#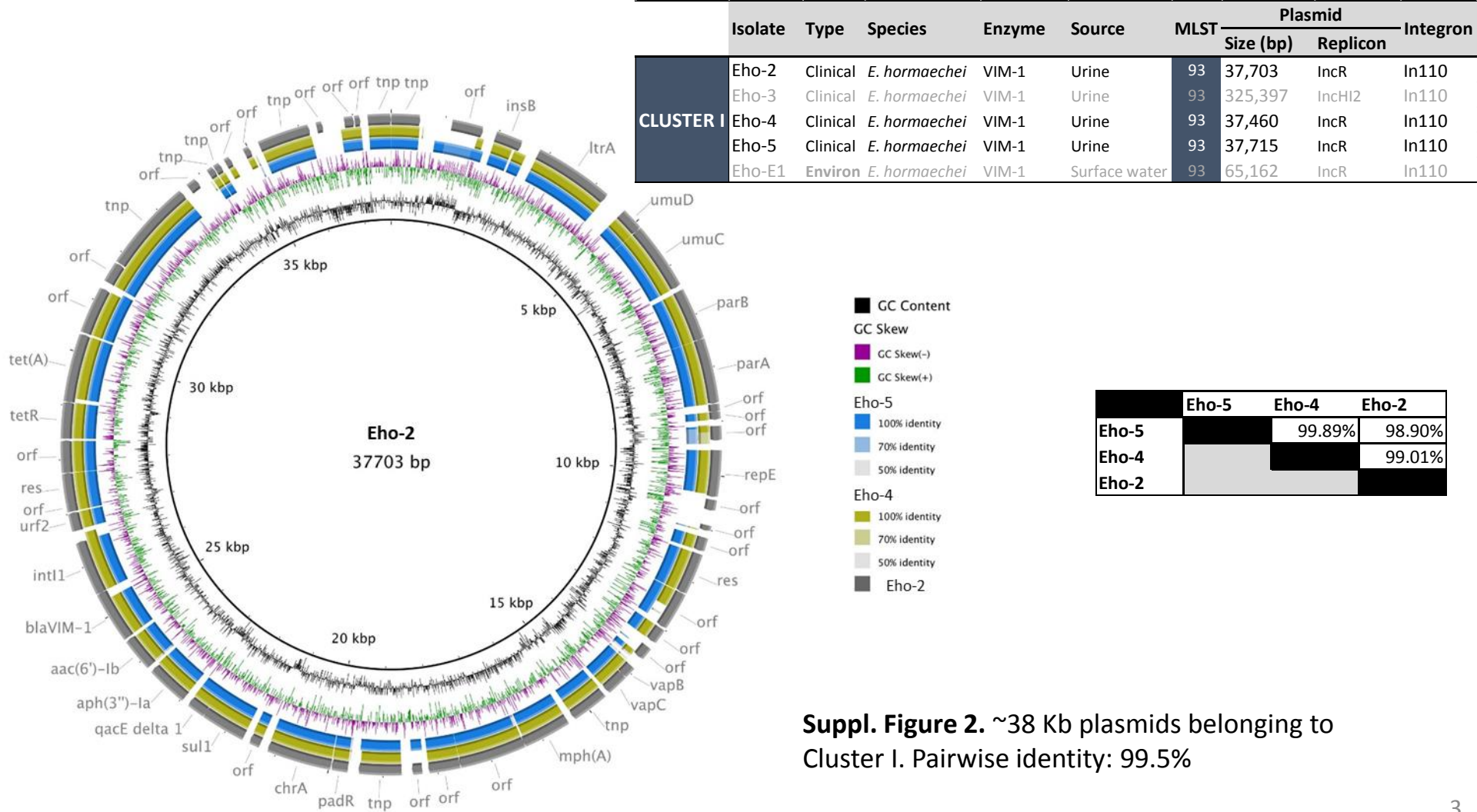 Dissemination of Verona Integron–encoded Metallo-β-Lactamase among Clinical and Environmental Enterobacteriaceae Isolates in Ontario, Canada

Philipp Kohler, Nathalie Tijet, Hyunjin C. Kim, Jennie Johnstone, Tom Edge, Samir N. Patel, Christine Seah, Barbara Willey, Brenda Coleman, Karen Green, Irene Armstrong, Kevin Katz, Matthew P. Muller, Jeff Powis, Susan M. Poutanen, David Richardson, Alicia Sarabia, Andrew Simor, Allison McGeer, Roberto G. Melano, for the Toronto Invasive Bacterial Diseases Network (TIBDN)

