

Antibiotic Pattern of Some Pathogenic Bacteria Isolated from Animal and It's Relation to Human Infections

¹A. Alhababi, ²M. Elgezery, ³G. Alnaimi and ³M. Soliman

¹King Fahad Military Medical Complex, Dhahran, Saudi Arabia

²Community Medicine, Alexandria University, Egypt

³King Fahad Military Medical Complex, Dhahran, Saudi Arabia

Abstract: The present study was done on 186 isolates obtained from both animals (107 isolates) and patients (79 isolates). The study aimed at comparing the antimicrobial patterns of pathogens isolated from animal and human cases (nosocomial infections). The isolates were identified and the antibiograms were reported. The results pointed out that *E. coli* was the most common isolate followed by *Pseudomonas aeruginosa* in human samples, while in animal samples the most common Gram negative isolates recovered were *Pseudomonas aeruginosa* followed by *Proteus* spp. On the other hand, *Staphylococcus aureus* was the most common Gram positive isolate in animal and human samples. Conclude that, both animal and human isolates were markedly resistant to different types of antibacterial agents. However, animal isolates compared with human isolates showed more resistance to most of the used antibacterial agents. This can be attributed to the fact that in our country, animals are generally exposed to several antibacterial agents either as therapeutic, prophylactic or as growth promoter agents.

Key words: Antibiotic, isolates, resistance, sensitivity

INTRODUCTION

Antibiotic resistance is a global public health problem. Although all countries are affected, the extent of the problem in the developing nations is certainly much more widespread and most of the data about the resistant bacterial strains are almost unknown (Richet *et al.*, 2001). Increasing travel and patient movement throughout the world have increased the transmission of drug-resistant organisms from one country to another leading eventually to serious worldwide problem.

Drug resistance is more frequently encountered in hospital-acquired pathogens (nosocomial infections). However, the incidence of antibiotic-resistant pathogens in community-acquired infections has been also on the rise in the recent years (Hooton and Levy, 2001).

Although antibiotic resistance is a natural expression of evolution and bacterial genetics, certain factors are thought to contribute immensely to enhance the expression and spreading of this bacterial inherent potentiality.

Antimicrobial resistance is a zoonotic health resulted threat. As in humans, the use of antimicrobial agents in animals resulted in the emergence and spread of resistant bacteria. Resistant bacteria of animals may be passed to humans via the food chain or direct animal contact, and may result in resistant infections. Increasing prevalence of

resistance to antimicrobial agents such as fluoroquinolones and third-generation cephalosporins, which are important for the treatment of infections caused by enteric pathogens, has significant public health implications. Controlling the spread of resistance requires the collaboration of several participants such as Veterinary, Medical, and Public Health Communities (Angulo *et al.*, 2004).

Antibiotic use whether for treatment or prophylaxis, or as performance enhancers will result in antibiotic resistant micro-organisms, not only among pathogens, but also among bacteria of the endogenous microflora of animals (Van den Bogaard and Stobberingh, 1999).

The long-term consequences of emergence of nosocomial strains for the outside community, however, still need to be assessed. The possibility that nosocomial pathogens may tend to be not only more resistant to antibiotics, but also more inherently virulent lends some urgency to this need.

Hospitals are places where antimicrobial with high selective pressure by antimicrobial agents. For this reason, bacteria producing nosocomial infections are not only virulent, but also resistant to antimicrobial agents (Rodriguez-Martinez, 2005).

The aim of the present study is to study the Antibacterial sensitivity testing of the animal and human isolates and to compare between bacterial resistance to

antibacterials in animal and human isolates in order to investigate the possible relationship between the drug-resistant veterinary bacterial pathogens and the corresponding human disease- etiologic agents.

MATERIALS AND METHODS

Cases: The samples were collected from two main hospitals in the Eastern Province - Kingdom of Saudi Arabia. The first hospital is the Veterinary Hospital of King Faisal University at Al Hasa area (for animal cases) and the second one is King Fahd Military Medical Complex at Dhahran Area (for human cases). All nosocomial infections reported in both hospitals during January and February 2007 were studied. A total of 186 isolates were studied. Animal isolates (107 isolates) constituted 57.53% of the sample while human isolates (79 isolates) constituted 42.47% of the sample.

Isolation and Identification of bacterial isolates:

Direct staining: Animals and human samples were prepared and stained with Grams stain. The type of organisms and density were reported.

Cultivation of samples: Both animal and human samples were cultured onto media according to the type of specimens by means of streaking loop technique using disposable sterile plastic loop. The media were incubated in the incubator to 18-24 h at 37°C and observed for cultural characteristics.

