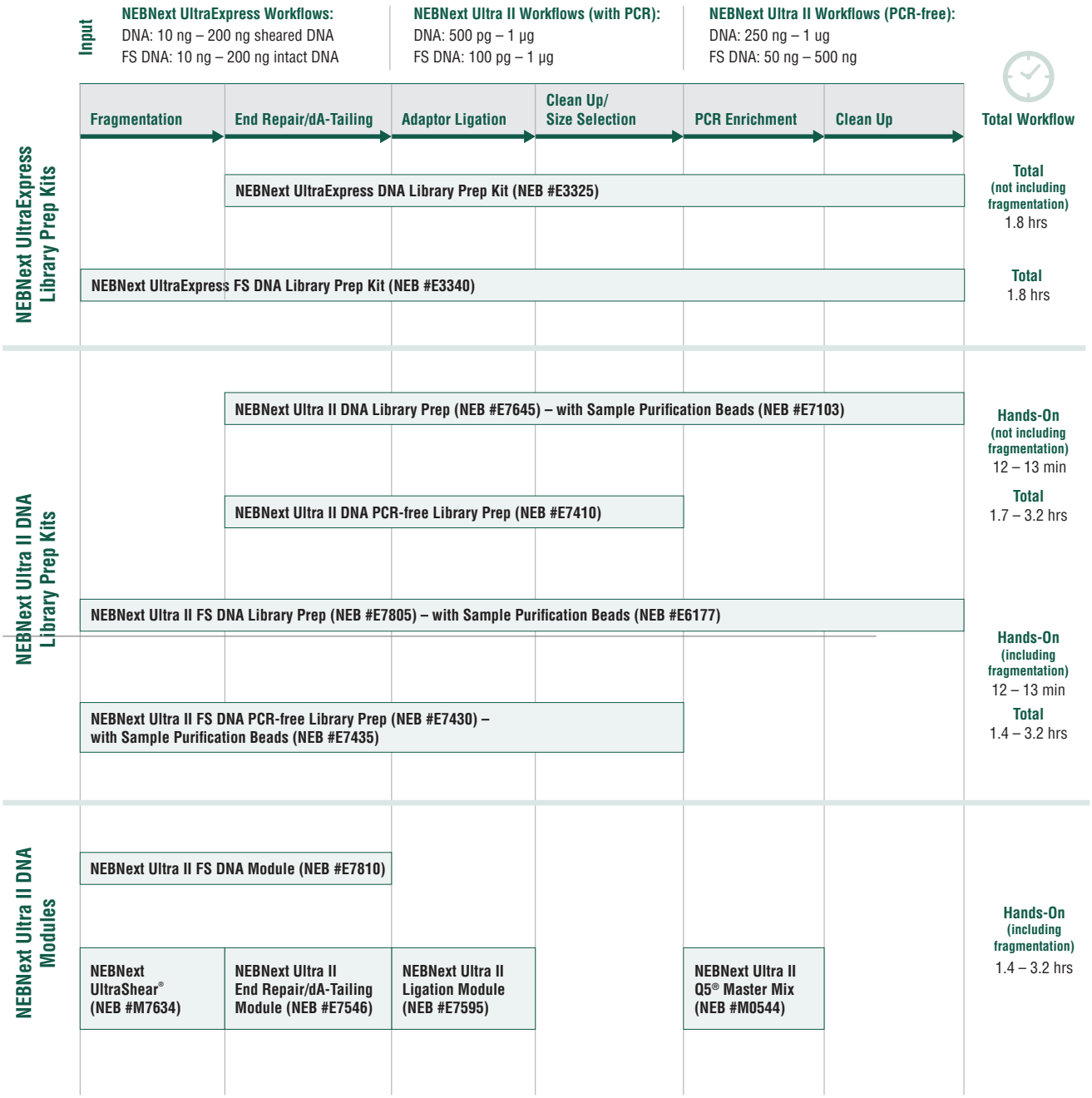
Your choice of NEBNext UltraExpress and NEBNext Ultra II reagents and workflows for library prep should now be clearer, but there are still more choices you can make. Will you be mechanically fragmenting your sample via sonication or shearing, or are you looking for an easy to use, robust enzymatic method that is scalable? NEBNext offers kits for both scenarios.



* As compared to NEBNext® Ultra™ II

NEBNext DNA Library Prep Reagents for Illumina Sequencing

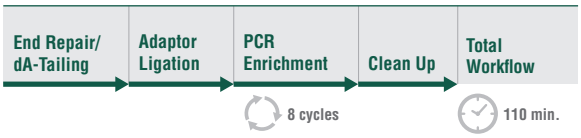
NEBNext UltraExpress and NEBNext Ultra II DNA library prep kits are available with or without integrated enzymatic DNA fragmentation. NEBNext Multiplex Oligos (Adaptors & Primers) are available in a range of options; learn more at neb.com/oligos. In addition to stringent QCs on individual components, the NEBNext DNA kits are also functionally validated by preparation of a library, followed by Illumina sequencing.



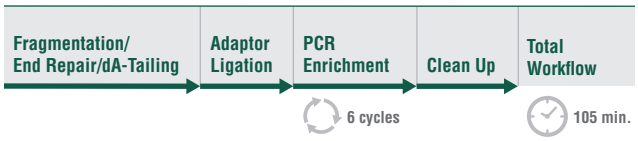
NEBNext UltraExpress DNA and FS DNA Library Prep Kits

The NEBNext UltraExpress DNA and FS DNA Library Prep Kits are the latest generation of NEBNext DNA library prep, with fast, streamlined workflows to generate high yields of high-quality libraries. The workflows allow processing of samples with a wide range of input amounts of pre-sheared DNA using a single protocol, without adjustment of reaction conditions.

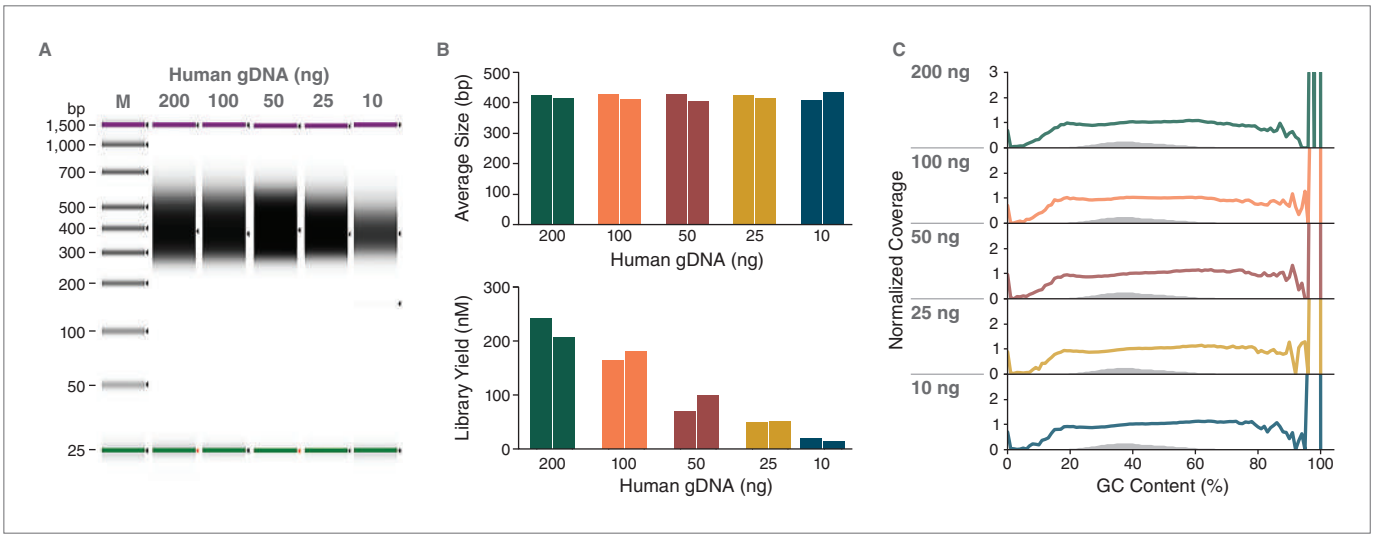
NEBNext UltraExpress DNA Workflow
Input: 10 ng – 200 ng pre-sheared DNA



NEBNext UltraExpress FS DNA Workflow
Input: 10 ng – 200 ng intact DNA

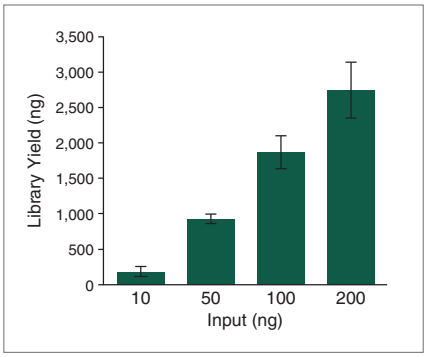


NEBNext UltraExpress DNA generates high yields of high quality libraries, across a broad input range.

