

SEQUENCE LISTING

<110> Tokushima University

<120> Target sequence-specific alteration technique using nucleotide target recognition

<130> 674277

<150> JP2017-158876

<151> 2017-08-21

<150> JP2017-236518

<151> 2017-12-08

<160> 45

<170> PatentIn version 3.5

<210> 1

<211> 224

<212> PRT

<213> Microcystis aeruginosa

<400> 1

Met Val His Ile Tyr Ser Cys Gln Leu Glu Leu His Asp Ser Leu Tyr
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Tyr Ala Thr Arg Glu Ile Gly Arg Leu Tyr Glu Ser Glu Pro Val Ile
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His Asn Tyr Ala Leu Cys Tyr Ala Leu Gly Leu Val Asn Ser Asp Ser
35 40 45

Tyr Arg Tyr Phe Cys Ser Glu Gln Ile Pro Gln Tyr Gln Glu His Leu
50 55 60

Asn Pro Leu Asn Glu Glu Lys Ile Tyr Val Thr Pro Ala Arg Ala Ile
65 70 75 80

Ala His Thr Ala Val Leu Asn Thr Trp Lys Tyr Ala Asn Asn Asn Tyr
85 90 95

His Val Glu Met Glu Lys Thr Gln Lys Asn Ile Pro Ser Phe Gly Arg
100 105 110

Ala Lys Glu Ile Ala Pro Glu Ser Ile Phe Glu Cys Phe Ile Ile Ser
115 120 125

His His Pro Leu Gln Leu Pro Lys Trp Ile Arg Leu Gly Lys Trp Met
130 135 140

Ser Lys Ala Glu Val Lys Leu Thr Glu Leu Ser Leu Ser Lys Gln Lys
145 150 155 160



Glu Asp Leu Phe Ile Tyr Pro Tyr Pro Leu Asn Pro Leu Asp Val Met
 165 170 175

Phe Thr His Gln Val Ile Gly Tyr Asp Val Ile Asn Met Pro Pro Val
 180 185 190

Ser Leu Ile Arg Asn Val Arg Met Arg Gly Glu Tyr Tyr Gln Ile Ser
 195 200 205

Asp Arg Pro Asp Leu Lys Ile Pro Ala Arg Leu Ser Tyr His Phe Gly
 210 215 220

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Val Ser Ser Val Asp Gln Glu Leu Gly Asn Ile Leu His Thr Ala Glu
 35 40 45

Ala Asp Lys Ala Phe Thr Leu Ser Pro Leu Gln Met Gln Ser Gly Gly
 50 55 60

Lys Thr Ile Asn Ser Pro Gln Trp Arg His Glu Arg Glu Ile Ala Ser
 65 70 75 80

Glu Thr Pro Cys Trp Trp Arg Ile Ser Leu Leu Asp Asp Arg Leu Phe
 85 90 95

Gly Lys Leu Thr Ser Leu Trp Leu Asn Leu Asn Pro Lys Gln Pro Trp
 100 105 110

His Leu Gly Ser Ala Asp Leu Val Ile Thr Ser Val Leu Ala Thr Pro
 115 120 125

Gln Ser Val Gln Pro Trp Ala Asn Ser Cys Thr Tyr Gln Tyr Leu Tyr
 130 135 140

Glu Asn Ala Ser Glu Thr Asn Arg Glu Phe Asp Phe Leu Phe Ala Thr
 145 150 155 160



Pro Val Thr Phe Arg Gln Gly Lys Phe Asp Ser Ala Leu Pro Thr Arg
165 170 175

Glu Leu Val Phe Asn Ser Leu Leu Gly Arg Trp Asn Arg Tyr Ser Gly
180 185 190

Ile Pro Phe Asp Ser Ile Ala Leu Glu Ser Ile Phe Pro Ser Phe Phe
195 200 205

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

Gly Cys Val Gly Glu Ile His Tyr Arg Leu Leu Gly Glu Val Glu Pro
225 230 235 240

Ala Lys Ile Lys Ala Ile Asn Ala Leu Ala Asp Phe Ala Leu Tyr Ala
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Ser Lys Asp Lys Arg
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<213> Microcystis aeruginosa

