SEQUENCE LISTING

<110> Iontas Limited

<120> Preparation of Libraries of Protein Variants Expressed in Eukaryotic Cells and Use for Selecting Binding Molecules

<130> SMW/CP7119530

<150> GB1407852.1
<151> 2014-05-02

<160> 142

<170> PatentIn version 3.5

<210> 1
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<212> DNA
<213> Artificial Sequence

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<223> pD1 dual promoter antibody expression cassette for surface expression

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<210> 2
<211> 233
<212> PRT
<213> Artificial sequence

<220>
<223> BM40 Leader, Humanised D1.3 VL and Human C Kappa sequence

<400> 2

Met Arg Ala Trp Ile Phe Phe Leu Leu Cys Leu Ala Gly Arg Ala Leu
1 5 10 15

Ala Ala Ser Asp Ile Gln Met Thr Gln Ser Pro Ser Ser Leu Ser Ala
20 25 30

Ser Val Gly Asp Arg Val Thr Ile Thr Cys Arg Ala Ser Gly Asn Ile
35 40 45

His Asn Tyr Leu Ala Trp Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys
50 55 60

Leu Leu Ile Tyr Tyr Thr Thr Thr Leu Ala Asp Gly Val Pro Ser Arg
65 70 75 80

Phe Ser Gly Ser Gly Ser Gly Thr Asp Tyr Thr Phe Thr Ile Ser Ser
85 90 95

Leu Gln Pro Glu Asp Ile Ala Thr Tyr Tyr Cys Gln His Phe Trp Ser
100 105 110

Thr Pro Arg Thr Phe Gly Gln Gly Thr Lys Val Glu Ile Lys Arg Thr
115 120 125

Ala Ala Ala Pro Ser Val Phe Ile Phe Pro Pro Ser Asp Glu Gln Leu
130 135 140

```
Lys Ser Gly Thr Ala Ser Val Val Cys Leu Leu Asn Asn Phe Tyr Pro
145                 150                 155                 160


Arg Glu Ala Lys Val Gln Trp Lys Val Asp Asn Ala Leu Gln Ser Gly
                165                 170                 175


Asn Ser Gln Glu Ser Val Thr Glu Gln Asp Ser Lys Asp Ser Thr Tyr
            180                 185                 190


Ser Leu Ser Ser Thr Leu Thr Leu Ser Lys Ala Asp Tyr Glu Lys His
        195                 200                 205


Lys Leu Tyr Ala Cys Glu Val Thr His Gln Gly Leu Ser Ser Pro Val
    210                 215                 220


Thr Lys Ser Phe Asn Arg Gly Glu Cys
225                 230


<210>  3
<211>  15
<212>  PRT
<213>  Artificial sequence

<220>
<223>  Leader 1 sequence

<400>  3

Met Ser Trp Ser Cys Ile Ile Leu Phe Leu Val Ala Thr Ala Thr
1               5                   10                  15


<210>  4
<211>  449
<212>  PRT
<213>  Artificial sequence

<220>
<223>  Leader 2 and D1.3 VH sequence, CH1 domain, Hinge, CH2 and CH3
       domains

<400>  4

Gly Ala Met Ala Gln Val Gln Leu Gln Glu Ser Gly Pro Gly Leu Val
1               5                   10                  15


Arg Pro Ser Gln Thr Leu Ser Leu Thr Cys Thr Val Ser Gly Ser Thr
            20                  25                  30


Phe Ser Gly Tyr Gly Val Asn Trp Val Arg Gln Pro Pro Gly Arg Gly
        35                  40                  45


Leu Glu Trp Ile Gly Met Ile Trp Gly Asp Gly Asn Thr Asp Tyr Asn
```

50 55 60

Ser Ala Leu Lys Ser Arg Val Thr Met Leu Val Asp Thr Ser Lys Asn
65 70 75 80

Gln Phe Ser Leu Arg Leu Ser Ser Val Thr Ala Ala Asp Thr Ala Val
85 90 95

Tyr Tyr Cys Ala Arg Glu Arg Asp Tyr Arg Leu Asp Tyr Trp Gly Gln
100 105 110

Gly Ser Leu Val Thr Val Ser Ser Ala Ser Thr Lys Gly Pro Ser Val
115 120 125

Phe Pro Leu Ala Pro Cys Ser Arg Ser Thr Ser Glu Ser Thr Ala Ala
130 135 140

Leu Gly Cys Leu Val Lys Asp Tyr Phe Pro Glu Pro Val Thr Val Ser
145 150 155 160

Trp Asn Ser Gly Ala Leu Thr Ser Gly Val His Thr Phe Pro Ala Val
165 170 175

Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val Pro
180 185 190

Ser Ser Asn Phe Gly Thr Gln Thr Tyr Thr Cys Asn Val Asp His Lys
195 200 205

Pro Ser Asn Thr Lys Val Asp Lys Thr Val Glu Arg Lys Cys Cys Val
210 215 220

Glu Cys Pro Pro Cys Pro Ala Pro Pro Val Ala Gly Pro Ser Val Phe
225 230 235 240

Leu Phe Pro Pro Lys Pro Lys Asp Thr Leu Met Ile Ser Arg Thr Pro
245 250 255

Glu Val Thr Cys Val Val Val Asp Val Ser His Glu Asp Pro Glu Val
260 265 270

Gln Phe Asn Trp Tyr Val Asp Gly Val Glu Val His Asn Ala Lys Thr
275 280 285

Lys Pro Arg Glu Glu Gln Phe Asn Ser Thr Phe Arg Val Val Ser Val
290 295 300

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Leu Thr Val Val His Gln Asp Trp Leu Asn Gly Lys Glu Tyr Lys Cys
305                 310                 315                 320

Ala Val Ser Asn Lys Gly Leu Pro Ala Pro Ile Glu Lys Thr Ile Ser
                325                 330                 335

Lys Thr Lys Gly Gln Pro Arg Glu Pro Gln Val Tyr Thr Leu Pro Pro
            340                 345                 350

Ser Arg Glu Glu Met Thr Lys Asn Gln Val Ser Leu Thr Cys Leu Val
        355                 360                 365

Lys Gly Phe Tyr Pro Ser Asp Ile Ala Val Glu Trp Glu Ser Asn Gly
    370                 375                 380

Gln Pro Glu Asn Asn Tyr Lys Thr Thr Pro Pro Met Leu Asp Ser Asp
385                 390                 395                 400

Gly Ser Phe Phe Leu Tyr Ser Lys Leu Thr Val Asp Lys Ser Arg Trp
                405                 410                 415

Gln Gln Gly Asn Val Phe Ser Cys Ser Val Met His Glu Ala Leu His
            420                 425                 430

Asn His Tyr Thr Gln Lys Ser Leu Ser Leu Ser Pro Gly Lys Gly Ser
        435                 440                 445

Lys
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<210> 5
<211> 60
<212> PRT
<213> Artificial

<220>
<223> Myc epitope, PDGFR spacer and PDGFR Transmembrane region (TM)

<400> 5

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Glu Gln Lys Leu Ile Ser Glu Glu Asp Leu Asn Ala Val Gly Gln Asp
1               5                   10                  15

Thr Gln Glu Val Ile Val Val Pro His Ser Leu Pro Phe Lys Val Val
            20                  25                  30

Val Ile Ser Ala Ile Leu Ala Leu Val Val Leu Thr Ile Ile Ser Leu
        35                  40                  45

Ile Ile Leu Ile Met Leu Trp Gln Lys Lys Pro Arg
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50 55 60

<210> 6
<211> 1542
<212> DNA
<213> Artificial Sequence

<220>
<223> pD1-huD1.3 donor construct fragment containing AAVS1 left homology arm, splice acceptor site, Blasticidin gene, SV40 polyA

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gcctccttca ggttccgtct tcctccactc cctcttcccc ttgctctctg ctgtgttgct 180

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gcaagctctt ccagcccccc gtcatggcat cttccagggg tccgagagct cagctagtct 660

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<210> 7
<211> 924
<212> DNA
<213> Artificial Sequence

<220>
<223> pD1-huD1.3 fragment containing AAVS1 right homology arm

<400> 7
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ccactaggga caggattggt gacagaaaag ccccatcctt aggcctcctc cttcctagtc 120

tcctgatatt gggtctaacc cccacctcct gttaggcaga ttccttatct ggtgacacac 180

ccccatttcc tggagccatc tctctccttg ccagaacctc taaggtttgc ttacgatgga 240

gccagagagg atcctgggag ggagagcttg gcaggggtg ggagggaagg gggggatgcg 300

tgacctgccc ggttctcagt ggccaccctg cgctaccctc tcccagaacc tgagctgctc 360

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acgcgtgtat actcgatctt tccg 924

<210> 8
<211> 132
<212> PRT
<213> Artificial Sequence

<220>
<223> Synthetic Blasticidin resistance gene

<400> 8

Met Ala Lys Pro Leu Ser Gln Glu Glu Ser Thr Leu Ile Glu Arg Ala
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Thr Ala Thr Ile Asn Ser Ile Pro Ile Ser Glu Asp Tyr Ser Val Ala
20 25 30

Ser Ala Ala Leu Ser Ser Asp Gly Arg Ile Phe Thr Gly Val Asn Val
35 40 45

Tyr His Phe Thr Gly Gly Pro Cys Ala Glu Leu Val Val Leu Gly Thr
50 55 60

Ala Ala Ala Ala Ala Ala Gly Asn Leu Thr Cys Ile Val Ala Ile Gly
65 70 75 80

Asn Glu Asn Arg Gly Ile Leu Ser Pro Cys Gly Arg Cys Arg Gln Val
85 90 95

Leu Leu Asp Leu His Pro Gly Ile Lys Ala Ile Val Lys Asp Ser Asp
100 105 110

Gly Gln Pro Thr Ala Val Gly Ile Arg Glu Leu Leu Pro Ser Gly Tyr
115 120 125

Val Trp Glu Gly
130

<210> 9
<211> 734
<212> DNA
<213> Artificial Sequence

<220>
<223> pD6 vector for scFv-Fc expression, fragment from Nco1 site to Pme1 site containing a fragment (last 4 amino acids) of the leader sequence, the complete CH2 domain and CH3 domain

<400> 9
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gctggacctt ccgtgttcct gttcccccca aagcccaagg acaccctgat gatcagccgg 120

acccccgaag tgacctgcgt ggtggtggac gtgtcccacg aggaccccga ggtgcagttc 180

aattggtacg tggacggcgt ggaagtgcac aacgccaaga ccaagcccag agaggaacag 240

ttcaacagca ccttccgggt ggtgtccgtg ctgaccgtgg tgcaccagga ctggctgaac 300

ggcaaagagt acaagtgcgc cgtctccaac aagggcctgc ctgcccccat cgagaaaacc 360

atcagcaaga ccaagggcca gcctcgcgag cctcaggtgt acacactgcc ccccagccgg 420

gaagagatga ccaagaacca ggtgtccctg acctgcctcg tgaagggctt ctaccccagc 480

gatatcgccg tggaatggga gagcaacggc cagcccgaga acaactacaa gaccacccca 540

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cggtggcagc agggcaacgt gttcagctgc agcgtgatgc acgaggccct gcacaaccac 660

