SUPPLEMENTAL MATERIAL

TABLE. S1 Library of Primers to Create Probable Promoter Sequence Amplicons. Primer numbers, sequences, 5' modification, and melting temperature (T_m) with and without modifications are included.

Primer Number	Sequence	5' Modification	T_m without Modifications (°C)	T_m with Modifications (°C)
Primer 1 (F1)	5' TCAGTGTCTAGAGTGCCACCAAAAGAGAAGTTGAACA 3'	XbaI + 6 upstream bases (5' TCAGTGTCTAGA 3')	58-60	61-63.9
Primer 2 (R1)	5' CTGACTGAATTCTTCACACAGGAAACAGCTATGACCA 3'	EcoRI + 6 upstream bases (5' CTGACTGAATTC 3')	58.3-60	59-63.2
Primer 3 (R2)	5' GACTCAGAATTCGAGCGCAACGCAATTAATGTGAGTTA 3'	EcoRI + 6 upstream bases (5' GACTCAGAATTC 3')	58.2-60	60-63.6
Primer 4 (R3)	5' CAGTCAGAATTCCTTTTTACGGTTCCTGGCCTTTT 3'	EcoRI + 6 upstream bases (5' CAGTCTGAATTC 3')	58.1-60	60-63
Primer 5 (F2)	5' ACTGTGTCTAGAGGGTGCCTAATGAGTGAGCTAACTC 3'	XbaI + 6 upstream bases (5' ACTGTGTCTAGA 3')	58.9-60	61-64.5
Primer 6 (F3)	5' TGACTGTCTAGAGGTCATAGCTGTTTCCTGTGTGAAA 3'	XbaI + 6 upstream bases (5' TGACTGTCTAGA 3')	57.3-60	60-63.5

TABLE. S2 Amplicons Created by Primers. Amplicon numbers, primers used, amplicon length, and amplicon details are included.

Amplicon Number	**Primers Used**	**Amplicon Length (BP)**	**Amplicon Details**
1	1 and 2 (F1 and R1)	847	Contains the sequence upstream of *brkA* and downstream of the *lac* promoter
2	1 and 3 (F1 and R2)	953	Contains the sequence upstream of *brkA*, up to and including the *lac* operon
3	1 and 4 (F1 and R3)	1209	Contains the sequence upstream of *brkA* up to just before the origin of replication
4	4 and 5 (R3 and F2)	325	Contains the sequence of the CAP binding site within the *lac* operon up to just before the origin of replication (Ideally, this amplicon would not have any part of the *lac* operon within it, but it was not feasible to make without the CAP binding site due to high primer melting temperatures. We do not expect this to have a large impact on our study due to the lack of the *lac* promoter and operator within this sequence)
5	4 and 6 (R3 and F3)	410	Contains the entire *lac* operon and downstream base pairs up to just before the origin of replication