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35 40 45

Val Arg Ala Gly Ile Asp Ser Lys Lys Thr Ile Ser Arg Leu Ser Met
50 55 60

Phe Lys Arg Lys Gln Ser Thr Pro Glu Arg Leu Val Gly Arg Glu Leu
65 70 75 80

Leu Arg Asn Tyr Gly Leu Ile Thr Ala Glu Glu Cys Glu Tyr Asn Val
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Lys Phe Ala Met Asn Asn Ala Asp Cys Ile Ile Tyr Gly Phe Ala Ile
100 105 110



Gly Asp Ser Gly Ser Glu Lys Ser Lys Val Val Val Asp Thr Ala Phe
 115 120 125
 Ser Ile Thr Pro Phe Asp Glu Ser His Glu Ser Phe Thr Leu Asn Ala
 130 135 140
 Pro Tyr Glu Asn Gly Thr Met Ala Ser Lys Gly Glu Asn Asn Thr Lys
 145 150 155 160
 Val Gly Glu Val Thr Ser Arg Ile Asn Gln Gln Asp His Ile Arg Pro
 165 170 175
 Gln Val Phe Phe Pro Ser Ile Val Thr Leu Lys Asp Pro Thr Glu Ala
 180 185 190
 Ser Phe Leu Tyr Val Phe Asn Asn Ile Leu Arg Thr Arg His Tyr Gly
 195 200 205
 Ala Gln Thr Thr Arg Thr Gly Arg Val Arg Asn Glu Leu Ile Gly Val
 210 215 220
 Ile Phe Ala Asp Gly Glu Ile Val Ser Asn Leu Arg Trp Thr Gln Ala
 225 230 235 240
 Ile Tyr Asp Arg Leu Pro Asp Glu Val Leu His Ser Ile Asp Pro Leu
 245 250 255
 Asp Glu Asp Leu Val Met Glu Lys Ala Thr Glu Ala Ile Gln Ala Leu
 260 265 270
 Met Ala Glu Glu Phe Ile Val His Thr Asp Phe Ile Gly Glu Asn Phe
 275 280 285
 Gln Pro Leu Leu Thr Glu Val Lys Thr Leu Thr Gly Thr Glu Ala Gly
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 Glu Gln Tyr Ile Glu Lys Lys Lys Ala Glu Lys Lys
 325 330

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 <213> Microcystis aeruginosa

