Sequencing Standards, BigDye[™] Terminator v3.1

SeqStudio[™] Flex, SeqStudio[™], and 3500 series instruments

Catalog Numbers 4404312

Pub. No. 4404319 Rev. D

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WARNING! Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. Safety Data Sheets (SDSs) are available from **thermofisher.com/support**.

Product description

The Sequencing Standards, BigDye[™] Terminator v3.1, contains DNA of a known sequence that is prepared with BigDye[™] Terminator v3.1. The DNA contained in the preparation has been lyophilized to maximize stability.

The kit contains Dye Set Z, which defines the number, dye color, and migration order of the dye peaks in the sample.

Use the kit to perform one of the following runs on the SeqStudio[™] Flex, SeqStudio[™], or 3500/3500xL Genetic Analyzers:

- · Installation run
- Installation run with spectral calibration
- Spectral calibration
- · Control sequencing run

Contents and storage

Contents	Amount	Storage
Sequencing Standards, BigDye [™] Terminator v3.1	4 tubes	Store at –15°C to –25°C.
		See the expiration date on the package. Do not use expired product.

Required materials not supplied

	Cat. No.	
Hi-Di™ Formamide	4311320	
MicroAmp™ Fast Optical 96-Well Reaction Plate, 0.1 mL		4346907
MicroAmp™ Optical 96-Well Reaction Plate N8010560		N8010560
Septa		
SeqStudio™ Flex and 3500 series	8-Strip Septa 3500/Flex Series (Qty 24)	4410701
	96-Well Septa 3500/Flex Series	4412614
SeqStudio™	Septa for SeqStudio™ Genetic Analyzer, 96 well	A35641

Control sequence

1	GAATTCCCCT	GCAGGCGTGG	CTGCAGCCTG	GTTATGATTA	СТСТТААТСТ	ТССТАСТАСТ	GCTGACAATG	СТССТССТСС
Ω1			TCCTTGAACA					
01								
161	CAGATCAGAA	CCACCACAGT	CAATATCACC	ACCTTCCTCT	TATAGATTCG	GAATCTCATG	ATAGGGGCTC	AGCCTCTGTG
241	CGAGTGGAGA	GAAGTTTGCA	GGCGAGCTGA	GGAGCAATTG	CAGGTGATAT	GATGTGCTCG	GCTCAAGAAG	CGGGCCCGGA
321	GAGGAAGAAG	TCGTGCCGGG	GCTAATTATT	GGCAAAACGA	GCTCTTGTTG	TAAACATTGA	TCCAACTGGA	ATGTCACTAA
401	TGGCGAATCA	ATATTCCATA	AGGCATGATG	GTTGCTCAGA	GGCAGGAGAA	GAGCAACGAA	TACGATCCTA	TAAAAGATAA
481	AACATAAATA	AACAGTCTTG	ATTATATTCT	GGGTATTAAA	GCCACAATCA	GAACAAATAT	ATGCTTTGTA	TCTTTTCTTG
561	CCTTCTTCAT	TACCAACTGC	TTCCGCGGCC	ACATTAAGAG	AACTTGTGGT	AAGATAAGAA	GATATTTTAT	TCGTTCTGCT
641	GACTTGCTGG	ATGTCGGGAA	ATATTCTGCA	TTTGATAAGA	GGCGGTTAAT	TGCAGATATA	ATTGGTAGTG	AAAAGGGTCG
721	TTGCTATGGT	CACCGTGAAG	CGAGTACAGC	AGCACAAGAA	TGTGTGCCGT	TCTCAGTTAA	TATTGTTTGA	ATATGGTAAC
801	CTGTTTTAGT	CGGTTTAAAG	GTAAGAAGAT	CTAACCAAAA	ACAACACTGC	AGTGACTGAT	TGTAGTATTT	ATTTTTTTAC
881	TTAATCTTAA	${\tt TTTTGGTGTA}$	AACATCAACG	GCGCACTTCA	ACCAATACTC	CAATGTTTTA	TCCATCGACA	TGACGTTCGA
961	GATAGGGTTG	AGTGTTGTTC	CAGTTTGGAA	CAAGAGTCCA	CTATTAAAGA	ACGTGGACTC	CAACGTCAAA	GGGCGAAAAA
1041	CCGTCTATCA	GGGCGATGGC	CCACTACGTG	AACCATCACC	CAAATCAAGT	TTTTTGGGGT	CGAGGTGCCG	TAAAGCACTA
1121	AATCGGAACC	CTAAAGGGAG	CCCCCGATTT	AGAGCTTGAC	GGGGAAAGCC	GGCGAACGTG	GCGAGAAAGG	AAGGGAAGAA
1201	AG							



Prepare the standard

- 1. Briefly centrifuge the sequencing standard tube to bring the contents to the bottom.
- Resuspend 1 tube of the sequencing standard with 300 µL of Hi-Di[™] Formamide.
- 3. Tightly cap the tube, vortex at full speed for 1 minute, then briefly centrifuge.
- 4. Heat the tube for 2 minutes at 95°C to denature the DNA fragments, then immediately place on ice for ≥2 minutes.
- 5. Dispense the prepared standard into the appropriate wells of a reaction plate. See "Well locations for the prepared standard" on page 2.
- 6. Cover the plate with septa, then centrifuge the plate to bring the mixture to the bottom and eliminate air bubbles.

For more information on setting up the run, see the instrument user guide.

Note: For dye set selection on the SeqStudio[™] Flex and SeqStudio[™] instruments, ensure that you select the **Sequencing** tab before you select the dye set.

Well locations for the prepared standard

Table 1 SeqStudio™ Flex Series Genetic Analyzer

Component	Well location for the prepared standard			
	96-well plate	384-well plate		
Prepared standard		Dispense 5 µL of the prepared standard into wells of a 384-well plate:		
	• 24-capillary array —24 wells (for example, A1–H3, A4–H6,	24-capillary array —24 wells (for example, A1, A3, A5; C1, C3, C5; E1, E3, E5; G1, G3, G5; I1, I3, I5; K1, K3, K5; M1, M3, M5; O1, O3, O5)		

Table 2 SegStudio™ Genetic Analyzer

Component Well location for the prepared standard	
Prepared standard	Dispense 10 µL of the prepared standard into wells of a 96-well plate:
	4 wells (for example, A1–D1)

Table 3 3500/3500xL Genetic Analyzer

Component	Well location for the prepared standard
Prepared standard	Data Collection Software v3 and later:
	Dispense 10 µL of the prepared standard into wells of a 96-well plate:
	8-capillary array — 8 wells (for example, A1–H1)
	• 24-capillary array – 24 wells (for example, A1–H3, A4–H6, A7–H9, or A10–H12)
	Note: If you place the standard in wells that do not correspond to injection position 1, specify the starting well position in the software.
	Data Collection Software v1, v1.1, and v2:
	Dispense 10 µL of the prepared standard into wells of a 96-well plate:
	8-capillary array—8 wells: A1–H1
	• 24-capillary array—24 wells: A1–H3

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Revision history: Pub. No. 4404319

Revision	Date	Description
D	,	Added the SeqStudio™ Flex Series Genetic Analyzer. Clarified the dye set; clarified runs that can be performed with the kit. Added required materials table. Consolidated "Prepare the standard" into one procedure. Added "Well locations for the prepared standard". Changed the manufacturing address to Vilnius.
С	04 May 2018	Corrected dye set for sequencing from E to Z
В	26 June 2017	Added support for SeqStudio™ Genetic Analyzer
A	6 March 2009	New document

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