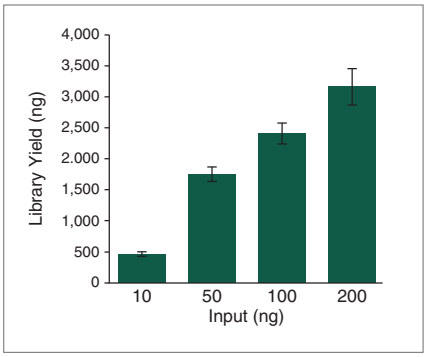


A-B. Libraries were made using 10–200 ng Covaris®-sheared Human NA19240 genomic DNA and the NEBNext UltraExpress DNA Library Prep Kit, with the same amount of adaptor and the same PCR conditions (8 cycles) for each. Libraries were pooled and sequenced on the Illumina MiSeq®.
C. 140,000 paired end reads were sampled (seqtk v1.3), adapter-trimmed (seqprep v0.1) and aligned to GRCh38 reference genome (bowtie2 v2.4.5). Libraries had high yields, uniform library profiles and even GC coverage.

The NEBNext UltraExpress DNA and FS DNA Library Prep Kits provide robust library yields over a wide input range



Libraries were prepared from 10, 50, 100 or 200 ng of Human NA19240 genomic DNA (Coriell Institute for Medical Research) using the same adaptor amount and 8 PCR cycles. Yields exceeded the minimum requirement (40 ng) for a single Illumina NovaSeq® 6000 run to achieve whole genome sequencing with at least 30X coverage.



Libraries were prepared in triplicate from 10, 50, 100 and 200 ng of a 9:1 Human NA19240 genomic DNA (Coriell Institute for Medical Research) and Escherichia coli gDNA (Lofstrand Labs Limited) mixed sample, using the NEBNext UltraExpress FS DNA single-protocol workflow (e.g., same adaptor amount and 6 PCR cycles for all input amounts). Yields exceeded the minimum requirement (40 ng) for a single Illumina NovaSeq 6000 run to achieve whole genome sequencing with at least 30X coverage.

ADVANTAGES

- Fast workflow (< 2 hours)
- Fewer steps and consumables
- Fewer cleanups
- Wide input range (10–200 ng pre-sheared DNA; FS DNA: 10–200 ng intact DNA)
- Single protocol for all inputs
- Automation friendly

PRODUCT	SIZE
NEBNext UltraExpress DNA Library Prep Kit (NEB #E3325S/L)	24/96 rxns
NEBNext UltraExpress FS DNA Library Prep Kit (NEB#E3340S/L)	24/96 rxns

NEBNext Ultra II DNA, FS and PCR-free DNA Library Prep Kits for Illumina

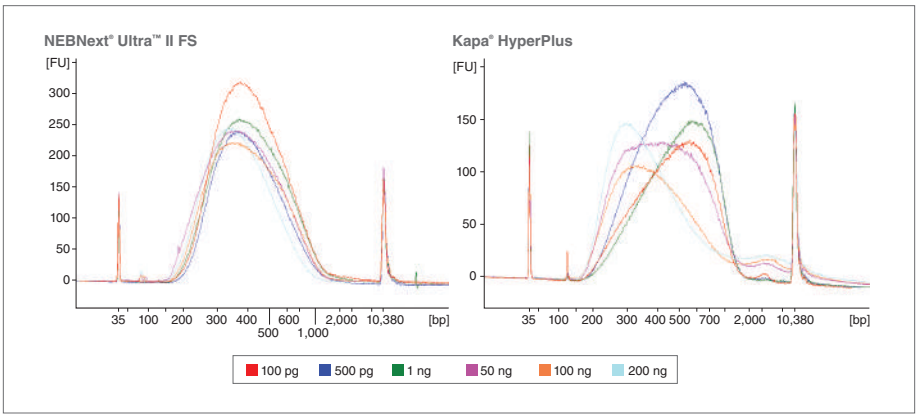
NEBNext Ultra II DNA Library Prep Kits for Illumina meet the challenge of constructing high quality libraries from ever-decreasing input quantities, enabling high yield preparation of high quality libraries from 100 picograms to 1 microgram of input DNA. Ultra II kits use a fast, streamlined, automatable workflow and enable use of fewer PCR cycles while also improving GC coverage. The kit is also effective with challenging samples such as FFPE DNA.

The Ultra II FS DNA Library Prep Kit combines robust enzymatic DNA fragmentation with end repair and dA-tailing, integrated into a streamlined library prep workflow.

PCR-free kits are now available for both the Ultra II DNA and Ultra II FS DNA workflows.

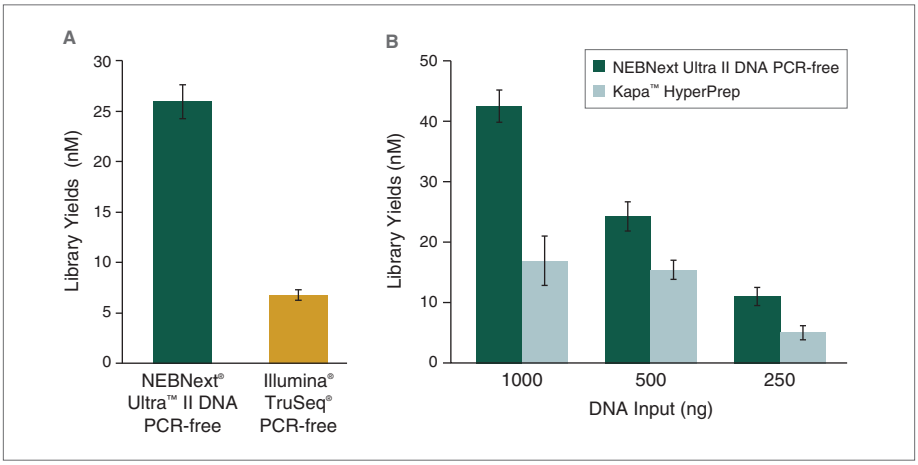
Ultra II kits are available with or without SPRIselect™ beads.

NEBNext Ultra II FS DNA provides consistent fragmentation regardless of input amount



Libraries were prepared from Human NA19240 genomic DNA using the input amounts shown. NEBNext Ultra II FS libraries (left) were prepared using a 20-minute fragmentation time. For Kapa® HyperPlus (right), input DNA was cleaned up with 3X beads prior to library construction, as recommended, and a 20-minute fragmentation time. Library size was assessed using the Agilent Bioanalyzer®. Low input (1 ng and below) libraries were loaded on the Bioanalyzer without a dilution. High input libraries were loaded with a 1:5 dilution in 0.1X TE.

NEBNext Ultra II DNA PCR-free Library Prep Kit generates libraries with higher yields



A. PCR-free libraries were prepared with NA19240 genomic DNA (Coriell Institute) using NEBNext Ultra II DNA PCR-free and Illumina TruSeq® PCR-free library prep kits and size selected for 350 bp inserts. DNA inputs were 1 µg.

B. Libraries of 150-200 bp inserts were prepared using NEBNext Ultra II DNA PCR-free and Roche Sequencing Kapa HyperPrep library prep kits coupled with Covaris shearing without size selection. NEBNext Unique Dual Index UMI Adaptors DNA Set 1, IDT for Illumina (TruSeq DNA UD Indexes) and Kapa Dual-Indexed Adaptors were used for the NEBNext, Illumina, and Kapa kits, respectively, following manufacturers' recommendations.

ADVANTAGES

- Get more of what you need, with the highest library yields
- Generate high quality libraries even with limited amounts of DNA, as low as 500 pg
- Prepare libraries from ALL of your samples, including GC-rich targets
- Save time with streamlined workflows, reduced hands-on time, and automation compatibility
- Access reliable and easy-to-use, scalable enzymatic DNA fragmentation, integrated into the workflow with the FS kit
- Enjoy the flexibility and reliability of the gold standard SPRIselect size selection and clean-up beads, supplied in just the amounts you need

PRODUCT	SIZE
NEBNext Ultra II DNA Library Prep Kit for Illumina (NEB #E7645S/L)	24/96 rxns
NEBNext Ultra II DNA Library Prep with Sample Purification Beads (NEB #E7103S/L)	24/96 rxns
NEBNext Ultra II DNA PCR-free Library Prep Kit for Illumina (NEB #E7410S/L)	24/96 rxns
NEBNext Ultra II FS DNA Library Prep Kit for Illumina (NEB #E7805S/L)	24/96 rxns
NEBNext Ultra II FS DNA Library Prep with Sample Purification Beads (NEB #E6177S/L)	24/96 rxns
NEBNext Ultra II FS DNA PCR-free Library Prep Kit for Illumina (NEB #E7430S/L)	24/96 rxns
NEBNext Ultra II FS DNA PCR-free Library Prep with Sample Purification Beads (NEB #E7435L)	96 rxns

