THE EVALUATION OF THE ANTIMICROBIAL RESISTANCE OF *ESCHERICHIA COLI* AND *SALMONELLA* SPP. STRAINS ISOLATED FROM RAW MEAT

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Abstract
The antimicrobial resistance of the most probable pathogen germs isolated from raw meat has not been tested thoroughly yet in our country. The transmissibility of this resistance from food to human has been previously described in the foreign literature that is why such a complex study is mandatory given the current situation. For the isolation and antimicrobial resistance assessment, the classical method was used, the confirmation being performed through molecular methods (simplex PCR). In the antimicrobial evaluation the automatic system TREK was used. The majority of the Escherichia coli strains isolated from the meat samples were confirmed as non-pathogenic ones but revealing a high number among them as being resistant to ampicillin and enrofloxacin. The Salmonella spp. bacteria isolated was found in a lower amount and with a high prevalence of resistance to cefazolin, cefuroxime and tetracycline. Although a lot of food poisoning episodes are treated in the infectious diseases hospitals with ampicillin, enrofloxacin and tetracycline it has been shown in this study that these antibiotics might not have the wanted effect. We recommend the antibiogram in every case given the fact that these bacteria have become more and more resistant due to improper use of antibiotics in animal feed, animal illnesses and human diseases.

***Key words*:** antibiogram, antimicrobial, raw meat, resistance.

INTRODUCTION

The improvements in antimicrobial treatments along with the sanitation, nutrition and immunization have finally lead to a decrease in the number of deaths and a major gain in life expectancies (WHO, 2007). But along with the increase use of antimicrobials this phenomenon of resistance (AMR – antimicrobial resistance) has proved to be one of the most serious threats to human health (WHO, 2007) and a major concern for public health, anima health and also food safety authorities (Tenover, 2006; Talbot, 2006; Courvalin, 2005; O'Brien, 2002; Marchese, 2007). In order to minimize the risk of occurrence, a few steps must be taken into account,, like the

antibiotic prescription control and use in animal growth, in veterinary practices, improving hygiene, epidemiological studies' making and applying measures for decreasing the probability of cross-contamination among resistant strains and healthy individuals. The control of this phenomenon implies sustained efforts from the involved authorities, directed towards the identification of resistant bacteria as well as towards the transmissibility pathways to humans. The purpose of this study was to evaluate the antimicrobial resistance of *E.coli* and *Salmonella* spp. bacterial strains isolated from pork and chicken meat and to evaluate the possible effect of their spread in meat destined for public consumption.

MATERIAL AND METHODS

This study was conducted on 40 samples of pork meat collected from the carcass surface after the protocol previously described by Dragomir (2012) and 30 samples of poultry carcasses according to the methods and working procedures described in the National and Comunity legislation, respectively Regulation (CE) no.2073/2005 regarding the microbiological criteria for food products modified with Regulation. (CE) no.1441/2007.
The *in vitro* antimicrobial susceptibility was tested by agar difussion test using the Muller-Hinton method and also by Trek automatic system method. The Trek Diagnostic System is a rapid testing waqy of the bacteria strains to a large variety of antibiotics. This system use the Sensititre MIC plates, each of them being dosed with antimicrobials agents in corresponding dilutions. The results have been read automatically using the fluorescence captured by the ARIS/ AutoReader apparatus.
The inoculation procedure:
With a sterile loop, 3-5 freshly obtained colonies have been emulsified in sterile demineralized water, adjusted to 0.5 McFarland standards. After this step, 30µl from the suspension was transferred in a Muller Hinton broth of 11 ml that contains TES tampon. Afterwards, 50µl were transferred in Trek plates and then incubated for 24h at 37°C.

RESULTS AND DISCUSSIONS

The antimicrobial susceptibility of the *E.coli* strains isolated from pork and poultry meat are shown in figure1:

