



LOCUS	Exported	6605 bp ds-DNA	circular SYN 06-
NOV-2020			
DEFINITION	synthetic circular DNA		
ACCESSION	.		
VERSION	.		
KEYWORDS	pCLIP-gRNA-EFS-Blasticidin		
SOURCE	synthetic DNA construct		
ORGANISM	synthetic DNA construct		
REFERENCE	1 (bases 1 to 6605)		
AUTHORS	Trial User		

```

TITLE      Direct Submission
JOURNAL    Exported Friday, Nov 6, 2020 from SnapGene 5.2.1
           https://www.snapgene.com
FEATURES   Location/Qualifiers
    source      1..6605
               /organism="synthetic DNA construct"
               /mol_type="other DNA"
    misc_feature 23..667
               /label=CMV-LTR
    misc_feature 720..745
               /label=psi
    misc_feature 746..840
               /label=HIV-1 Psi
               /note="packaging signal of human immunodeficiency
virus
    misc_feature type 1"
               1333..1566
               /label=RRE
               /note="The Rev response element (RRE) of HIV-1
allows for
    misc_feature Rev-dependent mRNA export from the nucleus to the
               2093..2210
               /label=cPPT/CTS
               /note="central polypurine tract and central
termination
    promoter    sequence of HIV-1"
               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
    CDS         2899..3297
               /codon_start=1
               /gene="Aspergillus terreus BSD"
               /product="blasticidin S deaminase"
               /label=BSD
               /note="confers resistance to blasticidin"

/translation="MAKPLSQEESTLIERATATINSIPISEDYSVASAALSSDGRIFTG
VNVYHFTGGPCAELVVLGTAAAAAAGNLTTCIVAIGNENRGILSPCGRCRQVLLDLHPGI
    misc_feature KAIVKDSGDGQPTAVGIRELLPSGYVWEG"
               3299..3304
               /label=MCS
    misc_feature 3311..3902
               /label=WPRE

```

```

misc_feature      4107..4342
                  /label=3'LTR
promoter          4436..4765
                  /label=SV40
                  /note="SV40 enhancer and early promoter"
rep_origin        complement(4854..5439)
                  /direction=LEFT
                  /label=ori
                  /note="high-copy-number ColE1/pMB1/pBR322/pUC origin
of

```

```

                  replication"
CDS               complement(5610..6470)
                  /codon_start=1
                  /gene="bla"
                  /product="beta-lactamase"
                  /label=AmpR
                  /note="confers resistance to ampicillin,
carbenicillin, and
                  related antibiotics"

```

```

/translation="MSIQHFRVALIPFFAAFCCLPVFAHPETLVKVKDAEDQLGARVGYI
ELDLSNGKILESFRPEERFPMMSSTFKVLLCGAVLSRIDAGQEQLGRRIHYSQNDLVEYS
PVTEKHLTDGMTVRELCSAAITMSDNTAANLLLTIGGPKELTAFLHNMGDHSVTRLDWR
EPELNEAIPNDERDITMPVAMATTLRKLTLGELLTLASRQQLIDWMEADKVAGPLLRS
LPAGWFIADKSGAGERGSRGIIAALGPDGKPSRIVVIYTTGSQATMDERNRQIAEIGAS
LIKHW"

```

```

promoter          complement(6471..6575)
                  /gene="bla"
                  /label=AmpR promoter

```

ORIGIN

```

      1 gacggatcgg gagatcctcg cgcgctgaca ttgattattg actagttatt
aatagtaatc
     61 aattacgggg tcattagttc atagcccata tatggagttc cgcgctacat
aacttacggt
    121 aaatggcccc cctggctgac cgcccaacga cccccgcca ttgacgtcaa
taatgacgta
    181 tgttcccata gtaacgcaa tagggacttt ccattgacgt caatgggtgg
agtatttacg
    241 gtaaactgcc cacttggcag tacatcaagt gtatcatatg ccaagtacgc
cccctattga
    301 cgtcaatgac ggtaaattgc cgcctggca ttatgccag tacatgacct
tatgggactt
    361 tcctacttgg cagtacatct acgtattagt catcgctatt accagctagc
gaggcgtggc
    421 ctgggcgggg ctggggagtg gcgagccctc agatcctgca tataagcagc
tgctttttgc
    481 ctgtactggg tctctctggt tagaccagat ctgagcctgg gagctctctg
gctaactagg
    541 gaaccactg cttaagcctc aataaagctt gccttgagtg cttcaagtag
tgtgtgcccc

