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The effects of egg powder application in pre-starter diet on serum metabolites of male broiler chickens

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ABSTRACT

A completely randomized design was conducted to evaluate the effect of egg powder application in pre-starter diet (0-7 d of age) of male broiler chickens. 320 male broiler chickens (ROSS 308) consumed different levels of egg powder (0, 2, 4 and 6 percent of diet) in their pre-starter diet from hatch until 7 d of age. Each treatment had 4 replicates with 20 male broiler chickens. Experimental sampling was carried out in 7 and 42 d of age. Results showed that relative weight of liver was not affected by egg powder application in pre-starter diet ($p>0.05$), while, increase in egg powder inclusion level of pre-starter diet led to gall bladder weight increase in both 7 and 42 d of age ($p\leq0.05$). At 7 d of age, concentration of Ca ion reduced ($p\leq0.05$) in serum of chickens which consumed egg powder in their pre-starter diet. Experimental treatments, however, had no effect on serum Ca concentration at the end of experiment ($p>0.05$). Cholesterol and triglyceride concentration in serum of chickens, which consumed egg powder in their pre-starter diet rose at 7 d of age ($p\leq0.05$); the reverse trend was observed at 42 d of age though ($p\leq0.05$). As a result of egg powder application at both 7 and 42 d of age, Serum concentration of total protein increased ($p\leq0.05$) and malondialdehyde (MDA) decreased ($p\leq0.05$). Result presented in this study showed that application of egg powder in pre-starter diet of broiler chicken significantly affected bird's metabolism which is reflected by change in serum metabolites.

Key words: egg powder, pre-starter diet, broiler, blood metabolites.

INTRODUCTION

Intensive research on genetic, nutrition, and management of broiler chickens during the past years has allowed the poultry industry to produce heavier and healthier chickens in a more efficient manner and the age to market has been gradually reduced in broiler chickens. The early days after hatch are considered as an important period of a broiler's life which a lot of research have been conducted on it. Whereas, at this period of time the gastrointestinal system of young chickens has not fully developed [1] and chickens at this age are not capable of utilizing common feed ingredients. Also at this time meeting nutritional needs of young chickens is essential. The main reason for this high level of requirement is high levels of growth and development in gastrointestinal system and related organs [2]. Since the first week of post-hatch represents a more critical part of the broiler's productive life, and as chickens digestive capabilities are limited, special attention is needed to be given to the nutritional requirements of the chicken in order to maximize their performance.

Before hatch time, all nutritional requirements of embryo are obtained from egg reserves. Immediately after hatch, the gastrointestinal tract of the chick passes a process of development and maturation [3]. On late days of incubation, residual yolk is internalized into the abdominal cavity. The yolk is composed of approximately 35 to 40% lipids, mainly acylglycerides [2], and is considered as a temporary energy source until the chick has access to feed.

Yolk sac residue is a good source of maternal antibody and plays a critical role in enhancement of birds' immune system and health [4]. Absorption of yolk sac residue occurs via intestinal system and in the absence of feed, young chicken utilizes the most valuable maternal antibodies as a source of energy. Although the yolk is absorbed regardless of feed intake [5], the presence of feed enhances yolk absorption via the intestinal system and helps the immune system to respond the undesirable antigens [2]. So immediate access to feed is the most critical point in chicken production and it helps the birds to save and use maternal antibodies for immune system. After the immediate access to feed, quality of feed ingredient is in the second importance. Conventional feed ingredients like grains and proteins from plant origin have limited digestibility for chickens in early days post-hatch [6]. Because of young chicken's incapability in gastrointestinal system and also its high levels of requirements for growth and development, feed ingredients used in early days after hatch must be highly digestible.

To achieve this goal we need to use pre-starter diets. These pre-starter diets must provide highly digestible ingredients that young chicks be able to utilize them more efficiently. Nowadays, a lot of researches conducted to develop new alternatives to feed young chicks.

