

PREVALENCE OF MYCOPLASMA INFECTION IN SLAUGHTERED DOGS IN JOS METROPOLIS**¹*Ogbu K. I., ²Ochai S. O., ¹Maimadu A. A., ³Olabode M. P., ⁴Olaolu O. S., ³Waziri I. A. and ³Oguche M. O.**¹Department of Animal Health, Federal College of Animal Health and Production Technology.²Faculty of Veterinary Medicine, University of Maiduguri, Maiduguri, Borno State Nigeria.³National Veterinary Research Institute, Vom, Plateau State, Nigeria.⁴Ahmadu Bello University Zaria, Kaduna State, Nigeria.⁵University of Agriculture, Makurdi, Nigeria.***Corresponding Author: Ogbu K. I.**

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ABSTRACT

The study was designed to determine the prevalence of Mycoplasma infection in slaughtered dogs in Jos Metropolis. Nasal swab (100) and trachea sample (50) were collected randomly at Jos dog market. The samples were cultured on Pleuro-pneumonia like organism agar and incubated anaerobically at 37°C for 72 hours using culture method by Morton and Lecce, (1953). The result were analyze using Chi Square method and presented in tables. The result of the study revealed that out of the 150 samples tested, 23 (15.3%) were positive. In relation to age, sex, location and sample type. There was a higher prevalence in puppies (16%), than in adult (15%), though not statistically significant $p > 0.05$. The prevalence was higher in females (16.2%) than males (14.5%) though not significant $p > 0.05$. The prevalence was higher in Jos North (20%) than Jos South (11.3%) though there was no significant difference $p > 0.05$. There was higher prevalence in nasal swab (16%) than trachea samples (14%) though not statistically significant $p > 0.05$. The study shows that there was no significant difference.

KEYWORD: Prevalence, Mycoplasma Infection, Dogs, Pleuro-pneumonia, Jos.**I. INTRODUCTION**

Mycoplasmas are bacteria that lack cell wall, but are enclosed by a lipid bilayer membrane. They colonize the mucus membranes of the respiratory and genital tracts as well as red blood cells and are found in many animals and humans.^[1] (Chalker and Brownlie, 2004). In dogs, *Mycoplasmas* are thought to be part of the normal bacterial flora in the upper respiratory tracts, but there are conflicting reports about the presence of *Mycoplasma* in the lower respiratory tracts of healthy dogs were colonized whereas other authors have failed to detect *Mycoplasmas* in the lower respiratory tract of healthy dogs. The role of individual *Mycoplasmas* species in the respiratory infections of dogs is not well understood, but they are thought to colonize the lung during pneumonia.^[2] (Rosendal, 1982). Because of the difficulty in identifying canine *Mycoplasma* species the majority of studies have investigated the presence or absence of *Mycoplasmas* in variety of clinical samples and little is known species about specific infection caused by *Mycoplasma* species. However, some studies have identified the present and a few have focused on individual species of *Mycoplasma* and natural or experimental infection.^[3,4,5,6,7,1] (Bowe *et al.*, 1982, Rosendal, 1974; Rosendal and Laber, 1973; Jang *et al.*, 1982; Barton *et al.*, 1985; Chalker and Brownlie, 2004).

Mycoplasma in dogs are fastidious in nature and some required cholesterol, sterol or urea for growth.^[8] (Razin and Tully, 1995). *Mycoplasma* approximately passes through 300 – 400nm membranes. The term '*Mycoplasmosis*' designates infectious diseases due to *Mycoplasmas*, which are known to be the smallest prokaryotes with autonomous replication, in fact, the general term '*Mycoplasma*' is a trivial name, referring to a group of microorganisms which differ from other bacteria by, among other things, lacking a cell wall. This was the reason for creating a new class, named Mollicutes (from the Latin *mollis* (soft) and *cutis* (skin)). This class comprises four orders, with different families and genera totaling approximately 160 species, which are parasitological to a broad range of hosts, from humans to animals, insects and plants. The list is far from exhaustive, as new species are currently being described and uncultured *Mycoplasma* like plant organisms, the phytoplasmas, are considered as Mollicutes too.^[9] (Whitford *et al.*, 1994). The decline in productivity and economic wastage cannot be unconnected to many problems of most diseases which are bacteria in nature. The disease cause respiratory failure, sterility and infertility. The prevention of such diseases as *mycoplasmosis* is necessary in dogs. It has been reported that dogs were infected with the diseases despite all the

precautionary measures taken; this could be attributed to inability to effectively diagnose and control the infection. *Mycoplasma* in dogs is thought to be part of the normal bacterial flora in the upper respiratory tracts, but there are conflicting reports about the presence of *Mycoplasmas* in the upper respiratory tracts of healthy dogs. The role of individual *Mycoplasmas* species in respiratory infections of dogs is not well understood. Therefore, this research will provide current status of *Mycoplasma* infection in dogs in the study area. This work is aimed at determining the prevalence of *Mycoplasma* species in dogs, isolation of *Mycoplasma* in dogs and to determine the prevalence of *Mycoplasma* in relation to age, sex, type of sample and location.

