Letter to the editor: *Escherichia coli* harbouring *mcr-1* gene isolated from poultry not exposed to polymyxins in Brazil

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To the editor: The recent paper by Fernandes et al. [1] described the presence of the *mcr-1* gene in *Escherichia coli* from pigs and poultry in Brazil. The authors stated that microbiology laboratories worldwide should be aware of *mcr-1* isolates resistant to polymyxins in patients living in or returning from Latin American countries and highlighted that the *mcr-1* gene dissemination results from polymyxins' misuse as growth promoter in food animals. In view of the concerning spread of antibiotic resistance, we screened *E. coli* isolates obtained from a poultry slaughterhouse in southern Brazil with official reports on antibiotic use.

Poultry rectal swabs were collected between August and October 2015. A total of 340 chickens farmed in Brazil and belonging to 17 batches were included in the study. All poultry had received bacitracin, narasin and nicarbazin during a first period of life (between the 2nd and the 18th day) and avilamycin and salinomycin during a second period (between the 20th and 35th day); the chickens of batches 10 and 11 had also received doxycycline during a total of 3 days, in the second period of life. Poultry included in this study were not exposed to polymyxin during their entire life (around 40 days).

A total of 343 isolates were evaluated by polymerase chain reaction (PCR) for the *mcr-1* gene [2] and 10 (3%) were positive. The *mcr-1* gene was confirmed by sequencing the PCR amplicon. The *mcr-1* positive isolates were obtained from 10 different chickens belonging to three batches from three different breeders. The polymyxin B minimum inhibitory concentrations (MIC) of the 10 *mcr-1* positive isolates were 2 mg/L (8 isolates), 1 mg/L and 0.25 mg/L and they could be classified as susceptible to polymyxin B, according to the European Committee on Antimicrobial Susceptibility

Testing (EUCAST) (resistance >2 mg/L). In contrast, most reports indicate that the *mcr-1* gene is usually found in isolates presenting resistance to polymyxins [2-6].

The *mcr-1* positive isolates were submitted to DNA macrorestriction typing by pulsed-field gel electrophoresis (PFGE) and five isolates, from the same batch, proved to be clonally related while the other five isolates were unrelated. Conjugation experiments with the *E. coli* J53 were successful for two *mcr-1* positive isolates which confirmed that the *mcr-1* gene was located in a plasmid. The transconjugants presented positive results by PCR for the *mcr-1* gene and had a polymyxin B MIC of 2 mg/L.

According to Brazilian law, all slaughterhouses must submit in advance to the Federal Inspection Service of the Ministry of Agriculture, the bulletin of health of each batch of animals to be slaughtered. It is of note that the chickens evaluated in this study have received antibiotics as growth promoters, but polymyxins were not included among these compounds. This goes against the hypothesis that the emergence of the *mcr-1* gene is linked to the use of polymyxins in animal feed in Brazilian livestock [1] and suggests that other compounds or factors may also be involved in the selection of this gene.

Finally, the fact that the *mcr-1* was originally described in China and thereafter in several other countries including Europe indicates that this gene is already widespread in the world. Therefore, isolates with *mcr-1* should be considered in any patient, regardless of whether they were living in or returning from Latin America or not.

*Authors' correction

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Conflict of interest

None declared.

Authors' contributions

Conceived the project: AFM; Managed sample collection: SAML, ASM, AFM; Performed laboratory investigations: SAML, DLM, LSN, VMLC; Drafted the article: DLM; Revised the article: APZ, ALB, AFM.

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