# Effect Of Kitasamycin On Mycoplasma Gallisepticum Infection And The Immune Response In Broiler Chickens

#### Seham Malhat, Azza G.M. Ayoub and Elen Mankarios

Zagazig Provincial Lab. Dept. of Biochemistry, Nutrition and Toxicology, Animal Health Research Institute, Zagazig, Egypt

### ABSTRACT

The activity of kitasamycin on the humoral and cellular immune response of chickens in presence and absence of mycoplasma gallisepticum infection was studied. For this purpose, one hundred (one – day old) chicks, mycoplasma free were raised. On the third week, they are divided into four equal groups as the following. The first one was non infected and non treated. The second group was non infected and treated with kitasamycin (1gm/kg.b.w.t.) in drinking water daily for 5-successive days. The third group was infected with Mycoplasma gallisepticum (0.2ml,  $2x10^8$  CFU/ml), in each bird inoculated in the air sac, this group remain non treated. The fourth group of chickens was infected as the third one and treated with kitasamycin as the second group.

Effectiveness of kitasamycin is assessed on different criteria (humoral and cellular immune response, clinical symptoms, weight gain and gross lesions in chickens either non-infected or mycoplasma gallisepticum infected ones.

Results revealed that, total serum proteins, and  $\delta$ -globulins, antibody titre, serum IgG, IgM, IgA and lymphocyte stimulation index, chemotactic index and the killing % of polymorphnuclear cells were significantly increased in all treated chickens with kitasamycin. It was also proved that kitasamycin was very active against Mycoplasma gallisepticum infection, reducing the clinical symptoms, gross lesions and increasing the weight gain of chickens.

#### INTRODUCTION .

Poultry industry is always under threat of major losses by poultry diseases. The cause of such problems are either bacteria, virus, fungi, parasites or nutritional disorders. The most serious problems facing broiler industry in Egypt is mycoplasmosis, which inducing CRD in broilers caused by mycoplasma (1). It causes high losses among affected broilers and it causes reduction in body weight and increases therapy costs (2). The diseases caused by mycoplasma with E.coli infection was more extensive than that caused by a single infection (3). Depending on their activity on mycoplasma and different microorganisms, macrolide antibiotics plays an important role in treatment and preventing respiratory diseases particularly mycoplasmosis in poultry (4).

Mycoplasma remain to be a major etiological factor in pathogenesis of respiratory diseases of chickens. Economic losses from mycoplasma gallisepticum infection in broilers arises from mortality, reduced weight gain and feed conversion and poor carcass quality which often lead to condemnation (5). Mycoplasma doesn't have a cell wall which renders them non susceptible to the action of penicillins (6).

Kitasamycin, a macrolide antibiotic particularly suited for use in mycoplasmosis, because it is concentrated intracellularly in phagocytic cells to reach a concentration which is higher than those in intracellular fluids (7).

The present study was designed to study the effect of kitasamycin in chickens, either mycoplasma infected or non infected ones. A special emphasis was directed to study its effect on cell mediated and humoral immune responses as well as its effects on the clinical symptoms, lesion scores, mortality rate and body weight of chickens.

#### MATERIAL AND METHODS

**Drug:** kitasamycin tartarate, water soluble powder (100gm/package) obtained from the Egyptian Co. for Chemicals and Pharmaceuticals (ADWIA). S.A.E.

**Dose:** 1gm/kg.b.w. administered in drinking water daily for 5 successive days (ADWIA).

Test organism: Mycoplasma gallisesticum (strain S6) with 2x10<sup>8</sup> C.F.U.,

0.2 ml inoculated via air sacs. It was obtained from Animal Health Research Institute, El-Dokki, Giza.

**Chickens:** a total one hundred (one day old chicks) were purchased from Sharkia Poultry Co. proved to be healthy and free from drugs. They were kept under hygienic conditions and fed on balanced ration free from antibiotics on 7<sup>th</sup> day of age chicks were vaccinated by Hitchiner B1 to protect chicks from Newcastle disease and on 14<sup>th</sup> day of age, they were vaccinated by Gumboro vaccine. To establish their freedom from mycoplasma, twenty chicks were taken randomly and examined bacteriologically for isolation of mycoplasma and serologically using slide agglutination test. On the third week of age, they were divided into four equal groups each one contain 20 birds as follows:

Group I: Non infected and no treated.

- Group II: Non infected and treated on the 28<sup>th</sup> day of age with kitasamycin (1gm/kg.b.w) in drinking water daily for 5-successive days.
- Group III: infected on the twenty first day of age with Mycoplasma gallisepticum  $(0.2\text{ml of } 2x10^8 \text{ CFU/ml})$  inoculated in the air sac of each bird and this group was non-treated.
- Group IV: Infected with Mycoplasma gallisepticum on  $21^{st}$  day of age inoculated in the air sacs with a dose of 0.2ml of  $2x10^8$  CFU/ml and treated with kitasamycin (1gm/kg.b.w) in drinking water daily for 5 successive days.

