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DICLAZURIL INTERACTION WITH AVOPARCIN AND VIRGINIAMYCIN AS GROWTH PROMOTERS IN GROWING RABBITS

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SUMMARY

The effect of feeding avoparcin and virginiamycin (10 ppm) alone or in combination with diclazuril (1 ppm) to growing rabbits for 10 successive weeks on absolute body weight, body gain and feed efficiency were studied. Moreover, their effects on blood picture and the activity of AST, ALT as well as urea and creatinine levels were also investigated. In addition, the residual pattern of avoparcin and virginiamycin were also demonstrated using the microbioiogical assay technique.

Avoparcin or virginiamycin supplementation with the basal ration significantly increased the absolute body weight (5.7 or 8.8%), amount of feed consumed (3.2 or 1.6 %) as well as plucked weight. Avoparcin was found to be compatible with diclazuril as their combination induced large improvement in the body weights (10.6 %) and the amount of feed consumed (6.2 %).

Combination of diclazuril with virginiamycin did not alter the absolute body weight and the weight gain, but increased the amount of feed consumed (4.5 %) as compared with group fed virginiamycin alone. Avoparcin or virginiamycin alone and combination of diclazuril with avoparcin significantly increased R.B. Cs count, hemoglobin content and PCV. Combination of diclazuril with avoparcin increased the activity of AST and ALT and that with virginiamycin increased AST and ALT activity as well as urea and creatinine levels. No residues of either avoparcin or virginiamycin were detected at the end of the experiment in organs and tissues of rabbits fed 10 ppm for 10 successive weeks.

In conclusion, feeding avoparcin or virginiamycin to growing rabbits at 10 ppm stimulates growth, feed efficiency and blood picture. Diclazuril (1ppm) was found to be more compatible with avoparcin than viginiamycin.

INTRODUCTION

Antiobiotics are added to feeds of several classes of livestock's to prevent diseases, stimulate growth and to improve feed efficiency.

Avoparcin (Avo) and virginiamycin (Virg) are among the antibiotics which have been used as feed additives to improve body weight gain (Pensak et al., 1982; Miles et al. 1984; hofshagen and kaldhusdal., 1992 and krinke and Jamroz, 1996). Efficacy studies have shown that avoparcin and virginiamycin significantly improve the body weight and the efficiency of food utilization in broiler chickens (Roth-Mair and Kirchgessener 1976, Lesson et al.; 1980; Foster and Stevenson, 1983; Shihata et al. 1989 and Abd El-Aziz and Agog, 1997). These studies showed improvements in the body weight gain and the efficiency of food utilization of about 4 % when avoparcin or viginiamycin were added to the diet at a rate of 10 PPM.

It is a common practice, however in feeding rabbits or broiler chickens to include anticoccidial compounds in the ration. Diclazuril is an anticoccidial commonly used to prevent coccidiosis in rabbits and broiler chickens (Brander et al., 1991).

This study was designed to evaluate the effect of avoparcin or virginiamycin on absolute body weight, weight gains, feed efficiency, blood picture and on the activity of some enzymes in growing rabbits. The interaction of diclazuril with avoparcin or virginiamycin was also investigated as their effects and combination in rabbits had not been fully documented.

Drugs:

1- Diclazuril (Clinacox) was obtained from Janessen Company, Breers, Belgium as 0.5 % premix.
2- Avoparcin (Avotan 50) was obtained from Hoffman-la-Roch Ltd. Basle, Switzerland as 5 % premix.
3- Virginiamycin (Stafac) was obtained from Smithkline Company. Bruxelles, Belgium as 50 % premix.

Rabbits:

A Total number of 60 male clinically healthy white Newzeland weaned rabbits of 530-550 g and of 4 weeks age were used. Rabbits were reared in cages each contains 5 animals. Feed and water were provided *ad-libitum*.

Experiment:

Rabbits were randomly divided into 5 equal groups each of 12 animals. A basal ration of about 17.6 % crude protein, 13 % crude fiber and of 2513 Kilo calorie. DE/Kg (Table 1) was formulated and unsupplemented (control group) or supplemented with 10 mg avoparcin / kg basal ration (Avo group); 10 mg avoparcin and 1 mg diclazuril / kg basal ration (Avo + Dicl. group);

10 mg virginiamycin / kg basal ration (Virg. group) and 10 mg virginiamycin and 1 mg diclazuril / kg basal ration (Virg + Diel. group). All groups were fed for a period of 10 weeks.

