



NEBNext[®] for Ion Torrent[™]

LIBRARY PREPARATION KITS

 NEW ENGLAND
BioLabs[®] Inc.

be INSPIRED
drive DISCOVERY
stay GENUINE

Table of Contents

- 3 Why choose NEBNext for Ion Torrent?
- 4–5 Workflow for Ion Torrent DNA Library Preparation
- 6 Product Selection and Product Details
- 7 Ordering Information

Why choose NEBNext for Ion Torrent?

The NEBNext suite of products supports Ion Torrent sequencing with library preparation tools that streamline workflows, minimize inputs, and improve library yields and diversity.

DNA library preparation is complete in approximately 2 hours, with minimal hands-on time. In addition to the extensive QCs on individual kit components, all NEBNext kits for Ion Torrent are functionally validated by preparation of a library, followed by Ion Torrent sequencing.

NEBNext kits for Ion Torrent include reagents for the entire library preparation workflow, and include master mix reagents that minimize the number of components and pipetting steps.

Bulk & 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.

For more information, please contact custom@neb.com.

The NEBNext Advantage

- Broad range of input amounts, from low ng to µg
- Fast workflows with minimal hands-on time
- Gel-free workflows
- High yields
- High library diversity
- Minimized GC bias
- Convenient formats include kits and modules
- All reagents undergo stringent quality controls, plus sequencing validation
- Value pricing



Learn more about NEBNext
www.NEBNext.com

Tools & Resources

Visit NEBNext.com to find:

- The full list of products available
- Protocols & FAQs
- Online tutorials to help with product selection, general handling tips and more
- NEBNext citations



Access to the NEBNext Selector Tool, our online tool for help with selecting the right NEBNext product.



Workflow for Ion Torrent DNA Library Preparation

RECOMMENDED INPUT AMOUNTS






NEBNext Fast DNA Library Prep Set for Ion Torrent

NEBNext Fast DNA Fragmentation & Library Prep Set for Ion Torrent

TOTAL DNA

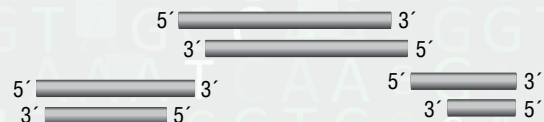
10 ng – 1 µg

10 ng – 1 µg

 DNA	 P1 Adaptor
 A Adaptor	 5' Phosphate
 PCR Primers	

1 DNA Fragmentation

The first step in DNA library preparation is generally fragmentation of purified DNA. Fragmentation can be accomplished by a number of methods including acoustic shearing, nebulization and enzyme-based methods, such as dsDNA Fragmentase[®]. All of these methods leave a mix of 3' and 5' ends (recessed, overhang, blunt) which may or may not be phosphorylated. NEBNext kits for Ion Torrent are available with or without enzyme-based fragmentation reagents, allowing a choice of fragmentation method.



