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Effects of Amprolium (Anticoccidial) on Some Productive and Reproductive Parameters of Rabbit Doe (*Oryctolagus cuniculus*)

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Abstract

This study was designed to study the effects of Amprolium (anticoccidial drug) on some productive and reproductive parameters in female rabbit. For this purpose, 24 nulliparous local breed rabbits, aged 7 months on average were divided into four groups of 6 animals each, and randomized into 24 cages. Amprolium was administered at doses of 0.00, 0.20, 0.32 and 0.80 g/L of drinking water for 8 weeks (4 weeks before and after mating). The average Amprolium intake from the calculated water consumption was 0.00, 32.00, 69.00 and 136.00 mg/kg body weight. The main results showed that the pregnancy length of treated rabbits decreased with increasing doses of amprolium, with a significant difference ($p<0.05$) between the lowest (32 mg/kg) and the highest doses (136 mg/kg). The litter weight and milk production decreased significantly ($p<0.05$) in rabbits treated with 32 and 69 mg of Amprolium/kg compared to those of control group. Whatever the physiological stage, there was no treatment-related effects of Amprolium ($p>0.05$) on feed and water consumption, body weight gain, feed efficiency, average weight at birth, sex-ratio and viability of young rabbits in any of the study groups. These data demonstrated that Amprolium exerts an adverse effect on female fertility at all the considered doses.

Keywords: Amprolium; Toxicity; Rabbit; Growth; Reproduction

Introduction

In developing countries, malnutrition is still relevant. Thus, animal proteins which have a very important place in the human diet are difficult to access for most households [1]. The resolution of this problem remains a major concern and it is in this light that the different states encourage breeding by programs that firstly aim to improve breeding conditions and secondly to promote the emergence of new breeding sectors, especially the so-called non-conventional species including cane rat, Japanese quail, Gambian rat, snail and rabbit. Regarding specifically the rabbit, this latter receives special attention for the quality of its white meat, its high prolificacy and its short production cycle [2]; its breeding can contribute to the reduction of the abovementioned food deficit [3]. However, rabbit breeding faces several difficulties due to the lack of control in production methods and the fragility of the animal, especially in tropical regions where atmospheric conditions are favorable to the development of pathogens, making diseases the most redoubtable constraints [4];

among other we have scabies, respiratory diseases, and coccidiosis. The latter is particularly dangerous because it often causes up to 90% mortality [5]. Coccidiosis is treated using a wide range of anticoccidials whose use has grown because of their significant contribution in reducing mortality due to coccidia. In fact, many producers chronically administer products and sometimes at doses higher than those recommended. This massive and uncontrolled use of these drugs may be a significant hindrance to production. Thus some researchers [6] noted the emergence of new strains of coccidia resistant to amprolium. In addition, problems with anorexia, diarrhea, weight loss and muscle weakness were revealed [7] after administration of high doses of anticoccidial (monesin) to hens.

This study's main objective is to contribute to a better understanding of the effects of amprolium on rabbits production and reproduction performance; it is specifically to study the effects of different doses of this product on growth parameters (feed and water consumption, body weight gain and feed conversion) and reproduction (gestation length, litter size and weight, pup viability and milk production).

Materials and Methods

Study Area

The study was conducted at the Teaching and Research Farm (TRF) of the University of Dschang in the Western Cameroon highlands (5-7° LN and 9-12° LE). The altitude is 1400 to 1500 m. The climate is tropical Sudano Guinean, tempered by altitude. The average rainfall is 2000 mm over two seasons: a dry season (mid-November to mid-March) and a long rainy season (mid-March to mid-November). The temperature ranges between 18 and 30°C and the relative humidity is between 80-98% depending on the season.

Animal

24 nulliparous local race does aged between 7-8 months were used. These animals were housed in galvanized iron maternity cages having the following dimensions: 96 cm long, 40 cm wide, and 15 cm high. They received a complete diet (Table 1) formulated *in situ* based on locally available ingredients.

The anticoccidian used

Amprolium-300 WS used in the context of the present study is produced by Interchemie Werken (Holland) and contains 200 mg of Amprolium hydrochloride and 150 mg Amprolium sulphaquinoxalin per gram of powder. Its chemical name is Chloride 5 - [(2-methylpyridin-1ium-1-yl) methyl] -2-propyl-pyrimidin-4-amine. Its molecular formula and molecular weight are C14H19N4Cl and 278.780 g/mol respectively.