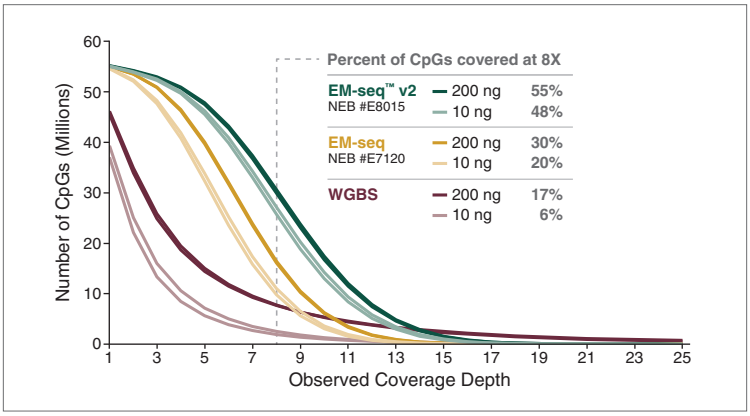
NEBNext Enzymatic Methyl-seq v2 (EM-seq)

NEBNext Enzymatic Methyl-seq (EM-seq) is a high-performance enzyme-based alternative to bisulfite sequencing for the identification of 5mC and 5hmC on the Illumina® platform. Unlike harsh bisulfite treatment, EM-seq minimizes DNA damage, resulting in superior detection of 5mC and 5hmC from fewer sequencing reads.

The new NEBNext Enzymatic Methyl-seq v2 Kit has a wider input range (as low as 100 pg), a faster, more streamlined workflow and improved performance compared to the original EM-seq kit (NEB #E7120).

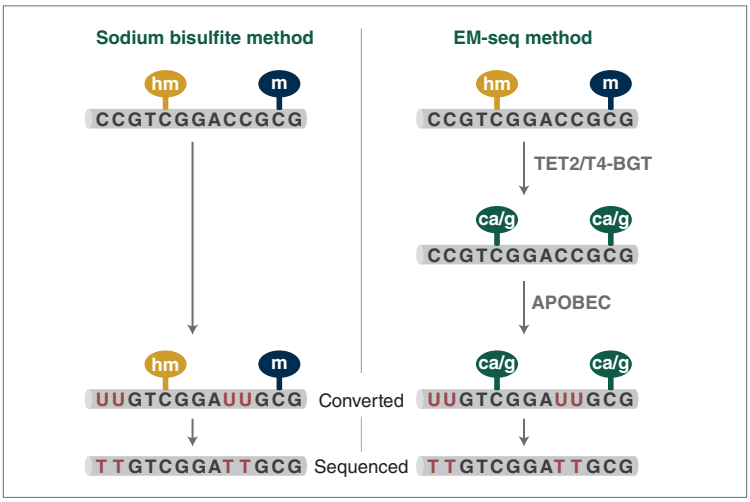
The NEBNext Enzymatic Methyl-seq v2 Kit includes conversion reagents, library prep reagents and the EM-seq Adaptor. Multiple sets of the required index primers (NEBNext LV Unique Dual Index Primers) are available separately, enabling greater flexibility in multiplexing. For enzymatic fragmentation of DNA designed for use with EM-seq see NEBNext UltraShear (page 13).

NEBNext EM-seq v2 identifies more CpGs than WGBS and the original EM-seq, at lower sequencing coverage depth



EM-seq v2 (NEB #E8015), EM-seq (NEB #E7120) and WGBS libraries were prepared from 200 ng and 10 ng of NA12878 DNA (sheared to ~350 bp), spiked with unmethylated lambda and CpG-methylated pUC19. Libraries were sequenced on an Illumina NovaSeq 6000. For accurate comparison of the original EM-seq and WGBS data with EM-seq v2 data, we evaluated data from approximately 625 million 100 base reads for each library aligned to a composite human T2T, lambda and pUC19 reference genome using bwa-meth.
The T2T genome covers a maximum of 67.8 million CpGs when the top and bottom strands are counted independently. EM-seq v2 and EM-seq covered over 54 million CpG sites for both 200 ng and 10 ng inputs; however, WGBS libraries covered only 46 million and 39 million for 200 ng and 10 ng inputs respectively at 1X coverage. The dashed lines represent coverage of 8X. The table lists the percentage of CpG sites covered by different libraries at 8X coverage level.

EM-seq conversion method



The harsh sodium bisulfite treatment deaminates unmodified cytosines to uracil. In the EM-seq workflow, 5mC and 5hmC are first protected using the enzymes TET2 and T4-BGT. Unmodified cytosines are then deaminated by the APOBEC enzyme to uracil, while the protected 5mC and 5hmC are not converted. During Illumina® sequencing, 5mCs and 5hmCs are represented as cytosine, while unmodified cytosines are represented as thymine.

ADVANTAGES

- Superior sensitivity of 5mC and 5hmC detection
- 100 pg – 200 ng input range
- Detection of more CpGs with fewer sequencing reads
- Even GC coverage
- High performance library preparation and larger library insert sizes
- Index primers supplied separately

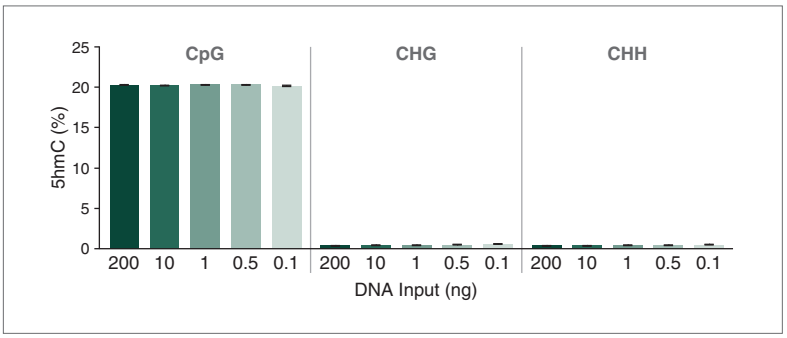
PRODUCT	SIZE
NEBNext Enzymatic Methyl-seq v2 Kit (NEB #E8015S/L)	24/96 rxns
NEBNext Enzymatic Methyl-seq v2 Conversion Module (NEB #E8020S/L)	24/96 rxns
NEBNext Q5U® Master Mix (NEB #M0597S/L)	50/250 rxns
NEBNext LV Unique Dual Index Primers Sets 2A, 2B (NEB #E3390, E3392)	24 rxns
NEBNext LV Unique Dual Index Primers Sets 1, 2, 3, 4, 5 (NEB #E3400, E3402, E3404, E3406, E3408)	96 rxns

NEBNext Enzymatic Methyl-seq is an enzymatic alternative to bisulfite conversion with superior performance. For more information, including extensive performance data, visit **NEBNext.com.**

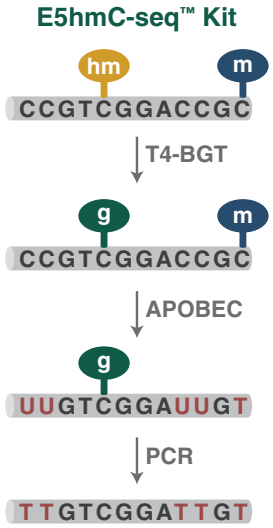
NEBNext Enzymatic 5hmC-seq (E5hmC-seq)

While NEBNext Enzymatic Methyl-seq (EM-seq) detects both 5mC and 5hmC, it does not distinguish between them. Specific detection of 5hmC sites is now enabled by the NEBNext Enzymatic 5hmC-seq Kit (E5hmC-seq). The kit includes NEBNext Ultra II library prep reagents, and 5hmC is detected using a two-step enzymatic conversion workflow, that minimizes damage to DNA and allows discrimination of 5hmC from both cytosine and 5mC after Illumina sequencing. E5hmC-seq data can also be subtracted from EM-seq data, allowing determination of the precise location of individual 5mC and 5hmC sites.

5hmC detected by E5hmC-seq in human brain gDNA is consistent across inputs

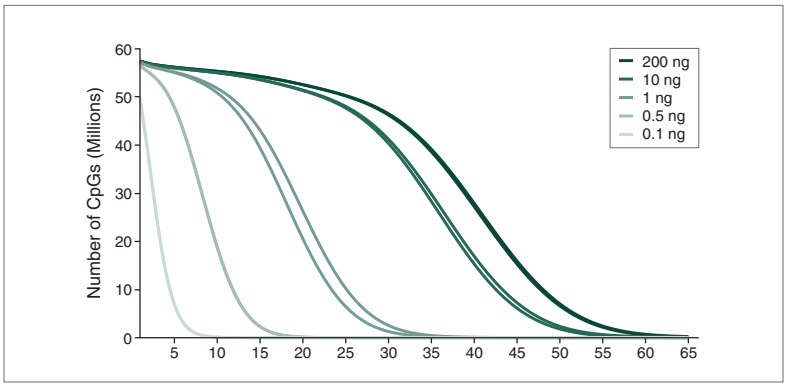


200–0.1 ng of human brain genomic DNA was sheared to 350 bp (Covaris ME220) and E5hmC-seq libraries were prepared and sequenced on an Illumina NovaSeq 6000 (2 x 150 bases). Approximately 1.9 billion (200 ng, 10 ng and 1 ng) or 715 million (0.5 ng and 0.1 ng) reads for each library were aligned to a composite human T2T, lambda and T4 reference genome using bwa-meth, and methylation information was extracted from the alignments using MethylDackel. Values shown are the average of two technical replicates and error bars show standard deviation. Detected 5hmC levels are similar between all inputs in the CpG, CHH and CHG contexts.



