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Effect of feed additive (Wefasan 2010) on growth performance and health status of chick

Hany Hefni* and Samy Zalat

Zoology Department, Faculty of Science, Suez Canal University, Ismailia, Egypt

ABSTRACT

This work aimed to evaluate the effect of feed additive (Wefasan 2010, an extract derived from palm lily) on growth performance and health status of chickens. Sixty 1-day-old chicks were divided equally into two experimental groups of similar mean weight. One group was fed the basal diet and the other a diet containing Wefasan 2010. After 30 days, Wefasan in the diet increased body weight gain by 15-29 %. No effect on morphometric parameters was noticed, except a significant increase in feather length. A mild adverse effect was found in the histological architecture of both liver and kidney. Protein electrophoretic patterns of liver, muscle and serum showed slight to mild changes when compared with chicks fed on the basal diet. Hematological and biochemical parameters were normal except aspartate aminotransferase (AST), which exceeded normal levels. Further investigations are needed before Wefasan 2010 can be recommended as a feed additive for chickens.

Keywords: Wefasan 2010, Growth Performance, Health Status of Chick

INTRODUCTION

Poultry health and live-production practices have traditionally been determined primarily by economic considerations (Brewer 1993). Today's poultry industry faces many challenges, including the acceptance and integration of biotechnology crops and feed additives in animal feed. While there are many opinions, facts support the conclusion that biotechnology crops and feed additives as well as new value-added crops developed through conventional breeding will benefit the poultry industry now and in the future (Coon 2004)

Wefasan 2010 is a feed additive used in stock breeding. It is produced from plant extracts (palm lilies) and therefore is a purely biological product. The active ingredients are saponines and steroid sapogenines, and it appears considerably to improve feed utilization and weight gain. This feed additive was recently introduced to the markets mainly of Egypt and some other Arab countries.

Since there is no previous work, in this study we aimed to evaluate the effect Wefasan 2010 on growth performance and health status of chicks under Egyptian conditions.

MATERIALS AND METHODS

One hundred 1-day-old chicks (commercial strain Lohmann brown) were divided equally into two groups. Group I was fed a basal diet (control), following the National Research Council (NRC, 1984). Wefasan 2010 extract (5.10%) was mixed with forage cereal (94.9%) to make up the diet of the experimental group, Group 2. The recommended dose is to add 330 g of Wefasan 2010 to 1 ton of standard diet. The experiment lasted 30 days.

The growth performance of the chicks was determined by measuring body weight and the lengths of the wing, feather, tail, peak, body and toes at 15 days intervals during

* Address for Correspondence

the experiment. The impact of the additive on anatomy and physiology was estimated as follows. Eight chicks from each group were scarified at day 30, and samples of liver and kidney of the control and treated groups were immediately removed, fixed in 10% neutral buffered formalin, dehydrated in ascending grades of ethanol, embedded in paraffin, sectioned at 5 µm thickness, and stained with haematoxylin and eosin (Bancroft & Stevens , 1990). Blood was collected directly from the heart at day 30 and then processed for haematological analysis (red blood cells, haemoglobin) according to Hall & Malia (1991). Serum from both groups of chicks at day 30 was calorimetrically analyzed for biochemical studies. Kits from Biomerieux lab reagents and products (France) were used for the determination of aspartate aminotransferase (AST) and aniline aminotransferase (ALT) (Reitmen & Frankle, 1957).

Preparations of samples (serum, muscle and liver) for SDS-PAGE protein electrophoresis analysis were done according to the procedure of Maeda *et al.* (1984). Three samples from each tissue were analyzed. A wide-range molecular weight prestained standard mixture (Bio-Rad) was used. Scanning of the gel was applied using Gel-Pro software (version. 3.0, Media Sci Image, USA, 1998).

All data were expressed as means ± S.E. Student's t-test implemented by SPSS 11 was used for assessment of significant differences between groups, with a level of significance of $p < 0.05$.

RESULTS

Table 1 shows the morphometric parameters measured in chicks from both control and treated groups. Wefasan 2010 resulted in significant increase in body weight gain ($p < 0.03$) at day 15 post-treatment, while a marginally significant difference was found at day 30 post-treatment ($p < 0.05$). There were no significant differences in the lengths of wing, tail, peak, body and toes, but there was in feather length ($p < 0.02$).