At 1<sup>st</sup> day, 1<sup>st</sup> week, 2<sup>nd</sup> week and 3<sup>rd</sup> week post treatment, five birds were randomly taken from each group, weighed and slaughtered and blood samples were taken and used for haematological and serological examination. Post-mortem examination was performed and the severity of air sacculitis lesions were assessed.

#### **Blood sampling:**

Two blood samples were collected from each bird after slaughtering at 1<sup>st</sup> day, 1<sup>st</sup> week, 2<sup>nd</sup> week and 3<sup>ru</sup> week post-treatment

Sample No.1: five ml of blood was collected in sterile plastic tube containing heparin (50 i.u/ml) to be used for total and differential leucocytic counts (8).

Sample No. 2: five ml of blood was collected from each bird without anticoagulant in a sterile plain test tubes for separation of serum and used for serological tests.

#### Indirect haematological test:

Slide haemagglutination and HIT for evaluating antibodies were performed by (9).

#### Cellular immune response:

Lymphocyte transformation test:

- Separation of lymphocytes was carried out (10).
- Viability of lymphocytes: was carried out by (11).
- -Standardization of the cell concentration was carried out by (12)
- Evaluation of lymphocyte transformation test was measured (13).
- -Phagocytic activity of monocytes was carried out by (14).

#### Humoral immune response:

- Determination of total serum proteins (15).

Electrophoretic analysis was carried out by (16) for determination of serum albumin, α, β and δ globulins.

#### Statistical analysis:

Student "t" test was employed according to, (17).

#### **RESULTS AND DISCUSSION**

All chickens of (non infected, non treated and non infected, treated with kitasamycin) showed no clinical symptoms throughout the experimental period. Seven days post infection all infected, non treated chickens displayed some clinical symptoms as loss of appetite, depression, respiratory symptoms including sneezing, gasping, mild conjunctivitis with frothy exudation in the eyes.

Infected chickens with mycoplasma and treated with kitasamycin showed milder degree of clinical symptoms than that of infected and non treated group. Mortality rate in each group was recorded throughout the experimental period and calculated as percent (Table 1). It was zero % in non infected, non treated chickens and non infected, treated group. Meanwhile, the mortality rate was 12% in the infected non treated group. The mortality rate was reduced in infected chickens and treated, and became 2%.

Post mortem examination of both dead or sacrificed chickens of all groups was carried out (Table 1). All chickens of both non infected, non treated and non infected treated were normal and revealed no lesions in different organs.

Infection with mycoplasma induced air sacculitis (80%), pericarditis (60%) and perihepatitis (50%). Meanwhile, infected and treated chickens with kitasamycin showed a less degree of pathological lesions.

The incidence of isolation of mycoplasma gallisepticum from trachea, air sacs and lungs in birds inoculated with Mycoplasma gallisepticum and kitasamycin are shown in (Table 2). The least rate of reisolation occurred in treated birds with kitasamycin.

Non infected and treated chickens with kitasamycin showed a significant increase in the body weight gain all over the experimental period compared with the control group (non infected and non treated) Infection with Mycoplasma gallisepticum in group III showed a significant reduction in the body weight gain all over the experimental period compared with the non infected, non treated chickens. Meanwhile, chickens which are infected with mycoplasma and treated with kitasamycin showed a significant increase in body weight gain compared with the infected non treated chickens (Table 3).

It had been shown that birds given kitasamycin showed greatest antibody titres on the 2<sup>nd</sup> week post-treatment (infected, treated group). Administration of the antibiotic auguments the magnitude of humoral immune response to Mycoplasma gallisepticum infection and make it faster. The peak of the titres was observed two weeks post treatment in infected chickens whose magnitude was greater than the infected ones (Table 4).

Blood of chickens infected with Mycoplasma galliserticum showed a significant increase of total leucocytic counts and heterophils and monocytes, while absolute number of lymphocytes was decreased. Treatment of infected birds with kitasamycin showed a significant increase of total leukocytic count and heterophils on the 2<sup>nd</sup> week post treatment in comparison to the infected, non treated chickens (Table 5, 6).

On the  $2^{nd}$  week post treatments in infected chickens a significant increase of the total protein, serum  $\delta$ -globulins, IgG, IgM and IgA was recorded in comparison with the infected non treated group (Table 7,8).

Administration of kitasamycin revealed a significant increase in the lymphocyte transformation index of polymorph-nuclear monocytes, phagocytesis %, chemotactic index and bacterial killing % on the 2<sup>nd</sup> week post treatment in infected chicken (Table 9).

