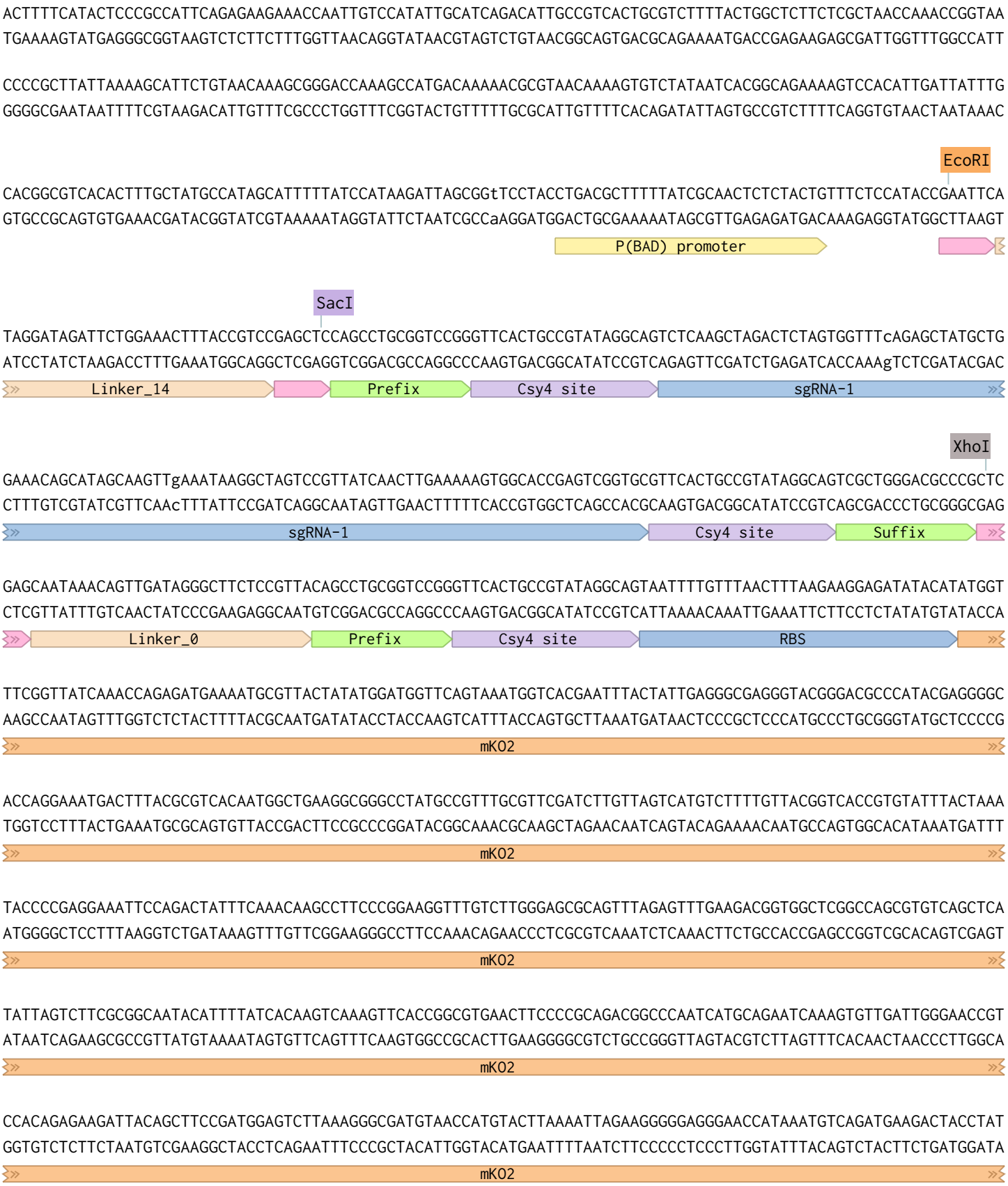
