

YWG UMI-Stubby Adapter & UDI Index Primer Set for illumina Operating instruction

(Cat#YG002,Version1.3)

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Introduction

YWG UMI-Stubby Adapter & UDI Primer Set for illumina is a DNA library construction adapter kit for illumina high-throughput sequencing platform. The YWG Stubby Adapter for Illumina is an incomplete double-ended UMI-Y type adapter that is compatible with a variety of T-A ligated library construction kits. After ligation, 4 to 5 nt of Unique Molecular Identifier (UMI) was added to each end of Insert DNA, which can be used to detect low-frequency mutations to LOD 0.1%. In addition, the YWG UDI Index Primer Set in the kit contains the Unique Dual Index (UDI), which can be used to double-check two completely independent Index after library amplification, minimizing index hopping & index misassignment. This ensures that the final reads entering the analysis process are the closest to the real situation of the samples.

The kit contains 96 different Truseq UDI Primers, and all the adapter provided in the kit have undergone strict quality control and functional validation to ensure the stability and reproducibility of the library construction to the maximum extent.

YWG UMI-Stubby Adapter adopts bi-directional HPLC and other multiple purification processes, with nucleic acid purity >95%, to improve the efficiency of DNA ligation during library construction. YWG UDI Index Primer Set adopts integrated synthesis and purification technology, which physically isolates the contamination between index primers and avoids the possibility of contamination between indexes in the purification process of HPLC, PAGE, etc. The contamination rate is reduced to less than 1 in 100,000.

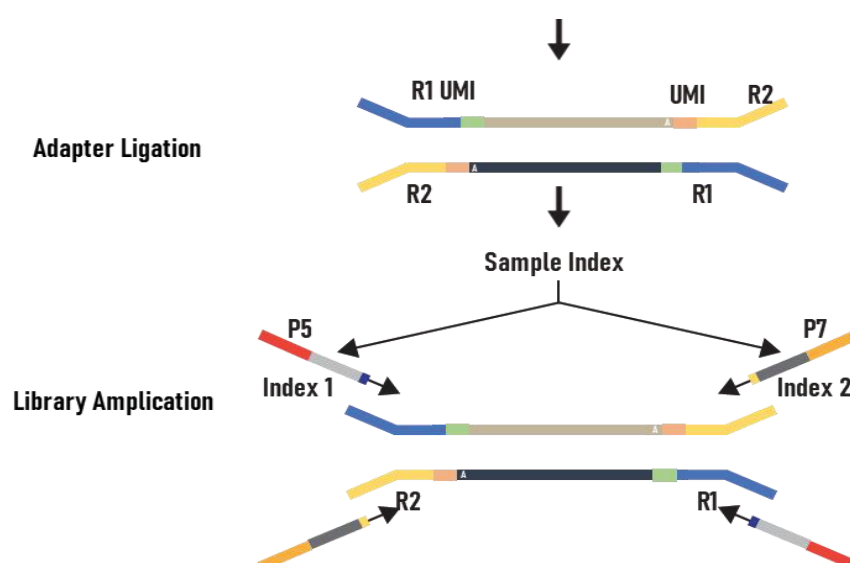



Figure 1: YWG UMI-Stubby Adapter & UDI Index Primer Set for illumina schematic

Component

Component	Concentration	Specification	CatLog
YWG UDI Index Primer plate	15 μmol/each	24 rxn/96 rxn	YG002
YWG UMI-Stubby Adapter*	15 μmol	24 rxn/96 rxn	

 * The amount of DNA library adapter input depends on the amount of DNA sample input, refer to the attached table for the specific concentration.

Storage conditions

-20°C Storage, 4°C delivery.

Application scope

DNA library construction for illumina platform , suitable for gDNA , cDNA , FFPE DNA , Cell-Free DNA and so on.

Sequencing Platform

Illumina MiSeq , MiniSeq , Nextseq , NovaSeq; GenoLab M; MGI T7 after adapter conversion;

Adaptor Structure

YWG UMI-Stubby Adapter for illumina

Name	Sequence
YWG UMI-Stubby Adapter	5'ACACTCTTTCCCTACACGACGCTCTTCCGATCTNNNN(N)-(S)-T-3'
	3'CTGACCTCAAGTCTGCACACGAGAAGGCTAGANNNN(N)-P-5'
P stand for phosphorylation modification, N stand for random random base , (S) stand for thio-modification	

YWG UDI Index-Primer

Name	Sequence
P5 PCR Primer	5'AATGATACGGCGACCACCGAGATCTACAC[i5]ACACTCTTTCCCTACA CGACGCTCTTCCGATC-(S)-T-3'
P7 PCR Primer	5'CAAGCAGAAGACGGCATAACGAGAT[i7]GTGACTGGAGTTCAGACGTG TGCTCTTCCGATC-(S)-T-3'

(S) stand for thio-modification, [i5] stand for 8 bp i5 Index, [i7] stand for 8 bp i7 Index.

