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Population structure of nontyphoidal Salmonella causing human infections by Multi Locus Sequence Typing

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Salmonellosis is one of the most common foodborne infections in Europe and multidrugresistant (MDR) Salmonella is emerging worldwide with increasing involvement of particular clones. Molecular typing methods are powerful tools in surveillance and outbreak investigation of Salmonella infections. In addition, Multi Locus Sequence Typing (MLST) is useful to obtain data on the evolution and population structure of contemporary Salmonella circulating from animals to food products and to humans in different regions and to increase the knowledge on the diversity of Salmonella epidemic lineages. The aim of our study was to assess genetic relationships between representative Salmonella isolates associated with human infections in different regions of Portugal and Brazil by using the technique of MLST with further analysis with the e-BURST software. The seven-locus scheme recommended in the Salmonella MLST database (http://mlst.ucc.ie/) was applied to 14 isolates from Brazil from different sources and 40 isolates from a large Portuguese collection (2002-2008), representative of MDR clones from human infections (n=22), food (n=14), and piggeries (n=4). The DNA was extracted using the InstaGene Matrix Kit™. PCR amplification was done with the AmpliTaq Gold® followed by purification. The Portuguese isolates studied represented 15 serotypes that accounted for 24 types of PFGE and 15 different STs. Most S. Typhimurium (n=13, 5 PFGE types) belonged to the worldwide spread ST19 (n=12) and to its SLV ST313 (n=1). They included the 3 most widespread clones in Portugal (DT104, OXA-30/CMY-2 and sul3) carrying different integron and plasmids types. The globally disseminated ST11 (n=4, 2 PFGE types) was identified in all S. Enteritidis isolates, including one carrying qnrS and belonging to a major clone. Isolates belonging to the emerging S. Rissen clone (n=5, 1 PFGE type) were assigned to the singleton ST469, only previously reported in European isolates of the same serotype. The remaining MDR isolates of different serotypes were assigned to STs belonging to already known clonal clusters (ST15, ST27, ST32, ST40, ST82 and ST334) or singletons (ST48, ST64, ST102, ST306 and ST358). Regarding the Brazilian isolates studied, they represented 5 serotypes which corresponded to 8 PFGE types and 7 different STs. As for the Portuguese isolates, all S. Typhimurium (n=5) and S. Enteritidis (n=2) belonged to worldwide spread clonal lineages, ST19 (n=3) or its SLV ST313 (n=2) and ST11, respectively. The remaining isolates of different serotypes were included in the already known ST10 (n=1), ST23 (S. Oranienburg; n=1), ST48 (S. Panama; n=2) and ST524 (S. Give; n=3). It is of note that most isolates of a particular serotype were assigned to a specific ST. This is the first study describing the population structure of nontyphoidal Salmonella in Portugal and Brazil. Our results confirm the prevalence of particular clusters enclosing major MDR clones that cause human infections, namely ST19 for S. Typhimurium and ST11 for S. Enteritidis. The selection and worldwide spread of particular STs might be influenced by the acquisition and horizontal transfer of specific resistance genes and genetic elements.

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