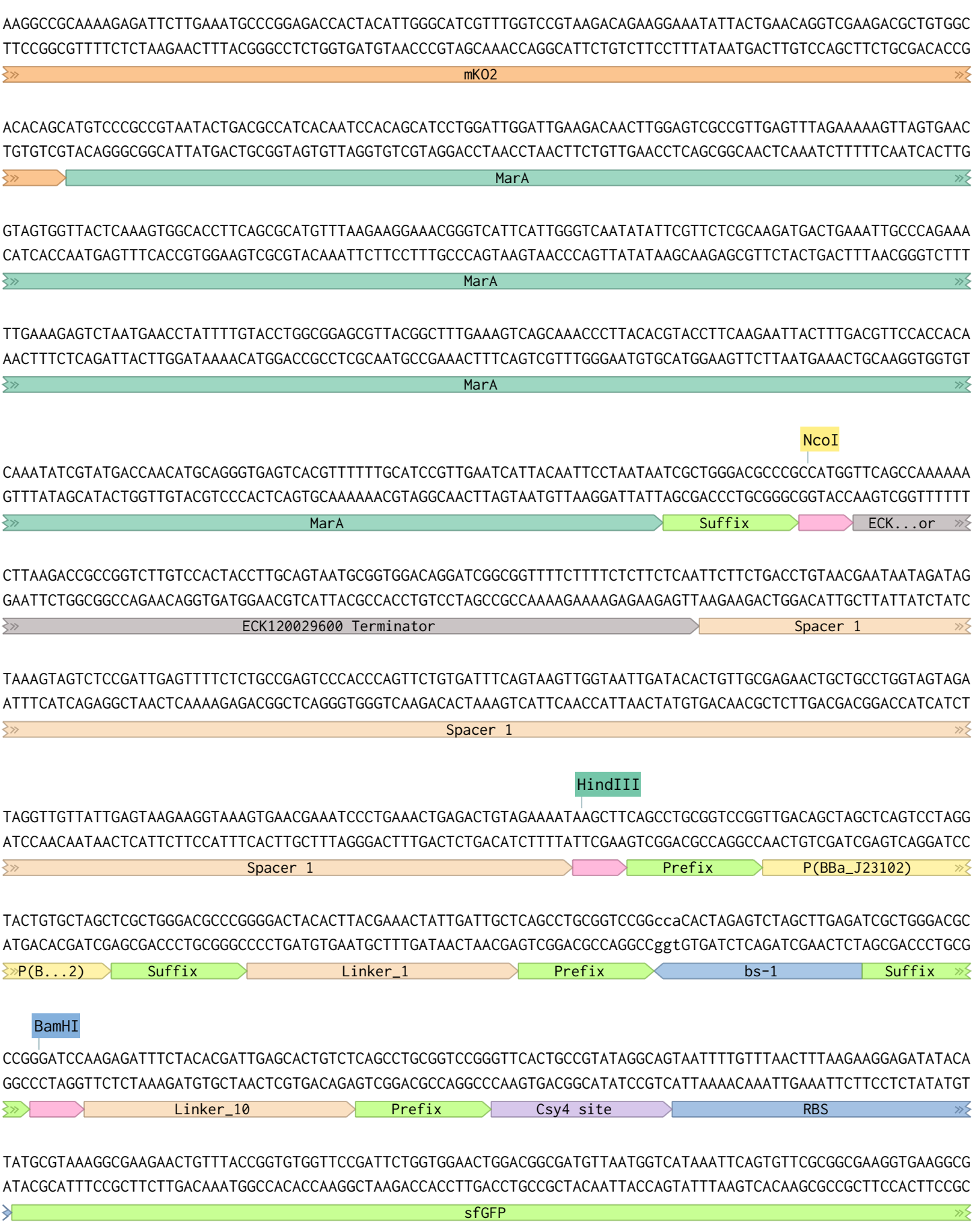
(from 1-1177 bp)

