SUPPORTING INFORMATION FOR

The *Yersinia pestis* Yersiniabactin Siderophore and the ZnuABC system both contribute to Zinc acquisition and the development of lethal septicemic plague in mice

By

Alexander G. Bobrov[a], Olga Kirillina[a], Jacqueline D. Fetherston[a], M. Clarke Miller[b], Joseph A. Burlison[b], and Robert D. Perry[a‡]

[a] Department of Microbiology, Immunology, and Molecular Genetics, University of Kentucky, Lexington, KY

[b] James Graham Brown Cancer Center, University of Louisville, Louisville, KY

[‡] For correspondence. E-mail rperry@uky.edu; Tel (+1) 859 323-6341; Fax (+1) 859 257 8994

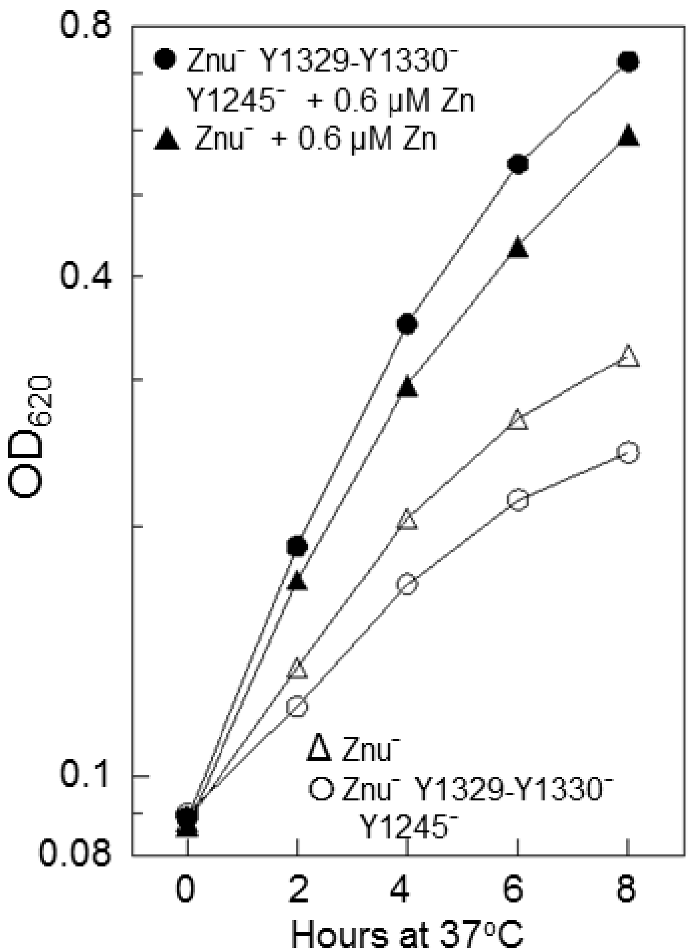

FIG. S1. The *Y. pestis* ZevAB orthologues (Y1329-Y1330) and Y1245 do not contribute significantly to *in vitro* Zn^{2+} acquisition by a *znuBC* mutant. Cells were grown at 37°C in Chelex-100-treated PMH2 with no additions (open symbols) or with 0.6 µM $ZnCl_2$ supplementation (closed symbols). Strains: KIM6-2077+ (Znu⁻ [*ΔznuBC*]) and KIM6-2077.17+ (Znu⁻ Y1329-Y1330⁻ Y1245⁻ [*ΔznuBC Δy1329-y1330::kan Δy1245::cam*]).