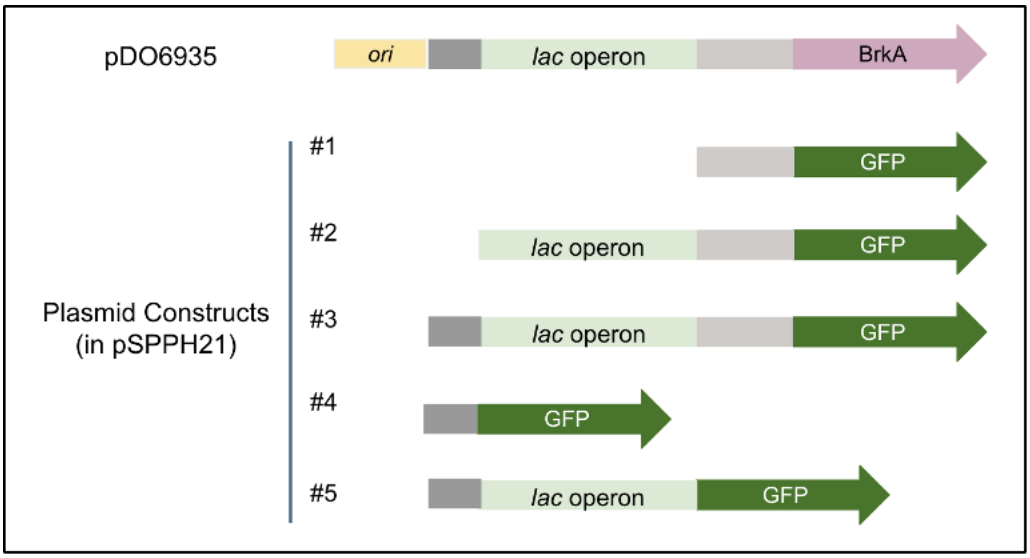

FIG. S1 Linearized Illustrations of Plasmid Constructs after Inserting Amplicons Upstream of GFP in pSPPH21. Amplicons generated using primers in Table S2 inserted various sections of the pDO6935 sequences being investigated for promoter function.