pJ2044 (7459 bp)



pJ2044 (7459 bp) (from 1178-2354 bp)



pJ2044 (7459 bp) (from 2355-3531 bp)

ATGCGACGAACGGCAAACTGACCCTGAAATTTATCTGCACCACGGGTAAACTGCCGGTCCCGTGGCCGACGCTGGTGACCACGCTGACCTATGGCGTTCAATGTTTT
TACGCTGCTTGCCGTTTGACTGGGACTTTAAATAGACGTGGTGCCCATTTGACGGCCAGGGCACCGGCTGCGACCACTGGTGCGACTGGATACCGCAAGTTACAAAA

sfGFP

GCGCGTTACCCGGATCACATGAAACAGCACGACTTTTTCAAATCGGCCATGCCGGAAGGCTATGTGCAGGAACGTACGATTAGCTTTAAAGACGATGGTACGTATAA
CGCGCAATGGGCCTAGTGTACTTTGTCGTGCTGAAAAAGTTTAGCCGGTACGGCCTTCCGATACACGTCCTTGCATGCTAATCGAAATTTCTGCTACCATGCATATT

sfGFP

AACCCGCGCGGAAGTGAAATTCGAAGGCGATACCCTGGTTAACCGTATCGAACTGAAAGGTATCGATTTCAAAGAAGACGGCAATATTCTGGGTCATAAACTGGAAT
TTGGGCGCGCCTTCACTTTAAGCTTCCGCTATGGGACCAATTGGCATAGCTTGACTTTCCATAGCTAAAGTTTCTTCTGCCGTTATAAGACCCAGTATTTGACCTTA

sfGFP

ATAACTTCAATTCCCACAACGTGTACATCACCGCGGATAAACAGAAAAACGGCATTAAAGCCAATTTCAAAATCCGCCATAATGTGGAAGATGGTAGCGTTCAGCTG
TATTGAAGTTAAGGGTGTTGCACATGTAGTGGCGCCTATTTGTCTTTTTGCCGTAATTTCGGTTAAAGTTTTAGGCGGTATTACACCTTCTACCATCGCAAGTCGAC

sfGFP

GCCGACCACTATCAGCAAAACACGCCGATTGGTGATGGCCCGGTCCTGCTGCCGGACAATCACTACCTGAGTACCCAGTCCGTGCTGTCAAAAGATCCGAACGAAAA
CGGCTGGTGATAGTCGTTTTGTGCGGCTAACCACTACCGGGCCAGGACGACGGCCTGTTAGTGATGGACTCATGGGTCAGGCACGACAGTTTTCTAGGCTTGCTTTT

sfGFP

ACGTGACCACATGGTCCTGCTGGAATTTGTGACGGCTGCGGGTATCACCCACGGCATGGACGAACTGTATAAAATGTCCCGCCGTAATACTGACGCCATCACAATCC
TGCACTGGTGTACCAGGACGACCTTAAACACTGCCGACGCCCATAGTGGGTGCCGTACCTGCTTGACATATTTTACAGGGCGGCATTATGACTGCGGTAGTGTTAGG

sfGFP MarAn20

SbfI KpnI

ACAGCATCCTGGATTGGATTGAAGACTAATAATCGCTGGGACGCCCGCCTGCAGGCTCGGTACCAAATTCCAGAAAAGAGGCCTCCCGAAAGGGGGGCCTTTTTTCG
TGTCGTAGGACCTAACCTAACTTCTGATTATTAGCGACCCTGCGGGCGGACGTCCGAGCCATGGTTTAAGGTCTTTTCTCCGGAGGGCTTTCCCCCCGGAAAAAAGC

MarAn20 Suffix L3S2P21 Terminator

TTTTGGTCCTAATAGATAAAGGATAGGTCTGGTAGTGTTGTTCGTTCTCGCAGGTAAATCAATAATACTCAGCAGTTCCGTAGACTTTTCAGTGGGACAGGGTAGCG
AAAACCAGGATTATCTATTTCCTATCCAGACCATCACAACAAGCAAGAGCGTCCATTTAGTTATTATGAGTCGTCAAGGCATCTGAAAAGTCACCCTGTCCCATCGC

