SUPPLEMENTARY MATERIAL

Table S1
Strains used in this study

Strain	*recAo*	*radA*	*recX*	Other relevant genotype	Source or derivation
CAG18642	+	+	+	*zfj-3131::Tn10*	*E. coli* Stock Center
GJ1989	+	+	+	*malE::Tn10-9 lexA3*	(11)
JC13509 [a]	+	+	+		
N3030	+	+	+	*gal-76::Tn10*	From R.G.Lloyd
SS775	+	+	+	*malE::Tn10-9 lexA3*	GJ1989→JC13509 [j]
SS996 [b]	+	+	+	*Ωgfp*	(8)
SS1153	+	+	+		pKD46→JC13509 [f]
SS1465	+	+	+	*gal-76::Tn10*	N3030→SS996 [i]
SS2642	*1403*	+	*del*	*ygaD1::kan recA4136,4155::gfp-901*	(10)
SS2647	*1403*	+	*del*	*ygaD1::kan recA4136,4155::gfp-901*	SS2642→JC13509 [h]
SS3047	*1403*	+	+	*ygaD1::kan recA4136,4155::gfp-901*	(10)
SS3085	*1403*	+	+	*ygaD1::kan recA4136,4155::gfp-901*	(9) [h]
SS3306	*1403*	+	+	*zfj-3131::Tn10 ygaD1::kan recA4136,4155::gfp-901*	CAG18642→SS3085 [i]
SS4074	+	+	+	*del(xthA)100::kan*	(1)
SS4279	+	*kan*	+		(1)
SS4414	+	+	+	*malE::Tn10-9 lexA3*	pKD46→SS775 [f]
SS4520	+	+	+	*del(xthA)100::kan*	SS4074→JC13509 [h]
SS4555	+	+	+	*del(xthA)200::frt*	This work [k]
SS4560	*1403*	+	+	*ygaD1::kan recA4136,4155::gfp-901 del(xthA)200::frt*	SS3047→SS4555 [h]
SS4857	+	+	+	*del(xthA)200::frt gal-76::Tn10 Ωgfp*	SS1465→SS4555 [i]
SS4989	+	+	+	*del(xthA)100::kan*	pKD46→SS4520 [f]
SS4959	+	+	*cat*		This work [d]
SS4999	+	+	+	*del(xthA)100::cat*	This work [g]
SS5841	*1403*	*del*	*cat*	*Ωgfp*	SS6186→SS7102 [j]
SS6013	+	+	+	*ygaD1::kan recA4142 Ωgfp*	(7)
SS6080	+	+	*cat*	*Ωgfp*	(7) [j]
SS6087	*1403*	+	+	*ygaD1::kan Ωgfp*	This work [e]
SS6088	*1403*	+	+	*ygaD1::kan Ωgfp*	SS6087→SS996 [h]
SS6156	*1403*	+	+	*ygaD1::kan recA4142 zfj-3131::Tn10 Ωgfp*	(6)
SS6186	*1403*	+	*cat*	*ygaD1::kan Ωgfp*	This work [d]
SS7102	+	*del*	+	*Ωgfp*	This work [k]
SS7108	+	*del*	+	*del(xthA)100::kan Ωgfp*	SS4074→SS7102 [h]
SS7118	+	*del*	+	*del(xthA)200::frt Ωgfp*	This work [k]
SS7128	*1403*	*del*	+	*del(xthA)200::frt Ωgfp*	SS6087→SS7118 [h]
SS7129	*1403*	*del*	*cat*	*del(xthA)200::frt Ωgfp*	SS6186→SS7118 [h]
SS7132	+	*del*	*cat*	*del(xthA)200::frt Ωgfp*	SS4959→SS7118 [j]
SS7136	*1403*	*del*	+	*Ωgfp*	SS6087→SS7102 [h]
SS7152	+	*del*	*cat*	*Ωgfp*	SS4959→SS7102 [j]
SS7155	*1403*	+	*cat*	*Ωgfp*	SS6186→SS996 [h]
SS7261	+	*del*	+	*recA4136,4155::gfp-901 zfj3131::Tn10*	SS3306→SS7255 [i]
SS8245		*op*		*srlC300::Tn10 recA730 sulB103*	This work [l]
SS8253	+	*op*	+	*Ωgfp*	SS8245→SS996 [j]
SS8254	+	*op*	+	*ygaD1::kan recA4142 Ωgfp*	SS8245→SS6013 [j]