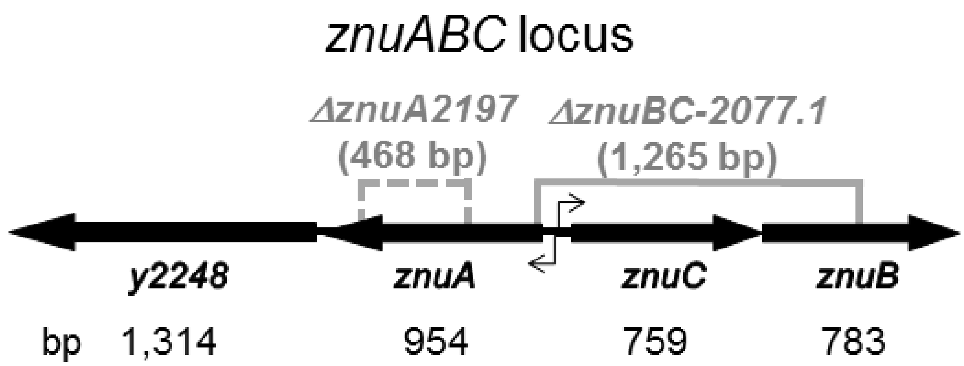

Fig. S2. Genetic organization of the *Y. pestis znuABC* locus. ORF arrows show the direction of transcription of the indicated genes and are drawn to scale. Small arrows indicate the divergent promoters in the 75 bp space between *znuA* and *znuC.* Locations and sizes of the *ΔznuBC2077.1* and *ΔznuA2197* mutations are shown.

Table S1. Bacterial strains, plasmids and primers used in this study.

Strain[a]	Relevant characteristics[b]	Reference or source
E. coli strains		
DH5α	Cloning strain	(Ausubel *et al.*, 1987)
DH5α λpir	Cloning strain for propagating plasmids with R6K origins; derived from DH5α	S.C. Straley
Y. pestis strains		
KIM5 (pCD1Ap)+	Ap^r Pgm$^+$ Lcr$^+$; pMT1, pCD1Ap (*yadA::bla*), pPCP1	(Gong *et al.*, 2001)
KIM5 (pCD1Ap)	Ap^r Pgm$^-$ (*Δpgm*; Ybt$^-$ Hms$^-$) Lcr$^+$; pMT1, pCD1Ap (*yadA::bla*), pPCP1	(Fetherston *et al.*, 2010)
KIM5-2045.6 (pCD1Ap)	Km^r Hms$^+$ Ybt$^-$ (*Δpsn::kan2045.6*) Lcr$^-$; pMT1, pPCP1	(Fetherston *et al.*, 2010)
KIM5-2046.1 (pCD1Ap)	Km^r Ybt$^-$ (*irp2::kan2046.1*) Hms$^+$ Lcr$^-$; pMT1, pPCP1	(Fetherston *et al.*, 2010)
KIM5-2077 (pCD1Ap)+	Ap^r Pgm$^+$ Znu$^-$ (*ΔznuBC2077*) Lcr$^+$; pMT1, pCD1Ap (*yadA::bla*),pPCP1; pCD1Ap electroporated into KIM6-2077+	(Desrosiers *et al.*, 2010)
KIM5-2077 (pCD1Ap)	Ap^r Pgm$^-$ (*Δpgm*; Ybt$^-$ Hms$^-$) Znu$^-$ (*ΔznuBC2077*) Lcr$^+$; pMT1, pCD1Ap (*yadA::bla*), pPCP1; pCD1Ap electroporated into KIM6-2077	This study
KIM5-2077.7 (pCD1Ap)	Ap^r Km^r Hms$^+$ Znu$^-$ (*ΔznuBC2077*) Ybt$^-$ (*irp2::kan2046.1*) Lcr$^+$; pMT1, pCD1Ap (*yadA::bla*), pPCP1; pCD1Ap electroporated into KIM6-2077.7	This study
KIM5-2077.8 (pCD1Ap)	Ap^r Hms$^+$ Ybt$^-$ (in-frame *Δirp2-2046.3*) Znu$^-$	This study

