Bioo Scientific®

FOR REFERENCE PURPOSES

This manual is for Reference Purposes Only. DO NOT use this protocol to run your assays. Periodically, optimizations and revisions are made to the kit and protocol, so it is important to always use the protocol included with the kit.

NEXTflex™ Cell Free DNA-Seq Kit

(Illumina Compatible)

Catalog #5150-01 (8 reactions)

 V14.09

This product is for research use only.

NEXTflex™ Cell Free DNA-Seq Kit - 5150-01

Product Overview

Cell free DNA has become a powerful marker in clinical applications due to the unique origin of DNA molecules present in plasma. Detection of fetal DNA from maternal plasma has proven to be a viable, non-invasive option to identify a variety of fetal traits including: sex determination, sex chromosome-linked disorders and aneuploidy events. Circulating tumor DNA extracted from plasma of symptomatic patients can be used as a non-invasive resource for diagnosis, prognosis, treatment decisions, and follow-up monitoring of cancer patients.

The NEXTflex™ Cell Free DNA-Seq Kit is designed for 3 hour DNA library construction of cell free fetal or circulating tumor DNA. The kit can be used to prepare single, paired-end and multiplexed DNA libraries for sequencing using Illumina® platforms. The NEXTflex™ 1-step End-Repair and Adenylation protocol simplifies workflow and shortens hands-on library construction time. In addition, the availability of up to 192 unique adapter barcodes facilitates high-throughput applications.

There are four main steps involved in preparing cell free DNA for sequencing: DNA End Repair & Adenylation, Adapter Ligation and PCR Amplification. The optional Gel-Free Size Selection step performed before End Repair & Adenylation is designed to enrich for 180 bp inserts. The NEXTflex™ Cell Free DNA-Seq Kit contains the necessary material to take the user's purified cell free DNA through library preparation and amplification for loading onto Illumina flow cells for sequencing. The NEXTflex™ Cell Free DNA-Seq Kit is intended for research use only.

Revision History

Version	Date	Description
V 14.07	July 2014	Initial Product Launch
V 14.09	September 2014	Added Optional Gel-Free Size Selection

Contents, Storage and Shelf Life

The NEXTflex™ Cell Free DNA-Seq Kit contains enough material to prepare 8 DNA samples for Illumina® compatible sequencing. The shelf life of all reagents is 12 months when stored properly. The NEXTflex™ Resuspension Buffer should be stored at room temperature. All other components can be safely stored at -20°C.

Kit Contents	Amount
CLEAR CAP	
NEXTflex™ End-Repair & Adenylation Buffer Mix	120 µL
NEXTflex™ End-Repair & Adenylation Enzyme Mix	24 µL
PURPLE CAP	
NEXTflex™ Ligase Enzyme Mix	380 µL
NEXTflex™ DNA-Seq Adapter 1	20 µL
GREEN CAP	
NEXTflex™ PCR Master Mix	96 µL
NEXTflex™ Primer Mix	16 µL
WHITE CAP	
Nuclease-free Water	1 mL
NEXTflex™ Resuspension Buffer	(2) 1 mL

Required Materials Not Provided

- 1-5 ng of Cell free DNA in up to 50 µL nuclease-free water.
- (Optional) NEXTflex™ DNA Barcodes – 6 / 12 / 24 / 48 (Cat # 514101, 514102, 514103, 514104) or NEXTflex-96™ DNA Barcodes (Cat # 514105, 514106) or NEXTflex™ Dual-Indexed DNA Barcodes (Cat # 514160, 514161)
- Ethanol 100% (room temperature)
- Ethanol 80% (room temperature)
- 96 well PCR Plate Non-skirted (Phenix Research, Cat # MPS-499) or similar
- 96 well Library Storage and Pooling Plate (Fisher Scientific, Cat # AB-0765) or similar
- Adhesive PCR Plate Seal (BioRad, Cat # MSB1001)
- Agencourt AMPure XP 60 mL (Beckman Coulter Genomics, Cat # A63880)
- Magnetic Stand -96 (Ambion, Cat # AM10027) / or / similar
- Thermocycler
- 2, 10, 20, 200 and 1000 µL pipettes / multichannel pipettes
- Nuclease-free barrier pipette tips
- Vortex