Avian mycoplasmosis are economically important, egg transmitted, hatchery dessiminated disease. Therefore, therapeutic and prophylactic treatment are essential as well as other control measures (18). Kitasamycin has been shown to be effective in the treatment of mycoplasmosis in chickens (19).

The present results showed that treatment of infected birds with kitasamycin great improvement from mycoplasmosis following its oral medication based on the reduction of lesion scores, high titre of antibodies, and increase of body weight gain these results are in agreement with those previously reported (20).

A variety of methods have been used to combat avian diseases in the commercial setting, including improved farm management practices, the use of antibiotics, the selection of disease resistant types of chickens and the manipulation of chickens immune system. In the latter category, the development of antibiotics which are immunostimulatns combat the major avian diseases has become a priority in the poultry industry (21).

The increased cell mediated and humoral immune responses observed could be attributed to activation of lymphoid organs in chickens. (22) studied the immunological role

#### Seham Malhat et al.

of different lymphoid organs in chickens. They claimed that the bursa of fabricia controlled the antibody-mediated immunity, including the level of circulating immunoglobulins and presence of plasma cells.

After oral administration of kitasamycin, it is completely absorbed and widely distributed in the body and became selectively concentrated in the lung tissues which favour its use for treatment of pulmonary infection (23,24). Kitasamycin, actively taken by polymorphnuclear leucocytes and macrophages to reach high concentration, which augument the bactericidal activity of phagocytic cells against intracellular mycoplasmosis (25).

It had been concluded that the immunostimulant effect of kitasamycin was most effective in reducing the severity of air saculitis and improving the immunological response to Mycoplasma gallisepticum.

Table (1): Effect of kitasamycin (1gm/kg b.w) administered in drinking water daily for 5	<b>)</b>
successive days, on the incidence of pathological lesions and mortality rate of	f
healthy and mycoplasma infected chickens.	

Crowns	Mortality	Lesion scores %					
Groups	Mortality	Air sacculitis	Pericarditis	Perihepatitis			
Non-infected, non treated	0,	0	0	0			
Non infected treated	0	0	0	0			
Infected, non treated	12	80	60	50			
Infected, treated	2	15	5	5			

*Table (2):* Number of chickens yielding Mycoplasma gallisepticum positive culture from respiratory organs post treatment.

Cround	Post treatment								
Groups	1D	1W	2W.	3W.					
Non infected, non treated	0	0	0	0					
Non infected, treated	0	0	0	0					
Infected, non treated	4	5	5	5					
Infected, treated	4	3	1	1					

**Table (3):** Effect of kitasamycin (1gm/kg b.w.) daily administered in drinking water for 5 successive days, on body weight and weight gain of healthy and mycoplasma infected chickens (n=5).

	DW hefere				Pos	t treatmen	t		
Groups	B.W. before experiment	I <sup>#</sup> Day		I <sup>st</sup> Week		2 <sup>nd</sup> Week		3 <sup>rd</sup> Week	
	experiment	<i>B.W.</i>	Gain	<i>B.W.</i>	Gain	<i>B.W.</i>	Gain	<i>B.W</i> .	Gain
Non infected,	325.6±	474.0±	148.3±	592.9±	267.2±	1221.1±	,895.4±	1697.4±	1371.7±
non treated	1.32	24.98	3.66	39.6	18.3	14.29	` 7.03	159.5	138.2
Non infected,	358.2±	521.4±	163.1±	652.2±	294.0±	1343.2±	984.9±	1863.5±	1508.8±
treated	23.42	27.4	4.03 <sup>•</sup>	43.6	20.1*	15.7	7.7	175.4	152.05
Infected, non	293.1±	426.6±	133.5±	533.6±	240.5±	1099±	805.8±	1527.6±	1234.5±
treated	19.1	22.4	3.29	35.7	16.5	12.0	6.3	143.5	124.4
Infected treated	309.4±	450.3±	140.9±	563.3±	253.9±	1160.1±	8506±	1612±	1303.1±
Infected freated	20.02	23.7	3.4*	37.6	$17.4^{+}$	13.57	6.6+	151	131.3 <sup>+</sup>

\* P < 0.05, compared with non infected, non treated.

+ P < 0.05, compared with infected, non treated.

# Zag. Vet. J.

Ground		Post treatment								
Groups	1 <sup>st</sup> Day	1 <sup>st</sup> Week	2 <sup>nd</sup> Week	3 <sup>rd</sup> Week						
Non infected, non treated	0	0	0	0						
Non infected, treated	0	0	0	0						
Infected, non treated	6±0.05	7±0.01	7.50±0.10	6.7±0.02						
Infected, treated	6.90±0.15	7.80±0.13	9.90±0.12 <sup>+</sup>	$7.90 \pm 0.60^+$						

# *Table (4):* Haemagglutination inhibition titres (titre counts) in chicken given kitasamycin after inoculation with Mycoplasma gallisepticum.

