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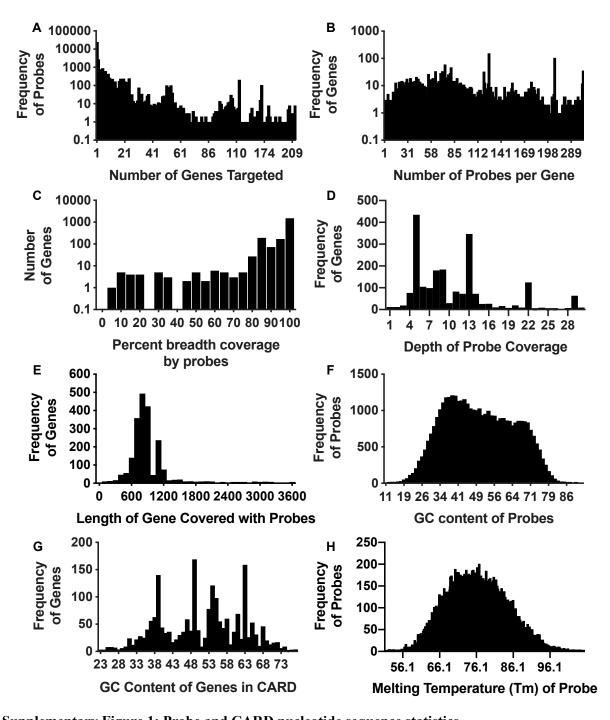
## Supplementary methods: Modified Library Preparation for Trial 2

The same genomic DNA (except for Pseudomonas aeruginosa C00060) used in Trial 1 was used for library construction through a modified library preparation protocol (1). Briefly,  $\sim$ 200 nanograms of sonicated genomic DNA (250 - 300 bp) was used for library preparation at a volume of 25 µL. Deviations from the protocol include the use of KAPA Pure Beads (KK8000, Roche Canada, Laval, QC) instead of AMPure beads, oligonucleotides ordered from Integrated DNA Technologies (IDT, Coralville, IA, USA) (the ready-to-use adapter mix was diluted to 10uM of each adapter prior to use), and a total reaction volume of 50 µL for blunt end repair. Briefly, blunt end repair with T4 polynucleotide kinase ( $10U/\mu L$ ) and T4 DNA polymerase ( $5U/\mu L$ ) was carried out for 15 min at 25°C and 15 min at 12°C in a modified buffer (50mM NaCl, 10mM Tris-HCl, 10 mM MgCl<sub>2</sub>, 100 µg/ml BSA, pH 7.9 at 25C) with 1 mM DTT, 100 µM dNTPs (2.5 mM each), and 1 mM ATP. Adapter ligation was performed using a final adapter concentration of  $2.5\mu$ M, with T4 DNA ligase (0.125U/ $\mu$ L) in T4 DNA ligase buffer (1X) and PEG-4000 (5%) for 15 hours at 16°C, then held at 4°C. The reaction was purified using KAPA Pure beads at 1.8X and eluted in 20 µL of EBT (10 mM Tris-Cl, pH 8.3, 0.05% Tween-20). This was followed with adapter fill-in with Bst polymerase, large fragment ( $8U/\mu L$ ) in ThermoPol reaction buffer (10X) with dNTPs (final concentration of 250 µM each) (30 mins at 37°C; 80°C for 20 mins; hold at 4°C). Purification with library size-selection was performed using KAPA Purebeads (0.6X-0.8X ratio of beads to reaction volume) and products were eluted in 30 µL of 10mM Tris-HCl, pH 8.3. Indexing PCR was performed using 12.5 µL of template, KAPA HiFi Hotstart DNA polymerase ready mix (2X), (750nM of each indexing primer -8 bp primers ordered from IDT diluted in nuclease free water at a stock concentration of  $100\mu M$ ) in a 40  $\mu L$  reaction with the following conditions: 1) 98°C for 45 sec; 2) 98°C for 15 sec; 3) 60°C for 45 sec; 4) 72°C for 30 sec; 5) Repeat

2-4 for 12 cycles total; 6) 72°C for 1 min. Libraries were purified with 1.5X KAPA Pure beads then eluted in 33  $\mu$ L 10 mM Tris pH 8.0. Re-amplification of libraries for 2 cycles was performed using 1  $\mu$ L of each library sample and KAPA HiFi Hotstart DNA polymerase ready mix (2X) and Library Amplification Primer Mix (10X) supplied by KAPA in the Library Amplification Kit with the followed conditions: 1) 98°C for 45 sec; 2) 98°C for 15 sec; 3) 60°C for 45 sec; 4) 72°C for 30 sec; 5) Repeat 2 - 4 for 2 cycles total; 6) 72°C for 1 min. Libraries were purified using 1.5X Kapa Purebeads and eluted in 33  $\mu$ L of 10 mM Tris-HCl pH 8.3. A negative control was also included consisting of a blank DNA extraction carried throughout the DNA extraction of bacterial DNA. Bioanalyzer and qPCR analyses were performed through the McMaster Farncombe Metagenomics Facility on all libraries including the blanks.

#### **Enrichment methods**

For Phase 1 Trial 1, the hybridization mix and blockers mix were prepared as described in the myBaits<sup>®</sup> manual version 3 (Arbor Biosciences, Ann Arbor, MI). 7 μL of DNA library (~100 ng) was added to the blockers mix and libraries were denatured at 95°C for 5 minutes. The hybridization mix and libraries with blockers were brought to the hybridization temperature of 65°C for 5 minutes. Hybridization mixes including the appropriate concentration of probeset were mixed with libraries and incubated at 65°C for 16 hours. After 16 hours, the wash buffer was prepared and heated to 65°C. Dynabeads MyOne Streptavidin C1 beads were prepared with binding buffer (10 mM Tris-HCl pH7.5, 1 mM EDTA, 2M NaCl in nuclease-free water) by three washes with binding buffer, and finally resuspended in binding buffer then heated to 65°C for 30 minutes with frequent mixing, then pelleted on a magnetic rack. Beads were washed with preheated wash buffer (Supplied in the myBaits<sup>®</sup> kit), incubated for 10 minutes at 65°C and the wash was repeated two times. The beads were finally resuspended in 30 µL 10mM Tris-Cl, 0.05% Tween-20, pH 8.3. Post-capture amplification was performed on 15  $\mu$ L of the bead-bound library with for 30 cycles of amplification using KAPA HiFi Hotstart DNA polymerase ready mix (2X), Library Amplification Primer Mix (10X) with the followed conditions: 1) 98°C for 45 sec; 2) 98°C for 15 sec; 3) 60°C for 45 sec; 4) 72°C for 30 sec; 5) Repeat 2-4 for 30 cycles total; 6) 72°C for 1 min. Reactions were purified with KAPA Purebeads (0.8X cleanup) and eluted in 30  $\mu$ L of 10 mM Tris-HCl, pH 8.3. These reaction products were purified with KAPA pure beads (1X) and eluted in 30 µL 10 mM Tris-HCl, pH 8.3. This is an over-amplified product and likely contains heterodimers of library fragments. An aliquot (2 µL) was used for an additional 3 cycles of amplification with the same primers and conditions as above. A 1X KAPA Purebeads purification was performed and products were eluted in 30 µL TE buffer. The concentration was verified by absorbance, followed by the Bioanalyzer and qPCR through the McMaster Metagenomics Facility. For Trial 2, conditions were similar to the enrichment for Trial 1 libraries except for postenrichment which was performed using 7.5 µL of enriched library, 12.5 µL of KAPA Ready Mix 2X, 1 µL of 10X primer mix (1) 98°C 45 sec; 2) 98°C 30sec; 3) 60°C for 45sec; 4) 72°C for 45sec; 5) Repeat step 2 - 4 for 17 cycles; 6) 72°C for 5 min; 7) 4°C hold. PCR products were purified using KAPA Pure beads (1.5X) and eluted in 10 mM Tris-HCl pH 8.0. The concentration was determined via absorbance and additional library quality control was performed through the McMaster Metagenomics Facility (Bioanalyzer and qPCR). Products were sequenced via Illumina MiSeq 2 x 250 bp.



**Supplementary Figure 1: Probe and CARD nucleotide sequence statistics** A) Number of genes targeted by probes through mapping with Bowtie2. B) Number of probes targeting genes through mapping with Bowtie2. C) Percent length coverage of genes with probes. D) Mean depth of probe coverage across individual genes in CARD. E) Length of sequence targeted by probes in genes in CARD. F) GC content of probes. G) GC content of genes in CARD. H) Melt temperature of final list of

probes. Figures generated using Prism 8 for macOS.

Supplementary Table 1: Bacterial strains used in control experiments.

Clinical bacterial isolates obtained through the Wright Clinical Collection. Bacterial genomes were sequenced, and draft genome assemblies were analyzed through the Resistance Gene Identifier in CARD to predict the number of resistance genes. The total probeset was mapped against the draft assembled genome and the number of genes with probe coverage, percentage of genome covered by probes and overlap between predicted RGI genes and probe coverage were determined.

Bacterial strain	Genome size (Mb)	GC Content (%)	Predicted genes by RGI	Region predicted by RGI (%)	# Probe- targeted sites	Length of probe-targeted site (average and range)	Region with probe coverage (%)	RGI genes with probes	Region predicted by RGI and targeted by probes (%)
Escherichia coli C0002	5.29	50.62	67	1.64	65	797.75 (80 - 3595)	0.97	43	0.81
Klebsiella pneumoniae C0006	5.45	57.23	30	0.55	35	331.54 (80 - 877)	0.21	17	0.17
Staphylococcus aureus C0018	2.92	32.66	16	0.55	13	1127.54 (140 – 2013)	0.50	12	0.41
Staphylococcus aureus C0033	2.92	32.77	16	0.64	14	1143.07 (155 – 2130)	0.52	13	0.44
Klebsiella pneumoniae C0050	5.60	57.05	34	0.63	40	346.18 (80 - 900)	0.25	18	0.19
Pseudomonas aeruginosa C0060	6.80	66.19	53	1.18	48	933.35 (97 - 3415)	0.66	33	0.54
Escherichia coli C0094	5.22	50.74	67	1.65	64	779.86 (80 – 3003)	0.95	41	0.79
Pseudomonas aeruginosa C0292	6.81	66.21	54	1.17	48	938.71 (97 - 3415)	0.66	33	0.57

# Supplementary Table 2: Library and sequencing information.

The amount in nanograms of each library and the corresponding amount of probes used for enrichment. The average size of library fragments prior to enrichment was determined through the Agilent Bioanalyzer 2100. The number of clusters (paired-end reads) that were generated for each library when sequenced by Illumina's MiSeq V2 2x250. Blanks for each trial were included and sequenced on a separate run; many of the blank libraries did not generate peaks on the Bioanalyzer nor any signal by quantitative PCR therefore their values are N/A. In Phase 2, three positive controls for enrichment were included with genomic DNA from *Escherichia coli* C0002 and varying library and probe amounts.

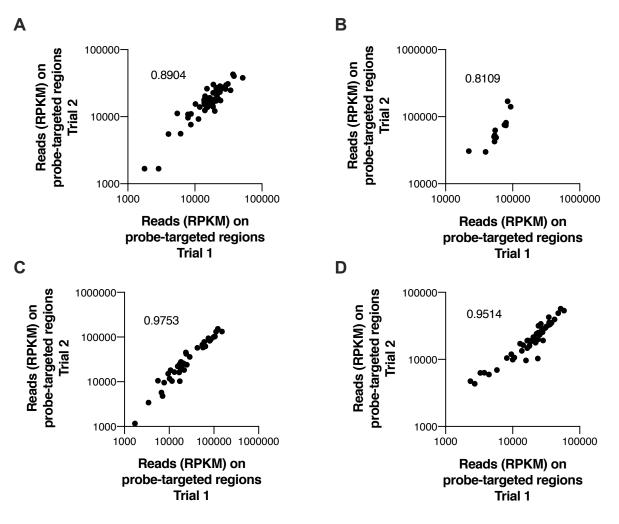
Phase	Trial/Set	Library	Amount of Probes (ng)	Amount of Library (ng)	Average Library Size (bp)	Clusters sequenced enriched	Clusters sequenced shotgun
		C0002	100	100	988	66926	
		C0018	100	100	994	75860	
		C0050	100	100	1222	73941	
	Trial 1	C0060	100	100	1225	81810	
		Pool 1	100	100	1257	61568	218008
		Pool 2	100	100	1158	61658	159059
		Pool 3	100	100	1216	58308	109194
Phase 1		Negative Control - Blank	100	N/A	632	170565	
r nase 1		C0002	100	100	435	99748	
		C0018	100	100	438	143804	
		C0050	100	100	416	153673	
	Trial 2	C0060	100	100	403	124971	
	111al Z	Pool 1	100	100	429	86023	29241
		Pool 2	100	100	413	124170	33488
		Pool 3	100	100	427	127682	32560
		Negative Control - Blank	100	N/A	345	44026	
		1 - 1	25	50	952	89768	
		1 - 2	50	50	968	77117	
		1 - 3	100	50	919	65746	
		1 - 4	50	100	1044	55783	
Phase 2	Set 1	1 - 5	100	100	972	64761	
Fliase 2	Set I	1 - 6	200	100	940	71099	3652948
		1 - 7	100	200	915	15211	4405779
		1 - 8	200	200	1020	59409	
		1 - 9	400	200	998	25911	
		Negative Control - Blank	50	N/A	276	2590	

Denitiere	C0002 – 1 - 1	100	50	986	80647	
Positive	C0002 – 1 - 2	50	50	939	116965	
Controls	C0002 – 1 - 3	25	50	976	112881	
	2 - 1	25	50	955	158710	
	2 - 2	50	50	887	100590	
	2 - 3	100	50	891	102689	
	2 - 4	50	100	902	120764	
G . + 2	2 - 5	100	100	956	141994	6151998
Set 2	2 - 6	200	100	941	159192	
	2 - 7	100	200	790	96211	
	2 - 8	200	200	944	129333	
	2 - 9	400	200	871	76195	7660355
	Negative Control - Blank	50	N/A	N/A	3804	
D	C0002 – 2 -1	100	33	993	139909	
Positive	C0002 – 2 - 2	50	50	935	235429	
Controls	C0002 – 2 - 3	25	50	876	129070	
	3 - 1	25	50	854	82778	5866495
	3 - 2	50	50	888	158968	
	3 - 3	100	50	910	65675	
	3 - 4	50	100	889	103671	
Set 3	3 - 5	100	100	882	78251	4213540
Set 5	3 - 6	200	100	943	68331	
	3 - 7	100	200	820	96722	
	3 - 8	200	200	934	79036	
	3 - 9	400	200	917	82375	
	Negative Control - Blank	50	N/A	N/A	5962	
Desitive	C0002 – 3 -1	100	38	846	54117	
Positive Controls	C0002 – 3 - 2	50	32	881	96258	
Controls	C0002 – 3 - 3	25	38	779	110746	

# Supplementary Table 3: Individual strain enrichment results.

Strains were enriched individually in two trials with different library sizes. For each strain the regions predicted to be targeted by probes were determined through mapping the probeset to each individual genome). Enrichment results across two trials were determined by mapping trimmed and filtered reads to genome, calculating the percentage on-target and normalizing reads and depth per kb per million reads for probe-targeted regions with at least 10 reads.