Spacer 2.5

SalI

ATAACAGATAGATTGTAATAAGACACAGTAGGTGCTCGTAGTTGCGTGAAGAGAACCGCTCAGGAAATCCAGTCAGAAGTATTGGTAATCGTTGAAAACTCAGTCGA
TATTGTCTATCTAACATTATTCTGTGTCATCCACGAGCATCAACGCACTTCTCTTGGCGAGTCCTTTAGGTCAGTCTTCATAACCATTAGCAACTTTTGAGTCAGCT

Spacer 2.5

AatII

CGCACTTACTGAAGACGTCCTATTACACTCGTCGTTGGAAACTGAAGATCAGCCTGCGGTCCGGGTTCACTGCCGTATAGGCAGTAATTTTGTTTAACTTTAAGAAG
GCGTGAATGACTTCTGCAGGATAATGTGAGCAGCAACCTTTGACTTCTAGTCGGACGCCAGGCCCAAGTGACGGCATATCCGTCATTAAAACAAATTGAAATTCTTC

Spa... 5 Linker_11 Prefix Csy4 site RBS

GAGATATACATATGAATCAGTCATTCATCTCGGACATCTTATATGCCGACATCGAATCGAAGGCTAAGGAACTTACAGTCAATTCCAACAATACTGTCCAGCCGGTC
CTCTATATGTATACTTAGTCAGTAAGTAGAGCCTGTAGAATATACGGCTGTAGCTTAGCTTCCGATTCCTTGAATGTCAGTTAAGGTTGTTATGACAGGTCGGCCAG

RBS RepA70

pJ2044 (7459 bp) (from 3532-4815 bp)

GCGCTTATGCGCTTAGGAGTTTTCGTTCCCAAACCTTCCAAGAGCAAAGGAGAAAGTAAGGAAATTGACGCCACCAAAGCCTTCTCTCAACTGGAGATTGCTAAAGC
CGCGAATACGCGAATCCTCAAAAGCAAGGGTTTGGAAGGTTCTCGTTTCCTCTTTCATTCCTTTAACTGCGGTGGTTTCGGAAGAGAGTTGACCTCTAACGATTTCG

RepA70

AGAGGGCatggttagtaaaggagaagaaaataacatggcaCTGATTAAGGAGAACATGCACATGAAGCTGTACATGGAGGGCACCGTGAACAACCACCACTTCAAGT
TCTCCCGtaccaatcatttcctcttcttttattgtaccgtGACTAATTCCTCTTGTACGTGTACTTCGACATGTACCTCCCGTGGCACTTGTTGGTGGTGAAGTTCA

mKate2

GCACATCCGAGGGCGAAGGCAAGCCCTACGAGGGCACCCAGACCATGAGAATCAAGgccGTCGAGGGCGGCCCTCTCCCCTTCGCCTTCGACATCCTGGCTACCAGC
CGTGTAGGCTCCCGCTTCCGTTCGGGATGCTCCCGTGGGTCTGGTACTCTTAGTTCcggCAGCTCCCGCCGGGAGAGGGGAAGCGGAAGCTGTAGGACCGATGGTCG

mKate2

TTCATGTACGGCAGCAAAACCTTCATCAACCACACCCAGGGCATCCCCGACTTCTTTAAGCAGTCCTTCCCTGAGGGCTTCACATGGGAGAGAGTCACCACATACGA
AAGTACATGCCGTCGTTTTGGAAGTAGTTGGTGTGGGTCCCGTAGGGGCTGAAGAAATTCGTCAGGAAGGGACTCCCGAAGTGTACCCTCTCTCAGTGGTGTATGCT

mKate2

AGACGGGGGCGTGCTGACCGCTACCCAGGACACCAGCCTCCAGGACGGCTGCCTCATCTACAACGTCAAGATCAGAGGGGTGAACTTCCCATCCAACGGCCCTGTGA
TCTGCCCCCGCACGACTGGCGATGGGTCCTGTGGTCGGAGGTCCTGCCGACGGAGTAGATGTTGCAGTTCTAGTCTCCCCACTTGAAGGGTAGGTTGCCGGGACACT

