

62132NAT - SEQ Listing.TXT
SEQUENCE LISTING

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Campana, Dario

<120> BLOCKADE OF CD7 EXPRESSION AND CHIMERIC ANTIGEN RECEPTORS FOR
IMMUNOTHERAPY OF T-CELL MALIGNANCIES

<130> 119419-5002-WO

<140> Filed Herewith
<141> 2017-11-22

<150> US 62/425,398
<151> 2016-11-22

<150> US 62/543,696
<151> 2017-08-10

<160> 40

<170> PatentIn version 3.5

<210> 1
<211> 117
<212> PRT
<213> Artificial Sequence

<220>
<223> VH domain of TH69 antibody (anti-CD7 scFv)

<400> 1

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

Ser Leu Lys Leu Ser Cys Ala Ala Ser Gly Leu Thr Phe Ser Ser Tyr
 20 25 30

Ala Met Ser Trp Val Arg Gln Thr Pro Glu Lys Arg Leu Glu Trp Val
 35 40 45

Ala Ser Ile Ser Ser Gly Gly Phe Thr Tyr Tyr Pro Asp Ser Val Lys
 50 55 60

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

Gln Met Ser Ser Leu Arg Ser Glu Asp Thr Ala Met Tyr Tyr Cys Ala
85 90 95

Arg Asp Glu Val Arg Gly Tyr Leu Asp Val Trp Gly Ala Gly Thr Thr
100 105 110

Val Thr Val Ser Ser
115

<210> 2
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<212> PRT
<213> Artificial Sequence

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<223> VL domain of TH69 antibody (anti-CD7 scFv)

<400> 2

Ala Ala Tyr Lys Asp Ile Gln Met Thr Gln Thr Thr Ser Ser Leu Ser
1 5 10 15

Ala Ser Leu Gly Asp Arg Val Thr Ile Ser Cys Ser Ala Ser Gln Gly
20 25 30

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

Lys Leu Leu Ile Tyr Tyr Thr Ser Ser Leu His Ser Gly Val Pro Ser
50 55 60

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

Asn Leu Glu Pro Glu Asp Ile Ala Thr Tyr Tyr Cys Gln Gln Tyr Ser
85 90 95

Lys Leu Pro Tyr Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Lys Arg
100 105 110

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<210> 3
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 <212> PRT
 <213> Artificial Sequence

<220>
 <223> intracellular signaling domain of 4-1BB

<400> 3

Lys Arg Gly Arg Lys Lys Leu Leu Tyr Ile Phe Lys Gln Pro Phe Met
 1 5 10 15

Arg Pro Val Gln Thr Thr Gln Glu Glu Asp Gly Cys Ser Cys Arg Phe
 20 25 30

Pro Glu Glu Glu Glu Gly Gly Cys Glu Leu
 35 40

<210> 4
 <211> 112
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> intracellular signaling domain of CD3zeta

<400> 4

Arg Val Lys Phe Ser Arg Ser Ala Asp Ala Pro Ala Tyr Gln Gln Gly
 1 5 10 15

Gln Asn Gln Leu Tyr Asn Glu Leu Asn Leu Gly Arg Arg Glu Glu Tyr
 20 25 30

Asp Val Leu Asp Lys Arg Arg Gly Arg Asp Pro Glu Met Gly Gly Lys
 35 40 45

Pro Arg Arg Lys Asn Pro Gln Glu Gly Leu Tyr Asn Glu Leu Gln Lys
 50 55 60

Asp Lys Met Ala Glu Ala Tyr Ser Glu Ile Gly Met Lys Gly Glu Arg
 65 70 75 80

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Arg Arg Gly Lys Gly His Asp Gly Leu Tyr Gln Gly Leu Ser Thr Ala
85 90 95