Strain	Average % mapping to genome	Average % mapping to probe- targeted sites	% of targeted regions with at least 10 reads	Average % coverage of targeted regions	Average reads per kb per million reads on probe- targeted region	Average depth per kb per million reads on probe- targeted region
Escherichia coli C0002	96.67 (±2.72)	95.07 (±1.54)	100	100	18975.73 (±414.91)	6192.13 (±297.27)
<i>Staphylococcus aureus</i> C0018	97.99 (±1.98)	94.89 (±2.31)	100	100	67615.06 (±4360.20)	19968.28 (±2670.37)
Klebsiella pneumoniae C0050	95.60 (±3.96)	85.74 (±4.68)	100	100	40531.43 (±2516.77)	17315.24 (±1630.66)
Pseudomonas aeruginosa C0060	91.45 (±5.49)	90.73 (±0.95)	100	100	22725.67 (±32.97)	6497.48 (±61.46)



Supplementary Figure 2: Consistency in library prep methods and trials.

Comparing read counts normalized in subsampled individual enrichment trials through different library prep methods. Reads from enrichment of individual genomes of *Escherichia coli* C0002 (A), *Staphylococcus aureus* C0018 (B), *Klebsiella pneumoniae* C0050 (C) and *Pseudomonas aeruginosa* C0060 (D) in Trial 2 were subsampled to same depth as reads in Trial 1. The reads were mapped to the respective bacterial genome, filtered for mapping quality and then the number of reads on each RGI and probe-targeted region were counted and normalized per kb per million reads. Pearson correlation coefficients are shown.

# Supplementary Table 4: Pooling of genomic DNA to create "mock metagenomes"

We pooled various nanogram amounts of genomic DNA from bacteria and estimated the percentage of each strain in the respective pools based on total genome size of each strain. With reads generated through shotgun sequencing and after enrichment, we calculated the percentage of reads mapping to a particular genome by mapping to a combined reference of the genomes used in a given pool and counting the reads that mapped to each respective genome (= reads mapping to genome A / reads mapping to all genomes). The percentage represented by targeted regions of each genome in each pool is determined by taking the percentage of each isolate's genome that is targeted by probes and the percentage of reads that map to that particular genome from the shotgun sequencing data.

sequenci	ng data.			% of reads	Targeted %	
D 1	сц. <sup>1</sup>	Amount of	Estimated %	mapping	of pool	% of reads
Pool	Strain	genomic DNA	of pool	from		mapping from enriched
		pooled (ng)	-	shotgun		from enriched
	C0002	312	21.98	24.82	0.24	52.55
Trial 1	C0018	312	40.00	12.06	0.060	32.12
Pool 1	C0050	312	20.74	27.18	0.068	8.86
	C0060	312	17.28	35.93	0.24	6.47
	C0002	112	18.77	22.30	0.22	33.95
Trial 2	C0018	174	53.01	65.29	0.33	62.88
Pool 1	C0050	106	16.79	4.39	0.011	1.54
	C0060	88	11.43	8.02	0.053	1.63
	C0002	1250	66.30	64.73	0.63	71.26
Trial 1	C0018	180	17.22	11.96	0.060	19.69
Pool 2	C0050	180	9.07	11.28	0.028	4.75
	C0060	180	7.41	12.03	0.079	4.30
	C0002	264	48.04	57.31	0.56	65.39
Trial 2	C0018	102	33.92	35.54	0.18	33.24
Pool 2	C0050	62	10.75	1.66	0.0042	0.44
	C0060	51	7.29	5.49	0.036	0.94
	C0002	125	11.01	13.91	0.13	38.50
	C0006	125	10.70	24.75	0.052	2.34
	C0018	125	19.88	6.54	0.033	11.62
Trial 1	C0033	125	19.88	11.59	0.060	22.81
Pool 3	C0050	125	10.40	12.75	0.032	2.73
	C0060	125	8.56	16.40	0.11	2.16
	C0094	125	11.01	6.90	0.066	18.78
	C0292	125	8.56	7.15	0.047	1.07
	C0002	46	8.65	9.84	0.095	14.80
	C0006	83	8.16	14.44	0.030	1.53
	C0018	43	28.17	11.49	0.057	12.49
Trial 2	C0033	36	28.15	34.36	0.18	34.58
Pool 3	C0050	45	7.68	0.60	0.0015	0.13
	C0060	83	5.20	2.02	0.013	0.42
	C0094	46	8.78	25.21	0.24	35.67
	C0292	36	5.21	2.04	0.013	0.39

# Supplementary Table 5: Enrichment results to probe-targeted regions in pooled samples

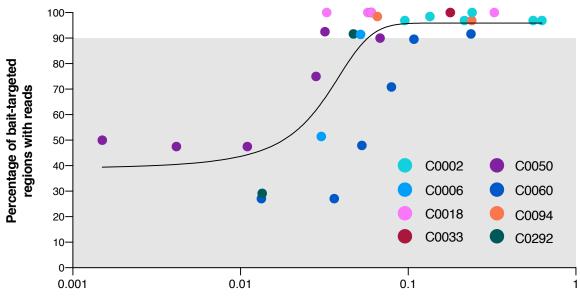
Genomic DNA from individual strains was pooled in various ratios to produce "mock metagenomes" for enrichment. For each strain, the regions predicted be targeted by probes (determined through mapping the probeset to each individual genome) are considered the targeted region for analysis. Trimmed and filtered reads from paired enriched and shotgun pools were subsampled to same read depth. The resulting reads were mapped to the individual strain's genomes, counted on-target and normalized per kb per million reads mapping. Percentage on-target, percentage of probe-targeted regions with at least 10 reads as well as their percent coverage, average reads, and average depth were determined for each strain at the probe-targeted region level. The fold enrichment is based on all genes regardless of read counts.

Sample	Strain	% of reads in Pool	% Mapping to probe- targeted regions	% of probe- targeted regions with reads	% coverage of probe- targeted regions	Average reads per kb per million reads	Average depth per kb per million reads	Fold-enrichment in reads (average and range)
	C0002	52.75	93.06	100	100	19097.95	6091.42	810.18 (2.66 - 16590.95)
Trial 1	C0018	20.05	94.84	100	100	67393.09	19715.42	135.84 (31.11 – 291.78)
Pool 1 Enriched	C0050	18.73	85.44	90	100	41944.82	16304.97	1341.88 (3.77 – 23020.26)
	C0060	3.40	90.26	91.67	98.73	24920.46	6697.48	994.87 (0 - 21945.61)
	C0002	21.61	1.56	18.46	90.13	671.09	153.52	
Trial 1 Pool 1	C0018	10.32	0.70	15.38	88.03	820.59	161.15	
Shotgun	C0050	23.56	0.82	25.00	100	762.34	190.87	
	C0060	28.70	0.81	12.50	84.54	301.55	44.92	
	C0002	35.84	98.90	96.92	100	20081.94	6630.47	4972.95 (2.84 - 35942.31)
Trial 2 Pool 1	C0018	56.55	98.56	100	100	74814.49	24542.74	144.41 (41.36 – 332.17)
Enriched	C0050	7.72	97.63	47.50	99.75	74609.44	24141.06	18991.42 (0 - 170582.07)
	C0060	1.31	93.37	47.92	83.22	30865.24	7310.50	17166.87 (0 - 70414.91)
	C0002	23.52	1.49	1.54	91.65	471.34	30.86	
Trial 2 Pool 1	C0018	57.30	0.71	76.92	79.03	570.56	98.30	
Shotgun	C0050	5.19	0.88	0	0	0	0	
	C0060	6.65	0.65	0	0	0	0	
	C0002	68.39	77.35	96.92	100	15928.54	4982.54	57.09 (2.57 - 192.18)
Trial 1 Pool 2	C0018	12.69	79.11	100	100	56570.38	16316.11	2614.81 (15.93 - 32565.71)
Enriched	C0050	12.61	74.13	75.00	99.93	41711.08	15702.37	2727.71 (0-39495.86)
	C0060	2.34	38.95	70.83	96.27	11523.24	2820.94	2382.94 (0 - 19387.19)
	C0002	58.69	1.34	58.46	96.92	321.15	81.43	
Trial 1 Pool 2	C0018	10.64	0.74	30.77	78.51	896.24	141.82	
Shotgun	C0050	11.48	1.33	20	100	1745.41	464.15	
	C0060	9.72	0.75	2.08	56.38	266.69	18.15	

	C0002	65.64	98.29	96.92	100	19970.52	6708.67	1190.08 (7.74 – 29085.20)
Trial 2 Pool 2	C0018	28.13	98.15	100	100	75034.93	24899.52	210.58 (32.41 - 596.02)
Enriched	C0050	10.26	98.23	47.50	100	77537.34	26906.17	8270.19 (0 - 50937.25)
	C0060	0.73	88.86	27.08	78.56	37440.00	8936.77	$18933.20 \ (0 - 106732.35)$
	C0002	56.47	1.38	20.00	73.49	404.35	72.86	
Trial 2 Pool 2	C0018	29.19	0.57	23.08	73.76	698.55	125.73	
Shotgun	C0050	3.01	4.51	2.50	79.03	10409.44	2093.37	
	C0060	4.27	0.73	0	0	0	0	
	C0002	38.74	94.12	98.46	100	19755.27	6312.06	2493.04 (3.05 - 22767.27)
	C0006	13.66	84.08	91.43	100	51010.68	22066.06	3295.94 (0 - 61249.67)
	C0018	29.65	95.22	100	100	63154.77	15991.26	2909.12 (54.61 - 35638.08)
Trial 1 Pool 3	C0033	33.17	94.82	100	100	56232.72	13178.66	156.78 (28.17 - 314.91)
Enriched	C0050	14.84	85.22	92.5	100	43478.45	18486.32	2475.78 (4.87 - 47799.65)
	C0060	2.45	91.97	89.58	98.78	26022.10	7430.52	3742.84 (3.65 - 62302.44)
	C0094	35.52	92.59	98.44	100	19949.59	6561.88	3526.16 (2.48 - 23220.26)
	C0292	2.78	84.96	91.67	99.29	28432.58	10574.24	4014.72 (0 - 54962.31)
	C0002	9.83	1.63	3.08	88.69	1449.60	308.97	\ /
	C0006	25.19	0.36	8.57	95.63	3450.36	1206.18	
	C0018	11.94	0.51	7.69	68.26	413.96	50.49	
Trial 1 Pool 3	C0033	12.81	0.59	7.14	68.25	424.67	47.08	
Shotgun	C0050	24.09	0.48	12.5	93.25	853.91	300.04	
C	C0060	17.84	0.90	4.17	64.69	222.28	16.28	
	C0094	8.25	1.67	3.125	88.69	1726.91	368.08	
	C0292	16.78	0.94	4.17	64.69	1141.24	84.87	
	C0002	32.65	98.09	96.92	99.97	20307.57	6847.06	7369.15 (4.14 – 66339.3)
	C0006	7.75	90.49	51.43	99.50	86220.71	36708.00	25683.46(0 - 271673.69)
	C0018	45.46	97.45	100	100	65485.29	17173.26	5819.09 (29.42 - 74023.04)
Trial 2 Pool 3	C0033	52.11	97.53	100	100	58846.80	13719.18	698.58 (72.34 - 8084.37)
Enriched	C0050	8.22	92.65	50.00	99.55	74207.10	29767.85	21813(0-256173.72)
	C0060	0.86	90.00	27.08	79.68	39544.66	8226.37	16172.91 (0 - 70505.29)
	C0094	34.91	97.65	96.87	100	20612.44	7021.48	7479.75 (2.67 – 61794.38)
	C0292	0.89	89.30	29.17	80.95	44281.92	13985.84	18128.93 (0 - 120321.02)
	C0002	16.88	1.38	0	0	0	0	
	C0006	15.36	0.47	0	0	0	0	
Trial 2 Pool 3	C0018	41.07	0.70	38.46	73.84	525.28	55.49	
Shotgun	C0033	44.54	0.79	50.00	77.22	703.43	113.13	
	C0050	12.76	0.64	0	0	0	0	
	_ 20000	12.70	0.01	v	v	0	0	

