If more than 8 DNA libraries need to be multiplexed on *one lane on an Illumina sequencer*, the following **additional 8 index pairs** may be purchased from IDT and used for Library PCR.

i5 index Primer	**Sequence**	**Index Sequence**
i5_Unique_9	AATGATACGGCGACCACCGAGATCTACAC AGAACGAG TCGTCGGCAGCGTC	AGAACGAG
i5_Unique_10	AATGATACGGCGACCACCGAGATCTACAC TGCTTCCA TCGTCGGCAGCGTC	TGCTTCCA
i5_Unique_11	AATGATACGGCGACCACCGAGATCTACAC CTTCGACT TCGTCGGCAGCGTC	CTTCGACT
i5_Unique_12	AATGATACGGCGACCACCGAGATCTACAC CACCTGTT TCGTCGGCAGCGTC	CACCTGTT
i5_Unique_13	AATGATACGGCGACCACCGAGATCTACAC ATCACACG TCGTCGGCAGCGTC	ATCACACG
i5_Unique_14	AATGATACGGCGACCACCGAGATCTACAC CCGTAAGA TCGTCGGCAGCGTC	CCGTAAGA
i5_Unique_15	AATGATACGGCGACCACCGAGATCTACAC TACGCCTT TCGTCGGCAGCGTC	TACGCCTT
i5_Unique_16	AATGATACGGCGACCACCGAGATCTACAC CGACGTTA TCGTCGGCAGCGTC	CGACGTTA

i7 index Primer	**Sequence**	**Index Sequence**
i7_Unique_9	CAAGCAGAAGACGGCATACGAGATATTAGCCGGTCTCGTGGGCTCGG	CGGCTAAT
i7_Unique_10	CAAGCAGAAGACGGCATACGAGATCGATCGATGTCTCGTGGGCTCGG	ATCGATCG
i7_Unique_11	CAAGCAGAAGACGGCATACGAGATGATCTTGCGTCTCGTGGGCTCGG	GCAAGATC
i7_Unique_12	CAAGCAGAAGACGGCATACGAGATAGGATAGCGTCTCGTGGGCTCGG	GCTATCCT
i7_Unique_13	CAAGCAGAAGACGGCATACGAGATGTAGCGTAGTCTCGTGGGCTCGG	TACGCTAC
i7_Unique_14	CAAGCAGAAGACGGCATACGAGATAGAGTCCAGTCTCGTGGGCTCGG	TGGACTCT
i7_Unique_15	CAAGCAGAAGACGGCATACGAGATGCTACTCTGTCTCGTGGGCTCGG	AGAGTAGC
i7_Unique_16	CAAGCAGAAGACGGCATACGAGATCTCTGGATGTCTCGTGGGCTCGG	ATCCAGAG

ORDER

1. Order oligos (single-stranded DNA) on IDT: https://www.idtdna.com/pages/support/how-to-order
2. Purification method: **standard desalting, 25 nmole**

LIBRARY PCR SETUP

3. Dilute each oligo to 4 µM with nuclease-free water (typically from 100 µM stock)
4. For each sample combine **5 µl of i5 index primer** and **5 µl of i7 index primer** (see Table 1 on the next page) → The combined primers represent the “V2 Index Primer”
5. Proceed as outlined in the User Guide