

# Colistin resistant *Escherichia coli* and spread of ESBL *E.coli* strains in samples collected from organic, antibiotic-free and conventional broilers in farms and at slaughter.



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## Introduction



The increase of antibiotic resistance is a global concern for humans and animal health. The use of antibiotics in poultry production have increased the selection pressure for antibiotic-resistant bacteria [1]. *Escherichia coli* is a commensal bacterium, ubiquitous in animals and humans and extended-spectrum  $\beta$ -lactamase (ESBL)/AmpC *E. coli* is frequently found as contaminant in broiler meat [2]. Furthermore, ESBL-producing *E. coli* are often resistant to fluoroquinolones, aminoglycosides and colistin. Currently, this antimicrobial previously used for prevention, treatment, metaphylaxis and growth promotion [3] is off labeled. Moreover it has recently been reassessed in human therapy following the diffusion of «superbug» bacteria.

## Aim



We aim to highlight the susceptibility of *E. coli* towards colistin and the spread of ESBL *E. coli*, isolated in the different typology of farming (conventional, antibiotic-free, organic) and in 2 slaughterhouses located in Central Italy.

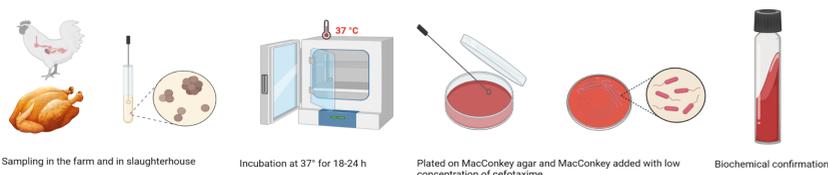
## Materials and Methods



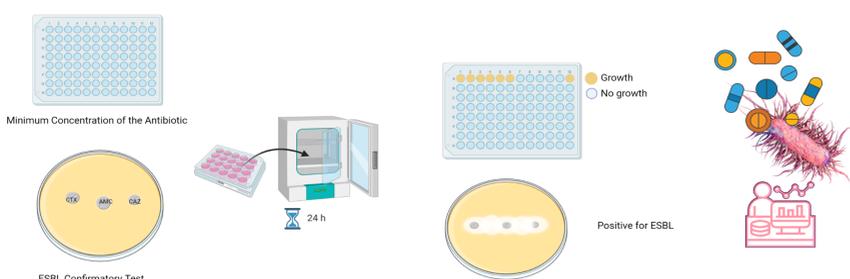
A total of 174 *E. coli* strains were isolated in farms and at slaughterhouses. To assess the antimicrobial susceptibility to colistin all *E. coli* isolates were analyzed by the minimum concentration of the antibiotic (MIC) using Euvsec plates. The susceptibility to colistin was evaluated using the FRCOL Plates (0.12-128  $\mu$ g/mL).

All results were evaluated through EUCAST guidelines. The presence of ESBL producing *E. coli* was confirmed by the combined disk test with cefotaxime and ceftazidime alone and in combination with clavulanic acid and by the microdilution method using Sensititre ESBL plates, according to EUCAST guidelines.