Supplementary Figures and Tables
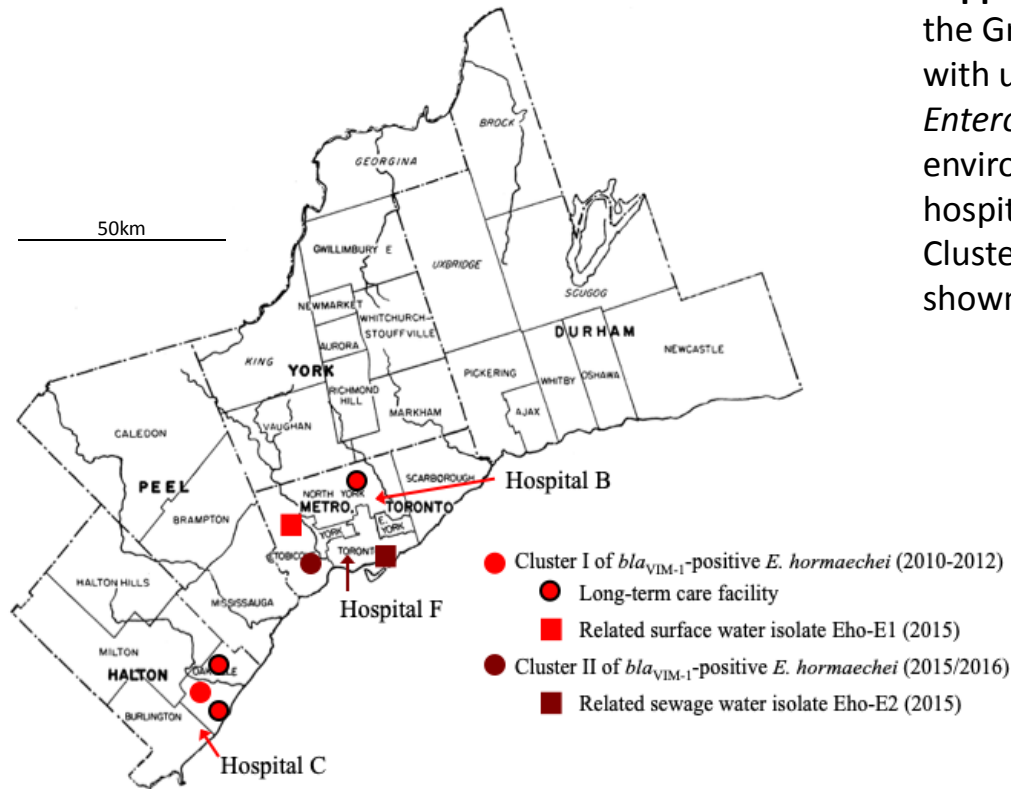
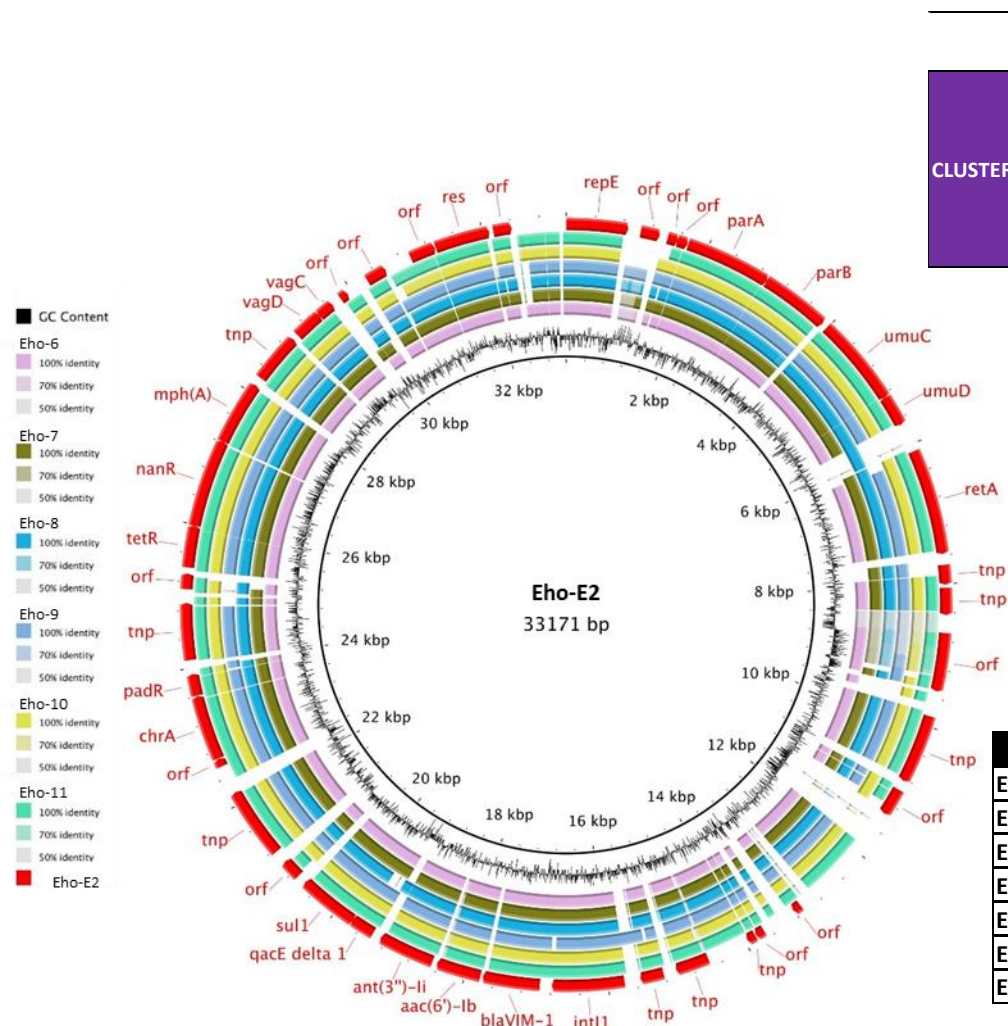**Supplementary Figure 1.** Place of residence within the Greater Toronto Area, Canada, of five patients with unexplained acquisition of bla_{VIM-1}-positive *Enterobacter hormaechei* (circles) and site of environmental water sampling (squares). Involved hospitals (arrows) are depicted by arrows. Clustering based on multilocus sequence typing is shown in bright (cluster I) and dark red (cluster II).

	Isolate	Type	Species	Enzyme	Source	MLST	Plasmid		Integron
							Size (bp)	Replicon	
CLUSTER I	Eho-2	Clinical	*E. hormaechei*	VIM-1	Urine	93	37,703	IncR	In110
	Eho-3	Clinical	*E. hormaechei*	VIM-1	Urine	93	325,397	IncHI2	In110
	Eho-4	Clinical	*E. hormaechei*	VIM-1	Urine	93	37,460	IncR	In110
	Eho-5	Clinical	*E. hormaechei*	VIM-1	Urine	93	37,715	IncR	In110
	Eho-E1	**Environ**	*E. hormaechei*	VIM-1	Surface water	93	65,162	IncR	In110

GC Content

GC Skew
- GC Skew(−)
- GC Skew(+)

Eho-5
- 100% identity
- 70% identity
- 50% identity

Eho-4
- 100% identity
- 70% identity
- 50% identity

Eho-2

	Eho-5	Eho-4	Eho-2
Eho-5		99.89%	98.90%
Eho-4			99.01%
Eho-2			

Suppl. Figure 2. ~38 Kb plasmids belonging to Cluster I. Pairwise identity: 99.5%

3

Isolate	Type	Species	Enzyme	Source	MLST	Plasmid		Integron
						Size (bp)	Replicon	
Eho-6	Clinical	*E. hormaechei*	VIM-1	Rectal	269	33,171	IncR	In110
Eho-7	Clinical	*E. hormaechei*	VIM-1	Urine	269	33,060	IncR	In110
Eho-8	Clinical	*E. hormaechei*	VIM-1	Dialysis fluid	269	32,990	IncR	In110
CLUSTER II Eho-9	Clinical	*E. hormaechei*	VIM-1	Dialysis fluid	269	33,024	IncR	In110
Eho-10	Clinical	*E. hormaechei*	VIM-1	Rectal	269	33,171	IncR	In110
Eho-11	Clinical	*E. hormaechei*	VIM-1	Rectal	269	32,697	IncR	In110
Eho-E2	Environ	*E. hormaechei*	VIM-1	Sewage water	269	33,171	IncR	In110

Suppl. Figure 3. ~33 Kb plasmids belonging to Cluster II. Pairwise identity: 99.2%

	Eho-7	Eho-9	Eho-11	Eho-10	Eho-E2	Eho-6	Eho-8
Eho-7		99.89%	98.90%	99.23%	99.67%	99.33%	99.33%
Eho-9			99.01%	99.34%	99.55%	99.22%	99.44%
Eho-11				99.66%	98.57%	98.24%	99.09%
Eho-10					98.90%	98.57%	99.43%
Eho-E2						99.67%	99.42%
Eho-6							99.09%
Eho-8							
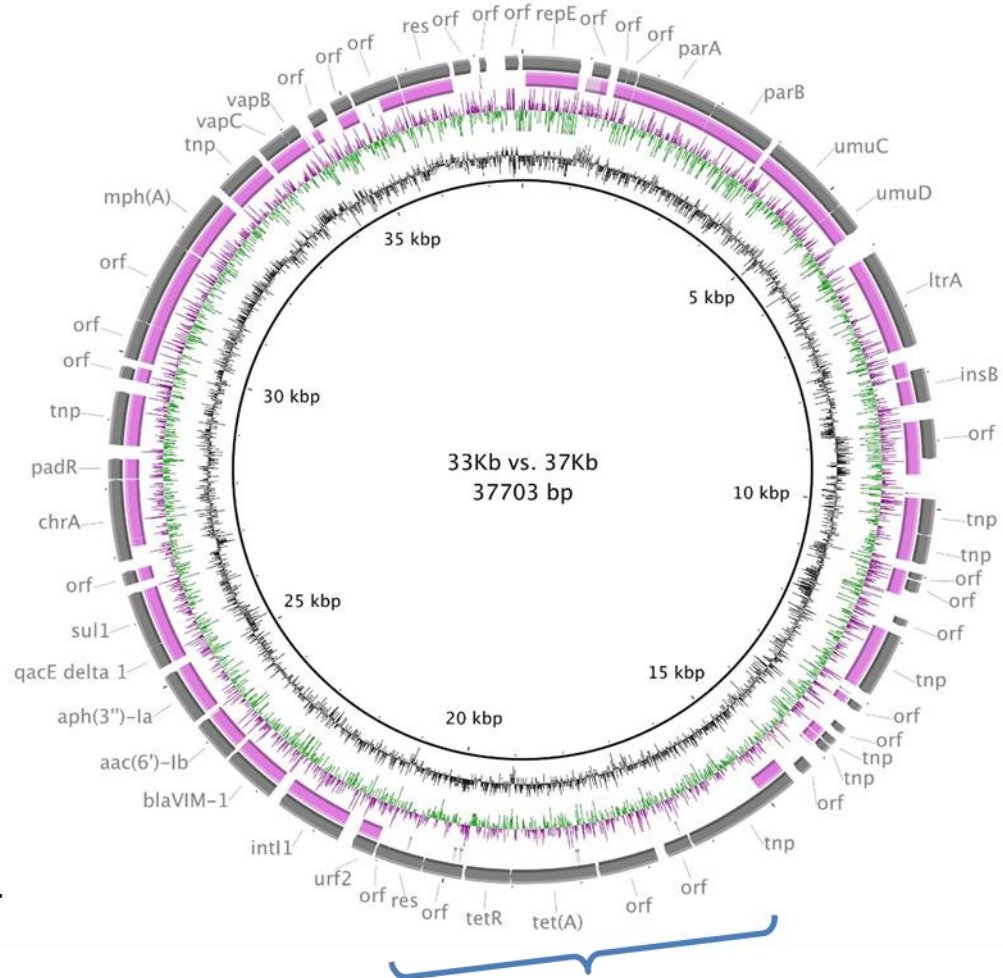
Suppl. Figure 4. Comparison of plasmids from Cluster I (Eho-2, ~37.7 kb) vs. Cluster II (isolate Eho-E2, ~33.2 kb). A ~6.3 kb DNA fragment containing the *tetAR* genes encoding for the tetracycline efflux protein TetA and its repressor TetR is missing in Eho-E2.