II. METHODOLOGY

This research work is limited to the detection of *Mycoplasma* in dogs. It covered Jos Metropolis and was restricted to nasal swab and tracheal tissues from 150 dogs from the LGAs.

Sample Collection: Nasal swabs were aseptically collected from the dogs in Jos metropolis. Dog markets where dogs are slaughtered were visited in the area. Immediately after the dogs were slaughtered and dropped into 2ml of pleuro-pneumonia like Organism broth, placed on an ice pack and transported to the laboratory for examination. The samples were stored at -20°C prior to examination (trachea samples), while the nasal swab were incubated immediately at 37°C for 72hours.

Tracheal Isolation of Mycoplasma: All of isolates were tracheal sample in pleuro-pneumonia like organism (PPLO) broth culture. After culture tube filtration, this media were incubated in pleuro-pneumonia like organism broth at 37°C for color change because of contamination. These isolates were inoculated in PPLO media agar plate at 37°C for 72 hours examining the presence of *Mycoplasma* colonies.^[10] (Evans *et al.*, 2009).

Preparation of the Transport Media: The media used was the pleuro pneumonia like organism agar and broth based from Difco laboratories[®]. The media were prepared based on manufacturer's instructions where 4.2g of the pleuro pneumonia like organism (PPLO) was measured and 140ml of distilled water was used to dissolve the PPLO powder. The solution was then poured in a clean container and then sterilized using the autoclave for 15minutes at a temperature of 121°C . The solution was then removed and allowed to cool to 50°C after sterilization. 40ml of Horse serum was added and 20ml of supplement (yeast, extracts, antibiotics, horse serum) was also added.^[11] (Morton and Lecce, 1953).

Preparation of the Agar Broth: The media used was the pleuro-pneumonia like organism and broth based from Difco laboratories. The media were prepared based on manufacturer's instructions where 3.5g of the pleuro-pneumonia like organism (PPLO) was measured and

65ml of distilled water was used to dissolve the PPLO powder. The solution was placed in an autoclave for sterilization at 121°C for 15minutes. The solution was removed and allowed to cool and 25ml of horse serum was added to the solution. 10ml of supplement was also added^[11] (Morton and Lecce, 1953).

Sample Processing: A small piece of each trachea samples was especially picked after the surface was starred with a red hot blade and dropped in 2ml pleuro-pneumonia like organism broth and then Incubated at 37°C for 72hours.

Culture and Isolation: 20 μl of the incubated broth samples was picked using a micro pipette and in 10 fold serial dilution was carried out in 180 μl broth in a micro titer plate to obtain a dilution of 10^{-6} after which a loop from the sixth dilution (10^{-6}) was taken and sub-cultured into the surface of the freshly prepared Pleuro-Pneumonia-Like Organism (PPLO) agar by spread plate method and allowed to absorb into the medium then Incubated an anaerobically in a Memmert[®] CO_2 Incubator at 37°C for 72hrs in 5% CO_2 .

Microscopic Examination: After 72hrs of incubation, the plates were examined using a stereo (inverted microscope) where the microscope was well focused to reveal any emerging colonies on the plates. The presumptive canine *Mycoplasma* colonies were identified according to standard morphological characteristic features of *Mycoplasma* showing a fried-egg appearance with a central nucleus protruding like a nipple.

Statistical Analysis

The results obtained were presented in tables and also analyzed using chi square test. Significant difference at $P < 0.05$ was determined.

III. RESULTS

The tables bellow shows the relationship and distribution of *Mycoplasma canis* in relation to sex, age, location and sample type collected.

Table no 1: Age.

Age	Positive	Negative	Total
Puppies	8(16%)	42(84%)	50(33.3%)
Adult	15(15%)	85(85%)	100(66.7%)
Total	23(15.3%)	127(84.7%)	150(100%)

$$(\chi^2)=0.0271 \quad \chi^2/=3.84 \quad P>0.05 \quad df=1$$

Table no 2: Sex.