E5hmC-seq conversion method. *To enable specific 5hmC detection, 5hmC is first glucosylated using T4-BGT. 5mC and unmodified cytosine are then deaminated by APOBEC to thymine and uracil, respectively, while the protected 5hmC is unconverted. During Illumina sequencing 5hmCs are represented as cytosine, while cytosine and 5mCs are represented as thymine.*

E5hmC-seq exhibits high CpG coverage across a range of inputs



200–0.1 ng of human brain genomic DNA was sheared to 350 bp (Covaris ME220) and E5hmC-seq libraries were prepared and sequenced on an Illumina NovaSeq 6000 (2 x 150 bases). Approximately 1.9 billion CpG specific file a cumulative coverage plot was generated for CpG sites covered using E5hmC-seq libraries across all inputs. The T2T genome covers a maximum of 67.8 million CpGs when the top and bottom strands are counted independently. E5hmC-seq covered over 56 million CpG sites for 0.5 ng to 200 ng inputs and roughly 48 million CpG sites for 0.1 ng input libraries.

ADVANTAGES

- Enzyme-based workflow enables high yields and high-quality data
- 100 pg – 200 ng inputs
- Minimal GC bias
- E5hmC-seq and EM-seq data can be combined
- Conversion module also available separately

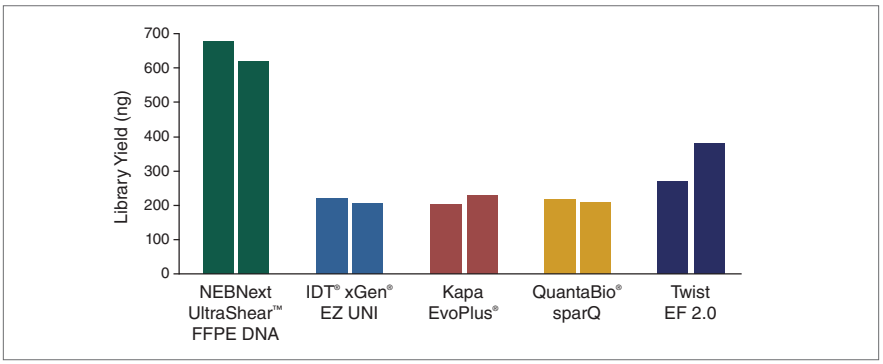
PRODUCT	SIZE
NEBNext Enzymatic 5hmC-seq Kit (NEB #E3350/L)	24/96 rxns
NEBNext Enzymatic 5hmC-seq Conversion Module (NEB #E3365/L)	24/96 rxns
NEBNext Oligos for Enzymatic 5hmC-seq (NEB #E3360S/L)	24/96 rxns

NEBNext UltraShear FFPE DNA Library Prep Kit & NEBNext FFPE DNA Library Prep

FFPE DNA poses many challenges for library preparation, including characteristically low input amounts and highly variable damage from fixation, storage, and extraction methods. Regions of interest are often enriched using hybrid capture-based approaches – these workflows require a high input of diverse, uniform DNA library.

The NEBNext FFPE DNA Library Prep Kit includes the NEBNext FFPE DNA Repair v2 Mix, an optimized cocktail of enzymes designed to repair FFPE DNA, library prep reagents featuring a new polymerase master mix, and a protocol optimized for FFPE DNA. The NEBNext UltraShear FFPE DNA Library Prep Kit also includes NEBNext UltraShear, a new solution designed for enzymatic fragmentation of challenging samples (e.g., FFPE DNA). This enzymatic shearing solution further increases library yields and quality, while improving scalability and ease of use.

NEBNext UltraShear FFPE DNA Library Prep Kit enables higher library yields



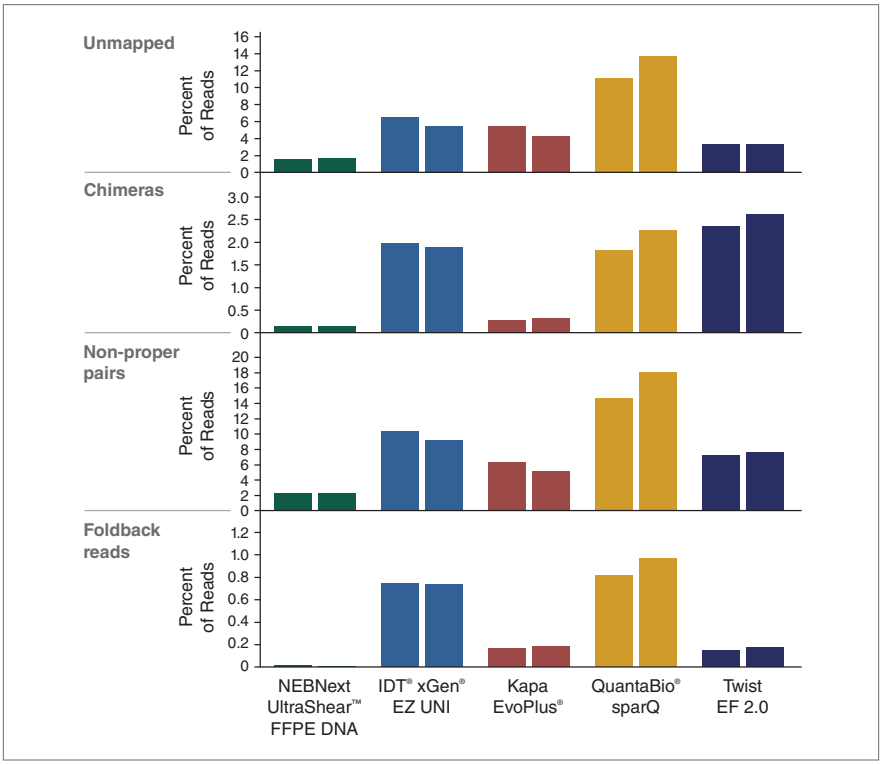
Libraries were prepared in duplicate from 100 ng of low quality, normal tissue FFPE DNA (DIN 1.8) and 9 PCR cycles, using the NEBNext UltraShear FFPE DNA Library Prep Kit. Results were compared to other enzymatic fragmentation-based library prep kits that have been validated for use with FFPE samples, using each vendor's own recommended adaptors (IDT® xGen® EZ UNI, Kapa EvoPlus® Library Prep Kit, QuantaBio® sparQ DNA Library Prep Kit, and Twist Library Preparation EF 2.0 kit). Library yields (total ng) were quantified using the Qubit High-Sensitivity dsDNA assay (Thermo Fisher Scientific®). The NEBNext UltraShear FFPE DNA Library Prep Kit enables higher library yields, sufficient for target enrichment library input.

ADVANTAGES

- Includes FFPE DNA repair reagents plus optimized library prep reagents and protocol
- Optional NEBNext UltraShear enzymatic fragmentation
- Increased library yields
- Improved sequencing metrics
- Greater sensitivity of somatic variant calling
- Automation-friendly workflows
- Input: 5–250 ng

PRODUCT	SIZE
NEBNext FFPE DNA Library Prep Kit (NEB #E6650S/L)	24/96 rxns
NEBNext UltraShear FFPE DNA Library Prep Kit (NEB #E6655S/L)	24/96 rxns

The NEBNext UltraShear FFPE DNA Library Prep Kit improves library quality



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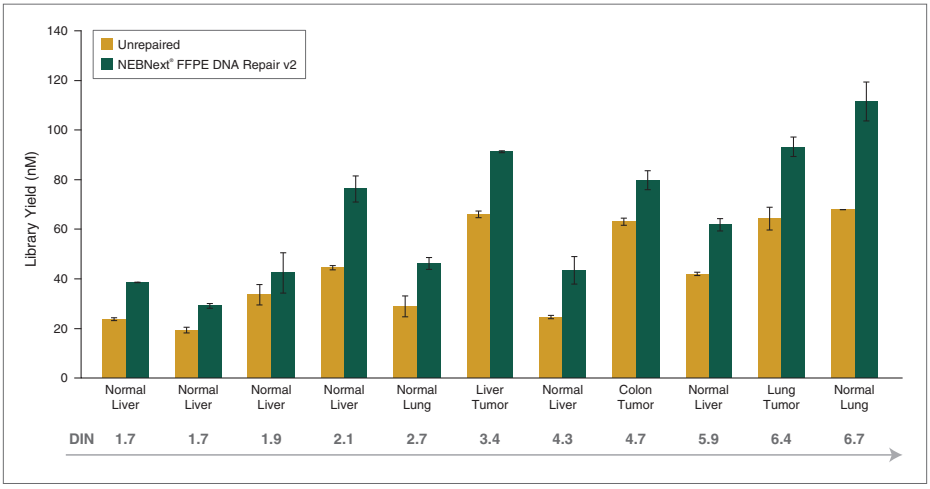
NEBNext FFPE DNA Repair v2 Module

The methods used for fixation and storage of Formalin-Fixed, Paraffin-Embedded (FFPE) DNA samples cause significant damage, making it challenging to obtain high quality sequence data. The NEBNext FFPE DNA Repair v2 Module is an optimized cocktail of enzymes designed to repair FFPE DNA, and supplied with optimized reagents to enable a streamlined workflow for NGS library preparation.

