



RESEARCH PAPER

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Effects of increasing the levels of a commercial vitamin premix on zoo technical performances and certain blood biochemical values in broiler Chickens

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Abstract

The present study aims to determine the influence of increasing vitamin premix levels on zoo technical and some biochemical parameters in broilers of ISA15strain. The tested vitamin premix is a commercial product administered in drinking water by many poultry farmers of Constantine region (East Algeria), as part of routine vitamin supplementation. A total of one hundred seventy chicks were randomly assigned to three experimental groups. Chicktonic® was administered as follow: control group (CH), treated with chicktonic® with dosages and duration of treatment as applied by many farmers of the region; group (CH+41) treated with an increased dose of chicktonic® starting from day 41st; group (CH+13) treated with an increased dose of chicktonic® starting from day13th. In animals consuming increased dosages of chicktonic®, mortality rates were lower (0.1% and 0.14%) than the control group (0.33%). No significant differences were recorded between the three groups concerning: feed intake (FI); average daily weight gain (ADWG); performance index (PI); and carcass yield (CY). A significant decrease in body weight at slaughter (BWs) was recorded in birds consuming higher levels of vitamin premix before the grower period. While a significant increase in abdominal fat (AF) was recorded in animals consuming higher levels of vitamin premix during the finisher period. In both groups receiving a high dose of chicktonic®, cholesterol and triglyceride values were higher than in the control group.

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Introduction

Vitamins affect directly the physiology of the cell and thus the functional metabolism of the organism (Castaing *et al.*, 2003). In fact, the fatty acids and the vitamins (D, A, E) act directly on the regulation of the functioning of the white blood cells, these particles can bind to intracellular receptors or modify the production of intracellular messengers (Latshaw, 1991; Klasing, 1998).

Vitamins form a cohesive group of organic compounds that are required in the diet in small amount (micrograms or milligrams per day) for the maintenance of normal health (Bender, 2003). They allow a more effective use of nutrients, which justifies their use at optimal dosages, with levels higher than those used merely to prevent clinical deficiencies (Pérez and al, 2003). Studies have often showed that stressed birds require higher vitamin and mineral supply, cited in 2007 by Lagana, in its publication, he suggest that organic trace mineral supplementation should be used instead of vitamin-mineral supplementation (Lagana, 2007). Others Studies confirm that vitamins contribute to growth, as well as the development of immunity defences and decrease in functional disorders and myopathies (Leeson, 2007).

Furthermore, the basal diet of animals is often enriched by polyunsaturated fatty acids with high oxidative power (Lessire, 1995), therefore, to minimize oxidative effects of these diets, many studies emphasise on the importance of vitamin supplementation, especially vitamin E (Cinar and *et al.*, 2010) and vitamin C, which is used as an antistress (Kassim and Norziha, 1995).

Protocols and products used in vitamin supplementation are different from one study to another, also, many formulated diets contain excess quantities of vitamins A, D₃ (cholecalciferol) and calcium, these diets can become dangerous for the health of animals. Most of the time, farmers follow protocols proposed by their veterinarians, who usually choose available products and establish the dose and the duration of the treatment according to the conditions of rearing, and particularly the results expected by the breeders.

In this context, it is not unusual for some farmers to abuse of vitamin premixes by increasing the recommended dosages hoping to obtain better results. Castaing in 2003, has tested the influence of a high-level OVNtm (Optimum Vitamin Nutrition) vitamin complex on zootechnical performances in males standard chickens ISA 15, it achieved an increase in growth performance of 2,2% and a better food efficiency of 1,2%.

In this experimental study, the effect of increasing the dosage of a commercial vitamin premix tested, the vitamin levels were increased either starting from day 13th or day 41st. The effect of dosage increase was studied through zoo technical performances, carcass characteristics at and certain blood biochemical parameters in broiler chicks raised from 1 to 50 days of age.

Materials and methods

Birds and housing

All procedures used in our experiment were approved by the scientific council of the Institute of Veterinary Sciences (University of Constantine, Algeria) and are conform to international guidelines concerning animal care and use in research and teaching (NIH publications no 85-93 revised 1985). A total of one hundred fifty, 1-day-old male chicks of ISA15 strain were used. Birds were purchased from a commercial local hatchery. Upon their reception chicks were weighed and randomly assigned to the different experimental groups.