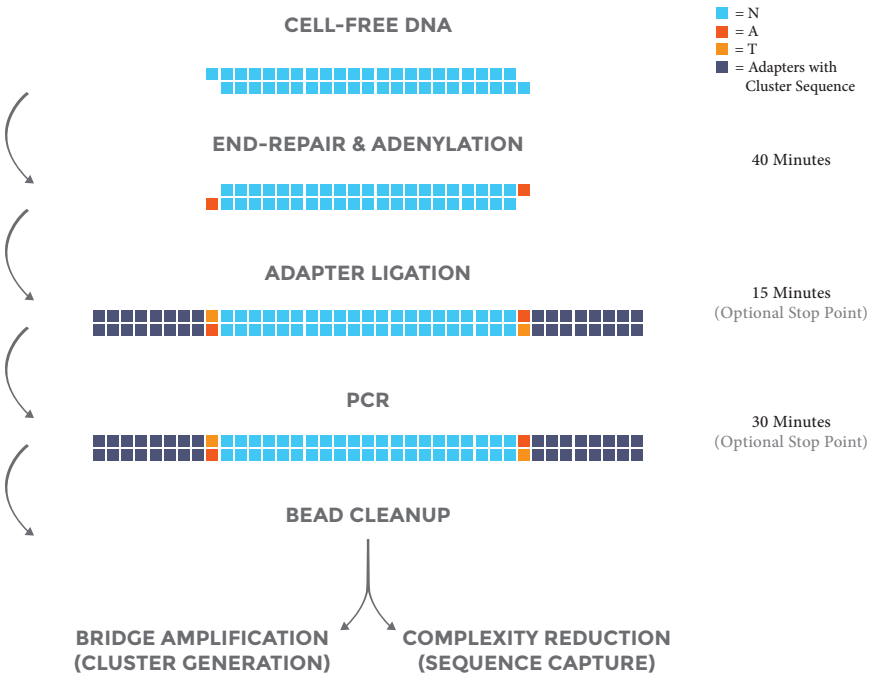
Warnings and Precautions

Bioo Scientific strongly recommends that you read the following warnings and precautions. Periodically, optimizations and revisions are made to the components and manual. Therefore, it is important to follow the protocol included with the kit. If you need further assistance, you may contact your local distributor, or contact Bioo Scientific at nextgen@biooscientific.com.

- Do not use the kit past the expiration date.
- DTT in buffers may precipitate after freezing. If precipitate is seen, vortex buffer for 1-2 minutes or until the precipitate is in solution. The performance of the buffer is not affected once the precipitate is in solution.
- Ensure pipettes are properly calibrated as library preparations are highly sensitive to pipetting error.
- Do not heat the DNA Adapter above room temperature.
- This kit contains a single barcoded DNA Adapter. To enable multiplexing, please use the appropriate combination of DNA Barcodes in place of the DNA Adapter during the Adapter Ligation step.
- Maintain a laboratory temperature of 20°–25°C (68°–77°F).
- Cell free DNA sample quality may vary between preparations. It is highly recommended fluorescent dyes be used as a means for cell free DNA sample quantification, as NanoDrop cannot accurately detect nucleic acids at concentrations found in pure cell free DNA sample preps (0.05 ng/µL to 1 ng/µL). The user should be aware that contaminating RNA, nucleotides and single-stranded DNA may affect the amount of usable DNA in a sample preparation.
- It is highly recommended that NEXTflex™ Primer Mix be used during PCR amplification. Inadvertent use of an incorrect primer sequence can potentially result in elimination of the index.

NEXTflex™ Cell Free DNA-Seq Flow Chart

Figure 1: Sample flow chart with approximate times necessary for each step.



Starting Material

The NEXTflex™ Cell Free DNA-Seq Kit has been optimized and validated using cell free DNA. Starting with 1-5 ng of high quality cell free DNA will allow you to perform at least 8 reactions (see page 3, Warnings and Precautions).

Reagent Preparation

1. Briefly spin down each component to ensure material has not lodged in the cap or side of tube. Keep on ice and vortex each NEXTflex™ component just prior to use. Sizing Solution and Resuspension Buffer can be stored at room temperature.
2. DTT in buffers may precipitate after freezing. If precipitate is seen in any mix, vortex for 1 minute or until the precipitate is in solution. The performance of the mix is not affected once the precipitate is in solution.
3. Allow Agencourt AMPure XP Beads to come to room temperature and vortex the beads until homogenous.

Gel-Free Size Selection: (Optional)

This optional size selection step is designed to isolate 180 bp inserts. Users who are not interested in enriching for 180 bp inserts can begin the protocol at Step A: End-Repair and Adenylation.