mKate2

TGCAGAAGAAAACACTCGGCTGGGAGGCCTCCACCGAGaccCTGTACCCCGCTGACGGCGGCCTGGAAGGCAGAgcCGACATGGCCCTGAAGCTCGTGGGCGGGGGC
ACGTCTTCTTTTGTGAGCCGACCCTCCGGAGGTGGCTCtggGACATGGGGCGACTGCCGCCGGACCTTCCGTCTcgGCTGTACCGGGACTTCGAGCACCCGCCCCCG

mKate2

CACCTGATCTGCAACTTGAAGACCACATACAGATCCAAGAAACCCGCTAAGAACCTCAAGATGCCCGGCGTCTACTATGTGGACAGAAGACTGGAAAGAATCAAGGA
GTGGACTAGACGTTGAACTTCTGGTGTATGTCTAGGTTCTTTGGGCGATTCTTGGAGTTCTACGGGCCGCAGATGATACACCTGTCTTCTGACCTTTCTTAGTTCCT

mKate2

GGCCGACAAAGAGACCTACGTCGAGCAGCACGAGGTGGCTGTGGCCAGATACTGCGACCTCCCTAGCAAACTGGGGCACAgAgtctaATAATCGCTGGGACGCCCGG
CCGGCTGTTTCTCTGGATGCAGCTCGTCGTGCTCCACCGACACCGGTCTATGACGCTGGAGGGATCGTTTGACCCCGTGTcTcagatTATTAGCGACCCTGCGGGCC

mKate2 Suffix

NotI

CGGCCGCggaaacacagAAAAAAGCCCGCACCTGACAGTGCGGGCTTTTTTTTTcgaccaaaggTAGCGAACGACGAGTCACTGTTGAGGATAAATACTTTCTCTAC
GCCGGCGcctttgtgtcTTTTTTCGGGCGTGGACTGTCACGCCCGAAAAAAAAAgctggtttccATCGCTTGCTGCTCAGTGACAACTCCTATTTATGAAAGAGATG

ECK120033737 Terminator Linker_8

KasI BbvCI AscI

TAGGCGCCTGTTACACAGGTCCTCAGCGGCGCGCCTTTGTCGGTGAACGCTCTCCTGAGTAGGACAAATCCGCCGGGAGCGGATTTGAACGTTGTGAAGCAACGGCC
ATCCGCGGACAATGTGTCCAGGAGTCGCCGCGCGGAAACAGCCACTTGCGAGAGGACTCATCCTGTTTAGGCGGCCCTCGCCTAAACTTGCAACACTTCGTTGCCGG

Spa...15

CGGAGGGTGGCGGGCAGGACGCCCGCCATAAACTGCCAGGCATCAAACTAAGCAGAAGGCCATCCTGACGGATGGCCTTTTTGCGTTTCAGATCTACCGGTaaacca
GCCTCCCACCGCCCGTCCTGCGGGCGGTATTTGACGGTCCGTAGTTTGATTCGTCTTCCGGTAGGACTGCCTACCGGAAAAACGCAAAGTCTAGATGGCCAtttggt

gcaatagacataagcggctatttaacgaccctgccctgaaccgacgacaagctgacgaccgggtctccgcaagtggcacttttcggggaaatgtgcgcggaacccct
cgttatctgtattcgccgataaattgctgggacgggacttggctgctgttcgactgctggcccagaggcgttcaccgtgaaaagccccctttacacgcgccttgggga

pJ2044 (7459 bp) (from 4816-6099 bp)

atttgtttatttttctaaatacattcaaatatgtatccgctcatgaattaattcttagaaaaactcatcgagcatcaaatgaaactgcaatttattcatatcaggat
taaacaaataaaaagatttatgtaagtttatacataggcgagtacttaattaagaatctttttgagtagctcgtagtttactttgacgttaaataagtatagtccta

KanR

tatcaataccatatttttgaaaaagccgtttctgtaatgaaggagaaaactcaccgaggcagttccataggatggcaagatcctggtatcggtctgcgattccgact
atagttatggtataaaaacttttcggcaaagacattacttcctcttttgagtggctccgtcaaggtatcctaccgttctaggaccatagccagacgctaaggctga