+ P < 0.05, compared with infected, non treated.

Table (5):	Totoal	leucocytic	count (	in the	ousands)	in	chickens	treated	with	kitasamycin	after
.,	inocu	ilation with	Mycopl	asam	gallisept	icu	m (mean =	⊦ SE, n =	= 5).	·	

	Post treatment									
Groups	1 <sup>st</sup> Day	1 <sup>st</sup> Week	2 <sup>nd</sup> Week	3 <sup>rd</sup> Week						
Non infected, non treated	25.2±1.3	26.90±2.5	28.0±2.9	28.60±4.5						
Non infected, treated	26.3±2.8	28±3.3	<u>31.2±3.9</u>	31.90±1.5						
Infected, non treated	29.8±4.1	39.9±1.8*	40.5±2.4*	40.6±4.2						
Infected, treated	27.9±1.1	35.9±3.1	$43.8 \pm 2.5^+$	45.9±3.9 <sup>+</sup>						

\* P < 0.05 compared with non infected, not treated.

+ P < 0.05 compared with infected, non treated.

Table (6): Differential	leucocytic counts	s in chickens	given	kitasamycin	after	inoculation	with
Mycoplas	m gallisepticum (N	$M \pm SE, n = 3$	5).	•			

				<u> </u>				ost tre	atme	ent				······		
Groups		181	Day			IstW					eek			3rd w	eek	
	Het.	Lymph.	Mon.	Eosin	Het.	Lymph.	Mon.	Eosin	Het.	Lymph.	Mon.	Eosin	Het.	Lymph.	Mon.	Eosin
Non	6.5	89.9	4.3	2.9	10.3	80.3	6.9	3.9	10.9	85.6	5.9	4.1	7.3	88.1	6.3	2.9
infected,	±	±	±	±	±	±	±	±	±	±	±	±	±	±	÷	±
non	0.05	6.7	0.06	0.02	0.2	3.6	0.05	0.01	0.3	3.5	0.1	0.2	0.1	9.1	0.1	0.1
treated		i					_									
Non	15.3	70.7	3.7	2.3	14.7	81.9	7.7	3.7	9.5	85.3	4.7	3.7	7.7	86.2	8.0	2.0
infected,	±	±	±	±	±	±	±	±	±	±	±	±	l ± ∣	t ±	±	±
treated	1.1	3.4	0.05	0.01	0.6	7.4	1.6	0.05	0.1	1.2	0.2	0.1	0.2	3.4	0.3	0.6
Infected,	16.6	65.9	4.9	2.3	14.8	74	6.0	3.9	12.5	82.6	7.8	3.3	9.7	74	9.7	2.5
non	±	±	±	±	±	±	±	±	±	±	±	±	±	\ ±	±	+
treated	3.6	5.5	0.01	0.05	0.4	3.2	2.3	0.01	0.3	2.3	0.6	0.1	0.1	2.6	0.7	0.2
Infected,	14.7	63.7	4.3	2.4	13.3	72	6.9	3.3	17.6	85.4	12.7	3.5	10.8	85	19.9	2.9
treated	±	±	±	±	÷±	±	±	±	±	±	±	±	] ±	±	±	±
	1.2	4.7	0.02	0.06	0.6	4.4	0.6	0.2	1.2	1.2*	$1.2^{+}$	0.2	$0.2^{+}$	7.1	1.6+	0.1

\* P < 0.05 compared with non infected, non treated + P < 0.05 compared with infected, non treated Het.: hetrophils Lymph: lymphocytes Mon.: monocytes Eosin: eosinphils