	(*ΔznuBC2077*) Lcr^+; pMT1, pCD1Ap (*yadA::bla*), pPCP1; pCD1Ap electroporated into KIM6-2077.8	
KIM5-2077.9 (pCD1Ap)	Km^r Ap^r Hms^+ Ybt^- (*Δpsn::kan2045.6*) Znu^- (*ΔznuBC2077*) Lcr^+; pMT1, pCD1Ap (*yadA::bla*),pPCP1; pCD1Ap electroporated into KIM6-2077.9	This study
KIM5-2077.10 (pCD1Ap)	Km^r Ap^r Pgm^- (*Δpgm*; Ybt^- Hms^-) Znu^+ (*ΔznuBC2077/ att*Tn*7::*mini-Tn7T-Km-znuABC$^+$) Lcr^+; pMT1, pCD1Ap (*yadA::bla*),pPCP1; pCD1Ap electroporated into KIM6-2077.10	This study
KIM5-2197 (pCD1Ap)+	Ap^r Pgm^+ Znu^- (in frame *ΔznuA2197*) Lcr^+; pMT1, pCD1Ap (*yadA::bla*), pPCP1; pCD1Ap electroporated into KIM6+	This study
KIM5-2197 (pCD1Ap)	Ap^r Pgm^- (*Δpgm*; Ybt^- Hms^-) $ZnuA^-$ (in frame *ΔznuA2197*) Lcr^+; pMT1, pCD1Ap (*yadA::bla*), pPCP1; *ΔznuA2197* was introduced into KIM5(pCD1Ap)	This study
KIM5-2197.1 (pCD1Ap)	Hms^+ $ZnuA^-$ (in frame *ΔznuA2197*) Ybt^-(in frame *Δirp2-2046.3*) Lcr^+; pMT1, pCD1Ap (*yadA::bla*), pPCP1; pCD1Ap electroporated into KIM6-2197.1	This study
KIM6+	Pgm^+ Lcr^-; pMT1, pPCP1	(Fetherston *et al.*, 1992)
KIM6	Pgm^- (*Δpgm*; Ybt^- Hms^-) Lcr^-; pMT1, pPCP1	(Fetherston *et al.*, 1992)
KIM6-2045.1	Hms^+ Ybt^- (*Δpsn2045.1*) Lcr^-; pMT1, pPCP1	(Fetherston *et al.*, 1995)
KIM6-2045.6	Km^r Hms^+ Ybt^- (*Δpsn::kan2045.6*) Lcr^-; pMT1, pPCP1	(Fetherston *et al.*, 1996)
KIM6-2046.1	Km^r Ybt^- (*irp2::kan2046.1*) Hms^+ Lcr^-; pMT1, pPCP1	(Fetherston *et al.*, 1995)
KIM6-2046.3	Hms^+ Ybt^- (in-frame *Δirp2-2046.3*) Lcr^-; pMT1, pPCP1	(Bearden *et al.*, 1997)
KIM6-2066	Hms^+ Ybt^- (*ΔybtQX2066*) Lcr^-; pMT1, pPCP1	(Fetherston *et al.*, 1999)
KIM6-2067	Hms^+ $YbtX^-$ (in-frame *ΔybtX2067*) Lcr^-; pMT1, pPCP1	Fetherston *et al.*, 1999)
KIM6-2071	Hms^+ Ybt^- (in frame *ΔybtU2071*) Lcr^-; pMT1, pPCP1	(Geoffroy *et al.*, 2000)
KIM6-2073+	Km^r Pgm^+ $TonB^-$ (*tonB::kan2073*) Lcr^-; pMT1, pPCP1	(Perry *et al.*, 2003b)
KIM6-2077+	Pgm^+ Znu^- (*ΔznuBC2077*) Lcr^-; pMT1, pPCP1	(Hazlett *et al.*, 2003)
KIM6-2077	Pgm^- (*Δpgm*; Ybt^- Hms^-) Znu^- (*ΔznuBC2077*) Lcr^-; pMT1, pPCP	(Desrosiers *et al.*, 2010)
KIM6-2077.7	Km^r Hms^+ Ybt^- (*irp2::kan2046.1*) Znu^- (*ΔznuBC2077*) Lcr^-; pMT1, pPCP1; derived from KIM6-2077+	This study