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tatataactt taaa 734

<210> 10
<211> 4
<212> PRT
<213> Artificial Sequence

<220>
<223> Vector pD6, fragment at the end of leader sequence

<400> 10

Gly Ala Met Ala
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<210> 11
<211> 219
<212> PRT
<213> Artificial Sequence

<220>
<223> Vector pD6 CH2 and CH3 domains

<400> 11

Ala Pro Pro Val Ala Gly Pro Ser Val Phe Leu Phe Pro Pro Lys Pro
1 5 10 15

Lys Asp Thr Leu Met Ile Ser Arg Thr Pro Glu Val Thr Cys Val Val
20 25 30

Val Asp Val Ser His Glu Asp Pro Glu Val Gln Phe Asn Trp Tyr Val
35 40 45

Asp Gly Val Glu Val His Asn Ala Lys Thr Lys Pro Arg Glu Glu Gln
50 55 60

Phe Asn Ser Thr Phe Arg Val Val Ser Val Leu Thr Val Val His Gln
65 70 75 80

Asp Trp Leu Asn Gly Lys Glu Tyr Lys Cys Ala Val Ser Asn Lys Gly
85 90 95

Leu Pro Ala Pro Ile Glu Lys Thr Ile Ser Lys Thr Lys Gly Gln Pro
100 105 110

Arg Glu Pro Gln Val Tyr Thr Leu Pro Pro Ser Arg Glu Glu Met Thr
115 120 125

Lys Asn Gln Val Ser Leu Thr Cys Leu Val Lys Gly Phe Tyr Pro Ser
130 135 140

Asp Ile Ala Val Glu Trp Glu Ser Asn Gly Gln Pro Glu Asn Asn Tyr
145 150 155 160

Lys Thr Thr Pro Pro Met Leu Asp Ser Asp Gly Ser Phe Phe Leu Tyr
165 170 175

Ser Lys Leu Thr Val Asp Lys Ser Arg Trp Gln Gln Gly Asn Val Phe
180 185 190

Ser Cys Ser Val Met His Glu Ala Leu His Asn His Tyr Thr Gln Lys
195 200 205

Ser Leu Ser Leu Ser Pro Gly Lys Gly Ser Lys
210 215

<210> 12
<211> 60
<212> DNA
<213> Artificial Sequence

<220>
<223> Fragment of sequence of pD4 vector preceding and including the start of the AAVS1 left homology arm

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<210> 13
<211> 3933
<212> DNA
<213> Artificial sequence

<220>
<223> Fragment of sequence of vector pD4 including the end of the AAVS1 LHA, FRT site, Lox2272 site, 1-Sce1 meganuclease site, GFP left TALEN binding site, GFP right TALEN binding site, T2A, GFP, PGK promoter, puromycin delta TK, loxP site and start of AAVS1 RHA

<400> 13
ttctgggtac ttttatctgt cccctccacc ccacagtggg gcaagatgca tgaagttcct 60

attccgaagt tcctattctc tagaaagtat aggaacttcg accataactt cgtataaagt 120

atcctatacg aagttatgcg atcgctcgcg cgtagggata acagggtaat aagtccaccg 180

gtcgccacca tggtgagcaa gggcgaggag ctgttcactt ctgacctctt ctcttcctcc 240

cacagggcct agagagatct ggcagcggag agggcagagg aagtcttcta acatgcggtg 300

acgtggagga gaatcccgga ccgatggtga gcaagggaga agaactcttc accggggtgg 360

tgcccatcct ggtcgagctg gacggcgacg tgaacggcca caagttcagc gtgtccggcg 420

agggcgaggg cgatgccacc tacggcaagc tgaccctgaa gttcatctgc accaccggca 480

agctgcccgt gccctggccc accctcgtga ccaccctgac ctacggcgtg cagtgcttca 540

gccgctaccc cgaccacatg aagcagcacg acttcttcaa gtccgccatg cccgaaggct 600

acgtccagga gcgcaccatc ttcttcaagg acgacggcaa ctacaagacc cgcgccgagg 660

tgaagttcga gggcgacacc ctggtgaacc gcatcgagct gaagggcatc gacttcaagg 720

aggacggcaa catcctgggg cacaagctgg agtacaacta caacagccac aacgtctata 780

tcatggccga caagcagaag aacggcatca aggtgaactt caagatccgc cacaacatcg 840

aggacggcag cgtgcagctc gccgaccact accagcagaa cacccccatc ggcgacggcc 900

ccgtgctgct gcccgacaac cactacctga gcacccagtc cgccctgagc aaagacccca 960

acgagaagcg cgatcacatg gtcctgctgg agttcgtgac cgccgccggg atcactcacg 1020

gcatggacga gctgtacaag aagctgagcc acggcttccc gccggcggtg gcggcgcagg 1080

atgatggcac gctgcccatg tcttgtgccc aggagagcgg gatggaccgt caccctgcag 1140

cctgtgcttc tgctaggatc aatgtgtagg tgatcagcct cgactgtgcc ttctagttgc 1200

cagccatctg ttgtttgccc ctccccgtg ccttccttga ccctggaagg tgccactccc 1260

actgtccttt cctaataaaa tgaggaaatt gcatcgcatt gtctgagtag gtgtcattct 1320

attctggggg gtggggtggg gcaggacagc aaggggggagg attgggaaga caatagcagg 1380

catgctgggg atggcccaat tctaccgggt aggggaggcg cttttcccaa ggcagtctgg 1440

agcatgcgct ttagcagccc cgctgggcac ttggcgctac acaagtggcc tctggcctcg 1500

cacacattcc acatccaccg gtaggcgcca accggctccg ttctttggtg gccccttcgc 1560

gccaccttct actcctcccc tagtcaggaa gttcccccc gccccgcagc tcgcgtcgtg 1620

caggacgtga caaatggaag tagcacgtct cactagtctc gtgcagatgg acagcaccgc 1680

tgagcaatgg aagcgggtag gcctttgggg cagcggccaa tagcagcttt gctccttcgc 1740

tttctgggct cagaggctgg gaaggggtgg gtccgggggc gggctcaggg gcgggctcag 1800

gggcggggcg ggcgcccgaa ggtcctccgg aggcccggca ttctgcacgc ttcaaaagcg 1860

cacgtctgcc gcgctgttct cctcttcctc atctccgggc ctttcgacct gcagccaacg 1920

ccaccatggg gaccgagtac aagcccacgg tgcgcctcgc cacccgcgac gacgtccccc 1980

gggccgtacg caccctcgcc gccgcgttcg ccgactaccc cgccacgcgc cacaccgtcg 2040

acccggaccg ccacatcgag cgggtcaccg agctgcaaga actcttcctc acgcgcgtcg 2100

ggctcgacat cggcaaggtg tgggtcgcgg acgacggcgc cgcggtggcg gtctggacca 2160

cgccggagag cgtcgaagcg ggggcggtgt tcgccgagat cggcccgcgc atggccgagt 2220

tgagcggttc ccggctggcc gcgcagcaac agatggaagg cctcctggcg ccgcaccggc 2280

ccaaggagcc cgcgtggttc ctggccaccg tcggcgtctc gcccgaccac cagggcaagg 2340

gtctgggcag cgccgtcgtg ctccccggag tggaggcggc cgagcgcgcc ggggtgcccg 2400

ccttcctgga gacctccgcg ccccgcaacc tccccttcta cgagcggctc ggcttcaccg 2460

tcaccgccga cgtcgaggtg cccgaaggac cgcgcacctg gtgcatgacc cgcaagcccg 2520

gtgccggatc catgcccacg ctactgcggg tttatataga cggtcctcac gggatgggga 2580

aaaccaccac cacgcaactg ctggtggccc tgggttcgcg cgacgatatc gtctacgtac 2640

ccgagccgat gacttactgg caggtgctgg gggcttccga gacaatcgcg aacatctaca 2700

ccacacaaca ccgcctcgac cagggtgaga tatcggccgg ggacgcggcg gtggtaatga 2760

caagcgccca gataacaatg ggcatgcctt atgccgtgac cgacgccgtt ctggctcctc 2820

atatcggggg ggaggctggg agctcacatg ccccgccccc ggccctcacc ctcatcttcg 2880

accgccatcc catcgccgcc ctcctgtgct acccggccgc gcgatacctt atgggcagca 2940

tgaccccca ggccgtgctg gcgttcgtgg ccctcatccc gccgaccttg cccggcacaa 3000

acatcgtgtt gggggccctt ccggaggaca gacacatcga ccgcctggcc aaacgccagc 3060

gccccggcga gcggcttgac ctggctatgc tggccgcgat tcgccgcgtt tacgggctgc 3120

ttgccaatac ggtgcggtat ctgcagggcg gcgggtcgtg gcgggaggat tggggacagc 3180

tttcggggac ggccgtgccg ccccagggtg ccgagcccca gagcaacgcg ggcccacgac 3240

cccatatcgg ggacacgtta tttaccctgt ttcgggcccc cgagttgctg gcccccaacg 3300

gcgacctgta caacgtgttt gcctgggcct tggacgtctt ggccaaacgc ctccgtccca 3360

tgcacgtctt tatcctggat tacgaccaat cgcccgccgg ctgccgggac gccctgctgc 3420

aacttacctc cgggatggtc cagacccacg tcaccacccc cggctccata ccgacgatct 3480

gcgacctggc gcgcacgttt gcccgggaga tgggggaggc taactgagct ctagagctcg 3540

ctgatcagcc tcgactgtgc cttctagttg ccagccatct gttgtttgcc cctcccccgt 3600

gccttccttg accctggaag gtgccactcc cactgtcctt tcctaataaa atgaggaaat 3660

tgcatcgcat tgtctgagta ggtgtcattc tattctgggg ggtggggtgg ggcaggacag 3720

caagggggag gattgggaag acaatagcag gcatgctggg gatgcggtgg gctctatggc 3780

ttctgaggcg gaaagaacca gctggggctc gagatccact agttctagcc tcgaggctag 3840

agcggccggc cctataactt cgtataatgt atgctatacg aagttatcag gtaagttaac 3900

agggcgcgcc cactagggac aggattggtg aca 3933

<210> 14
<211> 20
<212> DNA
<213> Artificial Sequence

<220>
<223> Fragment of pD4 including the end of the AAVS1 right homology arm (RHA)

<400> 14
tctgctctga cgcgtgtata 20

<210> 15
<211> 53
<212> PRT
<213> Artificial Sequence

<220>
<223> Fragment of pD4 comprising T2A and start of EGFP sequence

<400> 15

Gly Ser Gly Glu Gly Arg Gly Ser Leu Leu Thr Cys Gly Asp Val Glu
1 5 10 15

Glu Asn Pro Gly Pro Met Val Ser Lys Gly Glu Glu Leu Phe Thr Gly
20 25 30

Val Val Pro Ile Leu Val Glu Leu Asp Gly Asp Val Asn Gly His Lys
35 40 45

Phe Ser Val Ser Gly
50

<210> 16
<211> 69
<212> PRT
<213> Artificial Sequence

<220>
<223> Fragment of pD4 comprising terminal part of EGFP sequence and Mu ornithine decarboxylase PEST sequence