Thr Lys Asp Thr Tyr Asp Ala Leu His Met Gln Ala Leu Pro Pro Arg
100 105 110

<210> 5
<211> 45
<212> PRT
<213> Artificial Sequence

<220>
<223> Hinge region of anti-CD7 CAR

<400> 5

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
35 40 45

<210> 6
<211> 24
<212> PRT
<213> Artificial Sequence

<220>
<223> transmembrane region of anti-CD7 CAR

<400> 6

Ile Tyr Ile Trp Ala Pro Leu Ala Gly Thr Cys Gly Val Leu Leu Leu
1 5 10 15

Ser Leu Val Ile Thr Leu Tyr Cys
20

<210> 7
<211> 21

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<212> PRT

<213> Artificial Sequence

<220>

<223> CD8a signal peptide of anti-CD7 CAR

<400> 7

Met	Ala	Leu	Pro	Val	Thr	Ala	Leu	Leu	Leu	Pro	Leu	Ala	Leu	Leu	Leu
1				5				10					15		

His	Ala	Ala	Arg	Pro
			20	

<210> 8

<211> 14

<212> PRT

<213> Artificial Sequence

<220>

<223> Localization domain

<400> 8

Glu	Gln	Lys	Leu	Ile	Ser	Glu	Glu	Asp	Leu	Lys	Asp	Glu	Leu
1				5				10					

<210> 9

<211> 11

<212> PRT

<213> Artificial Sequence

<220>

<223> Localizing domain

<400> 9

Gly	Gly	Gly	Gly	Ser	Ala	Glu	Lys	Asp	Glu	Leu
1				5				10		

<210> 10

<211> 68

<212> PRT

<213> Artificial Sequence

<220>

<223> Localizing domain

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<400> 10

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
65

<210> 11

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> Localizing domain

<400> 11

Lys Tyr Lys Ser Arg Arg Ser Phe Ile Asp Glu Lys Lys Met Pro
1 5 10 15

<210> 12

<211> 20

<212> PRT

<213> Artificial Sequence

<220>

<223> Linker

<400> 12

Gly Gly Gly Gly Ser Gly Gly Gly Gly Ser Gly Gly Gly Gly Ser Gly
1 5 10 15

Gly Gly Gly Ser
20

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<210> 13
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 <212> PRT
 <213> Artificial Sequence

<220>
 <223> Localizing domain

<400> 13

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 Lys Tyr Lys Ser Arg Arg Ser Phe Ile Asp Glu Lys
 65 70 75 80

Lys Met Pro

<210> 14
 <211> 123
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> VH domain of 3alf antibody (anti-CD7 scFv)

<400> 14

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

Ser Val Lys Leu Ser Cys Lys Ala Ser Gly Tyr Thr Phe Thr Ser Tyr
 20 25 30

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Trp Met His Trp Val Lys Gln Arg Pro Gly Gln Gly Leu Glu Trp Ile
35 40 45

Gly Lys Ile Asn Pro Ser Asn Gly Arg Thr Asn Tyr Asn Glu Lys Phe
50 55 60

Lys Ser Lys Ala Thr Leu Thr Val Asp Lys Ser Ser Ser Thr Ala Tyr
65 70 75 80

Met Gln Leu Ser Ser Leu Thr Ser Glu Asp Ser Ala Val Tyr Tyr Cys
85 90 95

Ala Arg Gly Gly Val Tyr Tyr Asp Leu Tyr Tyr Tyr Ala Leu Asp Tyr
100 105 110

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

<210> 15
<211> 108
<212> PRT
<213> Artificial Sequence

<220>
<223> VL domain of 3alf antibody (anti-CD7 scFv)

<400> 15

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

Asp Ser Val Ser Leu Ser Cys Arg Ala Ser Gln Ser Ile Ser Asn Asn
20 25 30

Leu His Trp Tyr Gln Gln Lys Ser His Glu Ser Pro Arg Leu Leu Ile
35 40 45

Lys Ser Ala Ser Gln Ser Ile Ser Gly Ile Pro Ser Arg Phe Ser Gly
50 55 60

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Ser Gly Ser Gly Thr Asp Phe Thr Leu Ser Ile Asn Ser Val Glu Thr
65 70 75 80