KIM6-2077.8	Hms+ Ybt- (in-frame *Δirp2-2046.3*) Znu- (*ΔznuBC2077*) Lcr-; pMT1, pPCP1; derived from KIM6-2046.3	This study
KIM6-2077.9	Km^r Hms+ Ybt- (*Δpsn::kan2045.6*) Znu- (*ΔznuBC2077*) Lcr-; pMT1, pPCP1; derived from KIM6-2045.6	This study
KIM6-2077.10+	Km^r Pgm+ Znu+ (*ΔznuBC2077/ att*Tn*7::*mini-Tn7T-Km-znuABC+) Lcr-; pMT1, pPCP1; derived from KIM6-2077+	This study
KIM6-2077.10	Km^r Pgm- (*Δpgm*; Ybt- Hms-) Znu+ (*ΔznuBC2077/ att*Tn*7::* mini-Tn7T-Km-*znuABC*+) Lcr-; pMT1, pPCP1; derived from KIM6-2077	This study
KIM6-2077.11+	Km^r Pgm+ Znu- (*ΔznuBC2077*) TonB- (*tonB::kan2073*) Lcr-; pMT1, pPCP1; derived from KIM6-2073+	This study
KIM6-2077.12+	Km^r Pgm+ Znu- (*ΔznuBC2077*) TonB- (*tonB::kan2073*) HasB- (*ΔhasB2080*) Lcr-; pMT1, pPCP1; derived from KIM6-2077.11+	This study
KIM6-2077.13	Hms+ Ybt- (*ΔybtQX-2066*) Znu- (*ΔznuBC2077*) Lcr-; pMT1, pPCP1; derived from KIM6-2066	This study
KIM6-2077.14	Hms+ Znu- (*ΔznuBC2077*) Psn- (in-frame *Δpsn2045.1*) Lcr-; pMT1, pPCP1; derived from KIM6-2077+	This study
KIM6-2077.15	Km^r Hms+ Ybt- (in-frame *Δirp2-2046.3*) Znu+ (*ΔznuBC2077/ att*Tn*7::*mini-Tn7T-Km-znuABC+) Lcr-; pMT1, pPCP1; derived from KIM6-2077.8	This study
KIM6-2077.16+	Km^r Hms+ Znu- (*ΔznuBC2077*) *Δy1329-30::kan2199* Lcr-; pMT1, pPCP1; derived from KIM6-2077(pWL204)+	This study
KIM6-2077.17+	Km^r Cm^r Hms+ Znu- (*ΔznuBC2077*) *Δy1329-30::kan2199 Δy1245::cat2187. 1* Lcr-; pMT1, pPCP1; derived from KIM6-2077.16(pWL204)+	This study
KIM6-2077.18	Km^r Hms+ Znu- (*ΔznuBC2077*) Psn- (in-frame *Δpsn2045.1*) HMWP2- (*irp2::kan2046.1*) Lcr-; pMT1, pPCP1; derived from KIM6-2077.14	This study
KIM6-2077.19	Km^r Hms+ Znu- (*ΔznuBC2077*) Psn- (in-frame *Δpsn2045.1*) HMWP2- (*irp2::kan2046.1*) YbtX- (in-frame *ΔybtX2067*) Lcr-; pMT1, pPCP1; derived from KIM6-2077.18	This study
KIM6-2197+	Pgm+ ZnuA- (in frame *ΔznuA2197*) Lcr-; pMT1, pPCP1; derived from KIM6+	This study
KIM6-2197.1	Hms+ Znu- (in frame *ΔznuA2197*) Ybt- (in frame *Δirp2-2046.3*) Lcr-; pMT1, pPCP1; derived from KIM6-2046.3	This study
KIM6-2197.2	Hms+ Znu- (in frame *ΔznuA2197*) YbtX- (in frame *ΔybtX2067*) Lcr-; pMT1, pPCP1; derived from KIM6-2067	This study

