

Supplementary Table 1. Titers of bacteriophage T7 (PFU) during infection of *E. coli* strains with and without CbAgo.

Strain	Inoculation titer, temperature	1.5×10 ⁷ , 25°C	4×10 ¹⁰ , 25°C	1.5×10 ¹¹ , 20°C
	Time past infection	PFU/ml		
CbAgo WT	1 h	2.0×10 ⁵	4.0×10 ⁹	5.7×10 ⁹
	2 h	1.8×10 ⁶	4.6×10 ¹²	1.6×10 ¹⁰
	3 h	ND	1.6×10 ¹²	3.3×10 ¹¹
ΔCbAgo	1 h	2.1×10 ⁵	1.4×10 ⁹	3.6×10 ⁹
	2 h	5.3×10 ⁶	3.5×10 ¹²	4.7×10 ¹⁰
	3 h	ND	4.2×10 ¹²	4.0×10 ¹¹

ND, no data

Supplementary Table 2. Plasmids used in this study.

Name	Description	Source
pBAD/His B_CbAgo	ApR; derivative of the pBAD/His B vector (ThermoFisher Scientific); encodes codon-optimized N-terminally 6xHis-tagged CbAgo ⁷ under the control of <i>araBAD</i> promoter; used for pull-down assays and extraction of associated nucleic acids from CbAgo	This study
pBAD/His B_dCbAgo	ApR; encodes codon-optimized N-terminally 6xHis-tagged catalytically dead CbAgo under the control of <i>araBAD</i> promoter; used for pull-down assays and extraction of associated nucleic acids from CbAgo	This study
pSRKKm	KmR; used for construction of pChi and pNonChi plasmids	47
pChi	KmR; derivative of pSRKKm with deleted <i>lacI</i> and <i>lacZα</i> and with four tandem Chi sites inserted in the same direction at positions 1085-1116 in the resulting plasmid; used to test the effects of Chi-sites on plasmid processing	This study
pNonChi	KmR; derivative of pChi in which each T in the Chi octamers is substituted with A; used to test the effects of Chi sites on plasmid processing	This study
pSRKTc	TcR; derivative of pSRKKm; used for plasmid loss quantification	47
pSRKAmp	AmpR; derivative of pSRKKm; used for plasmid loss quantification	This study
pET28b	KmR; used for plasmid loss quantification	Novagen
RSF1010	SpR; used for plasmid loss quantification	48
pACYC184	TcR; used for plasmid loss quantification	49
pBR325	ApR, CmR, TcR; used for plasmid loss quantification	50
pKD46	ApR; used for gene deletions in <i>E. coli</i>	41

Supplementary Table 3. Bacterial Strains.