Effect on growth and feed efficiency:

Each rabbit in all groups was weighed separately at the begining of the experiment then every two weeks till the end of the experiment, which lasted 10 weeks. The body weight gain for each rabbit was calculated by the difference between the initial and the next weight. The amount of ration consumed for each group was recorded weekly for studying the feed efficiency.

At the end of the experiment, 5 rabbits from each group were slaughtered. Carcasses of slaughtered rabbits were then eviscerated to estimate the weight of meat and bone, giblets (liver, spleen, kidneys and heart), Fur and waste offal.

Determination of residual pattern:

Three animals from avoparcin or viginiamycin treated group were slaughtered at the end of the experiment then at the 1st, 2nd and the 5th day after stopping of the drug medication. Tissue samples (liver, kidney, heart, spleen, and muscle) were taken for studying the residue depletion using the microbiologial assay technique described by Arret et al. (1971) with *Bacillus Subtils* (ATCC 6633) as test organism. The detectable limits were 0.1 ug/g and 0.15ug/g for avoparcin and virginiamycin respectively.

Effect on blood criteria and enzymatic activities:

Two blood samples were taken from the ear vein at the end of the experiment from each rabbit in all groups. The first sample (1 ml) was collected directly into heparinized tubes for hematological examination. The second (2ml) was drawn into centrifuge tubes, allowed to clot and the sera were used for determination of the activity of some enzymes.

Estimation of hemoglobin content as described by Varly (1980), the packed cell volume (PCV) according to Schalm et al., (1975) and erythrocytic and leukocytic counts as explained by Wintrobe (1967) were carried out.

The activities of Asparate Amino Transferase (AST) and Alanine Amino Transferase (ALT) were determined as described by Reitman and Frankel (1957). The creatinine level was measured according to the method explained by Husdan and Rapoport (1968) while urea level by Patton and Crouch (1977)

Data are given as mean ± standard errors and analyzed by students "t" test (Snedecor and Cochran, 1967).

RESULTS

Effect on growth and feed efficiency:

The effects of dietary avoparcin and virginiamycin alone or in combination with

diclazuril for 10 successive weeks on the body gain, feed efficiency and carcass yield are presented in Tables 2,3 and 4.

It is clear from Tables (2) and (3) that combination of diclazuril with avoparcin increased significantly the absolute body weights and the body weight gain as compared with the group fed avoparcin alone. On the other hand, combination of virginiamycin with diclazuril did not alter the absolute body weight and the body gains as compared with the group fed virginiamycin alone. There was a significant ($P \geq 0.001$) increase in the amount of feed consumed in groups supplemented with avoparcin and virginiamycin alone or in combination with diclazuril (Table 2). Significant improvements in the feed conversion rate were recorded in virginiamycin fed group as compared with control ones, and in groups supplemented with combination of diclazuril with either avoparcin or virginiamycin as compared with groups fed avoparcin or virginiamycin alone.

Continuous feeding of avoparcin and virginiamycin increased significantly ($P \geq 0.001$) the body weight gains all over the experimental period as compared with the control group. The gains attributed to avoparcin and virginiamycin were 1541.8 ± 7.8 and 1578.3 ± 3.4 g, while those attributed to the combination of diclazuril with avoparcin or virginiamycin were 1627.5 ± 5.0 or 1590 ± 9.2 g respectively.

All groups fed with avoparcin or virginiamycin alone and the combination of diclazuril with avoparcin, increased significantly ($P \geq 0.001$) the weights of carcass, meat plus bone and giblets as compared with control group or the group fed avoparcin alone respectively (Table 4).

A significant ($P \geq 0.01$) increase in the weight of giblets was recorded in groups supplemented with combination of diclazuril and virginiamycin as compared with the group fed virginiamycin alone. (Table 4).