Glu Asp Phe Gly Met Tyr Phe Cys Gln Gln Ser Asn Ser Trp Pro Tyr
85 90 95

Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Lys Arg
100 105

<210> 16

<211> 120

<212> PRT

<213> Artificial Sequence

<220>

<223> VH domain of T3-3A1 antibody (anti-CD7 scFv)

<400> 16

Asp Val Gln Leu Val Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly
1 5 10 15

Ser Arg Lys Leu Ser Cys Ala Ala Ser Gly Phe Thr Phe Ser Ser Phe
20 25 30

Gly Met His Trp Val Arg Gln Ala Pro Glu Lys Gly Leu Glu Trp Val
35 40 45

Ala Tyr Ile Ser Ser Gly Ser Ser Thr Leu His Tyr Ala Asp Thr Val
50 55 60

Lys Gly Arg Phe Thr Ile Ser Arg Asp Asn Pro Lys Asn Thr Leu Phe
65 70 75 80

Leu Gln Met Thr Ser Leu Arg Ser Glu Asp Thr Ala Met Tyr Tyr Cys
85 90 95

Ala Arg Trp Gly Asn Tyr Pro His Tyr Ala Met Asp Tyr Trp Gly Gln
100 105 110

Gly Thr Ser Val Thr Val Ser Ser
115 120

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<210> 17
 <211> 111
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> VL domain of T3-3A1 antibody (anti-CD7 scFv)

<400> 17

Asp Ile Val Met Thr Gln Ser Pro Ala Ser Leu Ala Val Ser Leu Gly
 1 5 10 15

Gln Arg Ala Thr Ile Ser Cys Arg Ala Ser Lys Ser Val Ser Ala Ser
 20 25 30

Gly Tyr Ser Tyr Met His Trp Tyr Gln Gln Lys Pro Gly Gln Pro Pro
 35 40 45

Lys Leu Leu Ile Tyr Leu Ala Ser Asn Leu Glu Ser Gly Val Pro Ala
 50 55 60

Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Asn Ile His
 65 70 75 80

Pro Val Glu Glu Glu Asp Ala Val Thr Tyr Tyr Cys Gln His Ser Arg
 85 90 95

Glu Leu Pro Tyr Thr Phe Gly Gly Gly Thr Lys Leu Glu Ile Lys
 100 105 110

<210> 18
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<220>
 <223> Localizing sequence

<400> 18

Lys Asp Glu Leu
 1

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<210> 19
<211> 4
<212> PRT
<213> Artificial Sequence

<220>
<223> Localizing sequence

<220>
<221> misc_feature
<222> (3)..(4)
<223> Xaa can be any naturally occurring amino acid

<400> 19

Lys Lys Xaa Xaa
1

<210> 20
<211> 3
<212> PRT
<213> Artificial Sequence

<220>
<223> Localizing sequence

<220>
<221> misc_feature
<222> (2)..(2)
<223> Xaa can be any naturally occurring amino acid

<220>
<221> MISC_FEATURE
<222> (3)..(3)
<223> Xaa can be Asp or Glu

<400> 20

Lys Xaa Xaa
1

<210> 21
<211> 4
<212> PRT
<213> Artificial Sequence

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<220>

<223> Localizing sequence

<400> 21

Tyr Gln Arg Leu

1

<210> 22

<211> 4

<212> PRT

<213> Artificial Sequence

<220>

<223> Proteasome localizing sequence

<400> 22

Pro Glu Ser Thr

1

<210> 23

<211> 351

<212> DNA

<213> Artificial Sequence

<220>

<223> VH domain of TH69 antibody (anti-CD7 scFv)

