

Sequences head-to-tail cloning

Primer sequences in bold.

5'-overhangs marked in bold and italic.

pHisZ_GFP11

Backbone pHisZ:

CTCGAGGCGGCCGCATAACGCCGGTCGCTACCATTACCAACTTGTCTGGTGTCAAAAATAATAGGCCT
ACTAGTCGGCCGTACGGGCCCTTTTCGTCTCGCGCGTTTCGGTGATGACGGTGAAAACCTCTGACACATG
CAGCTCCCGGAGACGGTCACAGCTTGTCTGTAAGCGGATGCCGGGAGCAGACAAGCCCGTCAGGGCGCG
TCAGCGGGTGTGGCGGGTGTGGGGCTGGCTTAACTATGCCGCATCAGAGCAGATTGTAAGAGAGTG
CACCATATGCCGGTGTGAAATACCGCACAGATGCGTAAGGAGAAAATACCGCATCAGGCGGCCCTTAAGGG
CCTCGTGATACGCCTATTTTTATAGGTTAATGTCATGATAATAATGGTTTCTTAGACGTCAGGTGGCAC
TTTTCGGGGAAATGTGCGCGGAACCCCTATTTGTTATTTTTCTAAATACATTCAAAATATGTATCCGCT
CATGAGACAATAACCCTGATAAATGCTTCAATAATATTGAAAAAGGAAGAGTATGAGTATTCAACATT
TCCGTGTGCGCCTTATCCCTTTTTTTCGGCATTTTTGCCTTCCTGTTTTTGTCCACCCAGAAAACGCTGGT
GAAAGTAAAAGATGCTGAAGATCAGTTGGGTGCACGAGTGGGTTACATCGAACTGGATCTCAACAGCG
GTAAGATCCTTGAGAGTTTTGCCCCGAAGAAGCTTTTCCAATGATGAGCACTTTTAAAGTTCTGCTAT
GTGGCGCGGTATTATCCCGTATTGACGCGGGGAAGAGCAACTCGGTCGCCGCATACACTATTCTCAGA
ATGACTTGGTTGAGTACTCACCAGTCACAGAAAAGCATCTTACGGATGGCATGACAGTAAGAGAATTA
TGCAGTGCTGCCATAACCATGAGTGATAACACTGCGGCCAACTTACTTCTGACAACGATCGGAGGACCG
AAGGAGCTAACCGCTTTTTTGCACAACATGGGGGATCATGTAACCTCGCCTTGATCGTTGGGAACCGGAG
CTGAATGAAGCCATAACCAAACGACGAGCGTGACACCAGATGCCTGTAGCAATGGCAACAACGTTGCCG
AAACTATTAAGTGGCGAACTACTTACTTAGCTTCCCGGCAACAATTAATAGACTGGATGGAGGCGGAT
AAAGTTGCAGGACCACTTCTGCGCTCGGCCCTTCCGGCTGGCTGGTTTATTGCTGATAAATCTGGAGCC
GGTGAGCGTGGGTCTCGCGGTATCATTGCAGCACTGGGGCCAGATGGTAAGCCCTCCCGTATCGTAGTT
ATCTACACGACGGGGAGTCAGGCAACTATGGATGAACGAAATAGACAGATCGCTGAGATAGGTGCCTC
ACTGATTAAGCATTGGTAAGTGTGACAGCAAGTTTACTCATATATACTTTAGATTGATTTAAAACCTTCA
TTTTTAATTTAAAAGGATCTAGGTGAAGATCCTTTTTGATAATCTCATGACCAAATCCCTTAAAGTGA
GTTTTCGTTCCACTGAGCGTCAGACCCCGTAGAAAAGATCAAAGGATCTTCTTGAGATCCTTTTTTTTCT
GCGCGTAATCTGCTGCTTGCAAACAAAAAAACCACCGCTACCAGCGGTGGTTTGTGTTGCCGGATCAAGA
GCTACCAACTCTTTTTCCGAAGGTAAGTGGCTTTCAGCAGAGCGCAGATACCAAATACTGTCCTTCTAGT
GTAGCCGTAGTTAGGCCACCCTTCAAGAACTCTGTAGCACCAGCTACATACCTCGCTCTGCTAATCCTG
TTACCAGTGGCTGCTGCCAGTGGCGATAAGTCGTGCTTACCAGGTTGGACTCAAGACGATAGTTACCG
GATAAGGCGCAGCGGTGCGGCTGAACGGGGGGTTCGTGCACACAGCCAGCTTGGAGCGAACGACCTAC
ACCGAACTGAGATACCTACAGCGTGAGCTATGAGAAAGCGCCACGCTTCCCGAAGGGAGAAAGCGCGAC
AGGTATCCGGTAAGCGGCAGGGTTCGGAACAGGAGAGCGCACGAGGGAGCTTCCAGGGGAAACGCCTG
GTATCTTTATAGTCTGTGCGGTTTTCGCCACCTCTGACTTGAGCGTCGATTTTTGTGATGCTCGTCAGG
GGGGCGGAGCCTATGGAAAAACGCCAGCAACGCGGCCCTTTTTACGGTTCCTGGCCTTTTGTGCTGGCCTTT
TGCTCACATGTTCTTTCTGCGTTATCCCCTGATTTCTGTGGATAACCGTATTACCGCCTTTGAGTGAGC
TGATACCGCTCGCCGAGCCGAACGACCGAGCGCAGCGAGTCAGTGAGCGAGGAAGCGGAAGAGCGCCC
AATACGCAAACCGCTCTCCCCGCGGTTGGCCGATTCATTAATGCAGCTGGCAGCAGAGGTTTCCCGAC
TGGAAGCGGGCAGTGAGCGCAACGCAATTAATGTGAGTTAGCTCACTCATTAGGCACCCAGGCTTTA
CACTTTATGCTTCCGGCTCGTATGTTGTGTGGAATTGTGAGCGGATAACAATTTACACAGGAAACAGC
TATGACCATGATTACGCCAAGCTTGCATCATCATCATCATGTAGACAACAATTCACCAAAGAACA
ACAAAACCGGTTCTATGAGATCTTACATTTACCTAACTTAAACGAAGAACAACGAAACGCCTTCATCCA
AAGTTTAAAAGATGACCAAGCCAAAGCGCTAACTTGCTAGCAGAAGCTAAAAGCTAAATGATGCTC
AGGCGCCGAAACAGATCCAATCGGAT***CCCCGGTACCGTTCGCCACCGGTGGATCCGGT***

Insert GFP11 oligo:

CGGGTACCGGTGCCACCGGTGGATCCGGTTCGTGACCACATGGTCCTTCATGAGTACGTAAATGCTGCT
GGGATTACAGCGGCCGCACGACTC

Restriction enzyme for bead release: NotI

pHisZ_eGFP

Backbone pHisZ: same as for pHisZ_GFP11

Insert eGFP:

**CGGGTACCGGTCCGACCGGTGGATCCGGTATGGTGAGCAAGGGCGAGGAGCTGTTACCGGGGTGGT
GCCCATCCTGGTTCGAGCTGGACGGCGACGTAACCGGCCACAAGTTCAGCGTGTCCGGCGAGGGCGAGGG
CGATGCCACCTACGGCAAGCTGACCCTGAAGTTCATCTGCACCACCGGCAAGCTGCCCGTGGCCCTGGCCC
ACCCTCGTGACCACCCTGACCTACGGCGTGCAGTGTTCAGCCGCTACCCCGACCACATGAAGCAGCACG
ACTTCTTCAAGTCCGCCATGCCCCGAAGGCTACGTCCAGGAGCGCACCATCTTCTTCAAGGACGACGGCA
ACTACAAGACCCCGCGCCGAGGTGAAGTTCGAGGGCGACACCCTGGTGAACCGCATCGAGCTGAAGGGCA
TCGACTTCAAGGAGGACGGCAACATCCTGGGGCACAAGCTGGAGTACAACACTACAACAGCCACAACGTCT
ATATCATGGCCGACAAGCAGAAGAACGGGCATCAAGGTGAACCTCAAGATCCGCCACAACATCGAGGACG
GCAGCGTGCAGCTCGCCGACCACTACCAGCAGAACACCCCATCGGCGACGGCCCCGTGCTGCTGCCCGA
CAACCACTACCTGAGCACCCAGTCCGCCCTGAGCAAAGACCCCAACGAGAAGCGCGATCACATGGTCCT
GCTGGAGTTCGTGACCGCCCGGGATCACTCTCGGCATGGACGAGCTGTACAAGGGCGGCCGACGAC
TC**

Restriction enzyme for bead release: NotI

pHisZ_mCherry

Backbone pHisZ: same as for pHisZ_GFP11

Insert mCherry

**CGGGTACCGGTCCGACCGGTGGATCCGGTATGGTGAGCAAGGGCGAGGAGGATAACATGGCCATCAT
CAAGGAGTTCATGCGCTTCAAGGTGCACATGGAGGGCTCCGTGAACGGCCACGAGTTCGAGATCGAGGG
CGAGGGCGAGGGCCGCCCTACGAGGGCACCCAGACCGCCAAGCTGAAGGTGACCAAGGGTGGCCCCCT
GCCCTTCGCCTGGGACATCCTGTCCCTCAGTTCATGTACGGCTCCAAGGCCTACGTGAAGCACCCCGCC
GACATCCCCGACTACTTGAAGCTGTCTTCCCCGAGGGCTTCAAGTGGGAGCGCGTGAAGTTCGAG
GACGGCGGCGTGGTGACCGTGACCCAGGACTCCTCCCTGCAGGACGGCGAGTTCATCTACAAGGTGAAG
CTGCGCGGACCAACTTCCCTCCGACGGCCCCGTAATGCAGAAGAAGACCATGGGCTGGGAGGCCTCCT
CCGAGCGGATGTACCCCGAGGACGGCGCCCTGAAGGGCGAGATCAAGCAGAGGCTGAAGCTGAAGGACG
GCGGCCACTACGACGCTGAGGTCAAGACCACCTACAAGGCCAAGAAGCCCGTGCAGCTGCCCGGCGCCT
ACAACGTCAACATCAAGTTGGACATCACCTCCACAACGAGGACTACACCATCGTGGAACAGTACGAAC
GCGCCGAGGGCCGCCACTCCACCGGCGGCATGGACGAGCTGTACAAGGGCGGCCGACGACTC**

Restriction enzyme for bead release: NotI

PalkB_GFP_pZE

Backbone AlkS Alcanivorax NC_008260:

**CTCGAGTCCGACGTCCGCAACAAGACCCCTGTTGCATCTCCTCCAAAGACCTGCGTACTAAGGGATG
AATGCATATTGATATTATGTACAGAGCAAAAACCTACCCTTTGGAGTAGTTGTTTGGCCTCTATTTCTAG
CTAACCTACCCTGCCTACAACCTACAACCTCGTCCGCTACCAACATTACGCGGTGTGCGACAGCCTGT
AGAAACAATCAATATGATAAAAACAAGGTGATCCTAGAATTCATTAAAGAG**

Insert PalkB_GFP:

**AACAAGGTGATCCTAGAATTCATTAAAGAGGAGAAAGGTACCGCATGCGTAAAGGAGAAGAACCTTT
TCACTGGAGTTGTCCCAATTCTTGTTGAATTAGATGGTGATGTTAATGGGCACAAATTTTCTGTGCTGAGT
GAGAGGGTGAAGGTGATGCAACATACGGAAAACCTACCCTTAAATTTATTTGCACTACTGGAAAACCTA
CCTGTTCCATGGCCAACACTTGTCACTACTTTCCGTTATGGTGTTCATGCTTTGCGAGATAACCAGAT
CATATGAAAACAGCATGACTTTTTCAAGAGTGCCATGCCCCGAAGGTTATGTACAGGAAAGAACTATATT
TTTTCAAGATGACGGAACTACAAGACACGTGCTGAAGTCAAGTTTGAAGGTGATACCCTTGTTAATA
GAATCGAGTTAAAAAGGTATTGATTTTTAAAGAAGATGGAAACATTCTTGGACACAAATTTGGAATACAAC
TATAACTCACACAATGTATACATCATGGCAGACAAAACAAAAGAATGGAATCAAAGTTAACTTCAAAAT
TAGACACAACATTGAAGATGGAAGCGTTCAACTAGCAGACCATTATCAACAAAATACTCCAATTGGCG
ATGGCCCTGTCTTTTTACCAGACAACCATTACCTGTCCACACAATCTGCCCTTTGAAAGATCCCAACGA
AAAGAGAGACCACATGGTCTTCTTGAGTTTGTAAACAGCTGCTGGGATTACACATGGCATGGATGAACT
ATACAAAAGGCTGCAGCAAACGACGAAAACCTACGCTGCAGCAGTTTAATAAGCTTGATATCGAATTCC
TGCAGCCCGGGGATCCCATGGTACGCGTGTAGAGGCATCAAATAAAAACGAAAGGCTCAGTCGAAAGA**

