

## COLISTIN RESISTANCE IN *Escherichia coli* ISOLATED FROM BOVINE MILK AND FECES

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### INTRODUCTION

Antimicrobial resistance is one of the major concerns worldwide. Animal production has been pointed as an important source of resistant strains, since several antimicrobial bases are indiscriminately used to treat and prevent infectious diseases in herds. In dairy industry, bovine mastitis is the most prevalent disease, and it is responsible for the higher use of antimicrobials in dairy farms. Among the bacterial species that can cause this illness is *Escherichia coli* that has a high potential to acquire resistant phenotype, besides having great importance to public health, since it is a zoonotic pathogen.

The aim of this study was (1) assess the susceptibility profile to 12 antimicrobials, and (2) evaluate the presence of plasmid-mediated colistin resistance genes in of *E. coli* isolated from milk acquired from cows with mastitis and from the environment of dairy farms localized in Minas Gerais, Brazil.

### MATERIALS AND METHODS

Eighty-eight *E. coli* isolates were used in this study, including 57 strains isolated from milk of cows with mastitis, and 31 from bovine feces. The strains were obtained between 2004 and 2016 from farms localized in Minas Gerais, Brazil, and belong to the Microorganisms Collection from the Laboratório de Bacteriologia (Bacteriology Laboratory), Departamento de Medicina Veterinária, Universidade Federal de Lavras (UFLA). Twelve antimicrobial agents (Table 1) were used to assess the susceptibility profile and the minimal inhibitory concentration (MIC) of the isolates, employing the microdilution technique, according to the M100 and VET01S manual of the Clinical and Laboratory Standards Institute (CLSI). Strains were considered multidrug resistant (MDR) when they were resistant to three or more antimicrobial groups. A multiplex PCR assay were performed for detection of *mcr-1*, *mcr-2*, *mcr3* and *mcr-5* genes in all studied strains (Rebello et al, 2018).

### RESULTS AND DISCUSSION

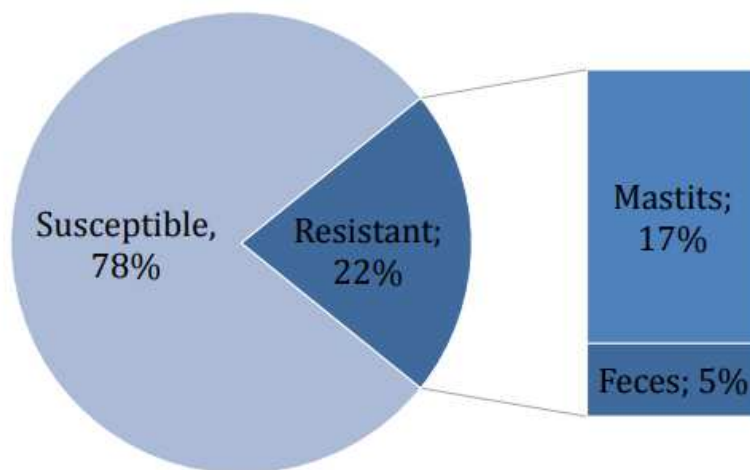
Results showed elevate levels of resistance (Table 1), with fifty-seven isolates [64.77% (57/88)] resistant to at least one antimicrobial tested, and 16 [18.88% (16/88)] multidrug resistant (Figure 1). Cephalosporins, phenicols, sulfonamides and tetracyclines are drugs commonly used in dairy farm routine and these results sustain that continuous exposure to antimicrobial principles generate selective pressure and, consequently, resistant strains. In addition, our results also showed high resistance to colistin, a drug not usually used in bovine treatment in Brazil, however, none of the *mcr* alleles prospected were found. A possible explanation for this result is the resistance mediated by the other alleles, as *mcr-9.1*, that was already reported in Brazil (Daza-Cardona et al., 2021). Nevertheless, other mechanisms associated with colistin resistance have already been reported, as drug efflux pumps and phenotypic alterations in the membrane cell (Godry et al., 2021).

Table 1 - Frequency of resistant *Escherichia coli* isolated from bovine milk and feces to several antimicrobial drugs, Minas Gerais, 2004-2016

Antimicrobial drug	% resistant isolates
Amikacin	0.00%
Ampicillin	7.95%
Cefazolin	12.50%
Cefoxitin	6.82%
Ceftiofur	6.82%
Ciprofloxacin	0.00%
Chloramphenicol	1.14%
Colistin	21.59%
Florfenicol	38.64%
Gentamycin	0.00%
Sulfamethoxazole/ Trimethoprim	18.18%
Tetracycline	17.05%

These mechanisms are more generic and can promote resistance to other antimicrobials principles, as antiseptics, for example (Mc Carlie, Boucher and Bragg, 2020). Furthermore, it is possible to speculate the role of the constant exposure to antiseptic in the milking routine (dipping) as a source of cross resistance to colistin and other antimicrobials, since mastitis isolates showed higher resistance levels than feces isolates (Figure 1). Additionally, the resistance to colistin is a public health concern, once colistin is a drug used as last-resource treatment for multidrug resistant Gram-negative infections in human.

Figure 1 - Colistin resistance in *Escherichia coli* isolated from bovine milk and feces, Minas Gerais, 2004-2016.



#### CONCLUSIONS AND FINAL CONSIDERATIONS

*E. coli* strains isolated from dairy cattle showed great levels of resistance to several drugs, including colistin, which draws attention to indiscriminate use of antimicrobials in animal production and to cross resistance with drugs important for human health.

#### BIBLIOGRAFIC REFERENCES

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