<400> 16

Asn Glu Lys Arg Asp His Met Val Leu Leu Glu Phe Val Thr Ala Ala
1 5 10 15

Gly Ile Thr His Gly Met Asp Glu Leu Tyr Lys Lys Leu Ser His Gly
20 25 30

Phe Pro Pro Ala Val Ala Ala Gln Asp Asp Gly Thr Leu Pro Met Ser
35 40 45

Cys Ala Gln Glu Ser Gly Met Asp Arg His Pro Ala Ala Cys Ala Ser
50 55 60

Ala Arg Ile Asn Val
65

<210> 17
<211> 18
<212> PRT
<213> Artificial Sequence

<220>
<223> Initial fragment of of purodelta TK sequence encoded by pD4

<400> 17

Met Gly Thr Glu Tyr Lys Pro Thr Val Arg Leu Ala Thr Arg Asp Asp
1 5 10 15

Val Pro

<210> 18
<211> 15
<212> PRT
<213> Artificial Sequence

<220>
<223> Terminal fragment of puromycin delta TK sequence encoded by pD4

<400> 18

Cys Asp Leu Ala Arg Thr Phe Ala Arg Glu Met Gly Glu Ala Asn
1 5 10 15

<210> 19
<211> 1098
<212> DNA
<213> Artificial Sequence

<220>
<223> Sequence of donor plasmid for integration into Flp/GFP TALEN sites (pD5), fragment preceding D1.3 antibody expression cassette

<400> 19
ggtaccgaat tcctagctct tccagccccc tgtcatggca tcttccaggg gtccgagagc 60

tcagctagtc ttcttcctcc aacccgggcc cctatgtcca cttcaggaca gcatgtttgc 120

tgcctccagg gatcctgtgt ccccgagctg ggaccacctt atattcccag ggccggttaa 180

tgtggctctg gttctgggta cttttatctg tcccctccac cccacagtgg ggcaagatgc 240

atgaagttcc tattccgaag ttcctattct ctagaaagta taggaacttc gaccataact 300

tcgtataaag tatcctatac gaagttatgc gatcgctcgc gcgtagggat aacagggtaa 360

taagtccacc ggtcgccacc atggtcttct gacctcttct cttcctccca cagggcatgg 420

caaaacctct gagccaggaa gaaagcacac tgattgaaag agcaaccgct actatcaaca 480

gcatccccat ctccgaagac tattctgtgg ctagtgccgc tctgtccagc gacgggagaa 540

tcttcaccgg tgtgaacgtc taccacttta caggcggacc atgcgcagag ctggtggtcc 600

tggggactgc agccgctgca gccgctggta atctgacctg tatcgtggcc attggcaacg 660

aaaatagggg catcctgtcc ccatgcggca ggtgtcggca ggtgctgctg gatctgcatc 720

ctggcatcaa ggcaattgtc aaagactctg atggacagcc taccgccgtc ggtatccgtg 780

aactgctgcc tagcggctat gtctgggagg gataatgaat gagcttggct tcgaaatgac 840

cgaccaagcg acgcccaacc tgccatcacg agatttcgat tccaccgccg ccttctatga 900

aaggttgggc ttcggaatcg ttttccggga cgccggctgg atgatcctcc agcgcgggga 960

tctcatgctg gagttcttcg cccaccccaa cttgtttatt gcagcttata atggttacaa 1020

ataaagcaat agcatcacaa atttcacaaa taaagcattt ttttcactgc attctagttg 1080

tggtttaatt aacaattc 1098

<210> 20
<211> 931
<212> DNA
<213> Artificial Sequence

<220>
<223> Sequence of donor plasmid for integration into Flp/GFP TALEN sites (pD5), fragment following D1.3 antibody expression cassette

<400> 20
caggcatgct ggggatggcc cgggcatgat aacttcgtat aatgtatgct atacgaagtt 60

atgtatacgg cgcgcccgag caagggcgag gagctgttca cttctgacct cttctcttcc 120

tcccacctga gcctagagag atctggcagc ggagagggca gaggaagtct tctaacatgc 180

ggtgacgtgg aggagaatcc cggaccgtga gtgagcaagg gagaagaact cttcaccggg 240

gtggtgccca tcctggtcga gctggacggc gacgtgaacg gccacaagtt cagcgtgtcc 300

ggcgagggcg agggcgatgc cacctacggc aagctgaccc tgaagttcat ctgcaccacc 360

ggcaagctgc ccgtgccctg gcccaccctc gtgaccaccc tgacctacgg cgtgcagtgc 420

ttcagccgct accccgacca catgaagcag cacgacttct tcaagtccgc catgcccgaa 480

ggctacgtcc aggagcgcac catcttcttc aaggacgacg gcaactacaa gacccgcgcc 540

gaggtgaagt tcgagggcga caccctggtg aaccgcatcg agctgaaggg catcgacttc 600

aaggaggacg gcaacatcct ggggcacaag ctggagtaca actacaacag ccacaacgtc 660

tatatcatgg ccgacaagca gaagaacggc atcaaggtga acttcaagat ccgccacaac 720

atcgaggacg gcagcgtgca gctcgccgac cactaccagc agaacacccc catcggcgac 780

ggccccgtgc tgctgcccga caaccactac ctgagcaccc agtccgccct gagcaaagac 840

cccaacgaga agcgcgatca catggtcctg ctggagttcg tgaccgccgc cgggatcact 900

cacggcatgg acgagcctga cgcgtgtata c 931

<210> 21
<211> 131
<212> PRT
<213> Artificial Sequence

<220>
<223> Sequence of Blasticidin resistance protein encoded by donor plasmid for integration into FLP/GFP TALEN sites (pD5)

<400> 21

Ala Lys Pro Leu Ser Gln Glu Glu Ser Thr Leu Ile Glu Arg Ala Thr
1 5 10 15

Ala Thr Ile Asn Ser Ile Pro Ile Ser Glu Asp Tyr Ser Val Ala Ser
20 25 30

Ala Ala Leu Ser Ser Asp Gly Arg Ile Phe Thr Gly Val Asn Val Tyr
35 40 45

His Phe Thr Gly Gly Pro Cys Ala Glu Leu Val Val Leu Gly Thr Ala
50 55 60

Ala Ala Ala Ala Ala Gly Asn Leu Thr Cys Ile Val Ala Ile Gly Asn
65 70 75 80

Glu Asn Arg Gly Ile Leu Ser Pro Cys Gly Arg Cys Arg Gln Val Leu
85 90 95

Leu Asp Leu His Pro Gly Ile Lys Ala Ile Val Lys Asp Ser Asp Gly
100 105 110

Gln Pro Thr Ala Val Gly Ile Arg Glu Leu Leu Pro Ser Gly Tyr Val
115 120 125

Trp Glu Gly
130

<210> 22
<211> 813
<212> DNA
<213> Artificial Sequence

<220>
<223> Sequence of I-Sce-1 meganuclease construct

<400> 22
ccatgggcta tccttacgat gtccctgatt acgccaacag tcctggtatc cctggtatgg 60

gtcctaaaaa gaagcgaaaa gtgggtagac tggaacccgg catgaagaac attaagaaaa 120

atcaggtgat gaacctggga cctaattcca agctgctgaa agagtacaag tctcagctga 180

tcgaactgaa cattgagcag tttgaagcag ggatcggtct gattctgggg gacgcctaca 240

tccggagcag ggatgagggc aagacttatt gcatgcagtt cgaatggaag aataaggcct 300

acatggacca cgtgtgtctg ctgtatgatc agtgggtcct gtctcccccc cacaagaaag 360

agagagtgaa ccatctgggc aatctggtca ttacttgggg agcacagacc ttcaagcatc 420

aggcctttaa caaactggct aacctgttca tcgtgaacaa caagaaaacc atccctaaca 480

atctggtcga aaactacctg acaccaatga gtctggccta ttggttcatg gacgatggcg 540

gaaaatggga ctacaacaag aacagcacaa acaaaagcat cgtgctgaat acccagtcct 600

tcacatttga ggaagtggag tatctggtca agggcctgcg gaacaaattc cagctgaact 660

gctacgtgaa gatcaacaag aacaagccaa tcatctacat cgattctatg agttacctga 720

tcttttataa cctgattaag ccatacctga tcccccagat gatgtataaa ctgcccaata 780

caatcagctc cgagactttc ctgaaggtct aga 813

<210> 23
<211> 269
<212> PRT
<213> Artificial Sequence

<220>
<223> Sequence encoded by I-Sce1 meganuclease construct: HA tag, NLS, and I-Sce1

<400> 23

Met Gly Tyr Pro Tyr Asp Val Pro Asp Tyr Ala Asn Ser Pro Gly Ile
1 5 10 15

Pro Gly Met Gly Pro Lys Lys Lys Arg Lys Val Gly Arg Leu Glu Pro
20 25 30

Gly Met Lys Asn Ile Lys Lys Asn Gln Val Met Asn Leu Gly Pro Asn
35 40 45

Ser Lys Leu Leu Lys Glu Tyr Lys Ser Gln Leu Ile Glu Leu Asn Ile
50 55 60

Glu Gln Phe Glu Ala Gly Ile Gly Leu Ile Leu Gly Asp Ala Tyr Ile
65 70 75 80

Arg Ser Arg Asp Glu Gly Lys Thr Tyr Cys Met Gln Phe Glu Trp Lys
85 90 95

Asn Lys Ala Tyr Met Asp His Val Cys Leu Leu Tyr Asp Gln Trp Val
100 105 110

Leu Ser Pro Pro His Lys Lys Glu Arg Val Asn His Leu Gly Asn Leu
115 120 125

Val Ile Thr Trp Gly Ala Gln Thr Phe Lys His Gln Ala Phe Asn Lys
130 135 140

```
Leu Ala Asn Leu Phe Ile Val Asn Asn Lys Lys Thr Ile Pro Asn Asn
145                 150                 155                 160


Leu Val Glu Asn Tyr Leu Thr Pro Met Ser Leu Ala Tyr Trp Phe Met
                165                 170                 175


Asp Asp Gly Gly Lys Trp Asp Tyr Asn Lys Asn Ser Thr Asn Lys Ser
            180                 185                 190


Ile Val Leu Asn Thr Gln Ser Phe Thr Phe Glu Glu Val Glu Tyr Leu
        195                 200                 205


Val Lys Gly Leu Arg Asn Lys Phe Gln Leu Asn Cys Tyr Val Lys Ile
    210                 215                 220


Asn Lys Asn Lys Pro Ile Ile Tyr Ile Asp Ser Met Ser Tyr Leu Ile
225                 230                 235                 240


Phe Tyr Asn Leu Ile Lys Pro Tyr Leu Ile Pro Gln Met Met Tyr Lys
                245                 250                 255


Leu Pro Asn Thr Ile Ser Ser Glu Thr Phe Leu Lys Val
            260                 265
```

<210> 24
<211> 67
<212> DNA
<213> Artificial Sequence

<220>
<223> pINT20 Vector fragment showing Nsi1 site, splice acceptor site and start of Puromycin resistance gene sequence