CTGGGCCTTTCGTTTTATCTGTTGTTTGTGCGGTGAACGCTCTCCTGAGTAGGACAAATCCGCCGCCCTA
GACCTAGGCGTTTCGGCTGCGGCGAGCGGTATCAGCTCACTCAAAGGCGGTAATACGGTTATCCACAGAA
TCAGGGGATAACGCAGGAAAGAACATGTGAGCAAAAGGCCAGCAAAAGGCCAGGAACCGTAAAAAGGC
CGCGTTGCTGGCGTTTTTCCATAGGCTCCGCCCCCTGACGAGCATCACAAAAATCGACGCTCAAGTCA
GAGGTGGCGAAACCCGACAGGACTATAAAGATACCAGGCGTTTCCCCCTGGAAGTCCCTCGTGCGCTC
TCCTGTTCCGACCCTGCCGCTTACCGGATACCTGTCCGCTTCTCCCTTCGGAAGCGTGCGCTTCT
CAATGCTCACGCTGTAGGTATCTCAGTTCGGTGTAGGTGCTTCGCTCCAAGCTGGGCTGTGTGCACGAA
CCCCCGTTCAGCCCGACCCTGCGCCTTATCCGTAACCTATCGTCTTGAGTCCAACCCGGTAAGACAG
ACTTATCGCCACTGGCAGCAGCCACTGGTAACAGGATTAGCAGAGCGAGGTATGTAGGCGGTGCTACAG
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AGCCAGTTACCTTCGAAAAAGAGTTGGTAGCTCTTGATCCGGCAAAACAAACCACCGCTGGTAGCGGTG
GTTTTTTTGTGTTGCAAGCAGCAGATTACGCGCAGAAAAAAGGATCTCAAGAAGATCCTTTGATCTTTT
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TTCTCACCAATAAAAAACGCCCGGGCAACCGAGCGTTCTGAACAAATCCAGATGGAGTTCTGAGGTC
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CAAGAAGGCGATAGAAGGCGATGCGCTGCGAATCGGGAGCGGCGATACCGTAAAGCACGAGGAAGCGG
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GCCACACCAGCCGGCCACAGTCGATGAATCCAGAAAAGCGGCCATTTTCCACCATGATATTCGGCAAG
CAGGCATCGCCATGGGTACGACGAGATCCTCGCCGTCGGGCATGCGCGCCTTGAGCCTGGCGAACAGT
TCGGCTGGCGCGAGCCCCTGATGCTCTTCGTCCAGATCATCTGATCGACAAGACCCGCTTCCATCCGAG
TACGTGCTCGCTCGATGCGATGTTTCGCTTGGTGGTGAATGGGCAGGTAGCCGGATCAAGCGTATGCA
GCCGCCGATTGATCAGCCATGATGGATACTTTCTCGGCAGGAGCAAGGTGAGATGACAGGAGATCCT
GCCCGGCACCTCGCCAATAGCAGCCAGTCCCTTCCCGCTTCAGTGACAACGTCGAGCACAGCTGCGCA
AGGAACGCCCCTCGTGGCCAGCCACGATAGCCGCGCTGCCTCGTCTGCAGTTCATTCAGGGCACCCGAC
AGGTGGTCTTGACAAAAAGAACCCGGCGCCCCTGCGCTGACAGCCGGAACACGGCGGCATCAGAGCAG
CCGATTGTCTGTTGTGCCAGTCATAGCCGAATAGCCTCTCCACCCAAGCGGCCGGAGAACCTGCGTGC
AATCCATCTTGTCAATGCGAAAACGATCCTCATCTGTCTCTTGATCAGATCTTGATCCCCTGCGCC
ATCAGATCCTTGGCGGCAAGAAAGCCATCCAGTTTACTTTGCAGGGCTTCCCAACCTTACCAGAGGGCG
CCCCAGCTGGCAATTCCGACGTCATGCAT

Restriction enzyme for bead release: AatII

2180012_2180013pAff8c

Backbone 2180012:

**CTCGAGGCGGCCGCAACAGCCAGGAAGGGAACACCTTCAAGCTCCAAGAGAGTATAGACCCCTGCAGG
TGAGGAGACCATGTCCCAACTTCTCCAACCTTAATGCCCGTATGCGAAGCCATTCTTTCTATTTCAGTC
AATATGAAAGACAAAGGTGGTATAGAAAAGTTGGAAAAGTATTTTTAAAGAAAGTCCCTGAGCCTACA
CCGGGCTACTAGTTCCCACTCTGTAGCAAAAAGAACCCAAAGCTCCTGCAGCCCCTGCCAACACCTTGCC
ATTGTTCTGATTCCAGAAGACCATCATCGTGTATAGACATCTATGTCTCTGCTATGGATGAGCTCCAC
TGTGATATAGGCTCAGGATCGAGCATTGA**

Insert 2180013pAff8c:

**CTGTGATATAGGCTCAGGATCGAGCATTGATTTTTGAGGACATCACCTCCATGGACACTAGATCTTTTT
CTTCAGACTACACCCACCTCCAGAATGCCAAAACCCCTGGGACTCAGAGCCTCCGATGTACCACACCAT
TGAGCGTTCCAAAAGTAGCCGCTACCTAGCCACCACGCCCTTTCTTCTAGAAGAGGCTCCCATTGTGAA
ATCTCATAGCTTTATGTTTTCCCCCTCAAGGAGCTATTATGCCAACTTTGGGGTGCCTGTAAAAACAGC
AGAATACACAAGTATTACAGACTGTATTGACACAAGGTGTGTCAATGCCCCCAAGCAATTGCGGACAG
AGCTGCCTTCCCTGGAGGTCTTGAGAGCAAAGTGGAGGACTTAACTTGCTGCCATCCAGAGCGAGAAGC
AGAAGTGAAGTACCCAGCTCTTAAGGCGCGCCCCACCGCTGAGCAATAACTAGCATAACCCCTTGGGG
CCTCTAAACGGGTCTTGAGGGGTTTTTTGCTGAAAGGAGGAACTATATCCGGATTGGCGAATGGGACGC
GCCCTGTAGCGGCGCATTAAAGCGCGCGGGTGTGGTGGTTACGCGCAGCGTGACCGCTACACTTGCCAG
CGCCCTAGCGCCCCTCCTTTCGCTTCTTCCCTTCTTCTCGCCACGTTGCGCGGCTTTCCCCGTCAAG
CTCTAAATCGGGGGCTCCCTTTAGGGTTCCGATTTAGTGCTTTACGGCACCTCGACCCCAAAAAAATTG
ATTAGGGTGTGTTACAGTGGGCTATCGCCCTGATAGACGGTTTTTCGCCCTTTGACGTTGGAGT
CCACGTTCTTTAATAGTGGACTCTTGTTCCAAACTGGAACAACACTCAACCCTATCTCGGTCTATTCTT
TTGATTTATAAGGGATTTTGGCGATTTCCGGCCTATTGGTTAAAAAATGAGCTGATTTAACAAAAATTT
AACCGCAATTTTAAACAAAATATTAACGTTTACAATTTACAGGTGGCACTTTTCCGGGAAATGTGCGCGG
AACCCCTATTTGTTTATTTTTCTAAATACATTCAAATATGTATCCGCTCATGAATTAATTTAGAAAA**

ACTCATCGAGCATCAAATGAAACTGCAATTTATTCATATCAGGATTATCAATACCATATTTTTGAAAA
AGCCGTTTCTGTAATGAAGGAGAAAACTCACCGAGGCAGTTCCATAGGATGGCAAGATCCTGGTATCGG
TCTGCGATTCCGACTCGTCCAACATCAATACAACCTATTAATTTCCCCTCGTCAAAAATAAGGTTATCA
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CCGCGTTTTCCAGACTTTACGAAACACGGAACCGAAGACCATTTCATGTTGTTGCTCAGGTGCGCAGACGT
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GATCCCGGTGCCTAATGAGTGAGCTAACTTACATTAATTTGCGTTGCGCTCACTGCCCCGTTTTCCAGTCG
GGAAACCTGTGCTGCCAGCTGCATTAATGAATCGGCCAACGCGCGGGGAGAGGGCGTTTTGCGTATTGGG
CGCCAGGGTGGTTTTTTCTTTTACCAGTGAGACGGGAACAGCTGATTGCCCTTACCAGCCTGGCCCTG
AGAGAGTTGCAGCAAGCGTCCACGCTGGTTTTGCCCCAGCAGGCGAAAATCCTGTTTGTGTTGTTAA
CGGCGGGATATAACATGAGCTGTCTCGGTATCGTCGATCCCACTACCGAGATATCCGCACCAACGCG
CAGCCCGACTCGGTAATGGCGCGCATTGCGCCAGCGCCATCTGATCGTTGGCAACCAGCATCGCAGT
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CGAGACAGAACTTAATGGGCCCCGCTAACAGCGCGATTTGCTGGTGACCCAATGCGACCAGATGCTCCAC
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AATGATCAGCCACTGACGCGTTGCGCGAGAAGATTGTGCACCGCGCTTTACAGGCTTCGACGCGCT
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GCAGAAACGTGGCTGGCCTGGTTTACCACGCGGGAAACGGTCTGATAAGAGACACCGGCATACTCTGCG
ACATCGTATAACGTTACTGGTTTTACATTCACCACCCTGAATTTGACTCTCTTCCGGGCGCTATCATGCC

ATACCGCGAAAGGTTTTGCGCCATTTCGATGGTGTCCGGGATCTCGACGCTCTCCCTTATGCGACTCCTGC
ATTAGGAAGCAGCCCAGTAGTAGGTTGAGGCCGTTGAGCACCGCCCGCAAGGAATGGTGCATGCAAG
GAGATGGCGCCCAACAGTCCCCGGCCACGGGGCTGCCACCATACCCACGCCGAAACAAGCGCTCATGA
GCCCGAAGTGGCGAGCCCGATCTTCCCATCGGTGATGTCCGGCATATAGGCGCCAGCAACCGCACCTG
TGGCGCCGGTGTATGCCGGCCACGATGCGTCCGGCGTAGAGGATCGAGATCTCGATCCCGCGAAATTAAT
ACGACTCACTATAGGGGAATTGTGAGCGGATAACAATCCCCCTCTAGAAATAATTTTGTAACTTTAA
GAAGGAGATATACCATGGGCAGCAGCCATCATCATCATCACAGCAGCGCCTGGTGCCGCGCGCA
GCCATATGGCTAGCGCGGCCGACGACTC

Restriction enzyme for bead release: NotI

2250322_2250323pAff8c

Backbone 2250322:

CTCGAGGCGGCCGATATTTGAATTATGGGCCCTACAGTTCCTTATGCACCGCATTATGACTCCACATT
TGCAAATATCAGCAAGGATGATTCTGATTTAATCTATTCAACCTATGGGGAAGACTCTGATCTTCCAAG
TGATTTCAAGCATCCATGAGTTTTTGGCCACGTGCCAAGATTATCCGTATGTCATGGCAGATAGTTACT
GGATGTTTTAAACAAAAGGAGGGCATTCCAGGACCCTACAAGAGATGGAGATGTCATTGCCTGAAGATG
AAGGCCATACTAGGACACTTGACACAGCAGGCTCAGGATCGGAGCCACCA

Insert 2250323pAff8c:

GACACAGCAGGCTCAGGATCGGAGCCACCAGGGCGTTTTGGACTCCAGTACTCAAGACAGGCTCATAGC
GCTGAAAGCAGTAAACAAATTTTGGCGTTCAGTTGAAGTTTTTACTCTGAAGAAGCTGAAATATTCC
AGAAGAACTTGATGAGACCACCAGATTGCTCAGGAACTCCAGGAAGCCCAGAATGAACGTTTGAGC
ACCAGACCCCTCCGAACATGATCTGTCTCTTGGGTCCCTCATAACAGAGAAATGCATCTTGCTGAACAA
GTGACCAATAATCTTAAATAAGGCGCGCCACCAGCTGAGCAATAACTAGCATAACCCCTTGGGGCCTC
TAAACGGGTCTTGAGGGTTTTTGGTGAAGGAGGAACTATATCCGGATTGGCGAATGGGACGCGCCC
TGTAGCGGCGCATTAAGCGCGGGGTGTGGTGGTTACGCGCAGCGTGACCGCTACACTTGGCAGCGCC
CTAGCGCCCGCTCCTTTCGCTTCTTCCCTTCTTCTCGCCACGTTTCGCGGCTTTCCCCGTCAAGCTCT
AAATCGGGGGCTCCCTTTAGGGTTCCGATTTAGTGCTTTACGGCACCTCGACCCCAAAAACTTGATTA
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GTTCTTTAATAGTGGACTCTTGTTCAAACTGGAACAACACTCAACCCTATCTCGGTCTATTCTTTTGA
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CGAATTTTAAACAAAATATTAACGTTTACAATTTACAGTGGCACTTTTTCGGGAAATGTGCGCGGAACCC
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TTCTGTAATGAAGGAGAAAACCTCACCGAGCGAGTTCCATAGGATGGCAAGATCCTGGTATCGGTCTGCG
ATTCCGACTCGTCCAACATCAATACAACCTATTAATTTCCCCTCGTCAAAAATAAGGTTATCAAGTGAG
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TGCGCCTGAGCGAGACGAAATACGCGATCGCTGTTAAAAGGACAATTACAAACAGGAATCGAATGCAA
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GATGGTCCGAAGAGGCATAAAATCCGTCAGCCAGTTTAGTCTGACCATCTCATCTGTAACATCATTTGGC
AACGCTACCTTTGCCATGTTTTAGAAACAACCTTGGCGCATCGGGCTTCCATAACAATCGATAGATTGT
CGCACCTGATTGCCGACATTATCGCGAGCCATTTATACCCATATAAATCAGCATCCATGTTGGAATT
TAATCGCGGCTAGAGCAAGACGTTTTCCGTTGAATATGGCTCATAACACCCCTTGTATTACTGTTTAT
GTAAGCAGACAGTTTTTATGTTTCATGACCAAAAATCCCTTAACGTGAGTTTTTCGTTCCACTGAGCGTCAG
ACCCCGTAGAAAAGATCAAAGGATCTTCTTGAGATCCTTTTTTTCTGCGGTAATCTGCTGCTTGCAAA
CAAAAAAACACCGCTACCAGCGGTGGTTTTGTTGCGGATCAAGAGCTACCAACTCTTTTTCCGAAGG
TAACTGGCTTACGAGAGCGCAGATAACAAATACTGTCTTCTAGTGTAGCCGTAGTTAGGCCACCACT
TCAAGAATCTGTAGCACCGCCTACATACTCGCTCTGCTAATCCTGTTACCAGTGGCTGCTGCCAGTGG
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TGAGCTATGAGAAAGCGCCACGCTTCCCGAAGGGAGAAAAGCGGACAGGTATCCGGTAAGCGGCAGGGT
CGGAACAGGAGAGCGCAGGAGGCTTCCAGGGGAAACGCCTGGTATCTTTATAGTCTGTGCGGTT
TCGCCACCTCTGACTTGAGCGTGCATTTTTGTGATGCTCGTCAGGGGGGCGGAGCCTATGGAAAAACGC
CAGCAACGCGGCCCTTTTTACGGTTCTTGGCCTTTTTGCTGACATGTTCTTCCCTGCGTTA
TCCCCTGATTCTGTGGATAACCGTATTACCGCCTTTGAGTGAGCTGATACCGCTCGCCGAGCCGAACG
ACCGAGCGCAGCGAGTCAAGTGGAGGAGGAAAGCGGAAGAGCGCCTGATGCGGTATTTTTCTCCTTACGCAT