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35 40 45

Val Ile Phe Asn Thr Ala Met Thr Gly Asp Gly Lys Ser Leu Ala Ala
50 55 60

Tyr Leu Arg Thr Leu Gln Gly Tyr Phe Pro Ile Met Gly Leu Tyr Pro
65 70 75 80

Thr Asn Glu Leu Ala Arg Asp Gln Arg Gly Gln Ile Glu Ala Tyr Ile
85 90 95

Gln Arg Phe Gln Pro Thr Asp Gln Pro Arg Val Asn Leu Leu Thr Gly
100 105 110

Pro Glu Leu Glu Leu Tyr Ala Glu Arg Asp Gly Lys Thr Lys Ala Ile
115 120 125

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

Asp Ile Phe His Tyr Leu His Arg Ala Ala Tyr Leu Thr Pro Tyr Asp
145 150 155 160

Asn Pro Asp Gln Leu Trp Asn Arg Ile Asp Lys His Phe Asp Leu Phe
165 170 175

Leu Phe Asp Glu Phe His Val Phe Gly Thr Pro Gln Val Ala Ser Ile
180 185 190

Ile Asn Thr Met Leu Leu Ile Arg Arg Ala Asn Arg Gly Lys Arg Tyr
195 200 205

Leu Phe Leu Ser Ala Thr Pro Asp Glu Gly Leu Leu Lys Arg Leu Asp
210 215 220

Lys Ala Gly Phe Arg Tyr Arg Ser Ile Asp Pro Val Arg Glu Gly Lys
225 230 235 240

Tyr Arg Phe Pro Asp Thr Pro Glu Glu Ala Asn Ser Leu Ala Gln Gln
245 250 255

Gly Trp Arg Gln Val Thr Ser Glu Ile Glu Leu Ser Phe Ile Pro Leu



260

265

270

Pro Ser Ser Phe Gln Thr Ser Glu Asn Trp Leu Lys Glu Asn Lys Glu
275 280 285

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

Ile Leu Asn Ser Ile Ala Ser Val Lys Arg Leu Leu Pro Ile Phe Arg
305 310 315 320

Glu Leu Leu Ala Thr Ile Gly Leu Thr Val Gly Glu Asn Thr Gly Leu
325 330 335

Ser Gly Thr Arg Glu Lys Leu Ala Ser Leu Asn Arg Asp Leu Val Ile
340 345 350

Gly Thr Ser Thr Ile Asp Val Gly Val Asp Phe Lys Ile Asn Phe Leu
355 360 365

Ile Phe Glu Ser Ser Asp Ala Gly Asn Phe Ile Gln Arg Phe Gly Arg
370 375 380

Leu Gly Arg His Ser Gly Tyr Asp Arg Lys Gly Thr Ala Val Lys Phe
385 390 395 400

Thr Asn Phe Thr Ala Ile Ala Leu Val Pro Lys Phe Phe Leu Glu Arg
405 410 415

Leu Phe Glu Lys Lys Asp Ala Pro Leu Gln Val Gly Glu Arg Tyr Asp
420 425 430

Arg Ile Gln Leu Gln Glu Ala Ile Lys Ser Asn Tyr Arg His Ile Asn
435 440 445

Asn Phe Glu Gly Tyr Tyr Gln Arg Trp Gly Ala Val Gln Ser Phe Gln
450 455 460

Leu Trp Trp Asn Leu Gly Ser Pro Lys Ile Lys Ser Gln Tyr Gly Glu
465 470 475 480

Ser Arg Gln Lys Phe Gln Gln Glu Cys Glu Glu Val Phe Asp Thr Ser
485 490 495

Leu Lys Arg Val Ala Gly Arg Val Lys Gly Trp Ala Asp Glu Trp Lys
500 505 510

Glu Leu Ser Gly Lys Asn Gly Asn Pro Ile Phe Glu Asp Ala Ser Ser
515 520 525



Phe Arg Gly Ser Ser Pro Leu Leu Cys Gly Leu Tyr Asp Ser Thr Glu
530 535 540

Pro Glu Glu Cys Asp Arg Phe Lys Thr Tyr Asp Leu Pro Ser Ile Leu
545 550 555 560

Gly Asn Leu Glu Val Glu Val Trp Arg Lys Gly Glu Phe Lys Arg Gln
565 570 575

Ile Glu Ala Thr Lys Thr Pro Ile Ala Arg Arg Arg Phe Asp Tyr Cys
580 585 590

Leu Ala Phe Leu Asn Leu Lys Gly Tyr Arg Glu Glu Arg Leu Asn Trp
595 600 605

Arg Phe Thr Tyr Asp Gly Asp Leu Gly Glu Ile Ala Ser Ala Trp Lys
610 615 620

Val Gln Val Leu Thr Gly Ile Gly Val Trp Gln Pro Asp Asn Pro Trp
625 630 635 640

Leu Asp Arg Ile Ser Arg Glu Leu Arg Asp Leu Ala Leu Val Ser Phe
645 650 655

Val Phe Ala Tyr Pro Val Ala Ala Val Arg Gln Arg Leu Gln Leu Pro
660 665 670

Met His Phe Gly Ile Tyr Pro Ile Ser Asp Glu Ser Ser Leu His Ser
675 680 685

Pro Leu Ser Pro Tyr Ser Ile Ala Ile Gly Gln Ala Ala Leu Leu Leu
690 695 700

Asp Thr Leu Ala His Arg Phe Lys Gly Lys Gly Gly Glu Val Trp Ile
705 710 715 720

Cys

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<212> PRT

<213> Microcystis aeruginosa

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Glu Asp Ile Asp Leu Glu Ser Leu Val Ser Glu Asp Leu Gly Ile Thr
35 40 45