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Suppl. Figure 5. Comparison of IncR plasmids from Cluster I (Eho-2, ~37.7 kb, vs. Eho-E1, ~65.2 kb)
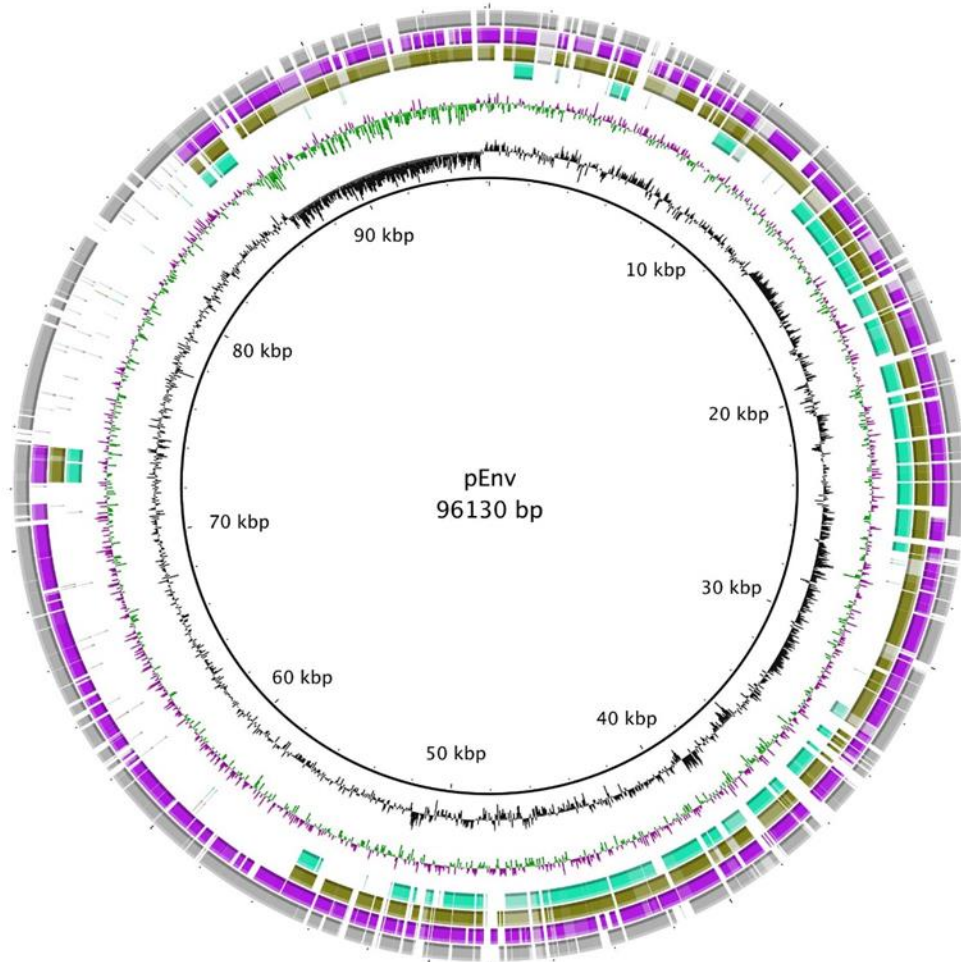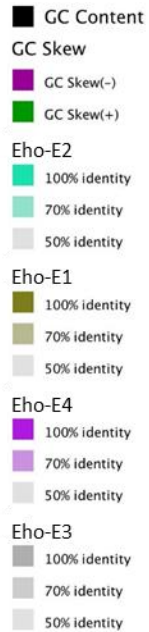
Suppl. Figure 6. bla_{VIM-1}, IncR plasmids from environmental isolates

Suppl. Figure 7. IncHI2 VIM-plasmid (~325 kb) found in clinical *E. hormaechei* isolate Eho-3 (from cluster I, ST93).

