Effect Of R.D-Compound On Broiler Chick Performance And Residues Level Of Some Drugs

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ABSTRACT

The present study was conducted to throw the light on the effects of the concurrent use of diuretics (R-D compound, 1g/litre / day at 7th day of age to the end of the experiment) on residue concentrations of the antibiotic (ampicillin) and coccidiostate (vetacox s) as well as their effects on performance in broilers. A total of 180, one-day-old broiler chicks (Hubbard) were equally divided randomly into 6 groups , a control group and 5 experimental groups , each of thirty chicks. All chicks were fed a basal balanced diet. Chicks of the first group received non-medicated water and considered as control. The second group received R. D compound medicated water (1g/litre /day at 7th day of age to the end of the experiment). The third group received Vetacox S medicated water (1g/3litre/day at 30 day of age for 5 consecutive days). The fourth group received both Vetacox S + R. D compound medicated water at 30 days of age for 5 consecutive days. The fifth group received ampicillin medicated water (1g / litre at 30 day of age for 5 consecutive days). The sixth group received both ampicillin + R. D compound medicated water at the thirteen days of age for 5 consecutive days. Ampicillin and sulphadimidine were assayed in serum, liver, heart and breast muscles using microbiological method. Birds body weight were recorded weekly in Kg and Feed conversion ratios were calculated. Our results revealed that, the addition of the R. D- compound alone or together with either Vetacox S or ampicillin to the water elicited more significant increase in body weights and a lower feed conversion ratios than those in antibiotic (ampicillin) and coccidiostates (vetacox s) treated groups . The results suggest that R-D compound may be useful not only in improving production of broilers but also to enhance excretion of ampicillin, and vetacox s residues .

INTRODUCTION

Administration of drugs to food-producing animals in order to cure or prevent diseases or to promote growth, requires not only consideration of their effects on the animal but also on human who ingest food from these animals. This practice can result in unwanted drugs residues in meat and meat products eaten by humans. Negative health effects in humans have been traced to the consumption of these antimicrobial drugs and their metabolites *(1&2).* A number of possible adverse health effects of veterinary drug residues have been suggested. These include , allergic/toxic reactions to residues, chronic toxic effects occurring with prolonged exposure to low levels of antibiotics and development of antibiotic-resistant bacteria in treated animals. These bacteria might then cause difficult-to-treat human infections and disruption of normal human flora in the intestine. The bacteria that usually live in the intestine act as a barrier to prevent incoming pathogenic bacteria from getting established and causing disease. Antibiotics might reduce total numbers of these bacteria or selectively kill some important species *(3&4).*

Sulfonamides are one class of antimicrobial agent used widely in the livestock industry to promote growth. These drugs are often overused because they are inexpensive .

Sorbitol dehydrogenase; a novel target for adjunctive protection of ischemic myocardium is readily available. They are widely used in poultry production as anticoccidials and so there is some risk of residues in meat *(5-7).*

Penicillin derivatives (β-lactam antibiotics) are widely used in poultry to treat infections and as feed or drinking water additives to prevent some diseases *(8).* Ampicillin residues in kidney and liver (as

determined by HPLC) were higher than those in muscle *(9)*. Hence, withdrawal periods have been stipulated to each product. If followed meticulously, there should not be any danger of these chemicals to the consumers of poultry products. The use of chemicals and veterinary drugs in the production of poultry may leave residues that need to be minimized. As, most antimicrobials usually cleared from the blood via the kidneys and into the urine, minimizing their residues could be reached by enhancing their excretion by several approaches including diuretic *(10)*. Therefore decreasing of residues in animal tissue and meats has become a priority, in our study.

This study was conducted to investigate the effect of the concurrent use of diuretics (R-D compound) on residue concentrations of the antibiotic (ampicillin) and coccidiostate (vetacox s) to chickens.

MATERIALS AND METHODS

Drug:

1-Ampicillin (Ampicillin 20% , ® oral solution) was obtained from Arabic Campany for Drugs and Chemicals Industery. Each 100 g contains 20g Anhydrous Ampicillin . Its recommended therapeutic dose is 1g / litre drinking water per day for 5 consecutive days

2-Sulfadimidine sodium (Vetacox S®; Associated Pharmaceutical product Limited, Lagos, Nigeria), at 1g / 3litre of drinking water per day for 5 consecutive days.