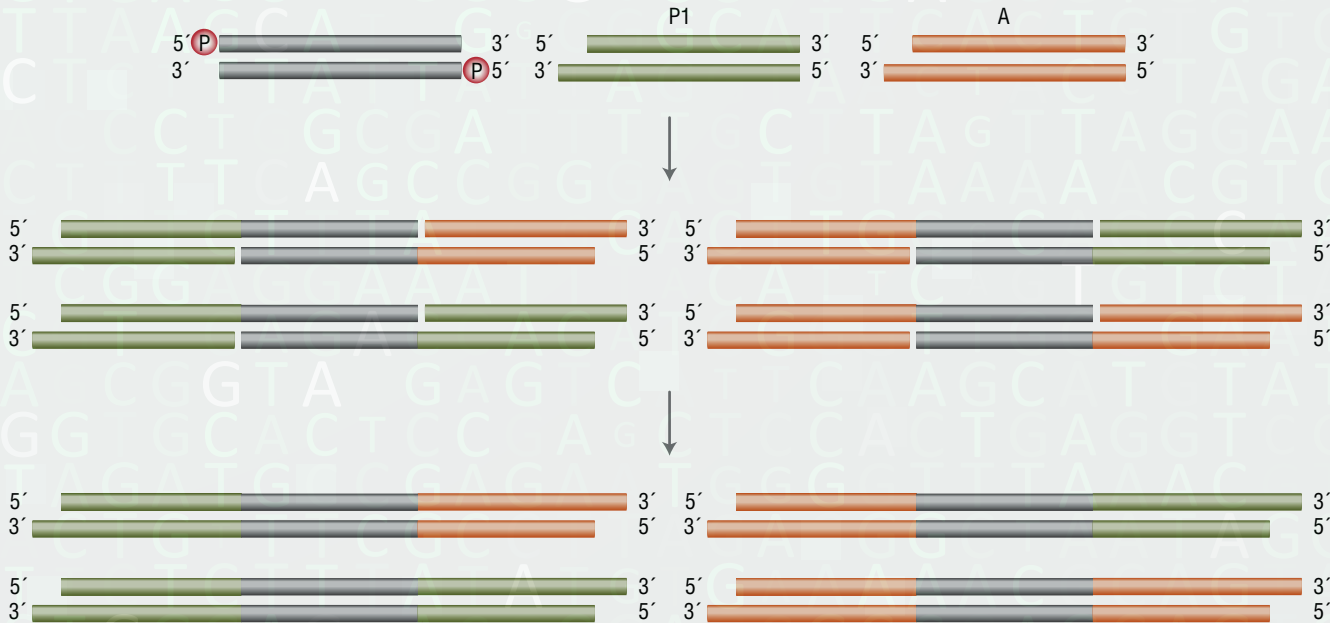
2 End Repair and 5' Phosphorylation

Blunt-ended fragments are created by filling in or chewing back 3' and 5' overhangs. Phosphorylation of the 5' ends ensures the fragments are suitable for ligation.



3 Adaptor Ligation and Nick Translation

Short adaptor sequences are added to the ends of the DNA fragments by blunt-end ligation. The adaptors are not 5' phosphorylated, in order to minimize adaptor-dimer formation. Since ligases require a 5' phosphate for ligation, this absence of 5' phosphate results in a nick on one strand at each ligation site. This nick is repaired by nick translation, using *Bst* 2.0 WarmStart® DNA Polymerase. The four possible reaction products are shown below.

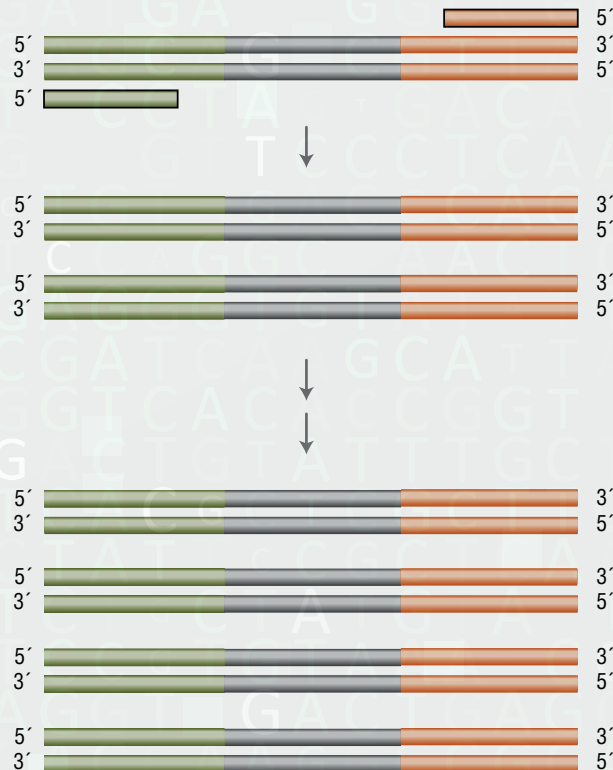


4 PCR Enrichment

Finally, amplification using a high-fidelity polymerase is performed, resulting in a robust library suitable for sequencing.

This step has multiple functions:

- Selection for molecules with the proper adaptor at each end
- Increase the amount of library
- Incorporation of sequences required downstream in the sequencing workflow, and, when desired, barcodes/indices to enable multiplexing. NEBNext kits for Ion Torrent are not supplied with barcode adaptors, but are compatible with adaptors from other sources, including Ion Xpress™ Barcode Adaptors from Life Technologies.

