

# CHARACTERIZATION OF MULTIRESISTANCE CASSETTE FOUND IN *SALMONELLA* ENTERITIDIS



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## Introduction

*Salmonella* Enteritidis is the main serovar causing animal-linked food poisoning. *S. Enteritidis* is predominantly antimicrobial susceptible, with a percentage of strains showing chromosomal mutation corresponding to quinolone resistance. Antimicrobial resistance (AMR) is a significant problem, and its increase is associated with negative health and economic effects. Gene mobilization via plasmids is one of the mechanisms of horizontal gene transfer (HGT). Monitoring of foodborne pathogens for AMR with transmission potential is crucial for food safety and public health. Thus in this study, we performed a hybrid assembly of the multiresistant *Salmonella* Enteritidis strain to assess mobilization and potential for transmission of antimicrobial resistance genes (ARGs).

## Methods

*Salmonella* Enteritidis strain was isolated from broiler farm boot swab in 2010. The strain was retrieved from the bacterial culture collection of National Veterinary Research Institute (PIWET). DNA was isolated on Maxwell RSC48 and sequenced on MiSeq Illumina platform (V3 2x300bp kit) and MinION Mk1B sequencer (R9.4.1 flow cell). Hybrid assembly was performed using Unicycler v0.4.8. The presence of ARGs and plasmids was tested using Abricate v.1.0.1 (GitHub) with databases: CARD, and PlasmidFinder. Multilocus sequence types (MLST) were established by mlst (GitHub). Genome annotation was done with Proksee and RAST web servers.



Figure 1. General scheme of the study.

## Results

Studied *Salmonella* Enteritidis belonged to ST11. Reconstruction of the genome revealed chromosome (4 684 438 bp) and two plasmids: IncI1-I(Alpha) 107 354 bp (Figure 2) and IncFII(S) 59 372 bp. **IncI1-I(Alpha)** plasmid carried the following resistance determinants: broad-spectrum class A beta-lactamase (**blaTEM-135**), class A tetracycline efflux pumps (**tet(A)**), resistance to sulfamethoxazole (**sul2**), and resistance to biocides (permease of the drug/metabolite transporter (**DMT**) superfamily). In addition, a gene associated with biofilm formation (**GlmM**) and phage tail protein were detected (Figure 2, 3).