For the duration of the study (from day 1 to day 50); birds were housed in the experimental poultry house of the Institute of Veterinary Sciences, University of Constantine (East Algeria). The facility has a controlled environment regarding: ventilation, humidity, temperature, hygiene and sanitary requirements. In order to separate the different experimental groups, adequate separations are used to divide the pen into smaller pens. Standard management practices of commercial broiler production were applied. The chicks were vaccinated against gumboro disease and Newcastle disease at the appropriate ages.

Diets

Birds were provided standard starter (day1 to day15); grower (day16 to day 45) and finisher (day 46 to day 50) diets. Feed provided by a local producer, was based on: corn, soybeans, and wheat bran, without additional vitamins. Daily feed consumption per experimental group was calculated by measuring the quantities provided for feeding and the leftover quantities.

All diets had been formulated to cover appropriately, the birds' energy and protein requirements during the three periods of breeding. Tables 1 represent the energy and protein values of the distributed diets, as analysed by a specialized laboratory (CATALYSE LAB).

Table 1. Energy and protein composition of the distributed diets (CATALYSE LAB, 2014).

	Starter	Grower	Finisher	Analyticalmethod
Proteins	17,93%	17,95%	17,08%	NA 2709
Sugars	22,4%	26,16%	30,77%	NA 5918
Lipids	9,81%	11,4%	16,98%	NA 1933

Treatments

The used drugs are produced by INVESA (International industrial Veterinaria, S.A., Spain). Treatments were dissolved in the drinking water just before watering. All treatments were withdrawn, 24 hours before slaughter. The tested commercial vitamin

premix is Chicktonic®; it is a liquid form complex of hydro and fat-soluble vitamins, which contains also all the essential amino acids and several non-essential ones. Table 02 shows vitamin composition of chicktonic® compared with NRC suggested requirements (NRC, 1994) and White head (2002).

Table 2. vitamins intake in chicktonic® (ml/l of water) in comparison with NRC recommendations (mg/Kg feed) (N RC, 1994) and White head (2002).

Vitamins	VitA (UI)	VitB1 (g)	VitB6 (g)	VitB2 (g)	VitB12 (mg)	VitD3 (UI)	VitE (g)	VitK (mg)	biotine (mg)
Chicktonic®	2500	0,0035	0,002	0,004	0,01	500	0,004	0,25	0,00002
NRC et Whitehead	1500	0,0018	-	0,0036	0,01	200	0,01	0,5	0,15

As a standard management practice, all animal groups received also a commercial antis tress, Betamint®; which is a mentholated oral solution, used in drinking water to alleviate dehydration symptoms associated with heat stress, and to improve metabolism in poultry. This product contains vitamin C (90g/L), and other nutrients such as betaine, electrolytes, menthol and sweeteners.

Experimental design

Birds were randomly assigned to three groups:

- Group (CH) (30 chicks): birds of this group received as a vitamin supplementation chicktonic® with dosages and duration of treatment as applied by many farmers of the region; this group is considered as control.

-Group (CH+ 41) (70 chicks): the dose of chicktonic® in this group was increased to 2,5ml/l during the period covering day 41stuntil day 48th.

-Group (CH+13) (70 chicks) : dosage of chicktonic® was increased gradually over three periods starting from day13th.

Table 3 shows dosages and periods of treatment in the three experimental groups. All experimental groups received as well, Betamint® with the same dose: 1ml/l for a total of 27 non-consecutive days covering the period starting from day 1 to day 40; and 2ml/l for 7 consecutive days, during the period starting from day 41to day 48.

Table 3. Dosages and durations of treatments for each experimental group.

