

USER GUIDE

HOLOGIC®
Diagenode

Premium Methyl UDI-UMI Adapters

Premium Methyl UDI-UMI Adapters Module - Set A

Cat. No. C02030040

Premium Methyl UDI-UMI Adapters Module - Set B

Cat. No. C02030041

Premium Methyl UDI-UMI Adapters Module – 96 rxns

Cat. No. C02030042

Content

Introduction	4
Usage recommendations.....	5
Premium RRBS V2 Construct.....	6
Sequencing Recommendations	7
Data Analysis Recommendations	8
Premium Methyl UDI-UMI Adapters Sequences.....	12
FAQ.....	17
Related products.....	18



Please read this manual carefully
before starting your experiment

Introduction

Premium Methyl UDI-UMI Adapters Modules include methylated full-length adapters with **unique dual indexes** (UDI) and **unique molecular identifiers** (UMI) for library multiplexing.

- The use of **Unique Dual Indexes** (unique i5 and i7 barcodes) is highly recommended to mitigate errors introduced by read misassignment, including index hopping frequently observed with patterned flow cells such as Illumina's NovaSeq platform.
- The use of **Unique Molecular Identifiers** (9-base sequences) is highly recommended to identify and remove PCR duplicates from your data, ensuring accurate methylation measurements. This feature is especially appreciable in case of low DNA amount analysis.

Our Methyl UDI-UMI adapters have been designed and validated for the Premium RRBS kit V2 (Cat. No. C02030036, C02030037), and are compatible with other bisulfite-based (BS-seq) or enzymatic-based (EM-seq) applications.

Please choose the format that matches your multiplexing needs among the compatible references:

Table 1. Premium Methyl UDI-UMI Adapters Modules

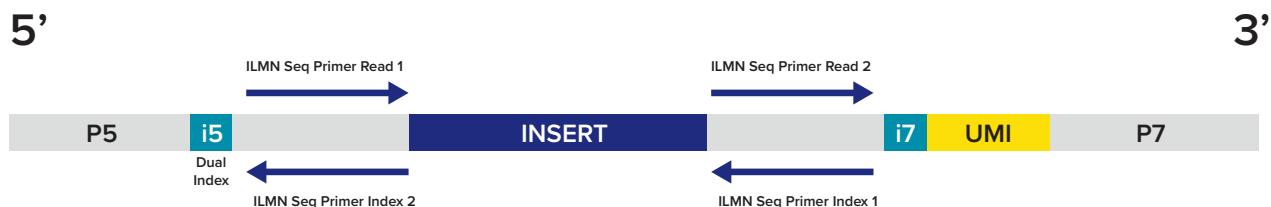
Premium Methyl UDI-UMI Adapters	Format	Cat. No.
Module – Set A	24 UDI-UMI, 24 rxns	C02030040
Module – Set B	24 UDI-UMI, 24 rxns	C02030041
Module – 96 rxns	96 UDI-UMI, 96 rxns	C02030042

Usage recommendations

All Premium Methyl UDI-UMI Adapters are supplied at 100 nM in a volume sufficient for one RRBS reaction (5 µl). Please note that depending on your application, dilution of the adapters may be necessary. Refer to the protocol for specific instructions.

Premium RRBS V2 Construct

Figure 1. Premium RRBS V2 library construct scheme. The final construct bears unique dual indexes (UDI) and a unique molecular identifier (UMI).



P5/P7 = Illumina adapters

UMI = Unique Molecular Identifier

i5/i7 = Unique Dual Indexes

Sequencing Recommendations

The library construct contains the UMI sequence immediately following the i7 index.

Consequently, a sequencing run mode consisting of n-8-17-n cycles must be carried out:

- n is the chosen read length
- 8 correspond to the i5 index sequence
- 17 correspond to the i7+UMI sequence

Data Analysis Recommendations

Samples demultiplexing

To preserve the UMI information, a 2-step process is required for the demultiplexing of the sequencing data:

1. Use bcl2fastq to generate Undetermined fastq files from bcl files, keeping the UDI and UMI information.
2. Use fumi_tools to demultiplex the Undetermined fastq based on UDI sequences and append the UMI information at the end of the read names.