<i>E. coli</i> strain	Relevant genotype ^a	Reference
BL21(DE3)	<i>E. coli</i> B F ⁻ ompT gal dcm lon hsdS _B (r _B ⁻ m _B ⁻) λ(DE3 [lacI lacUV5-T7p07 ind1 sam7 nin5]) [malB ⁺] _{K-12} (λ ^S)	51
MG1655	K-12 F ⁻ λ ⁻ ilvG ⁻ rfb-50 rph-1	42
MG1655 Z1	MG1655 Z1(LacR TetR SpR)	42
BL21(DE3) <i>recA</i>	BL21(DE3) Δ <i>recA</i> :: <i>kan</i>	This study
BL21(DE3) <i>recBrecD</i>	BL21(DE3) Δ <i>recB</i> :: <i>kan</i> <i>recD</i> ⁻ (<i>recD</i> expression is abolished by a frameshift)	This study
BL21(DE3) <i>recC</i>	BL21(DE3) Δ <i>recC</i> :: <i>cat</i>	This study
BL21(DE3) <i>Tus</i>	BL21(DE3) Δ <i>tus</i> :: <i>kan</i>	This study
NEB TURBO	F' <i>proA</i> ⁺ <i>B</i> ⁺ <i>lacI</i> ^q Δ <i>lacZ</i> M15 / <i>fhuA2</i> Δ(<i>lac-proAB</i>) <i>glnV</i> <i>galK16</i> <i>galE15</i> <i>R(zgb-210::Tn10)</i> <i>Tet</i> ^S <i>endA1</i> <i>thi-1</i> Δ(<i>hsdS-mcrB</i>)5	New England Biolabs (catalog # C2984)
DE170	<i>E. coli</i> NEB TURBO Z1::CbAgo- <i>cat</i>	This study
DL2917	BW27784 <i>lacZ</i> :: <i>I</i> -Scelcs <i>araB</i> ::PBAD- <i>I</i> -Scel <i>lacIq</i> <i>lacZ</i> _χ ⁻	29
DL2988	BW27784 <i>araB</i> ::PBAD- <i>I</i> -Scel <i>lacIq</i> <i>lacZ</i> _χ ⁻ <i>lacZ</i> ⁺	29
DL1777	MG1655 <i>lacIq</i> <i>lacZ</i> _χ ⁻ <i>fnr-267</i> (Δ <i>ynaJ</i> Δ <i>ydaA</i> Δ <i>frr</i> Δ <i>ogt</i> Δ <i>abgT</i> Δ <i>abgB</i> Δ <i>abgA</i> Δ <i>abgR</i> Δ <i>ydaL</i> Δ <i>ydaM</i> Δ <i>dboA</i> Δ <i>ydaO</i>)	30
DL2859	MG1655 <i>lacZ</i> ::246 <i>pal</i> <i>cynX</i> ::GmR <i>lacIq</i> <i>lacZ</i> _χ ⁻ <i>lacZ</i> (L8) <i>fnr-267</i> (Δ <i>ynaJ</i> Δ <i>ydaA</i> Δ <i>frr</i> Δ <i>ogt</i> Δ <i>abgT</i> Δ <i>abgB</i> Δ <i>abgA</i> Δ <i>abgR</i> Δ <i>ydaL</i> Δ <i>ydaM</i> Δ <i>dboA</i> Δ <i>ydaO</i>)	30
DL4977	BW27784 <i>lacZ</i> :: <i>I</i> -Scelcsmut1 <i>araB</i> ::PBAD- <i>I</i> -Scel <i>lacIq</i> <i>lacZ</i> _χ	D. Leach lab
DE157	DL1777 Z1::CbAgo- <i>cat</i>	This study
DE158	DL2859 Z1::CbAgo- <i>cat</i>	This study
DE163	DL2859 Z1::dCbAgo- <i>cat</i>	This study
DE159	DL2917 Z1::CbAgo- <i>cat</i>	This study
DE164	DL2917 Z1::dCbAgo- <i>cat</i>	This study
DE161	DL4977 Z1::CbAgo- <i>cat</i>	This study
DE178	MG1655 Z1::CbAgo- <i>cat</i>	This study
DE179	MG1655 Z1::dCbAgo- <i>cat</i>	This study
DE182	MG1655 Z1::noAgo- <i>cat</i>	This study

^a only the relevant genotype is shown in the case of mutant derivatives; *cat*, *kan* – kanamycin and chloramphenicol resistance cassettes

Supplementary Table 5. SmdNA and genomic DNA libraries.