Plasmids		
pACYC184	4.24 kb, Cm^r , Tc^r, low copy cloning vector	(Ausubel *et al.*, 1987)
pBGL2	4.8 kb, Ap^r, Tc^r; low copy cloning vector	(Perry *et al.*, 1990)
pBluescript-KS	3.0 kb, Ap^r; cloning vector	Stratagene
pBSZnuA	4.9 kb, Ap^r, *znuA*$^+$; 1.9 kb *Hind*II/*Pst*I fragment from pZnu2 ligated into the same sites in pBluescript-KS	This study
pBSΔZnuA	4.4 kb, Ap^r, in frame *ΔznuA2197*; *Hind*II/*Xmn*I and *Pst*I/*Fsp*I fragments from pBSznuA ligated into *Hind*I and *Pst*I sites of pBluescript-KS	This study
pCD1Ap	71.7 kb, Ap^r, Lcr^+; pCD1 with *bla* cassette inserted into *'yadA* downstream of the frameshift mutation in this pseudogene (*'yadA::bla*)	(Gong *et al.*, 2001)
pCIRP498.8	17.4 kb, Ap^r, Km^r, R6K ori, $SacB^+$, *irp2::kan2046.1*	(Fetherston *et al.*, 1995)
pCVDYbtX	6.3 kb, Ap^r, R6K ori, $SacB^+$, in frame *ΔybtX2067*	(Fetherston *et al.*, 1999)
pEUIrp2	15.4 kb, Spc^r, *irp2::lacZ* (β-gal$^+$);	(Perry *et al.*, 2003a)
pEUZnu1	15.5 kb, Spc^r, *znuA::lacZ* (β-gal$^+$); 255-bp PCR amplicon ligated into *Asp*718 of pEU730	(Desrosiers *et al.*, 2010)
pHas98	16.1 kb, Km^r, *hasRADEB*$^+$	(Rossi *et al.*, 2001)
pIrp2	14.3 kb, Ap^r, *irp2*$^+$; *irp2* gene cloned into pBGL2	(Bearden *et al.*, 1997)
pKD3	2.8 kb, Ap^r, Cm^r, template plasmid	(Datsenko and Wanner, 2000)
pKD4	3.3 kb, Ap^r, Km^r, template plasmid	(Datsenko and Wanner, 2000)
pKNG101	6.8 kb, Sm^r, R6K ori $SacB^+$ suicide vector	(Kaniga *et al.*, 1991)
pKNGΔtonB2[c]	8.7 kb, Sm^r, R6K ori $SacB^+$, *ΔhasB2080*; suicide vector; 1.8 kb *Dra*I/*Bam*HI fragment from pUC19ΔtonB2 ligated into the *Sma*I and *Bam*HI sites of pKNG101	(Perry *et al.*, 2003b)
pKNGΔznuA	8.3 kb Sm^r, R6K ori $SacB^+$, *ΔznuA2197*; suicide vector; 1.5 kb *Sal*I/*Xba*I fragment from pBSΔZnuA ligated into the same sites in pKNG101	This study
pPSN15	8.9 kb, Ap^r, R6K ori, $SacB^+$, *Δpsn2045.1*; suicide vector	(Fetherston *et al.*, 1995)
pSucZnu3.5	12.2 kb, Ap^r, *ΔznuBC2077 sacB*$^+$*,* R6K ori; in suicide vector pSUC1	(Hazlett *et al.*, 2003)
pTNS2	9.6 kb, Ap^r, R6K ori, Tn*7* transposase helper plasmid	(Choi *et al.*, 2005)
pUC18R6K-	4.5 kb, Ap^r, Km^r, mini-Tn*7*T-*kan*, R6K ori suicide vector	(Choi and

mini-Tn*7*T-Km		Schweizer, 2006)
pUC18R6K-ZnuABC-Km	9.8 kb, Ap^r, Km^r, mini-Tn*7*T-*kan*, R6K ori suicide vector carrying *znuABC*$^+$	This study
pUC19	2.7 kb, Ap^r, cloning vector	(Yanisch-Perron *et al.*, 1985)
pUC19ΔtonB2[c]	4.5 kb, Ap^r, *ΔhasB2080*; 2.3 kb *Bam*HI/*Hind*III fragment from pHas98 ligated into the same sites in pUC19, then digested with *Hinc*II/*Nru*I and the resulting 4.5 kb fragment religated	(Perry *et al.*, 2003b)
pWL204	8.2 kb, Ap^r *sacB*$^+$; λ-red recombinase helper plasmid containing	(Lathem *et al.*, 2007)
pYbtPQ	7.7 kb, Cm^r, *ybtPQ* cloned into pACYC184	(Fetherston *et al.*, 1999)
pYbtPQX	8.0 kb, Cm^r, *ybtPQX* cloned into pACYC184	(Fetherston *et al.*, 1999)
pYbtX	5.5 kb, Cm^r, 3.4 kb DNA fragment (*ybtPQ*) was deleted from pYbtPQX using reverse SOE PCR; *ybtX* expressed from native promoter in pACYC184	This study
pZnu2	16.1 kb, Km^r , *znuABC*$^+$ cloned into pWSK129	(Desrosiers *et al.*, 2010)
Primer name	*Primer sequence (5’→3’)*	Purpose
y1329red-Forw	GACTTACAACTTACATCACAGTAATTGCAGAATATC CAAGGGTTGAATAGTGTAGGCTGGAGCTGCTTC	construct *Δy1329-1330::kan*
y1330red-Rev	TCTGCAGCGTCAAGGTCGAAGGGTATCAGCGACC AAATGGACGAACAGCCATATGAATATCCTCCTTAGT	construct *Δy1329-1330::kan*
y1245-KMI	ATCTACCTGCTAATCGGCCTGTTGGTGGTGAATGG GGTGTAGGCTGGAGCTGCTTC	construct *Δy1245::cat*
y1245-KMII	AACTAACCAGCAACATATAAACATCCCTATAATCCA CATATGAATATCCTCCTTAGT	construct *Δy1245::cat*
ZnuC.5	CCGAAGCCAGATTAAAGG	prepare hybridization probe
ZnuC.3	GAAGGTACCGCAGAGAAAGGGAAATATCG	prepare hybridization probe
ybtPQdel_F	AGTTACTCCATCGCTCACCGTTTATCC	construct *ΔybtPQ*
ybtPQdel_R	GTGAGCGATGGAGTAACTGAATTTCCTGATGAA	construct *ΔybtPQ*
ZnuA5.3	GATCGCTTTATCACAGTTAC	confirm *znuABC* integration
attTn7Yp-Fwd	TCAGCTGCCACATGTCGAAG	confirm