Materials

Bioo Scientific Supplied

WHITE CAP - NEXTflex™ Resuspension Buffer

User Supplied

Cell free DNA in 50 µL or less Nuclease-free Water
96 well PCR Plate
Adhesive PCR Plate Seal
Agencourt AMPure XP Magnetic Beads
80% Ethanol, freshly prepared (room temperature)
Magnetic Stand

If sample volume is < 50 µL, bring to 50 µL with Nuclease-free Water

1. Add 37.5 µL of AMPure XP Beads to 50 µL cell-free DNA sample. Mix thoroughly until homogenized.
2. Incubate sample at room temperature for 5 minutes.
3. Place the 96 well PCR Plate on the magnetic stand at room temperature for 3 minutes or until sample appears clear.
4. Do not discard the sample in this step. Transfer 85 µL of clear supernatant to a new well.
5. Add 50 µL of AMPure XP Beads to each sample. Mix thoroughly until homogenized.
6. Incubate sample at room temperature for 5 minutes.
7. Place the 96 well PCR Plate on the magnetic stand at room temperature for 3 minutes or until sample is clear.
8. Remove and discard clear supernatant.
9. With plate on stand, add 200 µL of 80% ethanol to each magnetic bead pellet and incubate plate at room temperature for 30 seconds. Carefully remove ethanol by pipette.
10. Repeat step 9 for a total of 2 ethanol washes. Ensure all ethanol has been removed.
11. Remove the plate from the magnetic stand and let dry at room temperature for 3 minutes or until bead pellet is visibly dry.
12. Resuspend dried beads with 34 µL Resuspension Buffer. Mix thoroughly until homogenized
13. Incubate sample at room temperature for 2 minutes.
14. Place the 96 well PCR Plate on the magnetic stand at room temperature for 3 minutes or until sample is clear.
15. Do not discard the sample in this step. Transfer 32 µL of clear sample to a new well.
16. Proceed to Step A: End-Repair and Adenylation.

STEP A: End-Repair & Adenylation

Materials

Bioo Scientific Supplied

CLEAR CAP -NEXTflex™ End-Repair & Adenylation Buffer Mix, NEXTflex™ End-Repair & Adenylation Enzyme Mix

User Supplied

Cell free DNA in 32 µL nuclease-free water
96 well PCR Plate
Adhesive PCR Plate Seal
Agencourt AMPure XP Magnetic Beads
Microcentrifuge
Ice

1. For each sample, combine the following reagents on ice in a nuclease-free 96 well PCR Plate:

32 µL	Cell free DNA
15 µL	NEXTflex™ End-Repair & Adenylation Buffer Mix
3 µL	NEXTflex™ End-Repair & Adenylation Enzyme Mix
50 µL	TOTAL

2. Apply adhesive PCR plate seal and incubate on a thermocycler using the following program:

20 min	22 °C
20 min	72 °C
end	4 °C

3. Proceed to Step B: Adapter Ligation.

STEP B: Adapter Ligation

Materials

Bioo Scientific Supplied

PURPLE CAP - NEXTflex™ Ligase Enzyme Mix, NEXTflex™ DNA Adapter 1 (25 µM)
WHITE CAP - Nuclease-free Water

User Supplied

Thermocycler
50 µL of End Polished DNA (from STEP A)

Optional

NEXTflex™ DNA Barcodes – 6 / 12 / 24 / 48 (Cat # 514101, 514102, 514103, 514104) or NEXTflex-96™ DNA Barcodes (Cat # 514106) or NEXTflex™ Dual-Indexed DNA Barcodes (Cat # 514160, 514161)

1. Thaw NEXTflex™ Ligase Enzyme Mix to room temperature, and vortex for 5-10 seconds. Do not spin down tube, as this may cause components of the mix to separate and affect performance.

   For some samples, diluting the DNA barcoded adapter may be necessary. If the starting material was less than 5 ng, a 1:8 dilution with Nuclease-free Water is recommended. If the starting material was less than 1 ng, further adapter dilution may be necessary.

   The following reaction must be mixed thoroughly. The NEXTflex™ Ligase Enzyme Mix is very viscous. Thorough mixing of the reaction below is critical to obtaining optimal results. Suggestion: To mix, pipette up and down 15 times; visually inspect tubes to ensure proper homogenization.