```

601 tctgttgtgt gactctggta actagagatc cctcagacct ttttagtcag
tgtggaaaat
661 ctctagcagt ggcgcccga cagggacttg aaagcgaag ggaaaccaga
ggagctctct
721 cgacgcagga ctcggttgc tgaagcgcgc acggcaagag gcgaggggcg
gcgactgggtg
781 agtacgcaa aaatthtgac tagcggaggc tagaaggaga gagatgggtg
cgagagcgtc
841 agtattaagc gggggagaat tagatcgcga tgggaaaaaa 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 ctgttgcaac
tcacagtctg
1501 gggcatcaag cagctccagg caagaatcct ggctgtggaa agatacctaa
aggatcaaca
1561 gctcctgggg atttgggggt gctctggaaa actcatttgc accactgctg
tgccttggaa
1621 tgctagttagt agtaataaat ctctggaaca gatttggaaat cacacgacct
ggatggagtg
1681 ggacagagaa attaacaatt acacaagctt aatacactcc ttaattgaag
aatcgcaaaa
1741 ccagcaagaa aagaatgaac aagaattatt ggaattagat aaatgggcaa
gtttgtggaa
1801 ttggtttaac ataacaatt ggctgtggta tataaaatta ttcataatga
tagtaggagg
1861 cttggttagt ttaagaatag tttttgctgt actttctata gtgaatagag
ttaggcaggg
1921 atattcacca ttatcgtttc agaccacct cccaaccccg aggggacctg
acaggcccga
1981 aggaatagaa gaagaagggt gagagagaga cagagacaga tccattcgat
tagtgaacgg
2041 atcggcactg cgtgcgcaa ttctgcagac aaatggcagt attcatccac
aattttaaaa
2101 gaaaaggggg gattgggggg tacagtgcag gggaaagaat agtagacata
atagcaacag
2161 acatacaaac taaagaatta caaaaacaaa ttacaaaaat tcaaaattht
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 gttttagagc tagaaatagc aagttaaaat aaggctagtc
cgttatcaac
2581 ttgaaaaagt ggcaccgagt cggtgctttt 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 ggccaagcct ttgtctcaag aagaatccac
cctcattgaa
2941 agagcaacgg ctacaatcaa cagcatcccc atctctgaag actacagcgt
cgccagcgca
3001 gctctctcta gcgacggccg catcttcaact ggtgtcaatg tataatcatt
tactggggga
3061 ccttgtgcag aactcgtggt gctgggcact gctgctgctg cggcagctgg
caacctgact
3121 tgtatcgtcg cgatcggaaa tgagaacagg ggcatcttga gcccctgcgg
acggtgccga
3181 caggtgcttc tcgatctgca tcttgggatc aaagccatag tgaaggacag
tgatggacag
3241 ccgacggcag ttgggattcg tgaattgctg ccctctgggt atgtgtggga
gggctaaacg
3301 cgtctggaac aatcaacctc tggattacaa aatttgtgaa agattgactg
gtattcttaa
3361 ctatgttgct cttttacgc tatgtggata cgctgcttta atgcctttgt
atcatgctat
3421 tgcttcccgt atggctttca ttttctcctc cttgtataaa tcttggttgc
tgtctcttta
3481 tgaggagtgt tggcccgttg tcaggcaacg tggcgtggtg tgcaactgtgt
ttgctgacgc
3541 aacccccact ggttggggca ttgccaccac ctgtcagetc ctttccggga
ctttcgcttt
3601 ccccctccct attgccacgg cggaaactcat cgccgcctgc cttgcccgt
gctggacagg
3661 ggctcggctg ttgggactg acaattccgt ggtgttgcg gggaaactga
cgtcctttcc
3721 atggctgctc gctgtgttg ccacctggat tctgcgcggg acgtccttct
gctacgtccc
3781 ttggccctc aatccagcgg accttcttc ccgcccctg ctgccggctc
tgccgctct