As egg reserves are the only resource to supply embryo requirements at incubation period, then early days after hatch, egg and egg by-products could be utilize by chickens, and can meet their needs. Egg by-products include out comes from breaking facilities and unsalable eggs. Sparks [7] reported that these by-products are rich in fat, maternal antibodies, protein and bioactive nutrients. According to FAO reports, world egg production was about 67.455 million tons during 2002 [8]. It is estimated that about 10% of world egg production is not salable; thus, there are considerable amounts of by-product which could be utilized in newly hatched chickens' diet. Egg by-product is a rich source of high quality protein, fatty acids and several nutrients such as folic acid, choline, iron, selenium and vitamins A, B, D, E and K, antioxidant like carotenoids, lutein and zeaxanthin, and they could be offered as an excellent nutrient source to chickens with expecting positive effects [9, 10]. Some studies showed that dried egg powder can be used as an alternative to antibiotics due to its high content of antimicrobial proteins and eggs antibodies and may thus fed to large flocks, without negative effects on performance of chickens [11, 12].

The aim of this study was to evaluate egg powder as an alternative ingredient in pre-starter diet of broiler chicken and its influence on blood metabolites of chickens.

MATERIALS AND METHODS

Birds, diets and experimental design

Three hundred and twenty, one-day old male chickens (Ross 308) were housed over 16 wired floor pens and were provided with continuous light. Chemical composition (crude protein, ether extract, crude fiber, phosphorous and calcium) of egg powder and other feed ingredients were analyzed using AOAC [13]. Egg powder prepared from egg white and egg yolk that spray dried at 55º C. At this temperature most of bioactive components of egg keep their natural structure and functions. The experiment had 4 treatments including 4 levels of egg powder inclusion in pre-starter diet of chickens. Inclusion levels of egg powder in pre-starter (0 - 7 day age) diet were 0, 2, 4 and 6 percent of diet. Each treatment had 4 replicates of 20 male chickens.

Experiment was extended until day 42. All experimental diets were formulated according to Ross 308 management manual [14] to meet the exact nutritional requirements (table 1). The experimental period was divided into four phases; pre-starter (from hatch to 7 d), starter (from day 8 to day 10), grower (11 to 24 d) and finisher period (25 to 42 d). Feed and water was *ad libitum* throughout the experiment. All experimental diets were formulated to be isonitrogenous and isocaloric.

Table 1. Composition and calculated nutrient content of diets fed in this experiment

Feed Ingredients (g/kg)	Pre-starter (0-7 d)*				Grower	Finisher
	control	2%	4%	6%	(11-24 d)	(25-42 d)
Corn	515.50	525.50	534.10	542.70	587.50	590.70
Egg powder	-	20.00	40.00	60.00	-	-
Corn Gluten Meal	50.00	50.00	50.00	50.00	50.00	50.00
Soybean Meal (42%)	353.80	333.20	312.80	292.40	283.00	279.30
Vegetable Oil	31.90	24.70	18.00	11.30	36.70	42.00
DL-Methionine	2.46	1.94	1.50	1.06	1.80	1.10
L-Lysine HCl	3.04	2.23	1.42	0.61	2.60	1.00
L-Threonine	0.60	-	-	-	0.30	-
Di-calcium Phosphate	20.80	20.30	19.90	19.40	18.4 0	16.80
CaCO3	13.00	13.30	13.70	14.10	10.80	10.50
Na Bi-carbonate	1.60	1.20	0.70	0.30	1.40	0. 70
Common Salt	2.30	2.60	2.90	3.10	2.50	3.00
[a]Vitamin and Mineral Premix	5.00	5.00	5.00	5.00	5.00	5.00
calculated nutrient content (as-fed basis)						
Crude Protein (%)	23.60	23.60	23.60	23.60	20.92	20.77
ME (Kcal/kg)	3025.00	3025.00	3025.00	3025.00	3150.00	3200.00
Lysine (%)	1.28	1.28	1.28	1.28	1.08	0.95
Methionine (%)	0.59	0.58	0.58	0.58	0.50	0.42
Met+Cys (%)	0.92	0.94	0.97	1.00	0.80	0.72
Threonine (%)	0.81	0.81	0.87	0.93	0.69	0.65
Calcium (%)	1.05	1.05	1.05	1.05	0.90	0.85
Available Phosphorus (%)	0.50	0.50	0.50	0.50	0.45	0.42

[a]Premix provided the following per kilogram of diet: 4.8 mg of retinol acetate, 100 µg of cholecalciferol, 20 mg of DL-α-tocopheryl acetate, 4.5 mg menadione sodium bisulphate, 4.1 mg thiamine hydrochloride, 9.5 mg of riboflavin, 15 mg of calcium-D-pantothenate, 45 mg of nicotinic acid, 9 mg of pyridoxine hydrochloride, 2.2 mg of folic acid, 0.25 mg biotin, 12 mg ascorbic acid, 550 mg of choline chloride, 80 mg of Zn, 30 mg of Fe, 100 mg of Mn, 20 mg of Cu, 0.4 mg of Co, 1.2 mg of I, 0.4 mg of Se.

