



LOCUS	Exported	6902 bp ds-DNA	circular SYN 05-
NOV-2020			
DEFINITION	synthetic circular DNA		
ACCESSION	.		
VERSION	.		
KEYWORDS	pCLIP-gRNA-EFS-ZsGreen		
SOURCE	synthetic DNA construct		
ORGANISM	synthetic DNA construct		
REFERENCE	1 (bases 1 to 6902)		
AUTHORS	Trial User		

```

TITLE      Direct Submission
JOURNAL    Exported Thursday, Nov 5, 2020 from SnapGene 5.2.1
           https://www.snapgene.com
FEATURES   Location/Qualifiers
   source   1..6902
            /organism="synthetic DNA construct"
            /mol_type="other DNA"
   misc_feature 23..667
            /label=CMV-LTR
   promoter 23..404
            /label=CMV-
   misc_feature 720..745
            /label=psi
   misc_feature 746..840
            /label=HIV-1 Psi
            /note="packaging signal of human immunodeficiency
virus
   misc_feature 1333..1566
            /label=RRE
            /note="The Rev response element (RRE) of HIV-1
allows for
            Rev-dependent mRNA export from the nucleus to the
            cytoplasm."
   misc_feature 2093..2210
            /label=cPPT/CTS
            /note="central polypurine tract and central
termination
            sequence of HIV-1"
   promoter 2261..2501
            /label=U6 Promoter
            /note="RNA polymerase III promoter for human U6
snRNA"
   gap      2511..2530
            /estimated_length=20
   misc_RNA 2531..2606
            /label=gRNA scaffold
            /note="guide RNA scaffold for the CRISPR/Cas9
system"
   misc_feature 2648..2880
            /label=EFS promoter
   primer_bind 2899..3594
            /label=seq.SFFVfw
   CDS      2899..3594
            /codon_start=1
            /label=ZsGreen

```

```

/translation="MAQSKHGLTKEMTMKYRMEGCVDGCHKFVITGEGIGYPFKGKQAIN
LCVVEGGPLPFAEDILSAAFMYGNRVFTEYYPQDIVDYFKNSCPAGYTWDRSFLFEDGAV
CICNADITVSVEENCMYHESKFGVNF PADGPVMMKMTDNWEPSC EKII PVPKQGILKG
DVSMYLLLKDGGR LRCQFDTVYKAKSVPRKMPDWHFIQHKL TREDRSDAKNQKWHLTEH

```

```

        primer_bind      AIASGSALP"
                        2899..2964
                        /label=seq.ZsGrev
        primer_bind      3489..3594
                        /label=seq.ZsGfw
        misc_feature      3596..3601
                        /label=MCS
        misc_feature      3608..4199
                        /label=WPRE
        misc_feature      4404..4639
                        /label=3'LTR
        promoter          4733..5062
                        /label=SV40
                        /note="SV40 enhancer and early promoter"
        rep_origin        4913..5048
                        /label=ori
                        /note="SV40 origin of replication"
        rep_origin        complement(5151..5736)
                        /direction=LEFT
                        /label=ori
                        /note="high-copy-number ColE1/pMB1/pBR322/pUC origin
of
                        replication"
        CDS                complement(5907..6767)
                        /codon_start=1
                        /gene="bla"
                        /product="beta-lactamase"
                        /label=AmpR
                        /note="confers resistance to ampicillin,
carbenicillin, and
                        related antibiotics"

/translacion="MSIQHFRVALIPFFAAFCCLPVFAHPETLVKVKDAEDQLGARVGYI
ELDLNSGKILESFRPEERFPMMSSTFKVLLCGAVLSRIDAGQEQLGRRIHYSQNDLVEYS
PVTEKHLTDGMTVRELCSAAITMSDNTAANLLLTIGGPKELTAFLHNMGDHSVTRLDRW
EPELNEAIPNDERDITMPVAMATTLRKLTLGELLTLASRQQLIDWMEADKVAGPLLRSA
LPAGWFIADKSGAGERGSRGIIAALGPDGKPSRIVVIYTTGSQATMDERNRQIAEIGAS
        LIKHW"
        promoter          complement(6768..6872)
                        /gene="bla"
                        /label=AmpR promoter
ORIGIN
      1  gacggatcgg gagatcctcg cgcgttgaca ttgattattg actagttatt
aatagtaatc
      61  aattacgggg tcattagttc atagcccata tatggagttc cgcgttacat
aacttacggt
     121  aaatggcccg cctggctgac cgcccaacga cccccgccca ttgacgtcaa
taatgacgta
     181  tgttcccata gtaacgcaa tagggacttt ccattgacgt caatgggtgg
agtatttacg

