

***Escherichia coli* – *Staphylococcus aureus* Shuttle Vector pCN58, Recombinant in *Escherichia coli***

**Catalog No. NR-46159**

**Product Description:** NR-46159 is a culture of *Escherichia coli* (*E. coli*) DH5α (RN9668, NRS624) containing the *E. coli*-staphylococcal shuttle vector pCN58. Vector pCN58 contains the *E. coli* ColE1 replication origin, the *Staphylococcus aureus* (*S. aureus*) pT181 *cop-wt-repC* replicon, the luciferase report gene *luxAB* and the *blaZ* transcriptional terminator. Vector pCN58 was deposited as resistant to ampicillin and erythromycin in *E. coli* and resistant to erythromycin in *S. aureus*.

**Lot<sup>1</sup>: 63043371**

**Manufacturing Date: 31OCT2014**

TEST	SPECIFICATIONS	RESULTS
<b>Phenotypic Analysis</b> Cellular morphology Colony morphology <sup>2</sup>  Motility (wet mount)	Gram-negative rods Report results  Report results	Gram-negative rods Circular, low convex, entire and cream (Figure 1) Motile
<b>Confirmation of pCN58 Sequence</b> Illumina <sup>®</sup> MiSeq <sup>®</sup> sequence (Figure 2, Table 1)	Report results	Consistent with pCN58 vector description <sup>3,4</sup>
<b>Antibiotic Resistance</b> Erythromycin (encoded by the adenine methylase gene <i>ermC</i> ) Ampicillin (encoded by the beta-lactamase gene <i>amp</i> )	<i>ermC</i> sequence present  <i>amp</i> sequence present	<i>ermC</i> sequence confirmed  <i>amp</i> sequence confirmed
<b>Antibiotic Resistance</b> Ampicillin (100 µg/mL) <sup>2</sup>	Resistant (growth)	Growth observed
<b>Purity (post-freeze)<sup>5</sup></b>	Growth consistent with <i>E. coli</i>	Growth consistent with <i>E. coli</i>
<b>Viability (post-freeze)<sup>2</sup></b>	Growth	Growth

<sup>1</sup>NR-46159 was produced by inoculation of the deposited material in Luria-Bertani (LB) broth with 100 µg/mL ampicillin and incubated for 21 hours at 37°C in an aerobic atmosphere with shaking at ~ 250 rpm. Broth inoculum was passaged once in LB broth with 100 µg/mL ampicillin for 19 hours at 37°C in an aerobic atmosphere with shaking at ~ 250 rpm to produce this lot.

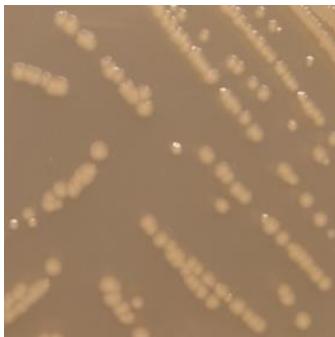
<sup>2</sup>23 hours at 37°C in an aerobic atmosphere on LB agar with 100 µg/mL ampicillin

<sup>3</sup>Illumina<sup>®</sup> MiSeq<sup>®</sup> sequence was analyzed with CLC Genomics Workbench Version 7.0.2.

<sup>4</sup>pCN58 was sequenced and annotated by BEI Resources and is consistent with the vector described in Charpentier, E., et al. "Novel Cassette-Based Shuttle Vector System for Gram-Positive Bacteria." *Appl. Environ. Microbiol.* 70 (2004): 6076-6085. PubMed: 15466553. The BEI Resources vector sequence was deposited into GenBank as NR-46159 (GenBank: KR781472).

<sup>5</sup>Purity of this lot was assessed for 7 days on Tryptic Soy agar with 5% defibrinated sheep blood at 37°C in an aerobic atmosphere

Figure 1: Colony Morphology



Date: 28 JUL 2016

Signature:

A handwritten signature in black ink, appearing to read "David C. Anderson".

BEI Resources Authentication

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC®'s knowledge.

