

Third-generation cephalosporin and fluoroquinolone resistance in *Salmonella enterica* associated with antimicrobial resistance genes

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Key words: *Salmonella enterica*, WGS, third-generation cephalosporin resistance, *blaCTX-M*

Motivation and Aim: *Salmonella enterica* is one of the most important global food-borne pathogens. Since ciprofloxacin (belonging to fluoroquinolones), ampicillin or third-generation cephalosporins (beta-lactams) are the common first-line antimicrobial drugs for the treatment of salmonellosis [1] it is important to study molecular mechanisms of resistance to these drugs and to monitor the dissemination of antimicrobial resistance (AMR) determinants. Thus, the goal of this work was to perform genomic analysis ciprofloxacin- and third-generation cephalosporin-resistant (CIP-3rd) isolates of food origin based on whole-genome sequencing (WGS).

Methods and Algorithms: Totally 10 CIP-3rd *Salmonella* isolates were obtained in Russia in 2021 from poultry products (n = 6), meat (n = 1) and ready-to-cook products (n = 4). Samples were received from Samara region (n = 1), Tula region (n = 1), Chelyabinsk region (n = 1), Magadan region (n = 1), Vologda region (n = 1), Primorsky Krai (n = 1), Sverdlovsk region (n = 2) and Novosibirsk region (n = 2). The isolates were tested for phenotypic antimicrobial resistance by the boundary concentration method on VITEK MS analyzer. WGS was performed on the Illumina NextSeq platform using Nextera DNA Sample Prep Kit (Illumina, USA). Genome assemblies and comparisons were performed using SPAdes, roary, dnadiff, and custom software.

Results: The majority of the analyzed *S. enterica* isolates belonged to ST32 and serovar O7 (n = 6), two were typed as ST548/O21, ST469/O7, while ST11/O9 was revealed in one isolate. All 10 isolates studied were characterized by AMR to ceftazidime (CZD) and ceftriaxone (CRO), which corresponds well to the revealed AMR determinants according to WGS data obtained. So, all *S. enterica* (ST32) carried *blaCTX-M-14* gene which was responsible for 3rd-generation cephalosporins resistance [2-4]. ST548, ST469, and ST11 carried β -lactamase genes *blaCMY-2*, *blaCTX-M-1*, and *blaCTX-M-55* respectively. Eight of the isolates presented also possessed resistance to cefepime (FEP) which belongs to fourth-generation cephalosporins. All FEP-resistant samples carried *blaCTX-M* gene family despite the different sequence types. Such dissemination raises concerns and requires further epidemiological surveillance of AMR transmission within *S. enterica* and investigations of the molecular mechanisms of resistance acquisition.

Moreover, all the isolates observed had ciprofloxacin (CIP) resistance. However, the presence of AMR genes to fluoroquinolones (*qnrB19*) and quinolones (*qnrS1*) was revealed only in two and three isolates, correspondingly. We suggest that these

distinctions could be associated with point mutations in a chromosomal virulence gene *gyrA* being the most common mechanism for resistance to quinolones in *Salmonella* [5]. *In silico* search for plasmids showed that all CIP-3rd ST32 samples were characterized by the presence of IncFIB plasmid, while ST548 carried IncC, ST469 had several types of plasmids simultaneously (Col(pHAD28) and IncI1), which was also revealed in ST11 (IncFIB, IncFII, IncI1).

Conclusion: Applying WGS technology enabled us to obtain unique genome features of foodborne CIP-3rd *S. enterica* isolates. Such features are important both from the standpoint of an increased level of AMR of the pathogens studied and evolutionary changes. The data obtained will contribute to further investigations in epidemiological surveillance of foodborne *S. enterica* isolates.

Acknowledgements: The study was supported by the industry research program of Rospotrebnadzor (2021–2025) “Scientific support of epidemiological monitoring and public health measures of the Russian Federation territory” and Russian Federation Government Resolution No. 3116-r.

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