Lybrary type	Strain	CbAgo, location	Plasmid	Growth Conditions	Growth Phase	No of replicas
Small DNAs (GSE148595)	<i>E. coli</i> BL21(DE3)	CbAgo, plasmid	pBAD HisB CbAgo	LB+Glc+Amp, L-Ara induction, 37°C -> 30°C	3h after induction	2
	<i>E. coli</i> BL21(DE3)	dCbAgo, plasmid	pBAD HisB dCbAgo	LB+Glc+Amp, L-Ara induction, 37°C -> 30°C	3h after induction	1
	<i>E. coli</i> BL21(DE3) <i>recA</i>	CbAgo, plasmid	pBAD HisB CbAgo	LB+Glc+Amp+Km, L-Ara induction, 37°C -> 30°C	3h after induction	2
	<i>E. coli</i> BL21(DE3) <i>recBrecD</i>	CbAgo, plasmid	pBAD HisB CbAgo	LB+Glc+Amp+Km, L-Ara induction, 37°C -> 30°C	3h after induction	2
	<i>E. coli</i> BL21(DE3) <i>recC</i>	CbAgo, plasmid	pBAD HisB CbAgo	LB+Glc+Amp+Cm, L-Ara induction, 37°C -> 30°C	3h after induction	2
	<i>E. coli</i> BL21(DE3) <i>tus</i>	CbAgo, plasmid	pBAD HisB CbAgo	LB+Glc+Amp+Km, L-Ara induction, 37°C -> 30°C	3h after induction	2
	<i>E. coli</i> BL21(DE3)	CbAgo, plasmid	pBAD HisB CbAgo + pChi	LB+Glc+Amp+Km (sublethal doses), L-Ara induction, 37°C -> 30°C	3h after induction	2
	<i>E. coli</i> BL21(DE3)	CbAgo, plasmid	pBAD HisB CbAgo + pNonChi	LB+Glc+Amp+Km (sublethal doses), L-Ara induction, 37°C -> 30°C	3h after induction	1
	DE158	CbAgo, genome	NA	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	2
	DE157	CbAgo, genome	NA	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	2
	DE170	CbAgo, genome	Phage M13	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	1
	DE178	CbAgo, genome	pET28b	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	1
	DE178	CbAgo, genome	NA	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	1
	DE159	CbAgo, genome	NA	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	2
	DE161	CbAgo, genome	NA	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	2
	DE164	dCbAgo, genome	NA	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	1
DE163	dCbAgo, genome	NA	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	1	
Genomic DNAs (GSE148594)	DL2859	without CbAgo	NA	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	1
	DE157	CbAgo, genome	NA	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	2
	DE158	CbAgo, genome	NA	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	2
	DE163	CbAgo, genome	NA	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	1
	DL2917	without CbAgo	NA	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	1
	DE159	CbAgo, genome	NA	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	2
	DE164	CbAgo, genome	NA	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	1
	DE161	CbAgo, genome	NA	LB+Glc+Cm, aTc induction, 18°C	OD600=1.0	2
	<i>E. coli</i> BL21(DE3)	Without CbAgo	NA	LB+Glc+Amp, L-Ara induction, 37°C -> 30°C	OD600=0.5	1

<i>E. coli</i> BL21(DE3)	CbAgo, plasmid	pBAD HisB CbAgo	LB+Glc+Amp, L-Ara induction, 37°C -> 30°C	OD600=0.5	1
<i>E. coli</i> BL21(DE3) <i>tus</i>	Without CbAgo	NA	LB+Glc+Amp, L-Ara induction, 37°C -> 30°C	OD600=0.5	1
<i>E. coli</i> BL21(DE3) <i>tus</i>	CbAgo, plasmid	pBAD HisB CbAgo	LB+Glc+Amp, L-Ara induction, 37°C -> 30°C	OD600=0.5	1
<i>E. coli</i> BL21(DE3)	Without CbAgo	NA	LB+Glc+Amp, L-Ara induction, 37°C -> 30°C	OD600=6.0	1
<i>E. coli</i> BL21(DE3)	CbAgo, plasmid	pBAD HisB CbAgo	LB+Glc+Amp, L-Ara induction, 37°C -> 30°C	OD600=6.0	1
<i>E. coli</i> BL21(DE3) <i>tus</i>	Without CbAgo	NA	LB+Glc+Amp, L-Ara induction, 37°C -> 30°C	OD600=6.0	1
<i>E. coli</i> BL21(DE3) <i>tus</i>	CbAgo, plasmid	pBAD HisB CbAgo	LB+Glc+Amp, L-Ara induction, 37°C -> 30°C	OD600=6.0	1

Glc, Glucose
Amp, Ampicillin
Km, Kanamycin
Cm, Chloramphenicol
L-Ara, L-Arabinose
aTc, anhydrotetracycline