3841 tccgcgtcct cgccttcgcc ctcagacgag tcggatctcc ctttgggccg
cctccccgcc
3901 tggaaattaat tctgcagtcg agacctagaa aaacatggag caatcacaag
tagcaataca
3961 gcagctacca atgctgattg tgcttgcta gaagcacaag aggaggagga
ggtgggtttt
4021 tccagtcaca cctcaggtac ctttaagacc aatgacttac aaggcagctg
tagatcttag
4081 ccacttttta aaagaaaaga ggggactgga agggctaatt cactcccaac
gaagacaaga
4141 tctgcttttt gcttgactg ggtctctctg gttagaccag atctgagcct
gggagctctc
4201 tggctaacta ggaacccac tgcttaagcc tcaataaagc ttgccttgag
tgcttcaagt
4261 agtgtgtgcc cgtctgttgt gtgactctgg taactagaga tccctcagac
ccttttagtc
4321 agtgtggaaa atctctagca gtagtagttc atgtcatctt attattcagt
attataact
4381 tgcaaagaaa tgaatatcag agagtgagag gccttgacat tgtttaaact
ggaatgtgtg
4441 tcagttaggg tgtggaaagt ccccaggctc cccagcaggc agaagtatgc
aaagcatgca
4501 tctcaattag tcagcaacca ggtgtggaaa gtccccaggc tccccagcag
gcagaagtat
4561 gcaaagcatg catctcaatt agtcagcaac catagtcccg cccctaactc
cgcccatccc
4621 gccctaact cgcgccagtt cgcgccatc tccgccccat ggctgactaa
tttttttat
4681 ttatgcagag gccgaggccg cctctgcctc tgagctattc cagaagtagt
gaggaggctt
4741 ttttggaggc ctaggctttt gcaaaaagct cccgggagct tgtatatcca
ttacatgtga
4801 gcaaaaggcc agcaaaaggc caggaaccgt aaaaaggccg cgttgctggc
gtttttccat
4861 aggctccgcc ccctgacga gcatcacaaa aatcgacgct caagtcagag
gtggcgaaac
4921 ccgacaggac tataaagata ccaggcgttt ccccctggaa gctccctcgt
gcgctctcct
4981 gttccgacct tgccgcttac cggatacctg tccgccttcc tcccttcggg
aagcgtggcg
5041 ctttctcata gctcacgctg taggtatctc agttcgggtg aggtcgttcg
ctccaagctg
5101 ggctgtgtgc acgaaccccc cgttcagccc gaccgctgcg ccttatccgg
taactatcgt
5161 cttgagtcca acccggaag acacgactta tcgccactgg cagcagccac
tggtaacagg
5221 attagcagag cgaggatgt aggcggtgct acagagttct tgaagtgggtg
gcctaactac
5281 ggctacacta gaagaacagt atttggtatc tgcgctctgc tgaagccagt
taccttcgga
5341 aaaagagttg gtagctcttg atccggcaaa caaacaccg ctggtagcgg
ttttttggtt
5401 tgcaagcagc agattacgcg cagaaaaaaa ggatctcaag aagatccttt
gatcttttct

```
5461 acggggtctg acgctcagtg gaacgaaaac tcacgttaag ggattttggt
catgagatta
5521 tcaaaaagga tcttcaccta gatcctttta aattaaaaat gaagttttta
atcaatctaa
5581 agtatatatg agtaaacttg gtctgacagt taccaatgct taatcagtga
ggcacctatc
5641 tcagcgatct gtctatttcg ttcattccata gttgcctgac tccccgtcgt
gtagataact
5701 acgatacggg agggcttacc atctggcccc agtgctgcaa tgataccgcg
agaccacgc
5761 tcaccggctc cagatttatc agcaataaac cagccagccg gaagggccga
gcgcagaagt
5821 ggtcctgcaa ctttatccgc ctccatccag tctattaatt gttgccggga
agctagagta
5881 agtagttcgc cagttaatag tttgcgcaac gttggtgcca ttgctacagg
catcgtggtg
5941 tcacgctcgt cgtttggtat ggcttcattc agctccgggt cccaacgatc
aaggcgagtt
6001 acatgatccc ccatgttgtg caaaaagcg gttagctcct tcggctcctc
gatcgttgtc
6061 agaagtaagt tggccgcagt gttatcactc atggttatgg cagcactgca
taattctctt
6121 actgtcatgc catccgtaag atgcttttct gtgactgggt agtactcaac
caagtcattc
6181 tgagaatagt gtatgcggcg accgagttgc tcttgcccgg cgtcaatacg
ggataatacc
6241 gcgccacata gcagaacttt aaaagtgctc atcattggaa aacgttcttc
ggggcgaaaa
6301 ctctcaagga tcttaccgct gttgagatcc agttcgatgt aaccactcg
tgacccaac
6361 tgatcttcag catcttttac tttcaccagc gtttctgggt gagcaaaaac
aggaaggcaa
6421 aatgccgcaa aaaaggaat aagggcgaca cggaaatggt gaatactcat
actcttcctt
6481 tttcaatatt attgaagcat ttatcagggt tattgtctca tgagcggata
catattgaa
6541 tgtattttaga aaaataaaca aataggggtt ccgcgcacat ttccccgaaa
agtgccacct
6601 gacgt
//
```