

***In vitro* sensitivity of various strains of *Mycoplasma synoviae* against five antibiotics.**

Based on a study by:

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Introduction

Mycoplasma synoviae (Ms) is one of the pathogens involved in Chronic Respiratory Disease (CRD). It usually causes a mild respiratory disease that can be exacerbated by other secondary pathogens. Ms also causes infectious synovitis (IS).

CRD is present wherever poultry are kept, worldwide (1). Recently, Ms was first isolated from Argentina (2).

The objective of the study was to evaluate, *in vitro*, the minimum inhibitory concentration (MIC) and the minimum mycoplasmacidal concentration (MMC) of 5 antibiotics to 8 isolated Ms strains. Two reference Ms strains were also included.

Materials and methods

The strains investigated were obtained from poultry flocks showing signs of either IS or CRD in the Buenos Aires and Entre Rios areas of Argentina. Various types of poultry were involved (see Table 1).

Table 1. Origins of the Ms strains

Strain	Category of Chicken	Farm	Location
100	Heavy breeders	1	Buenos Aires
117	Broilers	1	Buenos Aires
128	Heavy breeders	1	Buenos Aires
173	Heavy breeders	2	Buenos Aires
178	Heavy breeders	3	Buenos Aires
288	Heavy breeders	4	Buenos Aires
311	Heavy breeders	5	Entre Rios
312	Laying hens	6	Buenos Aires
WVU 1853	Reference strain		
2AS	Reference strain		

Antibiotics

The antibiotics used were:

- 3-acetyl-4-isovaleryltirosin (Aivlosin[®])
- tylosin tartrate (Tylan)
- tiamulin fumarate ((Tiamutin)
- oxytetracycline (OTC)
- chlortetracycline (CTC)

Incubation medium

MIC study: FM-4 sterilised by filtration

MMC study: FM-4 sterilised by filtration +1% agar solution

(see reference 3 for details)

Procedure:

A volume of 1.6ml of incubation medium was filled into sterilised haemolysis tubes. A further 0.2ml volume of the antibiotic dose and 0.2ml of a 24hrs-old culture medium for each of the strains was added. A control without antibiotic and a control without inoculum were used. Each tube was incubated at 37°C.

The concentrations of antibiotic used were 12.5, 6.25, 3.12, 1.56, 0.78, 0.39, 0.2, 0.1, 0.05 and 0.025 ug/ml. The MIC readings were done at the moment when growth of the organism was detected, indicated when the colour of the phenol red indicator turned yellow (about 24 hrs). Once the MIC had been determined, 0.01ml of each tube without growth was plated out onto a solid medium and incubated at 37°C in a humid environment. After 5 days of incubation, the plates were examined microscopically for the detection of Ms colonies.

Results

MIC, MIC₅₀, MIC₉₀ and MMC values are listed in Tables 2, 3 and 4.

TABLE 2. MIC values

ANTIBIOTICS					
STRAINS	AIVLOSIN[®]	TYLOSIN	TIAMULIN	OTC	CTC
100	0.025	0.05 ^a	0.39	1.56	1.56
117	0.05	0.05	0.39	1.56	1.56
128	0.05	0.1	0.78	0.78	0.78
173	0.05	0.05	0.78	1.56	1.56
178	0.05	0.1	0.2	0.1	0.39
211	0.05	0.1	0.78	1.56	3.12
311	0.025	0.1	0.78	0.78	12.5
312	0.025	0.1	3.12	6.25	6.25
2AS^b	0.05	0.05	0.2	0.2	0.78
WVU1853^b	0.05	1.56	0.78	1.56	1.56

a) ug/ml of antibiotics

b) reference strains

TABLE 3. MMC values

ANTIBIOTICS					
STRAINS	AIVLOSIN[®]	TYLOSIN	TIAMULIN	OTC	CTC
100	0.05	0.2	6.25	3.12	12.5
117	0.05	0.2	3.12	3.12	50
128	0.1	0.39	6.25	3.12	50
173	0.05	0.2	6.25	3.12	50
178	0.1	1.56	12.5	6.25	50
211	0.1	0.1	12.5	50	50
311	0.02	0.39	12.5	50	50
312	0.02	6.25	12.5	50	50
2AS^b	0.1	0.2	12.5	50	50
WVU1853^b	0.05	1.56	12.5	12.5	50

TABLE 4. values for MIC₅₀ and MIC₉₀

ANTIBIOTICS	RANGE	MIC₅₀	MIC₉₀
AIVLOSIN[®]	0.025 – 0.05	0.05	0.05
TYLOSIN	0.05 – 1.56	0.1	0.1
TIAMULIN	0.2 - 3.12	0.78	0.78
OTC	0.1 - 6.25	1.56	1.56
CTC	0.1 – 12.5	1.56	6.25

Discussion and conclusions

One strain, 311, was shown to be resistant to chlortetracycline (MIC value of 12.5ug/ml). The MIC values for oxtetracycline obtained in this study were higher than those reported previously (4). An explanation for this observation could be the massive and little-controlled use of antibiotics in the country, which may have increased the resistance of these microorganisms.

The results obtained from this study agree with previous publications showing that macrolide antibiotics continue to be the “first choice” for prevention and treatment of CRD and IS.

Aivlosin[®] was shown to have the best *in vitro* potency with MIC₉₀ values of 0.05ug/ml, with a range from 0.025 to 0.05ug/ml.

In addition Aivlosin[®] showed mycoplasmacidal action at similar values to the MIC whereas the other drugs tested required several times the MIC values to exert this effect e.g. Tiamulin MMC values were up to 62.5 x MIC values.

References:

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