Wefasan 2010 caused mild (62.5% of examined chicks) to moderate (37.5%) histopathological changes in the kidney and liver tissues. In the liver, histopathological alteration was represented by haemorrhage, nuclear necrosis and pyknosis, and lymphocytic infiltration (Figs. 1-3). Noticeable bleeding was observed in the kidney, especially in the cortical region with a normal appearance of the glomerulus. Tubular degeneration was found in the cortex and medulla of treated kidneys. This degeneration included vaculation and nuclear pyknosis (Figs. 4-6).

Haematological analysis revealed that blood indices were within normal range (Table 2). The enzymatic activity of aspartate aminotransferase (AST) was high in the treated compared to the normal group, while the level of alanine aminotransferase (ALT) and cholesterol level were in the normal range (Table 2).

The electrophoretic patterns of control serum showed that there were 13 bands ranged from 7.8 to 155.5 kD, while 10 bands ranging from 27.5 to 155.5 kD were observed in the treated group. Eight bands disappeared (MW: 123.6, 106.6, 92, 74.6, 61.2, 25.3, 19.9 and 7.8 kD) while 5 new bands appeared (MW: 120.7, 108.6, 94.78, 78 .7 and 64.65 kD) in the treated group (Fig. 7).

The electrophoretic patterns of the muscles of control chicks showed 19 bands ranging from 6.03 to 267.9 kD, but the treated group showed 27, ranging from 6.03 to 387.3 kD. Four bands disappeared (MW: 267.9, 258.2, 66.5 and 32.8 kD) while 13 new bands appeared (MW: 387.03, 225.7, 150.7, 95.4, 70.3, 64.3, 39.1, 33.8, 28.8, 24.7, 23.3, 22.2 and 11.2 kD) (Fig. 7).

The electrophoretic patterns of control livers showed 23 bands ranging from 15.2 to 188.06 kD, while treated livers had 22, ranging from 11.8 to 172.9 kD. Eight bands

disappeared (MW: 188.06, 88.9, 80.8, 41.4, 38.7, 30.7, 27.5 and 15.22 kD) while 7 new bands appeared (MW: 172.9, 92, 82.7, 40.8, 20.9, 17.1 and 11.8 kD) (Fig. 7).

Table 1: Morphometric measurements at day 15 & 30 of group I fed basal diet and group II fed basal diet supplemented by feed additive.

Parameter	15 days old Chicks		30 days old Chicks	
	Group I standard diet	Group II Feed Additive	Group I standard diet	Group II Feed Additive
	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE
Weight (g)	60.8 ± 1.6	**70 ± 3.1**	70.0 ± 3.8	90.5 ± 5.9
Wing length (cm)	6.1 ± 0.2	8.8 ± 0.9	8.2 ± 0.4	11.4 ± 1.9
Feather length (cm)	3.9 ± 0.2	**5.3 ± 0.3**	5.2 ± 0.5	**10.0 ± 1.4**
Peak length (cm)	1. 6 ± 0.1	1.5 ± 0.1	1.9 ± 0.1	1.7 ± 0.2
Shift length (cm)	9.7 ± 0.3	9.4 ± 0.3	10.8 ± 0.4	10.1 ± 1.9
Body length (cm)	13.7 ± 0.6	15.1 ± 0.7	17.1 ± 0.9	17.3 ± 2.9
Tail length (cm)	2.2 ± 1.4	1.3 ± 0.18	3.1 ± 0.2	2.5 ±0.3
Skull length (cm)	3.9 ± 0.1	4.3 ±0.1	4.7 ± 0.2	4.5 ±0.5
Toe length (cm)	2.9 ± 0.2	3 ± 0.1	3.3 ± 0.2	3.8 ± 0.3

n= 20 at day 15; n=15 at day 30. Figures in bold are statistically different compared to the corresponding control group; $p < 0.05$.