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Figure 2: Shuttle Vector pCN58

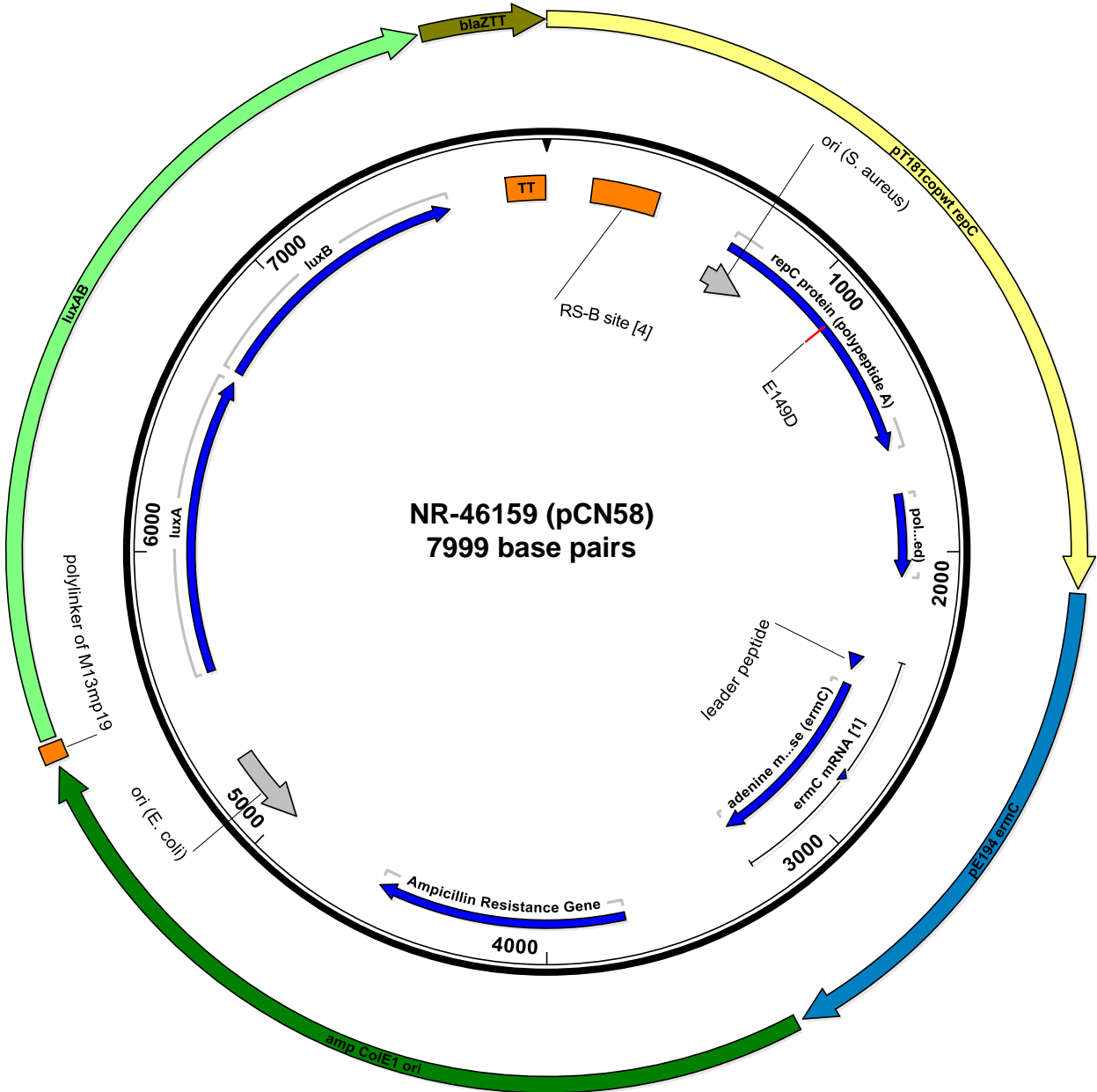


Table 1: Sequence of shuttle vector pCN58

1	CCTTTGCGGA	AAGAGTTAAT	AAGTTAACAG	AAGATGAACC	AAAACTAAAT	50
51	GGTTTAGCAG	GAAACTTAGA	TAAAAAAATG	AATCCAGAAT	TATATTCAGA	100
101	ACAGGAACAG	CAACAAGAAC	AACAAAAGAA	TCAAAAACGA	GATAGAGGTA	150
151	TGCACTTATA	GAACATGCAT	TTATGCCGAG	AAAACTTATT	GGTTGGAATG	200
201	GGCTATGTGT	TAGCTAACTT	GTTAGCGAGT	TGGTTGGACT	TGAATTGGGA	250
251	TTAATCCCAA	GAAAGTACCA	ACTCAACAAC	ACATAAAGCC	CTGTAGGTTT	300
301	CGACCAATAA	GGAAATTGGA	ATAAAGCAAT	AAAAGGAGTT	GAAGAAATGA	350
351	AATTCAGAGA	AGCCTTTGAG	AATTTTATAA	CAAGTAAGTA	TGTACTTGGT	400
401	GTTTTAGTAG	TTTTAACTGT	TTACCAGATA	ATACAAATGC	TTAAATAAAA	450
451	AAAGACTTGA	TCTGATTAGA	CCAAATCTTT	TGATAGTGTT	ATATTAATAA	500
501	CAAAATAAAA	AGGAGTCGCT	CACGCCCTGA	CCAAAGTTTG	TGAACGACAT	550
551	CATTCAAAGA	AAAAAACACT	GAGTTGTTTT	TATAATCTTG	TATATTTAGA	600
601	TATTAACCGA	TATTTAAATA	TACATCAAGA	TATATATTTG	GGTGAGCGAT	650
651	TCCTTAAACG	AAATTGAGAT	TAAGGAGTCG	ATTTTTTATG	TATAAAAACA	700
701	ATCATGCAAA	TCATTCAAAT	CATTTGGAAA	ATCACGATTT	AGACAATTTT	750
751	TCTAAAACCG	GCTACTCTAA	TAGCCGGTTG	GACGCACATA	CTGTGTGCAT	800