Sex	Positive	Negative	Total
Male	11(14.5%)	65(85.5%)	76(50.7%)
Female	12(16.2%)	62(83.8%)	74(49.3%)
Total	23(15.3%)	127(84.7%)	150(100%)

$$(\chi^2)=0.0862 \quad /\chi^2/=3.84 \quad P<0.05 \quad df=1$$

Table no 3: Location.

Location	Positive	Negative	Total
Jos North	14(20%)	56(80%)	70(46.7%)
Jos South	9(11.3%)	71(88.8%)	80(53.3%)
Total	23(15.3%)	127(84.7%)	150(100%)

$(X^2)=2.21$ $/X^2/=3.84$ $P>0.05$ $df=1$

Table no 4: Sample type.

Sample	Positive	Negative	Total
Nasal swab	16(16%)	84(84%)	100(66.7%)
Tracheal	7(14%)	43(80%)	50(33.3%)
Total	23(15.3%)	127(84.7%)	150(100%)

$(X^2)=0.1053$ $/X^2/=3.84$ $P>0.05$ $df=1$

IV. DISCUSSION

The prevalence of *Mycoplasma canis* in dogs did not differ significantly among the general prevalence of *Mycoplasma* ($P<0.05$). This is not in accordance with Chalker *et al.*, (2004)^[1] where they found out that worldwide predominantly. This may be as a result of mycoplasma species present in both healthy and chronic infectious respiratory disease (CIRD). Therefore depending on the prevalence of the organism colonizing the healthy dogs or those with chronic infectious respiratory disease may determine the overall prevalence.

The prevalence of *Mycoplasma canis* in dogs did not show any significant difference when compared to age ($P<0.05$), but younger dogs may be more susceptible to the mycoplasma infection than in older dogs. This is in accordance with Randolph *et al.*, (1993)^[12] whose later investigation noted a greater susceptibility to *Mycoplasma* infection in younger dogs. This may be due to low immunity in the systems of the young and also may be as a result of the absence of vaccine for the mycoplasma. Young dogs with concurrent infection with bordetella or streptococci in the lower airway were more likely to be infected with *Mycoplasma* and that the presence of *Mycoplasma* was associated with septic inflammation.^[12] (Randolph *et al.*, 1993). Again dogs are first exposed to and colonized by *Mycoplasmas* when passing through the birth canal.^[13] (Eberle and Kirchhoff, 1976).

The prevalence of *Mycoplasma canis* in dogs do not differ significantly when compared to sex ($P<0.05$). This is in accordance with L'Abée- *et al.*, (2003)^[14] whose study did not show whether sex is a factor to *Mycoplasma* infection but they stated that *Mycoplasma canis* has been isolated from dogs with urogenital disease and infertility, despite prolonged antibiotic therapy and has been cultured from the prostate, epididymis and the chronically inflamed bladder wall. Rosendal, (1982)^[2] also stated that experimental infection with *Mycoplasma canis* produced chronic urethritis, and epididymis in males tested and females enlarged uterus and endometritis was seen. This may be because *Mycoplasma* infection is not gender sensitive, it can affect any sex as far it come across a host.

The prevalence of *Mycoplasma canis* in dog did not differ significantly when compare between location ($P<0.05$). This is in disagreement to Nagatomo *et al.*, (2001)^[15] whose study stated that environment may be a factor or source of infection but considering the study location which is a cold and windy, *mycoplasma canis* may be easily transmitted from one location to another.

The prevalence of *Mycoplasma canis* in dogs did not differ significantly among sample type ($P<0.05$). This is in accordance with Barile *et al.*, (1970)^[16], Kirchner *et al.*, (1990)^[17], Chalker and Brownlie, (2003)^[18] where they found out that *Mycoplasmas* colonized the mucous membrane of the respiratory and genital tracts as well as red blood cells. This may be as a result of the predilection site since various *Mycoplasma* species can be isolated from dogs on different sites.

V. CONCLUSION

The study has isolated *Mycoplasma canis* but it was not prevalent according to age, sex, type of sample and location and so there was no significance. Our work is in disagreed with many work which shows no prevalence of the organism in the study areas.

VI. RECOMENDATION

Awareness creation on the presence and dangers of *Mycoplasma* infection in dogs which is one of the main causes of respiratory disease. Adequate use of antibiotics to prevent / treat this infection should be advocated. Proper biosecurity should also be adopted by the dog breeders to prevent the outbreak of this infection.

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