Glu Ser Val Ser Asp Arg Arg Val Glu Thr Val Arg Gln Leu Leu Thr
50 55 60

Leu Lys Leu Leu Arg Glu Ala Ile Arg Ala Glu Asn Pro Asp Asp Arg
65 70 75 80

Val Met Ala Asp Phe Ala Glu Met Val Leu Pro Asn Leu Leu Arg Leu
85 90 95

Ala Ile Gly Val Thr Ala Lys Gly Gly Asn Phe Ile Glu Ala Val Asp
100 105 110

Arg Gly Arg Glu Leu Arg Asn Lys Pro Lys Ala Lys Arg Asp Asn Ala
115 120 125

Gly Asp Gln Ser Leu Asn Thr His Leu Leu Asn Gly Leu Phe Pro Ala
130 135 140

Asn Leu Ile Glu Lys Arg Leu Gln Lys Leu Asn Thr Thr Val Arg Arg
145 150 155 160

Ile Ile Lys Glu Phe Glu Arg Arg Leu Ala Ile Ala Gly Phe Leu Val
165 170 175

His Asp Phe Glu Lys Phe Ser Tyr Asp Arg Phe Pro Ser Met Ser Glu
180 185 190

Arg Tyr Ile Gln Ile Gln Arg Asp Phe Ile Gln Asp Pro Phe Lys Asn
195 200 205

Gln Asp Pro Arg Lys Leu Ser Arg Glu Glu His Arg Glu Ile Leu Gln
210 215 220

Val Leu Ile Pro Glu Leu Gly Leu Asp Arg Phe Leu Phe Pro Asp Asn
225 230 235 240

Pro Glu Arg Trp Leu Glu Tyr Leu Asp Asp Leu Leu Tyr Ile Ala Lys
245 250 255

Asn Thr Gln Arg Arg Asn Asp Thr Asp Leu Asn Thr Ser Glu Asp Gly
260 265 270

Leu Asn Val Arg Leu Asn Asp Arg Val Ile Glu Ser Leu Cys Asp Leu



275

280

285

Ala Cys Leu Ala Asp Arg Leu Ala Ser Ile Ile Lys His Pro His Asp
290 295 300

Ala Glu Lys Ala Ser Leu Gln Asp Leu Leu Tyr Ser Leu Ser Asp Gly
305 310 315 320

Glu Leu Lys Phe Thr Tyr His Ser Ile Ala Glu Asn Arg Gly Val Leu
325 330 335

Thr Asn Val Leu Asn Asn Ala Val Met Glu Ala His Gln Glu Leu Asp
340 345 350

Tyr Gln Pro Leu Leu Tyr Leu Pro Thr Gly Val Val Tyr Ile Ala Pro
355 360 365

Lys Asn Ala Pro Glu Val Ser Leu Glu Thr Leu Pro Asn Arg Val Val
370 375 380

Asp Thr Ile Lys Ser Leu Cys Ser Gly Glu Leu Gln Arg Lys Gln Thr
385 390 395 400

Gly Phe Gly Arg Asp Gly Lys Gly Met Lys Tyr Ala Asp Tyr Tyr Ser
405 410 415

Gln Phe Phe Asp Asp Ala Gly Leu Met Arg Ala Ala Leu Asn Ala Thr
420 425 430

Leu Arg Ile Leu Gly Asp Asn Lys Ala Ser Val Ala Arg Ser Arg Gly
435 440 445

Glu Asn Leu Ile Lys Phe Gln Gln Gln Gly Val Leu Pro Thr Asp Tyr
450 455 460

Asp Phe His Cys Glu Asp Asp Ile Arg Ile Asp Arg Leu Ala Glu Phe
465 470 475 480

Gly Asp Val Val Thr Arg Lys Ile Trp Gly Asp Arg Leu Glu Lys Ile
485 490 495

Glu Gln Ala Arg Lys Leu Gln Lys Asn Leu Pro Ala Pro Pro Asp Leu
500 505 510

Asp Leu Ile Ser Glu Ile Ala His Tyr Trp Asn Leu Glu Asn Tyr Leu
515 520 525

Pro Gln Ile Arg Ala Ile Lys Arg Ile Asn Glu Ser Leu Lys Glu Leu
530 535 540



Lys Leu Lys Gly Asn Thr Gly Gly Val Pro Tyr Glu Trp Tyr Tyr Leu
 545 550 555 560

Ala Ala Gln Tyr Leu Lys Gln His Pro Gly Ile Glu Asp Ile Arg Pro
 565 570 575

Val Ala Glu Asp Leu Ile Ala Phe Leu Ala Ala Lys Ile Ala Ala Ile
 580 585 590

Val Ala Gly Tyr Asn Leu Pro Asp Gly Trp Glu Asp Leu Arg Glu Trp
 595 600 605

Val Asn Gln Val Val Gln Leu Pro Gly Arg Glu Leu Ala His Ser Ile
 610 615 620

Glu Thr Phe Gln Lys Glu Leu Asn His Tyr Asn Ala Ala Lys Lys Gln