Experimental design

A completely randomized block design of 4 treatments and six replications was used. Two weeks before the start of the experiment, animals were subjected to the experimental diet. Then they were weighed on an empty stomach, divided into four homogeneous groups of 6 animals each and randomly housed in 24 different cages. The 4 groups of animals were given *ad libitum* feed. Water was also served *ad libitum* and contained amprolium according to the dosage described in Table 2. The trial took 11 weeks and was conducted in three phases: Four weeks of treatment before mating; four weeks of gestation to assess the reproductive performances and three weeks following delivery to evaluate milk production and performance of young rabbits.

Table 1. Percentage composition of rabbit's diet.

Ingredients	Amount (%)
Corn	26
Middlings	6
Palm kernel cake	11
Soybean meal	12.5
Cotton seed meal	5
VNMC (5%)	5
Fish meal	5
Palm oil	4
Table salt	0.5
Pennisetum purpureum	25
Calculated nutrients analysis	
Nutrients	**Quantity**
Crude protein (%)	17.46
Digestible energy (kcal/kg)	2800
Crude fiber (%)	14.18
Ca (%)	1
P (%)	0.6
Na (%)	0.24
Lysine (%)	1
Methionine (%)	0.4

VNMC. Vitamin Nitrogen and Mineral Complex

Table 2. Dosage used for the administration of the Amprolium and calculation of corresponding doses.

Animal groups	Dosage	Dose g/l of water	Daily intake in mg / kg body weight
To	0 g (control group)	0.0	0
T1	100 g to 500 l of water	0.2	32
T2	100 g to 250 l of water	0.4	69
T3	100 g to 125 l of water	0.8	136

For each group, daily consumption was calculated using the following formula:

DC= (D x WC) / (DT x AW)

With:

DC = Daily consumption of Amprolium (mg/kg)

D = Dose Amprolium (g/ l)

WC = total average water consumption (l)
DT = duration of treatment (day)

AW = average weight of the group (kg)

Data Collection

During the test, the following parameters were evaluated:

The daily consumption of water and feed was obtained by the difference between the quantity served Q and the refusals R (C = Q - R);
The weekly body weight gain was obtained by weighing the fasting rabbits every week;

The litter size was obtained by counting the number of rabbits of each female after delivery; The gestation length was obtained by calculating the number of days between the date of the crossing and the date of delivery;
Milk production at 21 days was evaluated according to the methods described by Lebas et al. [8].

Calculated Parameters

The weight gain was obtained by the difference between the weights of two consecutive weeks. The Feed Conversion Ratio was calculated by dividing the feed consumption of a period by the body weight gain of the same period;
The fecundity was calculated as the ratio between the number of young rabbits born and the number of females having given birth;
The sex ratio of pups was obtained by the ratio between the number of males and females Mortality rate (MR) was calculated by the following formula: MR = Number of deaths x 100 / litter size;

Viability rate (VR) was calculated by the following formula: VR = Number of live rabbit's x 100

/ litter size.

Statistical Analysis

All data were expressed as mean ± standard deviation and the level of homogeneity between groups tested using one-way analysis of variance. When there were significant differences between the averages, the Duncan's multiple range tests was applied to separate at a 5% significance level. The statistical software SPSS 19.0 was used for these analysis.

Results

Effects of Amprolium on feed and water consumption, body weight gain and feed conversion ratio in non-pregnant rabbits

The effects of Amprolium on feed and water consumption, body weight gain and feed conversion ratio in non-pregnant rabbits are summarized in Table 3.

Table 3: Effects of Amprolium on feed and water consumption, body weight gain and feed conversion ratio in non-pregnant rabbits.

Parameters				
			69	136
Feed consumption (g)	3881.50±204.18	3686.25±610.14	3826.75±332.20	4000.50±114.86
Water consumption (l)	9.71±0.90	10.20±2.68	11.85±1.98	11.05±1.87
Body weight gain (g)	358.50±115.94	302.25±114.05	293.75±146.52	343.00±130.37
Feed Conversion Ratio	12.06±5.08	13.47±4.56		13.41±6.41

It appears from this table that although the average feed consumption was highest with the highest dose of Amprolium used, no significant difference (P>0.05) was observed between treatments. Water consumption was lower in rabbits of control group compared to those treated with Amprolium. However, the difference was not significant (P>0.05) between treatments. It also appears from Table 3 that although no significant difference (P>0.05) has been recorded among the treatments in terms of weight gain, the doses 32 and 69 mg/kg had the lowest values.