1. bcl2fastq

Following the completion of the sequencing run, the bcl2fastq converter tool must be run with default settings. An example is shown below:

```
bcl2fastq --runfolder-dir ./ --sample-sheet <bcl2fastq_sample_sheet.csv> --output-dir  
./Data/ \  
--ignore-missing-bcls \  
--ignore-missing-filter \  
--ignore-missing-positions \  
--ignore-missing-controls \  
--auto-set-to-zero-barcode-mismatches \  
--find-adapters-with-sliding-window \  
--adapter-stringency 0.9 \  
--loading-threads 4 \  
--processing-threads 10 \  
--writing-threads 4 \  
--mask-short-adapter-reads 22 \  
--minimum-trimmed-read-length 35
```

The sample sheet must not specify any UDI sequences at this step and should be formatted as follow:

```
[Header]
IEMFileVersion,5
Date,xx-xx-xx
WorkFlow,Generate FASTQ
Application, FASTQ Only
Instrument Type,NovaSeq6000
Assay,
Index Adapters,
Description,
Chemistry,
[Reads]
50
50
[Settings]
Adapter,
AdapterRead2,
[Data]
Sample_ID,Sample_Name,Sample_Plate,Sample_Well,Index_Plate_Well,I7_
Index_ID,index,I5_Index_ID,index2,Sample_Project
All_samples,,,,,NNNNNNNNNNNNNNNN,,NNNNNNNN, Project
```

Notes:

1. The sequence of the i7 index (7th field in the last line of the sample sheet displayed above and named index) **must** be 17 consecutive Ns, corresponding to the length of the i7 index (8 nucleotides long) and the UMI (9 nucleotides long).
2. The sequence of the i5 index (9th field in the last line of the sample sheet displayed above and named index2) **must** be 8 consecutive Ns, corresponding to *the length of the i5 index (8 nucleotides long)*.
3. Individual lanes can be selected using the --tiles s_x parameter in the bcl2fastq command line, where x represents the lane to be processed. Alternatively, the lane can be specified in the sample sheet as below:

```
Lane,Sample_ID,Sample_Name,Sample_Plate,Sample_Well,Index_Plate_Well,I7_Index_ID,index,I5_Index_ID,index2,Sample_Project
x,All_samples,,,,,NNNNNNNNNNNNNNNN,,NNNNNNNN,Project
```

Using this sample sheet, the bcl2fastq command generates one pair of Undetermined fastq files per processed lane (Undetermined_S0_L00x_ R1_001.fastq.gz

and Undetermined_S0_L00x_R2_001.fastq.gz), which are formatted with the <index7><UMI>+<index5> appended at the end of the read names as shown below for one read:

```
@A00801:392:HGNKCDRX3:1:1101:1036:1000 1:N:0:CTCTCGTCCCTATCCAC+AGGTTATA
NAAAAAACCTTCCCAACATAAACTTAATAACTTAAACAACACCCATC
#F, : FFFFFFFFFFFFFF : FFFFFFFFFFFFFFFF : FFFFFFFFFF
```

2. **fumi_tools**

The Undetermined fastq files can subsequently be used with the demultiplex function of **fumi_tools** using the following command (more information can be found on the gitlab page of the tool: https://ccb-gitlab.cs.uni-saarland.de/tobias/fumi_tools):

```
fumi_tools demultiplex -i Undetermined_S0_L00x_R1_001.fastq.gz \
--input-read2 Undetermined_S0_L00x_R2_001.fastq.gz \
-s <demultiplex_sample_sheet.csv> \
-o ./pathToDemultiplexLibrary /%s_S%i_L%l_R%r_001.fastq.gz \
--threads 6 --format-umi --tag-umi
```

The sample sheet used with **fumi_tools** must list the samples and the corresponding sequences of the UDI used (See **Premium Methyl UDI-UMI Adapters Sequences** section below for the UDI sequences):

Sample_ID	Sample_Name	Lane	index	index2
1	Sample1	1	CTCTCGTC	AGGTTATA
2	Sample2	1	GGCTTAAG	GGTCACGA
3	Sample3	2	GGCTTAAG	GGTCACGA
4	Sample4	2	AATCCGGA	AACTGTAG

Where the index column corresponds to the sequence of the i7 index and the index2 column corresponds to the sequence or the reverse complement of the i5 index depending on the type of sequencer used (see Tables 2, 3 and 4). The lane onto which the sample was loaded can be specified.