Residue depletion:

No residues of either avoparcin or virginiamycin were detected in tissues (taken at the end of the experiment or after stopping of medication) of rabbits fed 10 PPM of either drugs for 10 successive weeks.

Effect on blood picture and enzymatic activities:

It is clear from Table (5) that feeding avoparcin or virginiamycin alone or combination of avoparcin with diclazuril increased significantly R. B. Cs count, hemoglobin content and packed cell volume. Combination of diclazuril with either avoparcin or virginiamycin increased significantly ($P \geq 0.001$) the count of W. B. Cs (Table 5). Feeding virginiamycin (10 ppm) to growing rabbits for 10 successive weeks significantly ($P \geq 0.001$) decreased the urea level. Combination of diclazuril with avoparcin

Table 1: Composition and chemical analysis of the basal feed.

Ingredients	%	Items	Calculated chemical analysis (%)
Barely grain	15.0	Crude protein	17.6
Yellow corn	15.3	Crude fiber	13.0
Soybean meal	4.8	Ether extract	2.93
Wheat Bran	29.9	Calcium	0.82
Clover hay	12.5	Phosphorus	0.71
Sun flower meal	10.5	Lysine	0.8
Cotton seed meal	6.75	Methionin + Cystine	0.6
Molasses	3.0		
Lime stone	1.6		
Common salt	0.3		
Vitamin and mineral premix	0.3		
DL-methionin	0.05		

Table 2: Effect of feeding avoparcin (10 ppm) and virginiamycin (10 ppm) alone or in combination with diclazuril (1 ppm) for 10 successive weeks on the performance of growing rabbits. (Mean ± S. E., n = 10)

Items	Groups				
	Control	Avo	Avo + Dicl	Virg.	Virg. + Dicl
Initial live body weight (g)	530.83 ± 8.5	528.0 ± 8.15	535.0 ± 11.4	548.3 ± 7.10	545.0 ± 6.51
Final live body weight (g)	1953.83 ± 6.9	2069.8 ± 7.41 ***	2162.5 ± 8.6 ***	2126.6 ± 7.7 ***	2135.0 ± 6.9
Weight gain (g)	1423.0 ± 7.78	1541.80 ± 7.8 ***	1627.5 ± 5.0 ***	1578.3 ± 3.4 ***	1590.0 ± 9.2 ***
Feed Consumption (g)	6700.0 ± 18.7	6920.7 ± 27.2	7115.9 ± 8.7 *	6810.0 ± 8.6 *	7116.9 ± 5.7 *
Feed conversion rate	4.71 ± 0.12	4.47 ± 0.18	4.34 ± 0.09	4.28 ± 0.11	4.80 ± 0.10

* Significant at p≥ 0.05
** Significant at p≥ 0.01
*** Significant at p≥ 0.001

Table 3: Effect of feeding avoparcin (10 ppm) and virginiamycin (10 ppm) alone or in combination with diclazuril (1 ppm) for 10 successive weeks on body weight gain of growing rabbits. (Mean ± S. E., n = 10)

Groups	Body weight gain (g) after				
	2W	4W	6W	8W	10W
Control	258.0 ± 2.36	540.4 ± 7.44	872.0 ± 6.23	1140.2 ± 2.5	1423.0 ± 7.7
Avo.	*** 283.3 ± 2.18	*** 596.1 ± 5.05	*** 955.5 ± 4.35	*** 1261.8 ± 2.5	*** 1541.8 ± 7.8
Avo. + Dicl.	*** 303.21 ± 1.71	*** 632.6 ± 3.04	*** 990.1 ± 3.8	*** 1307.5 ± 6.1	*** 1627.2 ± 5.0
Virg.	*** 290.4 ± 3.73	*** 599.6 ± 7.03	*** 1002.2 ± 3.3	*** 1279.8 ± 5.4	*** 1578.3 ± 3.4
Virg. + Dicl	287.7 ± 5.49	595.8 ± 3.35	1018 ± 3.55	128.31 ± 6.2	1590.4 ± 9.2

*** Significant at p≥ 0.001

Table 4: Effect of feeding avoparcin (10 ppm) and virginiamycin (10 ppm) alone or in combination with diclazuril (1 ppm) for 10 successive weeks on carcass yield of growing rabbits. (Mean ± S. E., n = 5).