CTGTGCGGTATTTACACCCGCATATATGGTGCCTCTCAGTACAATCTGCTCTGATGCCGCATAGTTAA
GCCAGTATACTCCGCTATCGCTACGTGACTGGGTTCATGGCTGCGCCCCGACACCCGCCAACACCCGCT
GACGCGCCCTGACGGGCTTGTCTGCTCCCGGCATCCGCTTACAGACAAGCTGTGACCGTCTCCGGGAGCT
GCATGTGTCAGAGGTTTTACCGTCATCACCGAAACGCGCGAGGCAGCTGCGGTAAAGCTCATCAGCGT
GGTCGTGAAGCGATTACAGATGTCTGCTGTTTCATCCGCGTCCAGCTCGTTGAGTTTTCTCCAGAAGCG
TTAATGTCTGGCTTCTGATAAAGCGGGCCATGTTAAGGGCGGTTTTTTCCTGTTTGGTCACTGATGCCT
CCGTGTAAGGGGGATTCTGTTTCATGGGGTAATGATACCGATGAAACGAGAGAGGATGCTCACGATA
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ACAGGGTAGCCAGCAGCATCCTGCGATGCAGATCCGGAACATAATGGTGCAGGGCGCTGACTTCCGCGT
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ACGTGGCTGGCCTGGTTTACCACGCGGGAAACGGTCTGATAAGAGACACCGGCATACTCTGCGACATCG
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AGATATACCATGGGACGAGCCATCATCATCATCACAGCAGCGGCTGGTGCCGCGCGGCAGCCAT
ATGGCTAGCGGGCCGACGACTC

Restriction enzyme for bead release: NotI

3270055_3270054pAff8c

Backbone 3270055

**CTCGAGGCGGCCGACGCGGACGTCAGGAGGTGCGCCGCTTCTGCACCGCGAGCTGGTGCATCCCGAC
GCCCTCAACCGCTTCGGCAAGACGGCGCTGCAGGTCATGATGTTTGGCAGCACCGCCATCGCCCTGGAGC
TGCTGAAGCAAGGTGCCAGCCCCAATGTCCAGGACACCTCCGGTACCAGTCCAGTCCATGACGCGGC
TCAGGATCGTTCTGGAC**

Insert 3270054pAff8c

**CATGACGCGAGGCTCAGGATCGTTCTGGACACCCTGAAGGTCCTAGTGGAGCACGGGGCTGATGTCAA
CGTGCCTGATGGCACCGGGCACTTCCAATCCATCTGGCAGTTCAAGAGGGTCACACTGCTGTGGTCCAG
CTTTCTGGCAGCTGAATCTGATCTCCATCGCAGGGACGCCAGGTAAGGCGCGCCCCACCGCTGAGCAAT
AACTAGCATAACCCCTTGGGGCCTCTAAACGGGTCTTGGGGGTTTTTTGCTGAAAGGAGGAACTATAT
CCGGATTGGCGAATGGGACGCGCCCTGTAGCGGCGCATTAAAGCGCGGGGTGTGGTGGTTACGCGCAG**

CGTGACCGCTACACTTGCCAGCGCCCTAGCGCCCGCTCCTTTCGCTTCTTCCCTTCTTTCTCGCCAG
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CGAGATCTCGATCCCGCGAAATTAATACGACTCACTATAGGGGAATTGTGAGCGGATAACAATTTCCCT
CTAGAAATAATTTGTTAACTTTAAGAAGGAGATATACCATGGGCAGCAGCCATCATCATCATCA
CAGCAGCGGCCTGGTGGCGCGCGCAGCCATATGGCTAGCGGGCCGACGACTC

Restriction enzyme for bead release: NotI

HerceptinCH_pcDNA3.3_HumiraVH

Backbone HerceptinCH_pcDNA3.3:

CTCAGGCTAGCACCAAGGGCCATCGGTCTTCCCCCTGGCACCCCTCCTCAAGAGCACCTCTGGGGG
ACAGCGGCCCTGGGCTGCCTGGTCAAGGACTACTTCCCCGAACCGGTGACGGTGTCTGGAACCTCAGGC
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TGGTGACCGTGCCTCCAGCAGCTTGGGCACCCAGACCTACATCTGCAACGTGAATCACAAGCCCAGCA
ACACCAAGGTGGACAAGAAAGTTGAGCCCAAATCTTGGGACAAAACCTCACACATGCCACCGTGGCCAG
CACCTGAACTCCTGGGGGACCGTCACTTCTCTTCCCCCAAACCCAAAGGACACCCCTCATGATCTC
CCGACCCCTGAGGTACATGCGTGGTGGTGGACGTGAGCCACGAAGACCCCTGAGGTCAAGTTCAACTG
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GGGATTTCCAAGTCTCCACCCATTGACGTCAATGGGAGTTTTGTTTTGGCACAAAATCAACGGGACT
TTCCAAAATGTGTAACAACCTCCGCCCCATTGACGCAAAATGGGCGGTAGGCGTGTACGGTGGGAGGTCT
ATATAAGCAGAGCTCGTTTGTGTAACCGTCAGATCGCCTGGAGACGCCATCCACGCTGTTTTGACCTCC
ATAGAAGACACCGGGACCGATCCAGCCTCCGGACTCTAGAGGATCGAACCTT

Insert HumiraVH

CAGCTCCGGACTCTAGAGGATCGAACCTTGAGGTACAGCTGGTTGAATCCGGTGGTGGACTGGTCC
AACCTGGGAGAAGCCTGAGACTGAGCTGTGCAGCATCAGGGTTTACGTTTCGATGACTACCGGATGCATT

GGGTGCGTCAGGCTCCAGGAAAAGGGCTGGAATGGGTAGTGCCATCACATGGAATAGCGGCCACATCG
ACTATGCCGATTTCAGTGGAAGGCCGGTTCACCATTTCCAGGGACAATGCCAAGAAGTCCCTGTATCTCC
AGATGAACAGCCTTAGAGCCGAGGATACTGCAGTGTACTACTGCGCCAAGGTCAGCTACCTGAGCACAG
CCAGTAGCCTCGACTATTGGGGCCAAGGCACATTGGTGACTGTGAGTTCGGCTAGCCGACTC

Restriction enzyme for bead release: NheI

HerceptinCH_pcDNA3.3_OmnitargVH

Backbone HerceptinCH_pcDNA3.3: Same as for HerceptinCH_pcDNA3.3_HumiraVH

Insert OmnitargVH:

CAGCCTCCGACTCTAGAGGATCGAACCCCTTGAGGTGCAACTGGTTCGAAAGTGGCGGAGGACTCGTGC
AGCCAGGAGGTTCTCTGCGACTGAGTTGTGCCGCTTCTGGCTTTACTTTACCGACTATAACGATGGATT
GGGTGAGACAGGCTCCCGGAAAGGCCTCGAATGGGTAGCTGACGTCAACCCTAATTCAGGCGGGTCAA
TCTACAATCAGAGGTTTAAAGGCCGTTTACCTTGAGCGTGGATCGCAGCAAGAACACACTCTATCTGC
AGATGAACAGCCTTAGAGCCGAAGATACTGCAGTCTATTACTGTGCCAGGAATCTGGGGCCAAGCTTCT
ACTTCGACTATTGGGGACAAGGCACCCCTGGTTACGGTGTCTCTGCTAGCCGACTC

Restriction enzyme for bead release: NheI

HerceptinCL_pOptivec_HumiraVL

Backbone - HerceptinCL_pOptivec

CTCGAGCGTACGGTGGCTGCACCATCTGTCTTCATCTTCCCGCCATCTGATGAGCAGTTGAAATCTGG
AACTGCCTCTGTTGTGTGCCTGCTGAATAACTTCTATCCCAGAGAGGCCAAAAGTACAGTGGAAGGTGGA
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CCATCAGGGCCTGAGTTCGCCCCTCACAAAGAGCTTCAACAGGGGAGAGTGTTAACTCGAGAAGGGTTG
GATCCCTACCGGTGCTGCGGCCGCGCAGTTAACGCCGCCCTCTCCCTCCCCCCCCCTAACGTTACTGG
CCGAAGCCGCTTGAATAAAGGCCGGTGTGCGTTTGTCTATATGTTATTTTCCACCATATTGCCGTCTTT
TGGCAATGTGAGGGCCCGGAAACCTGGCCCTGTCTTGTGACGAGCATTCTAGGGGTCTTTCCCCTCTC
GCCAAAGGAATGCAAGGTCTGTTGAATGTCGTGAAGGAAGCAGTTCCTCTGGAAGCTTCTTGAAGACA
AACAACGTCTGTAGCGACCCCTTTCAGGCAGCGGAACCCCCACCTGGCGACAGGTGCCTCTGCGGCCA
AAAGCCACGTGTATAAGATACACTGCAAAGGCCGCAACCCAGTGCCACGTTGTGAGTTGGATAGT
TGTGGAAGAGTCAAATGGCTCTCTCAAGCGTATTCACAAGGGGTGAAGGATGCCCAGAAGGTACC
CCATTGTATGGGATCTGATCTGGGGCCTCGGTACACATGCTTTACATGTGTTTGTAGTCGAGGTTAAAAA
ACGTCTAGGCCCCCGAACCACGGGGACGTGGTTTTCTTTGAAAAACACGATGATAATATGGCCACAA
GATCTGCCACCATGGTTCGACCATTGAACTGCATCGTCGCCGTGTCCAAAATATGGGGATTGGCAAGA
ACGGAGACCTACCCTGGCCTCCGCTCAGGAACGAGTTCAGTACTTCAAAGAATGACCACAACCTCTT
CAGTGGAAGTAAACAGAATCTGGTGATTATGGGTAGGAAAACCTGGTTCTCCATTCTGAGAAGAAT
CGACTTTTAAAGGACAGAATTAATATAGTTCTCAGTAGAGAACTCAAAGAACCACCAGGAGGCTCAT
TTTCTTGCCAAAAGTTTGGATGATGCCTTAAAGACTTATTGAACAACCGGAATTGGCAAGTAAAGTAGA
CATGGTTTTGGATAGTCGGAGGCAGTCTGTTTACCAGGAAGCCATGAATCAACCAGGCCACCTCAGACT
CTTTGTGACAAGGATCATGCAGGAATTTGAAAGTGACACGTTTTTCCAGAAATTGATTTGGGGAAAT
ATAAACTTCTCCAGAATACCCAGGCGTCTCTCTGAGGTCCAGGAGGAAAAAGGCATCAAGTATAAGT
TTGAAGTCTACGAGAAGAAAGACTAAAACCGGTTAGTAATGAGTTTAAACGGGGGAGGCTAACTGAAA
CACGGAAGGAGACAATACCGGAAGGAACCCGCGCTATGACGGCAATAAAAAGACAGAATAAAACGCAC
GGGTGTTGGGTCGTTTGTTCATAAACCGGGGTTCCGGTCCCAGGGCTGGCACTCTGTGATACCCACC
GAGACCCATTGGGGCCAATACGCCCGCTTTCTTCTTTTCCCCACCCACCCCAAGTTCGGGTGAA
GGCCAGGGCTCGCAGCCAACGTCGGGGCGGCAGGCCCTGCCATTACCGTCGACCTCTAGCTAGAGCTTG
GCGTAATCATGGTCATAGCTGTTTCTGTGTGAAATTTGTTATCCGCTCACAATTCACACAACATACGA
GCCGGAAGCATAAAGTGTAAGCCTGGGGTGCCTAATGAGTGAGCTAACTCACATTAATTGCGTTGCGC
TCACTGCCCGCTTTCCAGTCGGGAAACCTGTCGTGCCAGCTGCATTAATGAATCGGCCAACGCGGGG
AGAGGCGTTTTGCGTATTGGGCGCTTTCGCTTCTCGCTCACTGACTCGCTGCGCTCGGTGCTTCCGC
TGCGGCGAGCGGTATCAGCTCACTCAAAGGCGGTAATACGGTTATCCACAGAATCAGGGGATAACGCAG
GAAAGAACATGTGAGCAAAAGGCCAGCAAAAGGCCAGGAACCGTAAAAAGGCCGCTTGTGGCGTTT
TTCCATAGGCTCCGCCCCCTGACGAGCATCACAAAATCGACGCTCAAGTCAGAGGTGGCGAAACCCG
ACAGGACTATAAAGATACCAGGCGTTTCCCCTGGAAGCTCCCTCGTGCGCTCTCTGTTCCGACCCTGC