 625 630 635 640

Gly Arg Gly Arg Gln Leu Leu Cys Ser Ile Ser His Ser Pro Tyr Ser
 645 650 655

Val Ser Glu Gln Met Glu Ser Ala Val Leu Phe Thr Pro Gln Val Tyr
 660 665 670

Thr Asn Lys Gln Met Leu Ala Gly Ser Asn Ala Lys Arg Asn Ile Ser
 675 680 685

Ser Ile Ala Gly Thr Glu Met Met Leu Arg Gln Ile Leu Met Asn Gln
 690 695 700

Thr Gln Ala Val Gly Lys Arg Phe Glu Asp Gly Lys Tyr Arg Tyr Leu
 705 710 715 720

Tyr Phe Tyr Pro Thr Tyr Tyr Phe Thr Pro Glu Thr Asn Ser Phe Leu
 725 730 735

Gln Lys Ala Tyr Ala Asn Ile Ala Gln Thr Arg Phe Asp Ser Ser Ile
 740 745 750

Lys Leu His Phe Val Asp Lys Asn Leu Val Ala Asn Phe Asp Arg Thr
 755 760 765

Arg Tyr Gln Ser Val Asp Ser Phe Leu Ile Asp Glu Lys Leu Arg Gln
 770 775 780

Lys Lys Glu Thr Ile Asn Glu Glu Glu Asp Gly Lys Lys Asp Arg Thr
 785 790 795 800



Phe Lys Leu Ser Tyr Pro Glu Asp Lys Pro Leu Thr Phe Tyr Phe Met
805 810 815

Ala Leu Pro Pro Gly Arg Asn Pro Thr Asp Thr Glu Ser Trp Val Met
820 825 830

Pro Ala Trp Leu Gly Leu Ala Phe Pro Met Ile Leu Asp Val Lys Thr
835 840 845

Val Val Ser Glu Ser Pro Ile Pro Pro Tyr Arg Asp Gly Ala Glu Phe
850 855 860

Glu Glu Thr Val Phe Leu Asp Ser Ala Pro Gln Ala Ile Arg Ser Leu
865 870 875 880

Thr Arg Cys Asp Arg Phe Arg Leu Asp Arg Val Leu Asn Pro Trp Gln
885 890 895

Asp Asn Asp Gly Lys Lys Tyr Ser Ala Pro Leu Asn Thr Leu Thr Ala
900 905 910

Ala Tyr Ser Ile His Leu Asp Val Asn Ser Lys Gln Gly Lys Thr Gly
915 920 925

Tyr Asp Pro Asn Trp Gly Lys Leu Thr Glu Leu Ala Ile Asn Leu Glu
930 935 940

Thr Ser Pro Leu Tyr Val Phe His Tyr Phe Lys Gln Trp Lys Arg Gly
945 950 955 960

Lys Asp Ala Asp Ile Pro Ser Ala Asn Arg Ile Ala Leu Tyr Leu Tyr
965 970 975

Asp Phe Tyr Pro Cys Phe Asp Pro Tyr Val Gln Ala Asn Arg Thr Asn
980 985 990

Leu Thr Ile Asp Met Thr Ala Glu Ser Pro Leu Asn His Pro Lys Asn
995 1000 1005

Leu Thr Glu Leu Tyr Arg Gln Phe Tyr Arg Ala Lys Ser Ser Lys
1010 1015 1020

Gly Lys Pro Ile Lys Ala Asn Ala Ile Leu Lys Pro Ile Asp Glu
1025 1030 1035

Ala Ala Asp Ile Ile Leu Lys Ala Asp Lys Ala Ile Ser Asp Asp
1040 1045 1050



Leu Thr Ser Leu Val Ala Ala Arg Leu Phe Lys Leu Met Asp Arg
1055 1060 1065

Val Arg Ser Gln Thr Ala Glu Gly Arg Tyr Val Ile Lys Glu Arg
1070 1075 1080

Asp Gln Glu Arg Glu Lys Ile Leu Asp Phe Ala Lys Tyr Phe Val
1085 1090 1095

Lys Asn Val Phe Glu Glu Ser Phe Glu Ser Asp Arg Ala Arg Leu
1100 1105 1110

Ala Gly Arg Gln Leu Asn Ile Ile Arg Asp Thr Cys Glu Phe Leu
1115 1120 1125

Tyr Arg Leu Glu Met Asp Lys Glu Arg Arg Gln Arg Gln Val Gln
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Pro Leu Asp Thr Ser Asn Ser Ser Ser Glu Glu Glu Glu
1145 1150 1155

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<220>
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nnnnnnnnnn nnguuccaau uaaucuuaag ccuauuagg gauugaaac 109

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<211> 675