Items	Groups				
	Control	Avo	Avo + Dicl	Virg.	Virg. + Dicl
Carcass (g)	1868.2 ± 9.43	*** 1985.5 ± 7.81	*** 2090.6 ± 4.76	*** 2016.25 ± 8.8	2025 ± 7.66
Meat & Bone (g)	1210.4 ± 3.47	*** 1309.4 ± 7.21	*** 1402.0 ± 3.72	*** 1338.3 ± 4.32	1340.0 ± 2.19
Giblets (g)	99.6 ± 1.24	*** 116.70 ± 1.35	*** 127.2 ± 1.40	*** 120.7 ± 1.68	** 128.0 ± 1.21
Fur (g)	224.0 ± 1.29	227.6 ± 1.56	229.5 ± 1.19	223.9 ± 1.29	225.0 ± 2.91
Waste offal (g)	334.01 ± 2.56	332.0 ± 1.4	331.9 ± 1.36	333.4 ± 1.2	332.1 ± 2.11

** Significant at p≥ 0.01
*** Significant at p≥ 0.001

Table 5: Effect of feeding avoparcin (10 ppm) and virginiamycin (10 ppm) alone or in combination with diclazuril (1 ppm) for 10 successive weeks on blood picture of growing rabbits. (Mean ± S. E., n = 10).

Groups	Rbcs (10^6/mm^3)	Hb (gm%)	PCV (%)	Wbcs (10^3/mm^3)
Control	6.16 ± 0.10	11.96 ± 0.13	40.6 ± 0.5	7.18 ± 0.22
Avo.	*** 7.27 ± 0.13	*** 13.6 ± 0.25	*** 43.9 ± 0.32	7.24 ± 0.18
Avo. + Dicl.	** 8.16 ± 0.16	*** 15.74 ± 0.34	*** 47.4 ± 0.12	*** 9.40 ± 0.27
Virg.	*** 7.71 ± 0.2	*** 14.7 ± 0.21	*** 43.6 ± 0.61	7.13 ± 0.25
Virg. + Dicl	7.67 ± 0.13	14.48 ± 0.17	43.9 ± 0.12	*** 11.5 ± 0.37

** Significant at p≥ 0.01
*** Significant at p≥ 0.001

Table 6: Effect of feeding avoparcin (10 ppm) and virginiamycin (10 ppm) alone or in combination with diclazuril (1 ppm) for 10 successive weeks on the activity of AST and ALT as well as urea and creatinine levels. (Mean ± S. E., n = 10).

Groups	AST Unit/ml	ALT Unit/ml	Urea mg%	Creatinine mg%
Control	45.0 ± 1.09	26.0 ± 1.15	21.4 ± 0.5	2.21 ± 0.08
Avo.	45.8 ± 1.56	26.5 ± 1.4	22.1 ± 1.31	2.3 ± 0.11
Avo. + Dicl.	** 51.9 ± 1.01	* 30.5 ± 0.62	22.9 ± 0.71	2.15 ± 0.2
Virg.	44.2 ± 0.92	24.9 ± 0.75	*** 17.1 ± 0.39	2.29 ± 0.19
Virg. + Dicl	*** 56.8 ± 1.2	*** 36.1 ± 0.93	*** 25.7 ± 0.51	*** 3.9 ± 0.17

* Significant at p≥ 0.05
** Significant at p≥ 0.01
*** Significant at p≥ 0.001

increased significantly the activity of AST and ALT and that with virginiamycin increased significantly ($P \geq 0.001$) the activity of AST and ALT as well as urea and creatinine levels. (Table 6).