** All experimental groups after day 7 fed the same starter diet until day 10 of age as control group consumed from beginning of experiment.*

Sampling:

At 7 and 42 d of age, four birds were randomly selected from each pen, and blood samples were obtained by cardiac puncture for subsequent determination of Ca in plasma, and cholesterol, triglyceride, malondialdehyde (MDA) and total protein in serum. Serum samples were analyzed by an automatic biochemical analyzer (RA-1000, Bayer Corp., Tarrytown, NY) using colorimetric methods, following the instructions of the manufacturer of the corresponding reagent kit (Pars Azmoon, Tehran, Iran). Plasma lipid peroxidation was estimated by spectrophotometric determination of TBARS and was expressed as nanomoles of MDA per milliliter of plasma [15].

After that these birds were slaughtered, and eviscerated. Subsequently, internal organs include liver and gall bladder were manually removed and individually weighed. Internal organ weights presented as percent of body weight.

Statistical analysis

Collected data were subjected to variance analysis using a completely randomized design (4 treatments and 4 replicates in each). Basic statistics and variance analysis were performed to test the significance between the treatments. To evaluate the differences between the experimental groups, significant means were further analyzed using Duncan's multiple range tests. Data were analyzed by ANOVA using PROC GLM [16]. In all cases, P-values ≤ 0.05 were considered significant.

RESULTS

Liver and gallbladder weight: Birds which consumed control diet had the lowest relative weight of gallbladder and by increasing the level of egg powder in pre-starter diet, relative weight of gall bladder increased ($p \leq 0.05$). This trend was observed in both 7 and 42 days of age. Liver weight was not affected by level of egg powder in pre-starter diet in both 7 and 42 d of ages ($p > 0.05$).

Table 2. Effect of consuming different levels of egg powder in pre-starter diet of male broiler chicken on relative weight (%) of liver and gall bladder

		Level of egg powder inclusion in pre-starter diet					
	Age (d)	control	2%	4%	6%	SE	P-value
Gall bladder	7	0.098[c]	0.116[bc]	0.136[ab]	0.149[a]	0.006	0.01
	42	0.043[c]	0.053[bc]	0.071[ab]	0.084[a]	0.005	0.02
Liver	7	3.344	3.532	3.609	3.736	0.103	0.64
	42	1.849	1.765	1.676	1.661	0.059	0.70

[ab] Means in the same rows with different letter are significantly different (p≤0.05).

Figure 1. Effect of egg powder application in pre-starter diet of male broiler chickens on plasma calcium (mg/dl).

