Effect of oestrogenic compounds on performance and biochemical parameters of chickens in Egypt

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Summary

Comparative studies of the effects of Nordette® and LutoFolone® on 15-day-old chickens were performed to determine their effects on growth performance, biochemical parameters and on hormonal residues in the liver and muscle. Sixty chickens were equally divided into three groups, with 20 chickens per group. Group 1 served as the control group. Groups 2 and 3 were treated daily with Nordette® (1 mg/kg body weight) mixed in the ration and LutoFolone® (0.5 mg/kg body weight) administered orally through a bent stainless steel feeding tube, respectively, for 30 days (from the 15th to the 45th day of age). The treated groups were left for a further period of 15 days without treatment. Blood samples were collected at 45 and 60 days of age and used for biochemical studies, while liver and muscles were excised from each chicken and used to prepare tissue homogenates for an estimation of hormonal residues (oestrogen and progesterone). Both drugs caused a gain in body weight. They also significantly increased (p<0.01) several serum variables, including alanine aminotransferase (ALT) (410% and 300%), aspartate aminotransferase (AST) (277.69% and 261.90%), cholesterol (16.91% and 17.19%), creatine kinase (CK) (72.47% and 27.46%), creatinine (62.22% and 42.22%) and uric acid (85.43% and 70.86%), and reduced total proteins (54.38% and 51.28%), albumin (60.38% and 52.08%) and globulin levels (50.22% and 49.36%) for Groups 2 and 3 respectively at 30 days post administration, in comparison with the control birds. Moreover, this study exhibited a significant increase in the levels of oestrogen residues in the liver (26.17% and 70.99%) and muscle (17.50% and 43.41%) for Groups 2 and 3, respectively. This indicated that the oestrogen level was much higher in the liver than in muscle in comparison to that of the controls. However, some of these findings showed insignificant changes 15 days after ceasing the administration of hormones. Data on the biochemical parameters and residue levels obtained from these results clearly indicate that anabolic agents in chickens may carry a specific risk to public health.

Keywords

Anabolic, Biochemical parameter, Chicken, Egypt, LutoFolone®, Nordette®, Oestrogen.

Effetto dei composti estrogenici su prestazione e parametri biochimici del pollame in Egitto

Riassunto

Studi comparati su Nordette® e LutoFolone® in esemplari di pollame di 15 giorni hanno permesso di valutare gli effetti dei due farmaci su crescita, parametri biochimici e residui ormonali in fegato e tessuto muscolare. Sessanta esemplari sono stati equamente divisi in tre gruppi di 20. Il Gruppo 1 è stato utilizzato come gruppo di controllo. I Gruppi 2 e 3 sono stati trattati su base giornaliera per 30 giorni (dal 15° al 45°giorno di vita) rispettivamente con Nordette® (1 mg/kg di peso corporeo) aggiunto al mangime e LutoFolone® (0,5 mg/kg di peso corporeo) per via orale,

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somministrati con sondino ricurvo in acciaio inox, e lasciati senza trattamento per altri 15 giorni. I campioni di sangue, raccolti a 45 e 60 giorni di vita, sono stati utilizzati per gli esami biochimici. I campioni di fegato e muscolo prelevati da ciascun esemplare sono stati impiegati per preparare gli omogenati tissutali per la stima dei residui ormonali (estrogeno e progesterone). Entrambi i farmaci hanno indotto un aumento ponderale. Rispetto al gruppo di controllo, a 30 giorni post-somministrazione i due farmaci hanno determinato aumenti significativi (p<0,01), rispettivamente nei Gruppi 2 e 3, di diversi parametri serici quali alanina aminotransferasi (ALT) (410% e 300%), aspartato aminotransferasi (AST) (277,69% e 261,90%), colesterolo (16,91% e 17,19%), creatinchinasi (CK) (72,47% e 27,46%), creatinina (62,22% e 42,22%) e acido urico (85,43% e 70,86%), e un calo delle proteine totali (54,38% e 51,28%), dell'albumina (60,38% e 52,08%) e dei livelli di globulina (50,22% e 49,36%). Lo studio ha rilevato un aumento significativo dei livelli di estrogeno residuo in fegato (26,17% e 70,99%) e tessuto muscolare (17,50% e 43,41%) rispettivamente nei Gruppi 2 e 3. Rispetto al gruppo di controlli è stato evidenziato un notevole aumento dei livelli di estrogeno nel fegato rispetto al tessuto muscolare. Tuttavia, alcune di queste variazioni sono risultate insignificanti 15 giorni dopo la sospensione del trattamento ormonale. I dati sui parametri biochimici e livelli dei residui hanno indicato chiaramente che la somministrazione di agenti anabolizzanti al pollame può comportare rischi per la Salute Pubblica.

