

Oligo Name	Sequence	Supplier	Length (bp)	Tm (°C)	GC %	Tm of homologous region (°C)	Tm used (°C)
Ultramers							
GFP ultramer F	<u>GAATGAGAGAAAGACGGACAGAACC</u> GATGGACAAACAAACACAACCTTAAAGATTTACAGCACAGCTTAGCTTTATTAAGTAGATCATTAGTTGAGTTACTTTAGTCGCCACCA TGGTGAGCAA	IDT	125	70.8	38.4	60.3	60
GFP ultramer R	<u>CCTCTCTGCTGAGCCTCCACACA</u> AAATCACTGAGGAGAAAAACCAGCGCTAACAGCCACAGCATCTTCATCCTCATCTCCATGATATCTGCAGAAATTCGCCC	IDT	100	72.2	51	52.5	60
iTOL ultramer F	<u>CGGCACGTAAGAGGTTCCA</u> ACTTTCACCATAATGAAATAAGATCACTACCGGGCGTATTTTTGAGTTATCGAGATTCCTGCTCGAGCCGGGCCC	IDT	96	71.7	48.9	73.4	60
iTOL ultramer R	<u>CATCCGCTTATTATCACTTATT</u> CAGGCGTAGCAACCAGGCGTTAAGGGCACCAATAACTATTATGATCCTCTAGATCAGATCT	IDT	84	69.3	41.6	45.3	60
Deletion reg.2 ultramer R	<u>GCTCAGTTTGT</u> CATCATCAGGTCGGCTCAGCCTGGCTCAGTTATATTTCGTAGAGCCAGTCCATGCATCCGTCGATTATGATCCTCTAGATCAGATCT	IDT	100	71.5	49	45.3	60
Ex63 ultramer F	<u>GACACAGACCTCTGACCCCTG</u> CAGCAGCGGAGCTGAGTGGCTCACACAAACAGCGGCAGCGTGAGTCTCACACAAAAGGTAGCTTGCAGTGGGCTT	IDT	100	74.8	60	64.2	60
BAC backbone ultramer R	<u>GGTCTCTCAGCGTATGGTTGTCG</u> CCGGATGTATAATTGCCTTCATCGATAAACTGCTGAACGTTCTGATATGTTTTCCGCTCATACTTTCCTTTTTCA	IDT	100	70.8	43	45.2	60
Ex4 ultramer F	<u>GATCTGGAGCGTTCCTGGAC</u> GGCGTGAACCTCCAGCCGTGGCAGTTTACGCCATTAGCGATACTGAGTGTGACGCAGGTAGCTTGCAGTGGGCTT	IDT	100	73.7	55	64.2	60
Ex2 ultramer F	<u>GAATCTGCCAGCAATGCCG</u> AAATCAGCACCAACGCCACCTGTGGCGATCCCGATCCCGAGATGTTCTGCAAACCTGGTGGAGGTAGCTTGCAGTGGGCTT	IDT					
Ex1 ultramer F	<u>GGAGATGAGGATGAAGATGCTGTGG</u> CTGTTAGCGCTGTTTTCTCCTCAGTGATTTGTGTGGAGGCTCAGCAGAGAGGTGAGGTAGCTTGCAGTGGGCTT	IDT	100	73	53	64.2	60
Ultramer end primers							

GFP end F	<i>GAATGAGAGAAAGACGGACAGAACC</i>	MWG	25	62	48		
GFP end R	<i>CCTCTCTGCTGAGCTCCACAC</i>	MWG	22	63.3	63.6		
iTOL end F	<i>CGGCACGTAAGAGGTTCCA</i>	MWG	19	60.5	57.9		
iTOL end R	<i>CATCCGCTTATTATCACTTATTCAGGC</i>	MWG	27	62	40.7		
Upstream reg 2 end R	<i>GCTCAGTTTGTATCATCAGGTCCG</i>	MWG	25	61.7	64.6		
Ex63 end F	<i>GACACACGACCTCTGACCCCTG</i>	MWG	22	64	63.6		
BAC backbone end R	<i>GGTCTCTCAGCGTATGGTTGTCG</i>	MWG	23	63.7	56.5		
Ex4 end F	<i>GATTCTGGAGCGTCCCTGGAC</i>	MWG	22	62.9	59.1		
Ex2 end F	<i>GAATCTGCCAGCAATGCCG</i>	IDT	20	60.2	60		
Ex1 end F	<i>GGAGATGAGGATGAAGATGCTGTGGC</i>	IDT	26	62.4	53.8		

Table 2.6: *Ultramer/primer sequences. Ultramer/primer sequences are in the 5' to 3' orientation. The part of the ultramer homologous to the plasmid/cassette is shown in italics, and the T_m of this homology region is indicated. End primers with homology to the ultramer 5' end are underlined on the ultramer sequence.*