CGCTTACCGGATACCTGTCCGCTTTCTCCCTTCGGGAAGCGTGGCGCTTTCTCATAGCTCACGCTGTAG
GTATCTCAGTTCGGTGTAGGTCGTTTCGCTCCAAGCTGGGCTGTGTGCACGAACCCCGTTCAGCCCGAC
CGCTGCGCTTATCCGGTAACTATCGTCTTGAGTCCAACCCGGTAAGACACGACTTATCGCCACTGGCA
GCAGCCACTGGTAACAGGATTAGCAGAGCGAGGTATGTAGGCGGTGCTACAGAGTCTTGAAGTGGTG
GCCTAACTACGGCTACACTAGAAGAACAGTATTTGGTATCTGCGCTCTGCTGAAGCCAGTTACCTTCGG
AAAAAGAGTTGGTAGCTCTTGATCCGGCAAACAAACCACCGCTGGTAGCGGTTTTTTTTGTTTGCAAGCA
GCAGATTACGCGCAGAAAAAAGGATCTCAAGAAGATCCTTTGATCTTTTCTACGGGGTCTGACGCTCA
GTGGAACGAAAACCTCACGTTAAGGGATTTTGGTCATGAGATTATCAAAAAGGATCTTCACCTAGATCC
TTTTAAATTAATAAATGAAGTTTTAAATCAATCTAAAGTATATATGAGTAAACTTTGGTCTGACAGTTAC
CAATGCTTAATCAGTGAGGCACCTATCTCAGCGATCTGTCTATTTTCGTTTCATCCATAGTTGCCTGACTC
CCCCTCGTGTAGATAACTACGATACGGGAGGGCTTACCATCTGGCCCCAGTGTGCAATGATACCGCGA
GACCCACGCTCACCGGCTCCAGATTTATCAGCAATAAACCCAGCCAGCCGGAAGGGCCGAGCGCAGAAGT
GGTCTGCAACTTTATCCGCTCCATCCAGTCTATTAATTGTTGCCGGGAAGCTAGAGTAAAGTAGTTTCG
CCAGTTAATAGTTTTCGCAACGTTGTTGCCATTGCTACAGGCATCGTGGTGTACGCTCGTCTGTTGGT
ATGGCTTCATTACGCTCCGTTTCCCAACGATCAAGGCGAGTTACATGATCCCCATGTTGTGCAAAAAA
GCGGTTAGCTCCTTCGGTCTCCGATCGTTGTCAGAAGTAAGTTGGCCGAGTGTATCACTCATGGTT
ATGGCAGCACTGCATAATTCTCTTACTGTCATGCCATCCGTAAGATGCTTTTCTGTGACTGGTGTAGTAC
TCAACCAAGTCATTCTGAGAATAGTGTATGCGGCGACCGAGTTGCTCTTGCCCGGCGTCAATACGGGAT
AATACCGCGCCACATAGCAGAACTTTAAAAGTGCTCATCATTGAAAACGTTCTTCGGGGCGAAAACCTC
TCAAGGATCTTACCGCTGTTGAGATCCAGTTCGATGTAACCCACTCGTGCACCCAACTGATCTTCAGCA
TCTTTTACTTTTACCAGCGTTTCTGGGTGAGCAAAAACAGGAAGGCAAAAATGCCGCAAAAAAGGGAAT
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TTATTGTCTCATGAGCGGATACATATTTGAATGTATTTAGAAAAATAAACAAATAGGGGTTCCGCGCA
CATTTCCCGAAAAGTGCCACCTGACGTCGACGGATCGGGAGATCAGTTGACATTGATTATTGACTAGT
TATTAATAGTAATCAATTACGGGGTCATTAGTTCATAGCCCATATATGGAGTTCGCGGTTACATAACTT
ACGGTAAATGGCCCGCTGGCTGACCGCCCAACGACCCCGCCCATTTGACGTCAATAATGACGTATGTT
CCCATAGTAACGCCAATAGGGACTTTCCATTGACGTCAATGGGTGGAGTATTTACGGTAAACTGCCAC
TTGGCAGTACATCAAGTGTATCATATGCCAAGTACGCCCCCTATTGACGTCAATGACGGTAAATGGCCC
GCCTGGCATTATGCCAGTACATGACCTTATGGGACTTTCTACTTGGCAGTACATCTACGTATTAGTC
ATCGCTATTACCATGGTGATGCGGTTTTGGCAGTACATCAATGGGCGTGGATAGCGGTTTACTCACGG
GGATTTCCAAGTCTCCACCCCATTTGACGTCAATGGGAGTTTTGTTTTGGCACAAAATCAACGGGACTTT
CCAAAATGTCGTAACAACTCCGCCCCATTGACGCAAAATGGGCGGTAGGCGTGTACGGTGGGAGGTCTAT
ATAAGCAGAGCTCGTTTTAGTGAACCGTCAGATCGCCTGGAGACGCCATCCACGCTGTTTTGACCTCCAT
AGAAGACACCGGGACCGATCCAGCCT**CCGGACTCTAGAGGATCCAACCCTT**

Insert HumiraVL:

AGCCTCCGGACTCTAGAGGATCCAACCCTTGACATCCAGATGACACAGTCTCCCTCTTCTCTGAGCGCT
AGCGTCGGGGATCGCGTGACCATTACGTGTAGGGCCTCCAGGGGATTTCGCAACTACCTCGCGTGGTAT
CAGCAGAAGCCTGGGAAAGCTCCAAGCTCTTGATCTATGCCGCTTCTACTCTGCAATCTGGCGTTCCC
AGTCGGTTTTTCCGGATCAGGCTCAGGAACCGATTTCACTCTGACCATATCCAGCCTTCAGCCAGAGGAC
GTAGCAACATACTACTGCCAGAGGTATAATCGAGCTCCGTACACCTTTGGCCAAGGGACCAAAGTGGGA
GATAAAGCGTACGCGACTC

Restriction enzyme for bead release: BsiWI

HerceptinCL_pOptivec_OmnitargVL

Backbone HerceptinCL_pOptivec: Same as for HerceptinCL_pOptivec_HumiraVL

Insert OmnitargVL:

CAGCTCCGGACTCTAGAGGATCCAACCCTTGACATCCAGATGACACAGAGCCCTTCTAGCTTGTCCGC
AAGTGTCCGAGATCGCGTAACCATTACTTGCAAAGCGAGTCAGGATGTGAGCATTGGCGTTGCTTGGTA
TCAGCAGAAAACCCGGGAAAGCCCTAAGCTGCTGATATACTCCGCAAGCTACCGGTATACAGGTGTTCC
ATCCCGTTTTTTCAGGGTCCGGATCTGGGACTGACTTCACACTGACTATTAGCTCCCTTCAGCCCGAGGA
CTTTGCCACCTACTATTGCCAGCAATACTACATCTACCCGTATACCTTCGGGCAAGGGACCAAAGTGG
AGATCAAGCGTACGCGACTC

Restriction enzyme for bead release: NheI

pcDNA3.3_HER2 signal peptide

Backbone pcDNA3.3:

CTCAGAGGCGCGCCATAACCCTACCGGTTAGTAATGAGTTTAAACGGGGGAGGCTAACTGAAACACG
GAAGGAGACAATACCGGAAGGAACCCGCGCTATGACGGCAATAAAAAGACAGAATAAAACGCACGGGT
GTTGGGTGTTTTGTTTCATAAACCGGGGTTTCGGTCCCAGGGCTGGCACTCTGTGATAACCCACCGAGA
CCCCATTGGGGCCAATACGCCCGCTTTCTTCCTTTTCCCCACCCACCCCAAGTTCGGGTGAAGGCC
CAGGGCTCGCAGCCAACGTGGGGCGGCAGGCCCTGCCATAGCAGATCTGCGCAGCTGGGGCTCTAGGG
GGTATCCCCACGCGCCCTGTAGCGGCGCATTAAAGCGCGGCGGGTGTGGTGGTTACGCGCAGCGTGACCG
CTACACTTGCCAGCGCCCTAGCGCCCGCTCCTTTTCGCTTTCTTCCCTTCTTTCTCGCCACGTTCCGCCGC
TTTTCCCGTCAAGCTCTAAATCGGGGCATCCCTTTAGGGTTCGATTTAGTGCTTTACGGCACCTCGACC
CCAAAAAAGTTGATTAGGGTGATGGTTCACGTAGTGGGCCATCGCCCTGATAGACGGTTTTTTCGCCCTT
TGACGTTGGAGTCCACGTTCTTTAATAGTGGACTCTTGTTCCAAACTGGAACAACACTCAACCCTATCT
CGGTCTATTCTTTTATTATAAGGGATTTTGGGGATTTCCGCCCTATTGGTTAAAAAATGAGCTGATTT
AACAAAAATTTAACGCGAATTAATTCTGTGGAATGTGTGTCAGTTAGGGTGTGGAAAGTCCCCAGGCT
CCCCAGCAGGCAGAAGTATGCAAAGCATGCATCTCAATTAGTCAGCAACCAGGTGTGGAAAGTCCCCAG
GCTCCCCAGCAGGCAGAAGTATGCAAAGCATGCATCTCAATTAGTCAGCAACCATAGTCCCGCCCTAA
CTCCGCCCATCCCGCCCTAACTCCGCCAGTTCCGCCCATTTCTCCGCCCATGGCTGACTAATTTTTTTT
TATTTATGCAGAGGCCGAGGCCGCTCTGCCTCTGAGCTATTCCAGAAGTAGTGAGGAGGCTTTTTTTGG
AGGCTAGGCTTTTGAAAAAGTCCCGGGAGCTTGTATATCCATTTTCGGATCTGATCAAGAGACAGG
ATGAGGATCGTTTTGCGATGATTGAACAAGATGGATTGCACGCAGGTTCTCCGGCCGCTTGGGTGGAGAG
GCTATTCCGGCTATGACTGGGCACAACAGACAATCGGCTGCTCTGATGCCGCCGTGTTCCGGCTGTCAGC
GCAGGGGCGCCCGTTCTTTTTGTCAAGACCGACCTGTCCGGTGCCTGAATGAACTGCAGGACGAGGC
AGCGCGGCTATCGTGGCTGGCCACGACGGGCGTTCCCTTGCAGCTGTGCTCGACGTTGTCACTGAAGC
GGGAAGGACTGGCTGCTATTGGGCGAAGTGCCGGGGCAGGATCTCCTGTCACTCACCTTGCTCCTGC
CGAGAAAGTATCCATCATGGCTGATGCAATGCGGCGGCTGCATACGCTTGATCCGGCTACCTGCCATT
CGACCACCAAGCGAAACATCGCATCGAGCGAGCACGTAATCGGATGGAAGCCGGTCTTGTGATCAGGA
TGATCTGGACGAAGAGCATCAGGGGCTCGCGCCAGCCGAACTGTTCCGCAAGGCTCAAGGCGCGCATGCC
CGACGGCGAGGATCTCGTCTGACCCATGGCGATGCCTGCTTCCGAATATCATGGTGGAAAAATGGCCG
CTTTTCTGGATTTCATCGACTGTGGCCGGCTGGGTGTGGCGGACCGCTATCAGGACATAGCGTTGGCTAC
CCGTGATATTGCTGAAGAGCTTGGCGGCGAATGGGCTGACCGCTTCTCCTGCTGCTTTACGGTATCGCCG
TCCCGATTTCGACGCGCATCGCCTTCTATCGCCTTCTTGACGAGTTCTTCTGAGCGGACTCTGGGGTTCC
CGAAATGACCGACCAAGCGACGCCAACCTGCCATCACAGATTTTCGATTCCACCGCCGCTTCTATGA
AAGGTTGGGCTTCGGAATCGTTTTCCGGGACCGCGGCTGGATGATCCTCCAGCGCGGGGATCTCATGCT
GGAGTTCTTCGCCCAACTTGTTTATTGAGCTTATAATGGTTACAAATAAAGCAATAGCATCAC
AAATTTACAAAATAAAGCATTTTTTCACTGCATCTAGTTGTGGTTTGTCCAAACTCATCAATGTATC
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GTGAAATTGTTATCCGCTCACAAATCCACACAACATACGAGCCGGAAGCATAAAGTGTAAGCCTGGGG
TGCCTAATGAGTGAGCTAACTCACATTAATTGCGTTGCGCTCACTGCCCGCTTTCCAGTCGGGAAACCT
GTCGTGCCAGCTGCATTAATGAATCGGCCAACGCGCGGGGAGAGGCGGTTTGCCTATTGGGCGCTCTTC
CGCTTCTCGCTCACTGACTCGCTGCGCTCGGTCTCGGCTGCGGCGAGCGGTATCAGCTCACTCAAAG
GCGGTAATACGGTTATCCACAGAATCAGGGGATAACGCAGGAAAGAATGTGAGCAAAAGGCCAGCA
AAAGGCCAGGAACCGTAAAAAGGCCGCTTGTGGCGTTTTTCCATAGGCTCCGCCCCCTGACGAGCA
TCACAAAAATCGACGCTCAAGTCAGAGGTGGCGAAACCCGACAGGACTATAAAGATACCAGGCGTTTCC
CCCTGGAAGCTCCCTCGTGCCTCTCCTGTTCCGACCTGCCGCTTACCGGATACCTGTCCGCTTTCTC
CCTTCGGGAAGCGTGGCGCTTTCTCAATGCTCACGCTGTAGGTATCTCAGTTCCGGTGTAGGTCGTTCCG
TCCAAGCTGGGCTGTGTGCACGAACCCCGTTCCAGCCGACCGCTGCGCTTATCCGGTAACTATCGTC
TTGAGTCCAACCCGTAAGACACGACTTATCGCCACTGGCAGCAGCCACTGGTAACAGGATTAGCAGAG
CGAGGTATGTAGGCGGTGCTACAGAGTTCTTGAAGTGGTGGCCTAACTACGGCTACACTAGAAGGACAG
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AACAAACCACCGCTGGTAGCGGTGGTTTTTTTTGTTTGAAGCAGCAGATTACGCGCAGAAAAAAGGAT
CTCAAGAAGATCCTTTGATCTTTTCTACGGGGTCTGACGCTCAGTGGAAACGAAAACTCACGTTAAGGA
TTTTGGTCATGAGATTATCAAAAAGGATCTTACCTAGATCCTTTTAAATTAATAAAGTAAAA
TCAATCTAAAGTATATATGAGTAACTTGGTCTGACAGTTACCAATGCTTAATCAGTGAGGCACCTATC
TCAGCGATCTGTCTATTTTCGTTTCATCCATAGTTGCCTGACTCCCGTCTGTAGATAACTACGATACGG
GAGGGCTTACCATCTGGCCCCAGTGCTGCAATGATACCGCGAGACCCACGCTCACCGGCTCCAGATTTA
TCAGCAATAAACCAGCCAGCCGGAAGGGCCGAGCGCAGAAGTGGTCTGCAACTTTATCCGCTCCATC
CAGTCTATTAATTGTTGCCGGGAAGCTAGAGTAAGTAGTTCGCCAGTTAATAGTTTGCACAACGTTGTT
GCCATTGCTACAGGCATCGTGGTGTACGCTCGTCTGTTGGTATGGCTTCATTCAGCTCCGGTTCCCAAC