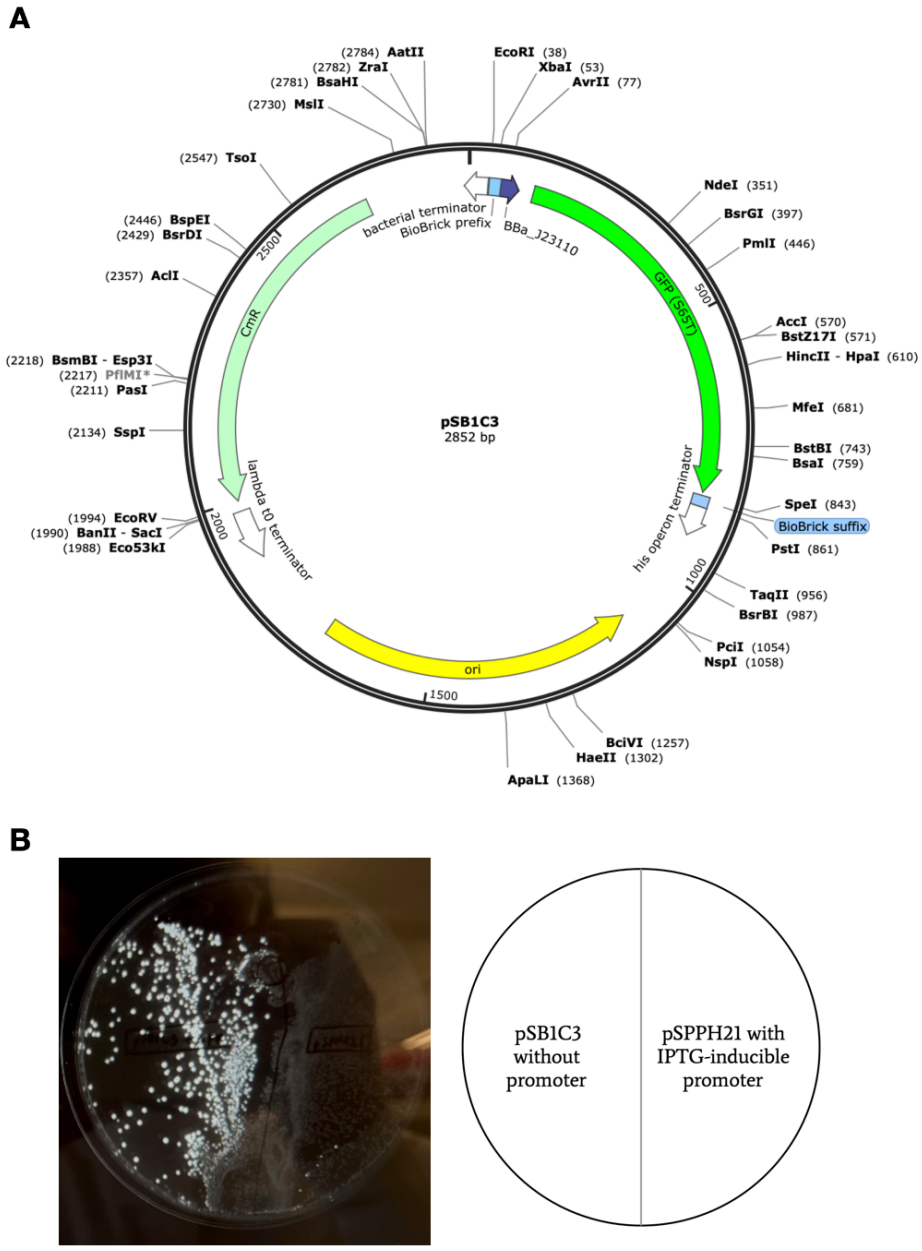

FIG. S2 The pSB1C3 vector induces GFP expression regardless of presence of *lac* promoter while pSPPH21 does not. (A) Image of pSB1C3 with sequence obtained from Plasmidsauraus. Full sequence can be found via the respective FASTA file. (B) GelDoc image showing GFP fluorescence of DH5α *E. coli* cells transformed with promoterless pSB1C3 or pSPPH21 (with IPTG-inducible promoter) and streaked on agar plates with 20 µg/mL chloramphenicol and 200 µL of 0.5 µM IPTG.

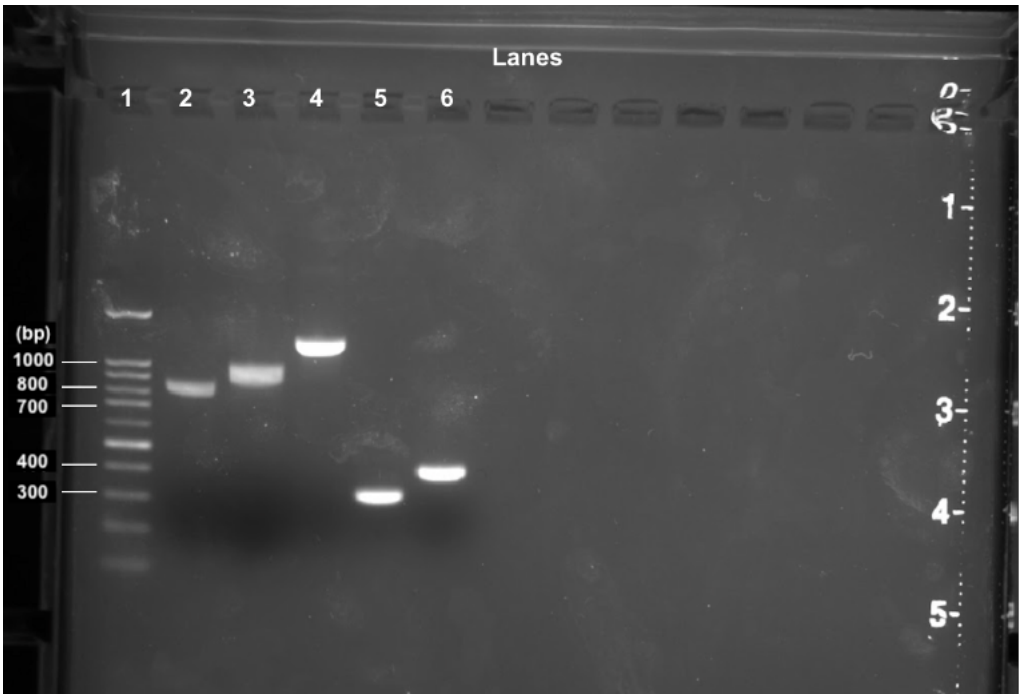

FIG. S3 Primers successfully amplified the expected amplicons. Primers were designed and PCR was used to obtain 5 amplicons. 1.5% agarose DNA gel electrophoresis was used to confirm the size of our promoter constructs. Lane 1 is the 100 bp DNA ladder. Lane 2 shows amplicon 1, which contains the region between *brkA* and the *lac* operon in pDO6935. Lane 3 shows amplicon 2, which includes all of amplicon 1, as well as the *lac* operon. Lane 4 shows amplicon 3, which contains the entire region between *brkA* and the origin of replication in pDO6935. Lane 5 shows amplicon 4, composed of the region between the origin of replication and the *lac* operon in pDO6935. Lane 6 shows amplicon 5, which contains all of amplicon 4 as well as the *lac* operon.