The `fumi_tools` demultiplex generates one pair of fastq files per sample, assigned based on the UDI sequences, in which the UMI sequences have been extracted and appended at the end of the read names as follow:

<index7><UMI>+<index5>:FUMII<UMI>|

```
@A00801 : 392 : HGNKCDRX3 : 1 : 1101 : 1036 :1000 1 : N : 0: CTCTCGTCCCTATCCAC+
AGGTTATA : FUMI | CCTATCCAC |
NAAAAAACCTTCCCAACATAAAACTTAATAACTTAAACAACACCCATC
NAAAAAACCTTCCCAACATAAAACTTAATAATAACTTAAACAACACCCATC
+
#F , : FFFFFFFFFFFFFF : FFFFFFFFFFFFFFFF : FFFFFFFFFF
```

Post-demultiplexing processing

After demultiplexing the samples and extracting the UMIs, the reads must be trimmed and aligned to the reference genome. Using the Premium Methyl UDI-UMI Adapter will result in directional libraries, but the sequencing adapters are applied in reverse order. As a result, unlike standard library construction, Read1 will correspond to the i5 index (index2) and Read2 to the i7 index, similar to libraries prepared using post-bisulfite adapter tagging (PBAT). From a bioinformatics perspective, this means that the trimmed read pairs should be aligned using Bismark with the `--pbat` option to account for the inversion of the libraries. This characteristic also specifically affects the trimming parameters of RRBS libraries. For more details, refer to the Data Analysis section of the RRBS-KIT-V2 Manual.

UMI-based deduplication

UMI-based deduplication of the coordinates-sorted and indexed alignment bam files can be carried out using the ‘dedup’ function available in either `fumi_tools` or `UMI tools` (<https://umi-tools.readthedocs.io/en/latest/reference/dedup.html>)

- `fumi_tools`:

```
fumi_tools dedup -i sorted.bam \
-o dedup.bam \
--threads 6 \
--memory 3G \
--paired
```

- `UMI tools`:

```
umi_tools dedup -l sorted.bam \
-S dedup.bam \
--paired
```

Note: the `--tag-umi` option of the ‘`fumi_tools demultiplex`’ command must not be set if the deduplication is being conducted using `UMI tools`.

Premium Methyl UDI-UMI Adapters Sequences

Table 2. Premium Methyl UDI-UMI Adapters Sequences— Set A (1-24)

