Antimicrobial drug susceptibility of clinical isolates of *Pseudomonas aeruginosa*

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Unlike other gram-negative bacterial pathogens, *Pseudomonas aeruginosa* is intrinsically insensitive to many antimicrobial drugs due to the low rate of passage of antibiotics across its outer membrane (1). Though antipseudomonal drugs, such as amikacin, gentamicin, and carbenicillin, are used against infections caused by this organism, comparatively less expensive drugs, such as tetracycline and neomycin, do have application in veterinary medicine (2,3). Monitoring of drug susceptibility trends may help to determine the possible use of a drug or a group of drugs in a specific animal species. Predominance of serotypes has been found to be different in one animal species compared to another, and an association between antimicrobial susceptibility and serotypes has been shown (5,6). Though individual antibiograms are necessary for successful therapy, a knowledge of general susceptibility patterns may be helpful in certain situations.

Two hundred and sixteen clinical isolates of *P. aeruginosa* were tested for in vitro antimicrobial drug susceptibility. One hundred and thirteen of these were from dogs, of which 60% were isolates from cases of otitis. Bovine isolates were the second most numerous, and the majority of these were of enteric origin. Most of the equine isolates were of respiratory origin, whereas isolates from pigs were mainly from internal organs. The majority of isolates from cats were from skin and wound infections. The susceptibilities of all isolates were determined by a standard disk diffusion procedure (7), using 16 antibiotics. The disks obtained from Unipath Inc., Nepean, Ontario, contained amikacin sulfate (Sefton Division of Glaxochem Ltd, Uxbridge, Middlesex, UK) 10 µg; amoxicillin/clavulanic acid (Beecham Pharmaceuticals Ltd, Worthing, Sussex, UK) 30 µg; carbenicillin (Beecham Pharmaceuticals Ltd) 100 µg; cephalothin (Sigma Chemical Company Ltd, Poole, Dorset, UK) 30 µg; cephalexin (Sefton Division of Glaxochem Ltd) 30 µg; chloramphenicol (Gee Lawson Chemicals Ltd., London, UK) 30 µg; gentamicin (Roussel Laboratories Ltd, Denham, Uxbridge, UK) 10 µg; kanamycin (Gee Lawson Chemicals Ltd) 30 µg; neomycin (Sigma Chemical Company Ltd) 30 µg; netilmicin (Essex Chemie AG, Luzern, Switzerland) 10 µg; oxytetracycline (Pfizer Chemical Division, Sandwich, Kent, UK) 30 µg; polymyxin B (Gee Lawson Chemicals Ltd) 300 units; streptomycin (Eli Lilly, Basingstoke, Hampshire, UK) 10 µg; or tobramycin (Eli Lilly) 10 µg. Ceftiofur sodium (Excenel, Upjohn Company-Animal Health, Orangeville, Ontario) 10 µg disks and enrofloxacin (Baytril, Bayvet Division, Chemagro Ltd, Etobicoke, Ontario) 5 µg disks were obtained from the manufacturers. Inhibition zones of ceftiofur and enrofloxacin were interpreted according to the technical information supplied by the manufacturers. For the ceftiofur disk, a zone diameter less than 19 mm indicated resistance; more than 24 mm, susceptibility; and a zone size in between, intermediate. For enrofloxacin, a zone size less than 17 mm meant resistance, and more than 22 mm indicated susceptibility. In addition, 58 isolates from dogs, cats, and horses originating from mucosal or internal infections were tested against 4 aminoglycoside drugs (gentamicin, amikacin, tobramycin, and netilmicin) by the E-test, which consists of an impervious strip impregnated with a continuous antimicrobial gradient, suitable for determining minimal inhibitory concentrations (MICs). The E-test strips (AB Biodisk, Solna, Sweden) were purchased from Unipath Inc. The test was conducted and interpreted according to the manufacturer's instructions. *Pseudomonas aeruginosa* strain 27853 (American Type Culture Collection) was used as a control in all of the assays.

