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EFFICACY OF FLOUROQUINOLONE (PEFLOXACIN) ON MYCOPLASMA INFECTION IN CHICKENS

MANAL ABO EL-MAKAREM AND MONA M. MOHAMED SHAKER
Mycoplasma Department, Animal Health Research Institute

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SUMMARY

The efficacy of the newer fluoroquinolone (pefloxacin) given alone or in-combination with levamisole against Mycoplasma gallisepticum (MG) in experimentally infected chickens was investigated. Two hundred one day old chicks were used. The use of levamisole as a non-specific immunostimulant with pefloxacin 10 mg/kg body weight showed good curative effect which was indicated by decrease in mortality rates, increase in the body weight gain, high feed conversion as well as low rate of re-isolated MG and low titre in HIT as compared with those received pefloxacin alone 10 mg/kg body weight or levamisole 2.5 mg/kg body weight alone.

INTRODUCTION

Mycoplasma gallisepticum infection is considered a major cause of economic losses to the poultry industry, previous works showed that the use of antibiotics do not result in complete elimination of mycoplasmas from host tissues (Hamdy et al., 1983). Among these new compounds fluoroquinolones have a bactericidal action on susceptible bacteria (Eliopoulos et al., 1984 and Anadon, 1993). Pefloxacin is a new member of flouroquinolones which penetrates intracellularly and acts on the DNA gyrase enzyme (intracellular enzyme) (Brown, 1996).

Also, Mycoplasma gallisepticum (MG) is considered as the primary cause of chronic respiratory disease (CRD), other organisms frequently cause complications, feed consumption is reduced and the birds losses its body weight (Shaker, 1995). MG has an immunosuppressive effect (Schwab, 1973 and Ata and El-Shabiny, 1993). The use of immunopotentiator was recommended by Bluming (1975). Levamisole was proved to be an immunostimulant agent and could be applied in poultry diseases e.g. Newcastle disease (Bastami et al., 1991); Salmonellosis (Burdarov et al., 1983 and Manal et al., 1998).

In this work, the non-specific immunostimulator (levamisole) was used simultaneously with pefloxacin to enhance the immune response of chickens experimentally infected with MG and enhance the efficacy of pefloxacin.

MATERIAL AND METHODS

I. Drugs:

1. Pefloxacin:-

It is a novel fluoroquinolone, possesses considerable in vitro potency against a broad range of veterinary pathogens including Mycoplasmas. It was obtained in the form of peflodad solution 10% (Pefloxacin 10%). It was orally administered for 5 successive days.

2. Levamisole:-

Levamisole hydrochloride (Pamisole ® Amoun for Pharmaceuticals Industries Co., Egypt) was used orally in a dose of 2.5 mg/kg body weight (Padmavattu et al., 1988).

II. Experimental birds:-

1. Chickens:-

Two hundred one day old breed chicks were used in this study. The birds were obtained form Middle East Company. They were mycoplasma-free as proved bacteriologically and serologically, they were kept on balanced suitable commercials ration and routine vaccination programs. After 15 days, the chicks were divided into five equal groups.

III. Strains:-

Mycoplasma gallisepticum virulent strain of MG (S6) was kindly obtained from Mycoplasma Research Department, Animal Health Research Institute.

IV. Inoculation:-

Fifteen day-old chicks were inoculated intranasally with 0.1 ml (containing 10^7 CFU) of MG as described by Talkington and Kleven (1985) and the chicks were left for one week.

V. Experimental design:-

Chickens were divided into 5 equal groups and treated as follows:-

1. **First group:** Inoculated with MG S6 and treated with pefloxacin 10 mg/kg body weight daily for 5 successive days.
2. **Second group:** Inoculated with MG S6 and given levamisole (2.5 mg/kg b.wt.) daily for 5 successive days.
3. **Third group:** Inoculated with MG S6 and given both levamisole (2.5 mg/kg b.wt.) and pefloxacin 10 mg/kg body weight daily for 5 successive days.
4. **Fourth group:** Inoculated with virulent MG (S6) and remained untreated (control positive).
5. **Non infected,** non treated group (control negative).

VI. Isolation and purification:-

Isolation of mycoplasma from lungs, trachea and air sacs and purification were performed as de-

scribed by Adler et al. (1958).

VII. Serological examination:-

- Slide agglutination test (SAT) (Adler et al., 1958)
- Haemagglutination inhibition test (HIT) (Yoder, 1980).

RESULTS

Ten chickens were slaughtered each week to examine the mean body weight, spleen weight, reisolation and identification of mycoplasma form lungs and air sacs as well as serological examinations were done.

The chickens in the fourth control group showed respiratory manifestations, decrease in body weight, poor physical condition, sinusitis and high mortality rates. Meanwhile at fourth week, the body and spleen weights were higher (590, 1.1 g) in fifth group (negative control), followed by third group (370, 1.9 g) (which treated with pefloxacin and levamisole), then the second group (340, 1.7 g) (which treated with levamisole) while the first group was (310, 1.3 g) which treated with pefloxacin) as shown in table (1).