   Combine the following in the PCR plate and mix thoroughly by pipette:

50 µL	End Polished DNA (from Step A)
47.5 µL	NEXTflex™ Ligase Enzyme Mix
2.5 µL	NEXTflex™ DNA Barcode
100 µL	TOTAL

2. Apply adhesive PCR plate seal and incubate on a thermocycler for 15 minutes at 22°C.
3. Proceed to Step C: Post-Ligation Cleanup.

STEP C: Post-Ligation Cleanup

Materials

Bioo Scientific Supplied
WHITE CAP - Resuspension Buffer

User Supplied
Agencourt AMPure XP Magnetic Beads (room temperature)
80% Ethanol, freshly prepared (room temperature)
Magnetic Stand
100 µL of Adapter Ligated DNA (from STEP B)

1. Add 60 µL of AMPure XP Beads to each sample. Mix thoroughly until homogenized.
2. Incubate sample at room temperature for 5 minutes.
3. Place the 96 well PCR Plate on the magnetic stand at room temperature for 5 minutes or until sample appears clear.
4. Remove and discard clear supernatant.
5. With plate on stand, add 200 µL of 80% ethanol to each magnetic bead pellet and incubate plate at room temperature for 30 seconds. Carefully remove ethanol by pipette.
6. Repeat step 5 for a total of 2 ethanol washes. Ensure all ethanol has been removed.
7. Remove the plate from the magnetic stand and let dry at room temperature for 3 minutes or until bead pellet is visibly dry.
8. Resuspend dried beads with 51 µL Resuspension Buffer. Mix thoroughly until homogenized.
9. Incubate sample at room temperature for 2 minutes.
10. Place the 96 well PCR Plate on the magnetic stand at room temperature for 5 minutes.
11. Do not discard the sample in this step. Transfer 50 µL of clear sample to a new well.
12. Add 40 µL of AMPure XP Beads to each sample. Mix thoroughly until homogenized.
13. Incubate sample at room temperature for 5 minutes.
14. Place the 96 well PCR Plate on the magnetic stand at room temperature for 3 minutes or until sample appears clear.
15. Remove and discard clear supernatant.
16. With plate on stand, add 200 µL of 80% ethanol to each magnetic bead pellet and incubate plate at room temperature for 30 seconds. Carefully remove ethanol by pipette.
17. Repeat step 16 for a total of 2 ethanol washes. Ensure all ethanol has been removed.
18. Remove the plate from the magnetic stand and let dry at room temperature for 3 minutes or until bead pellet is visibly dry.
19. Resuspend dried beads with 38 µL Resuspension Buffer. Mix thoroughly until homogenized.

20. Incubate resuspended beads at room temperature for 2 minutes.
21. Place the 96 well PCR Plate on the magnetic stand at room temperature for 3 minutes or until sample appears clear.
22. Transfer 36 µL of clear sample to a new well.
23. Proceed to Step D: PCR Amplification.

STEP D: PCR Amplification

Materials

Bioo Scientific Supplied

GREEN CAP - NEXTflex™ PCR Master Mix, NEXTflex™ Primer Mix
WHITE CAP - NEXTflex™ Resuspension Buffer, Nuclease-free Water

User Supplied

Thermocycler
96 Well PCR Plate
Agencourt AMPure XP Magnetic Beads (room temperature)
80% Ethanol, freshly prepared (room temperature)
Magnetic Stand
36 µL of Adapter Ligated DNA (from STEP C)

1. For each sample, combine the following reagents on ice in the PCR plate. **Mix thoroughly.**

_ µL	Ligated DNA
_ µL	Nuclease-free Water
12 µL	NEXTflex™ PCR Master Mix
2 µL	NEXTflex™ Primer Mix
50 µL	TOTAL

2. Apply adhesive PCR plate seal and place in thermocycler for the following PCR cycles:

2 min	98°C	
30 sec	98°C	
30 sec	65°C	Repeat for a total of 10-15 cycles
60 sec	72°C	
4 min	72°C	

3. Add 40 µL of AMPure XP Beads to each sample. Mix thoroughly until homogenized.
4. Incubate at room temperature for 5 minutes.
5. Place the 96 well PCR Plate on the magnetic stand at room temperature for 3 minutes or until sample is clear.
6. Remove and discard clear supernatant taking care not to disturb beads. Some liquid may remain in wells.
7. With plate on stand, add 200 µL of 80% ethanol to each magnetic bead pellet and incubate plate at room temperature for 30 seconds. Carefully remove ethanol by pipette.
8. Repeat step 7 for a total of 2 ethanol washes. Ensure all ethanol has been removed.
9. Remove plate from magnetic stand and let dry at room temperature for 3 minutes or until bead pellet is visibly dry.