801	ATCTGATCCA	AAATTAAGTT	TTGATGCAAT	GACGATCGTT	GGAAATCTCA	850
851	ACCGAGACAA	CGCTCAAGCC	CTTTCTAAAT	TTATGAGTGT	AGAGCCCCAA	900
901	ATAAGACTTT	GGGATATTCT	TCAAACAAAG	TTTAAAGCTA	AAGCACTTCA	950
951	AGAAAAAGTT	TATATTGAAT	ATGACAAAGT	GAAAGCAGAT	AGTTGGGATA	1000
1001	GACGTAATAT	GCGTATTGAA	TTTAATCCAA	ACAAACTTAC	ACGAGATGAA	1050
1051	ATGATTTGGT	TAAAACAAAA	TATAATAAGC	TACATGGAAG	ATGACGGTTT	1100
1101	TACAAGATTA	GATTTAGCCT	TTGATTTTGA	AGATGATTTG	AGTGACTACT	1150
1151	ATGCAATGTC	TGATAAAGCA	GTTAAGAAAA	CTATTTTTTA	TGGTCGTAAT	1200
1201	GGTAAGCCAG	AAACAAAATA	TTTTGGCGTG	AGAGATAGTA	ATAGATTTAT	1250
1251	TAGAATTTAT	AATAAAAAAGC	AAGAACGTAA	AGATAATGCA	GATGCTGAAG	1300
1301	TTATGTCTGA	ACATTTATGG	CGTGTAGAAA	TCGAACTTAA	AAGAGATATG	1350
1351	GTGGATTACT	GGAATGATTG	CTTTAGTGAT	TTACATATCT	TGCAACCAGA	1400
1401	TTGGAAAACT	ATCCAACGCA	CTGCGGATAG	AGCAATAGTT	TTTATGTTAT	1450
1451	TGAGTGATGA	AGAAGAATGG	GGAAAGCTTC	ACAGAAATTC	TAGAACAAAA	1500
1501	TATAAGAATT	TGATAAAAAGA	AATTTTCGCCA	GTCGATTTAA	CGGACTTAAT	1550
1551	GAAATCGACT	TTAAAAGCGA	ACGAAAAACA	ATTGCAAAAA	CAAATCGATT	1600
1601	TTTGCCAACA	TGAATTTAAA	TTTTGGAAAT	AGTGTACATA	TTAATATTAC	1650
1651	TGAACAAAAA	TGATATATTT	AAACTATTCT	AATTTAGGAG	GATTTTTTTA	1700