<400> 23

gaggtgcagc tggtcgaatc tggaggagga ctggtgaagc caggaggatc tctgaaactg 60

agttgtgccg cttcaggcct gaccttctca agctacgcca tgagctgggt gcgacagaca 120

cctgagaagc ggctggaatg ggtcgctagc atctcctctg gcgggttcac atactatcca 180

gactccgtga aaggcagatt tactatctct cgggataacg caagaaatat tctgtacctg 240

cagatgagtt cactgaggag cgaggacacc gcaatgtact attgtgccag ggacgaagtg 300

cgcggtctatc tggatgtctg gggagctggc actaccgtca ccgtctccag c 351

<210> 24

<211> 351

<212> DNA

<213> Artificial Sequence

<220>

<223> VL domain of TH69 antibody (anti-CD7 scFv)

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<400> 24
gaggtgcagc tggtcgaatc tggaggagga ctggtgaagc caggaggatc tctgaaactg      60
agttgtgccg cttcaggcct gaccttctca agctacgcca tgagctgggt gcgacagaca      120
cctgagaagc ggctggaatg ggtcgttagc atctcctctg gcgggttcac atactatcca      180
gactccgtga aaggcagatt tactatctct cgggataacg caagaaatat tctgtacctg      240
cagatgagtt cactgaggag cgaggacacc gcaatgtact attgtgccag ggacgaagtg      300
cgcggtctatc tggatgtctg gggagctggc actaccgtca cctctccag c              351
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<210> 25
<211> 369
<212> DNA
<213> Artificial Sequence
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<220>
<223> VH domain of 3alf antibody (anti-CD7 scFv)
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<400> 25
caggtccagc tgcaggagtc aggagctgag ctggtgaagc caggggcaag cgtcaaactg      60
tcctgcaagg cctctggata tacattcact agctactgga tgcactgggt gaaacagaga      120
cccggacagg gcctggagtg gatcggaaag attaacccta gcaatggcag gaccaactac      180
aacgaaaagt ttaaattcaa ggcaaccctg acagtggaca agagctcctc tacagcctac      240
atgcagctga gttcactgac ttcagaggat agcgcagtgt actattgcgc cagaggcggg      300
gtctactatg acctgtacta ttacgccctg gattattggg ggcagggaac cacagtgact      360
gtcagctcc                                369
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<210> 26
<211> 324
<212> DNA
<213> Artificial Sequence
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<220>
<223> VL domain of 3alf antibody (anti-CD7 scFv)
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<400> 26
gacatcgagc tgaccagag tcctgctaca ctgagcgtga ctccaggcga ttctgtcagt      60
ctgtcatgtc gggcaagcca gtccatctct aacaatctgc actggtacca gcagaaatcc      120
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catgaatctc cactgactgct gattaagagt gcctcacaga gcatctccgg cattccctcc	180
cggttctctg gcagtgggtc aggaactgac tttaccctga gtattaaactc agtggagaca	240
gaagatttcg gcatgtatatt ttgccagcag agcaattcct ggccctacac tttcggaggc	300
gggaccaaac tggagatcaa gcgg	324

<210> 27
 <211> 360
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> VH domain of T3-4A1 antibody (anti-CD7 scFv)

<400> 27	
gatgtgcagc tggtggagtc tgggggaggc ttagtgcagc ctggagggtc ccggaaactc	60
tcctgtgcag cctctggatt cactttcagt agctttggaa tgcactgggt tcgtcaggct	120
ccagagaagg ggctggagtg ggtcgcatac attagtagtg gcagtagtac cctccactat	180
gcagacacag tgaagggccg attcaccatc tccagagaca atcccaagaa caccctgttc	240
ctgcaaataa ccagtctaag gtctgaggac acggccatgt attactgtgc aagatgggggt	300
aactaccctc actatgctat ggactactgg ggtcaaggaa cctcagtcac cgtctcctca	360

<210> 28
 <211> 333
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> VL domain of T3-4A1 antibody (anti-CD7 scFv)