Parole chiave

Anabolizzante, Egitto, Estrogeno, LutoFolone®, Nordette®, Parametro biochimico, Pollame.

Introduction

Chicken meat is considered one of the most desirable sources of animal protein for human consumption (15). This is one of the reasons behind the development of the poultry industry on both private and government farms that is designed to meet the increasing demand for protein. Yet some poultry producers deliberately ignore certain health and safety rules in the process of production and in an attempt to make the greatest profit in the shortest possible time. There are now several hormones and hormone-like agents that have a marked ability to improve the rate of growth and efficiency of feed intake for farm animals. The Joint (Food and Agriculture Organization [FAO]/World Health Organization [WHO]) Codex Alimentarius Commission, in particular its Codex Committee for Veterinary Drug Residues in Food, has approved the use of a number of natural and synthetic growth promoters for use in food-producing animals, including the endogenous growth promoters oestradiol-17 beta, progesterone and testosterone, and the synthetic growth promoters, trenbolone acetate and zeranol (46). This is done by using oestrogenic compounds as food additives to increase poultry production either in meat or eggs (2). It is well documented that injecting synthetic oestrogen results in hyperlipemia in birds (35, 59). El-Allawy *et al.* (13) proved that contraceptives produced electrolyte imbalances and an elevated uric acid level in serum. The administration of oestrogen to chickens tends to decrease the metabolism (4) and fattening effects may be attributed, in part, to these factors. The aims of this study were to study the growth performance (body weight and gain), liver and kidney functions, and to analyse the levels of anabolic agents (hormonal residues) in the liver and muscles in chickens that had previously received Nordette® and LutoFolone® anabolic agents.

Materials and methods

Chicken

Sixty day-old broiler chickens (Hubbard breed) were purchased from the Al-Noubaria poultry company in the Alexandria Province of Egypt. The birds were housed in proper hygienic conditions, maintained on a commercial well-balanced ration and had access to water *ad libitum*. All chickens were vaccinated against Newcastle disease, using the Hitchner B_1 vaccine in water at 7 and 21 days of age, and against Gumboro disease in drinking water at 15 days of age according to Giambrone and Clay (18).

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Drugs

Two hormonal compounds, Nordette® (0.15 mg levonorgestrol and 0.03 mg ethinyl oestradiol) and LutoFolone® (2 mg oestradiol benzoate and 20 mg progesterone) were used in this experiment. Nordette® was produced by the Nile Company for pharmaceuticals in Cairo under licence from the American Home Food Products, Inc., in New York. LutoFolone® was produced by the Misr Pharmaceutical Industries Company in Cairo.

Experimental design

At 15 days of age, the chicks were randomly divided into three groups (20 chickens per group). Group 1 was kept as the control, Group 2 was treated daily with Nordette® mixed in the rations (1 mg/kg body weight) and Group 3 was treated orally with LutoFolone® (0.5 mg/kg body weight) each day using a bent stainless steel feeding tube for 30 days (from the 15th till 45th day of age). The treated groups were left for another 15 days without treatment.

Evaluation of growth performance

Body weight

The birds were weighed individually at two weeks of age to obtain the average initial body weight. The body weight was then recorded weekly to calculate the average body weight developments in each group.

Body weight gain

The body weight gain was calculated every week (6).

Sampling

Blood sample

At the end of each experimental period, ten birds were selected at random from each group; they were weighed and sacrificed by severing the jugular vein and blood was allowed to flow freely into labelled tubes without anticoagulant for separation of serum. The serum was kept deep-frozen prior to biochemical analysis. Serum total proteins (TP) and albumin were determined in accordance with the methods described by Doumas *et al.* (10) and Drupt (11); the serum globulins were calculated as the difference between TP and albumin. Serum alanine aminotransferase (ALT) and aspartate aminotransferase (AST) activities were determined colorimetrically (43). Serum cholesterol (53), creatine kinase (CK) (16), creatinine (48) and uric acid (47) were quantified according to the methods described in the citation that follows each parameter.

Tissue sample

Liver and muscle were removed at the end of experimental periods (at 45 and 60 days of age) and perfused with normal saline (0.9% w/v) to reduce red blood cell contamination. Samples were homogenised in a dark place in 100 mM potassium phosphate buffer (pH 7.5) containing 0.15 M KCl to obtain 10% homogenate, using a motor-fitted homogeniser. The homogenates were centrifuged in an ordinary centrifuge at 5 500 rpm for 10 min. The pellets were discarded and the final supernatant was used for the estimation of hormonal residues (28). The tissue residues for oestradiol II (E2) and progesterone II (P2) were determined in accordance with the methods of Lichtenberg *et al.* (33) and Guillaume *et al.* (22), respectively. The estimation of hormones was performed exactly as indicated in the instructions by using test kits by Roche (Pleasanton, California) and the electrochemiluminescence immunoassay 'ECLIA' 2010 analyser.