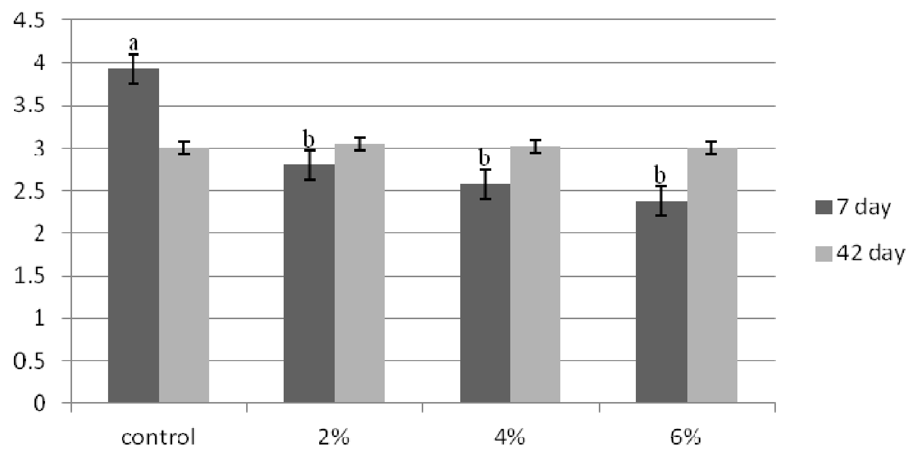
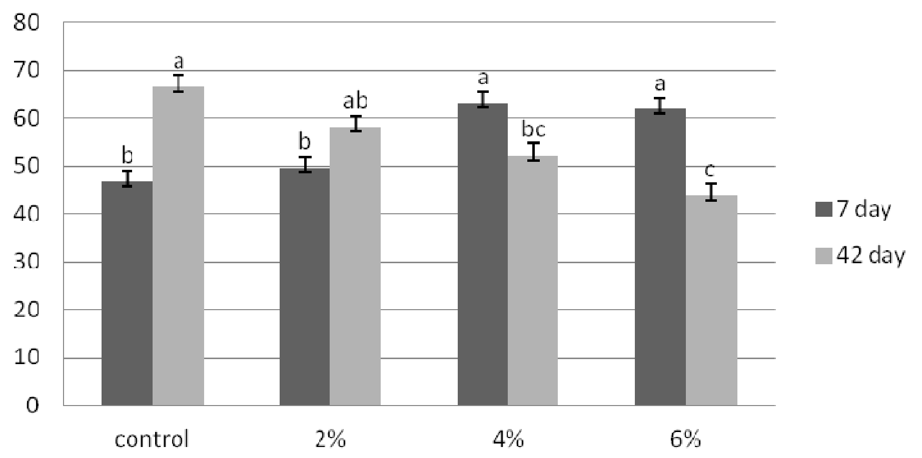


Figure 2. Effect of egg powder application in pre-starter diet of male broiler chickens on serum cholesterol (mg/dl).

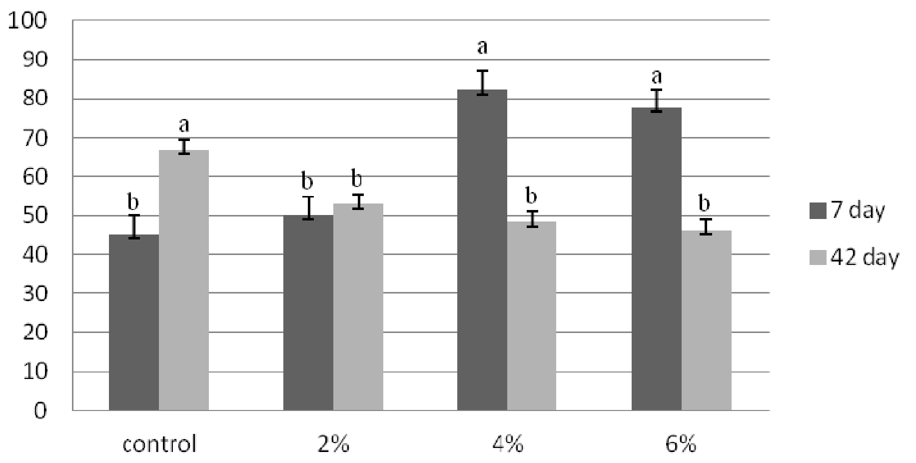


Blood metabolites: The effects of consuming egg powder in pre-starter diet of male broiler chickens on some blood metabolites are presented here. According to these results (figure 1), consumption of egg powder in birds pre-starter diet led to Ca concentration reduction in their plasma at 7 d of age (p≤0.05) However, all levels of egg powder application (2, 4 and 6 percent) in pre-starter diet resulted in the same concentration of Ca of blood. In 42 d of age, experimental diets had no effects on Ca concentration (p>0.05).

Cholesterol and triglyceride showed the same trend regarding egg powder inclusion in pre-starter diet. As presented in figure 2 and 3, application of egg powder in pre-starter diet increased both cholesterol and triglyceride level in 7 d of age ($p \leq 0.05$) and in 42 d of age the reverse trend was observed. In other words, in 42 d of age higher levels of egg powder resulted lower levels of cholesterol and triglyceride level in male broiler chickens plasma ($p \leq 0.05$).