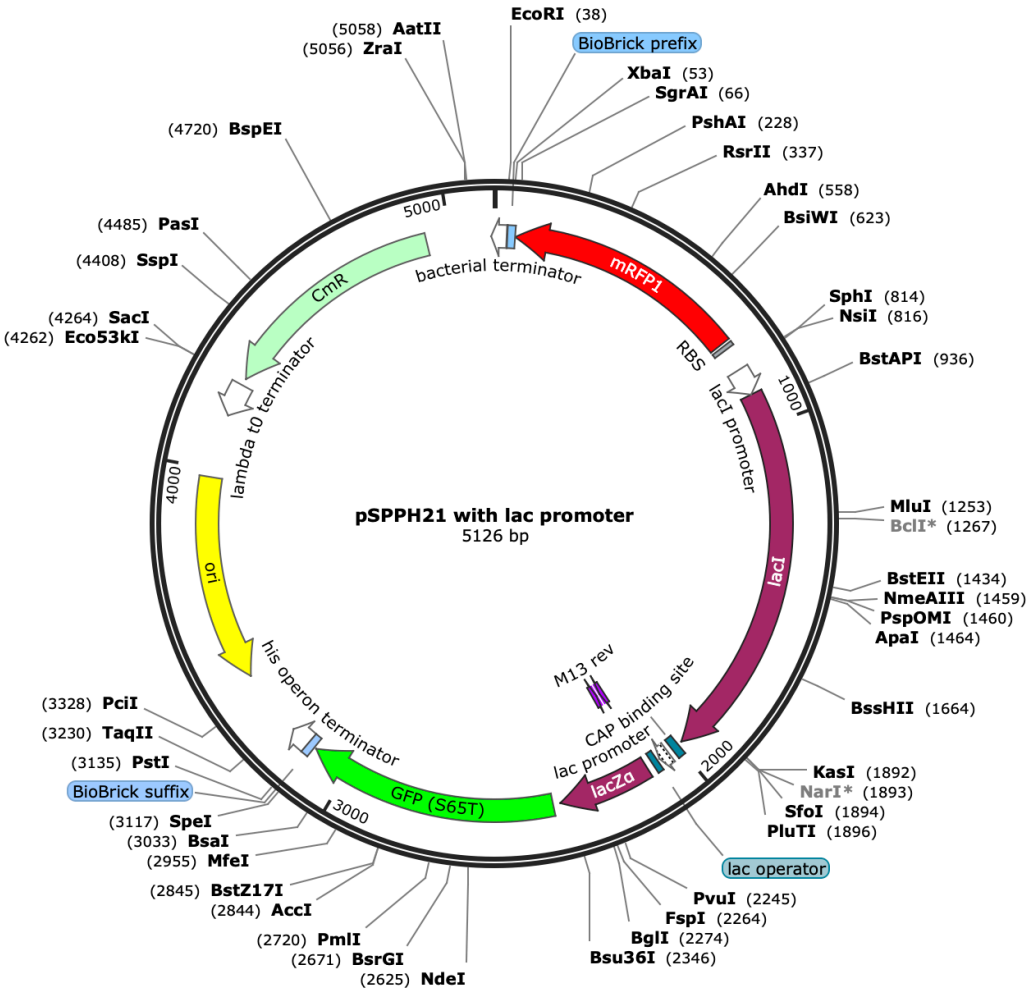

FIG. S4 Graphical Depiction of the pSPPH21 with lac promoter. Image of pSPPH21 with sequence obtained from Plasmidsauraus. Full sequence can be found via the respective FASTA file.