C0060	4.54	0.77	0	0	0	0
C0094	21.50	1.23	1.56	68.13	404.24	25.04
C0292	4.59	0.86	0	0	0	0

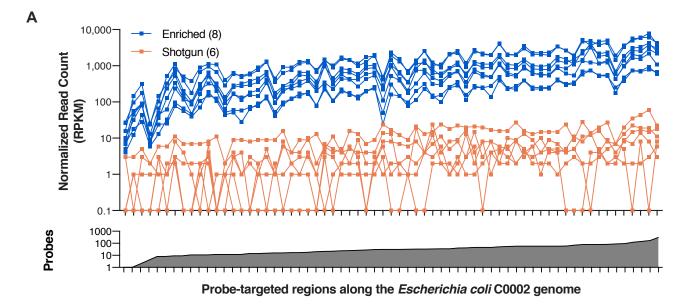
<u>U</u>

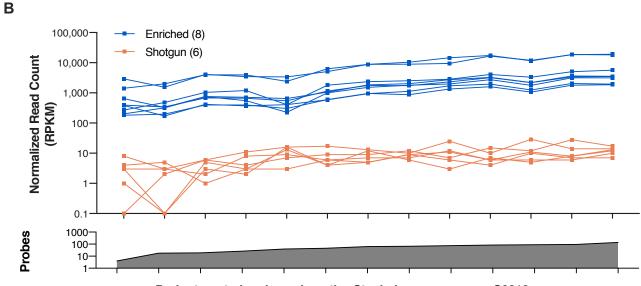


Percentage of mock metagenome represented by targeted regions within genome

Supplementary Figure 3

**Supplementary Figure 3: Sensitivity in recovery of bait-targeted regions with reads.** <u>Using</u> <u>the estimated percentage of a pool represented by each isolate's genome from the percentage of reads mapping in the shotgun dataset, the estimate percentage of the mock metagenome represented by that isolate's bait-targeted complement was determined. This represents the limit of detection or sensitivity for each isolate/genome tested. The Y-axis is the percentage of probe-targeted regions of a given isolate's genome that were successfully captured/recovered with at least 10 reads after filtering.</u>





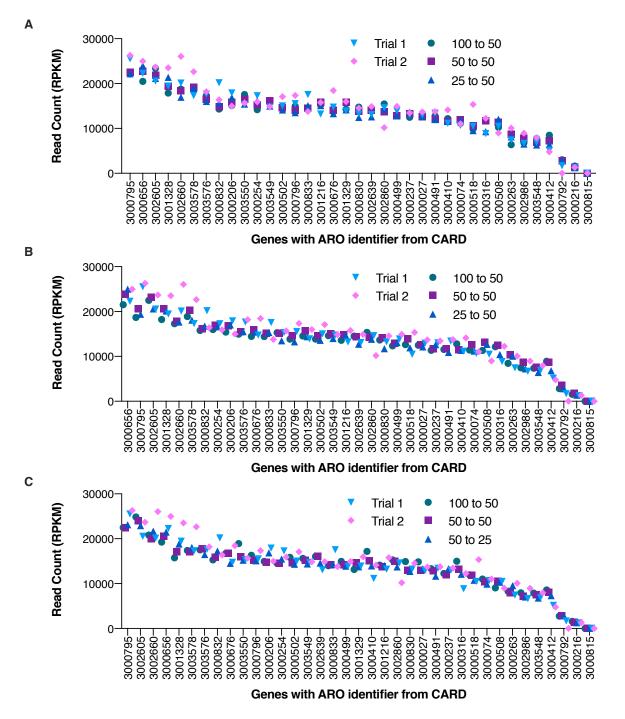
Probe-targeted regions along the Staphylococcus aureus C0018 genome

**Supplementary Figure 4: Enrichment results in higher read counts on antibiotic resistance genes compared to shotgun sequencing.** Raw read counts at each probe-targeted region within the *Escherichia coli* C0002 genome (A) and *Staphylococcus aureus* C0018 genome (B) in enriched and shotgun samples including individual and "mock metagenomes" of multiple strains. Among enriched and shotgun pairs, reads were subsampled to equal depths and mapped to the individual strain's genome. The predicted number of probes for each region along the genome are shown in the panels below. The Y axes are in the logarithmic scale.

# Supplementary Table 6: Control enrichment with *Escherichia coli* C0002.

Enrichment results from the positive control of *E. coli* C0002 control used in Phase 2. Trimmed and deduplicated reads were mapped to CARD using RGIBWT, filtered by genes with probe coverage, an average read mapping quality  $\geq 11$ , and percent length coverage of a gene with reads  $\geq 80\%$ .

	Probes (ng)	Library (ng)	% reads mapping to CARD	Total number of genes	Genes with map quality >=11	Genes with probes	Genes with length coverage with reads >=80%	Genes with probes and map quality >=11	Genes passing all filters
C0002	25	50	63.52	164	51	53	86	39	36
– Set 1	50	50	64.81	164	54	53	84	39	36
– Set 1	100	50	63.75	154	53	53	80	40	36
C0002 – Set2	25 50	50 50	61.10 65.77	179 195	62 60	54 59	82 84	42 44	36 36
	100	33	60.31	170	59	57	87	42	36
C0002	25 50	38 32	65.46 65.77	182 172	58 58	57 53	86 88	39 40	36 36
– Set 3	100	38	67.98	147	58 54	56	83	40	36



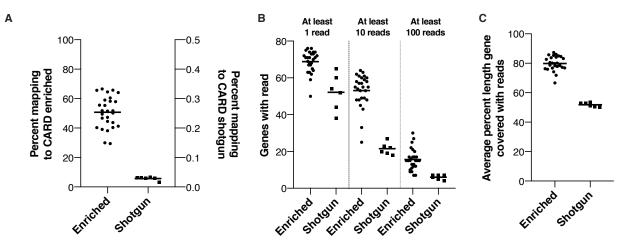
### Supplementary Figure 5: Genes identified in positive control enrichments.

Normalized read counts from C0002 control enrichments from *rgi bwt* results from 3 samples in each Set (A) Set 1, B) Set 2, C) Set 3) to the two trials of individual enrichment. Genes with reads were filtered based on read mapping quality greater than or equal to 11, percent length coverage of a gene with reads greater than or equal to 80% and genes with probes mapping. Reads were normalized per kb of reference gene per million reads mapping. Genes are ordered by sum of read counts from highest to lowers (left to right) with the ARO identifier shown along the X axis

# Supplementary Table 7: Phase 2 enrichment results with the full number of reads.

For the enriched samples, trimmed and deduplicated reads were mapped to CARD using RGIBWT, filtered by genes with at least 10 reads, those with probes, an average read mapping quality  $\geq 11$ , and length coverage of a gene with reads  $\geq 10\%$ . For the shotgun samples, trimmed and deduplicated reads were mapped to CARD using RGIBWT, filtered by genes with an average read mapping quality  $\geq 11$  and read length coverage of a gene  $\geq 10\%$ . EN = enriched, UN = shotgun.

Samul	Probes (ng)	Library (ng)	Reads mapping to CARD (%)	Total number of genes	Genes with read map quality >=11	Genes with probes	Genes with read length coverage >=10%	Genes passing all filters
Sampi	le Set 1 25	50	55.36	60	50	51	58	48
	23 50	30 50	65.73	60 62	50 54	52	58 60	48 49
	100	50	55.59	60	50	52 50	60	49 48
	50	100	65.63	56	30 47	30 46	55	48
EN	100	100	51.85	50 61	51	40 51	60	48
LIN	200	100	58.21	64	56	53	61	48 49
	100	200	51.52	34	26	27	34	25
	200	200	66.57	60	20 50	48	59	45
	400	200	49.44	45	30 37	36	43	33
	200	100	0.030	26	19		24	18
UN	100	200	0.030	20 32	22	N/A	29	20
Sampl	le Set 2	200	01020	52		1011		20
	25	50	64.07	78	67	64	76	61
	50	50	64.60	72	64	61	71	58
	100	50	57.96	75	64	61	74	57
	50	100	46.75	78	66	66	76	62
EN	100	100	58.99	79	69	64	77	61
	200	100	44.52	85	72	69	80	63
	100	200	60.43	76	66	62	73	59
	200	200	47.27	82	71	67	81	64
	400	200	41.22	70	59	58	69	55
UNI	400	200	0.016	41	28	N/A	37	27
UN	100	100	0.032	34	24	N/A	32	23
Sampl	le Set 3							
	25	50	50.16	72	63	61	70	58
	50	50	38.19	79	66	64	76	60
	100	50	51.73	69	59	59	68	55
	50	100	29.46	78	66	63	76	60
EN	100	100	40.28	74	65	60	72	57
	200	100	39.06	67	57	57	67	53
	100	200	29.97	69	57	58	68	54
	200	200	40.32	72	60	58	71	55
	400	200	43.74	69	58	56	67	53
UN	100	100	0.031	29	19	N/A	26	19
011	25	50	0.031	34	23	N/A	30	22



**Supplementary Figure 6: Comparing enriched and shotgun ARG recovery.** For the enriched and shotgun samples, the full number of reads for each sample were mapped to CARD using *rgi bwt*. A) The percentage of reads mapping to CARD. B) Genes were counted with at least 1, 10 and 100 reads and filtered for mapping quality (>=11), percent coverage by reads (>=10) and probes mapping (only for the enriched samples). C) The average percent coverage of all genes with at least 10 reads in each sample after the same filters used in B.

### Supplementary Table 8: Phase 2 enrichment results with subsampled reads.

For the enriched samples, reads were subsampled to 22,324 reads and mapped to CARD using RGIBWT. Results were filtered by genes with at least 10 reads, those with probes, an average read mapping quality  $\geq 11$ , and length coverage of a gene with reads  $\geq 10\%$ . For the shotgun samples, reads were subsampled to their paired enriched sample and mapped to CARD using RGIBWT. Results were filtered by genes with an average read mapping quality  $\geq 11$  and read length coverage of a gene  $\geq 10\%$ . EN = enriched, UN = shotgun.

Samula	Probes (ng)	Library (ng)	Reads mapping to CARD (%)	Total number of genes	Genes with read map quality >=11	Genes with probes	Genes with read length coverage >=10%	Genes passing all filters
Sample	25	50	55.24	34	26	27	34	25
	23 50	50 50	65.84	34 39	20 31	31	34	23
	100	50	56.11	46	37	37	45	34
	50	100	66.01	39	32	32	39	30
EN	100	100	51.94	40	32	32	37	28
	200	100	57.93	38	30	30	37	28
	100	200	51.52	34	26	27	34	25
	200	200	66.99	42	34	33	39	30
	400	200	49.39	33	26	26	33	24
UN	200	100	0.038	2	2	N/A	2	2
UN	100	200	0.054	0	0	N/A	0	0
Sample	e Set 2							
EN	25	50	64.25	41	33	34	40	32

	50	50	64.11	43	36	35	40	31
	100	50	58.80	43	36	35	43	33
	50	100	46.95	40	32	33	38	29
	100	100	59.13	42	35	34	41	31
	200	100	44.64	45	35	34	41	31
	100	200	60.55	50	42	42	49	39
	200	200	47.29	45	38	37	45	35
	400	200	41.56	43	34	35	41	32
UN	400	200	0.029	1	1	N/A	1	1
UN	100	100	0.035	2	2	N/A	2	2
Sample Set 3								
	25	50	50.64	37	29	30	36	27
	50	50	37.85	27	19	20	27	18
	100	50	51.41	36	27	28	33	24
	50	100	29.56	29	21	22	28	20
EN	100	100	40.77	34	26	26	33	24
	200	100	38.86	37	30	30	37	28
	100	200	30.08	31	23	24	30	21
	200	200	40.62	34	26	26	32	23
	400	200	44.35	37	30	29	35	26
UN	100	100	0.023	0	0	N/A	0	0
UIN	25	50	0.023	1	1	N/A	1	1

### Supplementary Table 9: Phase 2 overlapping genes with the full number of reads.

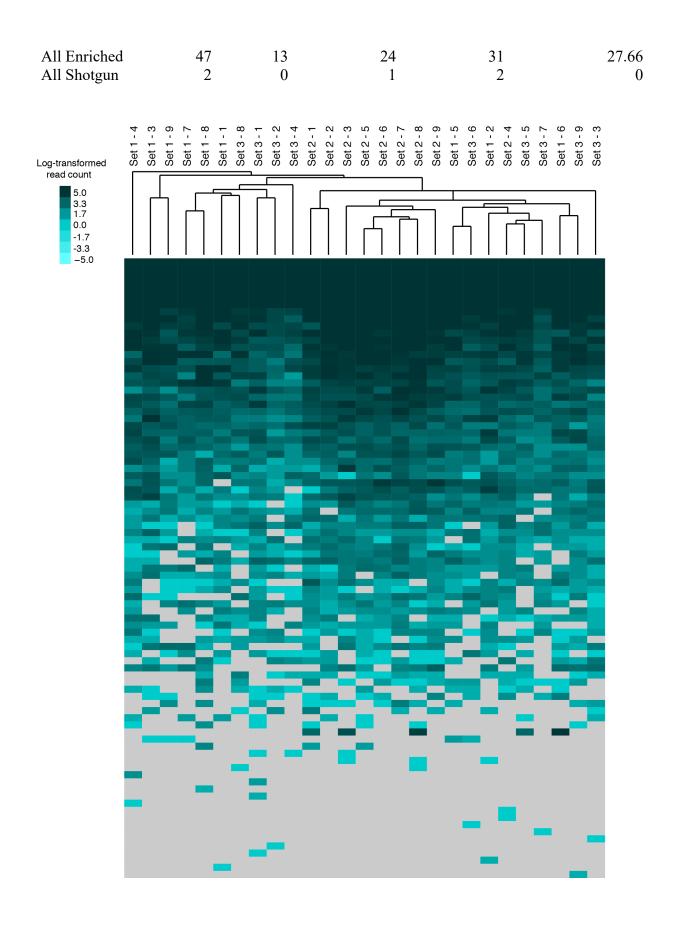
We calculated the overlap of genes with at least 10 reads passing the percent length coverage by reads ( $\geq$ =10%), average read mapping quality ( $\geq$ =11) and probe mapping (except for shotgun libraries) filters.