<400> 24
atgcatcttc tgacctcttc tcttcctccc acagggcatg accgagtaca agcccacggt 60

gcgcctc 67

<210> 25
<211> 10
<212> PRT
<213> Artificial Sequence

<220>
<223> Initial fragment of puromycin resistance protein encoded by pINT20 vector

<400> 25

```
Met Thr Glu Tyr Lys Pro Thr Val Arg Leu
1               5                   10
```

<210> 26
<211> 1125
<212> DNA
<213> Artificial Sequence

<220>
<223> Sequence of T cell receptor clone c12/c2 alpha chain construct containing Nhe1, Not1 and Acc651 restriction sites

<400> 26
gctagcaagc aggaagtgac tcagatccca gccgctctga gcgtgcctga gggagaaaac 60

ctggtcctga attgcagttt caccgactca gccatctata acctgcagtg gtttcgccag 120

gatccaggca agggactgac ctccctgctg ctgattcaga gctcccagag ggaacagaca 180

tctggcagac tgaatgctag tctggacaaa tctagtggac ggtctaccct gtacatcgca 240

gccagccagc ctggagattc cgcaacatat ctgtgcgccg tgcgcccact tacaggcgga 300

agctacattc ccaccttcgg gcgaggtaca agcctgatcg tgcacccaga catccagaat 360

ccggagcccg ccgtatacca gctgaaggac cccagaagcc aggacagcac cctgtgcctg 420

ttcaccgact tcgacagcca gatcaacgtg cccaagacaa tggaaagcgg caccttcatc 480

accgacaaga ccgtgctgga catgaaggct atggacagca agagcaacgg cgccattgcc 540

tggtccaacc agaccagctt cacatgccag gacatcttca aagagacaaa cgccacctac 600

cccagcagcg acgtgccctg tgatgccacc ctgaccgaga agtccttcga gacagacatg 660

aacctgaact tccagaacct gtccgcggcc gcaggcctgc tggatcccaa gctgtgctac 720

ctgctggacg ggatcctgtt catctacggt gtgatcctga ctgccctgtt cctgcgagtc 780

aaattttctc ggagtgccga cgctcctgca taccagcagg ggcagaacca gctgtataac 840

gagctgaatc tgggtcggag ggaggaatat gacgtgctgg ataagagacg cggcagggat 900

ccagaaatgg ggggcaagcc ccagcgacgg aaaaaccctc aggagggact gtataatgaa 960

ctgcagaagg acaaaatggc cgaggcttac tctgaaattg ggatgaaggg cgagaggaga 1020

cgcggcaaag gacacgatgg cctgtaccag ggactgagca ctgctaccaa ggacacatat 1080

gatgctctgc atatgcaggc actgcccccc agataataag gtacc 1125

<210> 27
<211> 369
<212> PRT
<213> Artificial Sequence

<220>
<223> Sequence of T cell receptor clone c12/c2 alpha chain

<400> 27

Lys Gln Glu Val Thr Gln Ile Pro Ala Ala Leu Ser Val Pro Glu Gly
1 5 10 15

```
Glu Asn Leu Val Leu Asn Cys Ser Phe Thr Asp Ser Ala Ile Tyr Asn
            20                  25                  30

Leu Gln Trp Phe Arg Gln Asp Pro Gly Lys Gly Leu Thr Ser Leu Leu
        35                  40                  45

Leu Ile Gln Ser Ser Gln Arg Glu Gln Thr Ser Gly Arg Leu Asn Ala
    50                  55                  60

Ser Leu Asp Lys Ser Ser Gly Arg Ser Thr Leu Tyr Ile Ala Ala Ser
65                  70                  75                  80

Gln Pro Gly Asp Ser Ala Thr Tyr Leu Cys Ala Val Arg Pro Leu Thr
                85                  90                  95

Gly Gly Ser Tyr Ile Pro Thr Phe Gly Arg Gly Thr Ser Leu Ile Val
            100                 105                 110

His Pro Asp Ile Gln Asn Pro Glu Pro Ala Val Tyr Gln Leu Lys Asp
        115                 120                 125

Pro Arg Ser Gln Asp Ser Thr Leu Cys Leu Phe Thr Asp Phe Asp Ser
    130                 135                 140

Gln Ile Asn Val Pro Lys Thr Met Glu Ser Gly Thr Phe Ile Thr Asp
145                 150                 155                 160

Lys Thr Val Leu Asp Met Lys Ala Met Asp Ser Lys Ser Asn Gly Ala
                165                 170                 175

Ile Ala Trp Ser Asn Gln Thr Ser Phe Thr Cys Gln Asp Ile Phe Lys
            180                 185                 190

Glu Thr Asn Ala Thr Tyr Pro Ser Ser Asp Val Pro Cys Asp Ala Thr
        195                 200                 205

Leu Thr Glu Lys Ser Phe Glu Thr Asp Met Asn Leu Asn Phe Gln Asn
    210                 215                 220

Leu Ser Ala Ala Ala Gly Leu Leu Asp Pro Lys Leu Cys Tyr Leu Leu
225                 230                 235                 240

Asp Gly Ile Leu Phe Ile Tyr Gly Val Ile Leu Thr Ala Leu Phe Leu
                245                 250                 255

Arg Val Lys Phe Ser Arg Ser Ala Asp Ala Pro Ala Tyr Gln Gln Gly
            260                 265                 270
```

```
Gln Asn Gln Leu Tyr Asn Glu Leu Asn Leu Gly Arg Arg Glu Glu Tyr
        275                 280                 285

Asp Val Leu Asp Lys Arg Arg Gly Arg Asp Pro Glu Met Gly Gly Lys
    290                 295                 300

Pro Gln Arg Arg Lys Asn Pro Gln Glu Gly Leu Tyr Asn Glu Leu Gln
305                 310                 315                 320

Lys Asp Lys Met Ala Glu Ala Tyr Ser Glu Ile Gly Met Lys Gly Glu
                325                 330                 335

Arg Arg Arg Gly Lys Gly His Asp Gly Leu Tyr Gln Gly Leu Ser Thr
            340                 345                 350

Ala Thr Lys Asp Thr Tyr Asp Ala Leu His Met Gln Ala Leu Pro Pro
        355                 360                 365

Arg
```

<210> 28
<211> 1226
<212> DNA
<213> Artificial Sequence

<220>
<223> Sequence of T cell receptor clone c12/c2 beta chain construct containing Nco1, Xho1 and Hind3 restriction sites

<400> 28

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ccatggccaa cgctggagtg actcagaccc ctaagttcca ggtcctgaaa actgggcaga       60

gtatgaccct gcagtgcgca caggacatga atcacgagta catgtcatgg tatcggcagg      120

atccagggat gggtctgagg ctgatccatt acagcgtggg cgctggaact accgaccagg      180

gcgaggtgcc caacggatat aatgtctcaa gaagcaccac agaagatttc ccactgcgac      240

tgctgagcgc cgctcctagc cagacatccg tgtacttttg cgccagctcc aatgtcggga      300

acaccggcga gctgttcttt ggggaaggtt cccgcctgac agtgctcgag gacctgagaa      360

acgtgacccc ccccaaggtg tccctgttcg agcctagcaa ggccgagatc gccaacaagc      420

agaaagccac cctcgtgtgc ctggccagag gcttcttccc cgaccacgtg gaactgtctt      480

ggtgggtcaa cggcaaagag gtgcacagcg gcgtgtccac cgatccccag gcctacaaag      540

agagcaacta cagctactgc ctgagcagca gactgcgggt gtccgccacc ttctggcaca      600

acccccggaa ccacttcaga tgccaggtgc agtttcacgg cctgagcgaa gaggacaagt      660

ggcccgaggg cagccctaag cccgtgaccc agaatatctc tgccgaagcc tggggcagag      720
```

ccgactgtgg cattaccagc gccagctacc agcagggcgt gctgtctgcc accatcctgt 780

acgaggtcgc gagcggactg ctggacccaa agctgtgcta cctgctggat gggatcctgt 840

tcatctacgg tgtgattctg acagccctgt tcctgcgagt caagttcagc cggagcgccg 900

acgcaccagc ataccagcag gggcagaatc agctgtataa cgagctgaat ctgggtcgga 960

gggaggaata cgacgtgctg gataagagac gcggcaggga tcccgaaatg ggcggaaagc 1020

ctcagcgacg gaaaaaccca caggagggac tgtacaatga actgcagaag gacaaaatgg 1080

ctgaggcata ttctgaaatc ggcatgaagg gagagaggag acgcggcaaa ggacacgatg 1140

ggctgtacca gggtctgagt acagccacta aggacaccta tgatgccctg catatgcagg 1200

ctctgccacc cagataataa aagctt 1226

<210> 29
<211> 404
<212> PRT
<213> Artificial Sequence

<220>
<223> Sequence of T cell receptor clone c12/c2 beta chain construct

<400> 29

Met Ala Asn Ala Gly Val Thr Gln Thr Pro Lys Phe Gln Val Leu Lys
1 5 10 15

Thr Gly Gln Ser Met Thr Leu Gln Cys Ala Gln Asp Met Asn His Glu
20 25 30

Tyr Met Ser Trp Tyr Arg Gln Asp Pro Gly Met Gly Leu Arg Leu Ile
35 40 45

His Tyr Ser Val Gly Ala Gly Thr Thr Asp Gln Gly Glu Val Pro Asn
50 55 60

Gly Tyr Asn Val Ser Arg Ser Thr Thr Glu Asp Phe Pro Leu Arg Leu
65 70 75 80

Leu Ser Ala Ala Pro Ser Gln Thr Ser Val Tyr Phe Cys Ala Ser Ser
85 90 95

Asn Val Gly Asn Thr Gly Glu Leu Phe Phe Gly Glu Gly Ser Arg Leu
100 105 110

Thr Val Leu Glu Asp Leu Arg Asn Val Thr Pro Pro Lys Val Ser Leu
115 120 125

Phe Glu Pro Ser Lys Ala Glu Ile Ala Asn Lys Gln Lys Ala Thr Leu
130 135 140

```
Val Cys Leu Ala Arg Gly Phe Phe Pro Asp His Val Glu Leu Ser Trp
145                 150                 155                 160

Trp Val Asn Gly Lys Glu Val His Ser Gly Val Ser Thr Asp Pro Gln
                165                 170                 175

Ala Tyr Lys Glu Ser Asn Tyr Ser Tyr Cys Leu Ser Ser Arg Leu Arg
            180                 185                 190

Val Ser Ala Thr Phe Trp His Asn Pro Arg Asn His Phe Arg Cys Gln
        195                 200                 205

Val Gln Phe His Gly Leu Ser Glu Glu Asp Lys Trp Pro Glu Gly Ser
    210                 215                 220

Pro Lys Pro Val Thr Gln Asn Ile Ser Ala Glu Ala Trp Gly Arg Ala
225                 230                 235                 240

Asp Cys Gly Ile Thr Ser Ala Ser Tyr Gln Gln Gly Val Leu Ser Ala
                245                 250                 255

Thr Ile Leu Tyr Glu Val Ala Ser Gly Leu Leu Asp Pro Lys Leu Cys
            260                 265                 270