#### Seham Malhat et al.

6		i neurity und my	1	$1 \text{ chickens} (\text{WI} \pm \text{S})$	o, n o j.
Item	Group			eatment	
Item	Group	1 <sup>st</sup> Day	1 <sup>st</sup> Week	2 <sup>nd</sup> Week	3 <sup>rd</sup> Week
Serum total	Ι	4.98±0.66	5.89±0.34	4.92±0.42	5.63±0.30
protein	II	4.59±0.73	5.63±0.38	6.53±0.47	6.36±0.34
	III	4.35±0.57	5.15±0.29	4.30±0.36	4.95±0.27*
	IV	4.66±0.15	5.52±0.29	$6.61 \pm 0.39^+$	$5.30\pm0.29^{+}$
Albumin	Ι	2.8±0.1	3.3±0.2	2.7±0.2	3.17±0.16
	II	2.15±0.41	2.7±0.2	2.0±0.2	2.56±0.18
	III	2.4±0.3	2.8±0.1	2.42±0.2	2.77±0.15
	IV	2.6±0.3	2.0±0.1	2.6±0.22	2.97±0.19
α-globulin	Ι	0.76±0.09	1.01±0.06	0.84±0.07	0.96±0.07
	II	0.86±0.11	1.13±0.069	0.95±0.08	1.08±0.09
	III	0.67±0.08	0.88±0.05	0.74±0.06	0.84±0.04
	IV	0.71±0.09	0.94±0.05	0.79±0.06	0.90±0.045
β-globulin	I	0.90±0.11	1.18±0.07	0.99±0.08	1.14±0.011
	II	0.04±0.13	$1.38 \pm 0.08$	1.15±0.09	1.32±0.05
	III	0.73±0.09	0.96±0.05	0.81±0.06	0.92±0.04
	IV	0.87±0.11	1.15±0.07	0.97±0.081	1.10±0.05
δ-globulin	Ι	1.07±0.13	1.26±0.07	1.06±0.08	1.22±0.061
_	II	1.21±0.15	$1.42 \pm 0.08$	2.19±0.09	2.37±0.061
	III	0.94±0.11	1.11±0.06	0.93±0.07	1.06±0.05
	IV	1.01±0.12	1.19±0.07	$2.99 \pm 0.08^{+}$	$2.14 \pm 0.05^{+}$

**Table (7):** Effect of Kitasamycin (1gm/kg.b.w) administered in drinking water daily for 5successive days on the serum total protein, albumin  $\alpha$ ,  $\beta$  and  $\delta$ -globulins (gm/100ml) of healthy and mycoplasma infected chickens (M ± SE, n = 5).

\* P < 0.05, compared with non infected, non treated.

+ P < 0.05, compared with infected, non treated.

I: non infected, non treated. III: infected, non treated II: non infected, treated.

IV: infected, treated.

Table (8): Effect of Kitasamycin (1gm/kg.b.w) administered in drinking water daily for 5-<br/>successive days on the serum IgG, M and A (gm/100ml) of healthy and<br/>mycoplasma infected chickens ( $M \pm SE$ , n = 5).

Item	Group		Post tre		
100441	Group	1 <sup>st</sup> Day	1 <sup>sr</sup> Week	2 <sup>nd</sup> Week	3 <sup>rd</sup> Week
IgG	I	1081.6±105.2	1121.6±84.0	1100±82.91	1120±84.2
	II	1216.8±118.35	1261.8±95.2	1237±93.2	1260±94.7
	III	1246.4±92.05	1281.4±24	96.5±72.5	980±73.6
	IV	1214.0±98.6	1251.5±79.3	1131.2±77.3 <sup>+</sup>	1050±78.9 <sup>+</sup>
IgM	I	240.34±23.11	244.21±17.8	248±16.8	247±16.79
	11	241.5±20.12	246±16.3	257±13.1	260±15.81
	III	240±11.1	240±17.10	220±11.2	235±13.20
	IV	248±10.12	245±13.11	260±10.20*	265±11.20 <sup>+</sup>
IgA	1	78.8±7.6	81.75±6.17	80.18±6.05	87±6.62
_	II	79.1±8.6	89±6.8	89.9±7.11	92±7.3
	III	78.10±7.3	88±7.1	69.1±6.3	79.11±6.5
	IV	80.9±6.6	89±6.8	75.8±5.4 <sup>+</sup>	90.3±8.7*

\* P < 0.05, compared with non infected, non treated.

+ P < 0.05, compared with infected, non treated.

I: non infected, non treated.

III: infected, non treated

II: non infected, treated. IV: infected, treated.

## Zag. Vet. J.

Table (9): Effect of Kitasamycin (1gm/kg.b.w) administered in drinking water daily for 5successive days on the lymphocyte transformation index, phagocytosis% and killing % and chemotactic % of healthy and mycoplasma infected chickens (M ± SE, n = 5).