1701	TGAAGTGTCT	ATTTAAAAAT	TTGGGGAATT	TATATGAGGT	GAAAGAATAA	1750
1751	TTTACCCCTA	TAAACTTTAG	TCACCTCAAG	TAAAGAGGTA	AAATTGTTTA	1800
1801	GTTTATATAA	AAAATTTAAA	GGTTTGTTTT	ATAGCGTTTT	ATTTTGGCTT	1850
1851	TGTATTCTTT	CATTTTTTAG	TGTATTAAT	GAAATGGTTT	TAAATGTTTC	1900
1901	TTTACCTGAT	ATTGCAAATC	ATTTTAATAC	TACTCCTGGA	ATTACAAACT	1950
1951	GGGTAAACAC	TGCATATATG	TTAACTTTTT	CGATAGGAAC	AGCAGTATAT	2000
2001	GGAAAATTAT	CTGATTATAT	AAATATAAAA	AAATTGTTAA	TTATTGGTAT	2050
2051	TAGTTTGAGC	TGCTTTGGTT	CATTGATTGC	TTTTATTGGG	CCCACCTAGG	2100
2101	AATTGAATGA	GACATGCTAC	ACCTCCGGAT	AATAAATATA	TATAAACGTA	2150
2151	TATAGATTTT	ATAAAGTCTA	ACACACTAGA	CTTATTTACT	TCGTAATTAA	2200
2201	GTCGTTAAAC	CGTGTGCTCT	ACGACCAAAA	CTATAAAAACC	TTTAAGAACT	2250
2251	TTCTTTTTTT	ACAAGAAAAA	AGAAATTAGA	TAAATCTCTC	ATATCTTTTA	2300
2301	TTCAATAATC	GCATCCGATT	GCAGTATAAA	TTTAACGATC	ACTCATCATG	2350
2351	TTCATATTTA	TCAGAGCTCG	TGCTATAAAT	ATACTAATTT	TATAAGGAGG	2400
2401	AAAAAATATG	GGCATTTTTA	GTATTTTTGT	AATCAGCACA	GTTTATTATC	2450
2451	AACCAAACAA	AAAATAAGTG	GTTATAATGA	ATCGTTAATA	AGCAAATTC	2500
2501	ATATAACCAA	ATTAAAGAGG	GTTATAATGA	ACGAGAAAAA	TATAAACAC	2550
2551	AGTCAAAACT	TTATTACTTC	AAAACATAAT	ATAGATAAAA	TAATGACAAA	2600