Samples	Total genes	Genes found in all	Genes found in 2/3 or more	Genes found in 1/3 or more	Overlap in All Samples (%)
Set 1 Enriched	62	24	38	53	38.71
Set 2 Enriched	68	50	57	64	73.53
Set 3 Enriched	70	41	53	60	58.57
All Enriched	70	24	52	60	34.28
All Shotgun	32	16	18	28	50.00

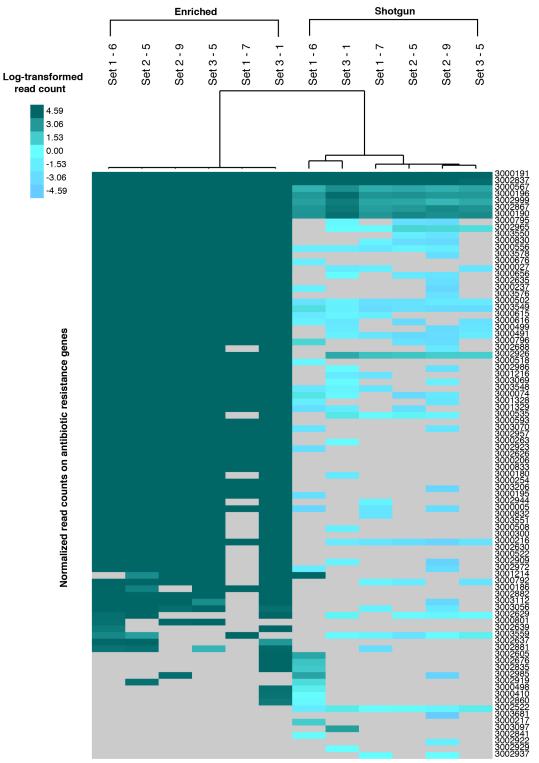
#### Supplementary Table 10: Phase 2 overlapping genes with subsampled reads.

Libraries were subsampled to the same number of reads within sets and overall (22,324 reads). Shotgun libraries were subsampled to the same number of reads as the lowest enriched library overall. Resulting genes with at least 10 reads were filtered for percent coverage by reads ( $\geq$ =10%), average mapping quality ( $\geq$ =11) and probe mapping (except for the shotgun samples).

Samples	Total genes	Genes found in all	Genes found in 2/3 or more	Genes found in 1/3 or more	Overlap in All Samples (%)
Set 1 Enriched	38	16	26	32	42.10
Set 2 Enriched	45	22	30	36	48.89
Set 3 Enriched	37	13	20	26	35.14



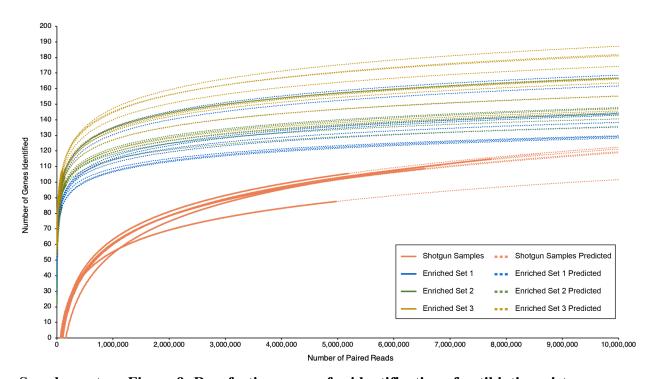
**Supplementary Figure 7: Hierarchical clustering of enriched libraries**. Enriched reads from 27 libraries were subsampled to 22,324 reads, mapped to CARD through *rgi bwt*. The reads were mapped to CARD through *rgi bwt* and filtered for genes with probes mapping, with greater than or equal to 10% length coverage by reads and an average read mapping quality >=11. Read counts were log-transformed and combined into a heatmap ordered by average read counts across the 27 enriched samples.



Supplementary Figure 8: Hierarchical clustering of enriched and shotgun libraries.

The full number of reads from the 6 enriched and shotgun pairs were mapped to CARD through *rgi bwt*. The results were filtered for genes with greater than or equal to 10% read length coverage and an average read mapping quality greater  $\geq 11$ . Read counts were normalized by kb of gene and reads available for mapping, log-transformed, and combined into a heatmap. Genes are

ordered by sum of read counts. ARO numbers from CARD are shown on the right-hand side of the heatmap.



**Supplementary Figure 9: Rarefaction curves for identification of antibiotic resistance genes.** The AmrPlusPlus Rarefaction Analyzer was used with subsampling every 1% of the total reads and a gene read length of at least 10% to identify antibiotic resistance genes. The solid lines show individual sequencing experiments and the dotted lines are the logarithmic extrapolations beyond the experimental sequencing depth.

Supplementary Table 11: Sequencing reads identified in the Blank samples. Enriched negative control blank libraries were
sequenced on separate MiSeq 2 x 250 runs. After de-multiplexing, we pulled the reads that were associated with various index
combinations used alongside the Blank Negative control throughout library preparation within the same trials and sets.

Sample	Samples processed alongside the blank library	Number of paired reads sequenced on run with Blank	Percentage of Blank
	C0002	1575	0.92
	C0018	0	0.00
	C0050	435	0.26
Blank	C0060	379	0.22
Trial 1	Pool1	3064	1.80
	Pool2	110959	65.05
	Pool3	36390	21.33
	Additional barcodes	2487	1.46
	Blank	15276	8.96
	C0002	6611	15.02
	C0018	11763	26.72
	C0050	5194	11.80
	C0060	4491	10.20
Blank	Pool1	1178	2.68
Trial 2	Pool2	4800	10.90
	Pool3	5862	13.31
	Additional barcodes	3044	6.91
	Blank	1083	2.46
	1 - 1	456	17.61
	1 - 2	94	3.63
	1 - 3	174	6.72
	1 - 4	101	3.90
	1 - 5	316	12.20
Blank Set 1	1 - 6	82	3.17
Dialik Set 1	1 - 7	683	26.37
	1 - 8	173	6.68
	1 - 9	35	1.35
	Negative Control - Blank	28	1.08
	C0002 – 1 - 1	120	4.63
	C0002 – 1 - 2	37	1.43

	C0002 – 1 - 3	291	11.24
	2 - 1	367	9.65
	2 - 2	22	0.58
	2 - 3	44	1.16
	2 - 4	119	3.13
	2 - 5	40	1.05
	2 - 6	0	0.00
Blank Set 2	2 - 7	39	1.03
	2 - 8	271	7.12
	2 - 9	137	3.60
	Negative Control - Blank	530	13.93
	C0002 - 2 - 1	207	5.44
	C0002 – 2 - 2	34	0.89
	C0002 – 2 - 3	1994	52.42
	3 - 1	224	3.76
	3 - 2	286	4.80
	3 - 3	71	1.19
	3 - 4	1653	27.73
	3 - 5	282	4.73
	3 - 6	23	0.39
Blank Set 3	3 - 7	42	0.70
	3 - 8	128	2.15
	3 - 9	1198	20.09
	Negative Control - Blank	0	0.00
	C0002 - 3 - 1	161	2.70
	C0002 – 3 - 2	817	13.70
	C0002 – 3 - 3	1077	18.06

Supplementary Table 12: Negative control enrichment with Blank samples. Enriched reds were divided among index combinations used during the respective Phase, Trial or Set (Supplementary Table 7). The reads belonging to each Negative Control – Blank library were trimmed and duplicates were removed then mapped to CARD through rgibwt. The number of genes with 1, at least 10 and at least 100 reads as well as genes with probes mapping, with average read mapping quality  $\geq 11$  and gene length coverage with reads  $\geq 10\%$  are shown. In Phase 2 Set 1, raw sequencing reads were used for analysis, in Set 2, deduplication was omitted, and for Set 3, there were no reads associated with the Blank indexes after sequencing.

Sample	Paired reads	Paired reads after trimming and de- duplication	Percent of reads mapping to CARD	Total genes with reads	Genes with 10 or more reads	Genes with 100 or more reads	Genes with at least 10 reads, >10% read coverage, MQ >=11 and probes			
Blank Phase 1 Trial 1	15276	2716	80.34	153	82	9	10: cpxA, mefA, arlS, mdtO, mdtE, mdtN, acrD, armA, AAC(3)-IV, APH(7'')-Ia,			
Blank Phase 1 Trial 2	1083	341	97.21	106	9	1	0			
Phase 2 Set 1			0	0	0	0	0			
Phase 2 Set 2*	530	412	76.46	94	26	0	<b>19</b> : APH(3'')-Ib, acrD, acrE, acrF, acrS, cpxA, dfrA17, emrK, emrY, eptA, evgS, mdtE, mdtF, mdtH, mdtO, mdtP, pmrF, tetQ, tolC			
Phase 2 Set 3	0	0	0	0	0	0	0			

\*Genes in Set 2 Blank found in enriched and shotgun libraries: *tetQ, acrF*. Genes found in blank and enriched: *acrD, acrE, acrS, cpxA, emrK, emrY, eptA, evgS, mdtE, mdtF, mdtH, mdtO, mdtP, pmrF, tolC*. Genes found in blank only: *APH(3'')-Ib, dfrA17,* 

**Supplementary Table 13: Genes identified through metagenomic analysis of enriched and shotgun samples.** Combining raw read counts across all 27 enriched and 6 shotgun sample at the full number of genes with the breakdown of gene, class and mechanisms identified. Genes were filtered based on genes with at least 10 reads mapping, percent coverage greater than or equal to 10%, mapping

quality greater than or equal to 11 and probes mapping (only for the enriched samples). This table is split into 4 parts with each part corresponding to a group of samples (Set 1, Set 2, Set 3 and the Shotgun samples). The first 6 columns are the same in all four parts.

ARO	Name	Class of Resistance	Present in Enriched	Present in Shotgun	Baits	Set 1 - 3	Set 1 - 4	Set 1 - 7	Set 1 - 6	Set 1 - 9	Set 1 - 8	Set 1 - 5	Set 1 - 2	Set 1 - 1
3000190	tetO	tetracycline antibiotic	27	6	Yes	2240	2088	655	3095	1195	2459	2613	2472	2909
3000191	tetQ	tetracycline antibiotic tetracycline	27	6	Yes	2174 7	21337	7489	30223	13830	27383	22368	25974	25651
3000196	tet32	antibiotic tetracycline	27	6	Yes	5306	4929	1610	7133	2788	6253	5760	5554	6339
3000567	tet(40)	antibiotic lincosamide	27	6	Yes	4375	3252	978	5835	1891	4454	4654	3774	4098
3002837	lnuC	antibiotic diaminopyrimid	27	6	Yes	2403	2223	828	2740	1202	2523	2240	2590	2884
3002867	dfrF	ine antibiotic	27	6	Yes	1093	1242	412	1185	485	1126	1232	1296	1770
3002999	cblA-1	cephalosporin glycopeptide	27	6	Yes	2531	2026	743	3297	1182	2927	2612	2258	2268
3002926	vanRG	antibiotic tetracycline	25	6	Yes	16	15	0	39	0	20	24	10	22
3000194	tetW	antibiotic macrolide antibiotic; lincosamide antibiotic; streptogramin	0	6	No	0	0	0	0	0	0	0	0	(
3000375	ermB Nocardia rifampin resistant beta- subunit of RNA polymerase	antibiotic peptide antibiotic; rifamycin	0	6	No	0	0	0	0	0	0	0	0	
3000501	(rpoB2)	antibiotic aminocoumarin	0	6	No	0	0	0	0	0	0	0	0	(
3002522	novA aac(6')-Ie-	antibiotic aminoglycoside	0	6	Yes	0	0	0	0	0	0	0	0	(
3002597	aph(2")-Ia Streptomyces rishiriensis <i>parY</i> mutant conferring resistance to	antibiotic	0	6	No	0	0	0	0	0	0	0	0	
	aminocoumari	aminocoumarin												
3003318	n	antibiotic	0	6	No	0	0	0	0	0	0	0	0	