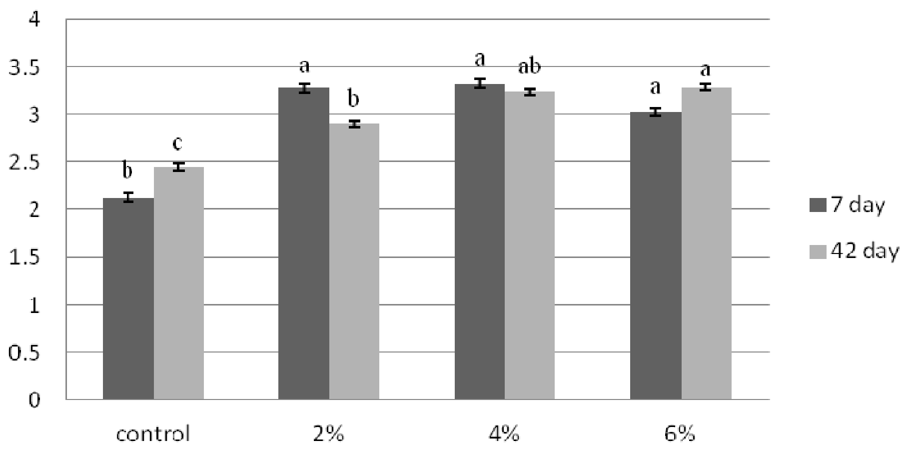
According to figure 3, application of 4 and 6 percent egg powder in pre-starter diet of chickens resulted the highest concentration of triglyceride at the age of 7 d (about 80 mg/dl) in 42 day of age a reverse trend was observed. So that both control group and experimental groups had the lower concentration of triglyceride in their serum.

Figure 3. Effect of egg powder application in pre-starter diet of male broiler chickens on serum triglyceride (mg/dl).



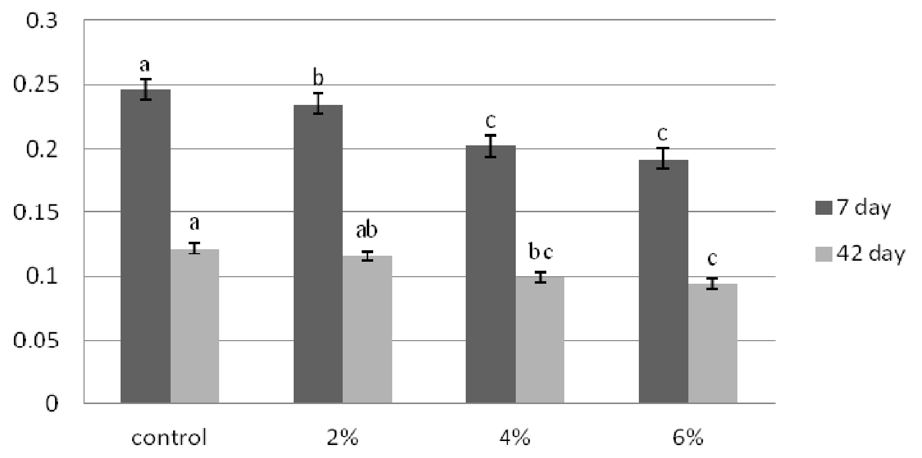
In case of plasma total protein, the birds which consumed higher levels of egg powder in their pre-starter diet, had higher levels of total protein ($p \leq 0.05$). This trend was observed in both times of sampling at 7 and 42 d of age (figure 4).

Figure 4. Effect of egg powder application in pre-starter diet of male broiler chickens on blood total protein (g/dl).



Malondialdehyde concentration in serum of male broilers significantly affected by consuming egg powder in pre-starter diet in both 7 and 42 d of age ($p \leq 0.05$). Birds that consumed higher levels of egg powder in their pre-starter diet had lower concentrations of MDA in their serum (figure 5).

Figure 5. Effect of egg powder application in pre-starter diet of male broiler chickens on serum MDA (nmol/dl).



DISCUSSION

According to the results presented in this study, application of egg powder in pre-starter diet of male broiler chickens profoundly affected blood metabolites and they showed that metabolism of birds was changed significantly as a result of consuming egg powder in pre-starter period.

Liver weight was not affected by experimental treatments; it might be due to high digestibility of egg powder that imposed no excess pressure to bird to utilize it. Although liver had to pack high levels of cholesterol and triglyceride existed in egg powder as lipoproteins, it didn't expend a lot of energy to overcome this matter because egg is one of the best protein sources and its chemical score is 100 [17]. Also egg yolk is a good source of lecithin which plays an important role in lipid metabolism [17]. Therefore, liver is not forced to experience excess pressure to synthesis new proteins and lecithin to pack high levels of cholesterol and triglyceride as lipoproteins. Consequently, the liver weight was not affected as a result of egg powder application.