GATCAAGGCGAGTTACATGATCCCCATGTTGTGCAAAAAAGCGGTTAGCTCCTTCGGTCTCCGATCG
TTGTCAGAAGTAAGTTGGCCGAGTGTATCACTCATGGTTATGGCAGCACTGCATAATTCTCTTACTG
TCATGCCATCCGTAAGATGCTTTTCTGTGACTGGTGAAGTCAACCAAGTCATTCTGAGAATAGTGTA
TGCGGCGACCGAGTTGCTCTTGCCCGCGTCAATACGGGATAATACCGCGCCACATAGCAGAACTTTAA
AAGTGCTCATCATTGAAAAACGTTCTTCGGGGCGAAAACTCTCAAGGATCTTACCCTGTTGAGATCCA
GTTTCGATGTAACCCACTCGTGCACCCAAGTATCTTCAGCATCTTTTACTTTTACCAGCGTTTCTGGGTG
AGCAAAAACAGGAAGGCAAAATGCCGCAAAAAAGGGAATAAGGGCGACACGGAAATGTTGAATACTCA
TACTCTTCCTTTTTCAATATTATTGAAGCATTATCAGGGTTATTGTCTCATGAGCGGATACATATTG
AATGTATTTAGAAAAATAAACAAATAGGGGTTCCGCGCACATTTCCCGAAAAGTGCCACCTGACGTCG
ACGGATCGGGAGATCTCCCGATCCCCTATGGTGCAGTCTCAGTACAATCTGCTCTGATGCCGCATAGTT
AAGCCAGTATCTGCTCCCTGCTTGTGTGTTGGAGGTGCGTGAAGTAGTGCAGGAGCAAAAATTAAGCTAC
AACAAAGCAAGGCTTGACCGACAATTGCATGAAGAATCTGCTTAGGGTTAGGCGTTTTGCGCTGCTTCG
CGATGTACGGGCCAGATATACGCGTTGACATTGATTATTGACTAGTTATTAATAGTAATCAATTACGG
GGTCATTAGTTCATAGCCCATATATGGAGTTCGCGTTACATAACTTACGGTAAATGGCCCGCCTGGCT
GACCGCCAACGACCCCGCCATTGACGTCAATAATGACGTATGTTCCCATAGTAACGCCAATAGGGA
CTTTCCATTGACGTCAATGGGTGGAGTATTTACGGTAAACTGCCCACTTGGCAGTACATCAAGTGTATC
ATATGCCAAGTACGCCCCCTATTGACGTCAATGACGGTAAATGGCCCGCCTGGCATTATGCCCAGTACA
TGACCTTATGGGACTTTCTACTTGGCAGTACATCTACGTATTAGTCATCGCTATTACCATGGTGATGC
GGTTTTGGCAGTACATCAATGGGCGTGGATAGCGGTTTACTCACGGGGATTTCCAAGTCTCCACCCCA
TTGACGTCAATGGGAGTTTGTTTTGGCACAAAATCAACGGGACTTTCCAAAATGTCGTAACAACCTCCG
CCCCATTGACGCAATGGGCGGTAGGCGTGTACGGTGGGAGGTCTATATAAGCAGAGCTCGTTTAGTGA
ACCGTCAGATCGCCTGGAGACGCCATCCACGCTGTTTTGACCTCCATAGAAGACACCGGGACCGATCCA
GCCTCCGGACTCTAGAGGATCGAACCTT

Insert oligo HER2 signal peptide:

AGCCTCCGGACTCTAGAGGATCGAACCTTATGGAGCTGGCGGCCCTTGTGCCGCTGGGGGCTCCTCCTCG
CTCTCTTGCCACCTGGAGCAGCTAGTGGCGGCCACGACTC

Restriction enzyme for bead release: *AscI*

pHisZ_VEGF_eGFP

Backbone pHisZ: same as for pHisZ_GFP11

Insert 1 VEGF:

**CGGGTACCGGTCCGACCGGTGGATCCGGTGAAGGAGGAGGGCAGAATCATCACGAAGTGGTGAAGTT
CATGGATGTCTATCAGCGCAGCTACTGCCATCCAATCGAGACCCTGGTGGACATCTTCCAGGAGTACCC
TGATGAGATCGAGTACATCTTCAAGCCATCCTGTGTGCCCTGATGCGATGCGGGGGCTGCTGCAATGA
CGAGGGCCTGGAGTGTGTGCCACTGAGGAGTCCAACATCACCATGCAGATTATGCGGATCAAACCTCA
CCAAGGCCAGCACATAGGAGAGATGAGCTTCTACAGCACAAACAATGTGAATGCAGACCAAAGAAAG
ATAGAGCAAGACAAGAAAAATGTGACAAGCCGAGGCGGGGCTCAGGATCG**

Insert2 eGFP:

**TGTGACAAGCCGAGGCGGGGCTCAGGATCGATGGTGAAGGCGAGGAGCTGTTACCGGGGTGG
TGCCCATCCTGGTTCGAGCTGGACGGCGACGTAACGGCCACAAGTTTCAAGCGTGTCCGGCGAGGGCGAGG
GCGATGCCACCTACGGCAAGCTGACCCTGAAGTTTCACTCTGCACCACCGGCAAGCTGCCCGTGCCTGGCC
CACCTCGTGACCACCTGACCTACGGCGTGCAGTGTTCAGCCGCTACCCCGACCACATGAAGCAGCAC
GACTTCTTCAAGTCCGCCATGCCCGAAGGCTACGTCCAGGAGCGCACCATCTTCTTCAAGGACGACGGC
AACTACAAGACCCGCGCCGAGGTGAAGTTTCGAGGGCGACACCCTGGTGAACCGCATCGAGCTGAAGGGC
ATCGACTTCAAGGAGGACGGCAACATCCTGGGGACAAGCTGGAGTACAACATAACAGCCACAACGTC
TATATCATGGCCGACAAGCAGAAGAACGGCATCAAGGTGAAGTTCAAGATCCGCCACAACATCGAGGAC
GGCAGCGTGCAGCTCGCCGACCACTACCAGCAGAACACCCCCATCGGGCGACGGCCCCGTGCTGCTGCCCG
ACAACCACTACCTGAGCACCCAGTCCGCCCTGAGCAAAGACCCCAACGAGAAGCGCGATCATATGGTCC
TGCTGGAGTTCGTGACCGCCGCGGGATCACTCTGGCATGGACGAGCTGTACAAGGCGGCCGACCA
CTC**

Restriction enzyme for bead release: *NotI*

pHisZ_VEGF_mCherry

Backbone pHisZ: same as for pHisZ_GFP11

Insert1 VEGF: Same as for pHisZ_VEGF_eGFP

Insert2 mCherry:

**TGTGACAAGCCGAGGGCGGGCTCAGGATCGATGGTGAGCAAGGGCGAGGAGGATAACATGGCCATCA
TCAAGGAGTTCATGCGCTTCAAGGTGCACATGGAGGGCTCCGTGAACGGCCACGAGTTCGAGATCGAGG
GCGAGGGCGAGGGCCGCCCTACGAGGGCAGCCAGACCGCCAAGCTGAAGGTGACCAAGGGTGGCCCCC
TGCCCTTCGCTGGGACATCCTGTCCCCTCAGTTCATGTACGGCTCCAAGGCCTACGTGAAGCACCCCGC
CGACATCCCCGACTACTTGAAGCTGTCTTCCCCGAGGGCTTCAAGTGGGAGCGCGTGATGAACTTCGA
GGACGGCGGGCGTGGTGACCGTGACCCAGGACTCCTCCCTGCAGGACGGCGAGTTCATCTACAAGGTGAA
GCTGCGCGGCACCAACTTCCCCTCCGACGGCCCCGTAATGCAGAAGAAGACCATGGGCTGGGAGGCCTC
CTCCGAGCGGATGTACCCCGAGGACGGCGCCCTGAAGGGCGAGATCAAGCAGAGGCTGAAGCTGAAGGA
CGGCGGCCACTACGACGCTGAGGTCAAGACCACCTACAAGGCCAAGAAGCCCGTGCAGCTGCCCGGCGC
CTACAACGTCAACATCAAGTTGGACATCACCTCCCACAACGAGGACTACACCATCGTGGAACAGTACGA
ACGCGCCGAGGGCCGCCACTCCACCGGCGGCATGGACGAGCTGTACAAGGGCGGCCGACGACTC**

Restriction enzyme for bead release: NotI

pHisZ_mCherry_EGFP

Backbone pHisZ: same as for pHisZ_GFP11

Insert1 mCherry:

**CGGGTACCGGTCGCCACCGGTGGATCCGGTATGGTGAGCAAGGGCGAGGAGGATAACATGGCCATCAT
CAAGGAGTTCATGCGCTTCAAGGTGCACATGGAGGGCTCCGTGAACGGCCACGAGTTCGAGATCGAGGG
CGAGGGCGAGGGCCGCCCTACGAGGGCAGCCAGACCGCCAAGCTGAAGGTGACCAAGGGTGGCCCCCT
GCCCTTCGCTGGGACATCCTGTCCCCTCAGTTCATGTACGGCTCCAAGGCCTACGTGAAGCACCCCGCC
GACATCCCCGACTACTTGAAGCTGTCTTCCCCGAGGGCTTCAAGTGGGAGCGCGTGATGAACTTCGAG
GACGGCGGGCGTGGTGACCGTGACCCAGGACTCCTCCCTGCAGGACGGCGAGTTCATCTACAAGGTGAAG
CTGCGCGGCACCAACTTCCCCTCCGACGGCCCCGTAATGCAGAAGAAGACCATGGGCTGGGAGGCCTCCT
CCGAGCGGATGTACCCCGAGGACGGCGCCCTGAAGGGCGAGATCAAGCAGAGGCTGAAGCTGAAGGACG
GCGGCCACTACGACGCTGAGGTCAAGACCACCTACAAGGCCAAGAAGCCCGTGCAGCTGCCCGGCGCCT
ACAACGTCAACATCAAGTTGGACATCACCTCCCACAACGAGGACTACACCATCGTGGAACAGTACGAAC
GCGCCGAGGGCCGCCACTCCACCGGCGGCATGGACGAGCTGTACAAGGGCTCAGGATCG**

Insert2 eGFP:

**ATGGACGAGCTGTACAAGGGCTCAGGATCGATGGTGAGCAAGGGCGAGGAGCTGTTACCGGGGTGG
TGCCCATCCTGGTTCGAGCTGGACGGCGACGTAACCGCCACAAGTTCAGCGTGTCCGGCGAGGGCGAGG
GCGATGCCACCTACGGCAAGCTGACCCTGAAGTTCATCTGCACCACCGGCAAGCTGCCCGTGCCTGGCC
CACCTCGTGACCACCTGACCTACGGCGTGAGTGTTCAGCCGCTACCCCGACCACATGAAGCAGCAC
GACTTCTTCAAGTCCGCCATGCCCCAAGGCTACGTCCAGGAGCGCACCATCTTCTTCAAGGACGACGGC
AACTACAAGACCCGCGCCGAGGTGAAGTTCGAGGGCGACACCCTGGTGAACCGCATCGAGCTGAAGGGC
ATCGACTTCAAGGAGGACGGCAACATCCTGGGGCACAAGCTGGAGTACAACACTACAACAGCCACAACGTC
TATATCATGGCCGACAAGCAGAAGAACGGCATCAAGGTGAAGTTCAGATCCGCCACAACATCGAGGAC
GGCAGCGTGCAGCTCGCCGACCACTACCAGCAGAACACCCCATCGGCGACGGCCCCGTGCTGCTGCCCG
ACAACCACTACCTGAGCACCCAGTCCGCCCTGAGCAAAGACCCCAACGAGAAGCGCGATCACATGGTCC
TGCTGGAGTTCGTGACCGCCCGGGGATCACTCTCGGCATGGACGAGCTGTACAAGGGCGGCCGACGCA
CTC**

Restriction enzyme for bead release: NotI

pcDNA3.3_HerceptinHeavy_GFP11

Backbone pcDNA3.3_HerceptinHeavy:

**CTCGAGGGCGCGCCATAACTCGAGAAGGGTTTCGATCCCTACCGGTTAGTAATGAGTTTAAACGGGGGA
GGCTAACTGAAACACGGAAGGAGACAATACCGGAAGGAACCCGCGCTATGACGGCAATAAAAAGACAG**