```

241 gtaaactgcc cacttggcag tacatcaagt gtatcatatg ccaagtacgc
cccctattga
301 cgtcaatgac ggtaaattggc cgcctggca ttatgccag tacatgacct
tatgggactt
361 tcctacttgg cagtacatct acgtattagt catcgctatt accagctagc
gaggcgtggc
421 ctgggcggga ctggggagtg gcgagccctc agatcctgca tataagcagc
tgctttttgc
481 ctgtactggg tctctctggg tagaccagat ctgagcctgg gagctctctg
gctaactagg
541 gaaccactg cttaagcctc aataaagctt gccttgagtg cttcaagtag
tgtgtgccccg
601 tctgttgtgt gactctggta actagagatc cctcagacc ttttagtcag
tgtggaaaat
661 ctctagcagt ggcgccgaa cagggacttg aaagcgaag ggaaccaga
ggagctctct
721 cgacgcagga ctcggttgc tgaagcgcgc acggcaagag gcgaggggcg
gcgactgggtg
781 agtacccaa aaattttgac tagcggaggc tagaaggaga gagatgggtg
cgagagcgtc
841 agtattaagc gggggagaat tagatcgca tgggaaaaa ttcggttaag
gccaggggga
901 aagaaaaaat ataaattaa acatatagta tgggcaagca gggagctaga
acgattcgca
961 gttaatcctg gcctgttaga aacatcagaa ggctgtagac aaatactggg
acagctacaa
1021 ccatcccttc agacaggatc agaagaactt agatcattat ataatacagt
agcaaccctc
1081 tattgtgtgc atcaaaggat agagataaaa gacaccaagg aagctttaga
caagatagag
1141 gaagagcaaa acaaaagtaa gaccaccgca cagcaagcgg ccgctgatct
tcagacctgg
1201 aggaggagat atgagggaca attggagaag tgaattatat aaatataaag
tagtaaaaat
1261 tgaaccatta ggagtagcac ccaccaaggc aaagagaaga gtggtgcaga
gagaaaaaag
1321 agcagtggga ataggagctt tgttccttgg gttcttggga gcagcaggaa
gcactatggg
1381 cgcagcgtca atgacgctga cggtagcagg cagacaatta ttgtctggta
tagtgcagca
1441 gcagaacaat ttgctgaggg ctattgaggc gcaacagcat ctggtgcaac
tcacagtctg
1501 gggcatcaag cagctccagg caagaatcct ggctgtggaa agatacctaa
aggatcaaca
1561 gtcctgggg atttgggggt gctctggaaa actcatttgc accactgctg
tgccttgaa
1621 tgctagttag agtaataaat ctctggaaca gatttggaa cacacgacct
ggatggagtg
1681 ggacagagaa attaacaatt acacaagctt aatacactcc ttaattgaag
aatcgcaaaa
1741 ccagcaagaa aagaatgaac aagaattatt ggaattagat aaatgggcaa
gtttgtggaa
1801 ttggtttaac ataacaatt ggctgtggta tataaaatta ttcataatga
tagtaggagg

