

# Sequencing Standards, BigDye™ Terminator v1.1

SeqStudio™ Flex, SeqStudio™, and 3500 series instruments

Catalog Number 4404314

Pub. No. 4404320 Rev. C

 **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](http://thermofisher.com/support).

## Product description

The Sequencing Standards, BigDye™ Terminator v1.1, contains DNA of a known sequence that is prepared with BigDye™ Terminator v1.1. The DNA contained in the preparation has been lyophilized to maximize stability.

The kit contains Dye Set E, 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 v1.1	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

Item	Cat. No.	
Hi-Di™ Formamide	4311320	
MicroAmp™ Fast Optical 96-Well Reaction Plate, 0.1 mL	4346907	
MicroAmp™ Optical 96-Well Reaction Plate	N8010560	
<b>Septa</b>		
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

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1 GAATCCCCT GCAGGCGTGG CTGCAGCCTG GTTATGATTA CTGTTAATGT TGCTACTACT GCTGACAATG CTGCTGCTGC
81 TTCTCCTCAC TGTCTCCACT TCCTTGAACA ATGCGCCGTC ATGCTTCTTT TGCTCCCGC TGCTCCAGAA AGCTAGGCCG
161 CAGATCAGAA CCACCACAGT CAATATCACC ACCTTCCTCT TATAGATTCG GAATCTCATG ATAGGGGCTC AGCCTCTGTG
241 CGAGTGGAGA GAAGTTTGA GCGGAGCTGA GGAGCAATTG CAGGTGATAT GATGTGCTCG GCTCAAGAAG CGGGCCCGGA
321 GAGGAAGAAG TCGTGCCGGG GCTAATTAT GGCAAAACGA GCTCTTGTG TAAACATTGA TCCAACTGGA ATGTCACTAA
401 TGGCGAATCA ATATTCCATA AGGCATGATG GTTGCTCAGA GGCAGGAGAA GAGCAACGAA TACGATCCTA TAAAAGATAA
481 AACATAAATA AACAGTCTTG ATTATATCT GGGTATTAAG GCCACAATCA GAACAAATAT ATGCTTTGTA TCTTTCTTG
561 CCTTCTTCAT TACCAACTGC TTCCGCGGCC ACATTAAGAG AACTTGTGGT AAGATAAGAA GATATTTAT TCGTTCCTGCT
641 GACTTGCTGG ATGTCGGGAA ATATTCTGCA TTTGATAAGA GCGGTTAAT TGCAGATATA ATTGGTAGTG AAAAGGGTCG
721 TTGCTATGGT CACCGTGAAG CGAGTACAGC AGCACAAGAA TGTGTGCCGT TCTCAGTTAA TATTGTTTGA ATATGGTAAC
801 CTGTTTTAGT CGGTTTAAAG GTAAGAAGAT CTAACCAAAA ACAACACTGC AGTGACTGAT TGTAGTATTT ATTTTTTTAC
881 TTAATCTTAA TTTTGGTGTA AACATCAACG GCGCACTTCA ACCAATACTC CAATGTTTTA TCCATCGACA TGACGTTCGA
961 GATAGGGTTG AGTGTGTTC CAGTTTGGAA CAAGAGTCCA CTATTAAGA ACGTGGACTC CAACGTCAA GGGCGAAAAA
1041 CCGTCTATCA GGGCGATGCC CCACTACGTG AACCATCACC CAAATCAAGT TTTTGGGGT CGAGGTGCCG TAAAGCACTA
1121 AATCGGAACC CTAAGGGGAG CCCCAGATT AGAGCTTGAC GGGGAAAGCC GGCGAACGTG GCGAGAAAGG AAGGGAAGAA
1201 AG
    
```

## Prepare the standard

1. Briefly centrifuge the sequencing standard tube to bring the contents to the bottom.
2. Resuspend 1 tube of the sequencing standard with 300  $\mu$ L 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  $\geq$ 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 10 $\mu$ L of the prepared standard into wells of a 96-well plate: <ul style="list-style-type: none"><li>• <b>8-capillary array</b>—8 wells (for example, A1–H1)</li><li>• <b>24-capillary array</b>—24 wells (for example, A1–H3, A4–H6, A7–H9, or A10–H12)</li></ul>	Dispense 5 $\mu$ L of the prepared standard into wells of a 384-well plate: <ul style="list-style-type: none"><li>• <b>24-capillary array</b>—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)</li></ul>

Table 2 SeqStudio™ Genetic Analyzer

Component	Well location for the prepared standard
Prepared standard	Dispense 10 $\mu$ L 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	<b>Data Collection Software v3 and later:</b> Dispense 10 $\mu$ L of the prepared standard into wells of a 96-well plate: <ul style="list-style-type: none"><li>• <b>8-capillary array</b>—8 wells (for example, A1–H1)</li><li>• <b>24-capillary array</b>—24 wells (for example, A1–H3, A4–H6, A7–H9, or A10–H12)</li></ul> <b>Note:</b> If you place the standard in wells that do not correspond to injection position 1, specify the starting well position in the software. <b>Data Collection Software v1, v1.1, and v2:</b> Dispense 10 $\mu$ L of the prepared standard into wells of a 96-well plate: <ul style="list-style-type: none"><li>• <b>8-capillary array</b>—8 wells: A1–H1</li><li>• <b>24-capillary array</b>—24 wells: A1–H3</li></ul>

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**Revision history:** Pub. No. 4404320

Revision	Date	Description
C	2 February 2022	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.
B	26 June 2017	Added support for SeqStudio™ Genetic Analyzer
A	6 March 2009	New document

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