Primer Plate-Index Sequence

Well Position	Well Position	Primer name	i5 Index	i5 Index	i7
			(Forward)	(Reverse Complement)	All illumina system
A1	1-A	YWG Index Primer, Index 1	ACCGACAA	TTGTCTGGT	ACGTTACC
B1	1-B	YWG Index Primer, Index 2	AGTGGCAA	TTGCCACT	ATAAGGCG
C1	1-C	YWG Index Primer, Index 3	CACAGACT	AGTCTGTG	CAGCGATT
D1	1-D	YWG Index Primer, Index 4	CGACACTT	AAGTGTCG	GTGCCATA
E1	1-E	YWG Index Primer, Index 5	GACTTGTG	CACAAGTC	GCACAACT
F1	1-F	YWG Index Primer, Index 6	GTGAGACT	AGTCTCAC	CTTCTGAG
G1	1-G	YWG Index Primer, Index 7	GTTCCATG	CATGGAAC	CTCTGGTT
H1	1-H	YWG Index Primer, Index 8	TAGCTGAG	CTCAGCTA	TTACCGAG
A2	2-A	YWG Index Primer, Index 9	CTTCGCAA	TTGCGAAG	CTGTGTTG
B2	2-B	YWG Index Primer, Index 10	GTGGTATG	CATACCAC	CTTACCTG
C2	2-C	YWG Index Primer, Index 11	CACTGTAG	CTACAGTG	TAGTGACC
D2	2-D	YWG Index Primer, Index 12	AGACGCTA	TAGCGTCT	CCTTGTAG
E2	2-E	YWG Index Primer, Index 13	CAACTCCA	TGGAGTTG	TTCTCTCG
F2	2-F	YWG Index Primer, Index 14	AACACGCT	AGCGTGTT	AACCGAAG
G2	2-G	YWG Index Primer, Index 15	TGGATGGT	ACCATCCA	GTTGATGG
H2	2-H	YWG Index Primer, Index 16	TTCGAAGC	GCTTCGAA	GTCCTAAG
A3	3-A	YWG Index Primer, Index 17	AACACCAC	GTGGTGTT	TGAGGTGT
B3	3-B	YWG Index Primer, Index 18	TGAGCTGT	ACAGCTCA	CGTTGCAA
C3	3-C	YWG Index Primer, Index 19	CACAGGAA	TTCCTGTG	CGAGACTA
D3	3-D	YWG Index Primer, Index 20	TGACAACC	GGTTGTCA	GCTGGATT