2601	TATAAGATTA	AATGAACATG	ATAATATCTT	TGAAATCGGC	TCAGGAAAAG	2650
2651	GCCATTTTAC	CCTTGAATTA	GTAAAGAGGT	GTAATTTTCGT	AACTGCCATT	2700
2701	GAAATAGACC	ATAAATTATG	CAAAACTACA	GAAAATAAAC	TTGTTGATCA	2750
2751	CGATAAATTC	CAAGTTTTAA	ACAAGGATAT	ATTGCAGTTT	AAATTTCCCTA	2800
2801	AAAACCAATC	CTATAAAATA	TATGGTAATA	TACCTTATAA	CATAAGTACG	2850
2851	GATATAATAC	GCAAAAATTGT	TTTTTGATAGT	ATAGCTAATG	AGATTTATTTT	2900
2901	AATCGTGGAA	TACGGGTTTG	CTAAAAGATT	ATTAAATACA	AAACGCTCAT	2950
2951	TGGCATTACT	TTTAATGGCA	GAAGTTGATA	TTTCTATATT	AAGTATGGTT	3000
3001	CCAAGAGAAT	ATTTTCATCC	TAAACCTAAA	GTGAATAGCT	CACTTATCAG	3050
3051	ATTAAGTAGA	AAAAAATCAA	GAATATCACA	CAAAGATAAA	CAAAAGTATA	3100
3101	ATTATTTTCGT	TATGAAATGG	GTTAACAAAG	AATACAAGAA	AATATTTTACA	3150
3151	AAAAATCAAT	TTAACAATTC	CTTAAAACAT	GCAGGAATTG	ACGATTTTAAA	3200
3201	CAATATTAGC	TTTGAACAAT	TCTTATCTCT	TTTCAATAGC	TATAAATTAT	3250
3251	TTAATAAGTA	AGTTAAGGGA	TGCATAAACT	GCATCCCTTA	ACTTGTTTTTT	3300
3301	CGTGTGCCCTA	TTTTTTTGTA	ATCGATTATG	TCTTTTGC	AGTCGGCTTA	3350
3351	AACCAGTTTT	CCGCGGCGCT	CGAGCGGCCG	CATAGTTAAG	CCAGCCCCGA	3400
3401	CACCCGCCAA	CACCCGCTGA	CGCGCCCTGA	CGGGCTTGTC	TGCTCCCGGC	3450
3451	ATCCGCTTAC	AGACAAGCTG	TGACCGTCTC	CGGGAGCTGC	ATGTGTCAGA	3500
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3551	CGCCTATTTT	TATAGCTTAA	TGTCATGATA	ATAATGGTTT	CTTAGACGTC	3600
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3651	TCTAAATACA	TTCAAATATG	TATCCGCTCA	TGAGACAATA	ACCCTGATAA	3700
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3851	CGAGTGGGTT	ACATCGAACT	GGATCTCAAC	AGCGGTAAGA	TCCTTGAGAG	3900
3901	TTTTTCGCC	GAAGAACGTT	TTCCAATGAT	GAGCACTTTT	AAAGTTCTGC	3950
3951	TATGTGGCGC	GGTATTATCC	CGTATTGACG	CCGGGCAAGA	GCAACTCGGT	4000
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4051	AGAAAAGCAT	CTTACGGATG	GCATGACAGT	AAGAGAATTA	TGCAGTGCTG	4100
4101	CCATAACCAT	GAGTGATAAC	ACTGCGGCCA	ACTTACTTCT	GACAACGATC	4150
4151	GGAGGACCGA	AGGAGCTAAC	CGCTTTTTTTG	CACAACATGG	GGGATCATGT	4200
4201	AACTCGCCTT	GATCGTTGGG	AACCGGAGCT	GAATGAAGCC	ATACCAAACG	4250
4251	ACGAGCGTGA	CACCACGATG	CCTGTAGCAA	TGGCAACAAC	GTTGCGCAAA	4300
4301	CTATTAAGTG	GCGAACTACT	TACTCTAGCT	TCCCGGCAAC	AATTAATAGA	4350
4351	CTGGATGGAG	GCGGATAAAG	TTGCAGGACC	ACTTCTGCGC	TCGGCCCTTC	4400
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4451	CGCGGTATCA	TTGCAGCACT	GGGCGAGAT	GGTAAGCCCT	CCCGTATCGT	4500
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4551	AGATCGCTGA	GATAGGTGCC	TCACTGATTA	AGCATTGGTA	ACTGTCAGAC	4600
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4701	CTTAACGTGA	GTTTTTCGTTT	CACTGAGCGT	CAGACCCCGT	AGAAAAGATC	4750
4751	AAAGGATCTT	CTTGAGATCC	TTTTTTTTCTG	CGCGTAATCT	GCTGCTTGCA	4800
4801	AACAAAAAAA	CCACCCTAC	CAGCGGTGGT	TTGTTTGCCG	GATCAAGAGC	4850
4851	TACCAACTCT	TTTTCCGAAG	GTAAGTGGCT	TCAGCAGAGC	GCAGATACCA	4900
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5051	TTACCGGATA	AGGCGCAGCG	GTCGGGCTGA	ACGGGGGGTT	CGTGACACACA	5100
5101	GCCCAGCTTG	GAGCGAACGA	CCTACACCGA	ACTGAGATAC	CTACAGCGTG	5150
5151	AGCTATGAGA	AAGCGCCACG	CTTCCCGAAG	GGAGAAAGGC	GGACAGGTAT	5200
5201	CCGGTAAGCG	GCAGGGTCGG	AACAGGAGAG	CGCACGAGGG	AGCTTCCAGG	5250