KanR

cgtccaacatcaatacaacctattaatttcccctcgtcaaaaataaggttatcaagtgagaaatcaccatgagtgacgactgaatccggtgagaatggcaaaagttt
gcaggttgtagttatgttggataattaaaggggagcagtttttattccaatagttcactctttagtggtactcactgctgacttaggccactcttaccgttttcaaa

KanR

atgcatttctttccagacttgttcaacaggccagccattacgctcgtcatcaaaatcactcgcatcaaccaaaccgttattcattcgtgattgcgcctgagcgagac
tacgtaaagaaaggtctgaacaagttgtccggtcggtaatgcgagcagtagttttagtgagcgtagttggtttggcaataagtaagcactaacgcggactcgctctg

KanR

gaaatacgcggtcgctgttaaaaggacaattacaaacaggaatcgaatgcaaccggcgcaggaacactgccagcgcatcaacaatattttcacctgaatcaggatat
ctttatgcgccagcgacaattttcctgttaatgtttgtccttagcttacgttggccgcgtccttgtgacggtcgcgtagttgttataaaagtggacttagtcctata

KanR

tcttctaatacctggaatgctgttttcccggggatcgcagtggtgagtaaccatgcatcatcaggagtacggataaaatgcttgatggtcggaagaggcataaattc
agaagattatggaccttacgacaaaagggcccctagcgtcaccactcattggtacgtagtagtcctcatgcctattttacgaactaccagccttctccgtatttaag

KanR

cgtcagccagtttagtctgaccatctcatctgtaacatcattggcaacgctacctttgccatgtttcagaaacaactctggcgcatcgggcttcccatacaatcgat
gcagtcggtcaaatcagactggtagagtagacattgtagtaaccgttgcgatggaaacggtacaaagtctttgttgagaccgcgtagcccgaagggtatgttagcta

KanR

agattgtcgcacctgattgcccgacattatcgcgagcccatttatacccatataaatcagcatccatgttggaatttaatcgcggcctagagcaagacgtttcccgt
tctaacagcgtggactaacgggctgtaatagcgctcgggtaaatatgggtatatttagtcgtaggtacaaccttaaattagcgccggatctcgttctgcaaagggca

KanR

tgaatatggctcatactcttccttttttcaatattattgaagcatttatcagggttattgtctcatgagcggatacatatttgaatgtatttagaaaaataaacaaat
acttataccgagtatgagaaggaaaaagttataataacttcgtaaatagtcccaataacagagtactcgcctatgtataaacttacataaatctttttatttgttta

KanR

aggcatgctagcgcagaaacgtcctagaagatgccaggaggatacttagcagagagacaataaggccggagcgaagccgtttttccataggctccgccccccctgacg
tccgtacgatcgcgtctttgcaggatcttctacggtcctcctatgaatcgtctctctgttattccggcctcgcttcggcaaaaaggtatccgaggcgggggggactgc

ColA ori

aacatcacgaaatctgacgctcaaatcagtggtggcgaaacccgacaggactataaagataccaggcgtttcccccctgatggctccctcttgcgctctcctgttccc
ttgtagtgctttagactgcgagtttagtcaccaccgctttgggctgtcctgatatttctatggtccgcaaaggggactaccgagggagaacgcgagaggacaaggg

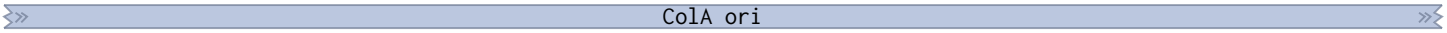
ColA ori

gtcctgcggcgtccgtgttgtggtggaggctttacccaaatcaccacgtcccgttccgtgtagacagttcgctccaagctgggctgtgtgcaagaacccccgttca
caggacgccgcaggcacaacaccacctccgaaatgggtttagtggtgcagggcaaggcacatctgtcaagcgaggttcgacccgacacacgttcttgggggggcaagt