Table 2: Hematological and biochemical parameters at day 30 of group I fed basal diet and group II fed basal diet supplemented by feed additive. (n=5)

Parameters	**Group I standard diet Mean ± SE**	**Group II Feed Additive Mean ± SE**	**Normal Range**
ALT	13.7 ± 6.4	11.7 ± 1.8	9.5-37.2 UL.
AST	133.3 ± 12.6	424.7 ±19.6	88 – 208 UL.
Cholesterol	143 ± 16.2	125.3 ± 3.7	52.0-148 mg/dl
RBCs x 10^6	2.08 ± 0.07	1.99 ± 0.09	1.5-4.5
HB	9.2 ± 0.51	8.8 ± 0.85	7- 13.1 g/dl

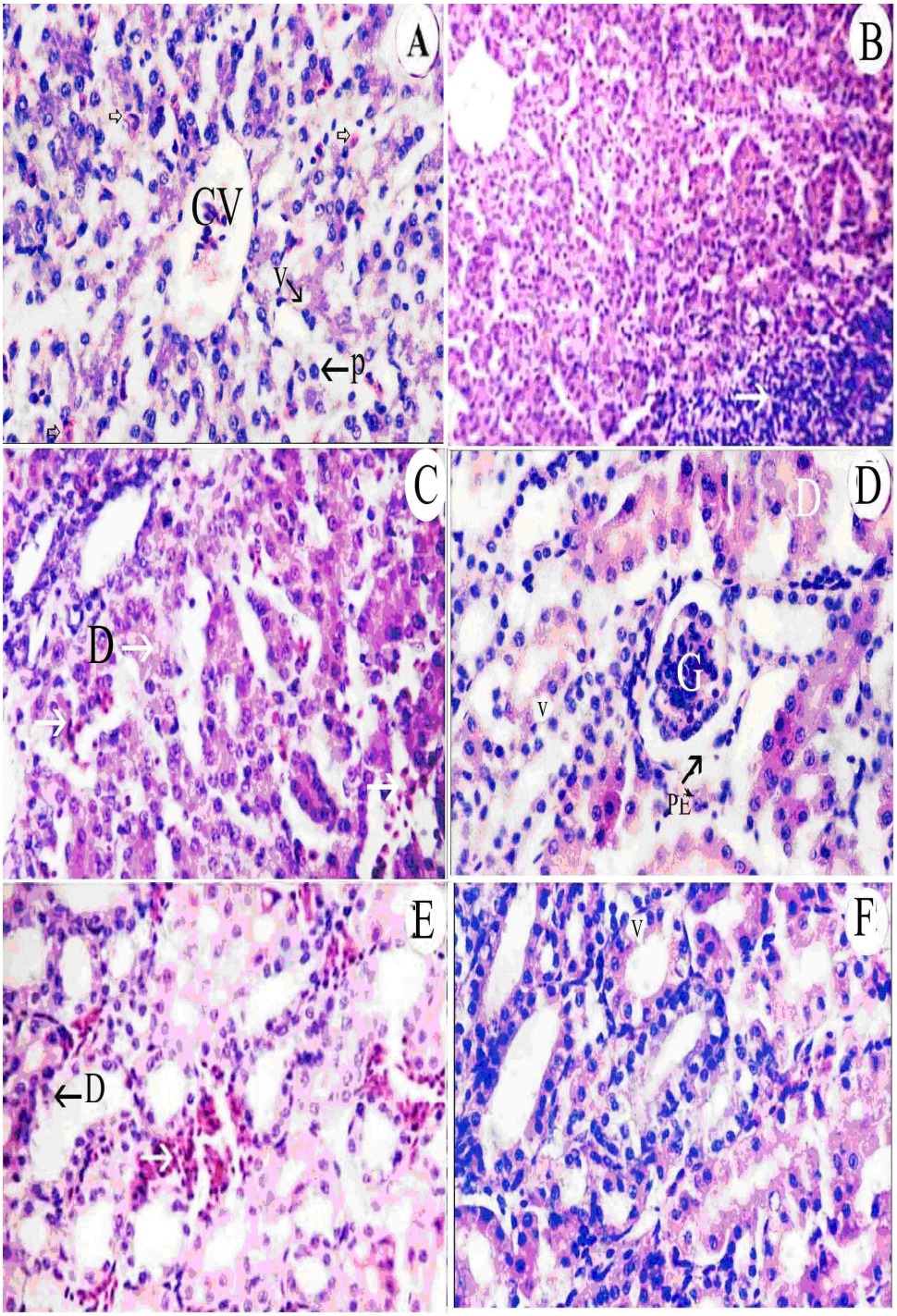

Fig.1: A, B & C: liver sections of 30-day old chicks fed the basal diet supplemented by feed additive, showing (A): pyknosis (p) hemorrhage (arrow), and vaculation (v). (B): lymphocytic infiltration (arrow). (C): Hepatocyte degeneration (D), haemorrhage (Arrow).
D, E & F: kidney sections of 30-day old chicks fed basal diet supplemented by feed additive showing (D): degeneration of cells of parietal layer of Bowmen capsule (PE), degeneration of some tubular cells of cortical layer (E) and vaculation (v), hemorrhage (arrow), (degeneration of some tubular cells of cortical layer (D). cv: central vein. G: glomerulus (F): degeneration (Arrow).