AATAAAACGCACGGGTGTTGGGTCGTTTGTTCATAAACGCGGGGTTCCGGTCCCAGGGCTGGCACTCTGT
CGATACCCACCGAGACCCCATTTGGGGCCAATACGCCCGGTTTCTTCCTTTTCCCAACCCACCCCA
AGTTCCGGGTGAAGGCCAGGGCTCGCAGCCAACGTCGGGGCGGCAGGCCCTGCCATAGCAGATCTGCGC
AGCTGGGGCTCTAGGGGGTATCCCACGCGCCCTGTAGCGGCGCATTAAAGCGCGGCGGGTGTGGTGGTT
ACGCGCAGCGTGACCGCTACACTTGCCAGCGCCCTAGCGCCCGCTCCTTTTCGCTTCTTCCCTTCTTTC
TCGCCACGTTTCGCGGGCTTTCCCGTCAAGCTCTAAATCGGGGCATCCCTTTAGGGTTCCGATTTAGTGC
TTTACGGCACCTCGACCCCAAAAAAATTGATTAGGGTGTAGGTTTACGTAAGTGGGCCATCGCCCTGATA
GACGGTTTTTCGCCCTTTGACGTTGGAGTCCACGTTCTTTAATAGTGGACTCTTGTTCAAAACCTGGAAC
AACACTCAACCCTATCTCGGTCTATTCTTTTGTATTTATAAGGGATTTTGGGGATTTCCGCCCTATTGGTT
AAAAAATGAGCTGATTTAACAAAAATTTAACGCGAATTAATTCTGTGGAATGTGTGTCAGTTAGGGTG
TGGAAAAGTCCCAGGCTCCCAGCAGGCAGAAAGTATGCAAAGCATGCATCTCAATTAGTCAGCAACCAG
GTGTGAAAAGTCCCAGGCTCCCAGCAGGCAGAAAGTATGCAAAGCATGCATCTCAATTAGTCAGCAAC
CATAGTCCCGCCCTAACTCCGCCATCCCGCCCTAACTCCGCCAGTTCGCCCTTCTCCGCCCATG
GCTGACTAATTTTTTTTATTTATGCAGAGGCCGAGGCCGCTCTGCCTCTGAGCTATTCCAGAAGTAGT
GAGGAGGCTTTTTTGGAGGCCTAGGCTTTTGCAAAAAGCTCCCGGGAGCTTGTATATCCATTTTCGGAT
CTGATCAAGAGACAGGATGAGGATCGTTTCGCATGATTGAACAAGATGGATTGCACGCAGGTTCTCCGG
CCGCTTGGGTGGAGAGGCTATTCCGGCTATGACTGGGCACAACAGACAATCGGCTGCTCTGATGCCCGG
TGTTCCGGCTGTACGCGCAGGGGCGCCCGTTCTTTTTGTCAAGACCGACCTGTCCGGTGCCTGAATG
AACTGCAGGACGAGGCAGCGCGGCTATCGTGGCTGGCCACGACGGGCGTTTCTTGGCAGCTGTGCTCG
ACGTTGTCACTGAAGCGGAAGGGACTGGCTGCTATTTGGGCGAAGTCCCGGGGAGGATCTCCTGTGAT
CTCACCTTGCTCCTGCCGAGAAAGTATCCATCATGGCTGATGCAATGCGGCGGCTGCATACGCTTGATC
CGGCTACCTGCCATTCCGACCAAGCGAAACATCGCATCGAGCGAGCACGTAAGTCCGATGGAAGCCG
GTCTTGTGATCAGGATGATCTGGACGAAGAGCATCAGGGGCTCGCGCCAGCCGAAGTTCGCCAGGC
TCAAGGCGCGCATGCCCGACGGCGAGGATCTCGTGTGACCCATGGCGATGCCTGCTTCCGAATATCA
TGGTGAAAAATGGCCGCTTTTCTGGATTTCATCGACTGTGGCCGGCTGGGTGTGGCGGACCGCTATCAGG
ACATAGCGTTGGCTACCCGTGATATTGCTGCAAGAGCTTGGCGGCGAATGGGCTGACCGCTTCTCTGTG
TTTACGGTATCGCCGCTCCCGATTCCGACGCAATCGCCTTCTATCGCCTTCTTACGAGTTCTTCTGAGC
GGGACTCTGGGGTTTCGCGAAATGACCGACCAAGCGACGCCCAACCTGCCATCACGAGATTTTCGATTTCA
CCGCCGCTTCTATGAAAGGTTGGGCTTCCGAATCGTTTTCCGGGACGCGGCTGGATGATCCTCCAGC
GCGGGGATCTCATGCTGGAGTTCTTCGCCACCCCAACTTGTATTATGACGCTTATAATGGTTACAAAT
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GTCTTCTCATGCTCCGTGATGCATGAGGCTCTGCACAACCACTACACGCAGAAGAGCCTCTCCCTGTC
CCCGGGTAAA

Insert GFP 11 oligo:

CAGAAGAGCCTCTCCCTGTCCCCGGTAAACGGGTACCGGTGCCACCGGTGGATCCGGTCTGACCAC
ATGGTCTTTCATGAGTACGTAAATGCTGCTGGGATTACAGCAGCTGCATAAGGCGCGCCCGACTC

Restriction enzyme for bead release: *AscI*

pOptivec_HerceptinLight_GFP11

Backbone pOptivec_HerceptinLight:

CTCGAGGGGGCGCCATAACTCGAGAAGGGTTGGATCCCTACCGGTGCTGCGGCCGCGCAGTTAACGCC
GCCCCCTCTCCCTCCCCCCCCCTAACGTTACTGGCCGAAGCCGCTTGAATAAGGCCGGTGTGCGTTTGT
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ACTCCCAGGAGAGTGTACAGAGCAGGACAGCAAGGACAGCACCTACAGCCTCAGCAGCACCTGACGC
TGAGCAAAGCAGACTACGAGAAACACAAAGTCTACGCCTGCGAAGTCACCCATCAGGGCTGAGTTCGC
CCGTCAAAAGAGCTTCAACAGGGGAGAGTGT

Insert GFP11 oligo:

GTCACAAAGAGCTTCAACAGGGGAGAGTGTGGGTACCGGTGCCACCGGTGGATCCGGTCGTGACCAC
ATGGTCCTTCATGAGTACGTAAATGCTGCTGGGATTACAGCAGCTGCATAAGGCGCGCCGACTC

Restriction enzyme for bead release: *AscI*

pHisZ_VEGF_AG_mCherry

Backbone pHisZ: same as for pHisZ_GFP11

Insert1 VEGF: Same as for pHisZ_VEGF_eGFP

Insert2 AG:

**TGTGACAAGCCGAGGGCGGGGCTCAGGATCGGCGCAACACGATGAAGCTCAACAAAATGCTTTTTATCA
AGTCTTAAATATGCCTAACTTAAATGCTGATCAACGCAATGGTTTTATCCAAAGCCTTAAAGATGATCC
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GCAACAAAATAACTTCAACAAAGATCAACAAAGCGCCTTCTATGAAATCTTGAACATGCCTAACTTAAA
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CAACTACTGAAGCTGTTGATGCTGCTACTGCAGAAAAAGTCTTCAAACAATACGTAACGACAACGGTG
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GAAACAATACTAAAGCAGTAGACGCAGAAACTGCAGAAAAAGCCTTCAAACAATACGTAACGACAA
CGGTGTTGATGGTGTGGACTTATGATGATGCGACTAAGACCTTTACGGTAACTGAAATGGTTACAG
AGGTTCTGGATCTAGTAGC**

Insert3 mCherry:

**ATGGTTACAGAGGTTCTGGATCTAGTAGCATGGTGAGCAAGGGCGAGGAGGATAACATGGCCATCA
TCAAGGAGTTCATGCGCTTCAAGGTGCACATGGAGGGCTCCGTGAACGGCCACGAGTTTCGAGATCGAGG
GCGAGGGCGAGGGCCGCCCTACGAGGGCACCCAGACCGCAAGCTGAAGGTGACCAAGGGTGGCCCCC
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CTACAACGTCAACATCAAGTTGGACATCACCTCCACAACGAGGACTACACCATCGTGGAACAGTACGA
ACGCGCCGAGGGCCGCCACTCCACCGGCGGCATGGACGAGCTGTACAAGGGCGGCCGACGACTC**

Restriction enzyme for bead release: *NotI*

96 different full length single chains to full-length antibodies heavy

Backbone HerceptinCH_pcDNA3.3

CTCGAGGCTAGCACCAAGGGCCCATCGGTCTTCCCCCTGGCACCCCTCCTCCAAGAGCACCTCTGGGGGC
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Insert scFv heavy

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XATGCAGACTCCGTGAAGGGCCGGTTCACCATCTCCCGTACAATTCCAAGAACACGCTGTATCTGCAA
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Restriction NheI

96 different full length single chains to full-length antibodies light

Backbone HerceptinCL_pOptivec

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Insert scFv light

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Restriction enzyme for bead release: BsiWI

Bispecific antibodies

Backbone Herceptin_pCDNA3.3

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4 amino acid linker to binder 1 for heavy chain

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GG

15 amino acid linker to binder 1 for heavy chain

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XXXXCTGGCCGAGGCCAAGGTGCTGGCCAACAGG

18 amino acid linker to binder 1 for heavy chain

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XXXXXXXXXXXXCTGGCCGAGGCCAAGGTGCTGGCCAACAGG

4 amino acid linker to binder 2 for heavy chain

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TGG

15 amino acid linker to binder 2 for heavy chain

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18 amino acid linker to binder 2 for heavy chain

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4 amino acid linker to binder 3 for heavy chain

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TTC

15 amino acid linker to binder 3 for heavy chain

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XXXXGTGGACAACAAGTTCAACAAGGAGGGCTTC

18 amino acid linker to binder 3 for heavy chain

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Backbone: Herceptin_pOptivec

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TGTTATCCGCTCACAATTCACACAACATACGAGCCGGAAGCATAAAGTGTAAGCCCTGGGTGCCTAA
TGAGTGAGCTAACTCACATTAATTGCGTTGCGCTCACTGCCCGCTTTCAGTCGGGAAACCTGTGCTGC
CAGCTGCATTAATGAATCGGCCAACCGCGGGGAGAGCGGTTTGGGTATTGGGCGCTTTCGCTTCC
TCGCTCACTGACTCGCTGCGCTCGGTGCTTGGCTGCGGCGAGCGGTATCAGCTCAAGGGCGGTA
ATACGGTTATCCACAGAATCAGGGGATAACGCGAGGAAAGAACATGTGAGCAAAAGGCCAGCAAAAGGC
CAGGAACCGTAAAAAGGCCGCTTGGTGGCGTTTTTCCATAGGCTCCGCCCCCTGACGAGCATCACAA
AAATCGACGCTCAAGTCAGAGGTGGCGAAACCCGACAGGACTATAAAGATACCAGGCGTTTCCCCCTGG
AAGCTCCCTCGTGCCTCTCCTGTTCCGACCTGCCGTTACCGGATACCTGTCCGCTTTTCTCCCTTCG
GGAAGCGTGGCGTTTTCTCATAGCTCACGCTGTAGGTATCTCAGTTCCGGTGTAGGTGCTTCCGCTCCAAG
CTGGGCTGTGTGCACGAACCCCGTTCCAGCCGACCGCTGCGCCTTATCCGGTAACTATCGTCTTGAGT
CCAACCCGGTAAGACACGACTTATCGCCACTGGCAGCAGCCACTGGTAACAGGATTAGCAGAGCGAGGT
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CCACCGCTGGTAGCGTTTTTTTTGTTTGAAGCAGCAGATTACGCGCAGAAAAAAGGATCTCAAGAAG
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TGAGATTATCAAAAAGGATCTTCACCTAGATCTTTTAAATTAATAAATGAAGTTTTAAATCAATCTAA
AGTATATATGAGTAAACTTGGTCTGACAGTTACCAATGCTTAATCAGTGAGGCACCTATCTCAGCGATC
TGTCTATTTGTTTCTCCATAGTTGCCTGACTCCCCGTCGTGTAGATAACTACGATACGGGAGGGCTTA
CCATCTGGCCCCAGTGCTGCAATGATACCGCGAGACCCACGCTCACCGGCTCCAGATTTATCAGCAATA
AACCAGCCAGCCGGAAGGGCCGAGCGCAGAAGTGGTCTGCAACTTTATCCGCTCCATCCAGTCTATT
AATTGTTGCCGGGAAGCTAGAGTAAGTAGTTCGCCAGTTAATAGTTTGGCAACGTTGTTGCCATTGCT
ACAGGCATCGTGGTGTACGCTCGTCTGTTGGTATGGCTTCACTCAGCTCCGGTCCCAACGATCAAGG
CGAGTTACATGATCCCCATGTTGTGCAAAAAAGCGGTTAGCTCCTTCGGTCTCCGATCGTTGTGAGA
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CCGAGTTGCTCTTGGCCGGCGTCAATACGGGATAATACCGCGCCACATAGCAGAACTTTAAAAGTGCTC
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TAACCCACTCGTGCACCCAAGTATCTTACGATCTTTTACTTTTACCAGCGTTTCTGGGTGAGCAAAA
ACAGGAAGGCAAAATGCCGCAAAAAAGGGAATAAGGGCGACACGGAAATGTTGAATACTCATACTCTT
CCTTTTTCAATATTATTGAAGCATTATCAGGGTATTGTCATGAGCGGATACATATTTGAATGTAT
TTAGAAAAATAACAAATAGGGTTCCGCGCACATTTCCCCGAAAAGTGCCACCTGACGTCGACGGATC
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AGCCCATATATGGAGTTCGCGTTACATAACTTACGGTAAATGGCCCGCTGGCTGACCGCCCAACGAC
CCCCGCCATTGACGTCAATAATGACGTATGTTCCCATAGTAACGCCAATAGGGACTTTCATTGACGT
CAATGGGTGGAGTATTTACGGTAAACTGCCACTTGGCAGTACATCAAGTGTATCATATGCCAAGTACG
CCCCCTATTGACGTCAATGACGGTAAATGGCCCGCTGGCATTATGCCAGTACATGACCTTATGGGAC
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GGTTCCACGTGGGATATTAGATGACGCGAGTCTCCCTCCTCCCTCAGTGCTTCCGTGGGAGATAGGGTT
ACTATTACCTGCCGGCCTCTCAAGACGTCAACACTGCAGTAGCCTGGTATCAGCAGAAGCCTGGTAAA
GCCCCAAAAGTCTGATCTACAGCGCAAGTTTCTTGTACTCCGGGGTGCCTCTAGATTTAGCGGCTCA
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CAACAGCACTATAACACACCTCCCACATTTGGCCAGGGTACTAAGGTGGAAATAAAGCGTACGGTGGCT
GCACCATCTGTCTTCATCTTCCC GCCATCTGATGAGCAGTTGAAATCTGGAAGTGCCTCTGTTGTGTGCC
TGCTGAATAACTTCTATCCCAGAGAGGCCAAAAGTACAGTGGAAAGTGGATAACGCCCTCCAATCGGGTA
ACTCCCAGGAGAGTGTACAGAGCAGGACAGCAAGGACAGCACCTACAGCCTCAGCAGCACCTGACGC
TGAGCAAAGCAGACTACGAGAAACAAAAGTCTACGCCTGCGAAGTCACCCATCAGGGCCTGAGTTTCGC
CCGTCAAAAGAGCTTCAACAGGGGAGAGTGT

