



# **NEXTFLEX® ChIP-Seq Kit**

(For Illumina® Platforms)
Catalog #NOVA-5143-01 (Kit contains 8 reactions)



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# NEXTflex™ ChIP-Seq Kit - 5143-01

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#### GENERAL INFORMATION

#### **Product Overview**

The NEXTflex™ ChIP-Seq Kit is designed to prepare single, paired-end and multiplexed DNA libraries from ChIP DNA, genomic DNA or cDNA for sequencing using Illumina® platforms. The enhanced NEXTflex ChIP-Seq Kit simplifies workflow by using master mixed reagents and magnetic bead based cleanup, reducing pipetting and eliminating time consuming steps in library preparation. In addition, the availability of up to 96 unique adapter barcodes and gel-free size selection allows for high-throughput, multiplexed sequencing.

The NEXTflex ChIP-Seq Kit contains specially designed NanoQ™ enzymes and buffers needed for sample preparation of chromatin immunoprecipitated or regular genomic DNA for next generation sequencing. ChIP-Seq is a useful technique that allows the user to examine DNA sequences bound by proteins in cells. After using an antibody to enrich for a specific DNA-protein complex, the user may use the ChIP-Seq kit to prepare a library ready for sequencing.

### Contents, Storage and Shelf Life

The NEXTflex™ ChIP-Seq Kit contains enough material to prepare 8 ChIP samples for Illumina® compatible sequencing. The shelf life of all reagents is 12 months when stored properly. All components can be safely stored at -20°C.

Kit Contents	Amount
CLEAR CAP	
NEXTflex <sup>™</sup> ChIP End Repair Buffer Mix	56 μL
NEXTflex <sup>™</sup> ChIP End Repair Enzyme Mix	24 μL
RED CAP	
NEXTflex <sup>™</sup> ChIP Adenylation Mix	36 μL
PURPLE CAP	
NEXTflex <sup>™</sup> ChIP Ligation Mix	220 μL
NEXTflex™ ChIP Adapter (0.6 μM)	16 μL
GREEN CAP	
NEXTflex $^{\text{\tiny ML}}$ ChIP Primer Mix (12.5 $\mu$ M)	16 μL
NEXTflex <sup>™</sup> ChIP PCR Master Mix	96 μL
WHITE CAP	
Nuclease-free Water	1.5 mL
Resuspension Buffer	(2) 1 mL
BLUE CAP	

5 μL

NEXTflex™ ChIP-Seq DNA Control

### **Required Materials not Provided**

- 1 ng 10 ng of fragmented DNA in up to 40 μL nuclease-free water
- (Optional) NEXTflex<sup>™</sup> ChIP-Seq Barcodes 6 / 12 / 24 / 48 or NEXTflex-96<sup>™</sup> ChIP-Seq Barcodes (Cat # 514120, 514121, 514122, 514123, 514124)
- Ethanol 100% (room temperature)
- Ethanol 80% (room temperature)
- · Covaris System (S2, E210) or other device for fragmenting DNA
- 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 5 mL (Beckman Coulter Genomics, Cat # A63880)
- Magnetic Stand -96 (Ambion, Cat # AM10027) or similar
- Heat block
- Thermocycler
- 2, 10, 20, 200 and 1000 μL pipettes / multichannel pipettes
- Nuclease-free barrier pipette tips
- · Microcentrifuge
- 1.5 mL nuclease-free microcentrifuge tubes
- Vortex

### **Revision History**

Version	Date	Description
v12.10	November 2012	Previous version.
v15.07	July 2015	All previous protocol options containing gel size-selection or column purification have been removed. Users now have the choice of a bead-based size-selection or not performing size-selection.

Customers who would like to follow the previous protocol utilizing column-based and gel-based purification should contact <a href="mailto:BiooNGS@BiooScientific.com">BiooNGS@BiooScientific.com</a> for assistance.