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ctgggcctgg tgaacagcga tagctatcgc tattttctgca gcgaacagat tccgcagtat 180
caggaacatc tgaacccgct gaacgaagaa aaaatttatg tgaccccggc gcgcgcgatt 240



gcgcataccg cggtgctgaa cacctggaaa tatgcgaaca acaactatca tgtggaaatg	300
gaaaaaaccc agaaaaacat tccgagcttc ggccgcgcga aagaaattgc gccggaaagc	360
attttcgaat gcttcattat tagccatcat ccgctgcagc tgccgaaatg gattcgctg	420
ggcaaatgga tgagcaaagc ggaagtgaaa ctgaccgaac tgagcctgag caaacagaaa	480
gaagatctgt tcatttatcc gtatccgctg aaccgctgg atgtgatgtt caccatcag	540
gtgattggct atgatgtgat taacatgccg ccggtgagcc tgattcgaa cgtgcgcatg	600
cgcggcgaat attatcagat tagcgatcgc ccggatctga aaattccggc acgtctgagc	660
tatcatttcg gctaa	675

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 <212> DNA
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 <223> Cas6d nucleotide sequence for expression in Escherichia coli

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cagagcggcg gcaaaacat taacagcccg cagtggcgcc atgaacgcga aattgcgagc	240
gaaaccccg tctggtggcg cattagcctg ctggatgac gcctgttcgg caaactgacc	300
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aacagcctgc tgggccgctg gaaccgctat agcggcattc cgttcgatag cattgcgctg	600
gaaagcattt tcccagcctt cttcgatatt cagaccaaac tggcggatga agcgtataaa	660
aaccagagca ttggctgcgt gggcgaaatt cattatcgcc tgctgggcga agtggaaaccg	720
gcgaaaatta aagcgattaa cgcgctggcg gatttcgcgc tgtatgcggg cgtgggccgc	780
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<210> 9
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 <212> DNA
 <213> Artificial

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 <223> Cas7d nucleotide sequence for expression in Escherichia coli

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accgatggcg aactgaacaa agcgcgctg cgcgcgggca ttgacagcaa gaaaaccatt	180
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cataccgatt tcattggcg aaacttccag ccgctgctg ccgaagtga aaccctgacc	900
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<211> 2166

<212> DNA

<213> Artificial

<220>

<223> Cas3d nucleotide sequence for expression in Escherichia coli

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gctcttaacg acccagacat cgacgtgatc ttcaacctg ctatgactgg agacggaaag	180
tctcttgctg cttatcttag aactcttcag ggatatttcc caatcatggg actttatcca	240
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aactggctta aggagaacaa ggagagaatc cttgactatt tcaagagata tccaggatct	900
aagggagcta tcatccttaa ctctatcgct tctgtgaaga gacttcttcc aatcttcaga	960
gagcttcttg ctactatcgg acttactgtg ggagagaaca ctggactttc tggaactaga	1020
gagaaacttg cttctcttaa cagagatctt gtgatcggaa cttctactat cgacgtggga	1080
gtggacttca agatcaactt ctttatcttc gagtcttctg acgctggaaa cttcatccag	1140
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gctggaagag tgaagggatg ggctgacgag tggaaggagc tttctggaaa gaacggaaac	1560
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<211> 35