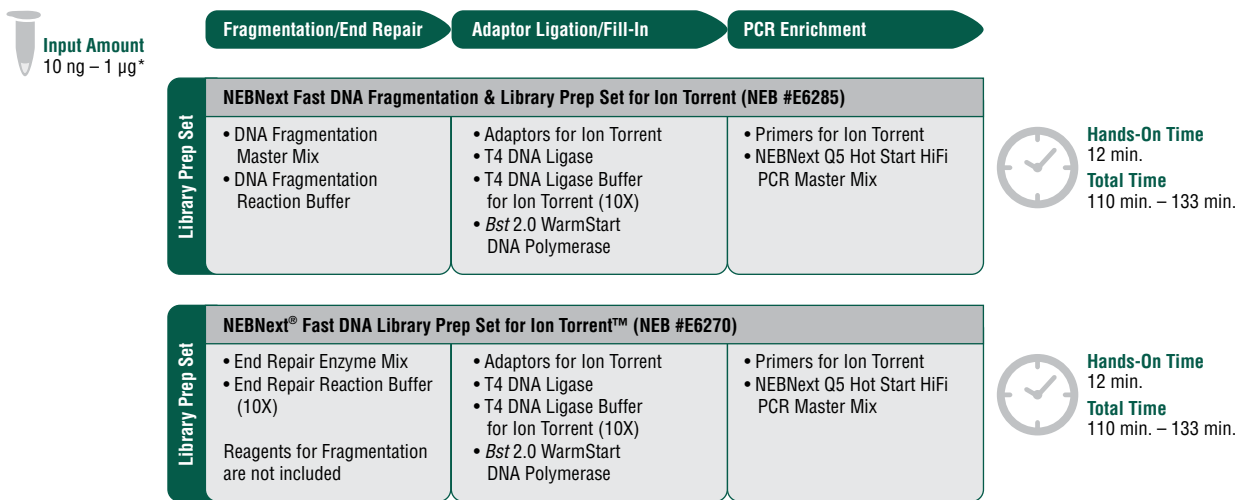


Product Selection

Designed with the user in mind, NEBNext kits maximize efficiency and convenience. For use with Ion Torrent, NEBNext kits are available with or without fragmentation reagents, allowing the user to choose the method of fragmentation. For maximum convenience and ease of scale-up, the NEBNext Fast DNA Fragmentation & Library Prep Set for Ion Torrent (NEB #E6285) includes a master mix for enzyme-based fragmentation. This set contains dsDNA Fragmentase, a mix of two enzymes for random DNA shearing, and this is combined with End Repair reagents for a streamlined workflow. When mechanical shearing methods, such as Covaris® acoustic shearing or nebulization are preferred, the NEBNext Fast DNA Library Prep Set for Ion Torrent (NEB #E6270) is the recommended choice. The components and protocol for this kit have been optimized for the volumes and concentrations of DNA sheared by mechanical methods. Both kits have the same fast workflow and low minimum input DNA requirements.

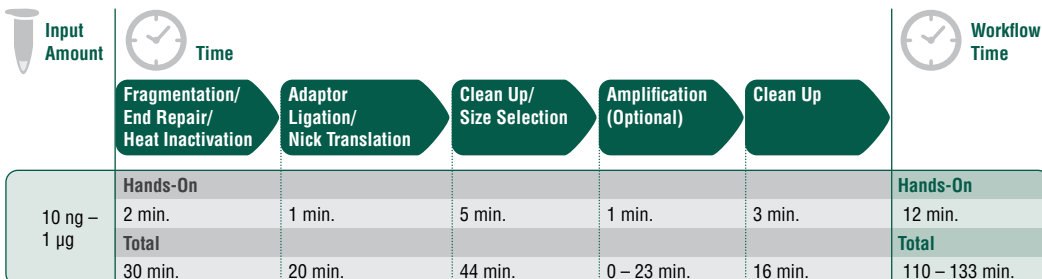
Product Details

In addition to stringent QCs on individual components, the NEBNext DNA kits are functionally validated by library preparation of a genomic DNA library, followed by Ion Torrent sequencing. Reagent lots are reserved specifically for inclusion in NEBNext kits. Most of these reagents are provided in master mix format, reducing the number of vials provided in the kits, and reducing pipetting steps. Adaptors and primers for singleplex libraries are supplied in the kits. For multiplexed libraries, the Ion Xpress Barcode Adaptors from Life Technologies can be used.