Comparison with pRH-R27 (GenBank acc. number LN555650, ~299 kb) and pSE15-SA01028 (CP026661, ~311 kb), bla_{VIM-1}-carrying plasmids found in *Salmonella enterica* serovar Infantis from Germany (99.92 %; 90% of query cover). A ~32 kb fragment (including a putative *lac* operon) disrupting an open reading frame encoding for a phosphoadenosine phosphosulfate reductase was detected in pEho-3. The lack of mobile elements flanking this fragment suggest a recombination event.
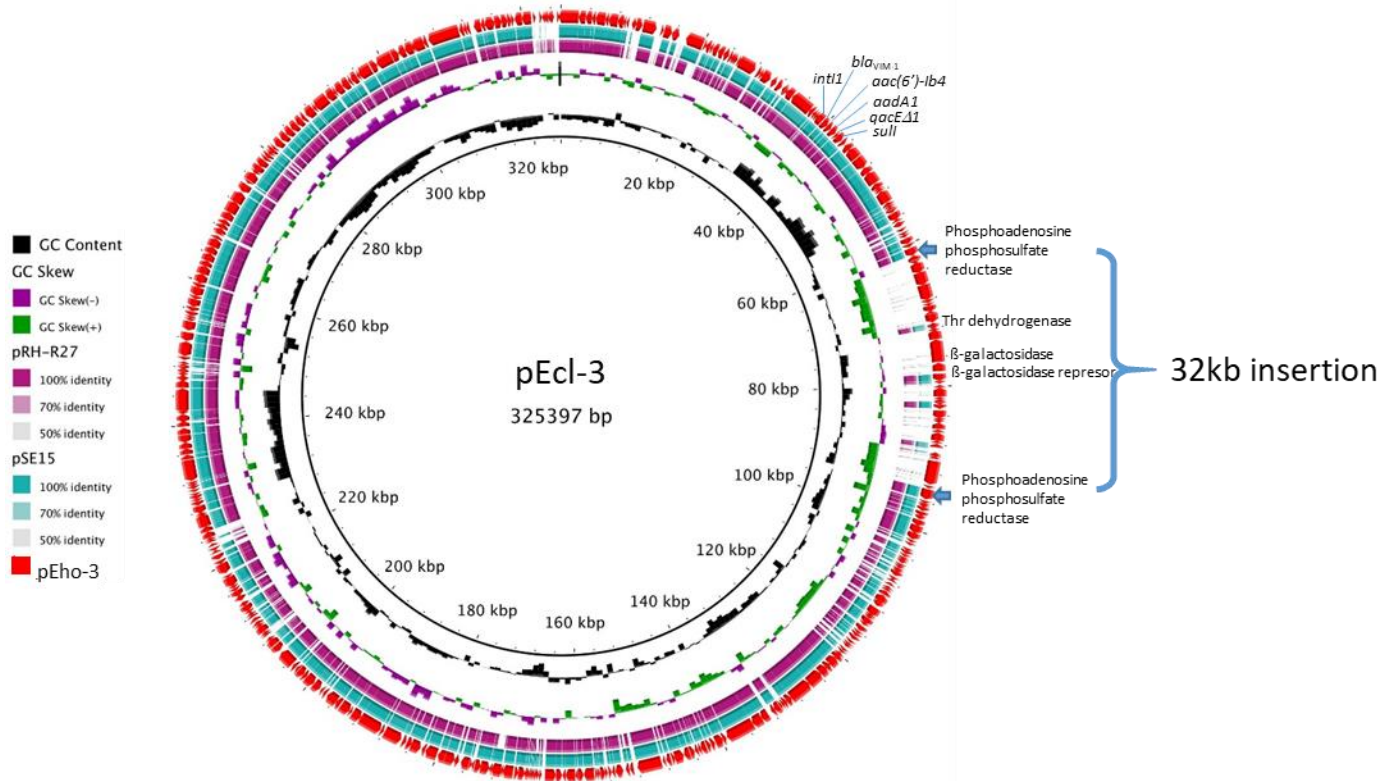
Table S1. Risk profile of patients with explained (i.e. healthcare visit abroad or epidemiologically and microbiologically established link) and patients with unexplained acquisition of blaVIM-1-positive Enterobacteriaceae

	All patients (N=14)	Explained (N=7)	Unexplained (N=7)	P-value
Demographics				
Male sex (%)	7 (50.0)	5 (71.4)	2 (28.6)	0.29
Age in years, median (range)	76.5 (24-90)	71 (24-80)	87 (65-90)	**0.03**
Clinical samples	8 (57.1)	4 (57.1)	4 (57.1)	1
Comorbidities				
Charlson score, median (range)	3 (0-7)	4 (0-5)	2 (1-7)	0.75
Diabetes mellitus	5 (35.7)	4 (57.1)	1 (14.3)	0.27
Cancer	4 (28.6)	1 (14.3)	3 (42.9)	0.56
Chronic kidney disease	5 (35.7)	3 (42.9)	2 (28.6)	1
Dialysis	3 (21.4)	3 (42.9)	0 (0.0)	0.19
Dementia	5 (35.7)	1 (14.3)	4 (57.1)	0.27
Epidemiological exposures*				
Community-acquired†	1 (7.1)	0 (0.0)	1 (14.3)	0.56
Nursing home resident	3 (21.4)	0 (0.0)	3 (42.9)	0.19
Intensive care stay	6 (42.9)	4 (57.1)	2 (28.6)	0.59
Outpatient visits, median (range)	2 (0-8)	1 (0-8)	3 (0-8)	0.9
Antibiotic resistant pathogens*				
MRSA	2 (14.3)	0 (0)	2 (28.6)	0.46
ESBL	4 (28.6)	1 (14.3)	3 (42.9)	0.56
Medical interventions*				
Colonoscopy	2 (14.3)	1 (14.3)	1 (14.3)	1
Gastroscopy	3 (21.4)	1 (14.3)	2 (28.6)	1
Foley catheter	7 (50.0)	3 (42.9)	4 (57.1)	1
Cystoscopy	2 (14.3)	1 (14.3)	1 (14.3)	1
Surgery	4 (28.6)	0 (0.0)	4 (57.1)	0.07
Central venous catheter	3 (21.4)	3 (42.9)	0 (0.0)	0.19
Blood transfusion	3 (21.4)	1 (14.3)	2 (28.6)	1
Medication*				
Antibiotics	5 (35.7)	7 (100.0)	5 (71.4)	0.46
Proton-Pump Inhibitors	6 (42.9)	3 (42.9)	3 (42.9)	1

*All within 1 year before VIM detection

†No previous hospital admission, no nursing home resident, no dialysis

Table S2. Characteristics of the VIM-positive isolates included in this study

	Isolate	Type	Species	Enzyme	Source	Isolation date	MLST	Plasmid Size (bp)	Plasmid Replicon	Integron
	Eho-1	Clinical	*E. hormaechei*	VIM-1	Rectal	1-Sep-2012	92	178'304	IncFIB-IncFII	In110
CLUSTER I	Eho-2	Clinical	*E. hormaechei*	VIM-1	Urine	12-Jul-2011	93	37'703	IncR	In110
	Eho-3	Clinical	*E. hormaechei*	VIM-1	Urine	10-Aug-2011	93	325'397	IncHI2	In110
	Eho-4	Clinical	*E. hormaechei*	VIM-1	Urine	18-Feb-2012	93	37'460	IncR	In110
	Eho-5	Clinical	*E. hormaechei*	VIM-1	Urine	20-Jun-2012	93	37'715	IncR	In110
	Eho-E1	Environ	*E. hormaechei*	VIM-1	Surface water	4-Aug-2015	93	65'162	IncR	In110
CLUSTER II	Eho-6	Clinical	*E. hormaechei*	VIM-1	Rectal	25-Feb-2014	269	33'171	IncR	In110
	Eho-7	Clinical	*E. hormaechei*	VIM-1	Urine	2-Jul-2014	269	33'060	IncR	In110
	Eho-8	Clinical	*E. hormaechei*	VIM-1	Dialysis fluid	2-Jun-2015	269	32'990	IncR	In110
	Eho-9	Clinical	*E. hormaechei*	VIM-1	Dialysis fluid	27-Sep-2015	269	33'024	IncR	In110
	Eho-10	Clinical	*E. hormaechei*	VIM-1	Rectal	18-Jan-2016	269	33'171	IncR	In110
	Eho-11	Clinical	*E. hormaechei*	VIM-1	Rectal	18-Jan-2016	269	32'697	IncR	In110
	Eho-E2	Environ	*E. hormaechei*	VIM-1	Sewage water	23-Jul-2015	269	33'171	IncR	In110
Other species	Cfr-12	Clinical	*C. freundii*	VIM-2	Sputum	28-Feb-2014	129	30'361	No hit found	In58
	Cfr-13a	Clinical	*C. freundii*	VIM-1	Rectal	21-Dec-2015	New	81'066	IncN-IncR	In110
	Kpn-13b	Clinical	*K. pneumoniae*	VIM-1	Rectal	14-Dec-2015	17	81'960	IncN-IncR	In110
	Kpn-14	Clinical	*K. pneumoniae*	VIM-1	Rectal	29-Sep-2014	376	150'505	IncA/C2	In110
	Eco-15	Clinical	*E. coli*	VIM-1	Rectal	26-Nov-2015	131	152'546	IncA/C	In110
Other Ecl env isolates	Eho-E3	Environ	*E. hormaechei*	VIM-1	Surface water	4-Aug-2015	93	96,194 bp	IncR	In1772
	Eho-E4	Environ	*E. hormaechei*	VIM-1	Surface water	4-Aug-2015	93	83,090 bp	IncR	In1772