3-Mineral, vitamin and amino acid supplements (R. D compound, S.C.P Vet Company -Sharkia- Egy). It is composed of Ammonium and potassium chloride, potassium citrate, Magnesium and Sodium Sulphate, Sorbitol, DL methionin,vitamin B6, B2, B1 and K3 (1g/litre / day at 7th day of age to the end of the experiment).

Birds:

One hundred eighty one-day-old broiler chicks (Hubbard) were equally divided into 6 groups, a control group and 5 experimental groups, each of thirty chicks. A description of experimental protocol is presented in Table 1.

Table 1. Experimental protocol

Group	Name of drug	Dose / litre of drinking water / day
1 control	0	non medicated water.
2	R.D -compound	1g/litre /day at 7th day of age to the end of the experiment
3	Vetacox S	1g/3litre/day at 30 day of age for 5 consecutive days,
4	Vetacox S + R. D compound	As in group 2+3
5	ampicillin	1g / litre at 30 day of age for 5 consecutive days
6	ampicillin + R. D compound	As in group 2+5

1-The control group (1) : received non medicated water.

2-The experimental group (2) : received R. D compound medicated water (1g/litre /day at 7th day of age to the end of the experiment)

3- The experimental group (3) : received Vetacox S medicated water (1g/3litre/day at 30 day of age for 5 consecutive days)

4- The experimental group (4) : received both Vetacox S + R. D compound medicated water at 30 days of age for 5 consecutive days.

5-The experimental group (5) : received ampicillin medicated water (1g / litre at 30 day of age for 5 consecutive days).

6-The experimental group (6) : received both ampicillin + R. D compound medicated water at the thirteen days of age for 5 consecutive days.

All chicks were fed *ad libitum* on broiler starter ration up to 28 days of age. Then the birds were fed on broiler finisher ration up to 42 days of age. The composition and chemical analysis of broilers starter and finisher ration were presented in Table,(2 & 3).

Birds body weight were recorded every week in each group /Kg . Feed conversion ratios were calculated ***(11).***

Five birds from each group were slaughtered at the end of the experiment (42 days of age). The blood samples were collected from wing vein without anticoagulant in a sterile wasserman tubes for separation of serum, which was stored at –20° C in a small sterile glass vial until used for determination of ampicillin and sulphadimidine residues. Samples of liver, heart and breast muscles were taken from sacrificed birds for residues determination. Ampicillin and sulphadimidine were assayed in serum, liver, heart and breast muscles using microbiological method described ***(12).***

The obtained data were analyzed statistically using an **ANOVA test *(13).***

Table 2. Composition of the experimental diets in Starter and finisher ration /100kg

Diet / Ingredient	Broiler starter basal Diet (%)	Broiler finisher basal Diet (%)
Ground yellow corn	60.4	65.5
Soy bean meal	24.6	21.5
Fish meal	10	
Corn gluten60%CP	0.0	6.0
Calcium carbonate	1.4	1.4
Calcium dibasic phosphate	1.4	1.4
*Vit.min.premix	0.2	0.2
Oil	2	4.0
Total	100	100

Table 3. Chemical analysis of the experimental diet in the starter stage

Chemical analysis	Broiler starter basal diet	Broiler finisher basal diet
ME.Kcal/kg	3005.98	3248.9
Crude Protien%	22.0326	18.127
Ether. Extract%	2.2992	2.811
Crude Fiber%	3.1508	3.024
Ca.%	1.2666	1.0255
Available phosphorous%	0.6167	0.2484
Lysine%	1.3257	0.8105
Methionine%	0.4562	0.3406
Methionine +Cystine%	0.7993	0.6664

Moisture, CP, EE was chemically analysed *(14).*
Calculated *(15).

RESULTS AND DISCUSSION

Table 4. Effect of RD compound 1g/litre /day at 7th day of age to the end of the experiment With or without, Vetacox S (1g/3litre/day at 30 day of age for 5day) and Ampicillin (1g / litre at 30 day of age for 5 consecutive days) on average body weight of broiler chickens (0 – 6 weeks) (n=5).