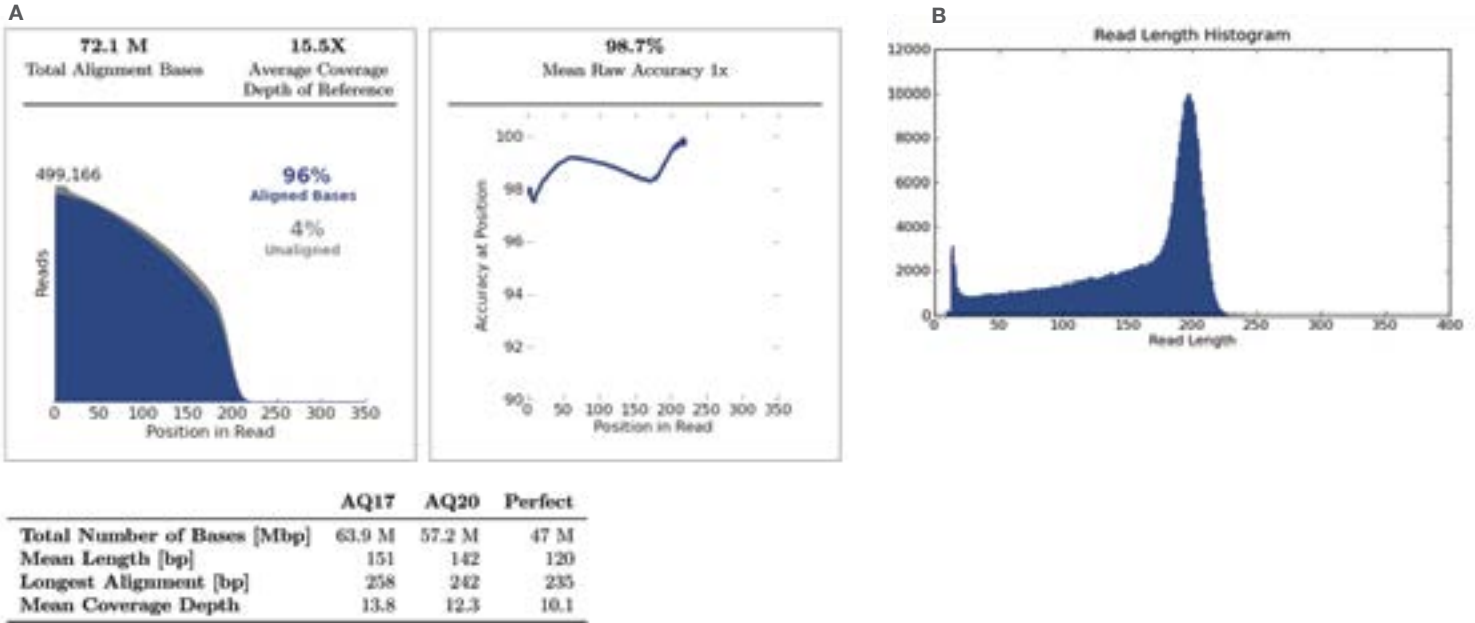


* Note that a minimum of 100 ng is recommended when used in conjunction with Ion Xpress Barcode Adaptors.

Library Preparation workflow for Ion Torrent is complete in approximately 2 hours