Solid media plates were examined macroscopically for growth and colonial morphology. Any changes of the medium such as haemolysis, change in colour of the media and colonial colour were recorded, then pure culture of bacteria were obtained by sub-culturing onto solid media till pure isolates were obtained.

Biochemical Identification of the isolates: Identification was done using Api system including 20 parameters in each individual strip, included and interpreted according to Api index guide using the numerical profile.

Antibiotic susceptibility patterns: Antibiotic susceptibility patterns were performed by agar disc diffusion technique (ADDT) Koneman *et al.* (1992) and Quinn *et al.* (2002).

Subculture from each isolate was tested to various antibiotics according to NCCLS guideline.

MIC assessment was done using E.test technique (AB biodisk):

Statistical analysis: Reporting and statistical analysis were done using SPSS program.

Table 1: Distribution of human and animal isolates according to gram stain and susceptibility to ampicillin

		Human isolate		Animal isolates	
		No.	%	No.	%
Gram -ve	Sensitive	12	20.34	17	34.7
	Resistant	47	79.66	32	65.3
	Total	59	100.00	49	100.00
$\chi^2 = 2.81$ (NS)					
		Human isolate		Animal isolates	
		No.	%	No.	%
Gram +ve	Sensitive	4	20.00	24	41.38
	Resistant	16	80.00	34	58.62
	Total	20	100.00	58	100.00
$\chi^2 = 2.95$ (NS)					

Table 2: Distribution of Human and animal isolates according to gram stain and susceptibility to Erythromycin

		Human isolate		Animal isolates	
		No.	%	No.	%
Gram -ve	Sensitive	39	66.10	11	22.45
	Resistant	20	33.90	38	77.55
	Total	59	100.00	49	100.00
$\chi^2 = 20.52$ (S)					
		Human isolate		Animal isolates	
		No.	%	No.	%
Gram +ve	Sensitive	10	50.00	17	29.31
	Resistant	10	50.00	41	70.69
	Total	20	100.00	58	100.00
$\chi^2 = 2.81$ (NS)					

Table 3: Distribution of human and animal isolates according to gram stain and susceptibility to gentamicin

		Human isolate		Animal isolates	
		No.	%	No.	%
Gram -ve	Sensitive	28	47.46	21	42.86
	Resistant	31	52.54	28	57.14
	Total	59	100.00	49	100.00
$\chi^2 = 0.28$ (NS)					
		Human isolate		Animal isolates	
		No.	%	No.	%
Gram +ve	Sensitive	9	45.00	36	62.07
	Resistant	11	55.00	22	37.93
	Total	20	100.00	58	100.00
$\chi^2 = 1.78$ (NS)					

RESULTS

Comparison of susceptibility patterns of human and animal isolates to various antibiotics were illustrated in Table 1-8.

High resistance pattern was observed among gram negative and gram positive human and animal isolates to Ampicillin, Penicillin and tetracycline (Table 1, 5 and 6, respectively). Half of the human isolates were resistant to Gentamycin (Table 3) and more than two thirds of gram negative human and animal isolates were resistant to

Table 4: Distribution of Human and animal isolates according to gram stain and susceptibility to Norfloxacin

		Human isolate		Animal isolates	
		No.	%	No.	%
Gram -ve	Sensitive	23	38.98	15	30.61
	Resistant	36	61.02	34	69.39
	Total	59	100.00	49	100.00
$\chi^2 = 9.7$ (S)					
		Human isolate		Animal isolates	
		No.	%	No.	%
Gram +ve	Sensitive	13	65.00	54	93.10
	Resistant	7	35.00	4	6.90
	Total	20	100.00	58	100.00
$\chi^2 = 9.7$ (S)					

Table 5: Distribution of Human and animal isolates according to gram stain and susceptibility to Penicillin

		Human isolate		Animal isolates	
		No.	%	No.	%
Gram -ve	Sensitive	15	25.42	10	20.40
	Resistant	44	74.58	39	79.60
	Total	59	100.00	49	100.00
$\chi^2 = 0.38$ (NS)					
		Human isolate		Animal isolates	
		No.	%	No.	%
Gram +ve	Sensitive	3	15.00	9	15.52
	Resistant	17	85.00	49	84.48
	Total	20	100.00	58	100.00
$\chi^2 = 0.6$ (NS)					

Table 6: Distribution of human and animal isolates according to gram stain and susceptibility to tetracyclin

		Human isolate		Animal isolates	
		No.	%	No.	%
Gram -ve	Sensitive	14	23.73	9	18.37
	Resistant	45	76.27	40	81.63
	Total	59	100.00	49	100.00
$\chi^2 = 0.46$ (NS)					
		Human isolate		Animal isolates	
		No.	%	No.	%
Gram +ve	Sensitive	6	30	20	34.48
	Resistant	14	70	38	65.52
	Total	20	100.00	58	100.00
$\chi^2 = 0.13$ (NS)					

Norfloxacin and Cotrimoxazole.(Table 4 and 8, respectively). Almost three quarters of the animal isolates were resistant to Erythromycin (Table 2) while these isolates were highly sensitive to cephalosporines (Table 7).