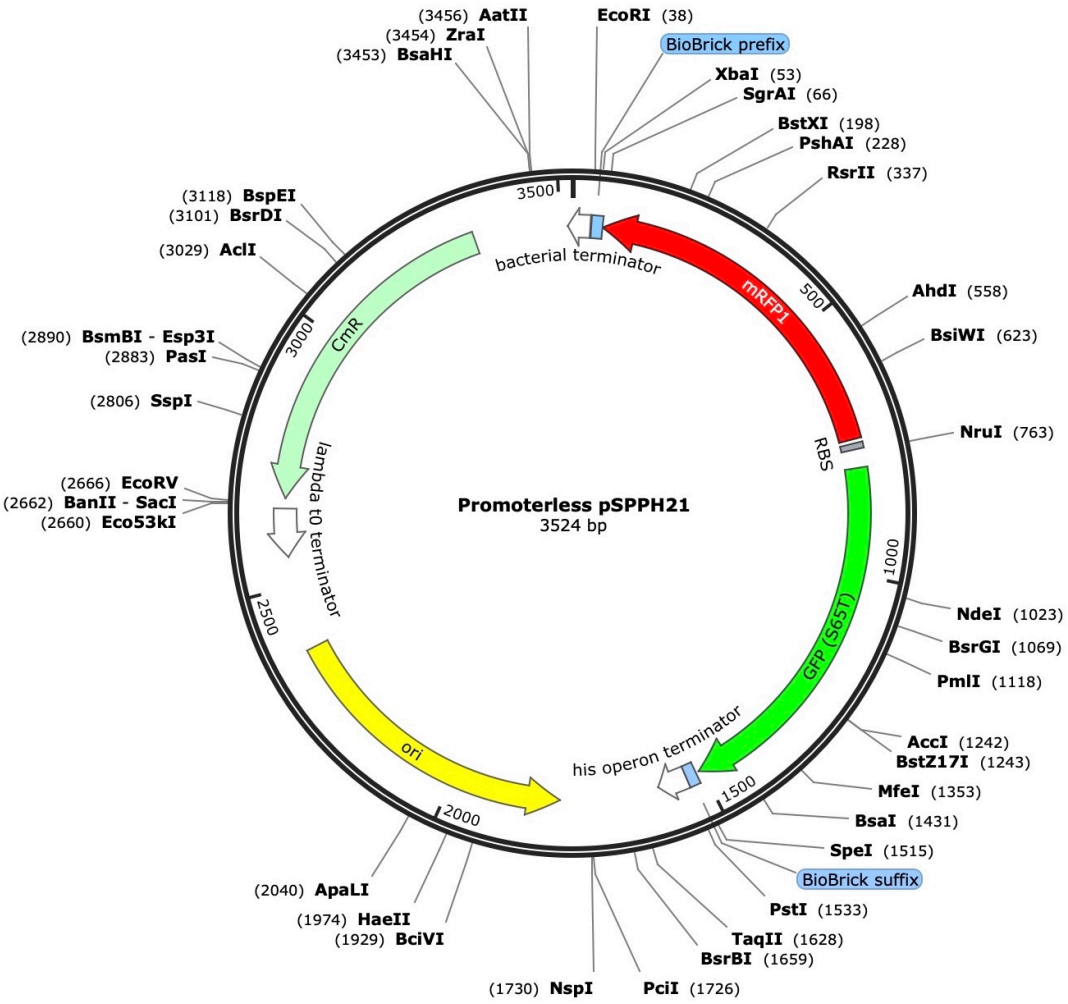

FIG. S5 Graphical Depiction of the promoterless pSPPH21. Image of promoterless pSPPH21 used as our negative control with sequence obtained from Plasmidsauraus. Full sequence can be found via the respective FASTA file.