10. Resuspend dried beads with 16 µL Resuspension Buffer. Mix thoroughly until homogenized.
11. Incubate resuspended beads at room temperature for 2 minutes.
12. Place the 96 well PCR Plate on the magnetic stand at room temperature for 5 minutes.
13. Transfer 15 µL of clear sample to a new well.
14. Examine your library by electrophoresis gel or Agilent Bioanalyzer.
15. qPCR is recommended to quantify DNA library templates for optimal cluster density. This can be performed using any qPCR quantification kit with the NEXTflex™ Primer Mix.

A)

B)

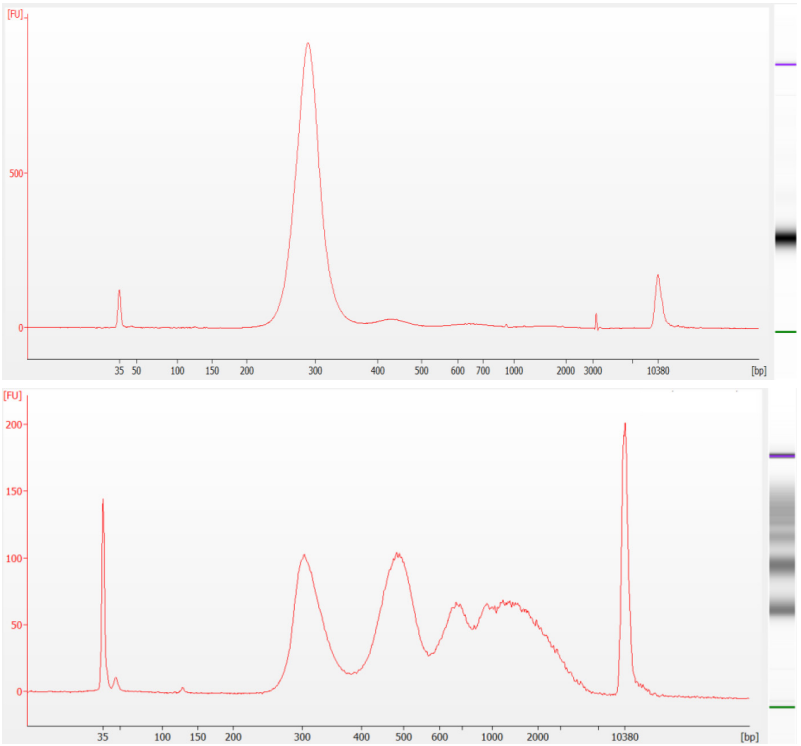


Figure 1: High Sensitivity DNA Electropherogram of Cell Free DNA-Seq Libraries

A) NEXTflex™ Cell Free DNA-Seq library generated from cell free DNA isolated from plasma of pregnant human female. Size selection option was used to enrich for 180bp insert (300bp library). During pregnancy, >90% of DNA fragments <300 bp are comprised of fetal DNA (1) and DNA fragments >300 bp are of maternal origin (2). Peaks in electropherogram represent double stranded libraries with Illumina compatible adapters.

B) NEXTflex™ Cell Free DNA-Seq library product generated from human male shows 300 and 500 bp peaks, characteristic of cell free DNA. DNA ladder-like smaller peaks are characteristic of apoptotic cell death, (3) products of cellular necrosis are represented by higher molecular weight DNA molecules >1 Kb. Both products of apoptotic and necrotic DNA are present in the plasma of cancer patients (4).

References

1) Fan H, et al. Noninvasive diagnosis of fetal aneuploidy by shotgun sequencing DNA from maternal blood. PNAS. 2008; 105(42): 16266–16271.

2) Li Y, Zimmermann B, Rusterholz C, et al. Size separation of circulatory DNA in maternal plasma permits ready detection of fetal DNA polymorphisms. Clin Chem. 2004; 50(6): 1002-1011.

3) Nagata S. Apoptotic DNA fragmentation. Exp. Cell Res. 2000; (256): 12-18.

4) Jahr S et al. DNA Fragments in the Blood Plasma of Cancer Patients: Quantitations and Evidence for Their Origin from Apoptotic and Necrotic Cells. Cancer Res. 2001; (61)1659.