Results of susceptibility to all 16 antibiotics are given in Table 1. Tobramycin, netilmicin, and polymyxin B were the only drugs to which 100% susceptibility was noted. All isolates were resistant to amoxicillin-clavulanic acid, cephalothin, cephalexin, and ceftiofur. In a disease problem with drug resistant *P. aeruginosa*, newer or third generation cephalosporins, such as cefoperozone and ceftazidime, should be considered, since the older cephalosporins are inactive against this bacterium. These drugs, however, are extremely expensive for veterinary use (3). Resistance to aminoglycosides was negligible, with the exception of streptomycin and kanamycin. The appearance of strains resistant to gentamicin and amikacin is of some concern. Fifty-eight isolates recovered from mucosal and internal sites of 37 dogs, 14 horses, and 7 cats were found to be susceptible to gentamicin, amikacin, tobramycin, and netilmicin in the E-test. Minimal inhibitory concentrations against 90% of the isolates (MIC_{90}) for gentamicin, amikacin, tobramycin, and netilmicin were equal to or below 4, 6, 1 and 6 µg/mL, respectively. Minimal inhibitory

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Table 1. Antimicrobial drug susceptibility of 216 isolates of *Pseudomonas aeruginosa*

Drug	Percent susceptible (intermediate)				
	Canine N = 113	Bovine N = 69	Equine N = 14	Porcine N = 12	Feline N = 8
(a) Aminoglycosides					
Streptomycin	5(44)	5(50)	7(50)	0(58)	0(62)
Neomycin	61(35)	76(23)	93(7)	67(33)	100
Kanamycin	0(35)	7(28)	7(14)	0(8)	0(50)
Gentamicin	95(3)	100	100	100	100
Amikacin	99(0)	100	100	100	100
Tobramycin	100	100	100	100	100
Netilmicin	100	100	100	100	100
(b) Other drugs					
Polymyxin B	100	100	100	100	100
Amoxicillin/ clavulanic acid	0	0	0	0	0
Cephalothin	0	0	0	0	0
Cephalexin	0	0	0	0	0
Ceftiofur	0	0	0	0	0
Enrofloxacin	36(54)	37(56)	29(64)	8(92)	62(25)
Carbenicillin	69(19)	88(7)	79(21)	100	62(25)
Tetracycline	4(59)	7(61)	14(86)	0(58)	12(63)
Chloramphenicol	4(30)	2(46)	0(29)	0(33)	0(50)

concentrations below 8 μg/mL indicate susceptibility to these drugs (3). While gentamicin is available for topical and systemic use in various animal species, amikacin is primarily available for treatment of mucosal and internal infections (3). Tobramycin, a newer aminoglycoside antibiotic, is useful against gentamicin-resistant strains (3). Only 5% of isolates from dogs were susceptible to streptomycin, unlike the results of another study (8) in which the percentage of susceptible *P. aeruginosa* strains from dogs, particularly those from the ear canal, was several times higher. Sixty-seven of 113 canine isolates in our study were from otitis cases, mostly otitis externa, and 55% of these were susceptible to neomycin. All 8 feline isolates were susceptible to neomycin.

Strains resistant to enrofloxacin were found in all species, except those from pigs. Nine percent of isolates from canine otitis cases were resistant to this drug. The majority of isolates from all species, except those from cats, showed only intermediate susceptibility. According to Miller (9), enrofloxacin can be used in the form of systemic therapy concurrently with topical treatment in canine otitis with a *P. aeruginosa* component, since this bacterium often invades deep tissues rapidly.

Overall susceptibility to carbenicillin was good, and only 8% of 216 isolates showed resistance to this drug. No resistant strains were found among equine and porcine isolates. Zschoek and Schliesser (10), however, found a high proportion of strains from various animal species resistant to this drug. Carbenicillin has application in horses, and dose recommendations for other species are also available (11).

The majority of isolates from all species showed intermediate susceptibility to tetracycline. In a recent study (12), the in vitro efficacy of oxytetracycline against *P. aeruginosa* isolates from dogs and horses was found to be greater than that reported in previous years. Of 6 urinary isolates from dogs in this study, 5 showed intermediate susceptibility to tetracycline. Since it has been shown that systemic administration of tetracycline results in very high concentrations in the urine (2), this drug may be efficacious in treating pseudomonal urinary infections.