Statistical analysis

Data were analysed using the one-way analysis of variance (ANOVA) (SPSS 11.0 for Windows) according to Tamhane and Dunlop (51). Significant differences found between the means were compared using Duncan's multiple range test (12).

Results

Body performance

The average body weight and gain increased significantly in Groups 2 and 3 at the 1st, 2nd, 3rd, 4th, 5th and 6th weeks, but there was no change in body gain at the 6th week in comparison to the controls (Group 1) (Tables I and II).

Table I
Average body weight (g)/chicken before and after administration of Nordette® at a dose of 1 mg/kg body weight (Group 2) and LutoFolone® at a dose of 0.5 mg/kg body weight (Group 3)

Group	At experimental initiation	No. of weeks Post-experimental initiation 1st week	2nd week	3rd week	4th week	5th week	6th week
1	238 ±2.5	353[c] ±12.21	480[c] ±20.06	624[c] ±8.71	793[c] ±14.54	955[c] ±16.66	1102[c] ±21.66
2	249 ±1.87	538[a] ±9.57	978[a] ±22.06	1360[a] ±10.12	1745[a] ±10.12	2007[a] ±26.01	2172[a] ±23.0
3	238 ±6.04	439[b] ±9.92	714[b] ±18.67	893[b] ±17.14	1100[b] ±17.15	1314[b] ±31.59	1461[b] ±29.55
Fisher test	NS	S	S	HS	S	S	S
LSD	–	32.75	62.58	38.67	57.21	78.61	76.96

Means in the same columns with different superscript letters are significantly different

NS non-significant difference
S significant at $p<0.05$
HS highly significant at $p<0.01$
LSD least significant difference

Table II
Average body weight gain (g)/chicken after administration of Nordette® at a dose of 1 mg/kg body weight (Group 2) and LutoFolone® at a dose of 0.5 mg/kg body weight (Group 3)

Group	No. of weeks 1st week	2nd week	3rd week	4th week	5th week	6th week
1	115[c] ±10.72	127[b] ±9.56	143[c] ±13.0	169[c] ±6.59	162[c] ±3.74	147 ±12.21
2	289[a] ±8.86	289[a] ±8.86	382[a] ±14.46	389[a] ±18.05	258[a] ±15.21	165 ±3.54
3	202[b] ±8.46	275[a] ±18.23	179[b] ±20.21	207[b] ±19.40	214[b] ±28.74	147 ±4.89
Fisher test	S	S	S	S	S	NS
LSD	28.96	39.88	26.89	28.59	28.23	–

Means in the same columns with different superscript letters are significantly different

S significant at $p<0.05$
NS non-significant difference
LSD least significant difference

Biochemical analysis

The serum TPs, albumin and globulin levels revealed a very significant decrease ($p<0.01$); however, the serum AST, ALT and CK activities, as well as cholesterol, creatinine and uric acid levels increased most significantly ($p<0.01$) in the Nordette® and LutoFolone® groups at 30 days post administration (Table III). At 15 days after stopping the administration of hormones (at 60 days of age), the serum proteinogram, creatinine and uric acid levels did not change significantly in any of the groups, while the serum enzymes and cholesterol kept increasing significantly (Table IV).

Hormonal residues in the liver and muscles

The administration of Nordette® and LutoFolone® for 30 days resulted in a highly significant increase in oestrogen residues in the liver and muscle tissue of chicken, while the progesterone level had no significant residues (Table V). The same results were seen 15 days after the cessation of drug administration, but at lower concentrations (Table VI).

Discussion

Female sex hormones, oestrogen and progesterone are commonly used in combination

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Table III
Serum biochemical parameters at 30 days post administration of Nordette® (Group 2) and LutoFolone® (Group 3) in chickens (45 days of age)

	Parameters								
Group	TP g/dl	Albumin g/dl	Globulin g/dl	AST units/l	ALT units/l	Cholesterol mg/dl	CK units/l	Creatinine mg/dl	Uric acid mg/dl
1	5.48$^{(a)}$ ±0.06	3.13$^{(a)}$ ±0.03	2.33$^{(a)}$ ±0.13	25.33$^{(b)}$ ±6.01	10.00$^{(c)}$ ±0.58	122.60$^{(b)}$ ±1.67	477.00$^{(c)}$ ±4.51	0.45$^{(c)}$ ±0.01	4.53$^{(b)}$ ±0.23
2	2.50$^{(b)}$ ±0.05	1.24$^{(c)}$ ±0.03	1.16$^{(b)}$ ±0.03	95.67$^{(a)}$ ±3.38	51.00$^{(a)}$ ±1.86	143.33$^{(a)}$ ±2.40	822.67$^{(a)}$ ±53.15	0.73$^{(a)}$ ±0.01	8.40$^{(a)}$ ±0.21
3	2.67$^{(b)}$ ±0.09	1.50$^{(b)}$ ±0.06	1.18$^{(b)}$ ±0.15	91.67$^{(a)}$ ±3.18	40.00$^{(b)}$ ±1.15	143.67$^{(a)}$ ±0.67	608.00$^{(b)}$ ±26.86	0.64$^{(b)}$ ±0.01	7.74 $^{(a)}$ ±0.29
Fisher test	HS	HS	S	HS	HS	S	HS	HS	HS
LSD	0.21	0.15	0.40	45.17	4.52	5.99	119.30	0.04	0.85