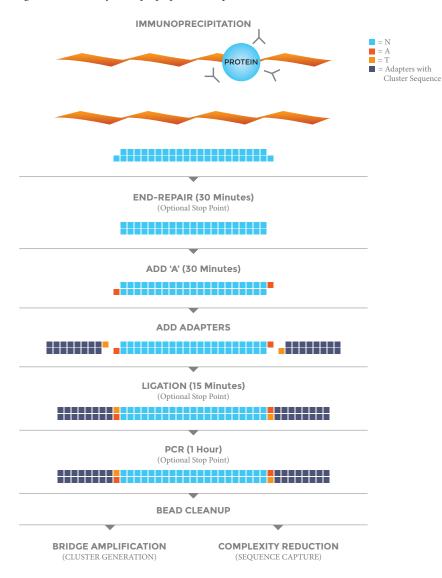
### **Warnings and Precautions**

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 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 precipitate is in solution.
- Ensure pipettes are properly calibrated as library preparations are highly sensitive to pipetting error.
- Do not heat the NEXTflex™ ChIP-Seq Barcode Adapters above room temperature.
- This kit contains a single barcoded NEXTflex™ ChIP Adapter. To enable multiplexing, please use the appropriate combination of NEXTflex™ ChIP-Seq Barcodes during the Adapter Ligation step (sold separately).
- Try to maintain a laboratory temperature of 20°-25°C (68°-77°F).
- DNA sample quality may vary between preparations. It is the user's responsibility to utilize high quality DNA. DNA that is heavily nicked or damaged may cause library preparation failure. Absorbance measurements at 260 nm are commonly used to quantify DNA and 260 nm / 280 nm ratios of 1.8 2.0 usually indicate relatively pure DNA. Other quantification methods using fluorescent dyes may also be used. The user should be aware that contaminating RNA, nucleotides and single-stranded DNA may affect the amount of usable DNA in a sample preparation.
- If starting with a DNA input amount greater or less than the range specified in the manual, adjust the NEXTflex™ ChIP DNA Adapter volume to preserve the insert to adapter ratio.
- It is critical that the user know the size of the starting fragmented ChIP or genomic DNA
  material. This is important to ensure that starting material falls within the range of the
  optional size-selection step, STEP B1: Size-Selection Cleanup.
- It is highly recommended that NEXTflex<sup>™</sup> Primer Mix be used during PCR amplification.
   Inadvertent use of an incorrect primer sequence can potentially result in elimination of the index.

### NEXTflex™ ChIP-Seq Sample Preparation Flow Chart

Figure 1: Flow chart for sample preparation steps:



### Starting Material

The NEXTflex™ ChIP-Seq Kit has been optimized and validated using 1 ng - 10 ng of qPCR verified ChIP DNA, genomic DNA and cDNA.

The NEXTflex™ ChIP-Seq Kit is a completely gel-free protocol that utilizes a magnetic bead based cleanup to size select DNA insert fragments between 180 – 280 bp. Alternately, users can choose to skip size-selection by following STEP B2: No Size-Selection Cleanup. STEP B2 is recommended if users have confirmed that their fragmented starting material falls within an acceptable range for their application.

If starting with poor quality or a very low amount of DNA (≤ 1 ng), STEP B2: No Size-Selection Cleanup) should be performed following End Repair.

If the user is performing the procedure for the first time, we recommend using the NEXTflex ChIP-Seq DNA Control included in the kit. When running a positive control reaction, the user should add 1  $\mu$ L of the NEXTflex ChIP-Seq DNA Control (20 ng/ $\mu$ L) in STEP A instead of the ChIP-Seq DNA sample.

### **Reagent Preparation**

- Briefly spin down each component to ensure material has not lodged in the cap or side of tube. Keep on ice and vortex each tube prior to use.
- 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 precipitate is in solution.
- 3. Allow Agencourt AMPure XP Beads to come to room temperature and vortex the beads until liquid appears homogenous before every use.