3003730	Bifidobacteria intrinsic <i>ileS</i> conferring resistance to mupirocin <i>Campylobacte</i> <i>r coli</i> chlorampheni col	mupirocin	0	6	No	0	0	0	0	0	0	0	0	0
	acetyltransfer	phenicol												
3004454	ase	antibiotic glycopeptide	0	6	No	0	0	0	0	0	0	0	0	0
3002965	vanWG	antibiotic macrolide	26	4	Yes	50	25	0	41	24	74	48	52	57
3000535	macB	antibiotic aminoglycoside	22	4	Yes	26	0	0	107	0	43	56	0	29
3002647	aph(3')-IIIa	antibiotic tetracycline	0	3	No	0	0	0	0	0	0	0	0	0
3000556	tet44	antibiotic macrolide antibiotic; fluoroquinolone antibiotic; tetracycline antibiotic; phenicol	27	2	Yes	82	111	28	90	27	111	101	91	144
3003056	smeE	antibiotic glycopeptide	8	2	Yes	16	0	0	13	0	10	0	0	0
3002937	vanSG	antibiotic aminoglycoside	0	2	Yes	0	0	0	0	0	0	0	0	0
3002983	<i>amrB</i> <i>cepA</i> beta-	antibiotic	0	2	No	0	0	0	0	0	0	0	0	0
3003559	lactamase	cephalosporin tetracycline	0	2	Yes	0	0	0	0	0	0	0	0	0
3004032	tetA(46)	antibiotic tetracycline	0	2	No	0	0	0	0	0	0	0	0	0
3004033	tetB(46)	antibiotic macrolide antibiotic; monobactam; tetracycline antibiotic; aminocoumarin	0	2	No	0	0	0	0	0	0	0	0	0
3004074	muxB	antibiotic macrolide antibiotic; fluoroquinolone antibiotic;	0	2	No	0	0	0	0	0	0	0	0	0
3004144	axyY	aminoglycoside	0	2	No	0	0	0	0	0	0	0	0	0

#### antibiotic; cephalosporin

		fluoroquinolone antibiotic; cephalosporin;												
3000502	acrF	cephamycin; penam aminocoumarin	27	1	Yes	190	107	28	181	51	140	141	127	130
3000793	mdtB	antibiotic aminocoumarin	0	1	No	0	0	0	0	0	0	0	0	0
3000794	mdtC	antibiotic	0	1	No	0	0	0	0	0	0	0	0	0
3003097	cfxA6	cephamycin fluoroquinolone	0	1	Yes	0	0	0	0	0	0	0	0	0
3000027	emrA	antibiotic macrolide antibiotic; fluoroquinolone antibiotic; cephalosporin; glycylcycline; cephamycin; penam; tetracycline antibiotic; aminocoumarin antibiotic; rifamycin antibiotic; phenicol antibiotic;	27	0	Yes	49	51	20	40	26	51	39	37	38
3000237	tolC	triclosan aminoglycoside	27	0	Yes	39	36	28	57	26	102	53	53	94
3000491	acrD	antibiotic macrolide	27	0	Yes	83	66	24	173	36	111	143	83	88
3000615	mefA	antibiotic macrolide antibiotic; streptogramin	27	0	Yes	57	27	13	55	27	68	33	36	43
3000616	mel	antibiotic macrolide antibiotic; fluoroquinolone antibiotic;	27	0	Yes	28	11	13	50	11	36	53	69	73
3000795	mdtE	penam	27	0	Yes	92	64	12	173	38	78	125	56	96

		macrolide antibiotic; fluoroquinolone antibiotic;												
3000796	mdtF	penam aminoglycoside	27	0	Yes	144	102	22	223	76	110	94	102	131
		antibiotic; aminocoumarin												
3000830	cpxA	antibiotic macrolide antibiotic; fluoroquinolone antibiotic;	27	0	Yes	93	40	12	97	49	56	66	54	49
		penam; tetracycline												
3000833	evgS	antibiotic fluoroquinolone	27	0	Yes	46	55	11	49	28	42	27	18	19
3001216	mdtH	antibiotic tetracycline antibiotic;	27	0	Yes	23	55	11	73	19	35	35	66	23
	Escherichia	benzalkonium chloride;												
3001328	coli mdfA	rhodamine nucleoside antibiotic;	27	0	Yes	44	28	11	22	17	42	37	19	32
3003549	mdtO	acridine dye nucleoside	27	0	Yes	75	91	20	104	36	93	112	79	118
3003550	mdtP	antibiotic; acridine dye peptide	27	0	Yes	73	44	34	118	53	83	74	74	57
3003576	eptA	antibiotic	27	0	Yes	59	76	16	112	30	65	65	68	91
3003578	pmrF	peptide antibiotic fluoroquinolone	27	0	Yes	68	25	11	71	30	42	46	53	47
3000074	emrB	antibiotic fluoroquinolone antibiotic; cephalosporin;	26	0	Yes	42	15	0	36	15	29	48	43	19
3000499	acrE	cephamycin; penam macrolide antibiotic; fluoroquinolone	26	0	Yes	68	37	0	76	24	40	66	56	48
3000518	crp	antibiotic; penam	26	0	Yes	31	10	0	47	17	28	21	16	10

		fluoroquinolone antibiotic; cephalosporin; glycylcycline; cephamycin; penam; tetracycline antibiotic; rifamycin antibiotic; phenicol antibiotic;												
3000656	acrS	triclosan aminoglycoside	26	0	Yes	23	28	0	36	26	18	16	11	37
3002635	aph(2")-IIa	antibiotic nucleoside antibiotic;	26	0	Yes	59	31	15	65	0	35	29	51	46
3003548	mdtN	acridine dye tetracycline	26	0	Yes	57	40	0	33	15	18	24	17	11
3000254	emrY	antibiotic	25	0	Yes	40	17	0	25	18	26	14	38	37
3001329	mdtG	fosfomycin peptide	25	0	Yes	24	38	0	36	0	12	30	60	12
3002986	bacA	antibiotic fluoroquinolone antibiotic; cephalosporin; glycylcycline; penam; tetracycline antibiotic; rifamycin antibiotic; phenicol antibiotic;	25	0	Yes	27	33	0	35	0	42	20	15	17
3000216	acrB	triclosan phenicol	24	0	Yes	13	13	0	35	0	25	14	16	0
3002688	catS	antibiotic tetracycline	24	0	Yes	0	13	0	14	0	25	24	14	21
3000195	tetB(P)	antibiotic lincosamide antibiotic; streptogramin antibiotic; pleuromutilin	23	0	Yes	15	0	0	15	0	0	19	19	15
3000300	lsaA	antibiotic	22	0	Yes	22	0	0	0	14	17	11	12	13

		macrolide antibiotic; fluoroquinolone antibiotic; cephalosporin; cephamycin; penam;												
		tetracycline												
3000676	H-NS	antibiotic	22	0	Yes	0	0	0	18	0	11	19	18	0
3003070	vanXD	glycopeptide antibiotic	22	0	Yes	25	19	0	22	0	0	19	0	0
3003070	VanAD	tetracycline	22	0	res	23	19	0	22	0	0	19	0	0
3000180	tetA(P)	antibiotic	21	0	Yes	15	15	0	0	0	0	0	12	31
5000100	<i>icial</i> (1)	macrolide antibiotic;	21	Ū	105	10	10	0	Ŭ	Ŭ	0	0	12	51
		lincosamide antibiotic;												
		streptogramin												
3000593	ermQ	antibiotic	21	0	Yes	0	0	0	10	0	12	0	23	15
	· · 2	aminoglycoside												
3002626	ant(6)-Ia	antibiotic	21	0	Yes	17	0	0	0	11	0	0	11	0
		glycopeptide												
3003069	vanXYG	antibiotic	21	0	Yes	27	0	0	13	0	0	12	0	0
		lincosamide												
		antibiotic;												
3003206	lsaE	pleuromutilin antibiotic	21	0	Yes	13	0	0	15	0	16	0	0	31
3003200	ISUL	tetracycline	21	0	res	15	0	0	15	0	10	0	0	51
3000206	emrK	antibiotic	20	0	Yes	0	0	0	0	0	14	12	0	13
3003551	emeA	acridine dye	20	0	Yes	0	22	0	0	0	0	13	16	0
3002923	vanRD	glycopeptide antibiotic	19	0	Yes	0	0	0	0	0	0	13	0	13
3002923	VanAD	glycopeptide	19	0	105	0	0	0	0	0	0	15	0	15
3002944	vanHD	antibiotic	19	0	Yes	15	0	0	17	0	0	12	0	0
		glycopeptide												
3000005	vanD	antibiotic	18	0	Yes	0	15	0	28	0	0	0	12	0
		macrolide												
		antibiotic;												
		lincosamide												
		antibiotic;												
3000522	ann C	streptogramin antibiotic	17	0	Yes	29	15	0	11	0	0	10	0	10
3000322	ermG	antibiotic	1 /	0	res	29	15	0	11	0	0	10	0	10

3000263	marA	fluoroquinolone antibiotic; monobactam; carbapenem; cephalosporin; glycylcycline; cephamycin; penam; tetracycline antibiotic; rifamycin antibiotic; phenicol antibiotic; triclosan; penem macrolide antibiotic; fluoroquinolone antibiotic; penam; tetracycline	14	0	Yes	0	0	0	0	0	0	0	10	10
3000832	evgA	antibiotic glycopeptide	14	0	Yes	0	15	0	0	0	0	0	0	0
3002972	vanTG	antibiotic aminoglycoside	14	0	Yes	0	0	0	0	0	0	0	10	14
3002630	ant(9)-Ia	antibiotic macrolide antibiotic; fluoroquinolone antibiotic;	12	0	Yes	0	0	0	0	0	0	0	15	0
3000508	gadX	penam lincosamide	11	0	Yes	0	0	0	21	0	0	0	0	0
3002882	lmrD	antibiotic	10	0	Yes	0	0	0	0	0	16	0	0	0
3002957	vanYD	glycopeptide antibiotic fluoroquinolone antibiotic; lincosamide antibiotic; nucleoside antibiotic; acridine dye; phenicol	10	0	Yes	0	0	0	0	0	0	0	10	14
3001214	mdtM	antibiotic	7	0	Yes	0	0	0	0	0	0	0	0	0
3002909	vanG	glycopeptide antibiotic	6	0	Yes	0	0	0	0	0	0	0	0	0

		1 1													
		lincosamide													
		antibiotic;													
		streptogramin antibiotic;													
		pleuromutilin													
3003112	lsaC	antibiotic	6	0	Yes	0	0	0	0	0	0	0	0	0	
5005112	isue	lincosamide	0	0	103	0	0	0	0	0	0	0	0	0	
3002881	lmrC	antibiotic	4	0	Yes	0	0	0	0	0	0	0	0	0	
		aminocoumarin													
3000792	mdtA	antibiotic	3	0	Yes	10	0	0	0	0	0	0	0	0	
		tetracycline													
3000186	<i>tetM</i>	antibiotic	2	0	Yes	0	0	0	0	0	0	0	0	0	
		macrolide													
		antibiotic;													
		fluoroquinolone													
		antibiotic; aminoglycoside													
		antibiotic;													
		cephalosporin;													
		penam;													
		tetracycline													
		antibiotic;													
		aminocoumarin													
		antibiotic;													
		diaminopyrimid													
		ine antibiotic;													
3000801	mexD	phenicol antibiotic	2	0	Yes	0	0	0	0	0	0	0	0	0	
5000801	mexD	aminoglycoside	2	0	105	0	0	0	0	0	0	0	0	0	
		antibiotic;													
		cephalosporin;													
		cephamycin;													
3003052	smeB	penam	2	0	Yes	0	0	0	0	0	0	0	0	0	
		aminoglycoside													
3002629	ant(6)-Ib	antibiotic	1	0	Yes	0	0	0	0	0	0	0	0	0	
			Present	Present		G 4 3	G 4 3	G ( )	G ( )	S / 3	S 4 3		G 4 3	G ( )	
ARO	Name	Class of Resistance	in Enriched	in Shotgun	Baits	Set 2 - 9	Set 2 - 3	Set 2 - 2	Set 2 - 6	Set 2 - 5	Set 2 - 1	Set 2 - 4	Set 2 - 8	Set 2 - 7	
AKU	Name	tetracycline	Enricheu	Shotgun	Daits	- 9	5	2	U	3	1	Set 2 - 4	0	1	
3000190	tetO	antibiotic	27	6	Yes	3478	4231	4417	6684	6400	5670	5324	6567	4717	
		tetracycline	_,	-		2267									
3000191	tetQ	antibiotic	27	6	Yes	4	32260	29099	46576	50381	46810	31557	36461	28754	
	-	tetracycline													
3000196	tet32	antibiotic	27	6	Yes	6678	8515	8709	12551	12546	11884	9807	11034	9021	
		tetracycline													
3000567	tet(40)	antibiotic	27	6	Yes	5956	7154	6153	10967	9134	7174	7321	9325	7143	
2002827	ImuC	lincosamide	27	Ĺ	Vac	2442	2407	2560	1857	5125	5272	2805	4082	2276	
3002837	lnuC	antibiotic	27	6	Yes	2443	3407	3560	4857	5135	5372	3895	4083	3376	
3002837 3002867	lnuC dfrF		27 27	6	Yes Yes	2443 1286	3407 1855	3560 2000	4857 2435	5135 2620	5372 3186	3895 2469	4083 2272	3376 1916	

