

TITLE: EMERGENCE OF COLISTIN RESISTANCE IN *E. coli* FROM HEALTHY PRODUCTION POULTRY IN RIO DE JANEIRO STATE, BRAZIL.

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ABSTRACT: The emergence of *mcr* genes associated with colistin resistance in *Enterobacteriaceae* has gained considerable attention as a significant concern threatening the use of a last resort drug for treating human disease. Colistin's use as growth promoter in small poultry farms was a reality in Brazil until 2016. In this study, a collection of 107 *E. coli* isolates from production poultry was examined for the presence of *mcr-1* and *mcr-2*. The isolates were collected between April 2015 and June 2016 from 30 to 41-day-old broiler chickens and 62 week old free range layer hens from poultry farms in the state of Rio de Janeiro, Brazil. All isolates were recovered from the trachea and cloaca of healthy birds and two additional isolates were recovered from two chickens diagnosed with colibacillosis (Blepharitis B46; pericarditis B157). All isolates were screened for the presence of *mcr-1* and *mcr-2* using PCR. Additionally, PFGE analysis, APEC virulence associated gene screening, replicon type analysis and additional antimicrobial resistance analysis and resistance gene screening were carried out in order to further characterize these isolates. The *mcr-1* gene was detected in a total of 62 isolates (61 healthy and 1 APEC); while *mcr-2* was not detected in any isolate. The *mcr-1* positive isolates were resistant to colistin using the agar dilution assay (>8ug/ml). PFGE analysis found that most of the isolates had unique fingerprints suggesting that the emergence of colistin resistance was not the result of clonal dissemination. We found in the *mcr-1* positive isolates that *Incl2*, *FIB* and *B/O* (38, 36 and 34% respectively) were the most prevalent replicon types detected; *tetA* and *tetB* (32 and 26% respectively) were the most prevalent antimicrobial resistance genes and *iutA*, was the most prevalent APEC virulence associated gene, present in 50% of the isolates, and 32% of the isolates examined could be considered APEC-like, based on the presence of 3 or more APEC virulence associated genes (*iroN*, *ompT*, *hlyF*, *iss*, *iutA*). This study has identified a high prevalence of *mcr-1* in poultry isolates, and suggests that animal husbandry practices could result in a potential source of resistance to the human food chain in countries where application of colistin in animal health is practiced. Emergence of the *mcr-1* gene and associated colistin resistance in production poultry warrants continued monitoring from the animal health and human health perspective.

Keywords: *mcr-1*, colistin, *Escherichia coli*, antimicrobial resistance.

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