Feed conversion ratio was not significantly affected by the used Amprolium doses.

Effects of Amprolium on feed and water consumption, body weight gain and feed conversion ratio in pregnant rabbits

The effects of Amprolium on feed and water consumption, body weight gain and feed conversion ratio in pregnant rabbits are summarized in Table 4.

Table 4. Effects of Amprolium on feed and water consumption, body weight gain and Feed conversion ratio in pregnant rabbits.

Parameters	Doses of amprolium (mg / kg) Parameters			
	0 (Control)	32	69	136
Feed consumption (g)	4330.22±334.37	4230.40±310.20	4394.71±362.62	4500.42±272.90
Water consumption (l)	9445.26±343.76	8430.98±1585.76	9740.78±794.21	9489.57±657.99
Body weight gain (g)	588.00±178.52	398.25±521.24	489.25±199.36	407.75±130.73
Feed Conversion Ratio	8.28±1.24	12.70±6.74	10.60±4.71	10.60±3.01

It appears from Table 4 that the feed and water consumptions were higher in rabbits that received the highest dose of Amprolium (136 mg/kg) and lowest with those treated with the lowest dose of Amprolium (32 mg/kg). However, the difference was not significant (P>0.05) between treatments.

As for the weight gain, the highest value was obtained with the control group, while the lowest value was obtained with the lowest dose of Amprolium (32 mg/kg). However, no significant difference (P>0.05) was found between treatments. It is also clear from table 4 that the highest feed conversion ratio was recorded in rabbits exposed to dose 32 mg/kg body weight and lowest in rabbits of the control group. No significant difference was recorded (P>0.05).

Effects of Amprolium on some reproductive parameters in rabbit does

The effects of Amprolium on some reproductive parameters in rabbits are summarized in Table 5. From this table, it appears that the rate of female mated, the fertility rate and viability at birth were not significantly affected (p>0.05) by doses of Amprolium considered. The duration of gestation recorded in treated animals decreased with increasing doses of Amprolium. However, this decrease was significant (p<0.05) only in rabbits that received the highest dose (136 mg/kg) of the product as compared to those treated with 32 mg/kg (Table 5). The litter weight and size generally decreased in rabbits exposed to Amprolium compared to control animals; however, statistical analysis, reveals a significant difference only between the litter weight of the rabbits of control group and those of animals that received 32 and 69 mg/kg body weight. Milk production recorded in control rabbits and in those treated with the highest dose of Amprolium (136 mg/kg) were significantly higher than those of females that ingested 32 and 69 mg of Amprolium / kg body weight.

Table 5. Effect of Amprolium on some reproductive parameters in rabbits.

Parameters	Doses of Amprolium (mg / kg)			
	0 (control)	32	69	136
Rate of female mated (%)	100.00±0.00	100.00±0.00	100.00±0.00	100.00±0.00
Fertility rate (%)	100.00±0.00	100.00±0.00	100.00±0.00	100.00±0.00
Pregnancy duration (d)	31.50±0.58^{ab}	32.00±0.82b	31.75±0.96ab	30,50±1.00
Litter size	8.50±2.08	7.00±1.83	6.75±2.75	7.75±0.96
Pups viable at birth (%)	100.00±0.00	100.00±0.00	100.00±0.00	100.00±0.00
Litter weight (g)	447.00±66.95^{b}	349.75±38.29a	347.00±71.56a	392.75±44.35ab
Milk production (g)	1622.00±195.15^{b}	1234.00±168.14a	1227.60±161.23a	1625.20±153.44b

a,b: Values with the same letters in the same line are not significantly different (P>0.05).

As shown in Table 6, performances of young rabbits were not significantly affected by the considered doses of Amprolium. However, the sex ratio and viabilities at 7, 14 and 21 days were relatively higher (P> 0.05) in rabbits of control group. On the other hand, the weight gain of 3 weeks young rabbits increases in the non-significant manner (P>0.05) with Amprolium doses when considering only the treated groups.