Oligonucleotide Sequences

NEXTflex™	Sequence
DNA Adapter 1	5'AATGATACGGCGACCACCGAGATCTACACTCTTTCCCTACACGACGCTCTTCCGATCT 5'GATCGGAAGAGCACACGTCTGAACTCCAGTCACCGATGTATCTCGTATGCCGTCTTCTGCTTG
Primer 1	5'AATGATACGGCGACCACCGAGATCTACAC
Primer 2	5'CAAGCAGAAGACGGCATACGAGAT

Illumina Compatible RNA NGS Kits and Adapters

Catalog #	Product
5138-01	NEXTflex™ Rapid RNA-Seq Kit (8 reactions)
5138-02	NEXTflex™ Rapid RNA-Seq Kit (48 reactions)
5138-07	NEXTflex™ Rapid Directional RNA-Seq Kit (8 reactions)
5138-08	NEXTflex™ Rapid Directional RNA-Seq Kit (48 reactions)
512911	NEXTflex™ RNA-Seq Barcodes –6
512912	NEXTflex™ RNA-Seq Barcodes – 12
512913	NEXTflex™ RNA-Seq Barcodes – 24
512914	NEXTflex™ RNA-Seq Barcodes – 48
512916	NEXTflex-96™ RNA-Seq Barcodes
5130-01	NEXTflex™ qRNA-Seq™ Kit 4 barcodes (8 reactions)
5130-02	NEXTflex™ qRNA-Seq™ Kit 24 barcodes - Set A (48 reactions)
5130-03	NEXTflex™ qRNA-Seq™ Kit 24 barcodes - Set B (48 reactions)
5130-04	NEXTflex™ qRNA-Seq™ Kit 24 barcodes - Set C (48 reactions)
5130-05	NEXTflex™ qRNA-Seq™ Kit 24 barcodes - Set D (48 reactions)
5130-01D	NEXTflex™ Rapid Directional qRNA-Seq™ Kit 4 barcodes (8 reactions)
5130-02D	NEXTflex™ Rapid Directional qRNA-Seq™ Kit 24 barcodes - Set A (48 reactions)
5130-03D	NEXTflex™ Rapid Directional qRNA-Seq™ Kit 24 barcodes - Set B (48 reactions)
5130-04D	NEXTflex™ Rapid Directional qRNA-Seq™ Kit 24 barcodes - Set C (48 reactions)
5130-05D	NEXTflex™ Rapid Directional qRNA-Seq™ Kit 24 barcodes - Set D (48 reactions)
5132-01	NEXTflex™ Small RNA Sequencing Kit (24 reactions)
5132-02	NEXTflex™ Small RNA Sequencing Kit (48 reactions)
5132-03	NEXTflex™ Small RNA Sequencing Kit v2 (24 reactions)
5132-04	NEXTflex™ Small RNA Sequencing Kit v2 (48 reactions)
513305	NEXTlfex™ Small RNA Barcode Primers -12 (Set A)
513306	NEXTlfex™ Small RNA Barcode Primers -12 (Set B)
513307	NEXTlfex™ Small RNA Barcode Primers -12 (Set C)
513308	NEXTlfex™ Small RNA Barcode Primers -12 (Set D)
512979	NEXTflex™ Poly(A) Beads (8 reactions)
512980	NEXTflex™ Poly(A) Beads (48 reactions)
512981	NEXTflex™ Poly(A) Beads (100 reactions)