3002999	cblA-1	cephalosporin	27	6	Yes	2970	3701	3263	5479	4788	4264	3771	4510	3451
3002926	vanRG	glycopeptide antibiotic	25	6	Yes	52	44	17	74	63	56	41	78	43
3000194	tetW	tetracycline antibiotic macrolide antibiotic; lincosamide antibiotic;	0	6	No	0	0	0	0	0	0	0	0	0
3000375	<i>ermB</i> Nocardia rifampin resistant beta- subunit of RNA polymerase	streptogramin antibiotic peptide antibiotic; rifamycin	0	6	No	0	0	0	0	0	0	0	0	0
3000501	(rpoB2)	antibiotic aminocoumarin	0	6	No	0	0	0	0	0	0	0	0	0
3002522	novA	antibiotic	0	6	Yes	0	0	0	0	0	0	0	0	0
3002597	aac(6')-Ie- aph(2")-Ia Streptomyces rishiriensis <i>parY</i> mutant conferring resistance to	aminoglycoside antibiotic	0	6	No	0	0	0	0	0	0	0	0	0
3003318	aminocoumari n Bifidobacteria intrinsic <i>ileS</i> conferring resistance to	aminocoumarin antibiotic	0	6	No	0	0	0	0	0	0	0	0	0
3003730	mupirocin Campylobacte r coli chlorampheni col	mupirocin	0	6	No	0	0	0	0	0	0	0	0	0
3004454	acetyltransfer ase	phenicol antibiotic	0	6	No	0	0	0	0	0	0	0	0	0
3002965	vanWG	glycopeptide antibiotic macrolide	26	4	Yes	86	121	91	193	184	120	109	178	135
3000535	macB	antibiotic aminoglycoside	22	4	Yes	106	84	62	167	95	66	65	96	75
3002647	aph(3')-IIIa	antibiotic tetracycline	0	3	No	0	0	0	0	0	0	0	0	0
3000556	tet44	antibiotic macrolide	27	2	Yes	115	172	200	252	283	279	271	277	210
3003056	smeE	antibiotic;	8	2	Yes	0	18	0	22	15	0	0	16	0

		fluoroquinolone antibiotic; tetracycline antibiotic; phenicol												
		antibiotic												
3002937	vanSG	glycopeptide antibiotic aminoglycoside	0	2	Yes	0	0	0	0	0	0	0	0	0
3002983	<i>amrB</i> cepA beta-	antibiotic	0	2	No	0	0	0	0	0	0	0	0	0
3003559	lactamase	cephalosporin tetracycline	0	2	Yes	0	0	0	0	0	0	0	0	0
3004032	tetA(46)	antibiotic tetracycline	0	2	No	0	0	0	0	0	0	0	0	0
3004033	<i>tetB(46)</i>	antibiotic macrolide	0	2	No	0	0	0	0	0	0	0	0	0
		antibiotic; monobactam; tetracycline antibiotic; aminocoumarin												
3004074	muxB	antibiotic macrolide antibiotic; fluoroquinolone antibiotic; aminoglycoside antibiotic;	0	2	No	0	0	0	0	0	0	0	0	0
3004144	axy Y	cephalosporin fluoroquinolone antibiotic; cephalosporin; cephamycin;	0	2	No	0	0	0	0	0	0	0	0	0
3000502	acrF	penam aminocoumarin	27	1	Yes	229	310	209	466	377	290	218	407	221
3000793	mdtB	antibiotic aminocoumarin	0	1	No	0	0	0	0	0	0	0	0	0
3000794	mdtC	antibiotic	0	1	No	0	0	0	0	0	0	0	0	0
3003097	cfxA6	cephamycin fluoroquinolone	0	1	Yes	0	0	0	0	0	0	0	0	0
3000027	emrA	antibiotic macrolide antibiotic; fluoroquinolone antibiotic; cephalosporin; glycylcycline; cephamycin;	27	0	Yes	94	119	98	139	123	91	98	149	121
3000237	tolC	penam;	27	0	Yes	75	100	102	186	142	113	127	158	69

		tetracycline antibiotic; aminocoumarin antibiotic; rifamycin antibiotic; phenicol antibiotic;												
		triclosan												
		aminoglycoside												
3000491	acrD	antibiotic	27	0	Yes	186	217	170	398	281	111	180	337	192
		macrolide												
3000615	mefA	antibiotic macrolide	27	0	Yes	82	75	92	134	168	100	107	114	118
		antibiotic;												
3000616	mel	streptogramin antibiotic	27	0	Yes	93	60	80	163	166	168	139	162	162
3000010	mei	macrolide	27	0	1 05	93	00	80	105	100	108	139	102	102
		antibiotic;												
		fluoroquinolone												
		antibiotic;												
3000795	mdtE	penam	27	0	Yes	127	176	127	293	271	148	199	228	134
		macrolide												
		antibiotic; fluoroquinolone												
		antibiotic;												
3000796	mdtF	penam	27	0	Yes	208	267	163	455	292	190	254	350	181
		aminoglycoside												
		antibiotic;												
		aminocoumarin	27	0	••				•		100	0.6		
3000830	cpxA	antibiotic macrolide	27	0	Yes	135	147	112	290	215	132	96	215	140
		antibiotic;												
		fluoroquinolone												
		antibiotic;												
		penam;												
2000022	G	tetracycline	27	0		12	(2)	0.0	100	120	107	76	116	50
3000833	evgS	antibiotic fluoroquinolone	27	0	Yes	43	62	88	120	130	126	76	116	53
3001216	mdtH	antibiotic	27	0	Yes	52	69	74	137	86	63	62	67	43
5001210	maili	tetracycline	27	0	105	52	0)	, .	157	00	05	02	07	15
		antibiotic;												
		benzalkonium												
2001220	Escherichia	chloride;	27	6		10	00	51	100			(0)	105	
3001328	coli mdfA	rhodamine nucleoside	27	0	Yes	49	98	51	100	44	77	60	105	66
		antibiotic;												
3003549	mdtO	acridine dye	27	0	Yes	129	197	164	290	262	119	145	241	162
		-												

		nucleoside												
2002550		antibiotic;	27	0							10.6	100	100	
3003550	mdtP	acridine dye peptide	27	0	Yes	135	140	116	252	266	136	102	192	154
3003576	<i>eptA</i>	antibiotic	27	0	Yes	121	121	109	234	171	139	111	208	91
	*	peptide												
3003578	pmrF	antibiotic	27	0	Yes	89	128	82	182	151	77	89	151	95
3000074	emrB	fluoroquinolone antibiotic	26	0	Yes	86	80	49	151	88	76	50	127	70
5000074	emib	fluoroquinolone	20	0	103	00	00	77	151	00	70	50	12/	70
		antibiotic;												
		cephalosporin;												
3000499	acrE	cephamycin; penam	26	0	Yes	90	107	76	178	102	151	82	149	82
5000477	uc/E	macrolide	20	0	103	70	107	70	170	102	151	02	147	02
		antibiotic;												
		fluoroquinolone												
3000518	crp	antibiotic; penam	26	0	Yes	36	80	35	90	48	54	47	54	29
5000510	crp	fluoroquinolone	20	0	103	50	00	55	70	10	54	-17	54	2)
		antibiotic;												
		cephalosporin;												
		glycylcycline; cephamycin;												
		penam;												
		tetracycline												
		antibiotic;												
		rifamycin antibiotic;												
		phenicol												
		antibiotic;												
3000656	acrS	triclosan	26	0	Yes	67	52	52	101	83	62	44	93	78
3002635	aph(2")-IIa	aminoglycoside antibiotic	26	0	Yes	50	43	69	84	102	97	138	90	93
5002055	<i>upn(2 )</i> 11 <i>u</i>	nucleoside	20	0	105	20	15	07	01	102	21	150	20	25
		antibiotic;												
3003548	mdtN	acridine dye tetracycline	26	0	Yes	43	76	47	105	85	41	57	81	25
3000254	emrY	antibiotic	25	0	Yes	28	11	23	71	31	49	33	61	44
3001329	mdtG	fosfomycin	25	0	Yes	42	47	44	97	94	28	74	113	44
5001529	maio	peptide	25	0	105	42	47	44	21	24	28	/4	115	
3002986	bacA	antibiotic	25	0	Yes	40	48	30	70	65	15	44	51	42
		fluoroquinolone												
		antibiotic; cephalosporin;												
		glycylcycline;												
		penam;												
		tetracycline												
3000216	acrB	antibiotic; rifamycin	24	0	Yes	45	40	28	61	36	17	22	34	22
3000210	ucrb	maniyem	∠4	0	1 08	43	40	20	01	30	1 /	22	34	22

		antibiotic; phenicol antibiotic; triclosan												
3002688	catS	phenicol antibiotic tetracycline	24	0	Yes	22	39	42	63	57	69	39	40	36
3000195	tetB(P)	antibiotic lincosamide antibiotic;	23	0	Yes	25	27	31	30	48	55	40	24	28
3000300	lsaA	streptogramin antibiotic; pleuromutilin antibiotic	22	0	Yes	17	15	28	28	24	30	37	23	35
3000300	ISUA	macrolide antibiotic;	22	0	1 es	17	15	28	28	24	30	57	25	33
		fluoroquinolone antibiotic; cephalosporin;												
		cephamycin; penam; tetracycline												
3000676	H-NS	antibiotic glycopeptide	22	0	Yes	50	37	20	56	56	37	27	62	41
3003070	vanXD	antibiotic tetracycline	22	0	Yes	12	27	16	32	26	12	24	39	22
3000180	tetA(P)	antibiotic macrolide	21	0	Yes	19	17	31	43	21	30	46	52	28
		antibiotic; lincosamide antibiotic; streptogramin												
3000593	ermQ	antibiotic aminoglycoside	21	0	Yes	11	13	11	33	34	29	25	16	18
3002626	ant(6)-Ia	antibiotic glycopeptide	21	0	Yes	14	17	14	29	25	18	26	27	28
3003069	vanXYG	antibiotic lincosamide antibiotic;	21	0	Yes	26	19	16	40	50	29	28	37	20
3003206	lsaE	pleuromutilin antibiotic tetracycline	21	0	Yes	20	30	25	34	27	53	28	42	30
3000206	emrK	antibiotic	20	0	Yes	29	29	24	39	25	22	35	34	32
3003551	emeA	acridine dye glycopeptide	20	0	Yes	22	28	26	31	16	29	34	39	44
3002923	vanRD	antibiotic glycopeptide	19	0	Yes	13	19	12	42	25	28	14	36	15
3002944	vanHD	antibiotic glycopeptide	19	0	Yes	0	18	37	37	30	17	32	26	23
3000005	vanD	antibiotic	18	0	Yes	0	26	16	29	28	44	17	19	26

3000522	ermG	macrolide antibiotic; lincosamide antibiotic; streptogramin antibiotic fluoroquinolone antibiotic; monobactam; carbapenem; cephalosporin; glycylcycline; cephamycin; penam; tetracycline antibiotic; rifamycin antibiotic;	17	0	Yes	0	22	23	16	15	27	27	21	19
3000263	marA	phenicol antibiotic; triclosan; penem macrolide antibiotic; fluoroquinolone antibiotic;	14	0	Yes	14	17	10	28	16	20	20	22	0
2000922	,	penam; tetracycline	14	0	V	10	0	12	22	15	12	12	26	17
3000832	evgA	antibiotic glycopeptide	14	0	Yes	10	0	12	23	15	12	13	26	17
3002972	vanTG	antibiotic aminoglycoside	14	0	Yes	12	13	0	24	26	12	14	13	19
3002630	ant(9)-Ia	antibiotic macrolide antibiotic; fluoroquinolone antibiotic;	12	0	Yes	0	11	0	10	0	13	22	16	0
3000508	gadX	penam lincosamide	11	0	Yes	16	0	12	16	17	0	19	0	11
3002882	lmrD	antibiotic	10	0	Yes	0	0	28	0	0	13	14	0	12
3002957	vanYD	glycopeptide antibiotic fluoroquinolone antibiotic; lincosamide antibiotic; nucleoside antibiotic;	10	0	Yes	0	0	12	10	10	0	0	11	0
3001214	mdtM	acridine dye;	7	0	Yes	10	0	0	15	0	16	11	16	0