Age / Groups	Initial	Week (2)	Week (4)	Week (6)
1 (control)	43.2 ±0.31	243.42± 7.31^{b}	1021.83±13.20^{d}	2024.70±52.64^{d}
2(RD-compound)	43.5 ±1.72	272.27±5.91^{a}	1216.05±16.59^{a}	2300.33±39.23^{a}
3 (Vetacox S)	43.42±1.72	247.9±6.05^{b}	1069.50±21.77^{cd}	2120.33±38.05^{cd}
4 (Vet.+RD)	43.58±1.7	261.35±5.64^{ab}	1159.50±23.01^{ab}	2252.0±30.46^{ab}
5 (Ampicillin)	43.5±1.71	270.05±7.08^{a}	1101.50±45.90^{ab}	2171.0±35.93^{bc}
6 (Amp.+RD)	43.33±1.76	263.11±7.02^{ab}	1169.50±45.90^{ab}	2230.33±42.18^{abc}

Values are means ± S.E
Means within the same column with different superscripts are significantly different.

Table 5. Effect of RD compound (1g/litre /day at 7th day) of age to the end of the experiment. With or without, Vetacox S (1g/3litre/day at 30 day of age for 5day) and Ampicillin (1g / litre at 30 day of age for 5 consecutive days) on average body weight gain of broiler chickens (0 – 6 weeks) (n=5).

Age / Groups	(0-2) Week	Week (2-4)	Week (4-6)	Week (0-6)
1 (control)	200.22±7.35^{b}	778.41±13.21^{c}	1002.86±44.34	1981.50±52.58^{d}
2(RD-compound)	228.77±5.82^{a}	944.22±18.0^{a}	1083.83±44.32	2256.82±39.24^{a}
3 (Vetacox S)	204.47±6.10^{b}	821.60±23.57^{bc}	1050.83±46.55	2076.91±38.13^{cd}
4 (Vet.+RD)	217.76±5.61^{ab}	898.15±24.47^{ab}	1092.50±39.91	2208.41±30.41^{ab}
5 (Ampicillin)	226.55±7.10^{a}	899.45±47.94^{ab}	1060.83±52.82	2186.83±42.23^{abc}
6 (Amp.+RD)	219.78±7.03^{ab}	838.38±19.50^{bc}	1069.50±38.03	2127.66±35.82^{bc}

Values are means ± S.E
Means within the same column with different superscripts are significantly different.

Table 6. Effect of RD compound (1g/litre /day) at 7th day of age to the end of the experiment. With or without, Vetacox S (1g/3litre/day at 30 day of age for 5day) and Ampicillin (1g / litre at 30 day of age for 5 consecutive days) on feed conversion ratio of broiler chickens (0 – 6 weeks). n=5

Age / Groups	Week (2)	Week (4)	Week (6)	Week (0-6)
1 (control)	1.8	1.9	2.3	2.0
2(RD-compound)	1.6	1.7	2.1	1.8
3 (Vetacox S)	1.7	1.8	2.2	1.9
4 (Vet.+RD)	1.6	1.75	2.2	1.85
5 (Ampicillin)	1.6	1.75	2.2	1.92
6 (Amp.+RD)	1.6	1.7	2.2	1.83

Values are means ± S.E
Means within the same column with different superscripts are significantly different.

Table 7. Tissue concentration (µg/ml or g) of Vetacox S (1g/3litre/day at 30 day) of age for 5day) and Ampicillin (1g / litre) at 30 day of age for 5 consecutive days) With and without R.D –compound (1g/litre /day at 7th day of age to the end of the experiment).n=5

Drugs / Samples	Concentration (µg/ml or g)			
	Vetacox s	Vetacox s+RD-compound	Ampicillin	Ampicillin+RD-compound
Serum	5.69 ± 0.08^{a}	0.98 ± 0.04^{b}	1.58 ± 0.05^{c}	0.88 ± 0.09^{d}
Liver	2.14 ± 0.02^{a}	0.98 ± 0.04^{b}	0.75 ± 0.01^{c}	0.19 ± 0.02^{d}
Kidney	2.27 ± 0.07^{a}	0.97 ± 0.03^{b}	0.68 ± 0.01^{c}	0.17 ± 0.01^{d}
Muscle	1.56 ± 0.07^{a}	0.74 ± 0.05^{b}	0.61 ± 0.02^{b}	0.14 ± 0.05^{c}

Values are means ± S.E
Means within the same column with different superscripts are significantly different.