Illumina Compatible DNA NGS Kits and Adapters

Catalog #	Product
4201-01	NEXTflex™ 16S V4 Amplicon-Seq Kit – 4
4201-02	NEXTflex™ 16S V4 Amplicon-Seq kit – 12
4201-03	NEXTflex™ 16S V4 Amplicon-Seq kit – 24
4201-04	NEXTflex™ 16S V4 Amplicon-Seq kit – 48
4201-05	NEXTflex™ 16S V4 Amplicon-Seq kit – 96
4201-06	NEXTflex™ 16S V4 Amplicon-Seq kit – 192
4201-07	NEXTflex™ 16S V4 Amplicon-Seq kit – 288
4202-01	NEXTlfex™ 16S V1-V3 Amplicon-Seq Kit - 4
4202-02	NEXTlfex™ 16S V1-V3 Amplicon-Seq Kit - 12
4202-03	NEXTlfex™ 16S V1-V3 Amplicon-Seq Kit - 48
4202-04	NEXTlfex™ 16S V1-V3 Amplicon-Seq Kit - 1-96
4202-05	NEXTlfex™ 16S V1-V3 Amplicon-Seq Kit - 97-192
4202-06	NEXTlfex™ 16S V1-V3 Amplicon-Seq Kit - 193-288
4202-07	NEXTlfex™ 16S V1-V3 Amplicon-Seq Kit - 289-384
5140-01	NEXTflex™ DNA Sequencing Kit (8 reactions)
5140-02	NEXTflex™ DNA Sequencing Kit (48 reactions)
5144-01	NEXTflex™ Rapid DNA-Seq Kit (8 reactions)
5144-02	NEXTflex™ Rapid DNA-Seq Kit (48 reactions)
5150-01	NEXTflex™ Cell Free DNA-Seq Kit (8 reactions)
5150-02	NEXTflex™ Cell Free DNA-Seq Kit (48 reactions)
514101	NEXTflex™ DNA Barcodes – 6
514102	NEXTflex™ DNA Barcodes – 12
514103	NEXTflex™ DNA Barcodes – 24
514104	NEXTflex™ DNA Barcodes – 48
514105	NEXTflex-96™ DNA Barcodes (Plate Format)
514106	NEXTflex-96™ DNA Barcodes (Tube Format)
514160	NEXTflex™ Dual-Indexed DNA Barcodes (1-96)
514161	NEXTflex™ Dual-Indexed DNA Barcodes (97-192)
5119-01	NEXTflex™ Bisulfite-Seq kit (8 reactions)
5119-02	NEXTflex™ Bisulfite-Seq kit (48 reactions)
511911	NEXTflex™ Bisulfite-Seq Barcodes – 6
511912	NEXTflex™ Bisulfite-Seq Barcodes – 12
511913	NEXTflex™ Bisulfite-Seq Barcodes - 24
5118-01	NEXTflex™ Methyl-Seq 1 Kit (8 reactions)
5118-02	NEXTflex™ Methyl-Seq 1 Kit (48 reactions)

511921	NEXTflex™ Msp 1 (8 reactions)
511922	NEXTflex™ Msp 1 (48 reactions)

5143-01	NEXTflex™ ChIP-Seq Kit (8 reactions)
5143-02	NEXTflex™ ChIP-Seq Kit (48 reactions)
514120	NEXTflex™ ChIP-Seq Barcodes – 6
514121	NEXTflex™ ChIP-Seq Barcodes – 12
514122	NEXTflex™ ChIP-Seq Barcodes – 24
514123	NEXTflex™ ChIP-Seq Barcodes – 48
514124	NEXTflex-96™ ChIP-Seq Barcodes

5140-51	NEXTflex™ Pre-Capture Combo Kit (6 barcodes)
5140-52	NEXTflex™ Pre-Capture Combo Kit (12 barcodes)
5140-53	NEXTflex™ Pre-Capture Combo Kit (24 barcodes)
5140-56	NEXTflex™ Pre-Capture Combo Kit (48 barcodes)
5140-54	NEXTflex™ Pre-Capture Combo Kit (96 barcodes)
514131	NEXTflex™ DNA Barcode Blockers - 6 for SeqCap
514132	NEXTflex™ DNA Barcode Blockers - 12 for SeqCap
514133	NEXTflex™ DNA Barcode Blockers - 24 for SeqCap
514136	NEXTflex™ DNA Barcode Blockers - 48 for SeqCap
514134	NEXTflex™ DNA Barcode Blockers - 96 for SeqCap

5142-01	NEXTflex™ PCR-Free DNA Sequencing Kit (8 reactions)
5142-02	NEXTflex™ PCR-Free DNA Sequencing Kit (48 reactions)
514110	NEXTflex™ PCR-Free Barcodes – 6
514111	NEXTflex™ PCR-Free Barcodes – 12
514112	NEXTflex™ PCR-Free Barcodes – 24
514113	NEXTflex™ PCR-Free Barcodes – 48

DNA Fragmentation

Catalog #	Product
5135-01	AIR™ DNA Fragmentation Kit (10 reactions)
5135-02	AIR™ DNA Fragmentation Kit (40 reactions)

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Your feedback is important to us. Tell us what you think of our kits by scanning the QR code or visiting our website at www.biooscientific.com/NGSfeedback. We can't wait to hear from you!

Bioo Scientific®

THE NGS EXPERTS™

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