*MIC, Minimal Inhibitory Concentration

FOX, Cefoxitin; CAZ, Ceftazidime; CTX, Cefotaxime; FEP, Cefepime; AZT, Aztreonam; ETP, Ertapenem; MEM, Meropenem;
IMI, Imipenem; AMK, Amikacin; GEN, Gentamicin; NAL, Nalidixic acid; CIP, Ciprofloxacin; TGC, Tigecyclin; FOS, Fosfomycin; SXT, Sulfamethoxazol-Trimethoprim; COL, Colistin

Table S2. Characteristics of the VIM-positive isolates included in this study

	Isolate	MIC* (µg/ml)															
		FOX	CAZ	CTX	FEP	AZT	ETP	MEM	IMI	AMK	GEN	NAL	CIP	TGC	FOS	SXT	COL
	Eho-1	≥256	≥256	≥256	96	8	2	4	12	64	96	32	6	0.5	24	≥32	0.125
CLUSTER I	Eho-2	256	≥256	192	32	0.12	≥8	16	8	2	24	≥256	≥32	1	32	≥32	0.125
	Eho-3	≥256	≥256	≥256	256	4	≥8	8	16	3	2	≥256	≥32	0.25	6	≥32	0.125
	Eho-4	≥256	≥256	≥256	256	1	8	16	8	2	32	≥256	≥32	0.75	32	≥32	0.094
	Eho-5	≥256	≥256	256	64	4	≥8	12	16	3	96	≥256	≥32	0.5	12	≥32	0.125
	Eho-E1	≥256	≥256	≥256	64	2	≥8	16	12	3	1	≥256	≥32	0.5	24	≥32	0.094
CLUSTER II	Eho-6	≥256	≥256	128	24	2	4	1.5	6	8	2	16	2	0.19	0.38	≥32	0.094
	Eho-7	≥256	≥256	256	24	4	2	8	8	8	2	16	3	0.19	0.094	≥32	0.125
	Eho-8	≥256	≥256	128	24	2	2	12	8	6	2	8	3	0.125	0.25	≥32	0.094
	Eho-9	≥256	≥256	256	64	2	2	2	4	6	2	12	4	0.19	0.5	≥32	0.094
	Eho-10	≥256	≥256	128	48	2	2	1.5	4	2	1.5	12	0.75	0.25	8	≥32	0.094
	Eho-11	≥256	≥256	192	32	2	2	1.5	4	12	3	16	4	0.38	0.75	≥32	0.094
	Eho-E2	≥256	≥256	192	64	0.25	2	1.5	16	8	2	16	2	0.19	12	≥32	0.094
Other species	Cfr-12	≥256	24	64	1	2	2	0.5	1.5	48	≥1024	≥256	≥32	0.38	0.5	0.5	0.125
	Cfr-13a	≥256	≥256	256	64	0.06	0.25	1	4	1.5	1.5	8	0.25	0.094	0.5	≥32	0.094
	Kpn-13b	128	≥256	48	12	0.03	1	1.5	8	2	1.5	8	1	0.19	32	≥32	0.125
	Kpn-14	≥256	≥256	≥256	≥256	≥256	≥8	≥32	≥32	128	64	≥256	≥32	0.25	≥1024	≥32	0.125
	Eco-15	32	48	8	4	8	0.06	16	0.5	2	1	6	0.032	0.094	1	≥32	0.094
Other Ecl env isolates	Eho-E3	≥256	≥256	≥256	64	4	≥8	12	8	4	1.5	≥256	≥32	1	24	≥32	0.094
	Eho-E4	≥256	≥256	256	64	2	≥8	4	8	4	1.5	≥256	≥32	0.75	16	≥32	0.094

*MIC, Minimal Inhibitory Concentration

FOX, Cefoxitin; CAZ, Ceftazidime; CTX, Cefotaxime; FEP, Cefepime; AZT, Aztreonam; ETP, Ertapenem; MEM, Meropenem; IMI, Imipenem; AMK, Amikacin; GEN, Gentamicin; NAL, Nalidixic acid; CIP, Ciprofloxacin; TGC, Tigecyclin; FOS, Fosfomycin; SXT, Sulfamethoxazol-Trimethoprim; COL, Colistin

Table S3. Resistance genotypes and predicted phenotypes as well as plasmid replicon types of all isolates.

	Isolate ID	Genotype	Predicted Phenotype	Plasmid
	Eho-1/GN1534	aac(3)-IIa, aac(6')-Ib-cr, aac(6')-Ib3, aadA1, aph(3')-VIa, blaACT-16, blaOXA-1, blaVIM-1, catB4, dfrA14, qnrB1, sul1	gentamicin, ciprofloxacin I/R, streptomycin, kanamycin, ampicillin, amoxicillin/clavulanic acid, cefoxitin, ceftriaxone, meropenem, chloramphenicol, trimethoprim, sulfisoxazole	IncFIB(pEhoA), IncFII(pEhoA), IncQ1
CLUSTER I	Eho-2/GN719	aac(6')-Ib-cr, aac(6')-Ib3, aadA1, ant(2'')-Ia, aph(6)-Id, blaACT-7, blaTEM-1B, blaVIM-1, dfrA14, fosA, mph(A), qnrS1, strA, sul1, tet(A)	ciprofloxacin I/R, gentamicin, streptomycin, kanamycin, ampicillin, amoxicillin/clavulanic acid, cefoxitin, ceftriaxone, meropenem, trimethoprim, fosfomycin, erythromycin, azithromycin, sulfisoxazole, tetracycline	IncFII(Yp), IncN, IncR
	Eho-3/GN738	aac(6')-Ib-cr, aac(6')-Ib3, aadA1, aph(6)-Id, blaACC-1, blaACT-7, blaVIM-1, catA1, dfrA14, fosA, qnrS1, strA, sul1	ciprofloxacin I/R, gentamicin, streptomycin, kanamycin, ampicillin, amoxicillin/clavulanic acid, cefoxitin, ceftriaxone, meropenem, chloramphenicol, trimethoprim, fosfomycin, sulfisoxazole	IncHI2, IncN
	Eho-4/GN825	aac(3)-IId, aac(6')-Ib-cr, aac(6')-Ib3, aadA1, aadA1b, ant(2'')-Ia, aph(6)-Id, blaACC-1, blaACT-7, blaTEM-1B, blaVIM-1, catA1, dfrA14, ere(A), fosA, mph(A), strA, sul1, tet(A)	gentamicin, ciprofloxacin I/R, streptomycin, kanamycin, ampicillin, amoxicillin/clavulanic acid, cefoxitin, ceftriaxone, meropenem, chloramphenicol, trimethoprim, erythromycin, azithromycin, fosfomycin, sulfisoxazole, tetracycline	Col156, IncFII(29), IncFII(Yp), IncHI2, IncHI2A, IncN, IncR
	Eho-5/GN882	aac(3)-IId, aac(6')-Ib-cr, aac(6')-Ib3, aadA1, aadA2, aph(6)-Id, blaACC-1, blaACT-7, blaTEM-1B, blaVIM-1, catA1, dfrA12, dfrA14, fosA, mph(A), qnrS1, strA, sul1, tet(A)	gentamicin, ciprofloxacin I/R, streptomycin, kanamycin, ampicillin, amoxicillin/clavulanic acid, cefoxitin, ceftriaxone, meropenem, chloramphenicol, trimethoprim, fosfomycin, erythromycin, azithromycin, sulfisoxazole, tetracycline	IncHI2, IncHI2A, IncN, IncR
	Eho-E1/GN1533	aac(6')-Ib-cr, aac(6')-Ib3, aadA1, aadA16, aph(6)-Id, ARR-3, blaACC-1, blaACT-7, blaVIM-1, dfrA27, floR, fosA, mph(A), qnrB6, strA, sul1, sul2, tet(A)	ciprofloxacin I/R, gentamicin, streptomycin, kanamycin, rifampicin, ampicillin, amoxicillin/clavulanic acid, cefoxitin, ceftriaxone, meropenem, trimethoprim, chloramphenicol, fosfomycin, erythromycin, azithromycin, sulfisoxazole, tetracycline	IncHI2, IncHI2A, IncN, IncR, IncX5