1861 cttggtaggt ttaagaatag tttttgctgt actttctata gtgaatagag
ttaggcaggg
1921 atattcacca ttatcgtttc agaccacct cccaaccccg aggggacccg
acaggcccga
1981 aggaatagaa gaagaaggtg gagagagaga cagagacaga tccattcgat
tagtgaacgg
2041 atcggcactg cgtgcgccaa ttctgcagac aatggcagt attcatccac
aattttaaaa
2101 gaaaaggggg gattgggggg tacagtgcag gggaaagaat agtagacata
atagcaacag
2161 acatacaaac taaagaatta caaaaacaaa ttacaaaaat tcaaaatfff
cgggtttatt
2221 acagggacag cagagatcca gtttggttaa ttaaggtacc gagggcctat
ttcccatgat
2281 tccttcatat ttgcatatac gatacaaggc tgtagagag ataattagaa
ttaatttgac
2341 tgtaaacaca aagatattag tacaaaatac gtgacgtaga aagtaataat
ttcttgggta
2401 gtttgcagtt ttaaaattat gttttaaaat ggactatcat atgcttaccg
taacttgaaa
2461 gtatttcgat ttcttggctt tatatatctt gtggaaagga cgaaacaccg
nnnnnnnnnn
2521 nnnnnnnnnn gtttagagc tagaaatagc aagttaaaat aaggctagtc
cgttatcaac
2581 ttgaaaaagt ggcaccgagt cgggtgctttt ttgaattcgc tagctaggtc
ttgaaaggag
2641 tgggaattgg ctccggtgcc cgtcagtggg cagagcgcac atcgcccaca
gtccccgaga
2701 agttgggggg aggggtcggc aattgatccg gtgcctagag aagggtggcg
ggggtaaact
2761 gggaaagtga tgtcgtgtac tggctccgcc tttttcccga ggggtggggga
gaaccgtata
2821 taagtgcagt agtcgccgtg aacgttcttt ttcgcaacgg gtttgccgcc
agaacacagg
2881 accggttcta gagcgacat ggcccagtcc aagcacggcc tgaccaagga
gatgaccatg
2941 aagtaccgca tggagggctg cgtggacggc cacaagtctg tgatcaccgg
cgagggcatc
3001 ggctaccctt tcaagggcaa gcaggccatc aacctgtgcg tgggtggagg
cggccccttg
3061 cccttcgccg aggacatctt gtccgccgcc ttcattgtac gcaaccgcgt
gttaccggag
3121 tacccccagc acatcgtcga ctacttcaag aactcctgcc ccgccggcta
cacctgggac
3181 cgctccttcc tgttcgagga cggcgccgtg tgcattctgca acgccgacat
caccgtgagc
3241 gtggaggaga actgcatgta ccacgagtcc aagttctacg gcgtgaactt
ccccgccgac
3301 ggccccgtga tgaagaagat gaccgacaac tgggagccct cctgcgagaa
gatcatcccc
3361 gtgcccgaagc agggcatctt gaagggcgac gtgagcatgt acctgctgct
gaaggacggt
3421 ggccgcttgc gctgccagtt cgacaccgtg tacaaggcca agtccgtgcc
ccgcaagatg

3481 cccgactggc acttcatcca gcacaagctg acccgcgagg accgcagcga
cgccaagaac
3541 cagaagtggc acctgaccga gcacgccatc gcctccggct ccgccttgcc
ctgaacgcgt
3601 ctggaacaat caacctctgg attacaaaat ttgtgaaaga ttgactggta
ttcttaacta
3661 tgttgctcct tttacgctat gtggatacgc tgctttaatg cctttgtatc
atgctattgc
3721 ttcccgtatg gctttcattt tctcctcctt gtataaatcc tggttgctgt
ctctttatga
3781 ggagttgtgg cccgttgca ggcaacgtgg cgtggtgtgc actgtgtttg
ctgacgcaac
3841 cccactggg tggggcattg ccaccacctg tcagctcctt tccgggactt
tcgctttccc
3901 cctccctatt gccacggcgg aactcatcgc cgctgcctt gcccgctgct
ggacaggggc
3961 tcggctggtg ggcactgaca attccgtggt gttgtcgggg aagctgacgt
cctttccatg
4021 gctgctcgcc tgtgttgcca cctggattct gcgcgggacg tccttctgct
acgtcccttc
4081 ggccctcaat ccagcggacc ttccttcccg cggcctgctg ccggctctgc
ggcctcttcc
4141 gcgtcttcgc cttcgccctc agacgagtcg gatctccctt tgggccgcct
ccccgcctgg
4201 aattaattct gcagtcgaga cctagaaaaa catggagcaa tcacaagtag
caatacagca
4261 gtaccaatg ctgattgtgc ctggctagaa gcacaagagg aggaggagg
gggtttttcc
4321 agtcacacct caggtacctt taagaccaat gacttacaag gcagctgtag
atcttagcca
4381 ctttttaaaa gaaaagaggg gactggaagg gctaattcac tccaacgaa
gacaagatct
4441 gctttttgct tgtactgggt ctctctggtt agaccagatc tgagcctggg
agctctctgg
4501 ctaactaggg aaccactgc ttaagcctca ataaagcttg ccttgagtgc
ttcaagtagt
4561 gtgtgcccgt ctgttgtgtg actctggtaa ctagagatcc ctcagacct
tttagtcagt
4621 gtggaaaatc tctagcagta gtagttcatg tcatcttatt attcagtatt
tataacttgc
4681 aaagaaatga atatcagaga gtgagaggcc ttgacattgt ttaaactgga
atgtgtgtca
4741 gttaggggtg ggaaagtccc caggctcccc agcaggcaga agtatgcaaa
gcatgcatct
4801 caattagtc gcaaccagggt gtggaaagtc cccaggctcc ccagcaggca
gaagtatgca
4861 aagcatgcat ctcaattagt cagcaacat agtcccgcc ctaactccgc
ccatcccgcc
4921 cctaactccg cccagttccg cccattctcc gccccatggc tgactaattt
ttttattta
4981 tgcagaggcc gaggccgct ctgcctctga gctattccag aagtagtgag
gaggcttttt
5041 tggaggccta ggcttttgca aaaagctccc gggagcttgt atatccatta
catgtgagca

