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Extended-spectrum beta-lactamase (ESBL) producing and ciprofloxacin-resistant Escherichia coli intestinal colonization of Brazilian cattle

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Background: Bacterial resistance in farm animals is currently one of the main targets of investigation by the scientific community, because of their relevance and impact on global public health. In this context, extended-spectrum beta-lactamases (ESBL) are important enzymes due to their ability to degrade oxyimino-beta-lactam antibiotics. Resistance to quinolones, namely ciprofloxacin, is important once quinolones are of the most used non beta-lactam antibiotics. Among animal production, cattle is an important component to the food chain, responsible for one high percentage of animal protein consumed around the world. In this context Brazil is one the biggest producers. Among the microorganisms showing bacterial resistance in farm animals, *Escherichia coli* is relevant considering it is the main facultative anaerobic commensal in animal intestinal flora. This study aimed to evaluate the presence of multidrug resistant *Enterobacteriaceae* in feces of healthy cattle in Brazil.

Material/methods: Isolates were obtained from bovine fecal samples of 22 productions in the North of Brazil. Selection of the isolates was performed on MacConkey agar with different antibiotics (beta-lactams, tetracycline, and ciprofloxacin). Samples were previously incubated in TSB. Susceptibility testing was achieved by disk diffusion method according to CLSI. Presumptive identification was performed with CHROMagar Orientation. ESBL producers were screened by the double disk synergy test. Resistance genes were screened by PCR with specific primers for ESBL: bla_{TEM}, bla_{OXA}, bla_{SHV} and bla_{CTX-M}; and for quinolone resistance (QR): *aac6'Ib-cr*, *qnrA*, *qnrB*, *qnrC*, *qnrD*, *qnrS*, *gyrA* and *parC*.

Results: From 188 cattle fecal samples, 267 *Enterobacteriaceae* isolates were obtained, showing resistance to amoxicillin (59.9%), amoxicillin+clavulanic acid (14.2%), cefotaxime (30.7%), tetracycline (73%), sulfamethoxazole+trimethoprim (44.9%), ciprofloxacin (33.7%), chloramphenicol (14.9%) and gentamicin (4.4%). A total of 176 isolates were multi-drug resistant (MDR). Twenty four of the selected

Enterobacteriaceae isolates (12.7%) were ESBL-producing and QR *Escherichia coli* (EC-ESBL/QR). Thirty five of the total fecal samples (8%) showed ESBL and QR producers with the following resistance genes profile: Beta-lactamase: bla_{SHV} (20.8%); bla_{CTX-M} Group 1 (G1) and bla_{OXA} (8.3%); bla_{TEM} and bla_{SHV} (8.3%); bla_{TEM} (8.3%); bla_{CTX-M} G1 and bla_{TEM} (4.2%); bla_{CTX-M} G1 (4.2%); bla_{CTX-M} Group 2 and bla_{OXA} (4.2%); bla_{CTX-M} Group 8 and bla_{SHV} (4.2%). QR: *gyrA* and *parC* (58.3%); *qnrB*, *gyrA* and *parC* (25%); *aac6'Ib-cr* and *parC* (8.3%); *aac6'Ib-cr*, *gyrA* and *parC* (4.2%); and *qnrS*, *gyrA* and *parC* (4.2%). The prevalent combination was bla_{SHV} + *gyrA* and *parC* (16.7%). Results showed the prevalence of bla_{CTX-M} present in 25% of EC-ESBL/QR. ESBL producers show different beta-lactamase types associated to quinolone resistances genes.

Conclusions: Results show a prevalence of ESBL-producing *E.coli*, presenting CTX-M and QR phenotype, colonizing these animals. Farm animals may act as reservoirs of MDR bacteria, highlighting the relevance of the "One Health" approach of this reality.