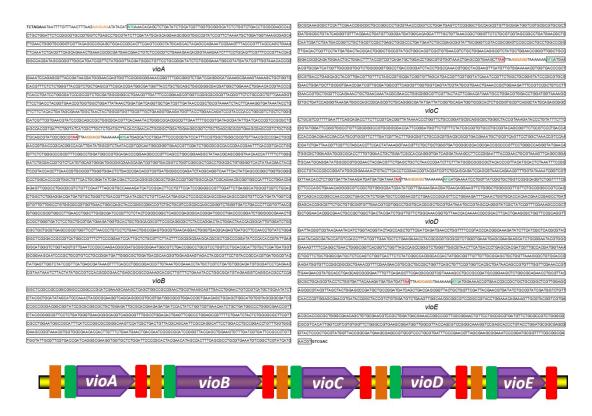
Supplementary data

Genetical control of violacein biosynthesis to enable a pigment-based

whole-cell lead biosensor

Strains, plasmids and Primers	Genotypes or description	Reference
E. coli stains		
BL21(DE3)pLysS	$F^- ompT hsdS_B (r_B^- m_B^-) gal dcm (DE3)$	Novagen
TOP10	F ⁻ Φ80 <i>lac</i> ZΔM15 Δ <i>lac</i> X74 recA1	Invitrogen
Plasmids		
pET-21a	Amp ^R , T7 promoter, lac operator	Novagen
pT-Ppbr	T vector carrying <i>pbrR</i> and Ppbr divergent promoter region	1
pET-vio	pET-21a derivative containing the violacien expression cassette (<i>vioABCDE</i>) inserted as a <i>NdeI/SacI</i> fragment	This study
pPpbr-vio	pET-vio derivative containing <i>pbrR</i> and Ppbr divergent promoter cloned as a <i>Bg/II/XbaI</i> fragment	This study
Primers		
F-pbr	GAAGATCTTTACCCAGATGTTTGA	This study
R-pbr	GCTCTAGAGAGTAACTCCTGAAAATC	This study

Table. S1 Bacterial strains and plasmids used in this study





The artificial violacein biosynthetic gene cluster spans seven kilobases and is comprised of *vioA* (1257 bp), *vioB* (3000 bp), *VioC* (1326 bp), *VioD* (1155 bp), and *vioE* (576 bp). All of the five genes are necessary for violacein biosynthesis from endogenous tryptophan in *E. coli*.

AGATCT TTACCCAGATGTTTGACTGTTCGTGGCACTTTCACCATGGCAATT GCCCAACCCTTG CAAAATGCCGCACGCCTCTACAGATCGAGAGCCAGAA CACTTCTCGCGCAAATCAACCAAGTGCCGTTTTAACTGCAACAGCGCGG ACACACGCATTTCCACCTGTTGAATATGGGCCTCCAGCAGCGTGATGACC pbrR TCCCCACAGTCCTGCATCGGGTTGTCTCGCAGACCCAGCAATGCGCGAA TCTCGCTCAACGTCATGTCGAGCGAACGGCAATGACGGATGAATTGCAA GCGCTCAATGTGCGCCTCACCGTACAACCGAAAGTTGCCACCGCTTCGC GCTGGCTTTGGCAGTAGCCCTTCCTTCTCGTAGTAGCGGATGGTCACGA CCTCGCACCCAGAGCGCTTGGCGAGGTCGCCAATTCTGATTTCCATGCA pbr bidirectional promoter region TCAATCTCCAATTATCACTTGACTCTATAGTGACTATAGAGATTTTAATGGA GGCTGAATAGAAGATTTTCAGGAGTTACTCTCTAGAAATAATTTTGTTTAA Violacein expression cassette CTTTAAGAAGGAGATATACATATG_vioA-vioB-vioC-vioD-vioE_TAAGA **GCTCCGTCGACAAGCTTGCGGCCGCACTCGAGCACCACCACCACCACC** ACTGAGATCCGGCTGCTAACAAAGCCCGAAAGGAAGCTGAGTTGGCTGC T7 terminator TGCCACCGCTGAGCAATAACTAGCATAACCCCTTGGGGCCTCTAAACGG GTCTTGAGGGGTTTTTTG

Fig. S2 The cloning/expression region of pPpbr-vio used in this study. DNA sequence and annotation data are all marked. The cassette containing the *pbrR* gene and the divergent *pbr* promoter was inserted in front of the violacein biosynthetic module.

References

1. Y. Guo, C. Y. Hui, L. Liu, H. Q. Zheng and H. M. Wu, Frontiers in microbiology, 2019, 10, 1454.