

SAMPLE PREPARATION

NEBNext® dA-Tailing Module

Instruction Manual

NEB #E6053S/L
20/100 reactions
Version 2.0 4/18



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The Module Includes:

The volumes provided are sufficient for preparation of up to 20 reactions (NEB #E6053S) and 100 reactions (NEB #E6053L). (All reagents should be stored at -20°C):

Klenow Fragment ($3' \rightarrow 5'$ exo⁻)

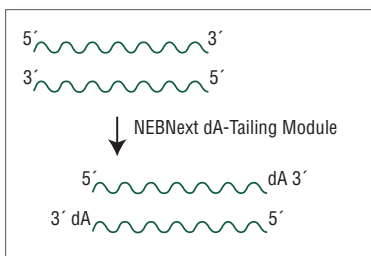
NEBNext dA-Tailing Reaction Buffer (10X)

Description:

The NEBNext dA-Tailing Module has been optimized to efficiently incorporate a non-templated dAMP on the 3' end of a blunt DNA fragment (1). 3'-dA DNA tailing prevents concatamer formation during subsequent ligation steps. DNA tailed with the NEBNext dA-Tailing module may be ligated to adaptors or cloning vectors with complementary dT overhangs. The NEBNext dA-Tailing Module is provided as a master mix to maximize efficiency and convenience in DNA sample preparation workflows.

The NEBNext dA-Tailing Module has been validated by sequencing with the Illumina Genome Analyzer II (Illumina, Inc.) in conjunction with the NEBNext End Repair Module, NEBNext Quick Ligation Module and Phusion® High-Fidelity PCR Master Mix.

For larger volume requirements, customized and bulk packaging is available by purchasing through the OEM/Bulks department at NEB. Please contact OEM@neb.com for further information.



Applications:

DNA sample preparation

dA-Tailing of 1–5 µg fragmented DNA

Advantages:

- **Efficient** – Converts 1–5 µg blunt DNA to DNA with 3'-dAMP overhangs
- **Convenient** – Reactions are provided in master mix format to reduce steps during DNA sample prep workflows
- **Automation Friendly**

References

1. Clark, J.M. et al. (1987) *J. Mol. Biol.*, 198, 123–127.

NEBNext dA-Tailing Module Protocol

Starting Material: 1–5 μg of end repaired, blunt DNA (100–1000 bp).

1. Mix the following components in a sterile microfuge tube:

End Repaired, Blunt DNA	variable
NEBNext dA-Tailing Reaction Buffer (10X)	5 μl
Klenow Fragment (3' \rightarrow 5' exo ⁻)	3 μl
Sterile H ₂ O	variable
<hr/>	
total volume	50 μl

2. Incubate in a thermal cycler for 30 minutes at 37°C.
3. Purify DNA sample on one spin column.

Kit Components

NEB #E6053S Table of Components

NEB #	PRODUCT	VOLUME
E6054A	Klenow Fragment (3' → 5' exo ⁻)	0.06 ml
E6055A	NEBNext dA-Tailing Reaction Buffer	0.1 ml

NEB #E6053L Table of Components

NEB #	PRODUCT	VOLUME
E6054AA	Klenow Fragment (3' → 5' exo ⁻)	0.30 ml
E6055AA	NEBNext dA-Tailing Reaction Buffer	0.5 ml

Revision History:

REVISION #	DESCRIPTION	DATE
1.1		3/12
2.0	Create "Kit Component – Table of Components" for small and large size kits. Delete individual component information pages.	4/18



USA

New England Biolabs, Inc.
240 County Road
Ipswich, MA 01938-2723
Telephone: (978) 927-5054
Toll Free: (USA Orders) 1-800-632-5227
Toll Free: (USA Tech) 1-800-632-7799
Fax: (978) 921-1350
e-mail: info@neb.com
www.neb.com

CANADA

New England Biolabs, Ltd.
Telephone: (905) 665-4632
Toll Free: 1-800-387-1095
Fax: (905) 665-4635
Fax Toll Free: 1-800-563-3789
e-mail: info.ca@neb.com
www.neb.ca

CHINA

New England Biolabs (Beijing), Ltd.
Telephone: 010-82378265/82378266
Fax: 010-82378262
e-mail: info@neb-china.com
www.neb-china.com

FRANCE

New England Biolabs France
Free Call: 0800-100-632
Free Fax: 0800-100-610
e-mail: 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)
e-mail: info.de@neb.com
www.neb-online.de

JAPAN

New England Biolabs Japan, Inc.
Telephone: +81 (0)3 5669 6191
Fax: +81 (0)3 5669 6192
e-mail: info.jp@neb.com
www.nebj.jp

SINGAPORE

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

UNITED KINGDOM

New England Biolabs (UK) Ltd.
Telephone: (01462) 420616
Call Free: 0800 318486
Fax: (01462) 421057
Fax Free: 0800 435682
e-mail: info.uk@neb.com
www.neb.uk.com