Premium Methyl UDI-UMI Adapter #	i5 index NovaSeq 6000 v1.0, MiSeq, HiSeq 2000/2500 systems	i5 index NovaSeq 6000 v1.5, iSeq, MiniSeq, NextSeq, HiSeq 3000/4000 systems	i7 index* (all Illumina systems)
1	AGCGCTAG	CTAGCGCT	CCGCGGTTNNNNNNNNNN
2	GATATCGA	TCGATATC	TTATAACCNNNNNNNNNN
3	CGCAGACG	CGTCTGCG	GGACTTGGNNNNNNNNNN
4	TATGAGTA	TACTCATA	AAGTCCAANNNNNNNNNN
5	AGGTGCGT	ACGCACCT	ATCCACTGNNNNNNNNNN
6	GAACATAC	GTATGTTC	GCTTGTCA>NNNNNNNNNN
7	ACATAGCG	CGCTATGT	CAAGCTAGNNNNNNNNNN
8	GTGCGATA	TATCGCAC	TGGATCGANNNNNNNNNN
9	CCAACAGA	TCTGTTGG	AGTCAGGNNNNNNNNNN
10	TTGGTGAG	CTCACCAA	GACCTGAANNNNNNNNNN
11	CGCGGTTC	GAACCGCG	TCTCTACTNNNNNNNNNN
12	TATAACCT	AGGTTATA	CTCTCGTC>NNNNNNNNNN
13	AAGGATGA	TCATCCTT	CCAAGTCTNNNNNNNNNN
14	GGAAGCAG	CTGCTTCC	TTGGACTC>NNNNNNNNNN
15	TCGTGACC	GGTCACGA	GGCTTAAGNNNNNNNNNN
16	CTACAGTT	AACTGTAG	AATCCGGANNNNNNNNNN
17	ATATTCAC	GTGAATAT	TAATACAGNNNNNNNNNN
18	GCGCCTGT	ACAGGCAC	CGGCGTGANNNNNNNNNN
19	ACTCTATG	CATAGAGT	ATGTAAGTN>NNNNNNNNNN
20	GTCTCGCA	TGCGAGAC	GCACGGACNNNNNNNNNN
21	AAGACGTC	GACGTCTT	GGTACCTTNNNNNNNNNN
22	GGAGTACT	AGTACTCC	AACGTTCC>NNNNNNNNNN
23	ACCGGCCA	TGGCCGGT	GCAGAATTNNNNNNNNNN
24	GTAAATTG	CAATTAAC	ATGAGGCC>NNNNNNNNNN

* In i7 index sequence, the 9N indicates the UMI sequence.

Table 3. Premium Methyl UDI-UMI Adapters Sequences – Set B (25-48)

Premium Methyl UDI-UMI Adapter #	i5 index NovaSeq 6000 v1.0, MiSeq, HiSeq 2000/2500 systems	i5 index NovaSeq 6000 v1.5, iSeq, MiniSeq, NextSeq, HiSeq 3000/4000 systems	i7 index* (all Illumina systems)
25	AACCGCGG	CCGCAGTT	ACTAAGATNNNNNNNN
26	GGTTATAA	TTATAACC	GTCGGAGCNNNNNNNN
27	CCAAGTCC	GGACTTGG	CTTGGTATNNNNNNNN
28	TTGGACTT	AAGTCCAA	TCCAACGCNNNNNNNN
29	CAGTGGAT	ATCCACTG	CCGTGAAGNNNNNNNN
30	TGACAAGC	GCTTGTCA	TTACAGGANNNNNNNN
31	CTAGCTTG	CAAGCTAG	GGCATTCTNNNNNNNN
32	TCGATCCA	TGGATCGA	AATGCCTCNNNNNNNN
33	CCTGAACT	AGTCAGG	TACCGAGGNNNNNNNN
34	TTCAGGTC	GACCTGAA	CGTTAGAANNNNNNNN
35	AGTAGAGA	TCTCTACT	AGCCTCATNNNNNNNN
36	GACGAGAG	CTCTCGTC	GATTCTGCNNNNNNNN
37	AGACTTGG	CCAAGTCT	TCGTAGTGNNNNNNNN
38	GAGTCAA	TTGGACTC	CTACGACANNNNNNNN
39	CTTAAGCC	GGCTTAAG	TAAGTGGTNNNNNNNN
40	TCCGGATT	AATCCGGA	CGGACAACNNNNNNNN
41	CTGTATTA	TAATACAG	ATATGGATNNNNNNNN
42	TCACGCCG	CGGCGTGA	GCGCAAGCNNNNNNNN
43	ACTTACAT	ATGTAAGT	AAGATACTNNNNNNNN
44	GTCCGTGC	GCACGGAC	GGAGCGTCNNNNNNNN
45	AAGGTACC	GGTACCTT	ATGGCATGNNNNNNNN
46	GGAACGTT	AACGTTCC	GCAATGCANNNNNNNN
47	AATTCTGC	GCAGAATT	GTTCCAATNNNNNNNN
48	GGCCTCAT	ATGAGGCC	ACCTTGGCANNNNNNNN

* In i7 index sequence, the 9N indicates the UMI sequence.

