Clinical Microbiology & Pathogen Genomics | Signal Rating: 2/5 Stars | Oct 29, 2026 | 10 min read

Enterobacteriaceae Core Chromosome Synteny, Resistome HGT & 16S Operon Phasing

Whole-contig tetranucleotide divergence and intragenomic rrsA-rrsH 16S operon phasing across Escherichia coli, Salmonella enterica, Klebsiella pneumoniae, and Acinetobacter baumannii resolve sister-species boundaries and isolate foreign resistance islands.

| Genomic Element | | Optimal Diagnostic Window | | Empirical Failure Boundary | | Corrective Assay Rescue Protocol |

| Phased 16S rRNA Operons + Whole-Contig Tetranucleotide D_KL | | Enteric Species Resolution & Horizontal Resistance Island Boundary Detection (7.4x D_KL separation ratio) | | Unphased short-read 16S V3-V4 amplicons collapse across Escherichia, Salmonella, and Klebsiella (61.5% bootstrap), while chromosomal housekeeping loci miss mobile plasmid/ICE transfers. | | Combine intragenomic rrsA-rrnH copy phasing with sliding-window tetranucleotide D_KL around tRNA integration junctions and plasmid replicon typing. |

Generated by: Native Rust Horizontal Gene Transfer, Tetranucleotide KL-Divergence & Resistome Scanner (hgt_bgc.rs)

Unphased single-copy 16S rRNA comparisons fail to separate closely related Escherichia coli and Salmonella enterica strains (negative barcoding gap = -0.0013; 61.5% bootstrap) because intragenomic divergence among the seven E. coli rrsA-rrsH operons (d_K2P = 0.0112) exceeds the inter-genus threshold. Evaluating 214,912 bp across E. coli, S. enterica, Klebsiella pneumoniae, and Acinetobacter baumannii suggests that phasing intragenomic 16S operons across the C2V-V4 region restores 94.0% clade bootstrap support, while whole-contig tetranucleotide divergence separates conserved enteric backbones from mobile Acinetobacter resistance islands by 7.4-fold.

Executive Summary: In clinical microbiology and pathogen genomics, treating multi-copy 16S rRNA operons (rrsA-rrsH) as an unphased single-copy barcode across closely related Enterobacteriaceae causes sister species to overlap. Unphased 16S comparisons across Escherichia coli K-12 (NC_000913.3), Salmonella enterica LT2 (NC_003197.2), and Klebsiella pneumoniae HS11286 (NC_016845.1) yield a negative barcoding gap (−0.0013; 61.5% bootstrap) because intragenomic divergence in E. coli rrsH (d intra = 0.0112) exceeds the minimum distance to S. enterica (d inter = 0.0099). Here we validate a dual-modality resolution across 13 loci (214,912 bp): coupling intragenomic rrsA-rrsH operon phasing across the C2V-V4 hypervariable region (94.0% bootstrap) with whole-contig tetranucleotide Kullback-Leibler divergence (D KL = 0.0255-0.0294 bits across 180 kb of enteric core chromosomes vs. 0.2175 bits in the 20 kb Acinetobacter baumannii NC_010410.1 mobile resistome donor) restores complete species separation and achieves a 7.40-fold HGT discrimination ratio .

Comparative Prokaryotic Enteric & Resistome Telemetry

| Accession & Coordinates | | Organism / Locus | | Locus Type | | Span (bp) | | GC (%) | | D_KL vs E. coli Core (bits) | | Intragenomic 16S SNPs | | G4Hunter Motifs | | Functional Classification |

| NC_000913.3:1-60000 | | Escherichia coli K-12 MG1655 | | Enteric Core Chromosome (thrL-dnaK) | | 60000 | | 51.23 | | 0.0000 | | 0 | | 24 | | Core Reference |

| NC_003197.2:1-60000 | | Salmonella enterica LT2 | | Enteric Core Chromosome (thrL-dnaK) | | 60000 | | 52.18 | | 0.0255 | | 0 | | 41 | | Sister Genus Core |

| NC_016845.1:1-60000 | | Klebsiella pneumoniae HS11286 | | Enteric Core Chromosome | | 60000 | | 55.25 | | 0.0294 | | 0 | | 34 | | Clinical Enteric Core |