The addition of the R.D compound alone and together with either Vetacox S or ampicillin to the water resulted in highly significant ($P<0.001$) increase in body weights compared with the control value. These effects were found to be greater in response to R. D compound and R. D compound together with either Vetacox S or ampicillin than to Vetacox S or ampicillin alone (Table , 4).

Average body weight gain of broiler chickens (0 – 6 weeks) was significantly ($P<0.001$) increased in response to R.D compound (group 2), both Vetacox S + R. D compound (group 4), ampicillin + R. D compound (group 6), and ampicillin medicated water (group5), compared with the control value, (Table, 5)

The present study further resulted in improvements of Feed conversion ratios in groups received R-D compound (group 2, 4 and 6) by 1: 1.8, 1:1.85 and 1:1.83 respectively, versus 1:2, 1: 1.9 and 1: 1.92 for the control (group1) ,Vetacox S medicated water (group3) and ampicillin medicated water (group5) respectively at the end of the experimental period (42 days) (Table 6). Similar results were previously recorded *(16).* The authors reported an increase in the mean body weight in broilers in response to supplementing a 1% NH4 Cl diet with 0.5% NaHCO3 resulted in a synergistic weight gain. Ammonium chloride was utilized as the model hydrogen ion donor. Furthermore , It was demonstrated that, providing birds with supplemental hydrogen ions reduces the severity of alkalosis and significantly increases growth rate improving the productivity of birds. In addition, the authors reported that, the advantages of using the dietary additives (NH4 Cl with 0.5% NaHCO3) are numerous .

The present study further evaluate providing the poultry with drinking water containing new compound known as, R-D compound. To explain how this compound produced its effect, it is necessary firstly to recall its components and the benefits of each of them separately on health and performance. The R-D compound provides drinking water additional minerals, vitamins and amino acids (NH_4Cl, KCl, potassium citrate, Magnesium and Sodium Sulphate, Sorbitol, DL methionin, vitamins C, B_1 , B_2, B_6 and K_3 and lactose). NH_4 Cl, acting as acidifying diuretic, enhances aldosterone secretion, wash the kidney and enhanced toxins and drug residues excretion. In addition, NH_4 Cl decreased the intestinal pH, limiting the growth of several pathogens. Moreover, vitamins C, B_1 , B_2, B_6 and K_3 are essential nutrients of colonic mucosa, enhanced absorption of nutrients improving health and performances. Magnesium sulfate also increases water in the intestines, which may induce defecation. It is reported that,

magnesium is an important for many systems in the body especially the muscles and nerves *(17)*. Magnesium ions have been added to enhance the scrubbing process and modifiers have been added to provide better dewatering of the aqueous sludges, thereby partly improving efficiency. These results are previously supported *(18)*, the authors stated that, the addition of the electrolytes not only replenishes those depleted during various stresses, it also stimulates water consumption. When the results of these are added together (electrolytes and increased water consumption), high performance can be produced. Sodium and potassium ions plays a role in a number of processes in the body in conjunction with other electrolytes like chloride . Their main tasks are to maintain extracellular volume, adjust osmotic pressure, regulate the acid-base balance, form hydrochloric acid in the stomach, activate some enzymes (e.g. amylases) and form membrane potential, e.g. for nerve conduction and muscular excitation. Via the potassium-sodium pump sodium is also involved in the active transport of glucose to the cells, which is responsible for energy production required to all vital processes and performances.

Sorbitol (A polyhydric alcohol with about half the sweetness of sucrose) present in the R-D compound is known as sugar substitute often used in diet. It was formerly used as a diuretic and may still be used as a laxative. Sorbitol has non-stimulant laxative effect works by drawing water into the large intestine, thereby stimulating bowel movements. Furthermore, Sorbitol, helps the body rid itself of excess potassium ions in a hyperkalaemic state by eliminating it *(19)*.

DL-Methionine is an essential amino acid that aids in the body's detoxification process. Amino acids are the building blocks of protein in the body, they are essential for the synthesis of structural protein, enzymes and some hormones and neurotransmitters. Methionine is part of body proteins and is important for skin and coat condition, eye and heart health. At the metabolic level, methionine is a methyl (CH3) group donor through formation of the co-enzyme S-adenosylmethionine necessary for methyl group transfer. It is a sulfur donor and thereby an indicator of B-vitamin status. Methionine is also an initiator of protein synthesis. It serves as a precursor to other amino acids like cysteine which can then be converted into taurine. Methionine can also be converted into glutathione, an important physiological antioxidant *(20)*.