Table 4. Premium Methyl UDI-UMI Adapters Sequences - 96 rxns (1-96)

Premium Methyl UDI-UMI Adapter #	Well position	i5 index (HiSeq® 2000/2500, MiSeq®, NovaSeq® systems)	i5 index (HiSeq 3000/4000/X, NextSeq®, MiniSeq®, iSeq® systems)	i7 index* (all Illumina systems)
1	A01	AGCGCTAG	CTAGCGCT	CCGCGGTTNNNNNNNNNN
2	A02	GATATCGA	TCGATATC	TTATAACCNNNNNNNNNN
3	A03	CGCAGACG	CGTCTGCG	GGACTTGGNNNNNNNNNN
4	A04	TATGAGTA	TACTCATA	AAGTCCAANNNNNNNNNN
5	A05	AGGTGCGT	ACGCACCT	ATCCACTGNNNNNNNNNN
6	A06	GAACATAC	GTATGTTTC	GCTTGTCA>NNNNNNNNNN
7	A07	ACATAGCG	CGCTATGT	CAAGCTAGNNNNNNNNNN
8	A08	GTGCGATA	TATCGCAC	TGGATCGANNNNNNNNNN
9	A09	CCAACAGA	TCTGTTGG	AGTCAGGNNNNNNNNNN
10	A10	TTGGTGAG	CTCACCAA	GACCTGAANNNNNNNNNN
11	A11	CGCGGTTTC	GAACCGCG	TCTCTACTNNNNNNNNNN
12	A12	TATAACCT	AGGTTATA	CTCTCGTC>NNNNNNNNNN
13	B01	AAGGATGA	TCATCCTT	CCAAGTCTNNNNNNNNNN
14	B02	GGAAGCAG	CTGCTTCC	TTGGACTC>NNNNNNNNNN
15	B03	TCGTGACC	GGTCACGA	GGCTTAAGNNNNNNNNNN
16	B04	CTACAGTT	AACTGTAG	AATCCGGANNNNNNNNNN
17	B05	ATATTCAC	GTGAATAT	TAATACAGNNNNNNNNNN
18	B06	GCGCCTGT	ACAGGCGC	CGCGTGANNNNNNNNNN
19	B07	ACTCTATG	CATAGAGT	ATGTAAGTNNNNNNNNNN
20	B08	GTCTCGCA	TGCGAGAC	GCACGGACNNNNNNNNNN
21	B09	AAGACGTC	GACGTCTT	GGTACCTNNNNNNNNNN
22	B10	GGAGTACT	AGTACTCC	AACGTTCC>NNNNNNNNNN
23	B11	ACCGGCCA	TGGCCGGT	GCAGAATTNNNNNNNNNN
24	B12	GTTAATTG	CAATTAAC	ATGAGGCCNNNNNNNNNN
25	C01	AACCGCGG	CCCGGGTT	ACTAAGATNNNNNNNNNN
26	C02	GGTTATAA	TTATAACC	GTCGGAGC>NNNNNNNNNN
27	C03	CCAAGTCC	GGACTTGG	CTTGGTATNNNNNNNNNN
28	C04	TTGGACTT	AAGTCAA	TCCAACGC>NNNNNNNNNN
29	C05	CAGTGGAT	ATCCACTG	CCGTGAAGNNNNNNNNNN
30	C06	TGACAAGC	GCTTGTCA	TTACAGGANNNNNNNNNN
31	C07	CTAGCTTG	CAAGCTAG	GGCATTCTNNNNNNNNNN
32	C08	TCGATCCA	TGGATCGA	AATGCCTC>NNNNNNNNNN
33	C09	CCTGAECT	AGTTCAAGG	TACCGAGGNNNNNNNNNN