<u> </u>		· · · · · · · · · · · · · · · · · · ·	Post tr	eatment		
Item	Group	1 <sup>sr</sup> Day	1 <sup>st</sup> Week	2 <sup>nd</sup> Week	3 <sup>rd</sup> Week	
Lymphocyte	I	1.24±0.12	$1.18 \pm 0.06$	1.056±0.056	1.12±0.06	
transformation	II	1.40±0.13	1.19±0.06	2.19±0.063	2.29±0.06	
index	III	1.09±0.11	0.92±0.05	0.90±0.049	0.85±0.02	
	ĪV	1.17±0.11	1.10±0.05	2.99±0.053 <sup>+</sup>	$2.08\pm0.5^+$	
Phagocytosis %	Ī	71.46±2.5	75.50±4.52	70.6±4.5	73.71±5.48	
	II	72.31±4.12	74.94±5.19	78.4±5.08	82.1±5.10	
	III	73.14±3.13	76.31±4.03	61.48±3.95	64.8±6.1	
· ·	IV	71.5±2.12	77±6.32	$76.88 \pm 4.5^+$	79.6±7.2 <sup>+</sup>	
Killing %	Ι	68.37±4.81	73.11±4.03	67.14±3.37	70.60±3.93	
-	II	66.16±3.28	71.12±4.54	77±1.61	78.11±5.10	
]	III	61.1±2.11	73.11±3.11	59.1±1.81	60.01±1.10	
	ĪV	67.2±3.12	77.12±4.11	75.12±3.11 <sup>+</sup>	79.31±3.10 <sup>+</sup>	
Chematoctic %	I	1.43±0.09	1.35±0.02	1.35±0.08	1.55±0.01	
	II	1.61±0.108	$1.63 \pm 0.03$	2.52±0.03	2.76±0.02	
	III	1.23±0.08	1.77±0.05	0.95±0.02	1.00±0.07	
	IV	1.34±0.09	1.33±0.06	2.27±0.06 <sup>+</sup>	2.31±0.05 <sup>+</sup>	

IV: infected, treated.

\* P < 0.05, compared with non infected, non treated.

+ P < 0.05, compared with infected, non treated. II: non infected, treated.

I: non infected, non treated.

III: infected, non treated

#### REFERENCES

- 1.Jordan, F.T. (1990): Poultry diseases, 3rd ed. Enterobacteriaceae and Avian mycoplasmosis chapters.
- 2.Clank, B.W.; John, H.; Uoder, M.W. (1991): Disease of poultry enterobacteriaceae and mycoplasmoses chapters.
- 3.Gale, G.O.; Layton, H.E.; Short, A. and Kemp, G.A. (1967): Effect of some antimicrobials on avian mycoplasmosis. Ann. New York, Acad. Sci., 143:239.
- 4. Villemin, P.; Brugere, H. and Brugere, J. (1984): Letraitement des infections respiratories des volailles. Recueil de Med. Vet., 160:117-1128.
- *H.W.* Mycoplasma 5.Yoder. (1978): gallisepticum infection in diseases of poultry. Iowa State Univ. Press, Ames, Iowa, U.S.A., PP. 236-250.
- 6.Razin, S. and Tully, J. (1983): Methods in mycoplasmology. Vol. I. Academic Press, N.Y. London.

- 7.Jordan, F.T. and Knight, D. (1984): The minimum inhibitory concentration of kitasamycin, tylosin and tiamulin for Mycoplasma gallisepticum and their protective effect on infected chicks. Av. Path., 2:151-162.
- (1986): Vet. *N.C.* Schalm's 8.Jain, Haematology 4<sup>th</sup> ed. Lea and Febiger. Philadelphia.
- 9. Thorn, C. J.; Morris, J. A. and Little, T. W. (1982): A spectrum of immune response and pathological conditions between certain animals species to experimental M. bovis infection . Br. J. Exp. Path. 63, 562-567.
- 10.Burrells, S. and Well, P. (1977): In vitro stimulation of avian lymphocytes. Res. Vet. Sci., 23:84-86.
- 11.Hanks, J.H. and Wallace, J.H. (1958): Determination of cell viability. Proc. Soc. Exp. Boil. Med., 98:183-192.
- 12.Hudson, L. and Hay, F. (1980): Immunology, 2<sup>nd</sup> Ed. Blackwell Scientific Publication, Oxford, London, Boston.

- 13. Charles, R.; Carpenter, B. and Bose, J. (1978): Suppression of the mitogens stimulated blastogenic response during reticuloendotheliosis of virus induced tumorigenesis. J. Immunol., 120:1313-1320.
- 14.Antley, P. and Hazen, K. (1988): Role of yeast cell growth temperature on Candida albicans virulence in mice. Infect. Immunity, 56:2884-2890.
- 15. Weichselbaum, T.E. (1946): An accurate and rapid method for determination of proteins in small amounts of blood serum and plasma. Am. J. Clin. Pathol. 16:40.
- 16.Davis, B.J. (1964): Disc electrophoresis, methods and application to human serum proteins. J. Ann. New York. Acad. Sci., 121:404-428.
- 17.Snedecor, G.W. and Cochran, G. (1967): Statistical method, 6<sup>th</sup> ed. Iowa State Univ. Press. Amer. Iowa, U.S.A.
- 18.Stipkovits, L. and Kempf, I. (1996): Mycoplasmosis in poultry. International office of Epizootics. Sci. and Tech. Rev., 15:1495-1525.
- 19.Kempf, I.; Cacou, P.M.; Guittet, M.; Ollivier, C. and Morin, M. (1988): Evaluation of the efficacy of kitasamycin in chickens experimentally infected with Mycoplasma gallisepticum Recueil. Med. Vet., 164:653-659.

