



POSTER

SHIGA-TOXIN-PRODUCING ESCHERICHIA COLI ISOLATED FROM FOOD IN LOMBARDY REGION: GENETIC DIVERSITY AND ANTIMICROBIAL RESISTANCE

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Aim. Shiga toxin-producing *Escherichia coli* (STEC) can cause a wide spectrum of clinical manifestations in humans, ranging from mild diarrhea to hemolytic uremic syndrome (HUS), posing a significant threat to public health and healthcare systems worldwide. According to EFSA, STEC is the fourth leading cause of foodborne illness in the European Union, despite being underdiagnosed and underreported. Ruminants, particularly cattle, serve as the natural reservoir for these bacteria, and the ingestion of contaminated food is considered the primary transmission route to humans. This study aimed to characterize the genetic diversity and antimicrobial resistance (AMR) profiles of STEC strains isolated from food products in Lombardy region between 2018 and 2026 to provide comprehensive insights into their distribution and resistance patterns to support evidence-based control, surveillance, and prevention strategies.

Methods. STEC detection was performed in accordance with the ISO/TS 13136:2012 + ISS-EU-RL VTEC Met 04 Rev 2 2021, using a real-time PCR targeting the *stx1* and *stx2* genes (encoding Shiga toxins 1 and 2), as well as the *eae* gene (encoding intimin). Positive samples underwent an additional real-time PCR to detect the serogroups most frequently associated with human infections in Europe (O111, O26, O103, O145, O157, and O104). STEC strains were isolated, and whole-genome sequencing (WGS) was performed on the Illumina MiSeq platform. Using the ARIES Galaxy Server, serogroups, virulence genes, and AMR genes were determined.

Results. A total of 162 STEC strains were isolated, mainly from matrices of bovine origin (cheese - 34%, milk - 17%, and meat - 11%). Virulence gene profiling revealed that

stx1 was present in 40% of the isolates, while *stx2* was detected in 37%. Notably, 78% of the strains harbored the *eae* gene which, in association to *stx* genes, confers more virulence to the strains. In fact, the co-occurrence of *stx2* and *eae* is generally correlated with severe clinical outcomes in humans. A variety of 43 different serogroups was identified among the isolates, suggesting a diversified circulation of strains within the environments of food production. In particular, 16% of the isolates belonged to 4 historically prevalent serogroups (O26 - 9%, O103 - 5%, O145 - 0.6%, O157 - 0.6%). No O111 or O104 strains were detected. Globally recognized emerging serogroups (O80, O113, and O128) were found in 16% of the isolates, underscoring the need of a continuous monitoring of the circulating serogroups, to assess their potential impact on public health. Regarding AMR, 50% of the strains exhibited resistance to at least one antimicrobial agent, and 22% were classified as multi-drug-resistant (MDR, resistance to ≥ 3 antimicrobial classes). Crucially, one MDR strain exhibited resistance to azithromycin, a bacteriostatic molecule that seems promising in preventing HUS progression in pediatric patients. The highest resistance rates were observed against β -lactams (61.4%), streptomycin and kanamycin (18.5%), and tetracycline (14.3%), emphasizing the importance of appropriate measures to control antibiotic use in livestock and of rigorous surveillance of AMR profiles in foodborne pathogens.

Conclusions. The genetic diversity of STEC isolated from food products in Lombardy region emphasizes the necessity of continuous integrated surveillance through a One Health approach to support Competent Authorities in developing targeted intervention strategies aimed at mitigating foodborne risks and protecting public health.