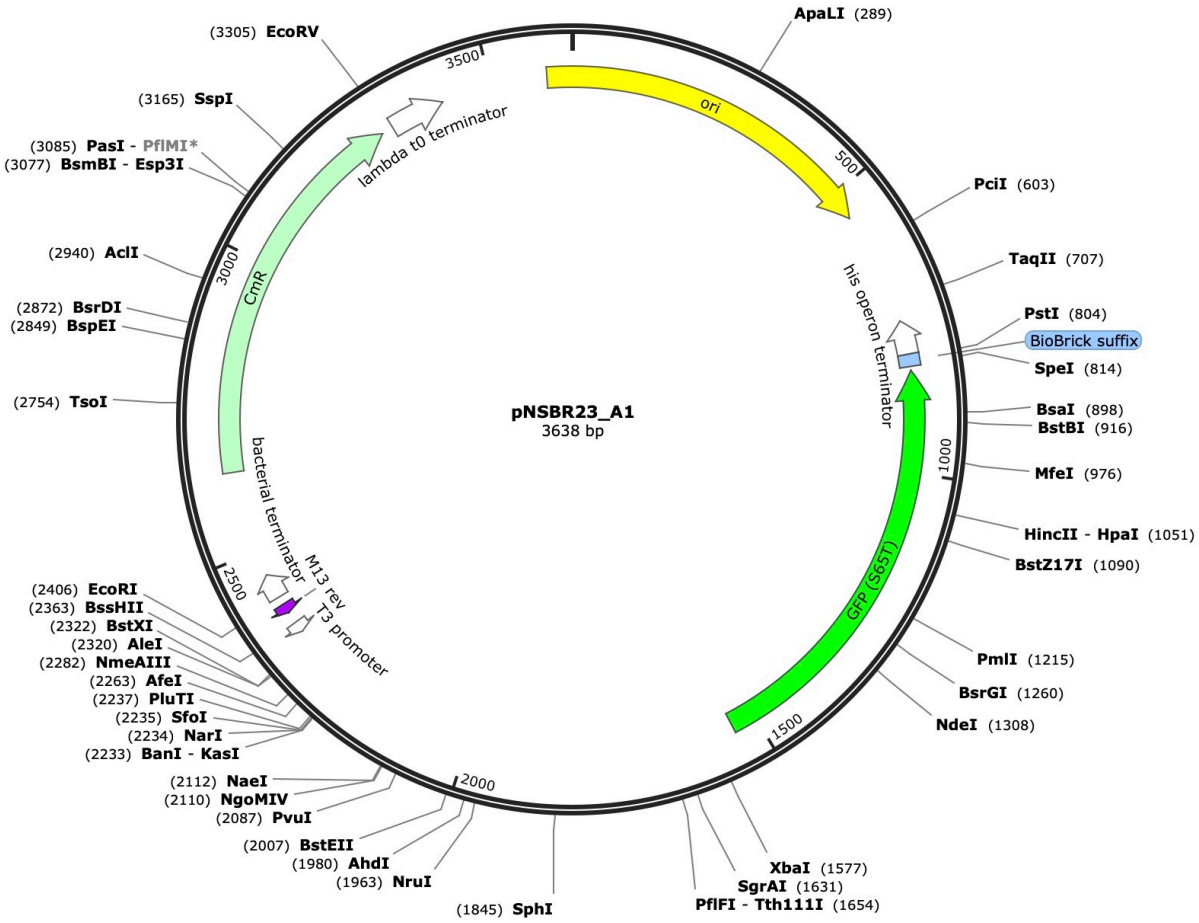

FIG. S6 Graphical Depiction of pNSBR23_A1. Image of pNSBR23_A1, obtained from Plasmidsauraus. Full sequence can be found via the respective FASTA file.