SS8272	*1403*	*op*	+	*zfj-3131::Tn10 ygaD1::kan recA4142 Ω2gfp*	SS6156→SS8253 [j]
SS9023	+	+	+	*ygaD1::kan recA4142 Ω2gfp*	SS6013→SS996 [h]
SS9024	+	*del*	+	*ygaD1::kan recA4142 Ω2gfp*	SS6013→SS7102 [h]
SS9040	*1403*	+	+	*ygaD1::kan del(xthA)200::frt gal-76::Tn10 Ω2gfp*	SS6088→SS4857 [h]
SS9041	+	+	*cat*	*del(xthA)200::frt gal-76::Tn10 Ω2gfp*	SS6080→SS4857 [j]
SS9043	*1403*	*del*	*del*	*ygaD1::kan recA4136,4155::gfp-901*	SS2647→SS7255 [h]
SS9045	*1403*	+	*cat*	*del(xthA)200::frt gal-76::Tn10 Ω2gfp*	SS6186→SS4857 [j]
SS9048	*1403*	*del*	*del*	*ygaD1::kan recA4136,4155::gfp-901 del(xthA)100::cat*	SS4999→SS9043 [j]

[a] The genotype for this strain is *sulB103Δattλ::sulApΩ2gfp-mut2 lacMS286 ϕ80dIIlacBK1 argE3 hi-4 thi-1 xyl-5 mtl-1 rpsL31 tsx.* The *lacMS286ϕ80dIIlacBK1* code for two partial non-overlapping deletions of the *lac* operon (5, 15).

[b] This is JC13509 with *Δattλ::sulApΩ2gfp-mut2.* This is the *sulAp-gfp* reporter gene inserted into the *attλ* site (8).

[c] This is SS1153 with *ygaD1::kan recAo1403 recA4136,4155::gfp-901* at the native *recA* locus (9).

[d] PCR using prSJS748 and prSJS749 and pACYC184 as template. The linear DNA fragment was then used to transform recombinationally competent bacteria SS1153. Cat^R was selected.

[e] Restrict pSJS1483 (7) with *Bam*HI and *Rsr*II and the linear DNA fragment was then used to transformed recombinationally competent bacteria. Kan^R was selected.

[f] Select Amp^R at 30°C. pKD46 is described in (2).

[g] PCR using prSJS748 and prSJS749 and pACYC184 as template. The linear DNA fragment was then used to transform recombinationally competent bacteria SS4989. Cat^R was selected, loss of Kan^R screened.

[h] Select for Kan^R and then screen for other marker phenotypically or by PCR.

[i] Select for Tet^R and then screen for other marker phenotypically or by PCR.

[j] Select for Cam^R and then screen for other marker phenotypically or by PCR.

[k] This deletion allele was created by first transducing the Kan resistant allele from the Kieo collection into the strain as indicated in the reference column. pLH29, carrying the *flp* gene, was then introduced and Kan sensitive derivatives were screened (3).

[l] This strain is in the AB1157 background.