### Isolation and Identification of *E. coli*



### Colistin Susceptibility Testing and ESBL Detection

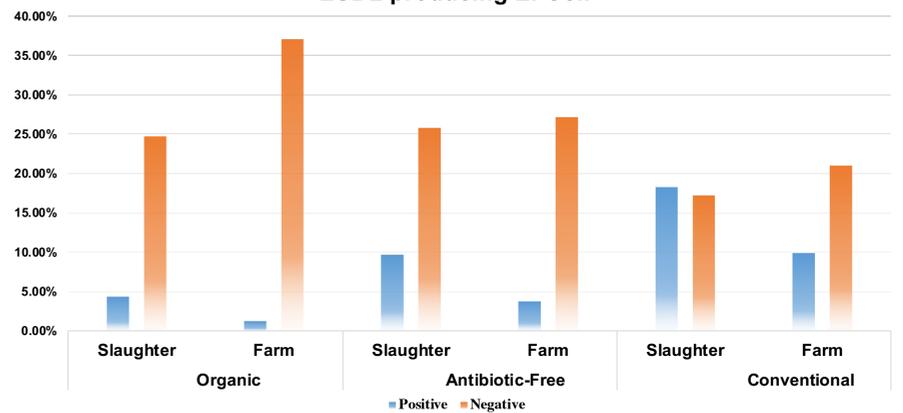


## Results



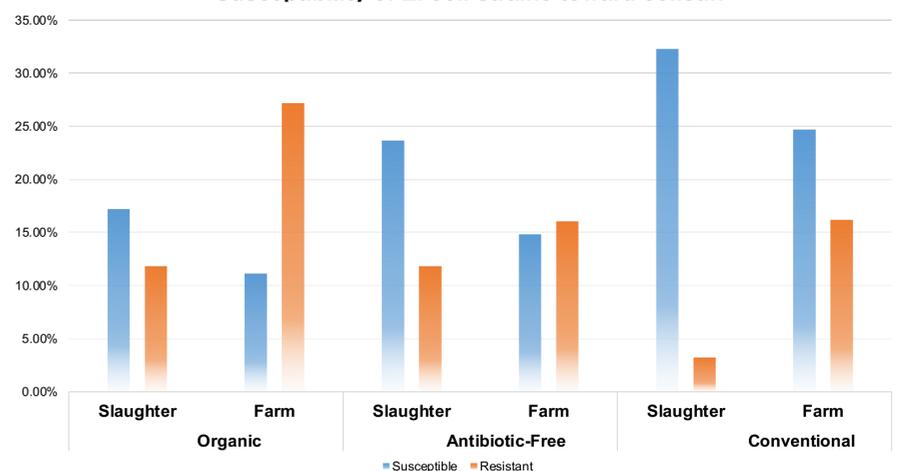
The number of ESBL *E. coli* strains was higher in slaughterhouses (OR 2.72; IC95%: 1.23-6.02) than in farms and samples from conventional carcasses presented the highest percentage of ESBL *E. coli* (18.27%). No statistical differences were found between organic and antibiotic free sample's carcasses (4.30%; 9.67%).

ESBL producing *E. Coli*



The highest number of colistin-resistant *E. coli* strains was found in the organic (OR 8.27; IC 95%: 3.26-20.96) and antibiotic-free (OR 4.73; IC 95%: 1.86-12.05) rearing systems.

Susceptibility of *E. coli* strains toward colistin



## Conclusions



Conventional carcasses showed the highest number of ESBL *E. coli* in relation to the large use of  $\beta$  lactams against *C. perfringens* infection in conventional farming, especially ampicillin. The absence of antibiotic treatment in organic and antibiotic-free farmings, in comparison to conventional one, could have favored the increase of the resistant *E. coli* strains in an intestinal microbiota "less pressured" by antimicrobials use. It should be also highlighted that the therapeutic protocol in conventional management is based on the use of other antimicrobial classes being the use of colistin off-labeled. Furthermore, as well as the possibility of vertical transmission of antimicrobial-resistance genes in all rearing systems, the role of the external environment contaminated also by antimicrobial resistant bacteria shed by wild animals should not be neglected in organic farms.

## REFERENCES

- [1] Diarra M. Antibiotics in Canadian poultry productions and anticipated alternatives. *Front. Microbiol.* 5:15, 2014. [2] Musa L. Antimicrobial Susceptibility of *Escherichia coli* and ESBL-Producing *Escherichia coli* Diffusion in Conventional, Organic and Antibiotic-Free Meat Chickens at Slaughter. *Animals (Basel)*. 10:1215, 2020. [3] EMA. Updated advice on the use of colistin products in animals within the European Union: development of resistance and possible impact on human and animal health. 2016. [4] Kempf I. Colistin use and colistin resistance in bacteria from animals. *Int. J. Antimicrob. Agents*, 48, 2016.