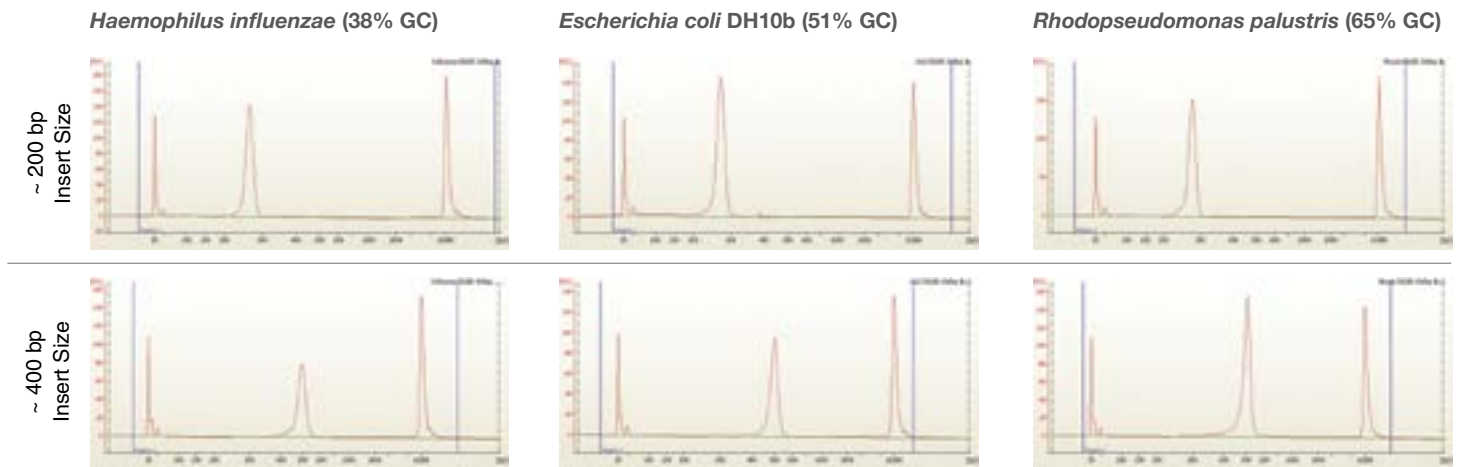


Sequencing Data



A typical Ion Torrent run report for libraries made from *E. coli* (K12 MG1655 strain) genomic DNA using NEBNext Fast DNA Fragmentation & Library Prep Set for Ion Torrent. A. Alignment Summary. 72 Mb of data was generated, with an average genome coverage of 15.5X, from approximately 0.5 Million 200 bp reads. B. Read length histogram from 200 bp run.

Varying GC Content Libraries



0.5 µg of DNA from 3 different genomes with varying GC content were used to construct 200 bp and 400 bp libraries using the NEBNext Fast DNA Fragmentation and Library Prep Set for Ion Torrent, analyzed by the Agilent® Bioanalyzer®.

Ordering Information

KITS FOR ION TORRENT DNA LIBRARY PREPARATION	NEB #	SIZE
NEBNext Fast DNA Library Prep Set for Ion Torrent	E6270S/L	10/50 rxns
NEBNext Fast DNA Fragmentation & Library Prep Set for Ion Torrent	E6285S/L	10/50 rxns

USA

New England Biolabs, Inc.
Telephone (978) 927-5054
Toll Free (USA Orders) 1-800-632-5227
Toll Free (USA Tech) 1-800-632-7799
email: info@neb.com

Australia & New Zealand

New England Biolabs (Australia) PTY
Telephone: 1800 934 218
email: info.au@neb.com
NEB New Zealand Ltd
Telephone: 0800 437 209
email: info.nz@neb.com

Canada

Telephone (905) 665-4632
Toll Free 1-800-387-1095
email: info.ca@neb.com
www.neb.ca

China

New England Biolabs (Beijing), Ltd.
Telephone 010-82378265/82378266
Free Call 400-811-2220
Free Call 400-690-3366
Fax 010-82378262
email: info@neb-china.com
www.neb-china.com

France

New England Biolabs France SAS
Telephone +33 (0) 1 69 90 87 15
Free Call 0800 100 632
email: info.fr@neb.com
www.neb-online.fr

Germany & Austria

New England Biolabs GmbH
Telephone +49 (0) 69/305 23140
Free Call 0800/246 5227 (Germany)
Free Call 00800/246 52277 (Austria)
Fax +49 (0) 69/305 23149
Free Fax 0800/246 5229 (Germany)
email: info.de@neb.com
www.neb-online.de

Japan

New England Biolabs Japan, Inc.
Telephone +81 (0) 3 4545 1422
Fax +81 (0) 3 5669 6194
email: cs.jp@neb.com
www.nebj.jp

Singapore

New England Biolabs Pte. Ltd.
Telephone +65 638 59623
Fax +65 638 59617
email: sales.sg@neb.com
www.neb.sg

United Kingdom

New England Biolabs (U.K.), Limited
Telephone +44 (0) 1462 420616
Free Call 0800 318486
Fax +44 (0) 1462 421057
Free Fax 0800 435682
email: info.uk@neb.com
www.neb.uk.com

www.neb.com



Products and content are covered by one or more patents, trademarks and/or copyrights owned or controlled by New England Biolabs, Inc. (NEB). The use of trademark symbols does not necessarily indicate that the name is trademarked in the country where it is being read; it indicates where the content was originally developed. See www.neb.com/trademarks. The use of these products may require you to obtain additional third-party intellectual property rights for certain applications. For more information, please email busdev@neb.com.

Your purchase, acceptance, and/or payment of and for NEB's products is pursuant to NEB's Terms of Sale at www.neb.com/support/terms-of-sale. NEB does not agree to and is not bound by any other terms or conditions, unless those terms and conditions have been expressly agreed to in writing by a duly authorized officer of NEB.

NEW ENGLAND BIOLABS®, NEBNEXT® and WARMSTART® are registered trademarks of New England Biolabs, Inc.

ION TORRENT™ and ION XPRESS™ are trademarks owned by Life Technologies, Inc.
AGILENT® and BIOANALYZER® are registered trademarks of Agilent Technologies.

COVARIS® is a registered trademark of Covaris.

KAPA HIFI® is a registered trademark of Kapa Biosystems.

B CORPORATION® is a registered trademark of B Lab IP, LLC, Inc.

© Copyright 2023, New England Biolabs, Inc.; all rights reserved