Experimental Group	Dosage of chicktonic®	period of administration	Total days of treatment.
(CH) control	1ml/l	day1 to day 38	19 days
(CH+41)	1ml/l	day1 to day 40	27 days
	2,5ml/l	day 41 to day 48	7 days
(CH+13)	1ml/l	day 1 to day 12	6 days
	2ml/l	day 13 to day 29	12 days
	2,5ml/l	day 30 to day 43	9 days
	3,5ml/l	day 44 to day 48	5 days

Zoo technical parameters

For each group, mortality rates were recorded as occurred. Birds, diets, and feed leftovers were weighted regularly to calculate the following performance parameters: Feed Conversion Ratio (FCR); Feed Intake (FI); Average Daily Weight Gain (ADWG); and Performance Index (PI).

At day 50th of age, 30% of the initial number of birds per group, were randomly selected for carcass yield, and relative abdominal fat weight assessments. The selected birds were first weighed to determine their live body weight at slaughter (BWs), than they were killed by cutting the jugular vein. The dead birds were defeathered and eviscerated, the obtained carcasses were immediately weighted to determine carcass yield (CY). Abdominal fat was removed, weighed and expressed as a percentage of the bird body weight at slaughter.

Biochemical parameters

At day 49th of age, five randomly selected birds from each group were slaughtered. Blood samples were collected early in the morning (8 am) from the jugular vein, using a sterilized syringe and heparinized tubes. Blood samples were immediately sent to the laboratory to separate the serum and analyse the following biochemical parameters: glycemia; total cholesterol; triglycerides; and alkaline phosphatases.

Statistical analysis

Data were analyzed using statistical software originating Pro 9.1 (Data analysis and graphing software). Results of zoo technical performances were compared using ANOVA (one way analysis of variance). Results of biochemical parameters were compared using Kruskal-Wallis test. Values of $p < 0.05$ were considered significant.

Results*Zoo technical performances*

In animals consuming increased dosages of chicktonic®, mortality rates were low: 0.1% for (CH+14) group and 0.14% for (CH+13) group, compared to 0,33% for the control group (CH).

After slaughtering the birds at the end of the experiment, no macroscopic abnormalities were recorded while inspecting their livers and kidneys. No significant differences were recorded between the three groups concerning: feed intake; (FI); average daily weight gain (ADWG); performance index (PI); and carcass yield (CY). The non-statistical changes recorded include: a decrease in ADWG, FI, and PI when vitamin dose is increased; and an increase of CY, with the decrease of vitamin doses; FCR decreased in (CH+14) group and increased in (CH+13) group. (Table 4)

Table 4. Zoo technical performances (Mean SD) of the three experimental groups (from day 1 to day 50).

Paramètre zootechnique	(CH) control	(CH+41)	(CH+13)
Mortality rates (%)	0,33	0,1	0,14
Daily feed intake (g)	145,48±115,93	106,08±73,10	93,68±67,29
Feed conversion ratio (FCR)	2,3±0,84	2,08±1,001	3,37±3,56
Average daily weight gain (g)	62,29±32,32	56,99±30,18	45,84±34,90
Body weight at slaughter (BWs) (g)	2787,3±254,45a	2607,5±220,74b	2316,5±215,24ab
Performance Index (PI)	263,09±133,20	358,24±313,25	253,06±218,40
Carcass	Weight	(1956g)	(1905g)
	yield (CY) (%)	70,83±9,43	72,65±4,47
Abdominal fat (AF)	Weight	(27g)	(33g)
	Percentage (%)	0,98±0,24a	1,28±0,37a

Results on the same row and with the same letter indicate that the differences are significant ($p < 0,05$). Concerning body weight at slaughter (BW_s), increasing the dose of vitamins, whether starting from day 41st or day 13th did not improve this parameter, the best BW_s was recorded in the control group. A significant decrease in BW_s was even recorded for the (CH+13) group compared with the control (CH) and (CH+41) groups. Regarding abdominal fat (AF), increasing chicktonic® dose starting from day 41st (CH+41 group) has significantly increased this parameter ($p < 0,05$); while increasing chicktonic®

dose starting from day 13th (CH+13 group) has significantly decreased this parameter ($p < 0,05$) compared to the control (CH) group.

Biochemical Profile

Increasing the dose of chicktonic® starting from day 13th, has led to an increase in cholesterol, triglycerides, and Alkaline phosphatase. The increase in the dose of vitamins tends to raise blood sugar level, but not statistically significant, The highest blood glucose level (2.54 g/l) was recorded in the control group.