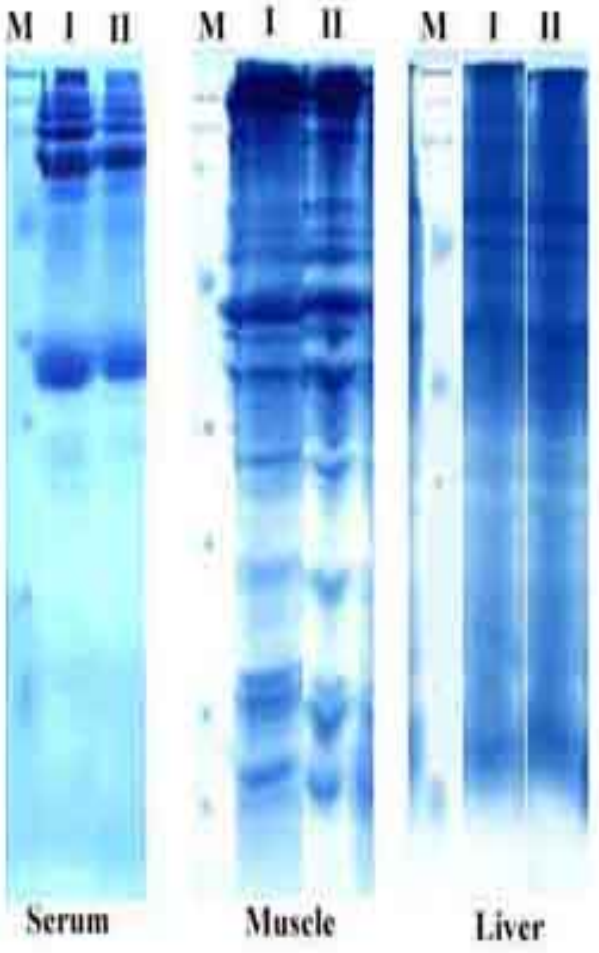

Fig. 2: Protein electrophoretic patterns of serum, muscle and liver of 30-day old chicks fed basal diet (I) and basal diet supplemented by feed additive (II).

DISCUSSION

This work aimed to evaluate the effect of Wefasan 2010 on the growth performance and health status of chicks. Wefasan in the diet increased body weight gain, probably by saving the energy used in the transformation of urea in the alimentary canal to ammonia. The active ingredients of Wefasan 2010 are saponins and steroid sapogenines, but tea saponin has been found to depress growth rates of chicks by decreasing feed intake (Ueda & Shigemizu 1998; Ueda *et al.* 2002). Other additives also are known to affect chick growth: a mixture of citric acid and sodium citrate improves growth in a dose-dependent manner (Boling *et al.* 2000), and nickel at the rate of 50 mg Ni/kg (Bersenyi *et al.* 2004) slightly improves weight gain and improves feed conversion efficiency.

Wefasan 2010 had moderate effects on the histological structure of both liver and kidney. Haraguchi *et al.* (2003) found degenerative changes in the striated skeletal muscle in the chest, myocardium and liver of chicks receiving high concentrations of *Senna occidentalis* seeds. Kim *et al.* (2003) found diffuse vacuolation of hepatocytes with individual cell necrosis in the liver of chicks treated by cyclophosphamide, with segmental necrosis of the lining

epithelial cells of the renal tubules, with eosinophilic material and necrotic detritus within the lumens. Santin *et al.* (2002) reported that ochratoxin A caused hepatocyte vacuolation and megalocytosis with accompanying hyperplasia of the biliary epithelium. Ramand & Sathyanesan (1987) found that liver histology of *Channa punctatus* exhibited various abnormalities, including hyperplasia, nuclear pyknosis, fatty necrosis and degeneration of hepatocytes leading to tumour and syncytium formation induced by mercurial fungicide.