3002909	vanG	phenicol antibiotic glycopeptide antibiotic lincosamide antibiotic; streptogramin antibiotic; pleuromutilin	6	0	Yes	0	0	0	0	15	14	16	12	0
3003112	lsaC	antibiotic lincosamide	6	0	Yes	0	0	0	17	12	0	18	15	14
3002881	lmrC	antibiotic aminocoumarin	4	0	Yes	0	0	0	0	0	0	0	12	17
3000792	mdtA	antibiotic	3	0	Yes	0	0	0	0	0	19	0	0	0
3000186	tetM	tetracycline antibiotic macrolide	2	0	Yes	0	0	0	0	0	0	0	11	0
3000801	mexD	antibiotic; fluoroquinolone antibiotic; aminoglycoside antibiotic; cephalosporin; penam; tetracycline antibiotic; aminocoumarin antibiotic; diaminopyrimid ine antibiotic; phenicol antibiotic aminoglycoside antibiotic; cephalosporin;	2	0	Yes	0	0	0	16	0	0	0	0	0
3003052	smeB	cephamycin; penam	2	0	Yes	0	0	0	0	0	0	0	0	0
3002629		aminoglycoside	-	0	Yes	0	0	0	0	0	0	0	0	
5002029	ant(6)-Ib	antibiotic	Present	Present	res	0	0	0	0	0	0	0	0	0
		Class of	in	in		Set 3	Set 3 -		Set 3 -	Set 3 -				
ARO	Name	Resistance	Enriched	Shotgun	Baits	- 9	6	8	7	5	3	Set 3 - 2	4	1
3000190	tetO	tetracycline antibiotic tetracycline	27	6	Yes	4389 3196	3143	4035	4083	3662	3459	5115	3742	4278
3000191	tetQ	antibiotic tetracycline	27	6	Yes	1	25807	27902	30217	30537	31375	57377	35805	38948
3000196	tet32	antibiotic tetracycline	27	6	Yes	8770	7045	8207	8497	8055	7484	12627	9006	9549
3000567	tet(40)	antibiotic	27	6	Yes	7844	5526	7038	6490	6893	5856	7884	5888	5971

		lincosamide												
3002837	lnuC	antibiotic diaminopyrimid	27	6	Yes	3591	2944	3308	3659	3351	3360	6483	4322	4901
3002867	dfrF	ine antibiotic	27	6	Yes	1624	1429	1733	2133	1746	1579	3276	2464	2945
3002999	cblA-1	cephalosporin glycopeptide	27	6	Yes	4441	3146	4007	4244	3884	3509	5435	3914	3948
3002926	vanRG	antibiotic tetracycline	25	6	Yes	49	50	29	19	29	45	21	18	21
3000194	tetW	antibiotic macrolide antibiotic; lincosamide antibiotic; streptogramin	0	6	No	0	0	0	0	0	0	0	0	0
3000375	ermB Nocardia rifampin resistant beta- subunit of RNA polymerase	antibiotic peptide antibiotic; rifamycin	0	6	No	0	0	0	0	0	0	0	0	0
3000501	(rpoB2)	antibiotic aminocoumarin	0	6	No	0	0	0	0	0	0	0	0	0
3002522	novA aac(6')-Ie-	antibiotic aminoglycoside	0	6	Yes	0	0	0	0	0	0	0	0	0
3002597	aph(2")-Ia Streptomyces rishiriensis <i>parY</i> mutant conferring resistance to	antibiotic	0	6	No	0	0	0	0	0	0	0	0	0
3003318	aminocoumari n Bifidobacteria intrinsic <i>ileS</i> conferring resistance to	aminocoumarin antibiotic	0	6	No	0	0	0	0	0	0	0	0	0
3003730	mupirocin Campylobacte r coli chlorampheni col	mupirocin	0	6	No	0	0	0	0	0	0	0	0	0
3004454	acetyltransfer ase	phenicol antibiotic	0	6	No	0	0	0	0	0	0	0	0	0
3002965	vanWG	glycopeptide antibiotic macrolide	26	4	Yes	109	78	107	89	76	80	94	70	107
3000535	macB	antibiotic aminoglycoside	22	4	Yes	92	71	82	77	63	87	0	51	51
3002647	aph(3')-IIIa	antibiotic	0	3	No	0	0	0	0	0	0	0	0	0

		tetracycline												
3000556	tet44	antibiotic macrolide	27	2	Yes	145	110	130	117	119	112	216	170	211
		antibiotic; fluoroquinolone												
		antibiotic;												
		tetracycline antibiotic;												
2002056		phenicol	0	2		0	0	0	0	20	0	0	0	0
3003056	smeE	antibiotic glycopeptide	8	2	Yes	0	0	0	0	20	0	0	0	0
3002937	vanSG	antibiotic aminoglycoside	0	2	Yes	0	0	0	0	0	0	0	0	0
3002983	<i>amrB</i> cepA beta-	antibiotic	0	2	No	0	0	0	0	0	0	0	0	0
3003559	lactamase	cephalosporin tetracycline	0	2	Yes	0	0	0	0	0	0	0	0	0
3004032	tetA(46)	antibiotic tetracycline	0	2	No	0	0	0	0	0	0	0	0	0
3004033	tetB(46)	antibiotic	0	2	No	0	0	0	0	0	0	0	0	0
		macrolide antibiotic;												
		monobactam;												
		tetracycline antibiotic;												
		aminocoumarin												
3004074	muxB	antibiotic macrolide	0	2	No	0	0	0	0	0	0	0	0	0
		antibiotic;												
		fluoroquinolone antibiotic;												
		aminoglycoside												
3004144	axvY	antibiotic; cephalosporin	0	2	No	0	0	0	0	0	0	0	0	0
3004144	axy1	fluoroquinolone	0	Z	INO	0	0	0	0	0	0	0	0	0
		antibiotic;												
		cephalosporin; cephamycin;												
3000502	acrF	penam	27	1	Yes	219	188	188	171	199	166	192	154	155
3000793	mdtB	aminocoumarin antibiotic	0	1	No	0	0	0	0	0	0	0	0	0
2000704		aminocoumarin	0	1	N	0	0	0	0	0	0	0	0	0
3000794	mdtC	antibiotic	0	1	No			0	0					
3003097	cfxA6	cephamycin fluoroquinolone	0	1	Yes	0	0	0	0	0	0	0	0	0
3000027	emrA	antibiotic macrolide	27	0	Yes	72	67	70	80	80	53	87	55	94
		antibiotic;												
3000237	tolC	fluoroquinolone antibiotic;	27	0	Yes	81	59	60	90	109	64	74	75	83
5000257	1010	antioione,	<i>∠ I</i>	U	1 05	01	37	00	90	109	04	/4	15	03

		cephalosporin; glycylcycline; cephamycin; penam; tetracycline antibiotic; aminocoumarin antibiotic; rifamycin antibiotic; phenicol antibiotic; triclosan												
3000491	acrD	aminoglycoside antibiotic	27	0	Yes	145	161	182	152	254	150	165	134	126
3000615	mefA	macrolide antibiotic macrolide antibiotic;	27	0	Yes	79	57	73	56	54	56	70	84	58
3000616	mel	streptogramin antibiotic macrolide antibiotic; fluoroquinolone antibiotic;	27	0	Yes	90	61	75	98	63	63	165	119	114
3000795	mdtE	antibiotic; penam macrolide antibiotic; fluoroquinolone antibiotic;	27	0	Yes	139	64	118	83	127	110	114	110	93
3000796	mdtF	penam aminoglycoside antibiotic; aminocoumarin	27	0	Yes	247	155	156	149	172	113	159	159	134
3000830	cpxA	antibiotic macrolide antibiotic; fluoroquinolone antibiotic; penam; tetracycline	27	0	Yes	142	101	91	94	133	131	108	90	85
3000833	evgS	antibiotic fluoroquinolone	27	0	Yes	36	47	47	59	64	28	66	68	61
3001216	mdtH	antibiotic tetracycline antibiotic; benzalkonium	27	0	Yes	72	31	38	39	34	37	56	47	45
3001328	Escherichia coli mdfA	chloride; rhodamine	27	0	Yes	40	47	46	54	48	42	46	43	50

		nucleoside												
		antibiotic;												
3003549	mdtO	acridine dye nucleoside antibiotic;	27	0	Yes	124	88	124	107	132	126	127	101	104
3003550	mdtP	acridine dye peptide	27	0	Yes	138	103	154	110	128	115	134	96	71
3003576	eptA	antibiotic peptide	27	0	Yes	125	87	75	76	113	79	84	107	107
3003578	pmrF	antibiotic fluoroquinolone	27	0	Yes	96	64	67	75	81	47	87	56	48
3000074	emrB	antibiotic fluoroquinolone antibiotic; cephalosporin; cephamycin;	26	0	Yes	37	53	44	76	55	63	65	47	62
3000499	acrE	penam macrolide antibiotic; fluoroquinolone antibiotic;	26	0	Yes	91	75	88	66	78	43	73	80	65
3000518	crp	penam fluoroquinolone antibiotic; cephalosporin; glycylcycline; cephamycin; penam; tetracycline antibiotic; rifamycin antibiotic; phenicol antibiotic;	26	0	Yes	44	17	45	27	23	26	39	23	16
3000656	acrS	triclosan aminoglycoside	26	0	Yes	33	32	54	29	32	18	68	38	40
3002635	aph(2")-IIa	antibiotic nucleoside antibiotic;	26	0	Yes	71	51	57	43	39	52	98	77	63
3003548	mdtN	acridine dye tetracycline	26	0	Yes	26	36	39	23	37	34	34	22	33
3000254	emrY	antibiotic	25	0	Yes	33	0	30	20	23	18	32	55	29
3001329	mdtG	fosfomycin peptide	25	0	Yes	31	44	32	45	45	38	40	15	27
3002986	bacA	antibiotic fluoroquinolone antibiotic; cephalosporin; glycylcycline;	25	0	Yes	42	49	29	41	36	15	20	28	33
3000216	acrB	penam;	24	0	Yes	28	15	34	17	29	23	24	26	10

		tetracycline antibiotic; rifamycin antibiotic; phenicol antibiotic; triclosan phenicol												
3002688	catS	antibiotic tetracycline	24	0	Yes	36	15	18	22	25	28	45	33	44
3000195	tetB(P)	antibiotic lincosamide antibiotic; streptogramin antibiotic; pleuromutilin	23	0	Yes	23	17	23	25	29	30	16	20	39
3000300	lsaA	antibiotic macrolide antibiotic; fluoroquinolone antibiotic; cephalosporin; cephamycin; penam; tetracycline	22	0	Yes	0	0	18	11	18	10	17	15	25
3000676	H-NS	antibiotic glycopeptide	22	0	Yes	41	31	19	17	30	37	35	30	27
3003070	vanXD	antibiotic tetracycline	22	0	Yes	16	13	12	11	18	19	54	21	25
3000180	tetA(P)	antibiotic macrolide antibiotic; lincosamide antibiotic; streptogramin	21	0	Yes	25	14	28	31	33	0	36	45	48
3000593	ermQ	antibiotic aminoglycoside	21	0	Yes	0	15	15	21	10	13	28	15	26
3002626	ant(6)-Ia	antibiotic glycopeptide	21	0	Yes	19	26	14	15	13	15	23	19	12
3003069	vanXYG	antibiotic lincosamide antibiotic; pleuromutilin	21	0	Yes	23	15	25	10	24	20	11	13	20
3003206	lsaE	antibiotic tetracycline	21	0	Yes	0	21	13	29	26	16	46	17	22
3000206	emrK	antibiotic	20	0	Yes	15	13	10	0	15	16	12	20	25
3003551	emeA	acridine dye glycopeptide	20	0	Yes	16	29	32	22	0	13	20	20	35
3002923	vanRD	antibiotic	19	0	Yes	20	16	0	11	14	13	18	11	23

		almaantida												
3002944	vanHD	glycopeptide antibiotic	19	0	Yes	15	22	0	37	14	19	32	11	32
3000005	vanD	glycopeptide antibiotic	18	0	Yes	14	0	19	20	19	0	15	13	14
3000003	vanD	macrolide	10	0	105	14	0	19	20	19	0	15	15	14
		antibiotic;												
		lincosamide												
		antibiotic;												
		streptogramin												
3000522	ermG	antibiotic	17	0	Yes	0	0	18	0	0	13	31	10	0
		fluoroquinolone												
		antibiotic;												
		monobactam;												
		carbapenem;												
		cephalosporin;												
		glycylcycline;												
		cephamycin;												
		penam; tetracycline												
		antibiotic;												
		rifamycin												
		antibiotic;												
		phenicol												
		antibiotic;												
		triclosan;												
3000263	marA	penem	14	0	Yes	17	0	0	0	0	0	14	12	12
		macrolide												
		antibiotic;												
		fluoroquinolone antibiotic;												
		penam;												
		tetracycline												
3000832	evgA	antibiotic	14	0	Yes	0	11	0	12	0	13	23	0	16
	0	glycopeptide												
3002972	vanTG	antibiotic	14	0	Yes	0	0	12	0	0	12	0	20	11
		aminoglycoside												
3002630	ant(9)-Ia	antibiotic	12	0	Yes	0	11	16	0	11	0	34	26	11
		macrolide												
		antibiotic; fluoroquinolone												
		antibiotic;												
3000508	gadX	penam	11	0	Yes	0	0	0	13	13	0	15	12	0
	8	lincosamide												
3002882	lmrD	antibiotic	10	0	Yes	19	0	0	0	0	10	18	14	12
		glycopeptide												
3002957	vanYD	antibiotic	10	0	Yes	0	19	0	0	12	0	12	0	13
		fluoroquinolone												
		antibiotic;												
3001214	mdtM	lincosamide antibiotic;	7	0	Yes	0	0	10	0	10	0	0	0	0
5001214	mann	annoione,	/	0	1 05	U	U	10	0	10	0	U	0	U

