



# Study on Antimicrobial Resistance of Escherichia Coli o157:H7/NM isolated from Raw Bovine, Camel, Water Buffalo, Caprine and Ovine milk

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## Abstract

*Escherichia coli O157:H7* has appeared as a food-producing pathogen in public health. Previously the various patterns of *E.coli*'s transition in bovine cattle have been mentioned such as long-term migration or transient cycle in host of bovine. The recognition of animals breeding *E.coli*, is difficult because some adult cattle do not appear signs of illness and shedding of it and they scatter diseases in everywhere. This study is aimed at determining antimicrobial resistance of *Escherichia coli o157:H7/NM* isolated from raw milk. The study was conducted on bovine, camel, water buffalo, caprine and ovine in Iran. According to Clinical and Laboratory Standards Institute (CLSI), 32 isolated milk experienced for susceptibility to antimicrobial drugs by Kirby-Bauer disk diffusion test using Muller-Hinton agar. Overall 26 cases of 32 *E.coli* isolates were resistant to one or more antimicrobial agents. Resistance to ampicillin was the most common finding (46.6%), and after that resistance to erythromycin (31.1%), gentamycin (9.4%), nalidixic acid (12.5%), doxycyclin (9.4%), streptomycin (9.4%), kanamycin (9.4%), tetracycline (9.4%), chloramphenicol (60.3%), and amoxicillin (3.1%). All *E. coli* isolates were sensitive to cefuroxime.

**Keywords:** *E.coli* O157:H7, *E.coli* O157: NM, milk, water buffalo (*bubalus bubalis*), bovine, camel, ovine, caprine, antimicrobial resistance.

## Introduction

The food supply is a certificated conveyer for specific kind of antimicrobial resistance and bacteria's pathogen. Antibiotics have saved millions existences of people and using of them have helped to improve the health of animals and human<sup>1</sup>. Using antibiotics in food-producing animals affected making healthier animals, reducing diseases and mortality. Also producing more quantities and qualities food stuff and reducing prices of food for human are other advantages of antibiotics<sup>2</sup>. On the other hand, there are the main concerns utilizing antibiotics for public health and producing healthy food in food-producing animals. Over the past two decades, the development of antimicrobial resistance, applied in agriculture, affecting human's treatments has become the main concern in public health<sup>3</sup>. Although resistance of *E.coli* to antimicrobial agents reported from all over the world but it seems this situation has become worse in developing countries because there are unchecked and common usage of antibiotics<sup>4</sup>. Recent studies have been conducted to recognition of antimicrobial resistance of *E.COLI* isolated from bovine, ovine, caprine, camel, water buffalo in Iran<sup>5</sup>.

## Methodology

Some samples selected randomly from 296 bovine, 207 ovine, 275 caprine, 121 camel, 109 water buffalo in the city of Esfahan, Chahmahal and Bakhtiari, Yazd, Kerman, Tehran, Khuzestan located in Iran. These samples with cooler and ice

packs have instantly been delivered to laboratory and one hour after collections were processed<sup>6</sup>. 10 ml of each sample synthesized with 90 ml of Tryptone Soya Broth and homogenized navabiocin and then it was incubated at 30<sup>0</sup>C for about 18-24 hours. 100 ml of culture was put on the sorbet MacConkey agar plate that was included compound of tellorim-sefecsim<sup>7</sup>.

After 24 hours incubation at 42<sup>0</sup>C, the negative sorbet clones were tested for antigens by compounds of Latex. Also more than 5 positive Agglutination were picked up *e.coli* was stabilized with PCR tests using encoding the o antigens of *E.coli* genes and general primers H7 Flagon which were mentioned before<sup>8</sup>. Antimicrobial susceptibility test:

A series of each sample of positive *E.Coli* is selected for sensitivity test to bacteria. Antimicrobial sensitive tests were conducted by Kirby-Bauer disc diffusion method using the Kirby-Bauer method which is formed by removing fiber in blood of sheep according to the Clinical and Laboratory Standards Institute. Performance Standards for Antimicrobial Disk Susceptibility tests were used for this experiment: Nalidixic acid (30µg), seforexim (30µg), aritomaysin (15µg), tetrasaykelin (15µg), streptomycin (30ag), gentamycin (10µg), amoxicillin (30µg), ampicillin (10µg), chloramphenicol (30µg)<sup>10</sup>. After 48hour of incubation at 30<sup>0</sup>C, the susceptibility of *E-coli* toward each antimicrobial agent was measured and results of it were expounded according to the expositive criteria by CLSL<sup>11</sup>.