4 amino acid linker to binder 1 for light chain

GTCACAAAGAGCTTCAACAGGGGAGAGTGTXXXXXXXXXXXXXCTGGCCGAGGCCAAGGTGCTGGCCAAC
AGG

15 amino acid linker to binder 1 for light chain

GTCACAAAGAGCTTCAACAGGGGAGAGTGTXXX
XXXXXCTGGCCGAGGCCAAGGTGCTGGCCAACAGG

18 amino acid linker to binder 1 for light chain

GTCACAAAGAGCTTCAACAGGGGAGAGTGTXXX
XXXXXXXXXXXXXXXXCTGGCCGAGGCCAAGGTGCTGGCCAACAGG

4 amino acid linker to binder 2 for light chain

GTCACAAAGAGCTTCAACAGGGGAGAGTGTXXXXXXXXXXXXXGTGGACAACAAGTTCAACAAGGAGATG
TGG

15 amino acid linker to binder 2 for light chain

GTCACAAAGAGCTTCAACAGGGGAGAGTGTXXX
XXXXXGTGGACAACAAGTTCAACAAGGAGATGTGG

18 amino acid linker to binder 2 for light chain

GTCACAAAGAGCTTCAACAGGGGAGAGTGTXXX
XXXXXXXXXXXXXXXXGTGGACAACAAGTTCAACAAGGAGATGTGG

4 amino acid linker to binder 3 for light chain

GTCACAAAGAGCTTCAACAGGGGAGAGTGTXXXXXXXXXXXXXGTGGACAACAAGTTCAACAAGGAGGGC
TTC

15 amino acid linker to binder 3 for light chain

GTCACAAAGAGCTTCAACAGGGGAGAGTGTXXX
XXXXXGTGGACAACAAGTTCAACAAGGAGGGCTTC

18 amino acid linker to binder 3 for light chain

GTCACAAAGAGCTTCAACAGGGGAGAGTGTXXX
XXXXXXXXXXXXXXXXXGTGGACAACAAGTTCAACAAGGAGGGCTTC

Binder 1

CTGGCCGAGGCCAAGGTGCTGGCCAACAGGXX
XXGG
CGCGCCACGACTC

Restriction enzyme for bead release: *AscI*

Binder 2

GTGGACAACAAGTTCAACAAGGAGATGTGGXX
XX
XX
XX
XX
XXXXXXXXXXXXXXXXXXXXTAAGGCGCGCCACGACTC

Restriction enzyme for bead release: *AscI*

Binder 3

GTGGACAACAAGTTCAACAAGGAGGGCTTCXX
XX
XX
XX
XX
XXXXXXXXXXXXXXXXXXXXTAAGGCGCGCCACGACTC

Restriction enzyme for bead release: *AscI*

Vector assembly

pHisZamp

Backbone: amp resistance

***CTCGAGGCGGCCGATAACGCCGGTCGCTACCATTACCAACTTGTCTGGTGTCAAAAATAATAGGCCT
ACTAGTCGGCCGTACGGGCCCTTTTCGTCTCGCGCGTTTCGGTGATGACGGTGAAAACCTCTGACACATG
CAGCTCCCGGAGACGGTCACAGCTTGTCTGTAAGCGGATGCCGGGAGCAGACAAGCCGTCAGGGCGCG
TCAGCGGGTGTGGCGGGTGTGGGGCTGGCTTAACTATGCGGCATCAGAGCAGATTGTACTGAGAGTG
CACCATATGCGGTGTGAAATACCGCACAGATGCGTAAGGAGAAAATACCGCATCAGGCGGCCTTAAGGG
CCTCGTGATACGCCTATTTTTATAGGTTAATGTCATGATAATAATGGTTTCTTAGACGTCAGGTGGCAC
TTTTTCGGGAAATGTGCGCGGAACCCCTATTTGTTTATTTTTCTAAATACATTCAAATATGTATCCGCT
CATGAGACAATAACCCTGATAAATGCTTCAATAATATTGAAAAAGGAAGAGTATGAGTATTCAACATT
TCCGTGTCGCCCTTATCCCTTTTTGCGGCATTTTGCCTTCCTGTTTTGCTCACCCAGAAACGCTGGT
GAAAGTAAAAGATGCTGAAGATCAGTTGGGTGCACGAGTGGGTTACATCGAACTGGATCTCAACAGCG
GTAAGATCCTTGAGAGTTTTGCCCCGAAGAACGTTTTCCAATGATGAGCACTTTTAAAGTTCTGCTAT***

GTGGCGGGTATTATCCCGTATTGACGCCGGGCAAGAGCAACTCGGTCGCCGCATACACTATTCTCAGA
ATGACTTGGTTGAGTACTCACCAGTCACAGAAAAGCATCTTACGGATGGCATGACAGTAAGAGAATTA
TGCAGTGCTGCCATAACCATGAGTGATAAACTGCGGCCAACTTACTTCTGACAACGATCGGAGGACCG
AAGGAGCTAACCGCTTTTTTGCACAACATGGGGGATCATGTAACCTCGCCTTGATCGTTGGGAACCGGAG
CTGAATGAAGCCATAACAAACGACGAGCGTGACACCACGATGCCTGTAGCAATGGCAACAACGTTGCGC
AACTATTAACCTGGCGAACTACTTACTCTAGCTTCCCGGCAACAATTAATAGACTGGATGGAGGCGGAT
AAAGTTGCAGGACCACTTCTGCGCTCGGCCCTTCCGGCTGGCTGGTTTATTGCTGATAAATCTGGAGCC
GGTGAGCGTGGGTCTCGCGGTATCATTGCAGCACTGGGGCCAGATGGTAAGCCCTCCCGTATCGTAGTT
ATCTACACGACGGGGAGTCAGGCAACTATGGATGAACGAAATAGACAGATCGCTGAGATAGGTGCCTC
ACTGATTAAGCATTGGTAACCTGTCAGACCAAGTTTACTCATATATACTTTAGATTGATTTAAAACCTCA
TTTTTAATTTAAAAGGATCTAGGTGAAGATCCTTTTTGATAATCTCATGACCAAAATCCCTTAACGTC
AGTTTTCGTTCCACTGAGCG

Insert 1: Origin of replication

CCTTAACGTGAGTTTTCGTTCCACTGAGCGTCAGACCCCGTAGAAAAGATCAAAGGATCTTCTTGAGA
TCCTTTTTTCTGCGCGTAATCTGCTGCTTGCAAACAAAAAAACCACCGCTACCAGCGGTGGTTTTGTTT
GCCGGATCAAGAGCTACCAACTCTTTTTCCGAAGGTAACCTGGCTTCAGCAGAGCGCAGATACCAAATAC
TGTCCTTCTAGTGTAGCCGTAGTTAGGCCACCACTTCAAGAACTCTGTAGCACCAGCTACATACCTCGCT
CTGCTAATCCTGTTACCAGTGGCTGCTGCCAGTGGCGATAAGTCGTGTCTTACCGGGTTGGACTCAAGA
CGATAGTTACCGGATAAGGCGCAGCGGTCCGGCTGAACGGGGGGTTTCGTGCACACAGCCAGCTTGGAG
CGAACGACCTACACCGAACTGAGATACCTACAGCGTGAGCTATGAGAAAGCGCCACGCTTCCGAAGGG
AGAAAGGCGGACAGGTATCCGGTAAGCGGCAGGGTCGGAACAGGAGAGCGCACGAGGGAGCTTCCAGG
GGGAAACGCCTGGTATCTTTATAGTCTGTCCGGTTTCGCCACCTCTGACTTGAGCGTCGATTTTTGTG
ATGCTCGTCAGGGGGGCGGAGCCTATGGAAAAACGCCAGCAACGCGGCCCTTTTACGGTTCCTGGCCTT
TTGCTGGCCTTTTGCTCACATGTTCTTTCTGCGTTATCCCTGATTCTGTGGATAACCGTATTACCGCC
TTTGAGTGAGCTGATACCGCTCGCCGCAGCCGAACGACCGAGCGCAGCGAGTCAGTGAGCGAGGAAGCG
GAAGAGCGCCCAATACGCAAACCGCCTCTCCCGCGCGTTGGCCGATT**CATTAATGCAGCTGGCAC**

Insert 2: Lac promoter

CGTTGGCCGATTCAATTAATGCAGCTGGCAGCAGAGGTTTCCCGACTGGAAAAGCGGGCAGTGAGCGCAA
CGCAATTAATGTGAGTTAGCTCACTCATTAGGCACCCAGGCTTTACACTTTATGCTTCCGGCTCGTAT
GTTGTGTGGAATTGTGAGCGGATAACAATTTACACACAGGAAACAGCTATGACCATGATTACGCCAAGCT
TGCATCATCATCATCATGTAGACAACAAATTAACAACAAAGAACAAACAAACCGCTTCTATGAGATCT
TACATTTACCTAACTTAAACGAAGAACAACGAAACGCCTTCATCCAAAGTTTAAAAGATGACCCAAGCC
AAAGCGCTAACTTGCTAGCAGAAGCTAAAAAGCTAAATGATGCTCAGGCGCCGAAACAGATCCAATCG
ATCCCCGGGT**ACCGGTCCGCCACCGGTGGATCCGGT**

Insert 3: eGFP

CGGGTACCGGTCCGCCACCGGTGGATCCGGTATGGTGAGCAAGGGCGAGGAGCTGTTACCGGGGTGGT
GCCCATCCTGGTGCAGCTGGACGGCGACGTAACCGGCCACAAGTTACGCGTGTCCGGCGAGGGCGAGGG
CGATGCCACCTACGGCAAGCTGACCCTGAAGTTTCTGCAACCACCGCAAGCTGCCCGTGGCCTGGCC
ACCCTCGTGACCACCCTGACCTACGGCGTGCAGTGCTTACGCCGCTACCCCGACCACATGAAGCAGCAG
ACTTCTTCAAGTCCGCCATGCCCCAAGGCTACGTCCAGGAGCGCACCATCTTCTTCAAGGACGACGGCA
ACTACAAGACCCGCGCCGAGGTGAAGTTTCGAGGGCGACACCCTGGTGAACCGCATCGAGCTGAAGGGCA
TCGACTTCAAGGAGGACGGCAACATCCTGGGGCACAAGCTGGAGTACAACACTACAACAGCCACAACGCTCT
ATATCATGGCCGACAAGCAGAAGAAGCGCATCAAGGTGAACCTCAAGATCCGCCACAACATCGAGGACG
GCAGCGTGCAGCTCGCCGACCACTACCAGCAGAACACCCCATCGGCGACGGCCCCGTGCTGCTGCCCGA
CAACCACTACCTGAGCACCCAGTCCGCCCTGAGCAAAGACCCCAACGAGAAGCGCGATCACATGGTCCT
GCTGGAGTTCGTGACCGCCCGGGATCACTCT**CGGCATGGACGAGCTGTACAAGGCGGCCGACGAC**
TC

Restriction enzyme for bead release: NotI

pHisZKm

Same as pHisZamp except backbone exchanged for:

Backbone:

***CTCGAGGCGGCCGATAAGGTCTGACGCTCAGTGGAAACGAAAACCTCACGTTAAGGGATTTTGGTCATG
AACAAATAAACTGTCTGCTTACATAAACAGTAATAACAAGGGGTGTTATGAGCCATATTTCAACGGGAAA
CGTCTTGCTCTAGGCCGCGATTAATTTCCAACATGGATGCTGATTTATATGGGTATAAATGGGCTCGCG
ATAATGTCGGGCAATCAGGTGCGACAATCTATCGATTGTATGGGAAGCCCGATGCGCCAGAGTTGTTTC
TGAAACATGGCAAAGGTAGCGTTGCCAATGATGTTACAGATGAGATGGTCAGACTAACTGGCTGACG
GAATTTATGCCTCTTCCGACCATCAAGCATTTTATCCGTA CTCTGATGATGCATGGTTACTCACCCT
GCGATCCCCGGGAAAACAGCATTCCAGGTATTAGAAGAATATCCTGATTCAGGTGAAAATATTGTTGA
TGCGCTGGCAGTGTTCTGCGCCGGTTGCATTGATTCTGTTTGTAAATTGTCCTTTTAAACAGCGATCG
CGTATTTCTGCTCTCGCTCAGGCGCAATCACGAATGAATAACGGTTTGGTTGATGCGAGTGATTTTGATGA
CGAGCGTAATGGCTGGCCTGTTGAACAAGTCTGGAAAAGAAATGCATAAACTTTTGCCATTCTCACCAGGA
TTCAGTCTGCTCACTCATGGTGATTTCTCACTTGATAACCTTATTTTTGACGAGGGGAAAATTAATAGGTTG
TATTGATGTTGGACGAGTCGGAATCGCAGACCGATACCAGGATCTTGCCATCCTATGGAACGCTCGG
TGAGTTTTCTCCTTATTACAGAAAACGGCTTTTTCAAAAAATATGGTATTGATAATCCTGATATGAATAA
ATTGCAGTTTCATTTGATGCTCGATGAGTTTTTCTAAGAATTAATTCATGAGCGGATACATATTTGAAT
GTATTTAGAAAAATAAACAAATAGGGGTTCCGCGCACATTTCCCCGAAAAGTGCCCTTAACTGAGTT
TTCGTTCCACTGAGCG***