DISCUSSION

Antibiotic resistance is a global problem both in human and animals. The current study aimed at comparing the antimicrobial effect between veterinary and human isolates by using both disc diffusion and MIC determination. In this study almost two thirds (63%) of

Table 7: Distribution of human and animal isolates according to gram stain and susceptibility to cephalosporines

		Human isolate		Animal isolates	
		No.	%	No.	%
Gram -ve	Sensitive	11	18.64	40	81.63
	Resistant	48	81.36	9	18.37
	Total	59	100.00	49	100.00
$\chi^2 = 42.6$ (S)					
		Human isolate		Animal isolates	
		No.	%	No.	%
Gram +ve	Sensitive	11	55.00	51	87.93
	Resistant	9	45.00	7	12.07
	Total	20	100.00	58	100.00
$\chi^2 =$ fisher exact test 0.003 (S)					

Table 8: Distribution of human and animal isolates according to gram stain and susceptibility to cotrimoxazole

		Human isolate		Animal isolates	
		No.	%	No.	%
Gram -ve	Sensitive	17	28.81	19	38.78
	Resistant	42	71.19	30	61.22
	Total	59	100.00	49	100.00
$\chi^2 = 1.2$ (NS)					
		Human isolate		Animal isolates	
		No.	%	No.	%
Gram +ve	Sensitive	17	85.00	32	55.17
	Resistant	3	15.00	26	44.83
	Total	20	100.00	58	100.00
$\chi^2 = 5.66$ (S)					

nosocomial infections in human were caused by gram negative bacilli mainly E. coli while other studies showed predominance of other isolates as Pseudomonas (Krause *et al.*, 1999), Acinetobacter (Mendes *et al.*, 2005) or Staph aureus (Sader *et al.*, 1998).

Resistance was noticed in 50% of G +ve organism to Penicillin, Clindamycin, Erythromycin, Tetracycline and Ciprofloxacin as were as resistant to ampicillin nitrofurantoin, levofloxacin piperacillin and cefuroxime was found in G +ve (Ahmed and Bahlas, 2009).

Respiratory pathogens acquire resistance as prevotella, H Influenza, M Catarlli but not to levafloxacin. In another study it was observed up to 34% resistance in prevotella in Jeddah and up to 56% in Kuwait (Memish *et al.*, 2007)

Regarding antibiotic Susceptibility testing of Penicillin, 1st generation cephalosporin and Tetracycline, we noticed significant relation between Gram Positive isolates compared to Gram negatives in human while insignificant relation in animals. On the other hand 2nd and 3rd generation cephalosporin showed insignificant relation in both. Quinolone and Aminoglycosides showed insignificant relation. The relation between Gram positive human and animal isolated to macrolides, penicillin and cephalosporin was significant.

Prevalence of resistance was more common among calves compared to adult cows (Sato *et al.*, 2005).

Antibiotics used as growth promoters in animals resulted in increased strain resistance as noticed from susceptibility pattern particularly to vancomycin and emergence of VRE. Accordingly it should be replaced with other agent or Probiotics.

The current study showed the importance of periodic antibiogram to monitor the pattern of resistance in the human and animals. Also the study indicated the importance of implementing hospital antibiotic policy which controls markedly its occurrence; the latter should be also controlled by avoiding misuses of antibiotic and appropriate combination of antibiotics.

In other study done in Makkah by Asghar and Faidih (2010). They reported higher rate of resistance among G -ve pathogens in comparison with other countries in the world which necessitates implementing monitoring program.

CONCLUSION

The results pointed out that *E. coli* was the most common isolate followed by *Pseudomonas aeruginosa*, while in animal samples the most common Gram negative isolates recovered were *Pseudomonas aeruginosa* followed by *Proteus* sp. and *Staphylococcus aureus* was the most common Gram positive isolate in animal and human samples. Generally, both animal and human isolates were markedly resistant to different types of antibacterial drugs. However, animal isolates compared with human isolates showed more resistance to most of the used antibacterial agents. This can be attributed to the fact that in our country, animals are generally exposed to several antibacterials either as therapeutic, prophylactic or as growth promoter agents.

