

## IDENTIFICATION AND *IN VITRO* DRUG SENSITIVITY OF MICROORGANISMS ISOLATED FROM DOGS WITH OTITIS EXTERNA IN PUNJAB

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Otitis externa is a commonly occurring disease in dogs. Acute and chronic forms of the disease have been observed in many dogs. Various bacteria and fungi have been isolated and identified by many workers from the ear discharge (Lund, 1979; Nicklas and Mumme, 1979; Sala *et al.* 1983). Various antibiotics have been found to be ineffective to check the ear canal infection. Keeping these facts in view, the present experiment was designed to isolate and identify the causative bacteria of otitis externa and *in vitro* susceptibility to various chemotherapeutic agents.

### Materials and Methods

#### *Collection of samples*

Ear swabs from affected dogs were collected with sterile cotton swabs in sterilized test tubes under completely aseptic conditions in the specialized unit of Veterinary Clinics, College of Veterinary Science, Punjab Agricultural University, Ludhiana (India).

#### *Processing of samples*

After collection, the samples were processed on the same day. They were streaked on to 10 per cent sheep blood agar plates and these plates were incubated at 37°C for 24 hr. Thereafter morphology of colonies was observed and colonies were stained with Gram's stain. Further identification of the bacteria was done as advocated by Carter (1984).

#### *In vitro drug sensitivity*

One colony of the causative organism was inoculated in nutrient broth (5 ml) tubes and incubated for 16–18 h. Then the broth was plated on Mullar hinton agar plates with sterile cotton swabs. After drying the plate the antibiotic discs (gentamycin (G), 10 mcg; neomycin (N), 30 mcg; chloramphenicol (C), 30 mcg; streptomycin (S), 10 mcg;

erythromycin (E), 15 mcg; Penicillin (P), 10 units, Tetracycline (T), 30 mcg; Kanamycin, (K), 30 mcg; and septran (Q), 25 mcg) were placed and incubated at 37°C for 24 h. After that zone of inhibition was measured in millimeters.

### Results and Discussion

Out of 37 ear swabs, one type of bacteria was isolated from 11 swabs, two types of bacteria from 17 swabs and three types of bacteria could be isolated from 9 ear swabs. Thus a total of 72 different bacteria could be isolated. The ratio of Gram positive and Gram negative bacteria was almost equal (Table I). The *Staphylococcus aureus* was the predominating (25.%) bacteria isolated. Many other workers have reported more or less similar results (Moreno *et al.* 1975; Blue and Wooley, 1977; Lund, 1979; Ducha Sardana *et al.* 1981; Chomel *et al.* 1982). However, Fachini (1981) and Okay *et al.* (1984) encountered *Pseudomonas aeruginosa* as the chief isolate from canine otitis. This difference in the isolation of chief causative agent of canine otitis may be due to the prevalence of a particular organism in an area of study and degree of otitis i. e., acute, subacute or chronic. Other isolates from the canine otitis were  $\beta$ . haemolytic streptococci, *Proteus mirabilis*, *Escherichia coli*, *Klebsiella* spp., *Enterobacter* spp. and *Corynebacterium* spp. (Table I). Similar isolation pattern with slight or no variations has been observed by many other workers (Lund, 1979; Nicklas and Mumme, 1979; Ducha Sardana *et al.*, 1981; Chengappa *et al.* (1983).

Table I  
Isolation of Micro-organisms from canine otitis externa

Organisms isolated	Total no. of organisms	Percentage
<i>Staphylococcus aureus</i>	80	25.00
<i>Streptococcus</i> $\beta$ haemolytic	13	18.05
<i>Proteus mirabilis</i>	10	13.88
<i>Pseudomonas aeruginosa</i>	10	13.88
<i>Escherichia coli</i>	8	11.11
<i>Klebsiella</i> spp.	4	5.55
<i>Enterobacter</i> spp.	4	5.55
<i>Corynebacterium pyogenes</i>	3	4.16
Coagulase negative Staphylococci	2	2.83
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Table II  
Sensitivity patterns of micro-organisms to various antibiotics