Table 5. Biochemical parameters (Mean SD) of the three experimental groups.

	(CH) Control group	(CH- 41) group	(CH-13) group
Blood glucose (g/l)	2,54±0,55	2,34±0,21	2,52±0,15
Total cholesterol (g/l)	1,05±0,08 ^a	1,23±0,17 ^a	1,31±0,12 ^a
Triglycerides (g/l)	1,05±0,25	1,09±0,54	1,22±0,39
Alkaline phosphatase (IU / l)	1644,4±592,83	1387,6±497,170 ^a	2037,316,69 ^a

Results on the same row and with the same letter indicate that the differences are significant ($p < 0, 05$).

Discussion

Zoo technical performances

The doses tested in our study, are far from the overdose which is six times the control dose in the avian species. Therefore, the increase in the recommended daily intake of chicktonic® starting from day 13 or day 41, is not supposed to negatively affect the functioning of the organism, neither create liver or kidney problems. Hypervitaminosis A causes disorders in the development of the bones of birds, such as: osteodystrophy (proliferative-maturation zone), metaphyseal sclerosis, hyperostéodosis and decreases numbers of osteoclasts. Hypervitaminosis causes mineralisation of parenchyme including liver, kidneys and heart. It's also increases the serum calcium levels which may affect the cardiac and muscular function. Excessive Calcium can lead the skeletal abnormalities in developing chicks (Harrison, 1994).

As an overall finding, increasing the dose of our vitamin premix, gave satisfactory zoo technical performances in comparison with ISA15 strain standards as well as for those of R308 and Arboracres strains (Arzour *et al.*, 2015, Hubbard-classic, 2015, Hoffmann *et al.*, 2013).

Our performances are also satisfactory compared with studies on supplementation by probiotics (Idoui *et al.*, 2009).

Increasing the dose of chicktonic® from 1ml to 2.5ml/l starting from day 41st has no significant effect ($p > 0,05$) on average daily weight gain (ADWG), feed conversion ratio (FCR), daily feed intake (FI) and carcass yield. Many studies on vitamin supplementation in broilers had reported non-significant variations in these zoo technical parameters, regardless of the growth rate of the used strain: ISA15, Cobb 500, Ross 308 (Alahgholi *et al.*, 2014, Shlig, 2009; Sun *et al.*, 2008, Javello, 2007, Castaing *et al.*, 2003).

The non statistical changes recorded in our study include a decrease in ADWG, FI, and PI with the increase of vitamin dose; and an increase of CY, with the decrease of vitamin doses. FCR decreased in (CH+14) group and increased in (CH+13) group. While body weight at slaughter (BW_s), did not improve with increasing the dose of vitamins.

Many published studies using other vitamin products, had reported changes in these zoo technical parameters: Laganá *et al.* (2007) have reported a same result for the FCR. Joavello, (2007) obtained with the vitamin complex Volihot® used from day 1 to day 42 in Cobb 500 broiler strain, an increase in ADWG and a decrease in FCR and (BWs). While Castaing *et al.*, (2003) with the Ovn™ vitamin complex, obtained in the ISA15 broiler strain, an increase of ADWG and BWs and a decrease of FCR. In another study, there has been an improvement in ADWG and live body weight at slaughter by supplementation with 0.5g/Kg of vitamin C (Rajput *et al.*, 2009).

In our study, the carcass yield (72.65%) has improved by 1.82% compared to the control group (70.83%). Castaing *et al.*, (2003) obtained an improvement of (1.1%), while Javello, 2007 found that carcass yield decreased by 0.5%. Rajput *et al.*, (2009) obtained with a vitamins association (A: 8500 IU/l; C: 0.5g/Kg; E: 0.003g/Kg and levamisole 2mg/kg) a carcass yield of 76.78% against 59.11% in the control group without vitamins.