- 20. Wieliczko, A.; Mazurkiewicz, M. and Pawiak, R. (1988): Efficacy of some antibiotics in controlling experimental mycoplasmosis in chicks. Meducuna. Vet., 2:88-91.
- 21.Lillehoj, E.P.; Yun, C. and Lillehoj, H. (2000): Vaccines against the avian enteropathogens. Anim. Health. Res. Rev., 1:47-65.
- 22. Warner, N.; Szenbrg, A. and Burent, F. (1962): The immunological role of different lymphoid organs in the chicken. Aust. J. Exp. Boil. Med. Sci., 40:373-382.
- 23. Kobayashi, H.; Sonmez, N. and Yamamoto, K. (1996): In vitro susceptibility of Mycoplasma hyosynoviae and M. hyorhinis to antimicrobial agents. Nat. Institute of Animal Health, Ibaraki, 305, Japan.
- 24.Cerda, R.; Giacoboni, G. and Landoni, M. (2002): In vitro antibiotic susceptibility of field isolates of Mycoplasma synoviae in Argentina. Avian Diseases, 46:215-218.
- 25.Laval, A.; Viso, M. and Berhanu, A. (1988): Immunomodulating effects of two antibiotics, chloramphenical and kitasamycin in the chicken. Annalis. Richer. Nat. Vet., 19:259-266.

تأثير الكيتاساميسين على مرض الميكوبلازما والاستجابة المناعية في كتاكيت التسمين

# \*سهام ملهط، \* عزه أيوب، \* إيلين منقريوس

# \* قسم الكيمياء الحيوية والنقص الغذائي والسموم - معهد بحوث صحة الحيوان بالزقازيق

الملخص

تسم في هذا البحث دراسة الكيتاساميسين، المضاد الحيوى من مجموعة الماكروليدز والذى يتمتع بفاعلية فى معظم الأمراض البكت يرية في الدجاج وبصفة خاصة ميكروب الميكوبلازما جليسيبتكم والتى تشكل خطرا حقيقيا على صناعة الدواجن ونظرا لأن هذا العقار لا نتوفر لدينا عنه كثير من الدراسات عن تأثيراته العلاجية المختلفة في الدجاج السليم أو المصاب فقد استهدفت هذه الدراسة استبيان المزيد من المعلومات ولهدف تقييم كفاءة عقار الكيتاساميسين لعلاج الإصابة معمليا بمسرض المسيكوبلازما فقد أجريت تجارب لدراسة تأثير الكيتاساميسين لعلاج المسليم أو الدجاج المعدي معلمي الكيتاساميسين المزيد من المعلومات ولهدف تقييم كفاءة عقار الكيتاساميسين لعلاج الإصابة معمليا بمسرض المسيكوبلازما فقد أجريت تجارب لدراسة تأثير الكيتاساميسين على كل من المناعة الخلوية والدموية في الدجاج المعدى صناعيا بالميكوبلازما والغير معدى وأيضا تمت دراسة هذا الدواء على الأعراض الناجمة وكذا على معدل الزيادة أو النقصان في وزن الكتاكيت.

تـم فـي هـذه الدراسـة استخدام مائه كتكوت من سلالة هبرد عمر يوم واحد. ولقد تم ذبح عشرون كتكوتا كعينة عشـوائية للـتأكد من خلوها من مرض الميكروبلازما وذلك بزراعة عينات من الجهاز النتفسى لعزل الميكروب – وأيضا بإجـراء الاختبارات السيرولوجية ومنها اختيار التلازن ولقد وجد أن الدجاج الغير مصاب بالميكوبلازما يعطى نتائج سلبية باختيار التلازن. وعند عمر ٢١ يوما تم نفسيم هذه الكتاكيت إلى أربعة مجمو عات متساوية كل منها عشرون كتكوتا.

المجموعة الأولى: وتستخدم كمجموعة ضابطة غير معداه وغير معالجة.

المجموعــة الثانية: غير معداة ومعالجة عند اليوم ٢٨ من العمر بدواء الكيتاساميسين الذي يضاف الى ماء الشرب بجرعة ١جم/كجم من وزن الجسم يوميا لمدة خمسة أيام متتالية.

**المجموعـــة** الثالــــثة: يـــتم عداوها نجريبيا بميكروب الميكوبلازما بحقنه في الأكياس الهوائية بجرعة ٠,٢ مللى عند اليوم الواحد والعشرون وغير معالجة.