Tyr Leu Leu Asp Gly Ile Leu Phe Ile Tyr Gly Val Ile Leu Thr Ala
        275                 280                 285

Leu Phe Leu Arg Val Lys Phe Ser Arg Ser Ala Asp Ala Pro Ala Tyr
    290                 295                 300

Gln Gln Gly Gln Asn Gln Leu Tyr Asn Glu Leu Asn Leu Gly Arg Arg
305                 310                 315                 320

Glu Glu Tyr Asp Val Leu Asp Lys Arg Arg Gly Arg Asp Pro Glu Met
                325                 330                 335

Gly Gly Lys Pro Gln Arg Arg Lys Asn Pro Gln Glu Gly Leu Tyr Asn
            340                 345                 350

Glu Leu Gln Lys Asp Lys Met Ala Glu Ala Tyr Ser Glu Ile Gly Met
        355                 360                 365

Lys Gly Glu Arg Arg Arg Gly Lys Gly His Asp Gly Leu Tyr Gln Gly
    370                 375                 380

Leu Ser Thr Ala Thr Lys Asp Thr Tyr Asp Ala Leu His Met Gln Ala
```

385 390 395 400

Leu Pro Pro Arg

<210> 30
<211> 677
<212> DNA
<213> Artificial Sequence

<220>
<223> Sequence of T cell receptor clone 4JFH alpha chain construct with initial Nhe1 and terminal Not1 restriction sites

<400> 30
gctagccaga aagaggtgga acagaacagc ggccctctga gcgtgccaga aggcgctatc 60

gccagcctga actgcaccta cagctttctg ggcagccaga gcttcttctg gtacagacag 120

tacagcggca agagccccga gctgatcatg ttcacctaca gagagggcga caaagaggac 180

ggcagattca ccgcccagct gaacaaggcc agccagcacg tgtccctgct gatcagagac 240

agccagccta gcgacagcgc cacctacctg tgcgccgtga atgatggcgg cagactgacc 300

tttggcgacg gcaccaccct gaccgtgaag cctgacatcc agaatccgga gcccgccgta 360

taccagctga aggaccccag aagccaggac agcaccctgt gcctgttcac cgacttcgac 420

agccagatca acgtgcccaa gacaatggaa agcggcacct tcatcaccga caagaccgtg 480

ctggacatga aggctatgga cagcaagagc aacggcgcca ttgcctggtc caaccagacc 540

agcttcacat gccaggacat cttcaaagag acaaacgcca cctaccccag cagcgacgtg 600

ccctgtgatg ccaccctgac cgagaagtcc ttcgagacag acatgaacct gaacttccag 660

aacctgtccg cggccgc 677

<210> 31
<211> 221
<212> PRT
<213> Artificial Sequence

<220>
<223> Sequence of T cell receptor clone 4JFH alpha chain construct

<400> 31

Gln Lys Glu Val Glu Gln Asn Ser Gly Pro Leu Ser Val Pro Glu Gly
1 5 10 15

Ala Ile Ala Ser Leu Asn Cys Thr Tyr Ser Phe Leu Gly Ser Gln Ser
20 25 30

Phe Phe Trp Tyr Arg Gln Tyr Ser Gly Lys Ser Pro Glu Leu Ile Met
35 40 45

```
Phe Thr Tyr Arg Glu Gly Asp Lys Glu Asp Gly Arg Phe Thr Ala Gln
    50                  55                  60

Leu Asn Lys Ala Ser Gln His Val Ser Leu Leu Ile Arg Asp Ser Gln
65                  70                  75                  80

Pro Ser Asp Ser Ala Thr Tyr Leu Cys Ala Val Asn Asp Gly Gly Arg
                85                  90                  95

Leu Thr Phe Gly Asp Gly Thr Thr Leu Thr Val Lys Pro Asp Ile Gln
            100                 105                 110

Asn Pro Glu Pro Ala Val Tyr Gln Leu Lys Asp Pro Arg Ser Gln Asp
        115                 120                 125

Ser Thr Leu Cys Leu Phe Thr Asp Phe Asp Ser Gln Ile Asn Val Pro
    130                 135                 140

Lys Thr Met Glu Ser Gly Thr Phe Ile Thr Asp Lys Thr Val Leu Asp
145                 150                 155                 160

Met Lys Ala Met Asp Ser Lys Ser Asn Gly Ala Ile Ala Trp Ser Asn
                165                 170                 175

Gln Thr Ser Phe Thr Cys Gln Asp Ile Phe Lys Glu Thr Asn Ala Thr
            180                 185                 190

Tyr Pro Ser Ser Asp Val Pro Cys Asp Ala Thr Leu Thr Glu Lys Ser
        195                 200                 205

Phe Glu Thr Asp Met Asn Leu Asn Phe Gln Asn Leu Ser
    210                 215                 220


<210>  32
<211>  353
<212>  DNA
<213>  Artificial Sequence

<220>
<223>  Sequence of T cell receptor clone4JFH beta chain construct with
       initial Nco1 restriction site and terminal Xho1 restriction site

<400>  32
ccatggccag ccagaccatc catcagtggc ctgccaccct ggtgcagcct gtgggatctc       60

ctctgagcct ggaatgcacc gtggaaggca ccagcaaccc caacctgtac tggtacagac      120

aggccgctgg cagaggcccc cagctgctgt tttactgggg ccccttggc cagatcagca       180

gcgaggtgcc ccagaacctg agcgccagca gaccccagga ccggcagttt atcctgagca      240
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gcaagaagct gctgctgagc gacagcggct tctacctgtg cgcttggagc gagacaggcc 300

tgggcatggg cggatggcag tttggcgagg gcagcagact gacagtgctc gag 353

<210> 33
<211> 117
<212> PRT
<213> Artificial Sequence

<220>
<223> Sequence of T cell receptor clone 4JFH beta chain construct

<400> 33

Met Ala Ser Gln Thr Ile His Gln Trp Pro Ala Thr Leu Val Gln Pro
1 5 10 15

Val Gly Ser Pro Leu Ser Leu Glu Cys Thr Val Glu Gly Thr Ser Asn
20 25 30

Pro Asn Leu Tyr Trp Tyr Arg Gln Ala Ala Gly Arg Gly Pro Gln Leu
35 40 45

Leu Phe Tyr Trp Gly Pro Phe Gly Gln Ile Ser Ser Glu Val Pro Gln
50 55 60

Asn Leu Ser Ala Ser Arg Pro Gln Asp Arg Gln Phe Ile Leu Ser Ser
65 70 75 80

Lys Lys Leu Leu Leu Ser Asp Ser Gly Phe Tyr Leu Cys Ala Trp Ser
85 90 95

Glu Thr Gly Leu Gly Met Gly Gly Trp Gln Phe Gly Glu Gly Ser Arg
100 105 110

Leu Thr Val Leu Glu
115

<210> 34
<211> 53
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer for mutation of CDR3 of c12/c2 TCR alpha chain

<220>
<221> misc_feature
<222> (20)..(21)
<223> n is a, c, g, or t

<220>
<221> misc_feature

<222> (23)..(24)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (31)..(31)
<223> n is a, c, g, or t

<400> 34
cacgcggcac gcgggtgaan nsnnscctwc natgtaaggg tggaagcccg ctc 53

<210> 35
<211> 52
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer used to mutate CDR3 of c12/c2 TCR alpha chain

<220>
<221> misc_feature
<222> (23)..(23)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (30)..(31)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (33)..(34)
<223> n is a, c, g, or t

<400> 35
ctcgcccgaa ggtgggaatg tangwtccsn nsnnaagtgg gcgcacggcg ca 52

<210> 36
<211> 17
<212> PRT
<213> Artificial Sequence

<220>
<223> Strategy used to mutate CDR3 of c12/c2 TCR alpha chain

<220>
<221> misc_feature
<222> (7)..(8)
<223> Xaa can be any naturally occurring amino acid

<400> 36

Cys Ala Val Arg Pro Leu Xaa Xaa Gly Ser Tyr Ile Pro Thr Phe Gly
1 5 10 15

Arg

<210> 37
<211> 17
<212> PRT
<213> Artificial Sequence

<220>
<223> Strategy used to mutate CDR3 of c12/c2 TCR alpha chain

<220>
<221> misc_feature
<222> (7)..(8)
<223> Xaa can be any naturally occurring amino acid

<400> 37

Cys Ala Val Arg Pro Leu Xaa Xaa Gly Thr Tyr Ile Pro Thr Phe Gly
1 5 10 15

Arg

<210> 38
<211> 50
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer used to mutate CDR3 of c12/c2 TCR beta chain

<220>
<221> misc_feature
<222> (20)..(21)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (29)..(30)
<223> n is a, c, g, or t

<400> 38
gtacttttgc gccagctccn nsstcgggnn saccggcgag ctgttctttg 50

<210> 39
<211> 49
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer used to mutate CDR3 of c12/c2 TCR beta chain