In our study, increasing the dose of chicktonic® lead to an increase in vitamin A (from 2500 IU /l to 6250 IU / l) and also in VitD₃ (500 IU /l to 1250 IU / l), these values ensure a good calcium fixation in the skeletal bones, hence the good physical appearance of the chickens in this experimental group. However, the excess of these vitamins promotes the increase of the calcium/phosphorus ratio which could lead to the decrease of the average daily weight gain (ADWG), feed conversion ratio (FCR), and Feed Intake (FI) values as shown in a study by (Magnin *et al.*, 2009).

In our study, abdominal fat (AF) is significantly higher ($p < 0.05$) when increasing vitamin levels; this increase reached 33g (yield 1.16%) in (CH+14) compared to the control group 27g (yield 0.98%). Conversely, in their study Perez *et al.*, (2003), when using a vitamin complex of optimum level OVN™ have obtained a decrease of (AF) in the Ross 308 strain, (41.71g (1.52%) against 47.14g (1.72%) in the control group.

Finally, knowing that chicktonic® contains amino acids such as D, L-methionine (5mg/ml of chicktonic) and L-lysine (2.5mg/ml of chicktonic®). Studies conducted by INRA (1992) showed that, the excess of amino acids leads to a decrease in lipogenesis as well as an increase of T₃. Similarly, excessive feeding leads to a reduction of lipogenesis as well as a growth slowdown (Larbier. M and Leclerq. B, 1992).

Biochemical parameters

In both groups receiving a high dose of chicktonic®, cholesterol and triglyceride values were higher than in the control group. Cholesterol increase was statistically significant ($p < 0.05$), going up from 1.05g/l for the control group (CH), to 1.23g/l for the (CH+14) group and 1.31g/l for the (CH+13) group.

According to different studies which has used different basal diets and broiler strains, total cholesterol values vary, from 0.7g/l for the Ross 308 strain, to 2.28g/l for Cobb 500 strain (Arzour, 2015, Ismail *et al.*, 2014, Tayeb *et al.*, 2012, Piotrowska *et al.*, 2011, Silva, 2007).

The alkaline phosphatases were significantly higher ($p < 0.05$) in the (CH+13) group, where chicktonic® dose was increased starting from day 13th (2037.3 IU/l) in comparison with the (CH+41) group where the chicktonic® dose was increased starting from day 41 (1387.6 IU/L). Normal values of alkaline phosphatases vary according to the strain and basal diets, ranging from 5103 IU/l (Silva, 2007), to 1218,70 IU/l (Murat *et al.*, 2001).

According to Zantop, (1997) cited by Khooshechin *et al.*, (2015), the increase in alkaline phosphatases is related to liver disease. The increase of chicktonic in (CH+13) group, before the grower period; could have caused a slight disorder in liver function but without causing signs of apparent disease.

In addition, the decrease in alkaline phosphatases when the dose of chicktonic® was increased starting from day 41 onwards, could be related to the increase in calcium intake (Kaya and Tuncer, 2009). The highest blood glucose level (2.54g/l) was recorded in the control group.

Several authors confirm that unlike mammals, the fast before slaughter has little or no effect on blood glucose. The recent reported blood glucose values in broiler chickens, show variation in basal glucose, the values given are 2,33g/l three days after hatching (Rideau and *et al.*, 2012). This value is almost identical to that recorded in the (CH-41) group. Blood glucose in this group is 2,34g/l, therefore an improvement in the value of blood glucose, when the dose of chicktonic was increased from j 41.

Conclusion

the vitamin premix tested in our study gave overall good zoo technical performances and blood parameters, with low mortality rates. Increasing the dose of the used treatment did not significantly affect feed intake; (FI); average daily weight gain (ADWG); performance index (PI); and carcass yield (CY). A significant decrease in body weight at slaughter BWs was recorded in birds consuming higher levels of vitamin premix before the grower period. While a significant increase in abdominal fat (AF) was recorded in animals consuming higher levels of vitamin premix during the finisher period. Changes in abdominal fat percentages appear to be related to plasma triglyceride levels rather than total cholesterol levels. Compared to the control group treated with doses as prescribed by many local veterinarians; increasing the dose of vitamin premix starting from day 41st gave better results concerning zoo technical performances : feed conversion ratio (FCR), Performance Index (PI), low mortality and Biochemical parameters: glycemia, cholesterol, triglycerides, and alkaline phosphatases.

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