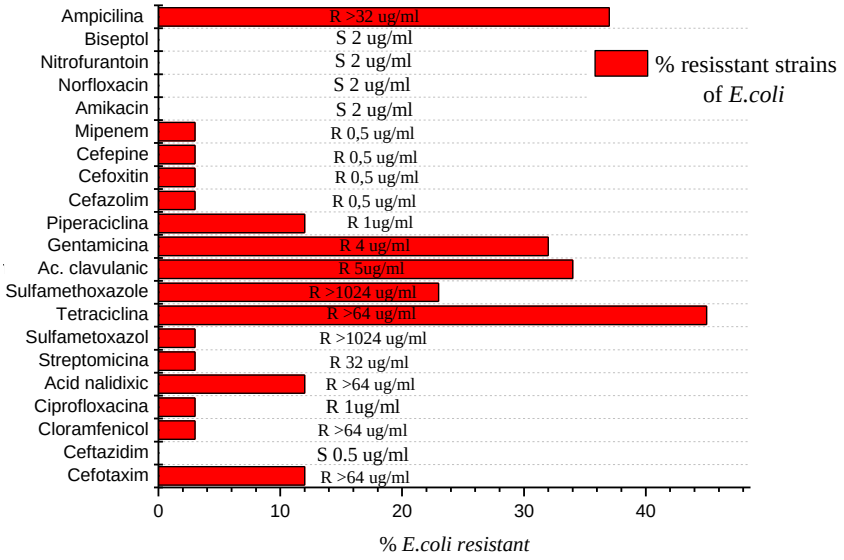

Figure 1. The percent of resistant *E.coli* isolated from pork and poultry meat

The number of *E.coli* strains resistant to some classes of antibiotics was relatively high given the fact that all the bacteria isolated came from raw meat samples destined for public consumption. The highest prevalence was revealed in the case of *E.coli* strains resistant to tetracycline and ampicillin. Thus, fig.1 shows that the *E.coli* strains were sensible to a series of antibiotics, the field literature stating the fact that there is a high prevalence of resistant among *E.coli* especially to fluoroquinolone (Andraud M. et al., 2011). The antibiotics that showed the highest percent in the case of resistant bacteria have proved to be the ones most frequently used in animal therapy at swine as well as birds.

The highest prevalence of *E.coli* resistant strains was found at poultry meat some authors claim that this is most commonly seen in pork meat (Kim et al., 2011). Our explanation for such a high prevalence in poultry meat is the fact that farmers still use in this intensive broiler growth systems antibiotics to reduce their mortality in the first days.

In case of *Salmonella* spp. identification, there was a low incidence of its occurrence in pork and poultry meat. Only one sample of pork meat was

identified with *Salmonella* spp. group C, that was sensible to all the antibiotics tested.

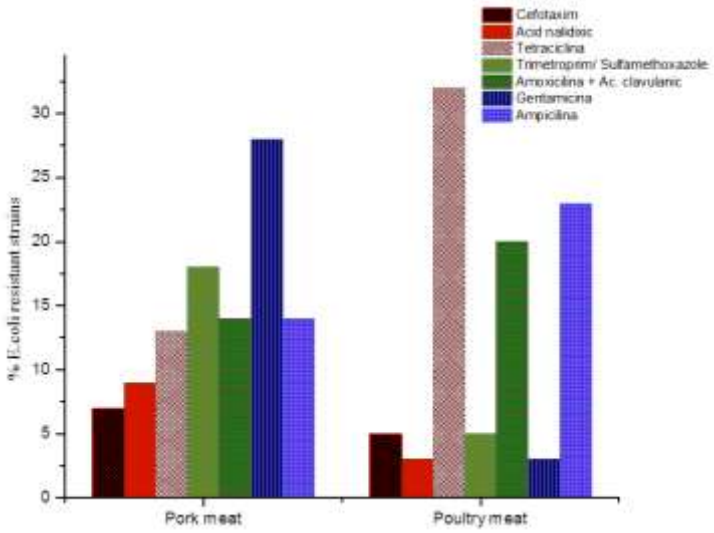

Figure 2. The frequency of *E.coli* resistant isolation among the two species studied

At the morphological exam of the selective media for the identification of *Salmonella* genus there were revealed red colored colonies with black center due to H_2S production on Rambach (Figure 3) but also on XLD media. In a study made by Marculescu et al. (2007) on *Salmonella* spp. prevalence at pig carcasses, the antimicrobial susceptibility was also evaluated. This study showed that there was a high prevalence of sensitive strains to ampicillin and amoxicillin (81.25%), with a low prevalence of resistant ones (18.75%). None of the poultry meat samples tested was found *Salmonella* positive which means that there is a low prevalence of occurrence in the studied slaughtering units.

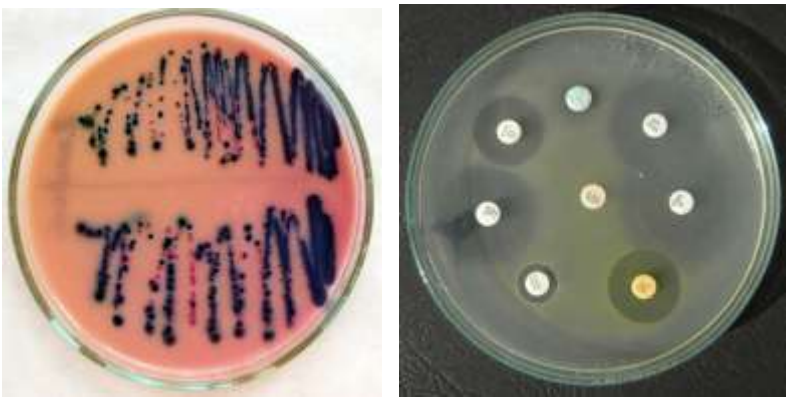

Figure 3. Identification of *Salmonella* spp. colonies on Rambach media and respectively the antimicrobial susceptibility tested by agar difussion

CONCLUSIONS

The prevalence of *E.coli* strains resistant to certain classes of antibiotics is relatively high in poultry meat. The most common resistance was observed at tetracycline (45%), over 30% of the poultry meat samples tested revealing *E.coli* resistant to tetracycline. The prevalence of *Salmonella* spp. in pork and poultry meat is low and the one investigated being sensible to every antibiotic tested. This study shows that there is an imperative need of keeping under control the transmissibility of *E.coli* resistant strains through food consumption given the great risk of contamination.

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