3002909	vanG		nucleoside antibiotic; acridine dye; phenicol antibiotic glycopeptide antibiotic lincosamide antibiotic; streptogramin antibiotic; pleuromutilin	6	0	Yes	12		0	0	0	0	0	14	0	0
3003112	lsaC		antibiotic	6	0	Yes	0		0	12	0	0	0	0	0	0
2002991	lC		lincosamide	4	0	V	11		0	0	1.4	0	0	0	0	0
3002881	lmrC		antibiotic aminocoumarin	4	0	Yes	11		0	0	14	0	0	0	0	0
3000792	mdtA		antibiotic	3	0	Yes	0		0	0	0	0	0	0	10	0
3000186	tetM		tetracycline antibiotic	2	0	Yes	0		0	0	0	0	0	0	13	0
3000186	mexD		macrolide antibiotic; fluoroquinolone antibiotic; aminoglycoside antibiotic; cephalosporin; penam; tetracycline antibiotic; aminocoumarin antibiotic; diaminopyrimid ine antibiotic; phenicol antibiotic aminoglycoside antibiotic; cephalosporin;	2		Yes	0		0	0	0	17	0	0	0	0
3003052	smeB		cephamycin; penam	2	0	Yes	0		32	0	0	0	38	0	0	0
3002629	ant(6)-1	TL	aminoglycoside antibiotic	1	0	Yes	0		0	0	0	0	0	10	0	0
3002029	<i>uni(0)-1</i>	lU	antibiotic	1	0	105	0		0	0	0	0	0	10	0	
ARO		Name	Class of Resistance	•	Present in Enriched		Present in Shotgun		Bait s	Set 1 - 6 Shotgun	Set 1 - 7 Shotgun		: 2 - 9 otgun	Set 2 - 5 Shotgun	Set 3 - 5 Shotgun	Set 3 - 1 Shotgun
300	00190	tetO	tetracycline antibio	ic	ź	27		6	Yes	127	14	46	296	281	179	211
300	0191	tetQ	tetracycline antibio	ic	í	27		6	Yes	654	77	74	1568	1314	790	1150
300	0196	tet32	tetracycline antibio		,	27		6	Yes	116	1:		238	221	133	227

3000567	tet(40)	tetracycline antibiotic	27	6	Yes	44	59	96	90	66	72
3002837	lnuC	lincosamide antibiotic diaminopyrimidine	27	6	Yes	94	114	208	174	84	152
3002867	dfrF	antibiotic	27	6	Yes	32	32	86	50	38	48
3002999	cblA-1	cephalosporin	27	6	Yes	46	50	60	66	44	76
3002926	vanRG	glycopeptide antibiotic	25	6	Yes	10	22	30	28	16	24
3000194	tetW	tetracycline antibiotic	0	6	No	546	635	1108	836	649	862
3000375	<i>ermB</i> Nocardia rifampin resistant beta-subunit of RNA	macrolide antibiotic; lincosamide antibiotic; streptogramin antibiotic	0	6	No	36	34	74	70	34	46
2000501	polymerase	peptide antibiotic;	0	(	N	97	120	12(	0.4	0(	100
3000501	( <i>rpoB2</i> )	rifamycin antibiotic aminocoumarin	0	6	No	86	120	136	94	96	108
3002522	novA aac(6')-Ie-	antibiotic aminoglycoside	0	6	Yes	12	14	14	24	22	16
3002597	aac(6)-Ie- aph(2")-Ia Streptomyce s rishiriensis	antibiotic	0	6	No	30	44	80	78	46	56
	s rishiriensis parY mutant conferring resistance to										
2002210	aminocouma	aminocoumarin antibiotic	0	(	N.	06	108	170	140	110	124
3003318	rin Bifidobacter	anubiolic	0	6	No	96	108	178	148	110	124
	ia intrinsic <i>ileS</i> conferring										
3003730	resistance to mupirocin <i>Campylobac</i> <i>ter coli</i> chloramphen icol	mupirocin	0	6	No	50	74	98	68	60	82
	acetyltransfe										
3004454	rase	phenicol antibiotic	0	6	No	14	16	22	26	10	22
3002965	vanWG	glycopeptide antibiotic	26	4	Yes	14	0	28	24	14	0
3000535	macB	macrolide antibiotic aminoglycoside	22	4	Yes	0	12	16	28	0	18
3002647	aph(3')-IIIa	antibiotic	0	3	No	0	12	0	10	0	10
3000556	tet44	tetracycline antibiotic	27	2	Yes	0	0	10	12	0	0

		macrolide antibiotic; fluoroquinolone antibiotic; tetracycline antibiotic; phenicol									
3003056	smeE	antibiotic	8	2	Yes	0	12	12	0	0	0
3002937	vanSG	glycopeptide antibiotic aminoglycoside	0	2	Yes	0	10	16	0	0	0
3002983	<i>amrB</i> cepA beta-	antibiotic	0	2	No	0	0	0	0	10	10
3003559	lactamase	cephalosporin	0	2	Yes	0	0	12	0	10	0
3004032	tetA(46)	tetracycline antibiotic	0	2	No	0	0	10	0	0	16
3004033	tetB(46)	tetracycline antibiotic	0	2	No	0	0	14	10	0	0
3004074	muxB	macrolide antibiotic; monobactam; tetracycline antibiotic; aminocoumarin antibiotic macrolide antibiotic; fluoroquinolone antibiotic;	0	2	No	0	0	0	15	0	18
3004144	axy Y	aminoglycoside antibiotic; cephalosporin	0	2	No	16	0	26	0	0	0
		fluoroquinolone antibiotic; cephalosporin;									
3000502	acrF	cephamycin; penam aminocoumarin	27	1	Yes	0	0	13	0	0	0
3000793	mdtB	antibiotic aminocoumarin	0	1	No	0	0	0	15	0	0
3000794	mdtC	antibiotic	0	1	No	0	0	10	0	0	0
3003097	cfxA6	cephamycin fluoroquinolone	0	1	Yes	0	0	0	0	0	43
3000027	emrA	antibiotic	27	0	Yes	0	0	0	0	0	0

3000237	tolC	macrolide antibiotic; fluoroquinolone antibiotic; cephalosporin; glycylcycline; cephamycin; penam; tetracycline antibiotic; aminocoumarin antibiotic; rifamycin antibiotic; phenicol antibiotic; triclosan	27	0	Yes	0	0	0	0	0	0
		aminoglycoside		0							
3000491	acrD	antibiotic	27	0	Yes	0	0	0	0	0	0
3000615	mefA	macrolide antibiotic	27	0	Yes	0	0	0	0	0	0
3000616	mel	macrolide antibiotic; streptogramin antibiotic	27	0	Yes	0	0	0	0	0	0
3000795	mdtE	macrolide antibiotic; fluoroquinolone antibiotic; penam	27	0	Yes	0	0	0	0	0	0
3000796 3000830	mdtF	macrolide antibiotic; fluoroquinolone antibiotic; penam aminoglycoside antibiotic; aminocoumarin antibiotic	27 27	0	Yes	0	0	0	0	0	0
3000830	cpxA		27	0	Yes	0	0	0	0	0	0
3000833 3001216	evgS mdtH	macrolide antibiotic; fluoroquinolone antibiotic; penam; tetracycline antibiotic fluoroquinolone antibiotic	27 27	0 0	Yes Yes	0 0	0 0	0 0	0 0	0 0	0 0
3001328	Escherichia coli mdfA	tetracycline antibiotic; benzalkonium chloride; rhodamine	27	0	Yes	0	0	0	0	0	0
3003549	mdtO	nucleoside antibiotic; acridine dye	27	0	Yes	0	0	0	0	0	0
3003550 3003576	mdtP eptA	nucleoside antibiotic; acridine dye peptide antibiotic	27 27	0 0	Yes Yes	0 0	0 0	0 0	0 0	0 0	0 0

3003578	pmrF	peptide antibiotic fluoroquinolone	27	0	Yes	0	0	0	0	0	0
3000074	emrB	antibiotic	26	0	Yes	0	0	0	0	0	0
		fluoroquinolone									
		antibiotic; cephalosporin;									
3000499	acrE	cephamycin; penam	26	0	Yes	0	0	0	0	0	0
		macrolide antibiotic;									
		fluoroquinolone									
3000518	crp	antibiotic; penam	26	0	Yes	0	0	0	0	0	0
		fluoroquinolone									
		antibiotic; cephalosporin;									
		glycylcycline;									
		cephamycin; penam; tetracycline antibiotic;									
		rifamycin antibiotic;									
2000/5/	G	phenicol antibiotic;	24	0		0	0	0	0	0	0
3000656	acrS	triclosan aminoglycoside	26	0	Yes	0	0	0	0	0	0
3002635	aph(2'')-IIa	antibiotic	26	0	Yes	0	0	0	0	0	0
		nucleoside antibiotic;									
3003548	mdtN	acridine dye	26	0	Yes	0	0	0	0	0	0
3000254	emrY	tetracycline antibiotic	25	0	Yes	0	0	0	0	0	0
3001329	mdtG	fosfomycin	25	0	Yes	0	0	0	0	0	0
3002986	bacA	peptide antibiotic	25	0	Yes	0	0	0	0	0	0
		fluoroquinolone									
		antibiotic; cephalosporin;									
		glycylcycline; penam; tetracycline antibiotic;									
		rifamycin antibiotic;									
	_	phenicol antibiotic;									
3000216	acrB	triclosan	24	0	Yes	0	0	0	0	0	0
3002688	catS	phenicol antibiotic	24	0	Yes	0	0	0	0	0	0
3000195	tetB(P)	tetracycline antibiotic	23	0	Yes	0	0	0	0	0	0
		lincosamide antibiotic;									
		streptogramin antibiotic;									
3000300	lsaA	pleuromutilin antibiotic	22	0	Yes	0	0	0	0	0	0

		macrolide antibiotic; fluoroquinolone antibiotic; cephalosporin; cephamycin; penam;									
3000676	H-NS	tetracycline antibiotic	22	0	Yes	0	0	0	0	0	0
3003070	vanXD	glycopeptide antibiotic	22	0	Yes	0	0	0	0	0	0
3000180	tetA(P)	tetracycline antibiotic	21	0	Yes	0	0	0	0	0	0
3000593	ermQ	macrolide antibiotic; lincosamide antibiotic; streptogramin antibiotic aminoglycoside	21	0	Yes	0	0	0	0	0	0
3002626	ant(6)-Ia	antibiotic	21	0	Yes	0	0	0	0	0	0
3003069	vanXYG	glycopeptide antibiotic	21	0	Yes	0	0	0	0	0	0
		lincosamide antibiotic;									
3003206	lsaE	pleuromutilin antibiotic	21	0	Yes	0	0	0	0	0	0
3000206	emrK	tetracycline antibiotic	20	0	Yes	0	0	0	0	0	0
3003551	emeA	acridine dye	20	0	Yes	0	0	0	0	0	0
3002923	vanRD	glycopeptide antibiotic	19	0	Yes	0	0	0	0	0	0
3002944	vanHD	glycopeptide antibiotic	19	0	Yes	0	0	0	0	0	0
3000005	vanD	glycopeptide antibiotic	18	0	Yes	0	0	0	0	0	0
3000522	ermG	macrolide antibiotic; lincosamide antibiotic; streptogramin antibiotic	17	0	Yes	0	0	0	0	0	0
3000263	marA	fluoroquinolone antibiotic; monobactam; carbapenem; cephalosporin; glycylcycline; cephamycin; penam; tetracycline antibiotic; rifamycin antibiotic; phenicol antibiotic; triclosan; penem	14	0	Yes	0	0	0	0	0	0
		macrolide antibiotic; fluoroquinolone antibiotic; penam;									
3000832	evgA	tetracycline antibiotic	14	0	Yes	0	0	0	0	0	0
3002972	vanTG	glycopeptide antibiotic	14	0	Yes	0	0	0	0	0	0

3002630	ant(9)-Ia	aminoglycoside antibiotic	12	0	Yes	0	0	0	0	0	0
3000508 3002882	gadX lmrD	macrolide antibiotic; fluoroquinolone antibiotic; penam lincosamide antibiotic	11 10	0 0	Yes Yes	0 0	0 0	0 0	0 0	0 0	0 0
3002957	vanYD	glycopeptide antibiotic	10	0	Yes	0	0	0	0	0	0
3001214	mdtM	fluoroquinolone antibiotic; lincosamide antibiotic; nucleoside antibiotic; acridine dye; phenicol antibiotic	7	0	Yes	0	0	0	0	0	0
3002909	vanG	glycopeptide antibiotic	6	0	Yes	0	0	0	0	0	0
3003112	lsaC	lincosamide antibiotic; streptogramin antibiotic; pleuromutilin antibiotic	6	0	Yes	0	0	0	0	0	0
3002881	lmrC	lincosamide antibiotic aminocoumarin	4	0	Yes	0	0	0	0	0	0
3000792	mdtA	antibiotic	3	0	Yes	0	0	0	0	0	0
3000186	tetM	tetracycline antibiotic macrolide antibiotic; fluoroquinolone antibiotic; aminoglycoside antibiotic; cephalosporin; penam; tetracycline antibiotic; aminocoumarin antibiotic; diaminopyrimidine antibiotic; phenicol	2	0	Yes	0	0	0	0	0	0
3000801	mexD	antibiotic	2	0	Yes	0	0	0	0	0	0
3003052	smeB	aminoglycoside antibiotic; cephalosporin; cephamycin; penam	2	0	Yes	0	0	0	0	0	0
3002629	ant(6)-Ib	aminoglycoside antibiotic	1	0	Yes	0	0	0	0	0	0

## References

1. Meyer M, Kircher M. 2010. Illumina Sequencing Library Preparation for Highly Multiplexed Target Capture and Sequencing. Cold Spring Harb Protoc 2010:pdb.prot5448-pdb.prot5448.