		znuABC integration
pBAD-y1329-start	CATCGACTTACAACTTACATCACAG	confirm *Δy1329-1330::kan*
Km2	CAATAGCAGCCAGTCCCTTC	confirm *Δy1329-1330::kan*
Y1245-3	TTGAAAATAATCTGGAGT	confirm *Δy1245::cat*
Cm-2	GAGATTTTCAGGAGCTAAGG	confirm *Δ1245::cat*
Irp Km1	AAAGTCGGAGGATATCGC	confirm *irp2::kan*
Mini-kan-1	TGCCTCTTCCGACCATCA	confirm *irp2::kan*
PP-7	GGTTATCGACATAGACGG	confirm *Δpsn*
PP-11	CCGCGAGAAGTTAAATTC	confirm *Δpsn*
Znu-delI	AGGCTCAGCACAAACATG	confirm *ΔznuBC*
ZnuC.5	CCGAAGCCAGATTAAAGG	confirm *ΔznuBC*
ZnuA 3.2	GTCCTTGTCCAATACTATAC	confirm *ΔznuA*
ZnuA C.3	CGCAGAGAAGGGAAATATCG	confirm *ΔznuA*
tonB2-P1	CCTGGCGAATAAGGCCTC	confirm *ΔhasB*
tonB2-P2	ATTTTGGCTAGTCGGGGC	confirm *ΔhasB*
pPQXvector_2100_seq	AGGAAGCAGCCCAGTAGTAG	confirm *ΔybtPQ*
YbtP-24	GCATAAACAGGGTTGTCG	confirm *ΔybtPQ*
P27	TGCATGAGTGATGTTCAG	confirm *ΔybtX*
P33	GCGAAATGGACTGGACAA	confirm *ΔybtX*

[a] For *Y. pestis* strains, a plus sign indicates an intact chromosomal 102-kb *pgm* locus. All other *Y. pestis* strains have a mutation within this locus or a deletion of the entire locus.

[b] Ap^r, Cm^r, Gm^r, Km^r, Spc^r, and Sm^r indicate resistance to ampicillin, chloramphenicol, kanamycin, gentamicin, spectinomycin, and streptomycin, respectively.

[c] Details on the construction of pUC19ΔtonB2 and pKNGΔtonB2 are included since these were omitted in Perry et al (Perry *et al.*, 2003b).

REFERENCES

Ausubel, F.M., Brent, R., Kingston, R.E., Moore, D.D., Seidman, J.G., Smith, J.A. and Struhl, K. (1987) Current Protocols in Molecular Biology. In. New York, N. Y.: John Wiley & Sons, pp.

Bearden, S.W., Fetherston, J.D. and Perry, R.D. (1997) Genetic organization of the yersiniabactin biosynthetic region and construction of avirulent mutants in *Yersinia pestis*. *Infect Immun* **65**: 1659-1668.

Choi, K.-H., Gaynor, J.B., White, K.G., Lopez, C., Bosio, C.M., Karkhoff-Schweizer, R.R. and Schweizer, H.P. (2005) A Tn7-based broad-range bacterial cloning and expression system. *Nat Methods* **2**: 443-448.

Choi, K.-H. and Schweizer, H.P. (2006) mini-Tn*7* insertion in bacteria with single *att*Tn*7* sites: example *Pseudomonas aeruginosa*. *Nat Protocols* **1**: 153-161.

Datsenko, K.A. and Wanner, B.L. (2000) One-step inactivation of chromosomal genes in *Escherichia coli* K-12 using PCR products. *Proc Natl Acad Sci U S A* **97**: 6640-6645.