It was observed also that the antibacterial agents of veterinary use only i.e., is not applied in human medicine showed lower resistance ratios when tested against Gram negative bacteria: florophenicol (6.3%), enrofloxacin (17.9%) and flumquine (42.4%). While in the resistance rate was Gram positive bacteria: florophenicol, enrofloxacin (10% for each) and flumquine (14.3%).

It was also observed that the antibacterial agents used only for treatment of human infections gave lower resistance ratios when used against Gram negative bacteria which affect human as: imipenem (8.8%), meropenem (9.6%), tazocin (46.4%) and cefpem (39.7%).

ACKNOWLEDGMENT

We would like to thanks all Microbiology staff of both King Fahd Military Medical Complex and College of Veterinary Medicine and animal Resources, King Faisal University for technical support.

REFERENCES

- Ahmed, M.M. and S. Bahlas, 2009. Bacteriological profile and antimicrobial resistance patterns of clinical bacterial isolates in a University Hospital. *Travel Med. Infect. Dis.*, 7(4): 235-238.
- Angulo, F.J., J.A. Nunnery and H.D. Bair, 2004. Antimicrobial resistance in zoonotic enteric pathogens. *Rev. Sci. Technol.*, 2: 485-96.
- Asghar, A.H. and H.S. Faidah, 2010. Frequency and antimicrobial susceptibility of gram-negative bacteria isolated from 2 hospitals in Makkah, Saudi Arabia., *Saudi Med. J.*, 31: 338.
- Hooton, T.M. and S.B. Levy, 2001. Antimicrobial resistance: A plan of action. *Am. Fam. Phys.*, 63: 1087-96.
- Koneman, E.W., S.D. Allen, W.M. Janda, P.C. Schreckenberger and W.C. Winn, 1992. Packaged in Kit Identification System. Color Atlas and Textbook of Diagnostic Microbiology. Koneman, E.W. (Eds.), 4th Edn., B. Lippincott Co., Philadelphia, PA., pp: 163-170.
- Krause, R., H. Mittermayer, G. Feierl, F. Allerberger, I. Wendelin, A. Hirschl and E.C. Reisinger, 1999. *In vitro* activity of newer broad spectrum beta-lactam antibiotics against enterobacteriaceae and non-fermenters: A report from Austrian intensive care units. Austrian carbapenem susceptibility surveillance group. *Wien Klin Wocheschr.*, 111: 549-554.
- Mendes, C., C. Oplustil, E. Sakagami, P. Turner, C. Kiffer and MYSTIC Brazil Group, 2005. Antimicrobial susceptibility in intensive care units: MYSTIC program Brazil 2002. *Braz. J. Infect. Dis.*, 9: 44-51.
- Memish, Z.A., A.O. Osoba, A.M. Shibl, E. Mokaddas, S. Venkatesh and V.O. Rotimi, 2007. Emergence and trends of penicillin non-susceptible *Streptococcus pneumoniae* in Saudi Arabia and Kuwait-perspective and outstanding issues, *J. Chemother.*, 19(5): 471-481.
- Quinn, P.J., B.K. Markey, M.E. Carter, W.J.C. Donnelly, F.C. Leonard and D. Maguire, 2002. Antimicrobial agents *Veterinary Microbiology and Microbial Disease*. Blackwell Science Ltd., UK, pp: 28-35.
- Richet, H.M., J. Mohammed, L.C. McDonald and W.R. Jarvis, 2001. Building communication networks: International network for the study and prevention of emerging antimicrobial resistance. *Emerg. Infect. Dis.*, 7(2): 319-322.
- Rodriguez-Martinez, J.M., 2005. Mechanisms of plasmid-mediated resistance to quinolones. *Enferm. Infec. Microbiol. Clin.*, 1: 25-31.

- Sader, H.S., R.N. Jones, A.C. Gales, P. Winokur, K.C. Kugler, M.A. Pfaller and G.V. Doern, 1998. Antimicrobial susceptibility patterns for pathogens isolated from patients in Latin American medical centers with a diagnosis of pneumonia: Analysis of results from the SENTRY Antimicrobial surveillance Program (1997). SENTRY latin america study group. *Diagn. Microbiol. Infect. Dis.*, 32: 289-301.
- Sato, K., P.C. Bartlett and M.A. Saeed, 2005. Antimicrobial susceptibility of *Escherichia coli* isolates from dairy farms using organic versus conventional production methods. *J. Am. Vet. Assoc.*, 226(4): 589-594.
- Van den Bogaard, A.E. and E.E. Stobberingh, 1999. Antibiotic usage in animals: Impact on bacterial resistance and public health. *Drugs*, 4: 589-607.