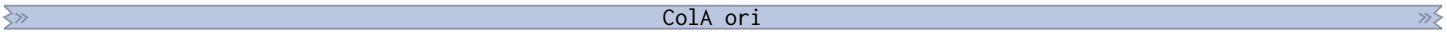
ColA ori

pJ2044 (7459 bp) (from 6100-7383 bp)

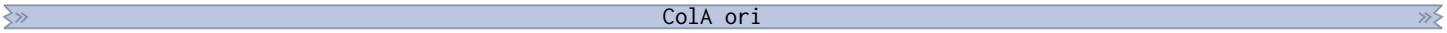
gcccgactgctgcgccttatccggtaactatcatcttgagtccaacccggaaagacacgacaaaacgccactggcagcagccattggtaactgagaattagtggatt
cgggctgacgacgcggaataggccattgatagtagaactcaggttgggccttctgtgctgttttgcggtgaccgtcgtcggtaaccattgactcttaatcacctaa



tagatatcgagagtcttgaagtggtggcctaacagaggctacactgaaaggacagtatttggtatctgcgctccactaaagccagttaccaggttaagcagttcccc
atctatagctctcagaacttcaccaccggattgtctccgatgtgactttcctgtcataaaccatagacgcgaggtgatttcggtcaatggtccaattcgtcaagggg

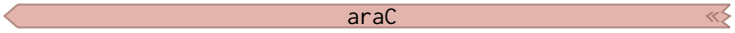


aactgacttaaccttcgatcaaaccgcctccccaggcggtttttttcgtttacagagcaggagattacgacgatcgtaaaaggatctcaagaagatcctttacggatt
ttgactgaattggaagctagtttggcggaggggtccgccaaaaaagcaaatgtctcgtcctctaatgctgctagcattttcctagagttcttctaggaaatgcctaa

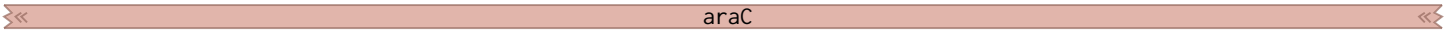


cccgacaccatcactctagatttcagtgcaatttatctcttcaaatgtagcacctgaagtcagccccatacgatataagttgtaattctcatgttagtcatgccccg
gggctgtggtagtgagatctaaagtcacgttaaatagagaagtttacatcgtggacttcagtcggggtatgctatattcaacattaagagtacaatcagtacggggc

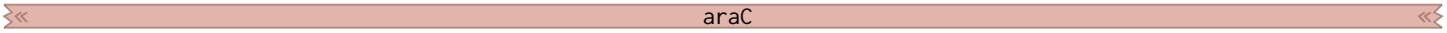
cgcccaccggaaggagctgactgggttgCTCCTAgGGTCTGATTCGTTACCAATTATGACAACTTGACGGCTACATCATTCACTTTTTCTTCACAACCGGCACGGAA
gcgggtggccttcctcgactgacccaacGAGGATcCCAGACTAAGCAATGGTTAATACTGTTGAACTGCCGATGTAGTAAGTGAAAAAGAAGTGTTGGCCGTGCCTT



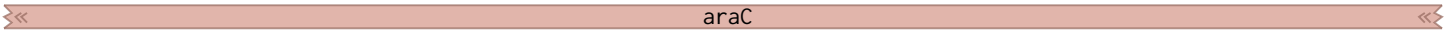
CTCGCTCGGGCTGGCCCCGGTGCATTTTTTAAATACCCGCGAGAAATAGAGTTGATCGTCAAAACCAACATTGCGACCGACGGTGGCGATAGGCATCCGGGTGGTGC
GAGCGAGCCCGACCGGGGCCACGTAAAAAATTTATGGGCGCTCTTTATCTCAACTAGCAGTTTTGGTTGTAACGCTGGCTGCCACCGCTATCCGTAGGCCCACCACG