		Genes	Resistance phenotype	Plasmid
CLUSTER II	**Eho-6/GN1550**	aac(6')-Ib-cr, aac(6')-Ib3, aadA1, ant(3'')-Ia, aph(3'')-Ib, aph(6)-Id, blaCTX-M-15, blaOXA-1, blaTEM-1B, blaVIM-1, catA1, catB4, dfrA14, mph(A), qnrB1, qnrS1, sul1, sul2	ciprofloxacin I/R, gentamicin, streptomycin, kanamycin, ampicillin, ceftriaxone, amoxicillin/clavulanic acid, cefoxitin, meropenem, chloramphenicol, chloramphenicol, trimethoprim, erythromycin, azithromycin, sulfisoxazole	IncHI2, IncHI2A, IncR
	Eho-7/GN1157	aac(6')-Ib-cr, aac(6')-Ib3, aadA1, ant(3'')-Ia, aph(3'')-Ib, aph(6)-Id, blaCTX-M-15, blaOXA-1, blaTEM-1B, blaVIM-1, catA1, catB4, dfrA14, mph(A), qnrB1, qnrS1, sul1, sul2	ciprofloxacin I/R, gentamicin, streptomycin, kanamycin, ampicillin, ceftriaxone, amoxicillin/clavulanic acid, cefoxitin, meropenem, chloramphenicol, chloramphenicol, trimethoprim, erythromycin, azithromycin, sulfisoxazole	IncHI2, IncHI2A, IncR
	Eho-8/GN1318	aac(6')-Ib-cr, aac(6')-Ib3, aadA1, ant(3'')-Ia, aph(3'')-Ib, aph(6)-Id, blaCTX-M-15, blaOXA-1, blaTEM-1B, blaVIM-1, catA1, catB4, dfrA14, mph(A), qnrB1, qnrS1, sul1, sul2	ciprofloxacin I/R, gentamicin, streptomycin, kanamycin, ampicillin, ceftriaxone, amoxicillin/clavulanic acid, cefoxitin, meropenem, chloramphenicol, chloramphenicol, trimethoprim, erythromycin, azithromycin, sulfisoxazole	IncHI2, IncHI2A, IncR
	Eho-9/GN1494	aac(6')-Ib-cr, aac(6')-Ib-cr, aac(6')-Ib-cr, aac(6')-Ib3, aadA1, ant(3'')-Ia, aph(3'')-Ib, aph(6)-Id, blaCTX-M-15, blaOXA-1, blaTEM-1B, blaVIM-1, catA1, catB4, dfrA14, mph(A), qnrB1, qnrS1, sul1, sul2	gentamicin, ciprofloxacin I/R, streptomycin, kanamycin, ampicillin, ceftriaxone, amoxicillin/clavulanic acid, cefoxitin, meropenem, chloramphenicol, chloramphenicol, trimethoprim, erythromycin, azithromycin, sulfisoxazole	IncHI2, IncHI2A, IncR
	Eho-10/GN1549	aac(6')-Ib-cr, aac(6')-Ib3, aadA1, ant(3'')-Ia, aph(3'')-Ib, aph(6)-Id, blaCTX-M-15, blaTEM-1B, blaVIM-1, catA1, dfrA14, mph(A), qnrB1, qnrS1, sul1, sul2	ciprofloxacin I/R, gentamicin, streptomycin, kanamycin, ampicillin, ceftriaxone, amoxicillin/clavulanic acid, cefoxitin, meropenem, chloramphenicol, trimethoprim, erythromycin, azithromycin, sulfisoxazole	IncHI2, IncHI2A, IncR
	Eho-11/GN1555	aac(6')-Ib-cr, aac(6')-Ib3, aadA1, ant(3'')-Ia, aph(3'')-Ib, aph(6)-Id, blaCTX-M-15, blaOXA-1, blaTEM-1B, blaVIM-1, catA1, catB4, dfrA14, mph(A), qnrB1, qnrS1, sul1, sul2	ciprofloxacin I/R, gentamicin, streptomycin, kanamycin, ampicillin, ceftriaxone, amoxicillin/clavulanic acid, cefoxitin, meropenem, chloramphenicol, chloramphenicol, trimethoprim, erythromycin, azithromycin, sulfisoxazole	IncHI2, IncHI2A, IncR
	Eho-E2/GN1532	aac(6')-Ib-cr, aac(6')-Ib3, aadA1, blaOXA-1, blaVIM-1, catB4, dfrA14, mph(A), qnrB1, qnrS1, sul1	ciprofloxacin I/R, gentamicin, streptomycin, ampicillin, amoxicillin/clavulanic acid, cefoxitin, ceftriaxone, meropenem, chloramphenicol, trimethoprim, erythromycin, azithromycin, sulfisoxazole	IncHI2, IncHI2A, IncR

