

GENETIC CHARACTERISATION OF MULTI-DRUG RESISTANT ENTEROBACTERIACEAE ISOLATED FROM WASTEWATER FROM IRISH FARMS.

Main author: Tetiana Garkavenko (State Scientific and Research Institute of Laboratory Diagnostics and Veterinary and Sanitary Expertise)

Co-authors: Olha Horbatiuk, Valentina Andriyashchuk

INTRODUCTION

The World Health Organization's Global Strategy for Containment of Antimicrobial Resistance (WHO, 2001) provides an action plan and preventive measures to prevent the emergence of new resistant bacteria and the spread of microorganisms with acquired antibiotic resistance. One of the priorities of this plan is to monitor antibiotic resistance in isolates from animals and humans.

The main field of application of antibiotics and the source of the spread of antibiotic resistant bacteria is not considered to be medicine but animal husbandry. After antibiotics had been added to feeds for more efficient conversion of feed to livestock and poultry products, and to improve husbandry, for a long time, the selective pressure of antibiotics contributed to the formation of resistance in commensal bacteria and pathogens, and diseases caused by them became very difficult to treat.

Therefore, an active monitoring of antimicrobial resistance in isolates of animal microorganisms, including commensal ones, circulating among farm animals and isolated from food of animal origin is relevant because it has strategic importance.

METHODOLOGY

The antimicrobial agents ampicillin (10 µg), ceftazidime (10 µg), cefotaxime (5 µg), meropenem (10 µg), tigecycline (15 µg) and trimethoprim (5 µg) were tested in this study. The antibiotic sensitivity of *Escherichia coli* and *Salmonella* spp. isolates was tested using the standard disk diffusion method according to the EUCAST guide. The direct colony suspension method was used to make a suspension of the organism in saline to the density of a 0.5 McFarland turbidity standard, which was evenly inoculated into Mueller-Hinton medium plates. Plates were inoculated by swabbing. Based on the inhibition zone size, the results were recorded as «Susceptible» or «Resistant» according to the interpretive criteria specified in EUCAST.

E. coli and *Salmonella* spp. isolates which demonstrated resistance to cefotaxime or ceftazidime or meropenem were tested using a second panel of antimicrobial substances

according to Commission Implementing Decision (EU) 2020/1729 of 17 November 2020. The method of combined discs was used for this research.

RESULTS

The results of the antibiotic resistance monitoring showed the widespread prevalence of antibiotic resistant isolates of *Salmonella* spp. and commensal *Escherichia coli* in livestock farms and in the private sector of the Kyiv, Vinnytsia, Kirovohrad, Poltava and Lviv regions.

In total, 27 resistant isolates (17.3 %) were detected from 156 investigated samples including 25 commensal *Escherichia coli* isolates (16.0 % of the total number of samples) from faeces, 1 (0.6 %) from the milk of cows with mastitis and 1 *Salmonella* spp. isolate (0.6 %) from faeces.

Six strains of commensal *Escherichia coli* (22.2 % of the total number of resistant isolates) were resistant to ceftazidime; 5 (18.5 %) to ampicillin; 4 (14.8 %) to tigecycline; 5 (18.5 %) to trimethoprim and 1 (3.7 %) to cefotaxime.

Salmonella spp. isolated from cattle faeces was resistant to ceftazidime (10 µg) and cefotaxime (5 µg) with an inhibition zone of 18 mm in both cases. AmpC production was confirmed in this isolate of *Salmonella* spp. after testing with a second panel of antibiotics since the inhibition zone to ceftazidime (30 µg) was 17 mm.

DISCUSSION

The results of our research demonstrate the prevalence of the ceftazidime-, cefotaxime- and meropenem-resistance of commensal *Escherichia coli* and *Salmonella* spp. isolates among resistant strains isolated from faeces and milk samples from cattle in Ukraine. This indicates an increase in the aggressive potential of the obtained pathogens and the rapid spread of acquired resistance.

Given the constant changes and levels of resistance of commensal *Escherichia coli* and *Salmonella* spp., the obtained data are significant where resistance to 3rd generation cephalosporins and carbapenems is a marker of the antibiotic resistance of Enterobacteria, as is claimed by other experts.

The policy for the use of antibiotics in livestock and poultry farming should be determined based on local data regarding antimicrobial resistance. Therefore, it is important to establish systems to monitor antimicrobial resistance at local, regional and national level. This is the focus of the actions of the Government of Ukraine and experts in this field.