<220>
<221> misc_feature
<222> (21)..(22)
<223> n is a, c, g, or t

<220>

<221> misc_feature
<222> (30)..(31)
<223> n is a, c, g, or t

<400> 39
caaagaacag ctcgccggts nncccgassn nggagctggc gcaaaagta 49

<210> 40
<211> 16
<212> PRT
<213> Artificial Sequence

<220>
<223> Strategy used to mutate CDR3 of c12/c2 TCR beta chain

<400> 40

Tyr Phe Cys Ala Ser Ser Asn Val Gly Asn Thr Gly Glu Leu Phe Phe
1 5 10 15

<210> 41
<211> 16
<212> PRT
<213> Artificial Sequence

<220>
<223> Strategy used to mutate CDR3 of c12/c2 TCR beta chain

<400> 41

Tyr Phe Cys Ala Ser Ser Asn Leu Gly Asn Thr Gly Glu Leu Phe Phe
1 5 10 15

<210> 42
<211> 462
<212> DNA
<213> Artificial Sequence

<220>
<223> Sequence of CD3 zeta in pINT21 CAR1

<400> 42
ggcgccatgg cccaggtcgc ggccgcaagc ggactgctgg acccaaagct gtgctacctg 60

ctggatggga tcctgttcat ctacggtgtg attctgacag ccctgttcct gcgagtcaag 120

ttcagccgga gcgccgacgc accagcatac cagcaggggc agaatcagct gtataacgag 180

ctgaatctgg gtcggaggga ggaatacgac gtgctggata agagacgcgg cagggatccc 240

gaaatgggcg gaaagcctca gcgacggaaa aacccacagg agggactgta caatgaactg 300

cagaaggaca aaatggctga ggcatattct gaaatcggca tgaagggaga gaggagacgc 360

ggcaaaggac acgatgggct gtaccagggt ctgagtacag ccactaagga cacctatgat 420

gccctgcata tgcaggctct gccacccaga taataaaagc tt 462

<210> 43

<211> 4
<212> PRT
<213> Artificial Sequence

<220>
<223> Fragment encoded by pINT21 CAR1 vector

<400> 43

Gly Ala Met Ala
1

<210> 44
<211> 141
<212> PRT
<213> Artificial Sequence

<220>
<223> CDR3 zeta sequence encoded by pINT21 CAR1

<400> 44

Ser Gly Leu Leu Asp Pro Lys Leu Cys Tyr Leu Leu Asp Gly Ile Leu
1 5 10 15

Phe Ile Tyr Gly Val Ile Leu Thr Ala Leu Phe Leu Arg Val Lys Phe
20 25 30

Ser Arg Ser Ala Asp Ala Pro Ala Tyr Gln Gln Gly Gln Asn Gln Leu
35 40 45

Tyr Asn Glu Leu Asn Leu Gly Arg Arg Glu Glu Tyr Asp Val Leu Asp
50 55 60

Lys Arg Arg Gly Arg Asp Pro Glu Met Gly Gly Lys Pro Gln Arg Arg
65 70 75 80

Lys Asn Pro Gln Glu Gly Leu Tyr Asn Glu Leu Gln Lys Asp Lys Met
85 90 95

Ala Glu Ala Tyr Ser Glu Ile Gly Met Lys Gly Glu Arg Arg Arg Gly
100 105 110

Lys Gly His Asp Gly Leu Tyr Gln Gly Leu Ser Thr Ala Thr Lys Asp
115 120 125

Thr Tyr Asp Ala Leu His Met Gln Ala Leu Pro Pro Arg
130 135 140

<210> 45
<211> 705
<212> DNA
<213> Artificial Sequence

<220>
<223> pINT21 CAR2 Sequence encoding CD3 zeta

<400> 45
gccatggccc aggtcgcggc cgcaacaaca accccagccc ccagacctcc tacccctgcc 60

cctacaattg ccagccagcc tctgagcctg aggcccgagg cttgtagacc agctgctggc 120

ggagccgtgc acaccagagg actggatttc gcctgcgaca tctacatctg ggcccctctg 180

gccggcacat gtggcgtgct gctgctgagc ctcgtgatca ccctgtactg caagcggggc 240

agaaagaaac tgctgtacat ctttaagcag cccttcatgc ggcccgtgca gaccacccag 300

gaagaggacg gctgctcctg cagattcccc gaggaagaag aaggcggctg cgagctgaga 360

gtgaagttca gcagatccgc cgacgccccc gcctacaagc agggccagaa ccagctgtac 420

aacgagctga acctgggcag acgggaagag tacgacgtgc tggacaagcg gagaggccgg 480

gacccagaga tgggcggaaa gcccagaaga aagaaccccc aggaaggcct gtataacgaa 540

ctgcagaaag acaaaatggc cgaggcctac agcgagatcg gaatgaaggg cgagcggaga 600

agaggcaagg ggcacgatgg cctgtaccag ggcctgagca ccgccaccaa ggacacctat 660

gacgccctgc acatgcaggc cctgcccccc agataataaa agctt 705

<210> 46
<211> 223
<212> PRT
<213> Artificial Sequence

<220>
<223> Sequence encoded by pINT21 CAR2: CD8 hinge / transmembrane region, 4-1BB and CD3 zeta