	Isolate	Resistance genes	Phenotypic resistance	Plasmid replicons
OTHER SPECIES	**Cfr-12/GN1092**	aac(3)-Ia, aac(3)-IId, aac(6')-Ib-cr, aac(6')-Ib3, aac(6')-Il, aph(3')-Ia, ARR-3, blaCMY-39, blaOXA-1, blaTEM-1B, blaVIM-2, catB4, mph(A), qnrA1, qnrB13, sul1	gentamicin, ciprofloxacin I/R, kanamycin, rifampicin, ampicillin, amoxicillin/clavulanic acid, cefoxitin, ceftriaxone, meropenem, chloramphenicol, erythromycin, azithromycin, sulfisoxazole	IncHI2, IncHI2A, IncR
	Cfr-13a/GN1762	aac(6')-Ib-cr, aac(6')-Ib3, aadA1, blaCMY-34, blaVIM-1, dfrA14, mph(A), qnrB9, qnrS1, sul1	ciprofloxacin I/R, gentamicin, streptomycin, ampicillin, amoxicillin/clavulanic acid, cefoxitin, ceftriaxone, meropenem, trimethoprim, erythromycin, azithromycin, sulfisoxazole	IncN, IncR
	Kpn-13b/GN1763	aac(6')-Ib-cr, aac(6')-Ib3, aadA1, blaSHV-11, blaVIM-1, dfrA14, fosA, mph(A), oqxA, oqxB, qnrS1, sul1	ciprofloxacin I/R, gentamicin, streptomycin, ampicillin, amoxicillin/clavulanic acid, cefoxitin, ceftriaxone, meropenem, trimethoprim, fosfomycin, erythromycin, azithromycin, chloramphenicol, sulfisoxazole	IncN, IncR
	Kpn-14/GN1761	aac(3)-IIa, aac(6')-Il, aadA15, ant(3'')-Ia, aph(3')-Ia, aph(3')-VIb, aph(6)-Id, blaCMY-2, blaCTX-M-14b, blaSHV-1, blaTEM-1B, blaVIM-1, catA1, dfrA1, fosA, oqxA, oqxB, qnrS1, strA, sul1, tet(B)	gentamicin, streptomycin, kanamycin, ampicillin, amoxicillin/clavulanic acid, cefoxitin, ceftriaxone, meropenem, chloramphenicol, trimethoprim, fosfomycin, ciprofloxacin I/R, sulfisoxazole, tetracycline	IncA/C2, IncFIB(K), IncFII(K), IncHI1B, IncL/M(pMU407)
	Eco-15/GN1765	aac(6')-Ib-cr, aac(6')-Ib3, aadA1, aph(3')-XV, blaSHV-12, blaVIM-1, catB2, dfrA14, mph(A), sul1, tet(B)	ciprofloxacin I/R, gentamicin, streptomycin, kanamycin, ampicillin, ceftriaxone, amoxicillin/clavulanic acid, cefoxitin, meropenem, chloramphenicol, trimethoprim, erythromycin, azithromycin, sulfisoxazole, tetracycline	Col156, IncA/C, IncFII(29)
Other Ecl env isolates	**Eho-E3/GN1768**	aac(6')-Ib-cr, aac(6')-Ib3, aadA1, aadA16, aph(6)-Id, ARR-3, blaACC-1, blaACT-7, blaVIM-1, dfrA27, mph(A), qnrB6, strA, sul1, sul2, tet(A)	gentamicin, ciprofloxacin I/R, streptomycin, kanamycin, rifampicin, ampicillin, amoxicillin/clavulanic acid, cefoxitin, ceftriaxone, meropenem, trimethoprim, erythromycin, azithromycin, sulfisoxazole, tetracycline	IncHI2, IncHI2A, IncN, IncR, IncX5
	Eho-E4/GN1766	aac(6')-Ib-cr, aac(6')-Ib3, aadA1, aadA16, aph(6)-Id, ARR-3, blaACC-1, blaACT-7, blaVIM-1, dfrA27, mph(A), qnrB6, strA, sul1, sul2, tet(A)	ciprofloxacin I/R, gentamicin, streptomycin, kanamycin, rifampicin, ampicillin, amoxicillin/clavulanic acid, cefoxitin, ceftriaxone, meropenem, trimethoprim, erythromycin, azithromycin, sulfisoxazole, tetracycline	IncHI2, IncHI2A, IncN, IncR, IncX5

Table S4. Results of PlasmidFinder

Isolate ID	Plasmid	%Identity	%Overlap	HSP Length/Total Length	Contig	Start	End	Accession
Eho-1/GN1534	IncFIB(pEhoA)	100	100	560/560	2	35915	36474	CP001919
Eho-1/GN1534	IncFII(pEhoA)	100	100	747/747	2	3	749	CP001919
Eho-1/GN1534	IncQ1	100	81.28	647/796	3	647	1	M28829.1
Eho-2/GN719	IncFII(Yp)	99.52	90.87	209/230	2	105039	105247	CP000670
Eho-2/GN719	IncN	99.61	100	514/514	3	39	552	AY046276
Eho-2/GN719	IncR	100	100	251/251	4	9889	10139	DQ449578
Eho-3/GN738	IncHI2	100	100	327/327	2	15542	15868	BX664015
Eho-3/GN738	IncN	99.61	100	514/514	4	39	552	AY046276
Eho-4/GN825	Col156	98.7	100	154/154	3	20270	20423	009781
Eho-4/GN825	IncFII(29)	99.61	100	259/259	3	45041	45299	CP003035
Eho-4/GN825	IncFII(Yp)	99.52	90.87	209/230	4	105039	105247	CP000670
Eho-4/GN825	IncHI2	100	100	327/327	2	329621	329295	BX664015
Eho-4/GN825	IncHI2A	100	100	630/630	2	211	840	BX664015
Eho-4/GN825	IncN	99.61	100	514/514	5	39	552	AY046276
Eho-4/GN825	IncR	100	100	251/251	6	25273	25523	DQ449578
Eho-5/GN882	IncHI2	100	100	327/327	2	295831	295505	BX664015
Eho-5/GN882	IncHI2A	100	100	630/630	2	211	840	BX664015
Eho-5/GN882	IncN	99.61	100	514/514	2	113678	114191	AY046276
Eho-5/GN882	IncR	100	100	251/251	3	22185	21935	DQ449578
Eho-6/GN1550	IncHI2	100	100	327/327	2	295128	294802	BX664015
Eho-6/GN1550	IncHI2A	100	100	630/630	2	211	840	BX664015
Eho-6/GN1550	IncR	100	100	251/251	4	13123	13373	DQ449578
Eho-7/GN1157	IncHI2	100	100	327/327	2	289263	288937	BX664015
Eho-7/GN1157	IncHI2A	100	100	630/630	2	211	840	BX664015
Eho-7/GN1157	IncR	100	100	251/251	6	24568	24818	DQ449578
Eho-8/GN1318	IncHI2	100	100	327/327	2	292818	292492	BX664015
Eho-8/GN1318	IncHI2A	100	100	630/630	2	211	840	BX664015
Eho-8/GN1318	IncR	100	100	251/251	5	3155	3405	DQ449578
Eho-9/GN1494	IncHI2	100	100	327/327	2	292818	292492	BX664015
Eho-9/GN1494	IncHI2A	100	100	630/630	2	211	840	BX664015
Eho-9/GN1494	IncR	100	100	251/251	6	20006	19756	DQ449578

Strain	Inc type	Col3	Col4	Col5	Col6	Col7
Eho-10/GN1549	IncHI2	100	100 327/327	3	272695	272369 BX664015
Eho-10/GN1549	IncHI2A	100	100 630/630	3	211	840 BX664015
Eho-10/GN1549	IncR	100	100 251/251	9	29830	30080 DQ449578
Eho-11/GN1555	IncHI2	100	100 327/327	2	295128	294802 BX664015
Eho-11/GN1555	IncHI2A	100	100 630/630	2	211	840 BX664015
Eho-11/GN1555	IncR	100	100 251/251	5	29644	29894 DQ449578
Eho-E1/GN1533	IncHI2	100	100 327/327	2	317217	316891 BX664015
Eho-E1/GN1533	IncHI2A	100	100 630/630	2	211	840 BX664015
Eho-E1/GN1533	IncN	99.81	100 514/514	5	39	552 AY046276
Eho-E1/GN1533	IncR	100	100 251/251	4	11271	11521 DQ449578
Eho-E1/GN1533	IncX5	100	100 576/576	6	7364	6789 MF062700.1
Eho-E2/GN1532	IncHI2	100	100 327/327	1	3734347	3734673 BX664015
Eho-E2/GN1532	IncHI2A	100	100 630/630	1	3719481	3718852 BX664015
Eho-E2/GN1532	IncR	100	100 251/251	3	642	392 DQ449578
Eho-E3/GN1768	IncHI2	100	100 327/327	2	117227	116901 BX664015
Eho-E3/GN1768	IncHI2A	99.21	100.16 631/630	2	132073	132701 BX664015
Eho-E3/GN1768	IncN	98.84	100.39 516/514	4	5167	4653 AY046276
Eho-E3/GN1768	IncR	99.6	100 251/251	3	72364	72614 DQ449578
Eho-E3/GN1768	IncX5	99.3	99.83 575/576	5	30014	29443 MF062700.1
Eho-E4/GN1766	IncHI2	99.69	100 327/327	2	76393	76718 BX664015
Eho-E4/GN1766	IncHI2A	99.84	100 630/630	2	61543	60915 BX664015
Eho-E4/GN1766	IncN	99.42	100 514/514	5	39	552 AY046276
Eho-E4/GN1766	IncR	99.6	100 251/251	3	47022	47272 DQ449578
Eho-E4/GN1766	IncX5	99.65	99.65 574/576	6	44119	43548 MF062700.1
Cfr-12/GN1092	IncHI2	100	100 327/327	2	366162	365836 BX664015
Cfr-12/GN1092	IncHI2A	100	100 630/630	2	211	840 BX664015
Cfr-12/GN1092	IncR	100	100 251/251	4	26371	26121 DQ449578
Cfr-13a/GN1762	IncN	99.61	100 514/514	3	39	552 AY046276
Cfr-13a/GN1762	IncR	100	100 251/251	3	15001	14751 DQ449578
Kpn-13b/GN1763	IncN	99.61	100 514/514	3	39	552 AY046276
Kpn-13b/GN1763	IncR	100	100 251/251	3	16006	15756 DQ449578
Kpn-14/GN1761	IncA/C2	100	100 417/417	4	45	461 JN157804
Kpn-14/GN1761	IncFIB(K)	98.93	100 560/560	3	859	300 JN233704