E3	3-E	YWG Index Primer, Index 21	TGTTCCGT	ACGGAACA	AACGGTCA
F3	3-F	YWG Index Primer, Index 22	CCTAGAGA	TCTCTAGG	TTCGTACC
G3	3-G	YWG Index Primer, Index 23	GCATAACG	CGTTATGC	GCTGTAAG
H3	3-H	YWG Index Primer, Index 24	CAGTGCTT	AAGCACTG	GAAGGTTC
A4	4-A	YWG Index Primer, Index 25	CGTATCTC	GAGATACG	GATCCATG
B4	4-B	YWG Index Primer, Index 26	CGTCAAGA	TCTTGACG	GATTCAGC
C4	4-C	YWG Index Primer, Index 27	CCATGAAC	GTTTCATGG	GACATGGT
D4	4-D	YWG Index Primer, Index 28	GGTACTTC	GAAGTACC	TAACGAGG
E4	4-E	YWG Index Primer, Index 29	ACCGCTAT	ATAGCGGT	ACAGACCT
F4	4-F	YWG Index Primer, Index 30	TTCCAGGT	ACCTGGAA	CTGTTAGG
G4	4-G	YWG Index Primer, Index 31	TCGAACCT	AGGTTCGA	GTCGAAGA
H4	4-H	YWG Index Primer, Index 32	TAGTGCCA	TGGCACTA	GAAGAGGT
A5	5-A	YWG Index Primer, Index 33	GGTACGAA	TTCGTACC	GCCTATCA
B5	5-B	YWG Index Primer, Index 34	AAGCATCG	CGATGCTT	TCACGTTC
C5	5-C	YWG Index Primer, Index 35	GCCAATAC	GTATTGGC	GCATGTCT
D5	5-D	YWG Index Primer, Index 36	CTGTATGC	GCATACAG	ATGGTTGC
E5	5-E	YWG Index Primer, Index 37	CTTAGGAC	GTCCTAAG	TCTCTTCC
F5	5-F	YWG Index Primer, Index 38	TCAGCCTT	AAGGCTGA	CACAAGTC
G5	5-G	YWG Index Primer, Index 39	ACATGCCA	TGGCATGT	GAGCTCAA
H5	5-H	YWG Index Primer, Index 40	GATGGAGT	ACTCCATC	TCTGAGAG
A6	6-A	YWG Index Primer, Index 41	CGATCGAT	ATCGATCG	AACAACCG
B6	6-B	YWG Index Primer, Index 42	TACTCCAG	CTGGAGTA	TGTGCGTT
C6	6-C	YWG Index Primer, Index 43	AGCTACCA	TGGTAGCT	ACTCCATC

D6	6-D	YWG Index Primer, Index 44	TCGACAAG	CTTGTCGA	CCTATACC
E6	6-E	YWG Index Primer, Index 45	TATGACCG	CGGTCATA	AGTGTTGG
F6	6-F	YWG Index Primer, Index 46	AGCCAACT	AGTTGGCT	TCTTGACG
G6	6-G	YWG Index Primer, Index 47	GATCTTGC	GCAAGATC	TGAACCTG
H6	6-H	YWG Index Primer, Index 48	CCTCGTTA	TAACGAGG	ACCGCATA
A7	7-A	YWG Index Primer, Index 49	AAGACACC	GGTGTCTT	ACTCGTTG
B7	7-B	YWG Index Primer, Index 50	GATACCTG	CAGGTATC	TAGTTGCG
C7	7-C	YWG Index Primer, Index 51	AACCGAAC	GTTCCGTT	TGTGACTG
D7	7-D	YWG Index Primer, Index 52	GCTGAATC	GATTCAGC	TTAGGTCG
E7	7-E	YWG Index Primer, Index 53	AGCTAGTG	CACTAGCT	TGGCATGT
F7	7-F	YWG Index Primer, Index 54	CTAGCTCA	TGAGCTAG	CGTCTTGT
G7	7-G	YWG Index Primer, Index 55	GTTAAGCG	CGCTTAAC	CCGACTAT
H7	7-H	YWG Index Primer, Index 56	CGATTGGA	TCCAATCG	GAAGTACC
A8	8-A	YWG Index Primer, Index 57	CATCTGCT	AGCAGATG	CCTATGGT
B8	8-B	YWG Index Primer, Index 58	ACCTCTTC	GAAGAGGT	AAGAGCCA
C8	8-C	YWG Index Primer, Index 59	ATCGCAAC	GTTGCGAT	CGAAGAAC
D8	8-D	YWG Index Primer, Index 60	AGTTGTGC	GCACAACT	GCAAGATC
E8	8-E	YWG Index Primer, Index 61	GAACGAAG	CTTCGTTC	AGAAGCGT
F8	8-F	YWG Index Primer, Index 62	GGAAGAGA	TCTCTTCC	CGTGATCA
G8	8-G	YWG Index Primer, Index 63	GTCATCGT	ACGATGAC	AGCTAACC
H8	8-H	YWG Index Primer, Index 64	CCAACGAA	TTCGTTGG	CAGGTATC
A9	9-A	YWG Index Primer, Index 65	CTCTCAGA	TCTGAGAG	TGTACACC
B9	9-B	YWG Index Primer, Index 66	ACGGACTT	AAGTCCGT	ACAGCTCA