The reisolation of MG from lungs was summarized in table (2) which shows 100% reisolation in group four at all weeks (positive control). Meanwhile, the third group which (treated with both drugs) showed lowest percentages at fifth week, 10%, then the first group (treated with pefloxacin only revealed low percentage 30% than that of the second group (treated with levamisole 50%).

Table (1) Changes in mean body weight, internal organs and mortality rate in chickens treated with pefloxacin and levamisole.

Group	3rd week		4th week		5th week		Mortality rate (%)
	b.wt. (g)	spleen	b.wt. (g)	spleen	b.wt. (g)	spleen	
1- Infected treated with pefloxacin	260	0.6	310	1.3	850	1.7	1
2- Infected treated with levamisole	290	0.8	340	1.7	960	2.1	15.4
3- Infected treated with both pefloxacin and levamisole	315	1.2	370	1.9	1010	2.5	-
4- Control positive	205	0.4	280	0.5	430	0.8	88
5- Control negative	205	0.2	340	0.7	430	1.1	-

Table (2) Reisolation and identification of Mycoplasma gallisepticum from lungs and air sac lesions.

Group	Reisolation from:											
	Lungs						Air sacs					
	3rd week		4th week		5th week		3rd week		4th week		5th week	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
1- Infected treated with pefloxacin	5	50	4	40	3	30	0	0	2	20	2	20
2- Infected treated with levamisole	6	60	5	50	5	50	1	10	2	20	3	30
3- Infected treated with both pefloxacin and levamisole	3	30	2	20	1	10	0	0	1	10	2	20
4- Control positive	10	100	10	100	10	100	10	100	10	100	10	100
5- Control negative	0	0	0	0	0	0	0	0	0	0	0	0

Table (3) Detection of HI antibody titers in serum of examined chickens.

Group	SAT			HIT					
				3rd		4th		5th	
	3rd	4th	5th	Min	Max	Min	Max	Min	Max
1- Infected treated with pefloxacin	+	+	+	1/64	1/128	1/32	1/64	1/16	1/64
2- Infected treated with levamisole	+	+	+	1/128	1/256	1/256	1/512	1/512	1/1024
3- Infected treated with both pefloxacin and levamisole	+	+	+	1/64	1/128	1/16	1/64	1/16	1/32
4- Control positive	+	+	+	1/128	1/256	1/256	1/512	1/256	1/1024
5- Control negative	-	-	-	1/8	1/8	1/8	1/8	1/8	1/8

N.B. Titer 1/8 was considered as negative result.
Titer 1/16 was considered as suspicious result.
Titer 1/32 was considered as positive result.

Serological examination by slide agglutination test were positive in all groups except the fifth group (control negative).

In fourth week, HIT was increased sharply in the fourth control positive group and the second group (treated with levamisole) but decreased in the first group (treated with pefloxacin) as shown in table (3).

DISCUSSION

The non-specific immunostimulant levamisole was used simultaneously with pefloxacin to enhance the immune response of chickens experimentally infected with MG and enhances the efficacy of pefloxacin. The present results revealed that levamisole given alone or in combination with pefloxacin in different doses used shows decreased mortality rates, high body weight gain and increased feed conversion when compared with other groups. These results are in agreement with that obtained by Afify (1987) and Manal et al. (1998) which may be attributed to improvement of healthy status of chickens or increase of digestibility and consequently increasing nutrients absorption. Also, similar findings were recorded by Padmavattu et al. (1988) and Manal et al. (1998).

In addition, chickens administered levamisole only showed a decreased isolation rate and decreased lesion scores in lungs and air sacs which may be attributed to increasing the phagocytosis (Soppi et al., 1979) or rising in polymorphonuclear cells or T-cells activation (Maheswaran et al., 1980 and Gorwnov and Bonoska, 1987).

Also, pefloxacin is a new member of fluoroquinolones which penetrates intracellularly and acts on the DNA gyrase enzyme (intracellular enzyme) (Kayser, 1985 and Brown, 1996).

Haemagglutination inhibition test (HIT) showed antibody titre against MG in chickens infected with virulent strains of MG with no difference from levamisole received group ones, which are in accordance to that obtained by Soppi et al. (1979) who recorded the absence of the effect of levamisole on antibody response to thymus independent antigen. These results are in agreement with Dajani (1980); Afify (1987) and Manal et al. (1998).

The first and third groups were positive for re-isolation of MG and antibodies detected by SAT and HIT were low. The present results proved that pefloxacin is of value in treatment and control of MG infection especially when used in combination with an immunostimulant e.g. levamisole. These results are agreed with that mentioned by Bradbury et al. (1994); Abdel Aziz et al. (1996)

and Ewing et al. (1998) who revealed that the newer fluoroquinolones-pefloxacin is one of them showed antibacterial activity and bactericidal action at low concentrations over naladixic acid and other quinolones.

These results indicated that levamisole in a dose of 2.5 mg/kg body weight for 5 days led to increased immuno-protection mechanism against the challenge with virulent MG strain. This may support that levamisole also acts as a non-specific immunostimulant in combination with pefloxacin 10 mg/kg body weight which yielded a good curative effect and manifested by decrease in the mortality rates, increases in the body weight gain, highest food conversion as well as low rate of re-isolation and increasing titres against MG.

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