#### **NEW PROTOCOL**

# Please see Revision History on page 3 before starting procedure.

### STEP A: End Repair

#### Materials

#### **Bioo Scientific Supplied**

CLEAR CAP - NEXTflex™ ChIP End Repair Buffer Mix, NEXTflex™ ChIP End Repair Enzyme Mix

WHITE CAP - Nuclease-free Water

#### **User Supplied**

ChIP or genomic DNA in 40  $\mu$ L (or less) nuclease-free water 96 well PCR Plate Thermocycler Adhesive PCR Plate Seal Ice

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

$_{-}\mu L$	Nuclease-free Water
$_{-}\mu L$	ChIP or genomic DNA (1 ng - 10 ng)
7 μL	NEXTflex™ ChIP End Repair Buffer Mix
$3\;\mu L$	NEXTflex™ ChIP End Repair Enzyme Mix
50 uL	TOTAL

- 50 μL TOTAL
- 2. Mix thoroughly by pipetting.
- 3. Apply adhesive PCR plate seal and incubate on a thermocycler for 30 minutes at 22°C.
- 4. Proceed to either STEP B1: Size-Selection Cleanup or STEP B2: No Size-Selection Cleanup.

### STEP B1: Size-Selection Cleanup

STEP B1 is designed to select for DNA insert fragments between 180-280 bp. Users who do not wish to size-select their libraries should proceed to STEP B2: No Size-Selection 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 50  $\mu$ L of End Repaired DNA (from STEP A)

- 1. Add 39  $\mu$ L of AMPure XP Beads to each sample and mix thoroughly by pipetting.
- 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 the supernatant appears clear.
- 4. Do not discard clear sample in this step. Transfer 87  $\mu$ L of clear sample to a new well. Be careful not to disrupt the magnetic bead pellet or transfer any magnetic beads with the sample.
- 5. Add 18 μL of AMPure XP Beads to each clear sample and mix thoroughly by pipetting.
- 6. Incubate sample at room temperature for 5 minutes.
- 7. Place the 96 well PCR Plate on the magnetic stand at room temperature for 5 minutes or until the supernatant appears clear.
- 8. Remove and discard clear supernatant taking care not to disturb beads. Some liquid may remain in wells.
- 9. With plate on stand, add 200  $\mu$ L of freshly prepared 80% ethanol to each magnetic bead pellet and incubate plate at room temperature for 30 seconds. Carefully, remove ethanol by pipette.
- 10. Repeat previous step, 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.
- 12. Resuspend dried beads with 17 µL of Resuspension Buffer. Mix thoroughly by pipetting. Ensure beads are no longer attached to the side of the well.
- 13. Incubate resuspended beads at room temperature for 2 minutes.
- 14. Place plate on magnetic stand for 5 minutes or until the sample appears clear.
- 15. Gently transfer 16 μL of clear sample to new well.
- 16. If you wish to pause your experiment, the procedure may be safely stopped at this step and samples stored at -20°C. To restart, always thaw your frozen samples on ice before proceeding to STEP C: 3' Adenylation.



### STEP B2: No Size-Selection Cleanup

STEP B2 is designed for users who do not wish to size-select their libraries. Cleanup steps throughout are designed to eliminate only unwanted low-molecular weight material. If you wish to size-select your libraries, please follow STEP B1: Size-Selection 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 50 μL of End Repaired DNA (from STEP A)

- 1. Add 90 μL of AMPure XP Beads to each sample and mix well by pipetting.
- 2. Incubate 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 the sample appears clear.
- 4. Gently remove and discard clear sample taking care not to disturb beads. Some liquid may remain in wells.
- With plate on stand, add 200 µL of freshly prepared 80% ethanol to each magnetic bead pellet and incubate plate at room temperature for 30 seconds. Carefully, remove ethanol by pipette.
- 6. Repeat previous step, for a total of 2 ethanol washes. Ensure all ethanol has been removed.
- Remove the plate from the magnetic stand and let dry at room temperature for 3 minutes.
- 8. Resuspend dried beads with 17  $\mu$ L Resuspension Buffer. Mix well by pipetting. Ensure beads are no longer attached to the side of the well.
- 9. Incubate resuspended beads at room temperature for 2 minutes.
- Place plate on magnetic stand at room temperature for 5 minutes or until the sample appears clear.
- 11. Transfer 16 μL of clear sample to new well.
- 12. If you wish to pause your experiment, the procedure may be safely stopped at this step and samples stored at -20°C. To restart, thaw frozen samples on ice before proceeding to STEP C: 3' Adenylation.