Desrosiers, D.C., Bearden, S.W., Mier, I., Jr, , Abney, J., Paulley, J.T., Fetherston, J.D.*, et al.* (2010) Znu is the predominant zinc importer in *Yersinia pestis* during *in vitro* growth but is not essential for virulence. *Infect Immun* **78**: 5163-5177.

Fetherston, J.D., Bearden, S.W. and Perry, R.D. (1996) YbtA, an AraC-type regulator of the *Yersinia pestis* pesticin/yersiniabactin receptor. *Mol Microbiol* **22**: 315-325.

Fetherston, J.D., Bertolino, V.J. and Perry, R.D. (1999) YbtP and YbtQ: two ABC transporters required for iron uptake in *Yersinia pestis*. *Mol Microbiol* **32**: 289-299.

Fetherston, J.D., Kirillina, O., Bobrov, A.G., Paulley, J.T. and Perry, R.D. (2010) The yersiniabactin transport system is critical for the pathogenesis of bubonic and pneumonic plague. *Infect Immun* **78**: 2045-2052.

Fetherston, J.D., Lillard, J.W., Jr. and Perry, R.D. (1995) Analysis of the pesticin receptor from *Yersinia pestis*: role in iron-deficient growth and possible regulation by its siderophore. *J Bacteriol* **177**: 1824-1833.

Fetherston, J.D., Schuetze, P. and Perry, R.D. (1992) Loss of the pigmentation phenotype in *Yersinia pestis* is due to the spontaneous deletion of 102 kb of chromosomal DNA which is flanked by a repetitive element. *Mol Microbiol* **6**: 2693-2704.

Geoffroy, V.A., Fetherston, J.D. and Perry, R.D. (2000) *Yersinia pestis* YbtU and YbtT are involved in synthesis of the siderophore yersiniabactin but have different effects on regulation. *Infect Immun* **68**: 4452-4461.

Gong, S., Bearden, S.W., Geoffroy, V.A., Fetherston, J.D. and Perry, R.D. (2001) Characterization of the *Yersinia pestis* Yfu ABC iron transport system. *Infect Immun* **67**: 2829-2837.

Hazlett, K.R.O., Rusnak, F., Kehres, D.G., Bearden, S.W., La Vake, C.J., La Vake, M.E.*, et al.* (2003) The *Treponema pallidum tro* operon encodes a multiple metal transporter, a zinc-dependent transcriptional repressor, and a semi-autonomously expressed phosphoglycerate mutase. *J Biol Chem* **278**: 20687-20694.

Kaniga, K., Delor, I. and Cornelis, G.R. (1991) A wide-host-range suicide vector for improving reverse genetics in Gram-negative bacteria: inactivation of the *blaA* gene of *Yersinia enterocolitica*. *Gene* **109**: 137-141.

Lathem, W.W., Price, P.A., Miller, V.L. and Goldman, W.E. (2007) A plasminogen-activating protease specifically controls the development of primary pneumonic plague. *Science* **315**: 509-513.

Perry, R.D., Abney, J., Mier, I., Jr., Lee, Y., Bearden, S.W. and Fetherston, J.D. (2003a) Regulation of the *Yersinia pestis* Yfe and Ybt iron transport systems. *Adv Exp Med Biol* **529**: 275-283.

Perry, R.D., Pendrak, M.L. and Schuetze, P. (1990) Identification and cloning of a hemin storage locus involved in the pigmentation phenotype of *Yersinia pestis*. *J Bacteriol* **172**: 5929-5937.

Perry, R.D., Shah, J., Bearden, S.W., Thompson, J.M. and Fetherston, J.D. (2003b) *Yersinia pestis* TonB: role in iron, heme and hemoprotein utilization. *Infect Immun* **71**: 4159-4162.

Rossi, M.-S., Fetherston, J.D., Létoffé, S., Carniel, E., Perry, R.D. and Ghigo, J.-M. (2001) Identification and characterization of the hemophore-dependent heme acquisition system of *Yersinia pestis*. *Infect Immun* **69**: 6707-6717.

Yanisch-Perron, C., Vieira, J. and Messing, J. (1985) Improved M13 phage cloning vectors and host strains: nucleotide sequences of the M13mp18 and pUC19 vectors. *Gene* **33**: 103-119.