**المجموعــة الرابعة: ي**تم عداوها تجريبيا بميكروب الميكوبلازما بنفس الطريقة السابقة وبنفس جرعة الميكروب وعند نفس العمــر ثم تعالج عند اليوم الثامن والعشرين من العمر بدواء الكيتاساميسين فى ماء الشرب يوميا لمدة خمسة أيام منتالية بجرعة ١جم/كجم من وزن الجسم.

ثم بعد آخر جرعة علاجية بيوم، وأسبوع وأسبوعين وثلاثة أسابيع تم وزن الكتاكيت، وملاحظة أعراض المرض وتم ذبح خمسة كتاكيت من كل مجموعة في كل مرة وتم فحص أعضاءها الداخلية ومدى معدل الإصابة وعلاجها وتم أخذ عينتيم مـن الـدم مـن كل كتكوت أحدهما تؤخذ على هيبارين المانع للتجلط وذلك لدراسة تأثير الدواء على العدد الكلى والمنوعى لكـرات الـدم البيضاء والأخرى بدون مانع للتجلط لفصل مصل الدم وذلك لدراسة تأثير الدواء المستخدم على المناعة بفرعيها الخلوية والدموية.

وتم تقييم تأثير الدواء على ميكروب الميكوبلازما بإجراء الاختبارات السيرولوجية – كما تم زرع عينات من الجهاز التنفسي لمعزل الميكروب وفحص دم الطيور وندوين عدد كرات الدم البيضاء وإجراء الفحص التشريحي بعد النبح لتحديد نسبة وشدة الإصابة داخل الجهاز التنفسي للطائر وفي الأعضاء الأخرى من الجسم – ثم أيضا ملاحظة أوزان الطيور قبل وأثناء وبعد التجربة وأوضحت النتائج أن أوزان الدجاج المعالج كانت أعلى الأوزان.

كما كان تأثير المضاد الحيوى هو تقليل شدة التغيرات في الأكياس الهوائية وكان معدل عزل ميكروب الميكوبلازما أقسل فسي الدجساج المعسالج كما أن الدواء أدى إلى تقليل نسبة الوفيات – كما أعطت عينات المصل المأخوذة من الدجاج المصساب بالمسيكوبلازما نستائج ايجابية في اختيار التلازن في حين التي لم تصاب بالعدوى وأيضا التي عولجت بالمضاد الحيوي أعطت نتائج سلبية.

كما أنه أوضحت نتائج اختيار مانع التجمع الدموى إنتاج الأجسام المضادة بنسبة عالية في الدجاج المعالج بالكيتاساميسين مما يثبت أن الدواء له تأثيرا منشطا للجهاز المناعى للطائر.

كما أنه تبيهن من هذه الدراسة أن دواء الكيتاساميسين يؤدى إلى حدوث زيادة معنوية في معامل تنشيط الخلايا الله يمفاوية في الدجاج المعداة بميكروب الميكوبلازما والغير معداة كما أحدث أيضا زيادة معنوية في نسبة القتل فى الخلايا البلعمية ومعامل الانجذاب والذي يعزي إلى تأثير الدواء.

وأيضــــا أحـــدث الـــدواء زيادة معنوية في العدد الكلى لكرات الدم البيضاء والخلايا الملتهمة الكبيرة وعدد الكريات الحامضية والخلايا القاعدية في الدجاج المعدى بالميكوبلازما.

ولقد أحدث الدواء زيادة معنوية فى مستوى البروتينيات الكلية فى المصل وكذا الجلوبيولنيات وبالتالى يستخلص من هذه الدراسة أن الكيتاساميسين علاج جيد ضد مرض الميكوبلازما ويتضح ذلك في اختفاء أعراض الميكوبلازما فى الدجاج المعالج وكذا قلة الإصابة فى الأعضاء الداخلية للدجاج المصاب والمعالج وكذا زيادة وزن الدجاج وأيضا أدى الدواء الى تقليل نسبة الوفيات التى يسببها ميكروب الميكوبلازما.

وفسوق كمل ذلسك فإن الدوأء يؤدى الى زيادة مناعة الطائر الخلوية والدموية في كل من الدجاج السليم والمريض بالميكوبلازما.

ولذلــك ينصـح باستخدام الكيتاساميسين للقضاء على ميكروب الميكوبلازما والتى تشكل خطورة حقيقية في صناعة الدواجن.

وعلم الأخص أن هذا الدواء منشطاً للمناعة لذلك يعطى أفضل النتائج لأنه يجعل الاستجابة المناعية للطائر أسرع وأقوى في القضاء على المرض.