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<213> Artificial

<220>

<223> J23108 synthesis promoter

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35

<210> 13

<211> 36

<212> DNA

<213> Artificial



<220>
 <223> Ribosomal binding sequence

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 aataattttg tttaacttta agaaggagat atacat 36

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 <211> 24
 <212> DNA
 <213> Artificial

 <220>
 <223> Terminator sequence STOP767

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 <210> 15
 <211> 19
 <212> DNA
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 <220>
 <223> Terminator sequence STOP768(1)

 <400> 15
 cgccagggtt ttcccagtc 19

 <210> 16
 <211> 19
 <212> DNA
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 <220>
 <223> Terminator sequence TOP768(2)

 <400> 16
 cgccagggtt ttcccagtc 19

 <210> 17
 <211> 47
 <212> DNA
 <213> Artificial

 <220>
 <223> T7 terminator sequence

 <400> 17
 tagcataacc ccttggggcc tctaaacggg tcttgagggg ttttttg 47

 <210> 18
 <211> 37
 <212> DNA
 <213> Artificial

 <220>
 <223> CRISPR repeat sequence

 <400> 18



gttccaatta atcttaagcc ctattagga ttgaaac

37

<210> 19

<211> 19

<212> DNA

<213> Artificial

<220>

<223> T7 promoter sequence

<400> 19

taatacgact cactatagg

19

<210> 20

<211> 109

<212> DNA

<213> Artificial

<220>

<223> crRNA expression cassette

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gttccaatta atcttaagcc ctattagga ttgaaacggt aataatacga ctactatag

60

ggagaaagga tcgttccaat taatcttaag ccctattagg gattgaaac

109

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<211> 581

<212> DNA

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<220>

<223> Synthesis ccdB gene expression cassette

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120

caatatctcc ggtctggtaa gcacaacat gcagaatgaa gcccgctgct tgcgtgccga

180

acgctggaaa gcggaaaatc aggaaggat ggctgaggtc gcccggttta ttgaaatgaa

240

cggctctttt gctgacgaga acagggactg gtgaaatgca gtttaagggtt tacacctata

300

aaagagagag ccgttatcgt ctgtttgtgg atgtacagag tgatattatt gacacgccc

360

ggcgacggat ggtgatcccc ctggccagt gacgtctgct gtcagataaa gtctcccggtg

420

aactttaccc ggtggtgcat atcggggatg aaagctggcg catgatgacc accgatatgg

480

ccagtgtgcc ggtctccgtt atcggggaag aagtggctga tctcagccac cgcgaaaatg

540

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581

<210> 22

<211> 18

<212> PRT

<213> Artificial

<220>



<223> Nuclear localizing signal (NLS) amino acid sequence

<400> 22

Asp Pro Lys Lys Lys Arg Lys Val Asp Pro Lys Lys Lys Arg Lys Val
1 5 10 15

Ser Gly

<210> 23

<211> 54

<212> DNA

<213> Artificial

<220>

<223> NLS nucleotide sequence

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<210> 24

<211> 20

<212> PRT

<213> Artificial

<220>

<223> Self-cleaving peptide 2A amino acid sequence

<400> 24

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

Asn Pro Gly Pro
20

<210> 25

<211> 60

<212> DNA

<213> Artificial

<220>

<223> Self-cleaving peptide 2A(1) coding sequence

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ggctctgagg gcagaggcag cctgctgacc tgcggcgacg tggaggaaaa ccctggccct 60

<210> 26

<211> 60

<212> DNA

<213> Artificial

<220>

<223> Self-cleaving peptide 2A(2) coding sequence

<400> 26

gggtctgagg gacgcggctc cctgctcacc tgttgagatg tggaagagaa ccaggcccc 60



<210> 27
 <211> 60
 <212> DNA
 <213> Artificial

 <220>
 <223> Self-cleaving peptide 2A(3) coding sequence

 <400> 27
 ggttctgaag gcagaggctc tctgctgaca tgtggggatg tggaggaaaa tcctggccct 60

 <210> 28
 <211> 60
 <212> DNA
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 <220>
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 <400> 28
 ggatccgagg gcagaggaag tctgctaaca tgcggtgacg ttgaggagaa tcccgggcca 60