Kpn-14/GN1761	IncFII(K)	98.65	100	148/148	3	119262	119115 CP000648
Kpn-14/GN1761	IncHI1B	99.47	100	570/570	2	655	86 JN420336
Kpn-14/GN1761	IncL/M(pMU407)	99.55	90.66	670/739	5	72383	73052 U27345
Eco-15/GN1765	Col156	98.7	100	154/154	3	20275	20428 009781
Eco-15/GN1765	IncA/C	98.32	100	417/417	2	45	461 FJ705807
Eco-15/GN1765	IncFII(29)	99.61	100	259/259	3	51867	52125 CP003035

Table S5. Summary of multilocus sequence typing results

Isolate ID	Scheme	Sequence Type	Locus 1	Locus 2	Locus 3	Locus 4	Locus 5	Locus 6	Locus 7
Eho-1/GN1534	ecloacae	92	dnaA(52)	fusA(20)	gyrB(19)	leuS(44)	pyrG(45)	rplB(12)	rpoB(32)
Eho-2/GN719	ecloacae	93	dnaA(9)	fusA(4)	gyrB(14)	leuS(61)	pyrG(37)	rplB(4)	rpoB(9)
Eho-3/GN738	ecloacae	93	dnaA(9)	fusA(4)	gyrB(14)	leuS(61)	pyrG(37)	rplB(4)	rpoB(9)
Eho-4/GN825	ecloacae	93	dnaA(9)	fusA(4)	gyrB(14)	leuS(61)	pyrG(37)	rplB(4)	rpoB(9)
Eho-5/GN882	ecloacae	93	dnaA(9)	fusA(4)	gyrB(14)	leuS(61)	pyrG(37)	rplB(4)	rpoB(9)
Eho-6/GN1550	ecloacae	269	dnaA(95)	fusA(56)	gyrB(112)	leuS(111)	pyrG(104)	rplB(4)	rpoB(63)
Eho-7/GN1157	ecloacae	269	dnaA(95)	fusA(56)	gyrB(112)	leuS(111)	pyrG(104)	rplB(4)	rpoB(63)
Eho-8/GN1318	ecloacae	269	dnaA(95)	fusA(56)	gyrB(112)	leuS(111)	pyrG(104)	rplB(4)	rpoB(63)
Eho-9/GN1494	ecloacae	269	dnaA(95)	fusA(56)	gyrB(112)	leuS(111)	pyrG(104)	rplB(4)	rpoB(63)
Eho-10/GN1549	ecloacae	269	dnaA(95)	fusA(56)	gyrB(112)	leuS(111)	pyrG(104)	rplB(4)	rpoB(63)
Eho-11/GN1555	ecloacae	269	dnaA(95)	fusA(56)	gyrB(112)	leuS(111)	pyrG(104)	rplB(4)	rpoB(63)
Eho-E1/GN1533	ecloacae	93	dnaA(9)	fusA(4)	gyrB(14)	leuS(61)	pyrG(37)	rplB(4)	rpoB(9)
Eho-E2/GN1532	ecloacae	269	dnaA(95)	fusA(56)	gyrB(112)	leuS(111)	pyrG(104)	rplB(4)	rpoB(63)
Eho-E3/GN1768	ecloacae	93	dnaA(9)	fusA(4)	gyrB(14)	leuS(61)	pyrG(37)	rplB(4)	rpoB(9)
Eho-E4/GN1766	ecloacae	93	dnaA(9)	fusA(4)	gyrB(14)	leuS(61)	pyrG(37)	rplB(4)	rpoB(9)
Cfr-12/GN1092	cfreundii	129	aspC(7)	clpX(6)	fadD(76)	mdh(45)	arcA(3)	dnaG(45)	lysP(63)
Cfr-13a/GN1762	cfreundii	not assigned	aspC(9)	clpX(~11)	fadD(4)	mdh(95)	arcA(59)	dnaG(~66)	lysP(106)
Kpn-13b/GN1763	kpneumoniae	17	gapA(2)	infB(1)	mdh(1)	pgi(1)	phoE(4)	rpoB(4)	tonB(4)
Kpn-14/GN1761	kpneumoniae	376	gapA(2)	infB(6)	mdh(1)	pgi(3)	phoE(8)	rpoB(1)	tonB(27)
Eco-15/GN1765	ecoli	131	adk(53)	fumC(40)	gyrB(47)	icd(13)	mdh(36)	purA(28)	recA(29)

Table S6. Settings

Key	Value
command_line	/ComputeShed/Applications/anaconda2/envs/staramr/bin/staramr search -o out11 GN1092.fasta GN1157.fasta GN1318.fasta GN1494.fasta GN1532.fasta GN1533.fasta GN1534.fasta GN1549.fasta GN1550.fasta GN1555.fasta GN1761.fasta GN1762.fasta GN1763.fasta GN1765.fasta GN1766.fasta GN1768.fasta GN719.fasta GN738.fasta GN825.fasta GN882.fasta
version	0.6.1
start_time	2019-12-12 11:40:22
end_time	2019-12-12 11:40:45
total_minutes	0.38
resfinder_db_dir	/ComputeShed/Applications/anaconda2/envs/staramr/lib/python3.7/site-packages/staramr/databases/data/dist/resfinder
resfinder_db_url	https://bitbucket.org/genomicepidemiology/resfinder_db.git
resfinder_db_commit	e8f1eb2585cd9610c4034a54ce7fc4f93aa95535
resfinder_db_date	Mon, 16 Jul 2018 16:58
pointfinder_db_dir	/ComputeShed/Applications/anaconda2/envs/staramr/lib/python3.7/site-packages/staramr/databases/data/dist/pointfinder
pointfinder_db_url	https://bitbucket.org/genomicepidemiology/pointfinder_db.git
pointfinder_db_commit	8706a6363bb29e47e0e398c53043b037c24b99a7
pointfinder_db_date	Wed, 04 Jul 2018 14:27
plasmidfinder_db_dir	/ComputeShed/Applications/anaconda2/envs/staramr/lib/python3.7/site-packages/staramr/databases/data/dist/plasmidfinder
plasmidfinder_db_url	https://bitbucket.org/genomicepidemiology/plasmidfinder_db.git
plasmidfinder_db_commit	81919954cbedaff39056610ab584ab4c06011ed8
plasmidfinder_db_date	Tue, 20 Nov 2018 08:51
mlst_version	2.11
pointfinder_gene_drug_version	050218
resfinder_gene_drug_version	050218.1