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5301	TTGAGCGTCG	ATTTTTGTGA	TGCTCGTCAG	GGGGCGGAG	CCTATGGAAA	5350
5351	AACGCCAGCA	ACGCGGCCTT	TTTACGGTTC	CTGGCCTTTT	GCTGGCCTTT	5400
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5451	TTACCGCCTT	TGAGTGAGCT	GGCGGCCGCT	GCATGCCTGC	AGGTCGACTC	5500
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5601	CCAGGTGAAA	CTCATAAGCA	AGTAATGGAT	CGCTTTGTTC	GGCTTGGTAT	5650
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5751	TTAGGAAGAA	CTAAAACATT	AAATGTTGGC	ACTATGGGGG	TTGTTATTCC	5800
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5851	TGTCGAAAGG	TCGTTTTAAT	TTTGGAACCG	TTTCGAGGGCT	ATACCATAAA	5900
5901	GATTTTCGAG	TATTTGGTGT	TGATATGGAA	GAGTCTCGAG	CAATTACTCA	5950
5951	AAATTTCTAC	CAGATGATAA	TGGAAAGCTT	ACAGACAGGA	ACCATTAGCT	6000
6001	CTGATAGTGA	TTACATTCAA	TTTCCTAAGG	TTGATGTATA	TCCCAAAGTG	6050
6051	TACTCAAAAA	ATGTACCAAC	CTGTATGACT	GCTGAGTCCG	CAAGTACGAC	6100
6101	AGAATGGCTA	GCAATACAAG	GGCTACCAAT	GGTTCTTAGT	TGGATTATTG	6150
6151	GTACTAATGA	AAAAAAGCA	CAGATGGAAC	TCTATAATGA	AATTGCGACA	6200
6201	GAATATGGT	ATGATATATC	TAAAATAGAT	CATTGTATGA	CTTATATTTG	6250
6251	TTCTGTTGAT	GATGATGCAC	AAAAGGCGCA	AGATGTTTGT	CGGGAGTTTC	6300
6301	TGAAAAAATTG	GTATGACTCA	TATGTAAATG	CGACCAATAT	CTTTAATGAT	6350
6351	AGCAATCAAAA	CTCGTGGTTA	TGATTATCAT	AAAGGTCAAT	GGCGTGATTT	6400
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6601	TCGCTCCTTT	CTTAAAAGAA	CCTAAATAAA	TTACTTATTT	GATACTAGAG	6650
6651	ATAATAAGGA	ACAAGTTATG	AAATTTGGAT	TATTTTTTCT	AAACTTTCAG	6700
6701	AAAGATGGAA	TAACATCTGA	AGAAACGTTG	GATAATATGG	TAAAGACTGT	6750
6751	CACGTTAATT	GATTCAACTA	AATATCATTT	TAATACTGCC	TTTGTTAATG	6800
6801	AACATCACTT	TTCAAAAAAT	GGTATTGTTG	GAGCACCTAT	TACCGCAGCT	6850
6851	GGTTTTTTTAT	TAGGGTTAAC	AAATAAATTA	CATATTGGTT	CATTAAATCA	6900
6901	AGTAATTACC	ACCCATCACC	CTGTACGTGT	AGCAGAAGAA	GCCAGTTTAT	6950
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7001	AGTGATTTTCG	AAATGGAATT	TTTTAGACGT	CATATCTCAT	CAAGGCAACA	7050
7051	ACAATTTGAA	GCATGCTATG	AAATAATTA	TGACGCATTA	ACTACAGGTT	7100
7101	ATTGTCATCC	CCAAAACGAC	TTTTATGATT	TTCCAAAGGT	TTCAATTAAT	7150
7151	CCACACTGTT	ACAGTGAGAA	TGGACCTAAG	CAATATGTAT	CCGCTACATC	7200
7201	AAAAGAAGTC	GTCATGTGGG	CAGCGAAAAA	GGCACTGCCT	TTAACATTTA	7250
7251	AGTGGGAGGA	TAATTTAGAA	ACCAAAGAAC	GCTATGCAAT	TCTATATAAT	7300
7301	AAAACAGCAC	AACAATATGG	TATTGATATT	TCGGATGTTG	ATCATCAATT	7350
7351	AACTGTAATT	GCGAACTTAA	ATGCTGATAG	AAGTACGGCT	CAAGAAGAAG	7400
7401	TGAGAGAATA	CTTAAAAGAC	TATATCACTG	AACTTACCC	TCAAATGGAC	7450
7451	AGAGATGAAA	AAATTAAGT	CATTATTGAA	GAGAATGCAG	TTGGGTCTCA	7500
7501	TGATGACTAT	TATGAATCGA	CAAAATTAGC	AGTGGAAAAA	ACAGGGTCTA	7550
7551	AAAATATTTT	ATTATCCTTT	GAATCAATGT	CCGATATTAA	AGATGTAAAA	7600
7601	GATATTATTG	ATATGTTGAA	CCAAAAATC	GAAATGAATT	TACCATAATA	7650
7651	AAATTAAGG	CAATTTCTAT	ATTAGATTGC	CTTTTTGGCG	CGCCTATTCT	7700
7701	AAATGCATAA	TAAATACTGA	TAACATCTTA	TATTTTGTAT	TATATTTTGT	7750
7751	ATTATCGTTG	ACATGTATAA	TTTTGATATC	AAAAACTGAT	TTTCCCTCTA	7800
7801	TTATTTTCGA	GATTTATTTT	CTTAATTCTC	TTTAACAAAC	TAGAAATATT	7850
7851	GTATATACAA	AAAATTATAA	ATAATAGATG	AATAGTTTAA	TTATAGGTGT	7900

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7901	TCATCAATCG	AAAAAGCAAC	GATCCTTATT	TAAAGTGCCT	TGCTTTTTTC	7950
7951	TCATTTATAA	GGTTAAATAA	TTCTCATATA	TCAAGCAAAG	TGACAGGCG	7999

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