### STEP C: 3' Adenylation

#### **Materials**

#### **Bioo Scientific Supplied**

RED CAP - NEXTflex™ ChIP Adenylation Mix

#### **User Supplied**

Thermocycler

16 μL of End Repaired DNA (from STEP B)

1. Combine the following in the 96 well PCR Plate:

$16~\mu L$	End-Repaired DNA (from Step B)
$4.5~\mu\mathrm{L}$	NEXTflex <sup>™</sup> ChIP Adenylation Mix
20.5 μL	TOTAL

- 2. Mix thoroughly by pipetting.
- 3. Apply adhesive PCR plate seal and incubate on a thermocycler for 30 minutes at 37°C.
- 4. Proceed to STEP D: Adapter Ligation.

### **STEP D: Adapter Ligation**

#### **Materials**

#### **Bioo Scientific Supplied**

PURPLE CAP - NEXTflex™ ChIP Ligation Mix (remove right before use and store immediately after use at -20°C), NEXTflex™ ChIP Adapter / or / NEXTflex™ ChIP-Seq Barcodes – 6 / 12 / 24 / 48 / 96 (Cat # 514120, 514121, 514122, 514123, 514124)

#### **User Supplied**

20.5 µL 3' Adenylated DNA (from STEP C)

 For each sample, combine the following reagents (in this order) in the 96-well PCR Plate:

```
20.5 μL 3' Adenylated DNA (from Step C)

27.5 μL NEXTflex™ ChIP Ligation Mix

2.0 μL NEXTflex™ ChIP Adapter

50 μL TOTAL
```

- 2. Mix thoroughly by pipetting.
- 3. Apply adhesive PCR plate seal and incubate on a thermocycler for 15 minutes at 22°C.
- Proceed to STEP E: Cleanup.



### STEP E: 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 50  $\mu$ L of Adapter Ligated DNA (from STEP D)

- 1. Add 40 μL of AMPure XP Beads to each sample and mix thoroughly by pipetting.
- 2. Incubate 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 the supernatant appears clear.
- 4. Remove clear supernatant taking care not to disturb beads. Some liquid may remain in wells.
- 5. With plate on stand, add  $200 \,\mu\text{L}$  of freshly prepared 80% ethanol to each sample and incubate plate at room temperature for 30 seconds. Carefully, remove ethanol by pipette.
- 6. Repeat previous step, for a total of 2 ethanol washes and ensure all ethanol has been removed.
- 7. Remove the plate from the magnetic stand and let dry at room temperature for 3 minutes. Note: Do not over dry the beads.
- Resuspend dried beads with 52 µL of Resuspension Buffer. Mix thoroughly by pipetting, ensuring beads are no longer attached to the side of the well.
- 9. Incubate resuspended beads at room temperature for 2 minutes.
- 10. Place plate on magnetic stand for 5 minutes or until the sample appears clear.
- 11. Gently transfer 50  $\mu$ L of clear sample to new well.
- 12. Remove the plate from the magnetic stand and add 40 μL of AMPure XP Beads to each sample and mix thoroughly by pipetting.
- 13. Incubate at room temperature for 5 minutes.
- 14. Place the 96 well PCR Plate on the magnetic stand at room temperature for 5 minutes or until the supernatant appears clear.
- Remove clear supernatant taking care not to disturb beads. Some liquid may remain in wells.
- 16. With plate on stand, add 200  $\mu$ L of freshly prepared 80% ethanol to each sample and incubate plate at room temperature for 30 seconds. Carefully, remove ethanol by pipette.