A majority of the isolates were resistant to chloramphenicol, a drug with little use for infections caused by *P. aeruginosa*. Polymyxin B, to which all our isolates were susceptible, has application in the treatment of otitis externa, as well as in infections of skin and mucous membranes (3). Most of our canine and feline isolates belonged to these categories. Strains with simultaneous resistance to antipseudomonal drugs, such as gentamicin, amikacin and carbenicillin, were not found among our isolates.

In summary, our results show that gentamicin is still very effective against *P. aeruginosa* strains of veterinary origin, irrespective of species and site of infection. Polymyxin B, to which all the isolates were susceptible, is a very useful topical drug against *P. aeruginosa*. Susceptibility to neomycin is unpredictable, and susceptibility testing should be performed. Enrofloxacin and tetracycline have application in some cases of otitis and urinary infections in dogs involving susceptible strains of *P. aeruginosa*. Only a minority of isolates of canine origin, including those from otitis cases, were susceptible to streptomycin and chloramphenicol, two drugs found in some commercial otic preparations. CVJ

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BOOK REVIEWS

REVUES DE LIVRE

Evans HE, ***Miller's Anatomy of the Dog, 3rd ed.*** WB Saunders, Toronto, 1993. 1113 pp. ISBN 0-7216-32009. $113.00.

Miller's Anatomy of the Dog established itself as the preeminent English language reference in canine anatomy almost immediately upon the publication of its first edition in 1964. In the third edition, the strengths of the original are maintained and augmented.

Miller's Anatomy uses a systemic approach to present the anatomy of the dog. There are one or more chapters for each anatomical system, including chapters on skin; bone; joints; muscle; digestive system; respiratory system; urogenital system; endocrine system; heart and arteries, veins, and lymphatics; several chapters on neuroanatomy; and chapters on the eye and ear. A strong point is the many anatomical drawings. Because the drawings depict regions of the body, many drawings of the vessels and nerves also contain useful information on the muscular system.

The third edition contains some additional and updated material; for example, sections by Robert K. Wayne, Juliet Clutton-Brock, and Peter Jewell. The section on muscle has a valuable addition on histochemical, fiber type classification. The section on the nervous system is rearranged and has new contributors, in the persons of R. Kitchell and A. Beitz. Dr. Kitchell's influence is seen in the addition of depictions of autonomous zones of cutaneous innervation. The sections on the nervous system also contain much updated material, reflecting our increased knowledge of neuroanatomy.

The section on the ear benefits from several new drawings and added detail on the external ear. However, it is questionable whether the duplication of the drawings of the auricular muscles with different names provided in the section on muscle and the section on the ear aids our understanding of that complex topic. The section on the eye benefits from the contribution of C.R. Murphy.

For a textbook of such great scope and size (1113 pages) to be perfect would be unlikely, even on statistical grounds. Fortunately, errors have been kept to a minimum and are restricted to less significant anatomical detail. Claiming that the median artery terminates as the palmar metacarpal arteries (page 643) is confusing, in the light of Figure 11-47. Another example is the retention of the term accessory interosseus for ulnar artery (page 640), an error that may have escaped proofreading.

As a reference, this book has great utility. A complete and detailed index makes searching out topics time-efficient. The many detailed drawings will aid veterinary surgeons and other specialists equally.

In the reviewer's opinion, it is also of value to consider the usefulness of this book within the context of current-day veterinary education and practice. It has almost become a cliché to speak of the information explosion; however, it exists and its consequences are being increasingly felt in education and practice. In education, anatomy is under pressure to give up time to allow teaching of further detail in other subjects. In practice, there is a trend toward more specialization, especially in the field of companion animal medicine. Under these circumstances, the value of the third edition of *Miller's Anatomy of the Dog* can be appreciated. It is a tribute to its several authors; its editor and author, Dr. Evans; and the memory of its originator, Dr. M.E. Miller.

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