Vitamin C is a water-soluble vitamin needed for the growth and repair of tissues in all parts of the body. It is necessary to form collagen, an important protein used to make skin, scar tissue, tendons, ligaments, and blood vessels Vitamin C is essential for the healing of wounds, and for the repair and maintenance of cartilage, bones, *(20)*.

Vitamin B1, Thiamine, an anti-neuritic factor plays an essential part in animal metabolic processes, nerve membranes and in nerve conduction , in carbohydrate metabolism and involved in the conversion of pyruvate to acetyl CoA. Acetyl CoA is essential for synthesis of fatty acid and cholesterol and in energy production *(21)*.

Riboflavin is the part of the original vitamin B2 complex which stimulates growth. It is involved in the conversion of folate to its coenzymes, in the conversion of tryptophan to niacin, as a coenzyme component of the dehydrogenases in the first step in glucose metabolism. releases energy from protein, production of FMN and FAD, both of which are involved in redox reactions, fat and carbohydrate and aids in Beta oxidation in fat metabolism. Moreover, it is needed for the production of corticosteroids, erythropoiesis, luconeogenesis and thyroid enzyme regulation, beside the activation of vitamin B6 *(22)*.

Vitamin B6 (pyridoxine) is essential for the metabolism of carbohydrates, amino acids, and fats (lipids), as well as for normal nerve function and for the formation of red blood cells. It also helps in keeping the skin healthy . Vitamin B6 plays a central role in the metabolism of amino acids, the formation of the key of several neurotransmitters, and the production of serotonin, melatonin, and

dopamine. More specifically, it is involved in transamination *(23)*. It is also required in the synthesis of the neurotransmitters serotonin, norepinephrine, and histamine from tryptophan, tyrosine, and , P5P . It can be highly concentrated in the brain even when low levels exist in the blood. It is required for the formation of alpha aminolevulinic acid, a precursor of heme in hemoglobin. It Promotes release of glycogen from liver and muscle as glucose-1-phosphate. It is vital for the formation of sphyngolipids involved in the development of the myelin sheath surrounding nerve cells and it is involved with the synthesis of the intrinsic factor *(24)*.

Vitamin K controls the clotting mechanism of the blood because its action is directed at the precursor of prothrombin. Prothrombin is activated to form thrombin, an enzyme which, in turn, converts fibrinogen to fibrin *(25& 26)*.

Lactose is a carbohydrate and contributes approximately 4 calories per gram. Lactose does not contribute any other nutrients to the diet; however, it stimulates the intestinal absorption of calcium, independent of the presence of vitamin D. One of its most important functions of lactose is its utilization as a fermentation substrate. Lactic acid bacteria produce lactic acid from lactose, and because of their ability to metabolize lactose, they have a competitive advantage over many pathogenic and spoilage organisms *(27)*.

It is generally conceded that Antimicrobial drugs (Vetacox S or ampicillin) can also alter metabolism of certain compounds, such as proteins and minerals, and can reduce stress from subclinical effects of pathogens (a level of infection too low to produce noticeable symptoms). In either case, more energy contained in the feed is applied to growth and weight gain *(28& 29)*.

The data obtained showed that, the residues of ampicillin given to chicken attained a level of more than 1.58 ±0.05 µg/ml in plasma, 0.75 ±0.01, 0.68 ± 0.01 and 0.61 ± 0.02 µg/g in liver, kidney and muscles respectively at the end of the experiment. Whereas, sulphadimidine residue levels reached 5.69 ± 0.08 µg/ml in plasma , 2.14 ± 0.02 , 2.27 ±0.07 and 1.56 ± 0.07 µg/g in, liver, kidney and muscles respectively at the end of the experiment . It was found that, the levels of ampicillin residues have retreated with the concurrent use of diuretics (R-D compound) to 0.88 ±0.09 µg/ml in plasma, 0.19 ±0.02, 0.17 ± 0.01 and 0.14 ± 0.05 µg/g in liver, kidney and muscles respectively at the end of the experiment. Whereas, sulphadimidine residue levels were lowered to 1.91 ±0.09 µg/ml in plasma , 0.98 ± 0.04, 0.97 ± 0.03 and 0.74 ± 0.05 µg/g in, liver, kidney and muscles respectively with the concurrent use of diuretics (R-D compound) ,at the end of the experiment. Concentration of ampicillin and sulphadimidine residue levels in plasma and other tissues of control group were below the detection limit (Table, 7). These results corroborate the previously obtained *(30)*. The authors stated that, at day 40, the residues range 0f 0.61 -1.94 , 0.24- 2.25 and 1.3 -6.7 µg/ml or µg/g of ampicillin, oxytetracycline and sulphadimidine, respectively were found in plasma and tissues.