<400> 46

Thr Thr Thr Pro Ala Pro Arg Pro Pro Thr Pro Ala Pro Thr Ile Ala
1 5 10 15

Ser Gln Pro Leu Ser Leu Arg Pro Glu Ala Cys Arg Pro Ala Ala Gly
20 25 30

Gly Ala Val His Thr Arg Gly Leu Asp Phe Ala Cys Asp Ile Tyr Ile
35 40 45

Trp Ala Pro Leu Ala Gly Thr Cys Gly Val Leu Leu Leu Ser Leu Val
50 55 60

Ile Thr Leu Tyr Cys Lys Arg Gly Arg Lys Lys Leu Leu Tyr Ile Phe
65 70 75 80

Lys Gln Pro Phe Met Arg Pro Val Gln Thr Thr Gln Glu Glu Asp Gly
85 90 95

```
Cys Ser Cys Arg Phe Pro Glu Glu Glu Glu Gly Gly Cys Glu Leu Arg
            100                 105                 110

Val Lys Phe Ser Arg Ser Ala Asp Ala Pro Ala Tyr Lys Gln Gly Gln
        115                 120                 125

Asn Gln Leu Tyr Asn Glu Leu Asn Leu Gly Arg Arg Glu Glu Tyr Asp
    130                 135                 140

Val Leu Asp Lys Arg Arg Gly Arg Asp Pro Glu Met Gly Gly Lys Pro
145                 150                 155                 160

Arg Arg Lys Asn Pro Gln Glu Gly Leu Tyr Asn Glu Leu Gln Lys Asp
                165                 170                 175

Lys Met Ala Glu Ala Tyr Ser Glu Ile Gly Met Lys Gly Glu Arg Arg
            180                 185                 190

Arg Gly Lys Gly His Asp Gly Leu Tyr Gln Gly Leu Ser Thr Ala Thr
        195                 200                 205

Lys Asp Thr Tyr Asp Ala Leu His Met Gln Ala Leu Pro Pro Arg
    210                 215                 220
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<210> 47
<211> 750
<212> DNA
<213> Artificial Sequence

<220>
<223> Sequence of FMC63 H-L (anti-CD19 antibody)

<400> 47

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gccatggccg aagtgaaact gcaggagtct ggacccggcc tggtggcccc atctcagtct       60

ctgagcgtga cctgtaccgt gtccggcgtg tccctgcctg actatggcgt gtcctggatc      120

agacagcccc ccagaaaggg cctggaatgg ctgggagtga tctggggcag cgaaaccacc      180

tactacaaca gcgccctgaa gtcccggctg accatcatca aggacaactc caagagccag      240

gtgttcctga agatgaacag cctgcagacc gacgacaccg ccatctacta ctgcgccaag      300

cactactact acggcggcag ctacgctatg gactactggg gccagggcac ctcggtcacc      360

gtctcgagtg gtggaggcgg ttcaggcgga ggtggctctg gcggtggcgc tagcgacatc      420

cagatgaccc agaccaccag cagcctgagc gccagcctgg gcgatagagt gaccatcagc      480

tgcagagcca gccaggacat cagcaagtac ctgaactggt atcagcagaa acccgacggc      540

accgtgaagc tgctgatcta ccacaccagc agactgcaca gcggcgtgcc cagcagattt      600
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tccggctctg gcagcggcac cgactacagc ctgaccatct ccaacctgga acaggaagat 660

atcgctacct acttctgtca gcaaggcaac accctgccct acaccttcgg cggagggacc 720

aagctggaga tcaaacgtac cgcggccgca 750

<210> 48
<211> 250
<212> PRT
<213> Artificial Sequence

<220>
<223> Sequence of FMC63 H-L (anti-CD19 antibody)

<400> 48

Ala Met Ala Glu Val Lys Leu Gln Glu Ser Gly Pro Gly Leu Val Ala
1 5 10 15

Pro Ser Gln Ser Leu Ser Val Thr Cys Thr Val Ser Gly Val Ser Leu
20 25 30

Pro Asp Tyr Gly Val Ser Trp Ile Arg Gln Pro Pro Arg Lys Gly Leu
35 40 45

Glu Trp Leu Gly Val Ile Trp Gly Ser Glu Thr Thr Tyr Tyr Asn Ser
50 55 60

Ala Leu Lys Ser Arg Leu Thr Ile Ile Lys Asp Asn Ser Lys Ser Gln
65 70 75 80

Val Phe Leu Lys Met Asn Ser Leu Gln Thr Asp Asp Thr Ala Ile Tyr
85 90 95

Tyr Cys Ala Lys His Tyr Tyr Tyr Gly Gly Ser Tyr Ala Met Asp Tyr
100 105 110

Trp Gly Gln Gly Thr Ser Val Thr Val Ser Ser Gly Gly Gly Gly Ser
115 120 125

Gly Gly Gly Gly Ser Gly Gly Gly Ala Ser Asp Ile Gln Met Thr Gln
130 135 140

Thr Thr Ser Ser Leu Ser Ala Ser Leu Gly Asp Arg Val Thr Ile Ser
145 150 155 160

Cys Arg Ala Ser Gln Asp Ile Ser Lys Tyr Leu Asn Trp Tyr Gln Gln
165 170 175

Lys Pro Asp Gly Thr Val Lys Leu Leu Ile Tyr His Thr Ser Arg Leu
180 185 190

His Ser Gly Val Pro Ser Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp
195 200 205

Tyr Ser Leu Thr Ile Ser Asn Leu Glu Gln Glu Asp Ile Ala Thr Tyr
210 215 220

Phe Cys Gln Gln Gly Asn Thr Leu Pro Tyr Thr Phe Gly Gly Gly Thr
225 230 235 240

Lys Leu Glu Ile Lys Arg Thr Ala Ala Ala
245 250

<210> 49
<211> 327
<212> DNA
<213> Artificial Sequence

<220>
<223> Sequence of lox1 binding Adhiron lox1A

<400> 49
gccatggctg ctacaggcgt gcgggctgtg cccggcaatg agaacagcct ggaaatcgag 60

gaactggcca gattcgccgt ggacgagcac aacaagaaag agaacgccct gctggaattc 120

gtgcgggtcg tgaaggccaa agagcagtgg agcgaggccg acaacgactg gcacaccatg 180

tactacctga ccctggaagc caaggacggc ggcaagaaga agctgtacga ggccaaagtg 240

tgggtcaagc tggacctgga aacctggcag cacttcaact tcaaagagct ccaggaattc 300

aagcccgtgg gcgacgctgc ggccgcg 327

<210> 50
<211> 109
<212> PRT
<213> Artificial Sequence

<220>
<223> Sequence of Lox1 binding Adhiron lox1A

<400> 50

Ala Met Ala Ala Thr Gly Val Arg Ala Val Pro Gly Asn Glu Asn Ser
1 5 10 15

Leu Glu Ile Glu Glu Leu Ala Arg Phe Ala Val Asp Glu His Asn Lys
20 25 30

Lys Glu Asn Ala Leu Leu Glu Phe Val Arg Val Val Lys Ala Lys Glu
35 40 45

Gln Trp Ser Glu Ala Asp Asn Asp Trp His Thr Met Tyr Tyr Leu Thr

50 55 60

Leu Glu Ala Lys Asp Gly Gly Lys Lys Lys Leu Tyr Glu Ala Lys Val
65 70 75 80

Trp Val Lys Leu Asp Leu Glu Thr Trp Gln His Phe Asn Phe Lys Glu
85 90 95

Leu Gln Glu Phe Lys Pro Val Gly Asp Ala Ala Ala Ala
100 105

<210> 51
<211> 333
<212> DNA
<213> Artificial Sequence

<220>
<223> Sequence of lox1 binding Adhiron lox1B

<400> 51
gccatggctg ctacaggcgt gcgggctgtg cccggcaatg agaacagcct ggaaatcgag 60

gaactggcca gattcgccgt ggacgagcac aacaagaaag agaacgccct gctggaattc 120

gtgcgggtcg tgaaggccaa agagcaggaa cagcccatcg gcgagcaccc cgtgaacgac 180

accatgtact acctgaccct ggaagccaag gacggcggca agaagaagct gtacgaggcc 240

aaagtgtggg tcaagcggtg gctgcggttc accgagatct acaacttcaa agagctccag 300

gaattcaagc ccgtgggcga cgctgcggcc gcg 333

<210> 52
<211> 111
<212> PRT
<213> Artificial Sequence

<220>
<223> Sequence of lox1 binding Adhiron lox1B

<400> 52

Ala Met Ala Ala Thr Gly Val Arg Ala Val Pro Gly Asn Glu Asn Ser
1 5 10 15

Leu Glu Ile Glu Glu Leu Ala Arg Phe Ala Val Asp Glu His Asn Lys
20 25 30

Lys Glu Asn Ala Leu Leu Glu Phe Val Arg Val Val Lys Ala Lys Glu
35 40 45

Gln Glu Gln Pro Ile Gly Glu His Pro Val Asn Asp Thr Met Tyr Tyr
50 55 60

Leu Thr Leu Glu Ala Lys Asp Gly Gly Lys Lys Lys Leu Tyr Glu Ala
65 70 75 80

Lys Val Trp Val Lys Arg Trp Leu Arg Phe Thr Glu Ile Tyr Asn Phe
85 90 95

Lys Glu Leu Gln Glu Phe Lys Pro Val Gly Asp Ala Ala Ala Ala
100 105 110

<210> 53
<211> 54
<212> DNA
<213> Artificial Sequence

<220>
<223> Adhiron_mut1 primer

<220>
<221> misc_feature
<222> (23)..(24)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (26)..(27)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (29)..(30)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (32)..(33)
<223> n is a, c, g, or t

<400> 53
cagggtcagg tagtacatgg tsnnsnnsnn snnctgctct ttggccttca cgac 54

<210> 54
<211> 48
<212> DNA
<213> Artificial Sequence

<220>
<223> Adhiron_mut2 primer

<220>
<221> misc_feature
<222> (20)..(21)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (23)..(24)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (26)..(27)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (29)..(30)
<223> n is a, c, g, or t

<400> 54
ctggagctct ttgaagttsn nsnnsnnsnn cttgacccac actttggc 48

<210> 55
<211> 54
<212> DNA
<213> Artificial Sequence

<220>
<223> Mutagenic primer for construction of library of binders within loop 1 (adhiron mut 1)

<220>
<221> misc_feature
<222> (22)..(23)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (25)..(26)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (28)..(29)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (31)..(32)
<223> n is a, c, g, or t

<400> 55
gtcgtgaagg ccaaagagca gnnsnnsnns nnsaccatgt actacctgac cctg 54

<210> 56
<211> 18
<212> PRT
<213> Artificial Sequence

<220>
<223> Adhiron mut1 loop library variant sequence

<220>
<221> misc_feature
<222> (8)..(11)
<223> Xaa can be any naturally occurring amino acid

<400> 56

Val Val Lys Ala Lys Glu Gln Xaa Xaa Xaa Xaa Thr Met Tyr Tyr Leu
1 5 10 15

Thr Leu

<210> 57
<211> 48
<212> DNA
<213> Artificial Sequence

<220>
<223> Mutagenic primer for construction of library of binders within Adhiron mut2 loop

<220>
<221> misc_feature
<222> (19)..(20)
<223> n is a, c, g, or t

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<222> (22)..(23)
<223> n is a, c, g, or t

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<222> (25)..(26)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (28)..(29)
<223> n is a, c, g, or t

<400> 57
gccaaagtgt gggtcaagnn snnsnnsnns aacttcaaag agctccag 48

<210> 58
<211> 16
<212> PRT
<213> Artificial Sequence

<220>
<223> Adhiron mut 2 loop library variant sequence

<220>
<221> misc_feature
<222> (7)..(10)
<223> Xaa can be any naturally occurring amino acid

<400> 58

Ala Lys Val Trp Val Lys Xaa Xaa Xaa Xaa Asn Phe Lys Glu Leu Gln
1 5 10 15

<210> 59
<211> 105
<212> DNA
<213> Artificial sequence

<220>
<223> Trypsin binding Knottin MCoTI-IIsequence

<400> 59
gccatggccg gtgtgtgccc caagatcttg aaaaagtgcc gccgtgacag cgattgtccc 60

ggcgcctgca tctgccgcgg caatggctat tgcggagcgg ccgca 105

<210> 60
<211> 32
<212> PRT
<213> Artificial Sequence

<220>
<223> Trypsin binding Knottin MCo-TI-II sequence

<400> 60

Ala Met Ala Gly Val Cys Pro Lys Ile Leu Lys Lys Cys Arg Arg Asp
1 5 10 15

Ser Asp Cys Pro Gly Ala Cys Ile Cys Arg Gly Asn Gly Tyr Cys Gly
20 25 30

<210> 61
<211> 99
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer for creation of library of Knottin mutants

<220>
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<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (14)..(14)
<223> n is a, c, g, or t

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<221> misc_feature
<222> (17)..(17)
<223> n is a, c, g, or t

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<222> (20)..(20)
<223> n is a, c, g, or t

<220>

<221> misc_feature
<222> (23)..(23)
<223> n is a, c, g, or t

<220>
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<222> (26)..(26)
<223> n is a, c, g, or t

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<222> (29)..(29)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (32)..(32)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (35)..(35)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (38)..(38)
<223> n is a, c, g, or t

<400> 61
ggtgtgtgcv nsvnsvnsvn svnsvnsvns vnsvnsvnst gccgccgtga cagcgattgt 60

cccggcgcct gcatctgccg cggcaatggc tattgcgga 99

<210> 62
<211> 99
<212> DNA
<213> Artificial Sequence

<220>
<223> Sequence varied in library of knottin mutants

<220>
<221> misc_feature
<222> (62)..(62)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (65)..(65)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (68)..(68)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (71)..(71)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (74)..(74)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (77)..(77)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (80)..(80)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (83)..(83)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (86)..(86)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (89)..(89)
<223> n is a, c, g, or t

<400> 62
tccgcaatag ccattgccgc ggcagatgca ggcgccggga caatcgctgt cacggcggca 60

snbsnbsnbs nbsnbsnbsn bsnbsnbsnb gcacacacc 99

<210> 63
<211> 33
<212> PRT
<213> Artificial Sequence

<220>
<223> Sequence of Knottin varied in library

<220>
<221> misc_feature
<222> (4)..(13)
<223> Xaa can be any naturally occurring amino acid

<400> 63

Gly Val Cys Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Cys Arg Arg
1 5 10 15

Asp Ser Asp Cys Pro Gly Ala Cys Ile Cys Arg Gly Asn Gly Tyr Cys
20 25 30

Gly

<210> 64
<211> 121
<212> DNA
<213> Artificial Sequence

<220>
<223> pD7-Sce1 sequence nucleotides 1 - 120

<400> 64
ggtaccgaat tctagggata acagggtaat atgcatcttc tgacctcttc tcttcctccc 60

acagggcatg gcaaaacctc tgagccagga agaaagcaca ctgattgaaa gagcaaccgc 120

t 121

<210> 65
<211> 18
<212> PRT
<213> Artificial Sequence

<220>
<223> Fragment of blasticidin resistance gene encoded by pD7-Sce1

<400> 65

Met Ala Lys Pro Leu Ser Gln Glu Glu Ser Thr Leu Ile Glu Arg Ala
1 5 10 15

Thr Ala

<210> 66
<211> 120
<212> DNA
<213> Artificial Sequence

<220>
<223> pD7-ObLiGaRe sequence fragment

<400> 66
ggtaccgaat tcttttctgt caccaatcct ggggccacta gggacactgt ggggtggagg 60

aaatgcatct tctgacctct tctcttcctc ccacagggca tggcaaaacc tctgagccag 120

<210> 67
<211> 7
<212> PRT
<213> Artificial Sequence

<220>
<223> Fragment of Blasticidin resistance gene encoded by pD7-ObLiGaRe