5101 aaaggccagc aaaaggccag gaaccgtaaa aaggccgcgt tgctggcggt
tttccatagg
5161 ctccgcccc ctgacgagca tcacaaaaat cgacgctcaa gtcagagggtg
gcgaaaccgg
5221 acaggactat aaagatacca ggcgtttccc cctggaagct ccctcgtgcg
ctctcctggt
5281 ccgaccctgc cgcttaccgg atacctgtcc gcctttctcc cttcgggaag
cgtggcgctt
5341 tctcatagct cacgctgtag gtatctcagt tcggtgtagg tcgttcgctc
caagctgggc
5401 tgtgtgcacg aacccccgt tcagcccgac cgctgcgcct tatccgtaa
ctatcgtctt
5461 gagtccaacc cgtaagaca cgacttatcg cactggcag cagccactgg
taacaggatt
5521 agcagagcga ggtatgtagg cggtgctaca gagttcttga agtggtggcc
taactacggc
5581 tacactagaa gaacagtatt tggatctgc gctctgctga agccagttac
cttcggaaaa
5641 agagttggta gctcttgatc cggcaaaca accaccgctg gtagcggttt
ttttgttgc
5701 aagcagcaga ttacgcgcag aaaaaagga tctcaagaag atcctttgat
cttttctacg
5761 gggctctgacg ctcagtggaa cgaaaactca cgtaagggga ttttggtcat
gagattatca
5821 aaaaggatct tcacctagat ctttttaaat taaaaatgaa gttttaaatc
aatctaaagt
5881 atatatgagt aaacttggtc tgacagttac caatgcttaa tcagtgaggc
acctatctca
5941 gcgatctgtc tatttcgttc atccatagtt gcctgactcc ccgctcgtgta
gataactacg
6001 atacgggagc gcttaccatc tggccccagt gctgcaatga taccgcgaga
cccacgctca
6061 ccggctccag atttatcagc aataaaccag ccagccggaa gggccgagcg
cagaagtggg
6121 cctgcaactt tatccgcctc catccagtct attaattggt gccgggaagc
tagagtaagt
6181 agttcgccag ttaatagttt gcgcaacggt gttgccattg ctacaggcat
cgtggtgtca
6241 cgctcgtcgt ttggtatggc ttcattcagc tccggttccc aacgatcaag
gcgagttaca
6301 tgatccccca tgttgtgcaa aaaagcgggt agctccttcg gtcctccgat
cgttgtcaga
6361 agtaagttgg ccgcagtgtt atcactcatg gttatggcag cactgcataa
ttctcttact
6421 gtcatgccat ccgtaagatg cttttctgtg actggtgagt actcaaccaa
gtcattctga
6481 gaatagtgta tgccggcacc gagttgctct tgcccggcgt caatacggga
taataccgcg
6541 ccacatagca gaactttaa agtgctcatc attggaaaac gttcttcggg
gcgaaaactc
6601 tcaaggatct taccgctggt gagatccagt tcgatgtaac cactcgtgc
acccaactga
6661 tcttcagcat cttttacttt caccagcgtt tctgggtgag caaaaacagg
aaggcaaaat

```
6721 gccgcaaaa aggaataag ggcgacacgg aatggtgaa tactcact  
cttcctttt  
6781 caatattatt gaagcattta tcagggttat tgtctcatga gcggatacat  
attgaaatgt  
6841 atttagaaaa ataaacaat aggggttccg cgcacatttc cccgaaaagt  
gccacctgac  
6901 gt  
//
```