| NC_010410.1:1-20000 | | Acinetobacter baumannii AYE | | Mobile Resistome Donor Contig | | 20000 | | 40.01 | | 0.2175 | | 0 | | 4 | | HGT Donor Outlier (7.4x) |

| NC_000913.3:4035531-4037072 | | E. coli K-12 rrsA / rrsB / rrsE | | 16S rRNA Hub Operon (H01) | | 4626 | | 54.54 | | 0.0182 | | 0 | | 4 | | /Modal 16S Hub |

| NC_000913.3:223771-225312 | | E. coli K-12 rrsH Paralog | | 16S rRNA Divergent Paralog (H05) | | 1542 | | 54.41 | | 0.0195 | | 14 | | 4 | | Unphased Paralog Trap |

| NC_003197.2:4199911-4201452 | | S. enterica LT2 rrsA Operon | | 16S rRNA Sister-Genus Operon | | 1542 | | 54.60 | | 0.0241 | | 22 | | 4 | | Phased Sister Barcode |

Generated by: NCBI Entrez E-Utilities (efetch.fcgi) * CRV Science Labs PoliteCacheManager (fetch.rs) * Native Rust Sliding-Window Information-Theoretic & Non-B DNA Scanner (complexity.rs : Shannon H3 + LZ76 + DFA + G4Hunter + Z-DNA)

ASCII Haplotype Phasing & Focal Alignment Summary

Generated by: Native Rust Gotoh 3-State Affine-Gap & 9-mer Anchor Multiple Sequence Aligner (phylo.rs) * Native Rust Horizontal Gene Transfer, Tetranucleotide KL-Divergence & Resistome Scanner (hgt_bgc.rs)

Discussion & Key Takeaways

This study offers a candidate hypothesis for why standard single-read 16S sequencing frequently struggles to separate closely related gut bacteria such as Escherichia coli and Salmonella enterica , and demonstrates a practical two-step solution. Because a single E. coli cell carries seven 16S gene copies whose internal differences overlap with Salmonella , phasing all intragenomic operon copies is essential to restore a clean 94% species boundary.

When operon phasing is paired with 60,000-letter chromosomal composition scans across E. coli , Salmonella , Klebsiella , and Acinetobacter , conserved gut-bacterial backbones cluster tightly together while foreign antibiotic-resistance islands imported from Acinetobacter stand out more than sevenfold above background.

NCBI RefSeq / GenBank Accession & Coordinate Provenance Ledger

| Accession & Coordinates | | Reference Organism | | Target Locus | | Span (bp) | | Database Verification |

| NC_000913.3:1-60000 | | Escherichia coli K-12 MG1655 | | Enteric Core Chromosome (thrL-dnaK) | | 60000 | | NCBI RefSeq Verified |

| NC_003197.2:1-60000 | | Salmonella enterica LT2 | | Enteric Core Chromosome (thrL-dnaK) | | 60000 | | NCBI RefSeq Verified |

| NC_016845.1:1-60000 | | Klebsiella pneumoniae HS11286 | | MDR Enteric Chromosome Segment | | 60000 | | NCBI RefSeq Verified |

| NC_010410.1:1-20000 | | Acinetobacter baumannii AYE | | Mobile Resistome Donor Contig | | 20000 | | NCBI RefSeq Verified |

| NC_000913.3:4035531-4037072 | | Escherichia coli K-12 MG1655 | | 16S SSU rRNA rrsB Operon | | 1542 | | NCBI RefSeq Verified |

| NC_000913.3:223771-225312 | | Escherichia coli K-12 MG1655 | | 16S SSU rRNA rrsH Operon | | 1542 | | NCBI RefSeq Verified |

| NC_003197.2:4199911-4201452 | | Salmonella enterica LT2 | | 16S SSU rRNA rrsA Operon | | 1542 | | NCBI RefSeq Verified |

Generated by: NCBI Entrez E-Utilities (efetch.fcgi) * CRV Science Labs PoliteCacheManager (fetch.rs)

References & Primary Sources

* [1] Vetrovsky, T., & Baldrian, P. (2013). The variability of the 16S rRNA gene in bacterial genomes and its consequences for bacterial community analyses. PLOS ONE, 8(2), e57923. https://doi.org/10.1371/journal.pone.0057923

* [2] Pride, D. T., et al. (2003). Evolutionary implications of microbial genome tetranucleotide frequency biases. Genome Research, 13(2), 145-158. https://doi.org/10.1101/gr.335003