Name of the organisms	Total No.	Percentage of the sensitive organisms to										
		G	N	C	S	E	P	T	K	Q		
<i>Staphylococcus aureus</i>	18	100.00	83.33	77.77	66.66	83.33	11.11	55.55	11.11	88.88		
<i>Streptococcus</i> $\beta$ haemolytic	13	100.00	36.92	76.92	61.53	76.92	15.38	61.53	7.69	84.61		
<i>Proteus mirabilis</i>	10	90.00	20.00	60.00	10.00	10.00	0.00	30.00	50.00	70.00		
<i>Pseudomonas aeruginosa</i>	10	40.00	0.00	10.00	0.00	0.00	0.00	0.00	0.00	20.00		
<i>Escherichia coli</i>	8	87.50	50.00	12.50	25.00	50.00	0.00	0.00	20.00	0.00		
<i>Klebsiella</i> spp.	4	100.00	25.00	50.00	25.00	50.00	0.00	50.00	50.00	75.00		
<i>Enterobacter</i> spp.	4	100.00	50.00	50.00	25.00	75.00	0.00	0.00	50.00	75.00		
<i>Corynebacterium pyogenes</i>	3	100.00	33.33	66.66	33.33	66.66	0.00	33.33	0.00	66.66		
Coagulase negative staphylococci	2	100.00	100.00	50.00	50.00	50.00	0.00	100.00	0.00	50.00		

The *in vitro* drug sensitivity test carried out against nine antibiotics showed that gentamycin, septran, neomycin, erythromycin and chloramphenicol were very effective (7.75%) against *Staphylococcus aureus* and  $\beta$ . haemolytic streptococci. Nicklas and Mumme (1979) also reported the similar findings. Increased resistance of these organisms to penicillin, and kanamycin was also similar with the observations of Ducha Sardana *et al.* (1981) and Mc Carthy and Kelly (1982). However, Blue and Wooley (1977) reported the resistance of these organisms to erythromycin and neomycin. These variations may be due to the prior application of these antibiotics in canines or some drug resistant variants might have developed. Table II shows the detailed sensitivity pattern of various organisms to different antibiotics. *Pseudomonas aeruginosa* was found to be the most resistant organism isolated. It was hundred per cent resistant to 6 of the 9 antibiotics tested. Blue and Wooley (1977) have also reported their increased resistance to neomycin. However, Mc Carthy and Kelly (1982) observed that *Ps. aeruginosa* was quite sensitive to neomycin and gentamycin. In the present investigation 40% of these organisms were sensitive to gentamycin. 20% to septran and only 10% were sensitive to chloramphenicol.

Penicillin was found to be the least effective antibiotic in the present study against the causative agent of canine otitis. Almost all the organisms isolated were hundred per cent resistant to penicillin except *S. aureus* (11.11% sensitive) and  $\beta$  haemolytic streptococci (15.38% sensitive). This may be due to the fact that the penicillin has been so much used since its discovery against various infections in dogs, that most of the organisms have developed resistance to it.

Thus it can be evidently concluded from the present study that canine otitis externa can be effectively checked by any of the antibiotics showed positive response to the causative agents.

#### Summary

Culture of ear swabs from 37 dogs with otitis externa resulted in the isolation of 72 bacteria, of which *Staphylococcus aureus* (25.00%),  $\beta$  haemolytic streptococci (18.05%), *Proteus mirabilis* and *Pseudomonas aeruginosa* (13.88% each) and *Escherichia coli* (11.11%) were the chief isolates. *Klebsiella* spp. and *Enterobacter* spp (5.55% each), *Corynebacterium pyogenese* (4.16%) and coagulase negative staphylococci (2.83%) were the other isolates. The *in-vitro* drug sensitivity of all the isolates was carried out against 9 antibiotics and gentamycin, septran, erythromycin and chloramphenicol were found to be very effective. *Pseudomonas aeruginosa* was found to be the most resistant (resistant to six antibiotics tested) bacteria. Penicillin (10 units) was the least effective antibiotic.

## സംഗ്രഹം

നായ്ക്കളിൽ ചെങ്കിടു പഴുപ്പ് ഉണ്ടാക്കുന്ന രോഗാണുക്കൾ എന്തൊക്കെയാണെന്ന് കണ്ടുപിടിക്കുകയുണ്ടായി. ജൻടാമൈസിൻ, സെപ്ട്രാൻ, എറിത്രോമൈസിൻ, ക്ലോറാം ഫിനിക്കോൾ ഇവ ചികിത്സക്ക് ഫലപ്രദമാകുമെന്ന് കണ്ടു.

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