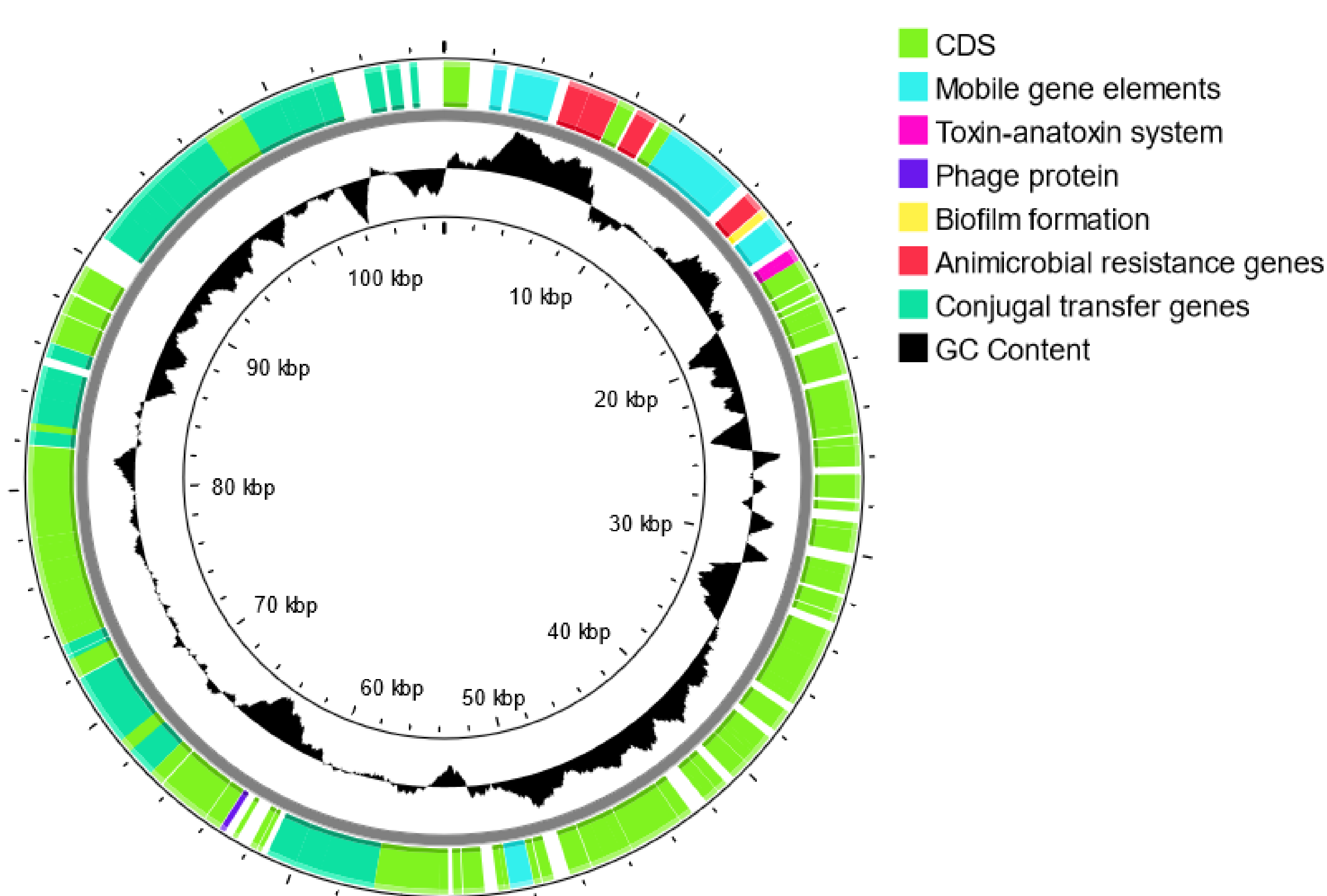


Figure 2. Composition of IncI1-I(Alpha) plasmid, annotated with Proksee online tool (Prokka and CARD database) and RAST. CDS - coding sequences.

Identified ARGs were closely located, constituting a resistance cassette, that was flanked by mobile genetic elements - insertion sequence (**IS91**) and transposons (**Tn3**).

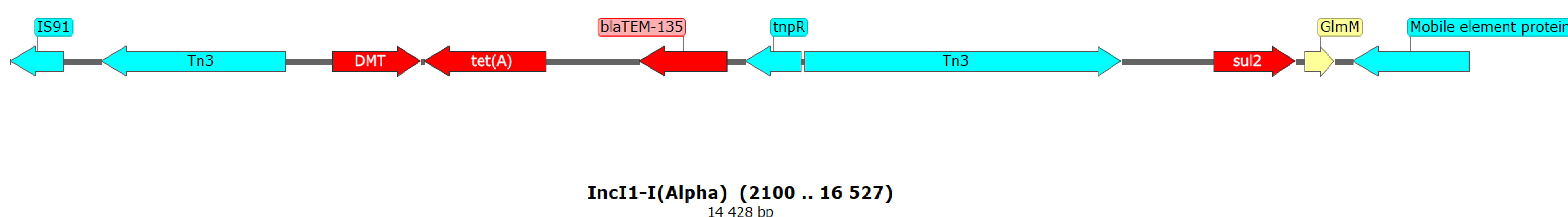


Figure 3. Multiresistance region of IncI1-I(Alpha) plasmid with gene cassette. Visualization made in SnapGene.

## Conclusion

ARGs found in tested *Salmonella* Enteritidis strain can be plasmid-mediated which raises concern due to the high risk of dissemination. Here, using a combination of short- and long-read sequencing, we were able to reconstruct chromosome and plasmids, and thus confirming the possible mobilization of ARGs. The genetic context is essential for understanding the emergence and dissemination of antimicrobial resistance.