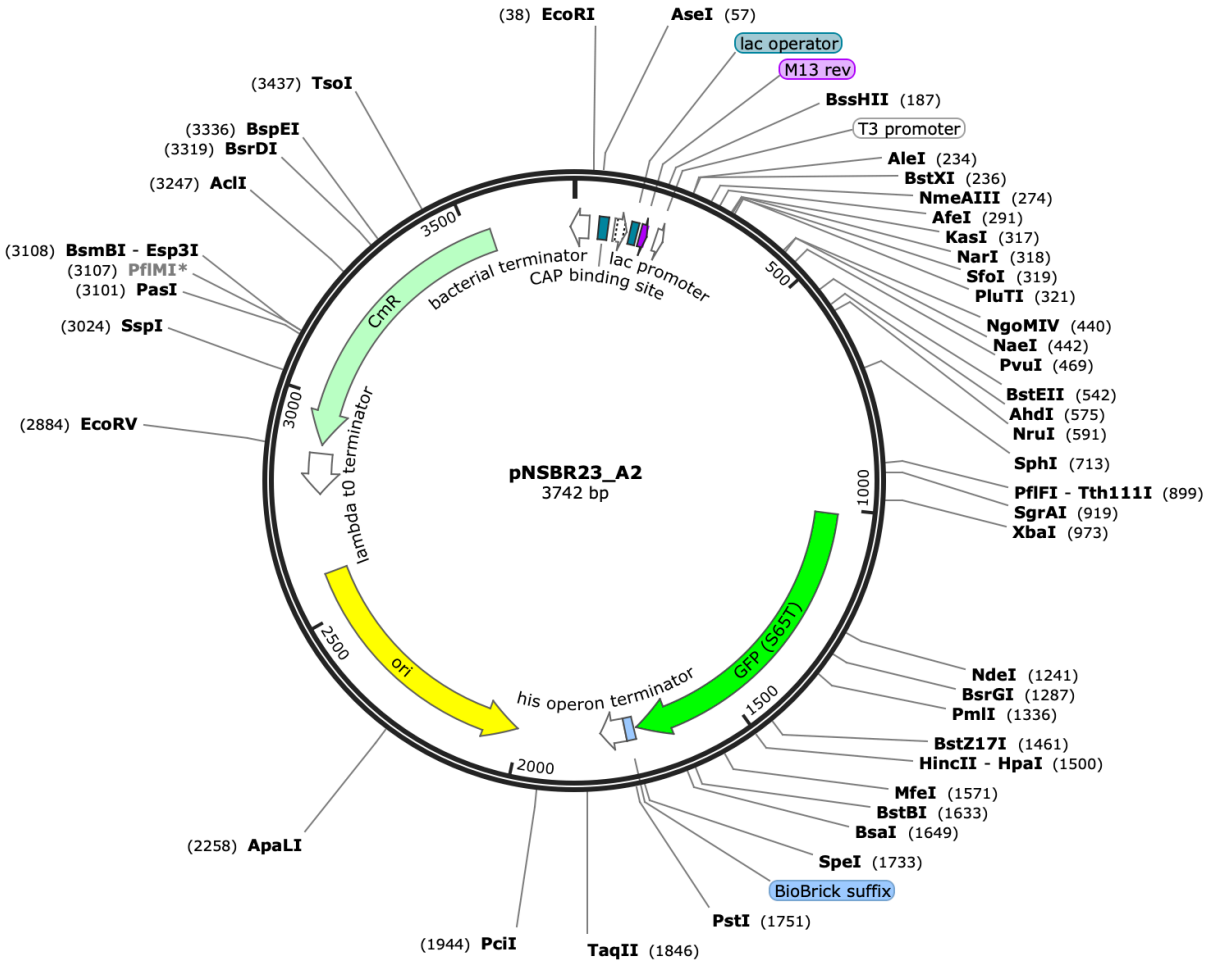

FIG. S7 Graphical Depiction of pNSBR23_A2. Image of pNSBR23_A2, obtained from Plasmidsauraus. Full sequence can be found via the respective FASTA file.