The NEBNext FFPE DNA Repair v2 Module improves upon the performance of the original NEBNext FFPE DNA Repair Mix, and offers higher efficiency, a more streamlined workflow, a more convenient reaction buffer and no cleanup is required between repair and library prep.

The NEBNext FFPE DNA Repair v2 Module enables robust library preparation from a broad range of FFPE DNA sample qualities



Libraries were prepared with 25 ng of Covaris acoustic-sheared FFPE DNA samples of different qualities and tissue sources. The NEBNext FFPE DNA Repair v2 Module was used, followed by NEBNext Ultra II DNA library preparation (NEB #E7645) with 9 PCR cycles. Libraries were quantified using the Agilent HS D1000 TapeStation. The NEBNext FFPE DNA Repair v2 Module improves the yield of FFPE libraries by varying degrees depending on the quality and damage types present in the input DNA. Error bars indicate the standard deviation of two replicates for each library sample.

FFPE Damage Type	Repaired by the FFPE DNA Repair v2 Module?
Deamination of cytosine to uracil	Yes
Nicks and gaps	Yes
Oxidized bases	Yes
Blocked 3´ ends	Yes
DNA fragmentation	No
DNA-protein crosslinks	No

ADVANTAGES

- Higher repair efficiency with FFPE DNA
- A more convenient reaction buffer containing all the required buffer components for both efficient FFPE DNA repair and downstream end repair and dA-tailing
- No cleanup is required between repair and library prep, through the use of Thermolabile Proteinase K

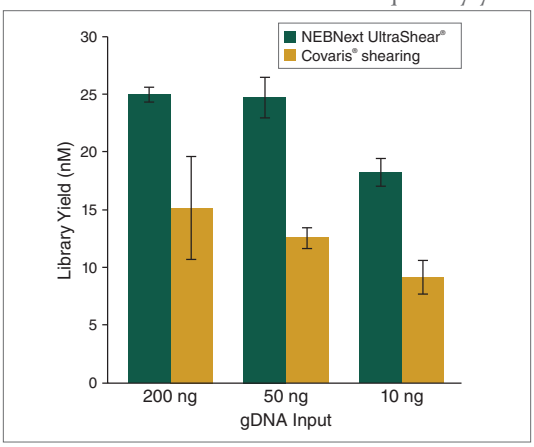
PRODUCT	SIZE
NEBNext FFPE DNA Repair v2 Module (NEB #E7360S/L)	24/96 rxns

NEBNext UltraShear

Enzymatic fragmentation of DNA as part of the library prep workflow provides many advantages compared to mechanical shearing. However, specialized fragmentation reagents are required for enzymatic shearing of challenging samples such as FFPE DNA, and in order to maintain methylation marks on samples for methylome analysis, including for use with NEBNext Enzymatic Methyl-seq (EM-seq).

NEBNext UltraShear is a mix of enzymes that has been designed and optimized to fragment these sample types upstream of library preparation. This improves library yields and diversity, and allows retention of methylation marks.

NEBNext UltraShear increases EM-seq library yields



200 ng, 50 ng and 10 ng of NA12878 DNA spiked with control DNA (CpG methylated pUC19 DNA and unmethylated lambda DNA) were fragmented by either NEBNext UltraShear (20 minutes at 37°C) or Covaris ME220 (350 bp protocol) followed by EM-seq library preparation. Library yields were quantified using Agilent TapeStation with the High Sensitivity D1000 ScreenTape®. EM-seq libraries fragmented by NEBNext UltraShear have higher yields than Covaris for the same number of PCR cycles for each input (200 ng = 4 cycles; 50 ng = 6 cycles; 10 ng = 8 cycles).

ADVANTAGES

- Compatible with methylation analysis workflows, including NEBNext Enzymatic Methyl-seq (EM-seq)
- Compatible with FFPE DNA
- Fast workflow with minimal hands-on time
- For methylation analysis, improves CpG coverage and sequencing metrics
- For FFPE DNA, increases usable reads and coverage uniformity

PRODUCT	SIZE
NEBNext UltraShear (NEB #M7634S/L)	24/96 rxns

NEBNext Adaptors and Primers for Illumina

Adaptors and Primers are an essential component of your NGS sample prep workflow, and NEBNext Multiplex Oligos offer flexibility in multiplexing; indexing options include unique dual indices (UDIs) with unique molecular identifiers (UMIs), unique dual indices (UDIs), combinatorial dual (CD) indices, and single indices in a range of formats and indexing strategies. For an overview of our Multiplex Oligos products, refer to the NEBNext Multiplex Oligos Selection Chart below.

Multiplex Oligos Selection Chart

	SINGLE INDEX	DUAL INDEX	UNIQUE DUAL INDEX	UNIQUE DUAL INDEX UMIs
NEB PRODUCTS	**#E7335 #E7500 #E7710 #E7730 #E6609**	**#E7600 #E7780**	**#E6440, #E6442, #E6444, #E6446 #E6448, #E7140, #E3400, #E3402 #E3390, #E3406, #E3408, #E3404 #E3392**	**#E7395 #E7874 #E7876 #E7878**
Contains UMI	No	No	No	Yes
Addresses Index Hopping	No	No	Yes	Yes
Indexing Strategy	Index Primer	Index Primer	Index Primer	Index Adaptor
Adaptor Included	NEBNext Adaptor (Loop)	NEBNext Adaptor (Loop)	Standard: NEBNext Adaptor (Loop) LV: None; adaptor provided with library prep kit	Unique Dual Index UMI Adaptor
Applications	DNA-seq, RNA-seq (except small RNA)	DNA-seq, RNA-seq (except small RNA)	DNA-seq, RNA-seq (except small RNA)	DNA-seq, RNA-seq (except small RNA)
Number of Indices for Multiplexing	up to 144	up to 384	up to 480	up to 96
Compatible with EM-seq	Yes*	Yes*	Yes*	No
Compatible with EpiMark® Bisulfite Sequencing	Yes**	Yes**	Yes**	No
Number of Sets Available;	Five;	Two;	Thirteen;	Four sets for DNA, One set for RNA;
Formats and Indices Available	Sets 1-4 (12 indices/set): Individual vials 96 Index: Premixed plate	Individual vials containing 8 i5 primers and 12 i7 primers for combinatorial mixing	24 or 96 indices in premixed, foil-sealed 96-well plates, including a version for EM-seq (up to 120 indices, either 96-well plate or 24 vial format)	Adaptors with 96 indices in premixed, foil-sealed 96-well plate (DNA-seq OR RNA-seq) and primers

* Requires the use of the EM-seq Adaptor; Single, dual and unique dual index are all compatible; NEB recommends using the Unique Dual Index Primers found in the NEBNext Enzymatic Methyl-seq Kit (NEB #E7120) or the NEBNext Multiplex Oligos for EM-seq (NEB #E7140), both supplied with the NEBNext EM-seq Adaptor; For higher levels of multiplexing, Unique Dual Index Primers Sets 3 and 4 (NEB #E6444 and #E6446) are also validated for EM-seq.

** Requires use of NEBNext EM-seq adaptor from NEBNext Multiplex Oligos for Enzymatic Methyl-seq (Unique Dual Index Primer Pairs, #E7140S/L.