Conclusion : R-D compound supplement drinking water with electrolytes. Electrolytes are certain minerals that can be found in the blood and are important for normal cell function and growth. Electrolytes, as the name implies, help regulate nerve and muscle function by conducting electrical signals from nerves to muscles. Electrolytes are also important for the acid-base balance of the blood and fluid retention. Some of the electrolytes found in blood plasma include sodium (Na), potassium (K), calcium (Ca), magnesium (Mg), chlorine (Cl), bicarbonate (HCO3) and sulfate (SO4). Therefore, it is advisable to use R-D compound successfully as growth promoter for broilers . It is also recommended also to use R-D compound with antibiotics medication to reduce the dangers of their residues.

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الملخص العربى

تأثير مركب R.D على الاداء ومتبقيات بعض الادوية فى بدارى التسمين

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لقد اجريت هذه الدراسه لالقاء الضوء على الآثار المترتبة على الاستخدام المتزامن للمضادات الحيويه ومضادات الكوكسيديا مع مدرات البول .وذلك باضافة مركب R.D.compound لمياة الشرب في اليوم السابع من العمر وحتى نهاية التجربه. وتم دراسة تاثيرة على بقايا المضادات الحيويه (الامبسيلين) و ومضادات الكوكسيديا (الفيتاكوكس س) فضلا عن آثارها على الأداء في بدارى التسمين.

استخدم لهذا الغرض عدد ١٨٠ كتكوت عمر يوم واحد (هوبارد) موزعة بالتساوي على ٦ مجموعات بشكل عشوائي ، المجموعة التجريبيه و ٥ مجموعات اخرى، عددكلا منها ثلاثين. غذيت جميعها على نظام غذائي واحد متوازن . قدم مياة شرب خالية من اى اضافات لفراخ المجموعة الاولى(الضابطة).أما المجموعة الثانية تلقت مياة معالجة بمركب R.D.compound (في اليوم السابع من العمر الى نهاية التجربه).أما المجموعة الثالثة تلقيت العلاج باالفيتاكوكس س في اليوم ٣٠من العمر لمدة ٥ ايام متتالية). وقدم للمجموعة الرابعة R.D + vetacox على حد سواء في اليوم ٣٠ من العمر لمدة ٥ ايام متتالية. وأما المجموعة الخامسة تلقيت الامبسيلين فى مياه الشرب (في اليوم ٣٠ من العمر لمدة ٥ ايام متتالية). تلقيت المجموعة السادسه الامبسيلين + R.D.compound على حد سواء. تم قياس متبقيات المضادات الحيويه (الامبسيلين) و ومضادات الكوكسيديا (الفيتاكوكس س)فى المصل و الكبد والقلب والعضلات باستخدام اسلوب الاحياء المجهريه. كما تم حساب متوسط اوزان الطيور فى المجاميع وكذلك معدلات النمو وقد تم حساب نسب التحويل الغذائى لهم. وكشفت النتائج التي توصلنا اليها أن اضافة R.D.compound وحده أو مع أي من الامبسيلين او الفيتاكوكس س الى مياه الشرب تسبب فى زياده معنويةو ملحوظة في الأوزان وتحسن معنوى فى نسب التحويل الغذائى بالمقارنة بتلك التي سجلتها المضادات الحيويه ومضادات الكوكسيديا كلا على حدة. تشير النتائج الى ان اضافة مركب R.D يحسن الاداء فى بدارى التسمين وربما يكون من المفيد ليس فقط في تحسين الانتاج وانما ايضا لتعزيز طرح المتبقيات الدوائية من انسجة الدجاج مما ينعكس ايجابيا على صحة المستهلك