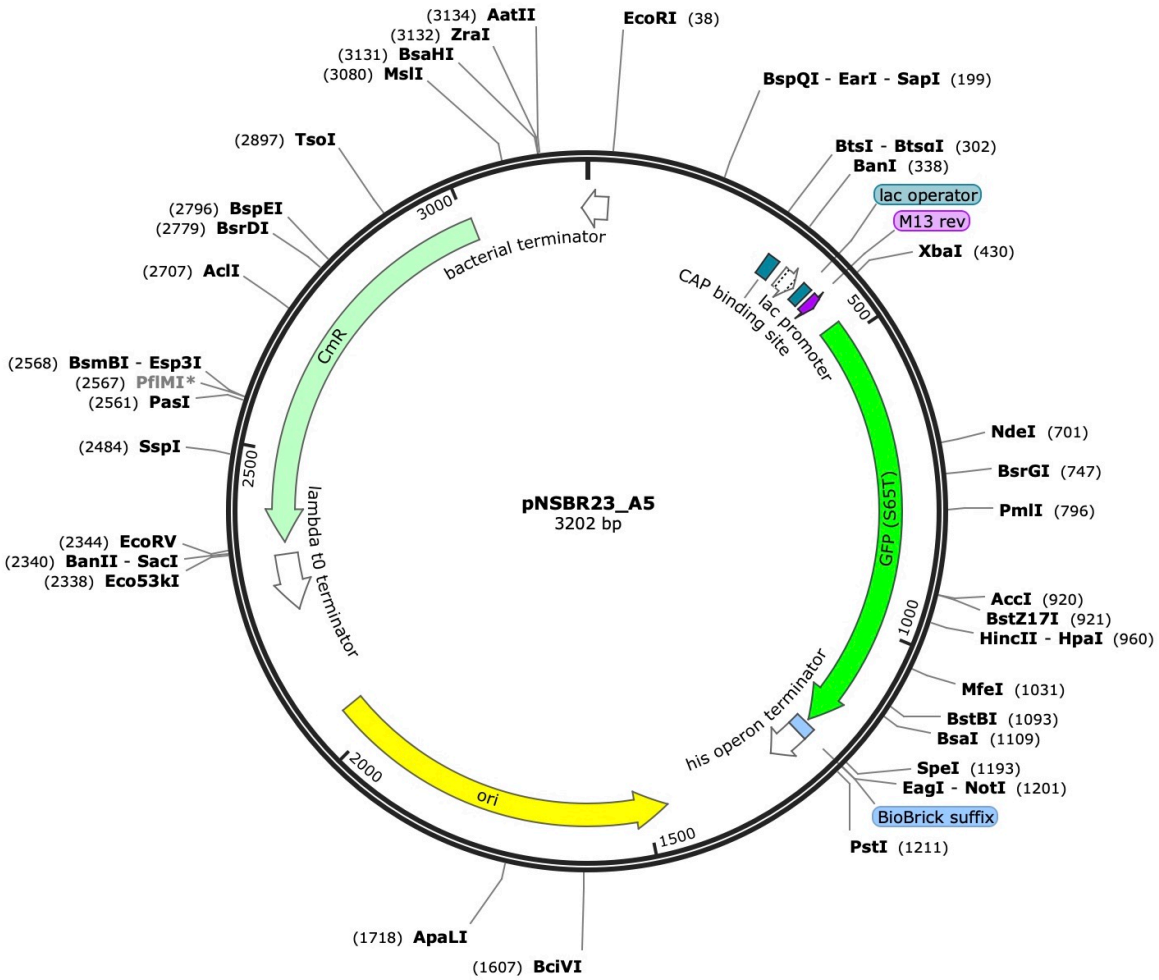

FIG. S8 Graphical Depiction of pNSBR23_A5. Image of pNSBR23_A5, obtained from Plasmidsauraus. Full sequence can be found via the respective FASTA file.

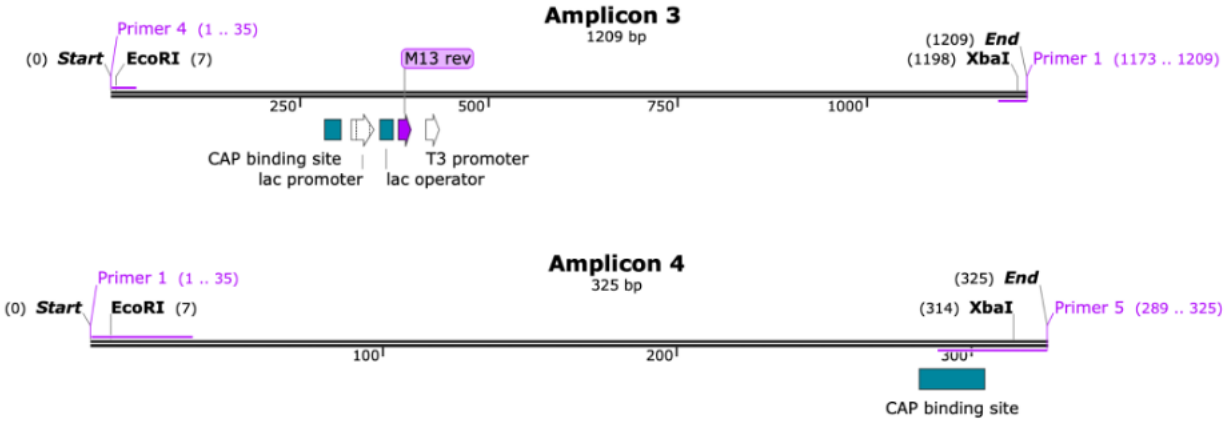

FIG. S9 Graphical Depiction of inserts for the creation of theoretical pNSBR23_A3 and pNSBR23_A4. As these amplicons were not successfully inserted there is no graphical depiction or sequence for plasmids pNSBR23_A3 and pNSBR23_A4.