Premium Methyl UDI-UMI Adapter #	Well position	i5 index (HiSeq® 2000/2500, MiSeq®, NovaSeq® systems)	i5 index (HiSeq 3000/4000/X, NextSeq®, MiniSeq®, iSeq® systems)	i7 index* (all Illumina systems)
34	C10	TTCAGGTC	GACCTGAA	CGTTAGAANNNNNNNNN
35	C11	AGTAGAGA	TCTCTACT	AGCCTCATNNNNNNNNN
36	C12	GACGAGAG	CTCTCGTC	GATTCTGCNNNNNNNNN
37	D01	AGACTTGG	CCAAGTCT	TCGTAGTGNNNNNNNNN
38	D02	GAGTCCAA	TTGGACTC	CTACGACANNNNNNNNN
39	D03	CTTAAGCC	GGCTTAAG	TAAGTGGTNNNNNNNNN
40	D04	TCCGGATT	AATCCGGA	CGGACAACNNNNNNNNN
41	D05	CTGTATTAA	TAATACAG	ATATGGATNNNNNNNNN
42	D06	TCACGCCG	CGGCGTGA	GCGCAAGCNNNNNNNNN
43	D07	ACTTACAT	ATGTAAGT	AAGATACTNNNNNNNNN
44	D08	GTCCGTGC	GCACGGAC	GGAGCGTCNNNNNNNNN
45	D09	AAGGTACC	GGTACCTT	ATGGCATGNNNNNNNNN
46	D10	GGAACGTT	AACGTTCC	GCAATGCANNNNNNNNN
47	D11	AATTCTGC	GCAGAATT	GTTCCAATNNNNNNNNN
48	D12	GGCCTCAT	ATGAGGCC	ACCTTGGCANNNNNNNNN
49	E01	ATCTTAGT	ACTAAGAT	ATATCTGNNNNNNNNN
50	E02	GCTCCGAC	GTCGGAGC	GCGCTCTANNNNNNNNN
51	E03	ATACCAAG	CTTGGTAT	AACAGGTTNNNNNNNNN
52	E04	GC GTTGGA	TCCAACGC	GGTGAACCNNNNNNNNN
53	E05	CTTCACGG	CCGTGAAG	CAACAATGNNNNNNNNN
54	E06	TCCTGTAA	TTACAGGA	TGGTGGCANNNNNNNNN
55	E07	AGAATGCC	GGCATTCT	AGGCAGAGNNNNNNNNN
56	E08	GAGGCATT	AATGCCTC	GAATGAGANNNNNNNNN
57	E09	CCTCGGTA	TACCGAGG	TGCGGCGTNNNNNNNNN
58	E10	TTCTAACG	CGTTAGAA	CATAATACNNNNNNNNN
59	E11	ATGAGGCT	AGCCTCAT	GATCTATCNNNNNNNNN
60	E12	GCAGAAC	GATTCTGC	AGCTCGCTNNNNNNNNN
61	F01	CACTACGA	TCGTAGTG	CGGAAC TGNNNNNNNNN
62	F02	TGTCGTAG	CTACGACA	TAAGGT CANNNNNNNNN
63	F03	ACCACTTA	TAAGTGGT	TTGCCTAGNNNNNNNNN
64	F04	GTTGTCCG	CGGACAAC	CCATTG CANNNNNNNNN
65	F05	ATCCATAT	ATATGGAT	ACACTAAGNNNNNNNNN
66	F06	GCTTGCAC	GCGCAAGC	GTGTCGGCANNNNNNNNN