Restriction enzyme for bead release: NotI

pAFF8C

Backbone: Origin of replication

***CTCGAGGCGGCCGCAAGCTTGTGCGACGGAGCTCGAATTCGGATCCATGTAGTGAGCGAAGGTACCAGG
TAATGCAGCTAAAATTTTCATCTATCAGTGCTTTTACACCTTCAACAGTTTTTGGCATTGTTGATTAGGTT
CTTGTAATAGTCACTTACTCCATATTTGTCAAGTCTCTGTTAGCTAAGACTTTAGCTTCAGCTAATTC
AATTGATTTAACAGTATCTTCAGCAGGTGTTTGTGATTTCAAGAAATCAGATAAGCCATCTGTTGCTTC
TGAAATACGCGCTTTCTTCGCTGATTCAACAACCTTGTGCTTGAAGGTCTTTTACACCTTCAACAGTTTT
GGCATTGTTGATTAGGTTCTTGTGATAGTCACTTACTCCATATTTGTCAAGTCTCTGTTAGCTAAGAC
TTTAGCTTCAGCTAAGCTAGCCATATGGCTGCCGCGCGCACACCAGGCCGCTGCTGTGATGATGATGAT
ATGGCTCACTGCCCATTGGTATATCTCCTTCTTAAAGTTAAACAAAATTAATTTCTAGAGGGGAATGTTAT
CCGCTCACAAATCCCCCTATAGTGAGTCGTATTAATTTCCGCGGATCGAGATCTCGATCTCAGGCCG
ACGCATCGTGGCCGCGCATCACCGGCCACAGGTGCGGTTGCTGGCGCCTATATCGCCGACATCACCGAT
GGGGAAGATCGGGCTCGCCACTTCGGGCTCATGAGCGCTTGTTCGGCGTGTTGTTGTTGTTGTTGTTGTTG
GTGGCCGGGGGACTGTTGGGCGCCATCTCCTTGATGCACCATTCTTGGCGCGCGGTTGCTCAACGGCC
TCAACCTACTACTGGGCTGCTTCCCTAATGCAGGAGTCGCATAAAGGGAGAGCGTCGAGATCCCGGACACC
ATCGAATGGCGCAAAACCTTTTCGCGGTATGGCATGATAGCGCCGGAAGAGAGTCAATTCAGGGTGGTG
AATGTGAAACCAGTAACGTTATACGATGTGCGAGAGTATGCCGGTGTCTCTTATCAGACCGTTTCCCGC
GTGGTGAACCAGGCCAGCCAGCTTTCTGCGAAAACCGGGAAAAAGTGAAGCGGCGATGGCGGAGCTG
AATTACATTTCCAACCGCTGGCACAACAACCTGGCGGGCAAAACAGTCGTTGCTGATTGGCGTTGCCACC
TCCAGTCTGGCCCTGCACGCGCCGTCGCAATTTGTCGCGCGGATTAATCTCGCGCCGATCAACTGGGT
GCCAGCGTGGTGGTGTGATGGTAGAACGAAGCGGCTCGAAGCCTGTAAAGCGGCGGTGCACAATCTT
CTCGCGCAACGCGTCAGTGGGCTGATCATTAACCTATCCGCTGGATGACCAGGATGCCATTGCTGTGGAA
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TTCTCCCATGAAGACGGTACGCGACTGGGCGTGGAGCATCTGGTGCATTGGGTACCAGCAAATCGCG
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ACCGAAGACAGCTCATGTTATATCCCGCGTTAACCACCATCAAACAGGATTTTTCGCTGCTGGGGCAA
ACCAGCGTGGACCGCTTGTGCAACTCTCTCAGGGCCAGGCGGTGAAGGGCAATCAGCTGTTGCCCGTC
TCACTGGTGAAGAAAAAACCACCTGGCGCCAATACGCAAACCGCCTCTCCCCGCGGTTGGCCGAT
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CTTCCGGTGGGCGCGGGCATGACTATCGTCCGCGCACTTATGACTGTCTTCTTTATCATGCAACTCGT
AGGACAGGTGCCGGCAGCGCTCTGGGTCATTTTTCGGCGAGGACCGCTTTCGCTGGAGCGCGACGATGAT
CGGCTGTGCTTGGCGTATTCCGAATCTTGACGCGCCTCGCTCAAGCCTTCGTCACTGGTCCCGCCACC***

AAACGTTTCGGCGAGAAGCAGGCCATTATCGCCGGCATGGCGGCCACGGGTGCGCATGATCGTGCTC
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TTCCGTGTTTTCGTAAAGTCTGGAAACGCGGAAGTCAGCGCCTGCACCATTATGTTCCGGATCTGCATC
GCAGGATGCTGCTGGCTACCCTGTGGAACACCTACATCTGTATTAACGAAGCGCTGGCATTGACCCTGA
GTGATTTTTCTCTGGTCCC GCCGCATCCATACCGCCAGTTGTTTACCCTACAACGTTCCAGTAACGGG
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ACAAGCCCGTCAGGGCGCGTCAGCGGGTGTGGCGGGTGTGGGGGCGCAGCCATGACCCAGTCACGTAG
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AACCGTAAAAAGGCCGCTTGGTGGCGTTTTTCCATAGGCTCCGCCCCCTGACGAGCATCACAAAAAT
CGACGCTCAAGTCAGAGGTGGCGAAACCGACAGGACTATAAAGATACCAGGCGTTTTCCCTTGGAAAGC
TCCCTCGTGCCTCTCTGTTCCGACCTGCCGCTTACCGGATACCTGTCCGCTTCTCCCTTCCGGAA
GCGTGGCGCTTCTCATAGCTCACGCTGTAGGTATCTCAGTTCGGTGTAGGTGCTTCCGCTCCAAGCTGG
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CCCGGTAAGACACGACTTATCGCCACTGGCAGCAGCCACTGGTAACAGGATTAGCAGAGCGAGGTATGT
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TGAACAATAAACTGTCTGCTTACATAAACAGTAATAACAAGGGGTGTTATGAGCATATTTCCGGGA
AACGCTTTGCTCTAGGCCGCGATTAATTC AACATGGATGCTGATTTATATGGGTATAAATGGGCTCG
CGATAATGTCCGGCAATCAGGTGCGACAATCTATCGATTGTATGGGAAGCCCGATGCGCCAGAGTTGTT
TCTGAAACATGGCAAAGGTAGCGTTGCCAATGATGTTACAGATGAGATGGTCAGACTAACTGGCTGA
CGGAATTTATGCCTCTTCCGACCATCAAGCATTATCCGTAATCCTGATGATGCATGGTTACTCACCA
CTGCGATCCCGGGAAAAACAGCATTCCAGGTATTAGAAGATATCCTGATTCAGGTGAAAAATATTGTTG
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GCGTATTTTCGTCTCGCTCAGGCGCAATCACGAATGAATAACGGTTTGGTTGATGCGAGTGATTTTGATG
ACGAGCGTAATGGCTGGCTGTTGAACAAGTCTGGAAAGAAATGCATAAACTTTTGCCATTCTCACCGG
ATTCAGTCGTCATCATGGTGATTTCTCACTTGATAACCTTATTTTTGACGAGGGGAAATTAATAGGTT
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AATTGCAGTTTTCATTTGATGCTCGATGAGTTTTTCTAAGAAATTAATTCATGAGCGGATACATATTTGA
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GGAACAAGAGTCCACTATTAAGAACGTGGACTCCAACGTCAAAGGGCGAAAAACCGTCTATCAGGGCG
ATGGCCACTACGTGAACCATCACCTAATCAAGTTTTTTGGGGTCGAGGTGCCGTAAAGCACTAAATC
GGAACCTAAAGGGAGCCCCGATTTAGAGCTTGACGGGGAAAGCCGGCGAACGTGGCGAGAAAGGAA
GGGAAGAAAGCGAAAGGAGCGGGCGCTAGGGCGCTGGCAAGTGTAGCGGTCACGCTGCGCGTAACCACC
ACACCCGCGCGCTTAATGCGCCGCTACAGGGCGCGTCCCATTCGCAATCCGGATATAGTTCTCTCTTT
CAGCAAAAAACCCCTCAAGACCCGTTTAGAGGCCCAAGGGTTATGCTAGTTATTGCTCAGCGGTGGG
GCGCGCTTATTTAAGATTATTGGTCACTTGTTCAGCAAGATGCATTTCTCTGTATGAGGGACCAAGA
GACAGATCATGTTCCGGAGGGGGTCTGGTGCTCAACGTTTCACTTCTGGGCTTCTGGAGTTCCCTGAGCA
ATCTGGTGGTCTCATCAAGTTTTCTTCTGGAATATTTTCAAGCTTCTTCAAGTCAAAAACTTCAACTGGAA
CGCAAAAATTTGTTACTGCTTTCAGCGCTATGAGCCTGTCTTGTGACTGGAGTCCAAACGCCCTGGTG
GCTCTGCGGCCGCACGACTC

Insert 1: Km resistance

GGTCTGACGCTCAGTGGAAACGAAAACTCACGTTAAGGGATTTTGGTTCATGAACAATAAACTGTCTGC
TTACATAAACAGTAATACAAGGGGTGTTATGAGCCATATTC AACGGGAAACGTTGCTCTAGGCCGCG
ATTAATTC AACATGGATGCTGATTTATATGGGTATAAATGGGCTCGCGATAATGTCCGGCAATCAG

GTGCGACAATCTATCGATTGTATGGGAAGCCCGATGCGCCAGAGTTGTTTCTGAAACATGGCAAAGGTA
GCGTTGCCAATGATGTTACAGATGAGATGGTCAGACTAAACTGGCTGACGGAATTTATGCCTCTCCGA
CCATCAAGCATTATCCGTACTCCTGATGATGCATGGTTACTCACCCTGCGATCCCCGGGAAAACAG
CATTCCAGGTATTAGAAGAATATCCTGATTCAGGTGAAAATATTGTTGATGCGCTGGCAGTGTTCCTGC
GCCGTTGCATTTCGATTCCTGTTTGTAAATTGTCCTTTTAAACAGCGATCGCGTATTTTCGTCTCGCTCAGG
CGCAATCACGAATGAATAACGGTTTGGTTGATGCGAGTGATTTTGTGATGACGAGCGTAATGGCTGGCCTG
TTGAACAAGTCTGGAAAGAAATGCATAAACTTTTGCCATTCTCACCGGATTCAGTCGTCACCTCATGGTG
ATTTCTCACTTGATAACCTTATTTTTGACGAGGGGAAATTAATAGGTTGTATTGATGTTGGACGAGTC
GGAATCGCAGACCGATACCAGGATCTTGCCATCCTATGGAAGTGCCTCGGTGAGTTTTCTCCTTCATTA
CAGAAACGGCTTTTTCAAAAATATGGTATTGATAATCCTGATATGAATAAATTGCAGTTTCATTTGAT
GCTCGATGAGTTTTTCTAAGAATTAATTCATGAGCGGATACATATTTGAATGTATTTAGAAAAATAAA
CAAATAGGGTTCCGCGCACATTTCCCGAAAAGTGC

Insert 2: f1 ori

GGGTTCCGCGCACATTTCCCGAAAAGTGCCACCTGAAATTGTAAACGTTAATATTTTTGTAAAATTC
GCGTTAAATTTTTGTAAAATCAGCTCATTTTTTAACCAATAGGCCGAAAATCCCTTATAAA
TCAAAAGAATAGACCGAGATAGGGTTGAGTGTGTTCCAGTTTGGAAACAAGAGTCCACTATTAAGAA
CGTGGACTCCAACGTCAAAGGGCGAAAAACCGTCTATCAGGGCGATGGCCCACTACGTGAACCATCACC
CTAATCAAGTTTTTTGGGGTCGAGGTGCCGTAAAGCACTAAATCGGAACCCTAAAGGGAGCCCCGATT
TAGAGCTTGACGGGAAAAGCCGGCGAACGTGGCGAGAAAGGAAGGAAGAAAGCGAAAGGAGCGGGCG
CTAGGGCGCTGGCAAGTGTAGCGGTACGCTGCGCGTAACCACCACCCGCGCGCTTAATGCGCCGCT
ACAGGGCGCGTCCCATTCGCAAT**CCGGATATAGTTCCTCCTTTCA**

Insert 3: PrEST 2250323

TCGCCAATCCGGATATAGTTCCTCCTTTAGCAAAAAACCCCTCAAGACCCGTTTAGAGGCCCAAGG
GGTTATGCTAGTTATTGCTCAGCGGTGGGGCGCGCCTTATTTAAGATTATTGGTCACTTGTTCAGCAAG
ATGCATTTCTCTGTATGAGGGACCCAAGAGACAGATCATGTTCCGAGGGGGTCTGGTGCTCAAACGTTT
ATTCTGGGCTTCTGGAGTTCCTGAGCAATCTGGTGGTCTCATCAAGTTTCTTCTGGAATATTTTCAGC
TTCTTCAGAGTCAAAAACTTCAACTGGAACGCCAAAATTTGTTACTGCTTTCAGCGCTATGAGCCTGTC
TTGAGTACTGGAGTCCAAACGCCCTGGTGGCTCTGCGGCCGC**ACGACTC**