Wefasan also affected the histological structure of the kidney of young broiler chickens. The histopathological changes included haemorrhage, vaculation and tubular cell necrosis. Huff *et al.* (1975) showed that the ochratoxin A gave the impression of oedema and some tubular necrosis in kidney sections, and pathological changes were observed at all dose levels. Santin et al (2002) found that kidneys treated by ochratoxin A showed hypertrophy of the renal proximal tubular epithelium, with thickening of the glomerular basement membrane. Biochemical analysis of serum revealed a significant increase in AST activity in the wefasan–treated group. Similarly, an increase in AST activity was recorded in chickens fed on a diet supplemented by 500 mg Ni/kg, soyabean meal, *Crotalaria pallida*, *Citrullus colocynthis* seed, or microbial phytase (Bersenyi *et al.* 2004, Aletor 1990, Diaz *et al.* 2003, Viveros *et al.* 2002, Bakhiet & Adam 1995). The increase in AST activity in the present study was correlated with histopathological alterations in the treated group, perhaps due to hepatocyte degeneration. This observation was confirmed histopathologically by necrosis of hepatic cells.

The present study suggests that the feed additive Wefasan 2010 increases the rate of protein synthesis, represented by the high intensity of some bands and the appearance of new ones. On the other hand, some protein bands disappeared in the Wefasan-treated group, and thus Wefasan may activate the umbiquitin degenerative system of protein. This effect of Wefasan 2010 may be attributed to the distraction of disulfide bonds (S-S) of inter and intra-chains of protein molecules (Guyton & Hall, 2000). The disrupted protein loses its function, leading to its candidature for the "umbiquitin degeneration" system; thus the intensity and number of bands decreases (Guyton & Hall, 2000). The reduction in the total protein content brought about by Wefasan may suggest disturbances in the functioning of the internal organs as a consequence of structural damage, which may lead to inhibition of protein synthesis and a general decrease in amino-acid concentrations. The increase in the number of bands (total protein) may be due to increasing γ globulin as a specific humeral immune response of chicks to Wefasan 2010. Several authors have also recorded a significant increase in total protein and γ globulin as a response to toxicants (Ezz-Eldin & Moussa 1998). Because of these detrimental effects, further investigations are needed before it is possible to recommend Wefasan 2010 as a safe feed additive.

REFERENCES

Aletor, VA (1990) Dietary fishmeal versus soyabean meal: an assessment of serum and liver enzyme response in the chicken. *Res. Vet. Sci.* 48(3): 267-270

Bakhiet AO, Adam SE. (1995) An estimation of *Citrullus colocynthis* toxicity for chicks. *Vet Hum Toxicol.* 37(4): 356-368

Bancroft JD & Stevens L (1990) Theory and practice of histology techniques. 3rd. Ed., Churchill Livingstone, London.

Bersenyi A, Fekete SG, Szilagyi M, Berta E, Zoldag L & Glavits R. (2004). Effects of nickel supply on the fattening performance and several biochemical parameters of broiler chickens and rabbits. *Acta Vet. Hung.* 52(2): 185-97

Boling SD, Webel DM, Mavromichalis I, Parsons CM & Baker DH (2000) The effects of citric acid on phytate-phosphorus utilization in young chicks and pigs. *J Anim Sci* 78(3): 682-9

Brewer RN (1993) Current concepts in broiler production. A publication of Alabama CooperativeExtension Service.

Coon C (2004) Poultry Feed and Biotechnology. A publication of University of Arkansas

Diaz GJ, Roldan LP & Cortes A (2003) Intoxication of *Crotalaria pallida* seeds to growing broiler chicks. *Vet Hum Toxicol.* 45(4): 187-199

Ezz-Eldin NM & Moussa WM (1998) Blood protozoa infecting *Claris lazera* in Lake Manzala with electrophoretic, haematological and biochemical studies on *Trypansoma mukasai* (Hoare, 1832). *Vet. Med. J. Giza* 46(4): 543-553

Guyton AC & Hall JE (2000) Textbook of Medical Physiology. WB Saunders, Philadelphia Pa. 10th edition, pp. 810-818

Hall R, & Malia RG (1991) Medical Laboratory Haematology 2nd ed. Butterworth Heinemann Ltd., Oxford.