## Results and Discussion

The pattern of antimicrobial resistance of E.coli isolated to 11 antimicrobial agent was experienced in the study and has been shown in table 1. Overall 26 cases of 32 E. Coli isolates were resistant to one or more antimicrobial factor. 9 groups of them resistant to one antibiotic and 12 groups to 2 antibiotics agents. Multi-resistance bacteria are called multi-bacteria due to their resistance to 3 or more tested drugs<sup>12</sup>.

Resistance to ampicillin was the most common finding (46.6%), and after that resistance to erythromycin (31.1%), gentamycin (9.4%), nalidixic acid (12.5%), doxycycline (9.4%), streptomycin (9.4%), kanamycin (9.4%), tetracycline (9.4%), chloramphenicol (60.3%), and amoxicillin (3.1%). All E. Coli O157:H7/NM isolates were susceptible to cefuroxime<sup>13</sup>.

Table-1: shows the chart of the antimicrobial resistance of E.coli isolated from raw bovine, camel, water buffalo, caprine and ovine milk in the city of Esfahan, Chaharmahal Bakhtiari, Yazd, Kerman, Tehran, Khuzestan located in Iran<sup>14</sup>. Antibiotics have saved millions existences of people and using of them have helped to improve the health of animals and human. Using antibiotics in food-producing animals affected making healthier animals, reducing diseases and mortality<sup>15</sup>. Also producing more quantities and qualities food stuff and reducing prices of food for human are other advantages of antibiotics. On the other hand, there are the main concerns utilizing antibiotics for public health and producing healthy food in food-producing animals. Over the past two decades, the development of antimicrobial resistance applied in agriculture which can be affected in human's treatments has become the main concern in public health<sup>16</sup>.

## Conclusion

The result of antimicrobial resistance tests in recent research indicate high resistance of E.coli to ampicillin, erythromycin and gentamycin and low resistance to nalidixic acid, kanamycin, streptomycin, doxycycline, amoxicillin, tetracycline, chloramphenicol. These results can be compared with other studies reported by researchers. However, the Byrne ET<sup>18</sup>. Al distributed the resistance of E.coli's class to three groups of erythromycin, streptomycin and tetracycline. The results of Goncuoglu indicated 100% of E.COLI isolated was resistant to sefalotodin and averagely was resistant to ampicillin, streptomycin and solphamethoxazol<sup>19</sup>. The resistance of antibiotics might be caused by alternative pressure or not taking of antibiotics advantage of antibiotics by human, or over using by farmer in dairy farm. Although antimicrobial resistance are very common, the effect of antibiotics is not clear in management of threatening diseases such as diarrhea. Multi-resistance quality is not observed in any experiments and all isolated bacteria were resistant to tetracycline and chloramphenicol using in cattle. In another research, Walsh et al. separated the E.COLI which was resistant to Streptomycin and

kanamycin from pieces of bovine meat. Differences between antimicrobial resistances of E.coli isolated from food sample reported in another research. This sample may be result of other sample, food storage, used technique, effect of season and/or used laboratory method and other studies. The result of finding of antimicrobial resistance in this study related to antibiotics used for cure of infection in animals' food<sup>20</sup>. Also high percentage of E.coli isolated was resistant to ampicillin which is an antibiotic used in treatment of human's infections.

Consequently, we find that raw milk is extensively embarrassed with antimicrobial resistance of E.coli. Therefore the supply of milk is more important but recognition of vector of pathogens diffusions and resistant agents is more important. Recognize the importance of health and basic mechanism of results are bound to accepting alternative purchases and controlling effort and proper mediation<sup>21</sup>.

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