In both times of measurements, due to consuming egg powder relative weight of gallbladder increased. According to studies [7, 2] egg yolk composed of approximately 40% lipid which is mainly acylglycerides. High levels of lipids in diet needs higher amount of bile salts and bile acids to be digested and utilized in intestinal tract of birds [18]. Also egg has high levels of cholesterol that is a precursor for bile salt synthesis. By synthesizing bile salts, gallbladder can reduce cholesterol concentration in blood. So bile role in blood cholesterol reduction and also in lipid utilization could explain the increase in gallbladder weight.

Blood calcium concentration reduced as a result of egg powder consumption in pre-starter diet of broiler chickens. This was only observed at 7 d of age and at the end of experiment (42 d of age).There was no difference among treatments in blood calcium concentration. High levels of lipids in egg powder [2] through the intestinal system of bird could interfere with minerals like calcium and suppress the absorption of these mineral ions. At the end of experiment in absence of egg powder, level of lipids especially free fatty acids, was low in intestinal system. So because the interference of lipids was limited, No difference was observed among the treatments.

Plasma concentration of cholesterol and triglyceride of chickens that consumed egg powder, in their pre-starter diet increased significantly at 7 d of age. The reverse trend was observed at 42 d of age and birds consumed egg powder at first week of age, showing lower levels of plasma concentration of cholesterol and triglyceride compared with control group. Egg yolk has high levels of cholesterol and triglyceride [7]; Therefore increase in plasma level of these metabolites after a week of egg powder consumption is expected. As showed in table 2, relative weight of gallbladder increased in chickens that consumed egg powder. Higher levels of cholesterol in plasma obliged the gallbladder to reduce plasma cholesterol by transforming it to bile salts. So increase in gallbladder relative weight was directly connected with its high activity. This high activity supports the metabolism of lipids which present in pre-starter diet of newly hatched chickens via producing bile salts and acids [18]. At the end of experiment and in the absence of lipid and cholesterol resource (egg powder) concentration of cholesterol and triglyceride in plasma of chickens from different treatments is expected to be equal. It is concluded that when an organism faces a challenge (high level of cholesterol and triglyceride in blood) its metabolism changes somehow to reduce the effect of the challenge. After the adaptation when the challenge or stressor is omitted, as a result of adaptation process, these organisms can behave more efficiently than those are in normal condition [19].

Total protein of plasma increased as a result of egg powder consumption in pre-starter diet of broilers. According to studies, blood total protein including albumin and globulin reflects the hepatic protein metabolism status in response to dietary treatments [20]. Egg is one of the best protein sources which contain high levels of albumin and globulin [2], with balanced amino acid profile. Then liver was not experienced any excess pressure to synthesize new proteins. In this study, liver weight was not affected by egg powder consumption. That is because of high quality of egg protein and also is a reasonable justification to increase in plasma total protein content of chickens which consumed egg powder.

Plasma level of MDA in birds consumed egg powder was reduced significantly. Malondialdehyde as a secondary oxidative product of lipid oxidation is produced in absence of antioxidants, also when the consumption of unsaturated fatty acids (like fatty acids exist in egg yolk) is high. Level of MDA in plasma represents an index that reflects antioxidant status of whole body. Egg contains components like carotenoids, lutein, zeaxanthin, vitamin E, as well as some pigments which have high antioxidant activity [9, 10]. Regardless of differences among treatments, MDA levels of serum at 7 d of age were about doubled compared with MDA levels at the end of experiment (figure 5). It was because of higher levels of unsaturated fatty acid consumption in pre-starter diet of birds via egg powder.

CONCLUSION

Results presented in this study indicated that metabolism of broiler chickens are significantly affected by application of egg powder in their pre-starter diet. Egg powder consumption in pre-starter diet affected most of plasma metabolites and this effect for some metabolites like MDA, cholesterol and triglyceride was positive. Regardless the changes in plasma metabolites concentration, the effect of these changes on bird’s performance, health and products quality should be surveyed.

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