ADVANTAGES

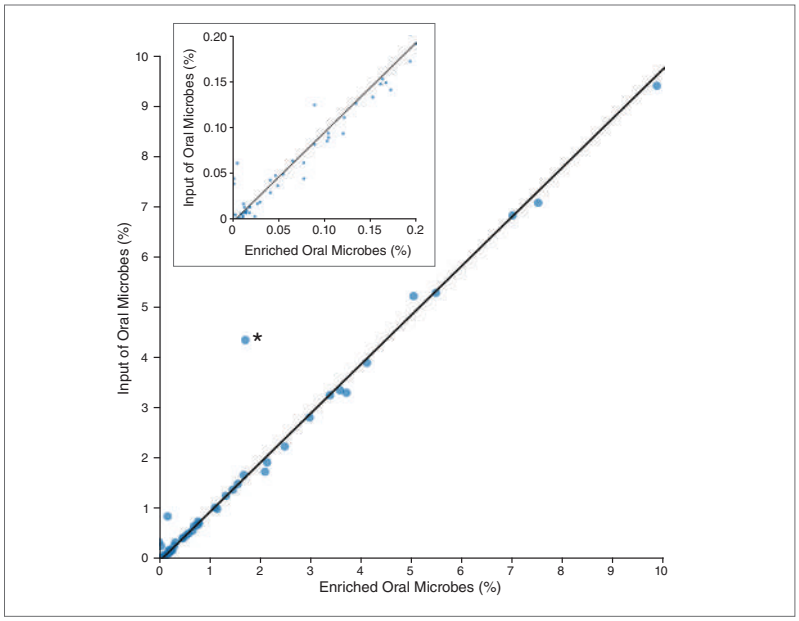
- Indexing strategies optimized by application
- Index Primers are available for NGS Library Prep workflows that include an amplification step
- Index Adaptors enable PCR-free workflows and incorporation of UMIs for error correction/ deduplication
- Extensively QC'd for purity and increased library yields
- Flexibility for use with NEBNext library preparation kits and other standard, Illumina-compatible library preparation methods
- Provided with index-pooling guidelines and sample sheets

PRODUCT	# INDICES	SIZE
NEBNext LV Unique Dual Index Primers Sets 2A, 2B (NEB #E3390, E3392)	24	24 rxns
NEBNext LV Unique Dual Index Primers Sets 1, 2, 3, 4, 5 (NEB #E3400, E3402, E3404, E3406, E3408)	96	96 rxns
NEBNext Multiplex Oligos for Illumina (Unique Dual Index UMI Adaptors DNA Set 1-4) (NEB #E7395S/L, #E7874S/L, #E7876S/L, #E7878S/L)	96 unique pairs	96/384 rxns
NEBNext Multiplex Oligos for Illumina (96 Unique Dual Index Primer Pairs or 96 Unique Dual Index Primer Pairs Set 1, 2, 3, 4, 5) (NEB #E6440S/L, #E6442S/L, #E6444S/L, #E6446S/L, #E6448S/L)	96 unique pairs	96/384 rxns
NEBNext Multiplex Oligos for Illumina (Dual Index Primers Set 1 or 2) (NEB #E7600S, #E7780S)	8 x 12	96 rxns
NEBNext Multiplex Oligos for Illumina (96 Index Primers) (NEB #E6609S/L)	96	96/384 rxns
NEBNext Multiplex Oligos for Illumina (Index Primers Set 1, 2, 3 or 4) (NEB #E7335S/L, #E7500S/L, #E7710S/L, #E7730S/L)	12	24/96 rxns
NEBNext Multiplex Oligos for Enzymatic Methyl-seq (Unique Dual Index Primer Pairs) (#E7140S/L)	96	24/96 rxns
NEBNext Multiplex Oligos for Illumina (Methylated Adaptor, Index Primers Set 1) (NEB #E7535S/L)	12	24/96 rxns
NEBNext Adaptor Dilution Buffer (NEB #B1430)		1 x 9.6 ml

NEBNext Microbiome DNA Enrichment Kit

Microbiome DNA analysis can be challenging due to the high percentage of host DNA present in many samples. The NEBNext Microbiome DNA Enrichment Kit facilitates enrichment of microbial DNA from samples containing methylated host DNA (including human), by selective binding and removal of the CpG-methylated host DNA. Importantly, microbial diversity remains intact after enrichment. If desired, the host DNA captured on the magnetic bead pellet can be eluted, and a protocol is provided for this.

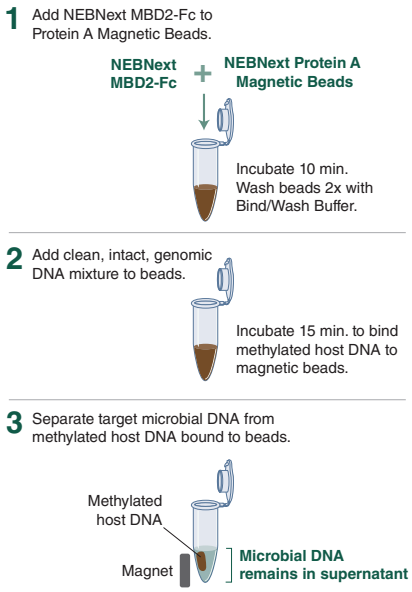
Microbiome diversity is retained after enrichment with the NEBNext Microbiome DNA Enrichment Kit



Libraries were prepared from unenriched and enriched samples, followed by sequencing on the SOLiD 4 platform. The graph shows a comparison between relative abundance of each bacterial species listed in HOMD before and after enrichment with the NEBNext Microbiome DNA Enrichment Kit. Abundance is inferred from the number of reads mapping to each species as a percentage of all reads mapping to HOMD. High concordance continues even to very low abundance species (inset). We compared 501M 50 bp SOliD 4 reads in the enriched dataset to 537M 50 bp SOLiD 4 reads in the unenriched dataset. Reads were mapped using Bowtie 0.12.7 with typical settings (2 mismatches in a 28 bp seed region, etc).

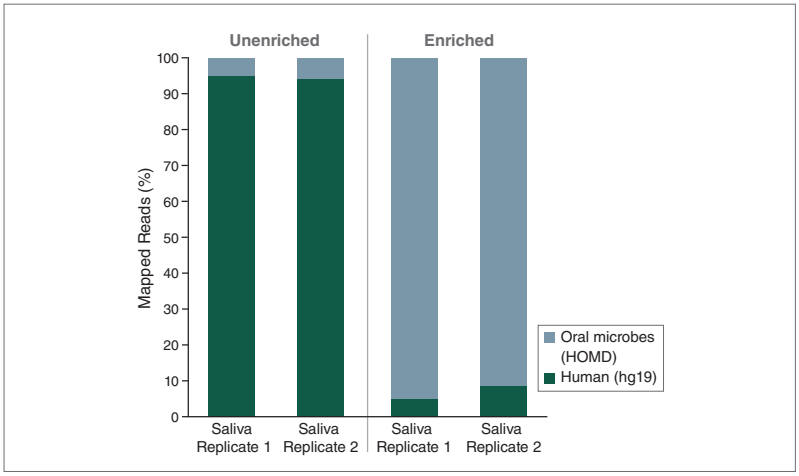
** Niesseria flavescens – This organism may have unusual methylation density, allowing it to bind the enriching beads at a low level. Other Niesseria species (N. mucosa, N. sicca and N. elognata) are represented, but do not exhibit this anomalous enrichment.*

Microbiome DNA Enrichment Kit workflow



The MBD2-Fc protein binds specifically to CpG methylated DNA. In the NEBNext Microbiome DNA Enrichment workflow, MBD2-Fc is attached to Protein A magnetic beads, enabling capture of methylated DNA, while the microbial DNA remains in the supernatant.

Salivary microbiome DNA enrichment



DNA was purified from pooled human saliva DNA (Innovative Research) and enriched using the NEBNext Microbiome DNA Enrichment Kit. Libraries were prepared from unenriched and enriched samples and sequenced on the SOLiD 4 platform. The graph shows percentages of 500 M–537 M SOLiD™4 50 bp reads that mapped to either the Human reference sequence (hg19) or to a microbe listed in Human Oral Microbiome Database (HOMD). (Because the HOMD collection is not comprehensive, ~80% of reads in the enriched samples do not map to either database.) Reads were mapped using Bowtie 0.12.7 with typical settings (2 mismatches in a 28 bp seed region, etc.).

ADVANTAGES

- Effective separation of microbial DNA from host DNA
- Enables microbiome whole genome sequencing, even for samples with high levels of host DNA
- Compatible with downstream applications including next generation sequencing on all platforms, qPCR and end-point PCR
- No requirements for live cells

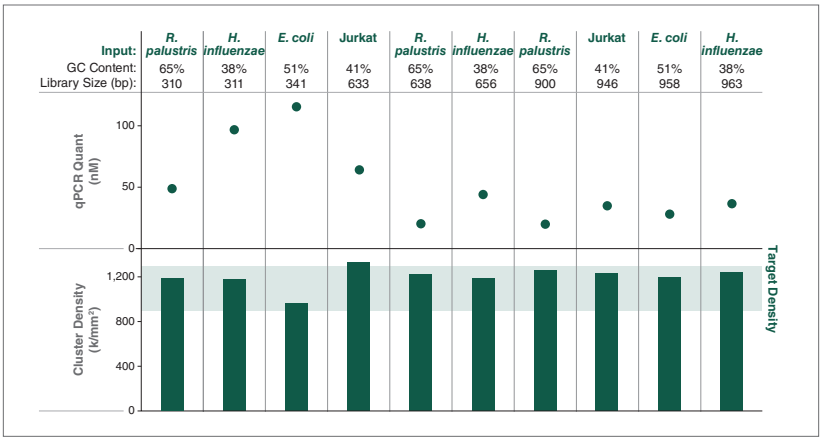
PRODUCT	SIZE
NEBNext Microbiome DNA Enrichment Kit (NEB #E2612S/L)	6/24 rxns

NEBNext Library Quant Kit for Illumina

Accurate quantitation of next-generation sequencing libraries is essential for maximizing data output and quality from each sequencing run. For Illumina sequencing specifically, accurate quantitation of libraries is critical to achieve optimal cluster densities, a requirement for optimal sequence output. qPCR is considered to be the most accurate and effective method of library quantitation, providing considerably higher consistency and reproducibility of quantitation. qPCR-based methods quantitate only those molecules that contain both adaptor sequences, thereby providing a more accurate estimate of the concentration of the library molecules that can be sequenced. The NEBNext Library Quant Kit delivers significant improvements to qPCR-based library quantitation for next-generation sequencing.