Table S2
Oligonucleotides used for constructing the *radAop* mutation

Name	5' – 3' sequence	Purpose
prSJS 1049	GTAGCGCGTACGGAAGTTCCTATTCTCTAGAAAGTA TAGGAACTTCGGCGAAAATGAGACGTTGATC	Forward primer with homology to cat gene on pACYC184 and adds Flip site
prSJS 1050	GTTACGCCGTACGGAAGTTCCTATACTTTCTAGAGA ATAGGAACTTCTCAGGCGTAGCACCAGGCG	Reverse primer with homology to cat gene on pACYC184 and adds Flip site
prSJS 1119	GCTACTCGACATCCTAGCTACTCAGGGAAGTTCCTAT ACTTTCTAGAGAATAGGAACTTCTCAGGCGTAGCAC CAGGCGTTTAAGG	Reverse primer with Flip site and adds an arbitrary sequence and *recAp* sequences
prSJS 1137	TAATACCGGATAGTCAATATGTTCTGTTGAAGCAATT ATACTGCATGCTCATACGGTATCAAGTGACTCGACA TCCTAGCTACTCAGG	First reverse primer PCR for *priCop* Cat^R cassette
prSJS 1143	CCTTCCAGTTTTTCCAGCAGCAGGGCGGTTTTCATTT TCTTCCTCCTTCATGCCGGGTAATACCGGATAGTCAA TATGTTCTGTTG	Second reverse primer PCR for *priCop* Cat^R cassette
prSJS 1144	CGAGCACTTAGCGTGGCGAACTGTGACACCGGGGCA CAACGCTGACGCAGCGTAGCGAGCTGTCCTTCCAGT TTTTCCAGCAGCAGG	Third reverse primer PCR for *priCop* Cat^R cassette
prSJS 1145	AACAATTATCATTTTCATTGAGGTCTTATCCGTACGG AAGTTCCTATTCTCTAGAAAGTATAGGAACTTCGGC GAAAATGAGACGTTG	First forward primer PCR for *priCop* Cat^R cassette
prSJS 1146	GATGATTAAAATGATTCGTTGCATATTGAGTGTTGTC AGCTTACAGAGTGGCTACTTTAGCATAACAATTATC ATTTTCATTGAGG	Second forward primer PCR for *priCop* Cat^R cassette
prSJS 1147	GGCGTCGTCGGTAATACCGGTAATACCACGCCCAGC GTACCCAGCACTACCGCCAGCCAGCCAATGATGATT AAAATGATTCGTTGC	Third forward primer PCR for *priCop* Cat^R cassette
prSJS 1243	GCCTGAATCAGAAGTAATTGCTCGCCCGCCATCCTG CGGGCGGCACAGCATTAACGAGGTACACCCGTACGG AAGTTCCTATTCTCTAG	Forward primer for *radAop*
prSJS 1244	CCTGCCAGCGCGGATAATCGGCCCCGCATTCATTAC AAACAAAGGCGCGTTTTGGAGCTTTTGCCATTTTCTT CCTCCTTCATGCCGGG	Reverse primer for *radAop*

Construction of the *radA* overproducer. The *radA* overproducer mutation was made from a *priC* overproducer mutation (*priCop*). The original construction for *priCop* used four sets of PCR reactions. Each one adding sequences to the *cat* gene from pACYC184. They add sequences for the *FRT* sites, the *recA* promoter (*recAo1401* and *recAo281*) and RBS sequences as well as the region of homolog before and after the *priC* start codon. The first PCR reaction used prSJS1049 and prSJS1050 (Table S1) and pACYC184 as a template to create a DNA fragment with the cat gene flanked by Flip (FRT) sites. Then using this PCR fragment just generated as the template, PCR was done using primers prSJS1049 and prSJS1119. This step added an arbitrary sequence after the FRT site that allowed for more specific amplification in subsequent steps. Then using the DNA fragment just generated as the template, PCR was done again with prSJS1145 and prSJS1137 to generate a fragment of DNA where sequences upstream of the *priC* gene were added on the upstream side of the FRT-*cat*-FRT fragment and the arbitrary sequence and part of the *recA* promoter were added on downstream side. This fragment was then used as the template in a PCR reaction with prSJS1146 and prSJS1143 to generate a fragment of DNA where more upstream *priC* sequences and the remainder of the *recA* promoter, RBS, ATG and *priC* coding sequences were added. It should be noted that the *recA* promoter had *recAo1401* and *recAo281* mutations that eliminated SOS regulation (6, 7, 8) and the RBS was modified from the wild type to one that had been optimized. This fragment was then used in a final PCR reaction with prSJS1147 and prSJS1144 that further added regions of *priC* sequences upstream and downstream of the ATP. This fragment was then recombined onto the chromosome inserting the FRT-*cat*-FRT-*recAp*-RBS in front of the *priC* start codon by standard methods. The *priC* start

codon was also changed from a GTG to an ATG in the process.

Construction of the *radA* overproducer mutation. This sequence of DNA was constructed by using prSJS1243 and prSJS1244 with the strain that contained the *priCop* construction described above as a template in a PCR reaction. This fragment was then used to transform a recipient cell expressing the *exo, bet* and *gam* proteins (2). Chloramphenicol resistant recombinants were selected. One was selected and sequenced and named SS8245 (Table S1). This construct was confirmed by DNA sequencing.

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