- 17. Repeat previous step, for a total of 2 ethanol washes and ensure all ethanol has been removed.
- 18. Remove the plate from the magnetic stand and let dry at room temperature for 3 minutes. Note: Do not over dry the beads.
- 19. Resuspend dried beads with 38  $\mu$ L of Resuspension Buffer. Gently, pipette entire volume up and down 10 times mixing thoroughly. Ensure beads are no longer attached to the side of the well.
- 20. Incubate resuspended beads at room temperature for 2 minutes.
- 21. Place plate on magnetic stand for 5 minutes or until the sample appears clear.
- 22. Gently transfer 36 μL of clear sample to new well.
- 23. If you wish to pause your experiment, the procedure may be safely stopped at this step and samples stored at -20°C. To restart, always thaw your frozen samples on ice before proceeding to STEP F: PCR Amplification.

### **STEP F: PCR Amplification**

#### **Materials**

#### **Bioo Scientific Supplied**

GREEN CAP - NEXTflex™ ChIP Primer Mix, NEXTflex™ ChIP PCR Master Mix WHITE CAP - Resuspension Buffer

#### **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 Purified Adapter Ligated DNA (from STEP E)

1. For each sample, combine the following reagents on ice in the 96 well PCR plate:

```
    36 μL Purified Adapter Ligated DNA (from Step E)
    12 μL NEXTflex™ ChIP PCR Master Mix
    2 μL NEXTflex™ ChIP Primer Mix
    50 μL TOTAL
```

- 2. Mix thoroughly by pipetting.
- 3. PCR Cycles:

2 min	98°C	
30 sec	98°C	
30 sec	65°C	*Repeat 11-15 cycles
60 sec	72°C	
4 min	72°C	

<sup>\*</sup>PCR cycles will vary depending on the amount of starting material and quality of your sample. Further optimization may be necessary. Always use the least number of cycles possible.

- 4. Add 44 μL of AMPure XP Beads to each sample and mix thoroughly by pipetting.
- 5. Incubate at room temperature for 5 minutes.
- 6. Place the 96 well PCR Plate on the magnetic stand at room temperature for 5 minutes or until the supernatant appears clear.
- Remove clear supernatant taking care not to disturb beads. Some liquid may remain in wells.
- 8. With plate on stand, add 200  $\mu L$  of freshly prepared 80% ethanol to each sample and

- incubate plate at room temperature for 30 seconds. Carefully, remove ethanol by pipette.
- Repeat previous step, for a total of 2 ethanol washes and ensure all ethanol has been removed.
- 10. Remove the plate from the magnetic stand and let dry at room temperature for 3 minutes. Note: Do not over dry the beads.
- Resuspend dried beads with 52 µL of Resuspension Buffer. Mix thoroughly by pipetting, ensuring beads are no longer attached to the side of the well.
- 12. Incubate resuspended beads at room temperature for 2 minutes.
- 13. Place plate on magnetic stand for 5 minutes or until the sample appears clear.
- 14. Gently transfer 50 µL of clear sample to new well.
- 15. Remove the plate from the magnetic stand and add  $44 \mu L$  of AMPure XP Beads to each sample and mix thoroughly by pipetting.
- 16. Incubate at room temperature for 5 minutes.
- 17. Place the 96 well PCR Plate on the magnetic stand at room temperature for 5 minutes or until the supernatant appears clear.
- 18. Remove clear supernatant taking care not to disturb beads. Some liquid may remain in wells.
- 19. With plate on stand, add 200  $\mu$ L of freshly prepared 80% ethanol to each sample and incubate plate at room temperature for 30 seconds. Carefully, remove ethanol by pipette.
- 20. Repeat previous step, for a total of 2 ethanol washes and ensure all ethanol has been removed.
- 21. Remove the plate from the magnetic stand and let dry at room temperature for 3 minutes. Note: Do not over dry the beads.
- Resuspend dried beads with 16 µL of Resuspension Buffer. Mix thoroughly by pipetting.
   Ensure beads are no longer attached to the side of the well.
- 23. Incubate resuspended beads at room temperature for 2 minutes.
- 24. Place plate on magnetic stand for 5 minutes or until the sample appears clear.
- 25. Gently transfer 15 μL of clear sample to a well of a new 96 well PCR Plate.
- 26. With an input amount of 1 ng -10 ng, your product yield may be too low to quantify by gel. We recommend quantifying your library with a fluorometer and checking the size using an Agilent Bioanalyzer. If on the Bioanalyzer trace there are two bands, one of expected size and one higher molecular weight band, a portion of your adapter ligated inserts have annealed to each other. This occurs due to the long adapter length and is