DISCUSSION

Under the conditions of this study, the addition 10 PPM avoparcin or virginiamycin to the ration of growing rabbits produced an increase in the absolute body weight (5.7 & 8.8 %), body weight gains (9.02 & 10.89 %) and food consumption (3.2 & 1.6 %) respectively. These results are comparable to those found by other workers for avoparcin. (Hofshagen, et al. 1992; Pensack et al., 1992 and Krinke et al., 1996) in broiler chickens and for virginiamcyin by Shihata et al. (1989), and Abd El-Aziz and Agag (1996), in chickens and by Soliman et al., (1995) in rabbits. On the other hand these findings are somewhat higher than those reported by Foster and Stevenson (1983) for avoparcin, in whose exmperiment improvement in the efficiency of food utilization and body weights were of order 2%, with littlé change in feed consuption. In addition to the increase in the live body weight there was a significant increase in plucked weight because of the addition of avoparcin or virginiamycin. The inclusion of avoparcin or virginiamycin resulted in an increase in the weight of saleable products in agreement with the work of Spoerl and Kirchgessner, (1978). Shihata et al., (1989) and Proudfoot et al., (1990) for broiler chickens and Soliman, et al. (1995) for growing rabbits.

The growth promoting effect induced by supplementation of avoparcin or virginiamycin could be attributed to their antibacterial effect against Gram-Positive organism which interfere with the absorption of nutrients (Eyssen and Se Somer 1963) and / or due to its sparing effect of the metabolizable energy from the diet (March et al. (1978). In addition, virginiamycin may improve the digestive utilization of nitrogen and amino acids (Canale et al., 1980) or retard the rate of passage of ingesta, so increase the absorptive capacity of the intestine (Fausch, 1981). Moreover Krinke et al., (1996) reported that, the growth promoting effect of avoparcin is related to a restriction in the host animals of response to intestinal bacteria.

Avoparcin (10 ppm) was found to be compatible with diclazuril and large improvements in the body weight, (10.6 %) and the amount of food consumed (6.2 %) were observed. In addition, combination of diclazuril with avoparcin produced proportionally greater carcass weight but reduced the offal weights. Similar findings were previously recorded by Fairly et al., (1985) for combination of avoparcin with lasalocid or salinomycin in broiler chickens. On the other hand, combination of diclazuril with

virginiamycin did not alter the body weight gains and the live body weights but increased the amount of feed consumed (4.5 %) as compared with the group supplemented with virginiamycin alone. Moreover, inclusion of diclazuril and virginiamycin to the feed of growing rabbits induced an increase in the weight of edible giblets, while the weight of carcass and waste offal did not altered. These findings indicated that, combination of diclazuril with virginiamycin was less compatible thant that with avoparcin as growth promoter in growing rabbits.

Our results showed that no detectable level of either avoparcin or virginiamycin were determined at the end or at the 1st , 2nd and 5th day after stopping of medication in tissues of rabbits. This finding suggested even zero withdrawal time for avoparcin and virginiamycin. Similar finding was previously recorded for viginiamycin in broilers (Mulder et al. al., 1976) and pigs (Lauridsen et al., 1988). In addition, Bishop (1996) mentioned that, the withdrawal period was nil for avoparcin and virginiamycin in cattle, lambs, broilers and turkeys.

Our results showed that supplementation of avoparcin and virginiamycin alone or combination of diclazuril with avoparcin to the diet of growing rabbits significantly increased R. B. Cs count, hemoglobin content, and PCV. Similar findings were previously recorded for avoparcin in broilers (Lesson et al., 1980) and for virginiamycin in calves (Skrivanova and Marounek, 1993). Combination of diclazuril with either avoparcin or virginiamycin significantly increase W. B. Cs count. This effect may be attributed to the stimulant effect of the combination on the immune response.

Feeding virginiamycin to growing rabbits significantly decreased the urea level in consistent with the finding reported previously in calves (Skrivanova and Marounek, 1993). In addition, combination of diclazuril with avoparcin increase the activity of AST and ALT and that with virginiamycin increased AST and ALT activity as well as Urea and Creatinine levels. These findings indicated that a slight hepatotoxic and nephrotoxic effects resulted from the combination of diclazuril with avoparcin or virginiamycin.

Conclusively, feeding avoparcin or virginiamycin to growing rabbits at 10 PPM stimulates growth, improves feed efficiency and blood picture. Diclazuril (1 ppm) was found to be compatible with avoparcin as it induces great improvements in the body weights and feed efficiency. On the other hand, diclazuril was less compatible with virginiamycin as their combination may induce slight hepatotoxic and nephrotoxic effects as will as did not alter its effect on growth and body weights.

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