C9	9-C	YWG Index Primer, Index 67	GTTGCTGT	ACAGCAAC	GGTGTCTT
D9	9-D	YWG Index Primer, Index 68	TGTCGACT	AGTCGACA	AGAGCCTT
E9	9-E	YWG Index Primer, Index 69	CGTCTAAC	GTTAGACG	AGCGGAAT
F9	9-F	YWG Index Primer, Index 70	AACACTGG	CCAGTGTT	CCAAGTTG
G9	9-G	YWG Index Primer, Index 71	TCAGACAC	GTGTCTGA	GCCTTGTT
H9	9-H	YWG Index Primer, Index 72	AGAAGGAC	GTCCTTCT	TCTCTAGG
A10	10-A	YWG Index Primer, Index 73	TCGTCTGA	TCAGACGA	GTATGCTG
B10	10-B	YWG Index Primer, Index 74	CATGTGTG	CACACATG	GTAAAGGC
C10	10-C	YWG Index Primer, Index 75	TCTAGTCC	GGACTAGA	AAGAAGGC
D10	10-D	YWG Index Primer, Index 76	AAGGCTCT	AGAGCCTT	GCAATGGA
E10	10-E	YWG Index Primer, Index 77	AACCAGAG	CTCTGGTT	TAACCGGT
F10	10-F	YWG Index Primer, Index 78	ACTATCGC	GCGATAGT	GTACCTTG
G10	10-G	YWG Index Primer, Index 79	GTCCTAAG	CTTAGGAC	AACTTGCC
H10	10-H	YWG Index Primer, Index 80	TGACCGTT	AACGGTCA	AAGCACTG
A11	11-A	YWG Index Primer, Index 81	CAATAGCC	GGCTATTG	TGATGTCC
B11	11-B	YWG Index Primer, Index 82	TGCCTCAA	TTGAGGCA	AAGCCACA
C11	11-C	YWG Index Primer, Index 83	GACGAACT	AGTTCGTC	AGGTTCGA
D11	11-D	YWG Index Primer, Index 84	CCTAACAG	CTGTTAGG	CTGGAGTA
E11	11-E	YWG Index Primer, Index 85	CGCCTTAT	ATAAGGCG	CATGGAAC
F11	11-F	YWG Index Primer, Index 86	ACAACAGC	GCTGTTGT	GACTATGC
G11	11-G	YWG Index Primer, Index 87	AGACCTTG	CAAGGTCT	CAATGTGG
H11	11-H	YWG Index Primer, Index 88	GCGTTAGA	TCTAACGC	CCAAGCAA
A12	12-A	YWG Index Primer, Index 89	CATTCGTC	GACGAATG	GTCCTTCT

B12	12-B	YWG Index Primer, Index 90	ATCTGACC	GGTCAGAT	ACACGGTT
C12	12-C	YWG Index Primer, Index 91	TTCGTACG	CGTACGAA	CATGTTCC
D12	12-D	YWG Index Primer, Index 92	AAGACGAG	CTCGTCTT	GAACATCG
E12	12-E	YWG Index Primer, Index 93	CTCGTTCT	AGAACGAG	ATGGTCCA
F12	12-F	YWG Index Primer, Index 94	TGTGGCTT	AAGCCACA	TGGATCAC
G12	12-G	YWG Index Primer, Index 95	AGACATGC	GCATGTCT	AAGGCTGA
H12	12-H	YWG Index Primer, Index 96	TCTAGGAG	CTCCTAGA	TGTTTCGAG

Illumina Index Form

Forward	NovaSeq® 6000 (v1.0 reagent kits)
	HiSeq® 2000, 2500
	NextSeq® 1000, 2000 (using v2 Sample Sheet)
	MiSeq®
	MiniSeq® (rapid reagent kits)
	GenoLab M (SE075-D)
	FASTASeq 300 (SE075-D)
Reverse Complement	NovaSeq® 6000 (v1.5 reagent kits)
	HiSeq® 3000, 4000, X
	NextSeq® 1000, 2000 (using v1 Sample Sheet)
	NextSeq® 500, 550
	MiniSeq® (standard reagents)
	iSeq® 100
	GenoLab M (150/300 cycles)
	FASTASeq 300 (150/300 cycles)

Appendix

The amount of library adapter to be used depends on the amount of DNA required by the different kits, please refer to the table below if you are using the matching library construction kits, if cfDNA input, donot do the adapter dilution.

Input DNA	Dilution times	Concentration
1 µg~50 ng	NO	15 µM
49 ng ~25 ng	2X dilution	7.5 µM
24 ng ~10 ng	5X dilution	3 µM
9 ng ~ 5 ng	10X dilution	1.5 µM
< 5 ng	20X dilution	0.75 µM

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