<400> 67

Met Ala Lys Pro Leu Ser Gln
1 5

<210> 68
<211> 1061
<212> DNA
<213> Artificial Sequence

<220>
<223> ROSA 26 left homology arm sequence

<400> 68
gcgatcgcgc tgattggctt cttttcctcc cgccgtgtgt gaaaacacaa atggcgtgtt 60

ttggttggcg taaggcgcct gtcagttaac ggcagccgga gtgcgcagcc gccggcagcc 120

tcgctctgcc cactgggtgg ggcgggaggt aggtggggtg aggcgagctg gacgtgcggg 180

cgcggtcggc ctctggcggg gcgggggagg ggagggaggg tcagcgaaag tagctcgcgc 240

gcgagcggcc gcccaccctc cccttcctct gggggagtcg ttttacccgc cgccggccgg 300

gcctcgtcgt ctgattggct ctcggggccc agaaaactgg cccttgccat tggctcgtgt 360

tcgtgcaagt tgagtccatc cgccggccag cgggggcggc gaggaggcgc tcccaggttc 420

cggccctccc ctcggccccg cgccgcagag tctggccgcg cgcccctgcg caacgtggca 480

ggaagcgcgc gctgggggcg gggacgggca gtagggctga gcggctgcgg ggcgggtgca 540

agcacgtttc cgacttgagt tgcctcaaga ggggcgtgct gagccagacc tccatcgcgc 600

actccgggga gtggagggaa ggagcgaggg ctcagttggg ctgttttgga ggcaggaagc 660

acttgctctc ccaaagtcgc tctgagttgt tatcagtaag ggagctgcag tggagtaggc 720

ggggagaagg ccgcaccctt ctccggaggg gggaggggag tgttgcaata cctttctggg 780

agttctctgc tgcctcctgg cttctgagga ccgccctggg cctgggagaa tcccttcccc 840

ctcttccctc gtgatctgca actccagtct ttctagaatg cattaaggga tctgtagggc 900

gcagtagtcc agggtttcct tgatgatgtc atacttatcc tgtccctttt ttttccacag 960

ctcgcggttg aggacaaact cttcgcggtc tttccagtgg ggatcgacgg tatcgtagag 1020

tcgaggccgc tctaggaatt cacgccgcca ccatgaccga g 1061

<210> 69
<211> 703
<212> DNA
<213> Artificial Sequence

<220>
<223> ROSA 26 right homology arm sequence

<400> 69
gtatacggga attgaacagg tgtaaaattg gagggacaag acttcccaca gattttcggt 60

tttgtcggga agttttttaa taggggcaaa taaggaaaat gggaggatag gtagtcatct 120

ggggttttat gcagcaaaac tacaggttat tattgcttgt gatccgcctc ggagtatttt 180

ccatcgaggt agattaaaga catgctcacc cgagttttat actctcctgc ttgagatcct 240

tactacagta tgaaattaca gtgtcgcgag ttagactatg taagcagaat tttaatcatt 300

tttaaagagc ccagtacttc atatccattt ctcccgctcc ttctgcagcc ttatcaaaag 360

gtattttaga acactcattt tagccccatt ttcatttatt atactggctt atccaacccc 420

tagacagagc attggcattt tccctttcct gatcttagaa gtctgatgac tcatgaaacc 480

agacagatta gttacataca ccacaaatcg aggctgtagc tggggcctca acactgcagt 540

tcttttataa ctccttagta cacttttgt tgatcctttg ccttgatcct taattttcag 600

tgtctatcac ctctcccgtc aggtggtgtt ccacatttgg gcctattctc agtccaggga 660

gttttacaac aatagatgta ttgagaatcc aacctcctgc agg 703

<210> 70
<211> 18
<212> DNA
<213> Artificial Sequence

<220>
<223> Left TALEN recognition sequence

<400> 70
tcccctccac cccacagt 18

<210> 71
<211> 15
<212> DNA
<213> Artificial Sequence

<220>
<223> TALEN spacer

<400> 71
ggggccacta gggac 15

<210> 72
<211> 18
<212> DNA
<213> Artificial Sequence

<220>
<223> Right TALEN target sequence

<400> 72
aggattggtg acagaaaa 18

<210> 73
<211> 18
<212> DNA
<213> Artificial Sequence

<220>
<223> Right TALEN sequence

<400> 73

ttttctgtca ccaatcct 18

<210> 74
<211> 20
<212> DNA
<213> Artificial Sequence

<220>
<223> AAVS-Left-arm-junction-PCR-Forw (9625) primer

<400> 74
ccggaactct gccctctaac 20

<210> 75
<211> 23
<212> DNA
<213> Artificial Sequence

<220>
<223> BSD_Junction PCR-rev (9626) primer

<400> 75
tagccacaga atagtcttcg gag 23

<210> 76
<211> 21
<212> DNA
<213> Artificial Sequence

<220>
<223> Donor_plasmid_seq_PDGFRTM-2 Forw primer

<400> 76
acacgcagga ggccatcgtg g 21

<210> 77
<211> 20
<212> DNA
<213> Artificial Sequence

<220>
<223> AAVS1_right arm_junction_PCR_rev primer

<400> 77
tcctgggata ccccgaagag 20

<210> 78
<211> 54
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2598

<400> 78
tttttttaa ttaagattat tgactagtta ttaatagtaa tcaattacgg ggtc 54

<210> 79
<211> 52
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2619

<400> 79
tttttgttt aaacttacct tggatccctt gccggggctc aggctcaggg ac 52

<210> 80
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> M13Leadseq primer

<400> 80
aaattattat tcgcaattcc tttggttgtt cct 33

<210> 81
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Notmycseq primer

<400> 81
ggccccattc agatcctctt ctgagatgag 30

<210> 82
<211> 22
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2623

<400> 82
taaagtaggc ggtcttgaga cg 22

<210> 83
<211> 22
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2624

<400> 83
gaaggtgctg ttgaactgtt cc 22

<210> 84
<211> 19
<212> DNA

<213> Artificial Sequence

<220>
<223> Primer 2595

<400> 84
gagggctctg gcagctagc 19

<210> 85
<211> 20
<212> DNA
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<220>
<223> Primer 2597

<400> 85
tcgagactgt gacgaggctg 20

<210> 86
<211> 19
<212> DNA
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<220>
<223> Primer 2625

<400> 86
ccttggtgct ggcactcga 19

<210> 87
<211> 40
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 1999

<400> 87
aaaaagcagg ctaccatgag ggcctggatc ttctttctcc 40

<210> 88
<211> 44
<212> DNA
<213> Artificial Sequence

<220>
<223> TALEN recognition sequence

<400> 88
tccaccggtc gccaccatgg tgagcaaggg cgaggagctg ttca 44

<210> 89
<211> 33
<212> DNA
<213> Artificial Sequence

<220>

<223> AAVS1_HA-L_Nested_Forw1 primer

<400> 89
gtgcccttgc tgtgccgccg gaactctgcc ctc 33

<210> 90
<211> 23
<212> DNA
<213> Artificial Sequence

<220>
<223> EGFP_Synthetic_gene_Rev_Assembly primer

<400> 90
ttcacgtcgc cgtccagctc gac 23

<210> 91
<211> 21
<212> DNA
<213> Artificial Sequence

<220>
<223> Purotk_seq_fow2 primer

<400> 91
tccataccga cgatctgcga c 21

<210> 92
<211> 27
<212> DNA
<213> Artificial Sequence

<220>
<223> J44 primer

<400> 92
aaaagcgcct cccctacccg gtagaat 27

<210> 93
<211> 27
<212> DNA
<213> Artificial Sequence

<220>
<223> J46 primer

<400> 93
ggcgacacgg aaatgttgaa tactcat 27

<210> 94
<211> 27
<212> DNA
<213> Artificial Sequence

<220>
<223> J48 primer

<400> 94

cactacaccc agaagtccct gagcctg 27

<210> 95
<211> 20
<212> DNA
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<220>
<223> pSANG10pelB primer

<400> 95
cgctgcccag ccggccatgg 20

<210> 96
<211> 26
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2097

<400> 96
gatggtgatg atgatgtgcg gatgcg 26

<210> 97
<211> 38
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2423

<400> 97
ttttttccat gggccggccc tccttcagtt tagttgag 38

<210> 98
<211> 39
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2437

<400> 98
ttttttgcgg ccgcggaagc cgtgatctcc ttctctctc 39

<210> 99
<211> 20
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<220>
<223> Primer 2622

<400> 99
gaacaggaac acggaaggtc 20

<210> 100
<211> 22
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 41679

<400> 100
atgagttgga gctgtatcat cc 22

<210> 101
<211> 18
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2621

<400> 101
gcattccacg gcggccgc 18

<210> 102
<211> 9
<212> PRT
<213> Artificial Sequence

<220>
<223> Peptide SLLMWITQV (NY-ESO-1 157-165) recognised by c12c2

<400> 102

Ser Leu Leu Met Trp Ile Thr Gln Val
1 5

<210> 103
<211> 10
<212> PRT
<213> Artificial Sequence

<220>
<223> Peptide MART-1 26-35 recognised by MEL5 TCR

<400> 103

Glu Leu Ala Gly Ile Gly Ile Leu Thr Val
1 5 10

<210> 104
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 4JFH-Valpha-F

<400> 104
acacacgcta gccagaaaga ggtggaacag 30

<210> 105
<211> 50
<212> DNA
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<220>
<223> Primer 2770

<220>
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<223> n is a, c, g, or t

<220>
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<222> (30)..(31)
<223> n is a, c, g, or t

<400> 105
caaagaacag ctcgccggts nncccgassn nggagctggc gcaaaagtac 50

<210> 106
<211> 53
<212> DNA
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<220>
<223> Primer 2771

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<223> n is a, c, g, or t

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<222> (30)..(31)
<223> n is a, c, g, or t

<220>
<221> misc_feature
<222> (33)..(34)
<223> n is a, c, g, or t

<400> 106
ctcgcccgaa ggtgggaatg tangwtccsn nsnnaagtgg gcgcacggcg cac 53

<210> 107
<211> 22
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2780

<400> 107
ctggcagcta gcaagcagga ag 22

<210> 108
<211> 22
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2781

<400> 108
tacattccca ccttcgggcg ag 22

<210> 109
<211> 26
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2782

<400> 109
ttttttgcgg ccgcggacag gttctg 26

<210> 110
<211> 27
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2783

<400> 110
cgtaagctgg taccttatta tctaggg 27

<210> 111
<211> 27
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2785

<400> 111
ccctagataa taaggtacca gcttacg 27

<210> 112
<211> 19
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2787

<400> 112
accggcgagc tgttctttg 19

<210> 113
<211> 26

<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2788

<400> 113
agtgacaagc ttttattatc tgggtg 26

<210> 114
<211> 19
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2789

<400> 114
caggtcctcg agcactgtc 19

<210> 115
<211> 25
<212> DNA
<213> Artificial Sequence

<220>
<223> CRISPR1 double-stranded DNA insert strand

<400> 115
ggggccacta gggacaggat gtttt 25

<210> 116
<211> 26
<212> DNA
<213> Artificial Sequence

<220>
<223> CRISPR1 double-stranded DNA insert strand

<400> 116
catcctgtcc ctagtggccc ccggtg 26

<210> 117
<211> 25
<212> DNA
<213> Artificial Sequence

<220>
<223> CRISPR2 double-stranded DNA insert strand

<400> 117
gtcaccaatc ctgtccctag gtttt 25

<210> 118
<211> 25
<212> DNA
<213> Artificial Sequence

<220>
<223> CRISPR2 double-stranded DNA insert strand

<400> 118
ctagggacag gattggtgac cggtg 25

<210> 119
<211> 102
<212> RNA
<213> Artificial Sequence

<220>
<223> CRISPR 1 RNA

<400> 119
gggggggccac uagggacagg auguuuuaga gcuagaaaua gcaaguuaaa auaaggcuag 60

uccguuauca acuugaaaaa guggcaccga gucggugcuu uu 102

<210> 120
<211> 102
<212> RNA
<213> Artificial Sequence

<220>
<223> CRISPR 2 RNA

<400> 120
gggucaccaa uccugucccu agguuuuaga gcuagaaaua gcaaguuaaa auaaggcuag 60

uccguuauca acuugaaaaa guggcaccga gucggugcuu uu 102

<210> 121
<211> 25
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2723

<400> 121
cgcgccagaa gtctcaccaa gccca 25

<210> 122
<211> 25
<212> DNA
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<220>
<223> Primer 2724

<400> 122
cgcgtgggct tggtgagact tctgg 25

<210> 123
<211> 60
<212> DNA
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<220>
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<400> 123
aattctcccc tccaccccac agtagggaca gtggggccag gattggtgac agaaaatgca 60

<210> 124
<211> 52
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2769

<400> 124
ttttctgtca ccaatcctgg ccccactgtc cctactgtgg ggtggagggg ag 52

<210> 125
<211> 28
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2778

<400> 125
aattctaggg ataacagggt aatatgca 28

<210> 126
<211> 20
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2779

<400> 126
tattaccctg ttatccctag 20

<210> 127
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<400> 127
aattcttttc tgtcaccaat cctggggcca ctagggacac tgtggggtgg aggggatgca 60

<210> 128
<211> 57
<212> DNA
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<220>
<223> Primer 2809

<400> 128
tcccctccac cccacagtgt ccctagtggc cccaggattg gtgacagaaa agaattg 57

<210> 129
<211> 41
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer J60

<400> 129
acacacggta ccgcgatcgc gctgattggc ttcttttcct c 41

<210> 130
<211> 35
<212> DNA
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<220>
<223> Primer 2706

<400> 130
ttttttatgc attctagaaa gactggagtt gcaga 35

<210> 131
<211> 18
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2715

<400> 131
gagcgtccgc ccaccctc 18

<210> 132
<211> 18
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2716

<400> 132
gagggtgggc ggacgctc 18

<210> 133
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2709

<400> 133
ttttttatgc attaagggat ctgtagggcg cag 33

<210> 134
<211> 21
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2710

<400> 134
gtgaattcct agagcggcct c 21

<210> 135
<211> 50
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer 2745

<400> 135
gaggccgctc taggaattca cgccgccacc atgaccgagt acaagcccac 50

<210> 136
<211> 48
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer J59

<400> 136
aaaaaaagat ctgtgtgttt cgaatcaggc accgggcttg cgggtcat 48

<210> 137
<211> 36
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer J61

<400> 137
ttttttgtat acgggaattg aacaggtgta aaattg 36

<210> 138
<211> 43
<212> DNA
<213> Artificial Sequence

<220>
<223> Primer J62

<400> 138
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