<400> 28	
gacattgtga tgaccagtc tcctgcttcc ttagctgtat ctctggggca gagggccacc	60
atctcatgca gggccagcaa aagtgtcagt gcatctggct atagttatat gcactgggtac	120
caacagaaac caggacagcc acccaaactc ctcatctatc ttgcatcaa cctagaatct	180
gggggtccctg ccaggttcag tggcagtggg tctgggacag acttcaccct caacatccat	240
cctgtggagg aggaggatgc tgtaacctat tactgtcagc acagtaggga gcttccgtac	300
acgttcggag gggggaccaa gctggaata aaa	333

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<210> 29
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<220>
 <223> Polypeptide linker

<400> 29

Gly Gly Gly Gly Ser
 1 5

<210> 30
 <211> 63
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> CD8alpha signal peptide

<400> 30
 atggctctgc ctgtgaccgc actgctgctg cccctggctc tgctgctgca cgccgcaaga 60
 cct 63

<210> 31
 <211> 60
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> VH-VL Linker

<400> 31
 ggaggaggag gaagcggagg aggaggatcc ggaggcgggg gatctggagg aggaggaagt 60

<210> 32
 <211> 42
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> ER localization domain, KDEL tethered to scFv with myc ("myc KDEL")

<400> 32
 gagcagaaac tgattagcga agaggacctg aaagatgaac tg 42

62132NAT - SEQ Listing.TXT

<210> 33
 <211> 78
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> localization domain, "link.(20)AEKDEL"

<400> 33
 ggtgggtggcg gcagtggtgg cggtggctca ggcggtgggtg gctccggtgg cggtggctct 60
 gcagaaaaag atgagttg 78

<210> 34
 <211> 249
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> localization domain, "mb DEKKMP"

<400> 34
 accactacac ctgcaccaag gcctcccaca cccgctccca ctatcgcttc ccagccactg 60
 tccctgaggc ccgaggcctg caggccagca gctggcggag ccgtgcatac tagggggctg 120
 gacttcgctt gcgacatcta catctgggcc ccactggcag ggacatgcgg agtcctgctg 180
 ctgtccctgg tcatcacact ttacaaatac aaaagcagac gcagttttat tgatgaaaag 240
 aaaatgcct 249

<210> 35
 <211> 126
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Nucleic acid sequence of intracellular signaling domain of 4-1BB

<400> 35
 aagcgggggc gcaaaaaact gctgtatatc tttaagcagc ctttcatgag accagtgcag 60
 acaaccagg aggaagatgg gtgctcatgc cggtttcccg aggaggagga aggcggctgc 120
 gagctg 126

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

<220>
 <223> intracellular signaling domain of CD3zeta

<400> 36
 aggggtgaagt tttcccgctc agcagatgct cctgcctacc agcagggcca gaaccagctg 60
 tataatgagc tgaacctggg cagacgcgaa gagtatgatg tgctggacaa aaggcgggga 120
 agagaccccg aaatgggagg gaagccaagg cggaaaaacc cccaggaggg cctgtacaat 180
 gagctgcaga aggacaaaat ggcagaggct tacagtgaga ttgggatgaa gggagagaga 240
 cggaggggga aagggcacga tggcctgtac caggggctga gcacagcaac caaagatact 300
 tatgacgcac tgcacatgca ggcactgcca cccaga 336

<210> 37
 <211> 204
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Hinge and transmembrane domain of CD8alpha

<400> 37
 accactacac ctgcaccaag gcctcccaca cccgctccca ctatcgcttc ccagccactg 60
 tccctgaggc ccgaggcctg caggccagca gctggcggag ccgtgcatac tagggggctg 120
 gacttcgctt gcgacatcta catctgggcc cactggcag ggacatgcgg agtcctgctg 180
 ctgtccctgg tcatcacact gtac 204

<210> 38
 <211> 15
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> synthetic oligonucleotide primer

<400> 38
 atggccgggc ctccg 15

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<210> 39
 <211> 18
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> synthetic oligonucleotide primer

<400> 39
 tcactggtac tggttggg 18

<210> 40
 <211> 10
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> Localizing domain

<400> 40

Glu Gln Lys Leu Ile Ser Glu Glu Asp Leu
 1 5 10