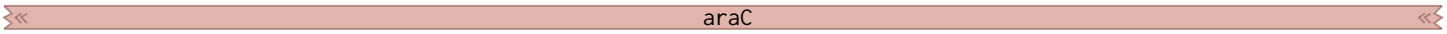
TCAAAAGCAGCTTCGCCTGGCTGATACGTTGGTCCTCGCGCCAGCTTAAGACGCTAATCCCTAACTGCTGGCGGAAAAGATGTGACAGACGCGACGGCGACAAGCAA
AGTTTTCGTCGAAGCGGACCGACTATGCAACCAGGAGCGCGGTCGAATTCTGCGATTAGGGATTGACGACCGCCTTTTCTACACTGTCTGCGCTGCCGCTGTTCGTT



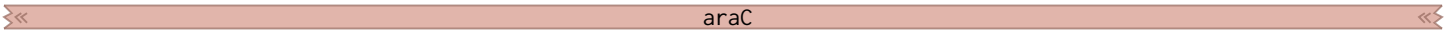
ACATGCTGTGCGACGCTGGCGATATCAAAATTGCTGTCTGCCAGGTGATCGCTGATGTACTGACAAGCCTCGCGTACCCGATTATCCATCGGTGGATGGAGCGACTC
TGTACGACACGCTGCGACCGCTATAGTTTTAACGACAGACGGTCCACTAGCGACTACATGACTGTTCGGAGCGCATGGGCTAATAGGTAGCCACCTACCTCGCTGAG



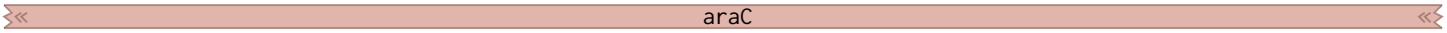
GTTAATCGCTTCCATGCGCCGCAGTAACAATTGCTCAAGCAGATTTATCGCCAGCAGCTCCGAATAGCGCCCTTCCCCTTGCCCGGCGTTAATGATTTGCCCAAACA
CAATTAGCGAAGGTACGCGGCGTCATTGTTAACGAGTTCGTCTAAATAGCGGTCGTCGAGGCTTATCGCGGGAAGGGGAACGGGCCGCAATTACTAAACGGGTTTGT



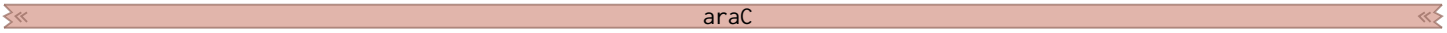
GGTCGCTGAAATGCGGCTGGTGCGCTTCATCCGGGCGAAAGAACCCCGTATTGGCAAATATTGACGGCCAGTTAAGCCATTCATGCCAGTAGGCGCGCGGACGAAAG
CCAGCGACTTTACGCCGACCACGCGAAGTAGGCCCGCTTTCTTGGGGCATAACCGTTTATAACTGCCGGTCAATTCGGTAAGTACGGTCATCCGCGCGCCTGCTTTC



TAAACCCACTGGTGATACCATTCGCGAGCCTCCGGATGACGACCGTAGTGATGAATCTCTCCTGGCGGGAACAGCAAAATATCACCCGGTCGGCAAACAAATTCTCG
ATTTGGGTGACCACTATGGTAAGCGCTCGGAGGCCTACTGCTGGCATCACTACTTAGAGAGGACCGCCCTTGTCGTTTTATAGTGGGCCAGCCGTTTGTTTAAGAGC



TCCCTGATTTTTCACCACCCCCTGACCGCGAATGGTGAGATTGAGAATATAACCTTTCATTCCCAGCGGTCGGTCGATAAAAAAATCGAGATAACCGTTGGCCTCAA
AGGGACTAAAAAGTGGTGGGGGACTGGCGCTTACCACTCTAACTCTTATATTGGAAAGTAAGGGTCGCCAGCCAGCTATTTTTTTAGCTCTATTGGCAACCGGAGTT



pJ2044 (7459 bp) (from 7384-7459 bp)

TCGGCGTTAAACCCGCCACCAGATGGGCATTAAACGAGTATCCCGGCAGCAGGGGATCATTTTGCGCTTCAGCCAT
AGCCGCAATTTGGGCGGTGGTCTACCCGTAATTTGCTCATAGGGCCGTCGTCCCCTAGTAAAACGCGAAGTCGGTA