Premium Methyl UDI-UMI Adapter #	Well position	i5 index (HiSeq® 2000/2500, MiSeq®, NovaSeq® systems)	i5 index (HiSeq 3000/4000/X, NextSeq®, MiniSeq®, iSeq® systems)	i7 index* (all Illumina systems)
67	F07	AGTATCTT	AAGATACT	TTCCTGTTNNNNNNNNN
68	F08	GACGCTCC	GGAGCGTC	CCTTCACCNNNNNNNNN
69	F09	CATGCCAT	ATGGCATG	GCCACAGGNNNNNNNNN
70	F10	TGCATTGC	GCAATGCA	ATTGTGAANNNNNNNNN
71	F11	ATTGGAAC	GTTCCAAT	ACTCGTGTNNNNNNNNN
72	F12	GCCAAGGT	ACCTTGGC	GTCTACACNNNNNNNNN
73	G01	CGAGATAT	ATATCTCG	CAATTAACNNNNNNNNN
74	G02	TAGAGCGC	GCGCTCTA	TGGCCGGTNNNNNNNNN
75	G03	AACCTGTT	AACAGGTT	AGTACTCCNNNNNNNNN
76	G04	GGTCACCC	GGTGAACC	GACGTCTTNNNNNNNNN
77	G05	CATTGTTG	CAACAATG	TGCGAGACNNNNNNNNN
78	G06	TGCCACCA	TGGTGGCA	CATAGAGTNNNNNNNNN
79	G07	CTCTGCCT	AGGCAGAG	ACAGGCGCNNNNNNNNN
80	G08	TCTCATTTC	GAATGAGA	GTGAATATNNNNNNNNN
81	G09	ACGCCGCA	TGCGGCGT	AACTGTAGNNNNNNNNN
82	G10	GTATTATG	CATAATAC	GGTCACGANNNNNNNNN
83	G11	GATAGATC	GATCTATC	CTGCTTCCNNNNNNNNN
84	G12	AGCGAGCT	AGCTCGCT	TCATCCTTNNNNNNNNN
85	H01	CAGTTCCG	CGGAACTG	AGGTTATANNNNNNNNN
86	H02	TGACCTTA	TAAGGTCA	GAACCGCGNNNNNNNNN
87	H03	CTAGGCAA	TTGCCTAG	CTCACCAANNNNNNNNN
88	H04	TCGAATGG	CCATTGCA	TCTGTTGGNNNNNNNNN
89	H05	CTTAGTGT	ACACTAAG	TATGCACNNNNNNNNN
90	H06	TCCGACAC	GTGTCGGA	CGCTATGTNNNNNNNNN
91	H07	AACAGGAA	TTCCTGTT	GTATGTTNNNNNNNNN
92	H08	GGTGAAGG	CCTTCACC	ACGCACCTNNNNNNNNN
93	H09	CCTGTGGC	GCCACAGG	TACTCATANNNNNNNNN
94	H10	TTCACAAT	ATTGTGAA	CGTCTGCGNNNNNNNNN
95	H11	ACACGAGT	ACTCGTGT	TCGATATCNNNNNNNNN
96	H12	GTGTAGAC	GTCTACAC	CTAGCGCTNNNNNNNNN

FAQ

1. Do we have to use special sequencing kits or primers?

No, the libraries generated with our Premium Methyl UDI_UMI adapters are compatible with classical Illumina sequencing kits and primers. However, to allow UMI sequence reading during sequencing, it is necessary to specify a particular run mode/recipe to your sequencing provider. UMI sequence is following the i7 sequencing, thus the appropriate run mode is : cycle n-8-(i5) and17-(i7)-n, where n is the read length. As a consequence, you should ask for delivery of the reads unattributed and undemultiplexed and perform the indexes and UMI processing yourself following our data analysis pipeline recommendations and tools (see section Data Analysis Recommendations).

2. Is your kit designed for directional sequencing?

Yes, the libraries generated with the Premium UDI-UMI adapters are directional.

3. Are your adapters compatible with EM-seq (Enzymatic Methyl-seq) protocol?

Our adapters are methylated, therefore it is possible to use them in the EM-seq workflow.

Related Products

Product	Cat. No.
Premium Methyl UDI-UMI Adapters Modules – Set A	C02030040
Premium Methyl UDI-UMI Adapters Modules – Set B	C02030041
Premium Methyl UDI-UMI Adapters Modules – 96 rxns	C02030042
Premium RRBS Kit V2 (24 rxns)	C02030036
Premium RRBS Kit V2 (96 rxns)	C02030037
Bisulfite conversion reagent for RRBS	C02030035
DNA methylation control package V2	C02040019
MethylTaq Plus 2X Master Mix	C09010012

Revision history

Version	Date of modification	Description of modifications
Version 1 12_2024	December 2024	Manual creation

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