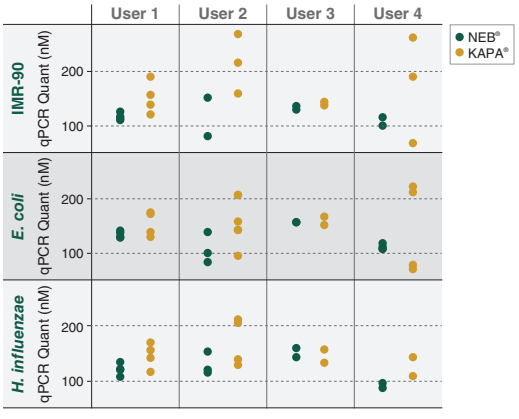
	Reagent Preparation	Library Dilution	Set Up	qPCR	Data Analysis	Total Workflow
Hands-On	5 min.	10 min.	25 min.	1 min.	10 min.	**51 min.**
Total	5 min.	10 min.	25 min.	60 min.	10 min.	**1 hr. 50 min.**

With NEBNext, optimal cluster density is achieved from quantitated libraries with a broad range of library size and GC content

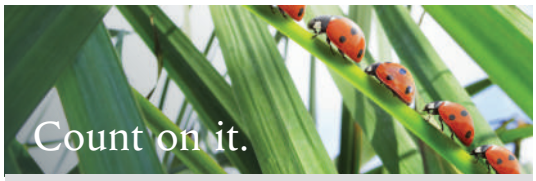


Libraries of 310–963 bp from the indicated sources were quantitated using the NEBNext Library Quant Kit, then diluted to 8 pM and loaded onto a MiSeq (v2 chemistry; MCS v2.4.1.3). Library concentrations ranged from 7–120 nM, and resulting raw cluster density for all libraries was 965–1300 k/mm2 (ave. =1199). Optimal cluster density was achieved using concentrations determined by the NEBNext Library Quant Kit for all library sizes.

Three 340–400 bp libraries were quantitated by 4 different users 2–4 times using either the NEBNext or Kapa Library Quantification Kit (Universal)



A notable improvement in quantitation consistency was observed for concentrations determined by the NEBNext Kit versus those from the Kapa kit.



- Be confident in your quant values, as our kit provides **more accurate and reproducible results** than other methods and kits
- Get up and running quickly with our **easy-to-use kit**, containing Library Dilution Buffer, optimized master mix, 6 standards and ROX dye
- Simplify your reaction setup with **fewer pipetting steps** and a **single extension time** for all libraries
- Enjoy the flexibility to use **4 or 6 standards**
- **Use with all your libraries**, regardless of insert size, GC content and preparation method
- Save money with our value pricing

TOOLS & RESOURCES

Use NEBioCalculator at **NEBioCalculator.neb.com** to calculate your qPCR-based library quant values

Download our application note, "Improved library quantitation for a broad range of library types using the NEBNext Quant Kit for Illumina" at **www.neb.com/E7630**

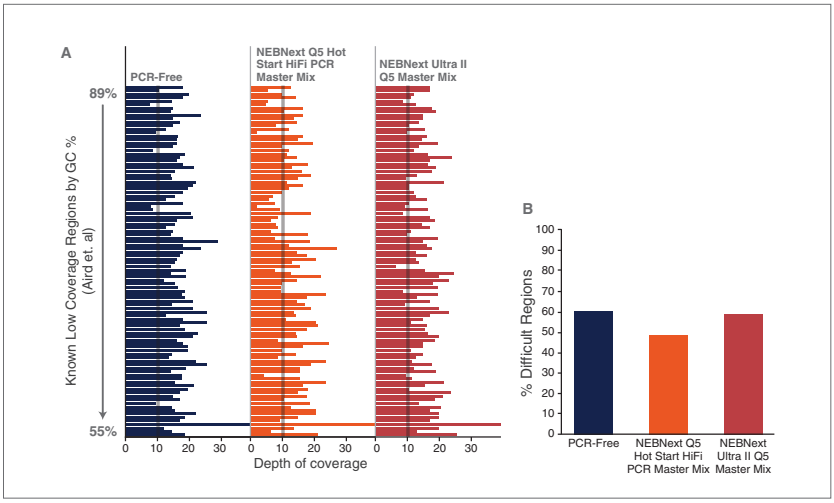
PRODUCT	SIZE
NEBNext Library Quant Kit for Illumina (NEB #E7630S/L)	100/500 rxns
NEBNext Library Dilution Buffer (NEB #B6118S)	7.5 ml

NEBNext Ultra II Q5 Master Mix

To ensure that sequence data reflects exactly the sequence of the original sample, it is essential that amplification of libraries be performed uniformly and with high fidelity. Historically, high-fidelity polymerases have been more susceptible to difficulties in PCR amplification of GC-rich and other challenging regions. If such bias occurs in library amplification, this can lead to uneven sequence coverage, challenges in sequence assembly and even "missing" sequence.

The NEBNext Ultra II Q5 Master Mix is the latest formulation of Q5 DNA polymerase that has been optimized for robust, high-fidelity amplification of next-generation sequencing (NGS) libraries. This formulation further improves the uniformity of amplification of libraries, including superior performance with GC-rich regions.

NEBNext Ultra II Q5 Master Mix provides improved coverage of known low-coverage regions of the human genome



Libraries were prepared from Human NA19240 genomic DNA. One library was not amplified. The other two libraries were amplified using 5 cycles of PCR with NEBNext Q5 Hot Start HiFi PCR Master Mix (NEB #M0543) or with NEBNext Ultra II Q5 Master Mix (NEB #M0544). Libraries were sequenced on an Illumina NextSeq® 500. 420 million 75 bp reads were randomly extracted from each dataset, representing an average coverage of 10X. Reads were mapped to the GRCh37 reference genome using Bowtie 2.2.4. Reads on each region were counted using bedtools v2.19.1.

A. The number of reads overlapping distinct low coverage regions of the human genome (1) are shown for each library.

B. From the 420 million 75 bp reads randomly extracted from each dataset, 10X coverage was expected. The % of difficult regions covered at > 10X are shown for each library. The NEBNext Ultra II Q5 Master Mix provides improved coverage of these known low coverage regions, without drop-outs, and shows similar coverage to the unamplified sample. (1) Popatov, V. and Ong, J.L. (2017). Examining Sources of Error in PCR by Single-Molecule Sequencing. PLoS ONE. 12(1):e0169774.

ADVANTAGES

- Optimized for high yields in NGS library amplification
- Minimizes GC bias, with superior performance across the GC spectrum
- Ultra-high-fidelity amplification with Q5, the highest-fidelity polymerase (2)
- Aptamer-based hot start without a separate activation step, for room-temperature reaction set-up

PRODUCT	SIZE
NEBNext Ultra II Q5 Master Mix (NEB #M0544S/L/X)	50/250/500 rxns

Reference:

1. *Aird, D. et al. (2011). Analyzing and minimizing PCR amplification bias in Illumina sequencing libraries. Genome Biology 12(2), R18.*
2. *Popatov, V. and Ong, J.L. (2017). Examining Sources of Error in PCR by Single-Molecule Sequencing. PLoS ONE. 12(1):e0169774.*

NEBNext Direct® Genotyping Solution

Do you need high-throughput targeted genotyping for Illumina Sequencing? The NEBNext Direct Genotyping Solution combines highly multiplexed, capture-based enrichment with maximum efficiency next generation sequencing to deliver cost-effective, high-throughput genotyping for a wide variety of applications. Applicable for ranges spanning 100-5,000 markers, pre-capture multiplexing of up to 96 samples combined with dual indexed sequencing allows over 3.8 million genotypes in a single Illumina sequencing run.

The NEBNext Direct Genotyping Solution begins with 25–100 ng of purified genomic DNA. The DNA molecules are enzymatically fragmented and 5´ tagged with an Illumina-compatible P5 adaptors, incorporating both an inline sample index to tag each sample prior to pooling and an inline Unique Molecular Identifier (UMI) to mark each unique DNA fragment within the samples. Up to 96 samples are subsequently pooled together prior to hybridization-based enrichment using biotinylated baits and captured on streptavidin beads. For the remainder of the protocol, up to 96 samples are processed as a single pool through ligation of a 3´ adaptor, removal of off-target sequence and final PCR, which amplifies the material and adds a second pool index to produce the final sequencing-ready fragment.