 <210> 29
 <211> 820
 <212> DNA
 <213> Artificial

 <220>
 <223> 2 x cauliflower mosaic virus 35S gene promoter + omega sequence

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 caacaaacaa caacaacat tacaattact atttacaatt 820

 <210> 30
 <211> 447



<212> DNA
 <213> Artificial

 <220>
 <223> Arabidopsis shock protein 18.2kDa gene terminator

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 gtaagatctc attataatga ataaacaaat gtttctataa tccattgtga atgttttggt 180

 ggatctcttc tgcagcatat aactactgta tgtgctatgg tatggactat ggaatatgat 240

 taaagataag atgggctcat agagtaaaac gaggcgaggg acctataaac ctcccttcat 300

 catgctattt catgatctat ttataaaaat aaagatgtag aaaaaagtaa gcgtaataac 360

 cgcaaaacaa atgatttaaa acatggcaca taatgaggag attaagttcg gtttacgttt 420

 attttagtac taattgtaac gtgagac 447

 <210> 31
 <211> 97
 <212> DNA
 <213> Artificial

 <220>
 <223> crRNA expression cassette

 <400> 31
 gttccaatta atcttaagcc ctattaggga ttgaaacgga gaccctcaat tgcggtctc 60

 gttccaatta atcttaagcc ctattaggga ttgaaac 97

 <210> 32
 <211> 448
 <212> DNA
 <213> Artificial

 <220>
 <223> Arabidopsis U6 snRNA-26 gene promoter sequence

 <400> 32
 aagcttcgtt gaacaacgga aactcgactt gccttccgca caatacatca tttcttctta 60

 gctttttttt ttcttcttcg ttcatacagt ttttttttgt ttatcagctt acattttctt 120

 gaaccgtagc tttcgttttc ttctttttta ctttccattc ggagtttttg tatcttgttt 180

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<211> 723
<212> DNA
<213> Artificial

<220>
<223> 2xNLS + Cas5d

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<210> 34
<211> 882
<212> DNA
<213> Artificial

<220>
<223> 2xNLS + Cas6d

<400> 34
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<210> 35
 <211> 1053
 <212> DNA
 <213> Artificial

<220>
 <223> 2xNLS + Cas7d

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 <211> 2217
 <212> DNA
 <213> Artificial

<220>
 <223> 2xNLS + Cas3d

<400> 36



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ccaatcgcta gaagaagatt cgactattgc cttgctttcc ttaaccttaa gggatataga	1860
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<220>
 <223> 2xNLS + Cas10d

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<210> 38
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 <213> Nicotiana benthamiana

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<210> 39
 <211> 35
 <212> DNA
 <213> Nicotiana benthamiana

<400> 39	
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<210> 40
 <211> 35
 <212> DNA
 <213> Solanum lycopersicum

<400> 40	
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<210> 41
 <211> 811
 <212> DNA
 <213> Artificial

<220>
 <223> Cytomegalovirus enhancer + universal chicken beta-actin gene hybrid promoter

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<210> 42
 <211> 223
 <212> DNA
 <213> Artificial

<220>
 <223> Bovine-derived growth hormone gene terminator sequence

<400> 42	
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ttgcatcgca ttgtctgagt aggtgtcatt ctattctggg gggtaggggtg gggcaggaca	180
gcaaggggga ggattgggaa gagaatagca ggcatgctgg gga	223

<210> 43
 <211> 249
 <212> DNA
 <213> Artificial

<220>
 <223> Human U6 snRNA gene promoter

<400> 43	
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ataattggaa ttaatttgac tgtaaacaca aagatattag taaaaatac gtgacgtaga	120
aagtaataat ttcttgggta gtttgcagtt ttaaaattat gttttaaaat ggactatcat	180
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cgaacacc	249

<210> 44
 <211> 35
 <212> DNA
 <213> Homo sapiens

<400> 44	
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<210> 45
 <211> 35
 <212> DNA
 <213> Homo sapiens

<400> 45	
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