more prevalent when there are too many PCR cycles. This type of double band will not affect your sequencing results as the double stranded product will be denatured prior to cluster generation. As an extra verification step, a portion of your product can be denatured manually by heating the sample to 95°C for 5 minutes and then placing it on ice. The denatured product should appear as a single band on a Bioanalyzer RNA Pico 6000 Chip Kit.

- 27. qPCR is recommended to quantitate DNA library templates for optimal cluster density. This can be performed using any qPCR quantification kit with the NEXTflex™ ChIP Primer Mix.
- 28. The library is now ready for cluster generation per the standard Illumina protocol. Proceed to cluster generation or seal with Adhesive PCR Plate Seal and store at -20°C.

#### LIBRARY VALIDATION

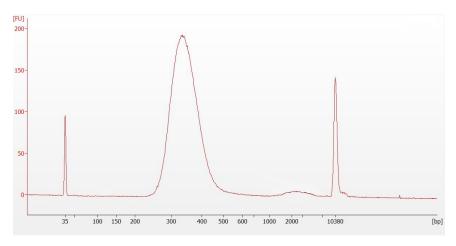


Figure 2: Example of DNA Library Size Distribution. 1 ng of input DNA was used in the End-Repair step. Size Selection was performed to select for insert sizes of 180-280 bp and a total of 15 cycles of PCR were performed. 1 µL of the resulting library was run on an Agilent High Sensitivity DNA chip to verify size. Using a Qubit\* 2.0 Fluorometer & Qubit\* dsDNA HS Assay Kit, the concentration of the library was determined to be > 10 nM.

### **APPENDIX A**

# Oligonucleotide Sequences

NEXTflex™	Sequence (5' → 3')
ChIP Adapter	5'AATGATACGGCGACCACCGAGATCTACACTCTTTCCCTACACGACGCTCTTCCGATCT 5'GATCGGAAGAGCACACGTCTGAACTCCAGTCAC <u>CGATGT</u> ATCTCGTATGCCGTCTTCTGCTTG
ChIP Primer 1	5'AATGATACGGCGACCACCGAGATCTACAC
ChIP Primer 2	5'CAAGCAGAAGACGGCATACGAGAT

### **RELATED PRODUCTS**

# 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 v2 - 4 barcodes (8 reactions)
5130-02	NEXTflex" qRNA-Seq" Kit v2 - 24 barcodes - Set A (48 reactions)
5130-03	NEXTflex" qRNA-Seq" Kit v2 - 24 barcodes - Set B (48 reactions)
5130-04	NEXTflex" qRNA-Seq" Kit v2 - 24 barcodes - Set C (48 reactions)
5130-05	NEXTflex" qRNA-Seq" Kit v2 - 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	NEXTIfex™ Small RNA Barcode Primers -12 (Set A)
513306	NEXTIfex™ Small RNA Barcode Primers -12 (Set B)
513307	NEXTIfex™ Small RNA Barcode Primers -12 (Set C)
513308	NEXTIfex™ Small RNA Barcode Primers -12 (Set D)
512979	NEXTflex <sup>™</sup> 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* 168 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 <sup>™</sup> 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 <sup>™</sup> Pre-Capture Combo Kit (12 barcodes)
5140-53	NEXTflex <sup>™</sup> Pre-Capture Combo Kit (24 barcodes)
5140-56	NEXTflex" Pre-Capture Combo Kit (48 barcodes)
5140-54	NEXTflex <sup>™</sup> 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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