PLANT	ANIMAL	HUMAN
Marker assisted selection / breeding	Mouse Genotyping	Biobanking
Quantitative Trait Locus (QTL) Screening	Livestock Breeding	NGS Sample Tracking

Advantages:

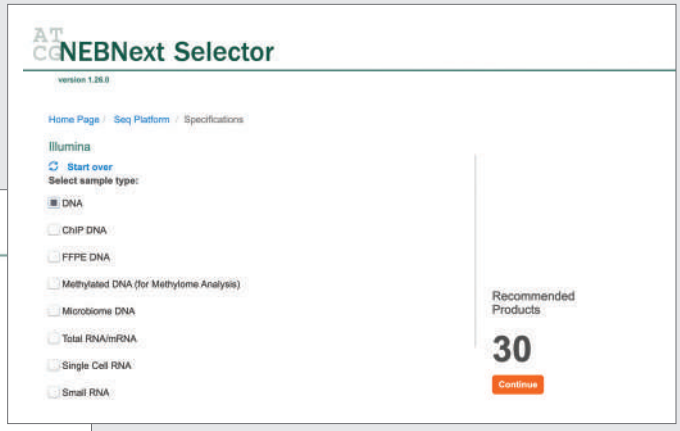
- Ideal solution for genotyping hundreds to thousands of markers
- Reduce costs and streamline workflow through pre-capture pooling of up to 96 samples
- Maximize sequencer efficiency through dual barcode sample indexing plus Unique Molecular Identifier
- Unparalleled target coverage uniformity through unique capture-based enrichment
- Eliminate marker dropouts with finely tuned bait design
- Increase sample throughput using the 1-day, automatable workflow

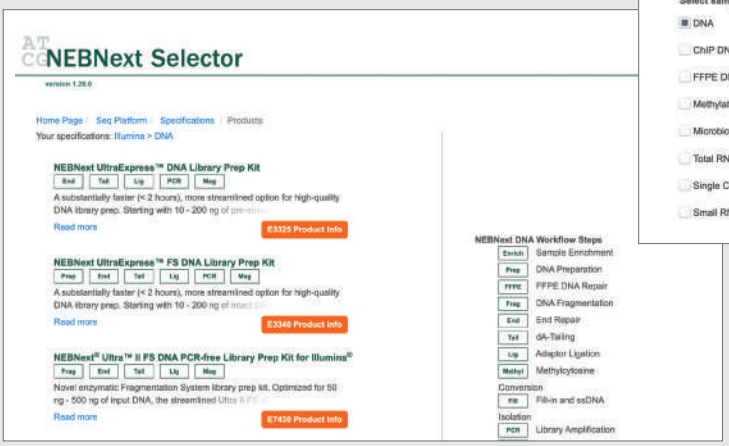
PRODUCT	SIZE
NEBNext Direct Genotyping Solution(NEB #E9500B-S)	96 rxns
NEBNext Direct Genotyping Solution (NEB #E9530B-S)	8 rxns

For research use only, not intended for diagnostic use.

NEBNext Selector

Use this tool to guide you through selection of NEBNext reagents for next generation sequencing sample preparation. Try it out at **NEBNextSelector.neb.com**





Ordering Information

KITS FOR ILLUMINA DNA LIBRARY PREPARATION		NEB #	SIZE
DNA	NEBNext UltraExpress DNA Library Prep Kit	E3325S/L	24/96 rxns
	NEBNext UltraExpress FS DNA Library Prep Kit	E3340S/L	24/96 rxns
	NEBNext Ultra II DNA Library Prep Kit for Illumina	E7645S/L	24/96 rxns
	NEBNext Ultra II DNA Library Prep with Sample Purification Beads	E7103S/L	24/96 rxns
	NEBNext Ultra II FS DNA Library Prep Kit for Illumina	E7805S/L	24/96 rxns
	NEBNext Ultra II FS DNA Library Prep with Sample Purification Beads	E6177S/L	24/96 rxns
	NEBNext Ultra II DNA PCR-free Library Prep Kit for Illumina	E7410S/L	24/96 rxns
	NEBNext Ultra II FS DNA PCR-free Library Prep Kit for Illumina	E7430S/L	24/96 rxns
	NEBNext Ultra II FS DNA PCR-free Library Prep with Sample Purification Beads	E7435L	96 rxns
	NEBNext Enzymatic Methyl-seq v2 Kit	E8015S/L	24/96 rxns
	NEBNext Enzymatic 5hmC-seq Kit	E3350S/L	24/96 rxns
	NEBNext FFPE DNA Library Prep Kit	E6650S/L	24/96 rxns
	NEBNext UltraShear FFPE DNA Library Prep Kit	E6655S/L	24/96 rxns
	NEBNext Ultra DNA Library Prep Kit for Illumina	E7370L	96 rxns
MODULES & ENZYMES		**NEB #**	**SIZE**
DNA	NEBNext Enzymatic Methyl-seq v2 Conversion Module	E8020S/L	24/96 rxns
	NEBNext Enzymatic 5hmC-seq Conversion Module	E3365S/L	24/96 rxns
	NEBNext FFPE DNA Repair v2 Module	E7360S/L	24/96 rxns
	NEBNext FFPE DNA Repair Mix	M6630S/L	24/96 rxns
	NEBNext Microbiome DNA Enrichment Kit	E2612S/L	6/24 rxns
	NEBNext UltraShear	M7634S/L	24/96 rxns
	NEBNext Ultra II FS DNA Module	E7810S/L	24/96 rxns
	NEBNext Ultra II End Repair/ dA-Tailing Module	E7546S/L	24/96 rxns
	NEBNext Ultra II Ligation Module	E7595S/L	24/96 rxns
	NEBNext Ultra II Q5 Master Mix	M0544S/L/X	50/250/500 rxns
	NEBNext dsDNA Fragmentase	M0348S/L	50/250 rxns
	NEBNext End Repair Module	E6050S/L	20/100 rxns
	NEBNext dA-Tailing Module	E6053S/L	20/100 rxns
	NEBNext Quick Ligation Module	E6056S/L	20/100 rxns
	NEBNext Q5 Hot Start HiFi PCR Master Mix	M0543S/L	50/250 rxns
	NEBNext High-Fidelity 2X PCR Master Mix	M0541S/L	50/250 rxns
	NEBNext Q5U Master Mix	M0597S/L	50/250 rxns
	NEBNext dsDNA Fragmentase Reaction Buffer v2	B0349S	6 ml

ADAPTORS & PRIMERS	NEB #	SIZE
NEBNext Multiplex Oligos for Illumina Unique Dual Index UMI Adaptors DNA Sets (1,2,3,4)	E7395S/L E7874S/L E7876S/L E7878S/L	96/384 rxns 96/384 rxns 96/384 rxns 96/384 rxns
NEBNext LV Unique Dual Index Primer Sets (2A, 2B)	E3390S E3392S	24 rxns
NEBNext LV Unique Dual Index Primer Sets (1, 2, 3, 4, 5)	E3400S E3402S E3404S E3406S E3408S	96 rxns
NEBNext Multiplex Oligos for Illumina 96 Unique Dual Index Primer Pairs Sets (1, 2, 3, 4, 5)	E6440S/L E6442S/L E6444S/L E6446S/L E6448S/L	96/384 rxns 96/384 rxns 96/384 rxns 96/384 rxns 96/384 rxns
NEBNext Multiplex Oligos for Enzymatic Methyl-seq (Unique Dual Index Primer Pairs)	E7140S/L	24/96 rxns
NEBNext Multiplex Oligos for Illumina (Dual Index Primers Set 1)	E7600S	96 rxns
NEBNext Multiplex Oligos for Illumina (Dual Index Primers Set 2)	E7780S	96 rxns
NEBNext Multiplex Oligos for Illumina Index Primers Sets (1, 2, 3, 4)	E7335S/L E7500S/L E7710S/L E7730S/L	24/96 rxns
NEBNext Multiplex Oligos for Illumina (96 Index Primers)	E6609S/L	96/384 rxns
NEBNext Adaptor Dilution Buffer	B1430S	1 x 9.6 ml
TARGET ENRICHMENT	**NEB #**	**SIZE**
NEBNext Direct Genotyping Solution	E9500B-S	96 rxns
NEBNext Direct Genotyping Solution	E9530B-S	8 rxns
LIBRARY QUANTITATION	**NEB #**	**SIZE**
NEBNext Library Quant Kit for Illumina	E7630S/L	100/500 rxns
NEBNext Library Dilution Buffer	B6118S	7.5 ml
MAGNETIC SEPARATION	**NEB #**	**SIZE**
NEBNext Magnetic Separation Rack	S1515S	24 tubes

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