Haraguchi M, Dagli ML, Raspantini PC & Gorniak SL (2003) The effects of low doses of *Senna occidentalis* seeds on broiler chickens. *Vet Res Commun.* 27(4): 321-8

Huff, WE, Wyatt RD & Hamilton PB (1975) Nephrotoxicity of dietary ochratoxin A in broiler chickens. *Appl. Microbiol.* 30(1): 48-51

Kim Y, Brown TP & Pantin-Jackwood MJ (2003) Lesions induced in broiler chickens by cyclophosphamide treatment. *Vet Hum Toxicol.* 45 (3): 121-233

Maeda S, Irie Y & Yasuraoka K (1984) Comparison of protein composition between schistosomula of *Schistosoma japonicum* and *Schistosoma mansoni*. *Parasitol.* 89: 453-459

Marron L, Bedford MR & McCracken KJ (2001) The effects of adding xylanase, vitamin C and copper sulphate to wheat-based diets on broiler performance. *Br Poult Sci.* 42(4): 493-500

National Research Council (1984) Nutrient requirement of chickens. 8th rev. ed. National Academy Press, Washington, DC.

Ramand RN & Sathyanesan AG (1987) Histopathological and biochemical changes in the liver of teleost fish, *Channa punctatus* (Bloch) induced by mercurial fungicide. *Environ. Poll.* 47: 135-145

Reitman S & Frankel S (1957) A colourimetric method for determinations of serum glutamic oxaloacetic and glutamic pyruvic transaminases. *Amer. J. Clinc. Pathol*. 28: 56-59

Santin E, Paulillo AC, Maiorka PC, Alessi AC, Krabbe EL & Maiorka A (2002) The effects of ochratoxin/aluminosilicate interaction on the tissues and humoral immune response of broilers. *Avian Pathol.* 31(1): 73-79

Ueda, H & Shigemizu G (1998) Effects of tea saponin, cholesterol and oils on the growth and feed passage rates in chicks. *Anim Sci & Technol* 69(1): 14-21

Ueda H, Takagi A, Katou K & Matsumoto S (2002) Feeding behavior in chicks fed tea saponin and quinine sulfate. *J. Poultr Sci* 39(1): 34-41

Viveros A, Brenes A, Arija I & Centeno C (2002) Effects of microbial phytase supplementation on mineral utilization and serum enzyme activities in broiler chicks fed different levels of phosphorus. *Poultr. Sci.* 81(8): 1172-1183

الملخص العربى

تاثير الاضا فات الغذائية (ويفاسان ٢٠١٠) على النمو والحالة الصحية للكتاكيت

هانئ أحمد حفنى و سامى محمد زلط

قسم علم الحيوان – كلية العلوم – جامعة قناة السويس – الإسماعيلية - مصر

يهدف البحث إلى تقييم تأثير الأضافات الغذائية الطبيعية المستخلصة من زيت النخيل (ويفاسان ٢٠١٠) على النمو والحالة الصحية للكتكوت .تم أختيار ستين كتكوتاً عمر يوم واحد ونفس الوزن وقسمت الكتاكيت الى مجموعتين كلاً منهما ثلاثون كتكوت احداهما تم تغذيته بالغذاء الاساسى والآخر بالغذاء الأساسى مضافاً إليه المستخلص الطبيعى ويفاسان ٢٠١٠. وتم تغذية المجموعتين لمدة ثلاثين يوما. وقد لوحظ أن تأثير المستخلص سبّب زيادة ملحوظة في الوزن بنسبة تصل الي (١٥– ٢٩%) وبالنسبة للشكل الظاهرى لوحظ زيادة في طول الريش.

تم دراسة التأثيرات النسيجية ووجد أن تأثير المستخلص على الكبد والكلية تمثلا في احتقان الأوعية الدموية وتحلل الأنوية وانتشار الخلايا الليمفاوية بين خلايا نسيج الكبد والكلية. وبدراسة التحليل البروتيني للكبد والعضلات والبلازما أوضحت الدراسة اختلاف طفيف في العينات التي تتغذى علي المستخلص عن العينات التي تتغذي علي الغذاء الاساسى .وبالنسبة لتحليل الدم فقد لوحظ ذيادة (AST) عن معدله الطبيعي. والخلاصة أن المستخلص فى حاجة لمزيد من الدراسة والتدقيق قبل التوصية بإستخدام المستخلص على نطاق واسع.