Table 6. Effect of Amprolium on young rabbits reproduction parameters.

Parameters				
			69	136
Average weight at birth (g)	53.81 ± 8.06	51.70 ± 9.29	55.31 ± 13.77	51.05 ± 7.07
Sex ratio	0.92 ± 0.30	0.75 ± 0.46	0.60 ± 0.37	0.78 ± 0.16
Viability at birth (%)	100.00 ± 0.00	100.00 ± 0.00	100.00 ± 0.00	100.00 ± 0.00
Viability at 7 days (%)	100.00 ± 0.00	94.44 ± 11.11	97.50 ± 5.00	96.88 ± 6.25
Viability to 14 days (%)	100.00 ± 0.00	94.44 ± 11.11	97.50 ± 5.00	96.88 ± 6.25
Viability to 21 days (%)	100.00 ± 0.00	94.44 ± 11.11	97.50 ± 5.00	96.88 ± 6.25
Weight gain in 3 weeks (g)	193.73 ± 49.17	202.54 ± 59.17	219.50 ± 66.61	220.95 ± 12.35

Discussion

Whatever the physiological stage, feed consumption of rabbits was not significantly affected by different doses of Amprolium used, however we noticed a high appetite in rabbits treated with the highest dose of Amprolin (136 mg/kg). These results contradict those reported by Polin et al. [9] in laying chickens fed with diet containing amprolium (5.8 ppm) and comparable to those obtained by Mc Dougald and Mc Quistion [10] who noted an improved feed consumption in chickens treated with Lasalocid and Salinomycin compared to controls. Boka [11] also found that anticoccidial ionophores (90 ppm lasalocid sodium (15%), 100 ppm monesin (20%), 70 ppm of narasin (10%)) increase feed consumption of chickens treated compare to control. Indeed, as stated by Curasson [12], we can have a shelf or exacerbation of appetite, this in order to compensate for nutrient input deficits caused by intestinal lesions. The same trend was noted with water consumption; this is linked to the dry state of the feed that stimulates thirst after consumption. The body weight gain of non-pregnant rabbits was relatively higher

in animals receiving the highest dietary intakes. This result could be explained by the greater bioavailability of nutrients necessary for tissue formation. In pregnant rabbits, the body weight gain was higher in the control group. This can explain why the litter size and / or weight was higher in these rabbits. Regardless of the stage of development, the feed conversion ratio was better in rabbits of control group. Thus, Amprolium at the used doses reduced the use capabilities of feed.

The gestation length of rabbits remained in the range 28-32 days as provided for Djago and Kpodekon [3] and Tchoumboué and Téguia [2]; However, The value of this parameter recorded in treated animals appeared to decrease with increasing doses of amprolium; so therefore, young rabbits would grow faster as demonstrated by their growth after delivery. Indeed, the young rabbits from the treated females showed a higher weight gain than the rabbits of control group; this weight gain increased with the dose of amprolium. This could be explained by the fact that the amprolium would act on the hormones responsible for growth regulation, accelerate embryonic development and pup growth after delivery.

Amprolium induced a nonsignificant decrease in litter size of rabbits; also at delivery, the litter weights of all animals remained in the interval of 30-80 advocated by Lebas [13]; However, this parameter was significantly lower for animals treated with doses 32 mg/kg and 69 mg/kg compared to the control group. Milk production was lower in rabbit groups that presented the lowest litter and weight size, namely groups corresponding to smaller doses of amprolium used (32 mg/kg and 69 mg/kg). Indeed, milk production is dependent of the litter size [13].

The average birth weight, sex ratio and viability of young rabbits were not significantly influenced by different doses of amprolium used. This results are in agreement with those obtained by Onbasilar and Yalçin [14] which after inclusion of 1000 ppm of probiotic (Bioteksin™) and / or 66 ppm of anticoccidial agent (robenidine hydrochloride Cycostat® 66G) in the rabbits feed found no significant effect for these parameters.

Conclusion

After this study that examined the effects of Amprolium on some production and reproductive parameters of local breed rabbits, the following conclusions can be drawn:

- A high dose (136 mg / kg), Amprolium reduces the duration of gestation;
- Administered at doses 32 and 69 mg / kg, Amprolium leads to lower litter weight and milk production.

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