Means in the same columns with different superscript letters are significantly different

TP total protein
AST aspartate aminotransferase
ALT alanine aminotransferase
CK creatine kinase
HS highly significant at $p<0.01$
S significant at $p<0.05$
LSD least significant difference

Table IV
Serum biochemical parameters at 15 days post administration of Nordette® (Group 2) and LutoFolone® (Group 3) in chickens (60 days of age)

	Parameters								
Group	TP g/dl	Albumin g/dl	Globulin g/dl	AST units/l	ALT units/l	Cholesterol mg/dl	CK units/l	Creatinine mg/dl	Uric acid mg/dl
1	5.87 ±0.17	3.70 ±0.09	2.17 ±0.15	18.53^{b} ±0.29	12.00^{b} ± 1.50	117.00^{b} ± 5.00	421.00^{b} ±53.00	0.45 ±0.02	4.53 ±0.50
2	5.70 ±0.21	3.67 ±0.09	2.13 ±0.13	22.33$^{(a)}$ ±0.67	15.00$^{(a)}$ ± 2.30	138.00$^{(a)}$ ± 6.80	485.00$^{(a)}$ ± 64.19	0.50 ± 0.30	4.40 ±0.30
3	5.77 ± 0.17	3.53 ±0.15	2.14 ± 0.33	24.00$^{(a)}$ ± 0.29	15.40$^{(a)}$ ± 1.80	141.10$^{(a)}$ ± 10.33	488.00$^{(a)}$ ± 85.53	0.50 ±0.25	4.77 ±0.90
Fisher test	NS	NS	NS	S	S	S	S	NS	NS
LSD	–	–	–	2.6	1.7	15	61	–	–

Means in the same columns with different superscript letters are significantly different

TP total protein
AST aspartate aminotransferase
ALT alanine aminotransferase
CK creatine kinase
NS non-significant difference
S significant at $p<0.05$
LSD least significant difference

with most contraceptive preparations (20). These compounds are used on poultry farms as anabolic agents to improve body performance and, in turn, economic income (5, 17, 21, 24). Our study was conducted to illustrate the impact of selected hormones on the performance and biochemical parameters, and the residual levels of the hormones in liver and muscles of chickens.

The administration of Nordette® and LutoFolone® resulted in a significant increase in body performance (weight and gain) from the first to the fifth week, in comparison with the controls. These improvements in body weight observed throughout all periods were directly related to time. The increase in body weight and gain of treated chickens may be a reflection of increased feed intake, increased

Table V
Hormonal residues in the liver and muscle tissues at 30 days post administration of hormones in chickens

Group	Parameters			
	Oestradiol (pg/g tissue)		Progesterone (pg/g tissue)	
	Liver	Muscle	Liver	Muscle
1	612.43[(c)] ± 6.56	87.60[(c)] ±3.40	1.23 ±0.14	0.20 ±0.001
2	772.73[(b)] ±5.19	102.93[(b)] ±2.02	1.11 ±0.05	0.20 ±0.004
3	1047.20[(a)] ±26.19	125.63[(a)] ±3.45	1.20 ±0.13	0.21 ±0.004
Fisher test	S	S	NS	NS
LSD	54.95	10.49	–	–

Means in the same columns with different superscript letters are significantly different

S significant at $p<0.05$
NS non-significant difference
LSD least significant difference

Table VI
Hormonal residues in the liver and muscle tissues at 15 days after stopping the administration of hormones in chickens

Group	Parameters			
	Oestradiol (pg/g tissue))		Progesterone (pg/g tissue))	
	Liver	Muscle	Liver	Muscle
1	444.53[b] ±4.76	20.93[c] ±0.59	2.09 ±0.07	0.15 ±0.01
2	564.67[(a)] ±8.95	25.38[b] ±0.74	2.04 ±0.21	0.14 ±00.01
3	555.45[(a)] ±11.59	29.40[(a)] ±0.46	2.06 ±0.06	0.135 ±0.03
Fisher test	S	S	NS	NS
LSD	30.46	2.09	–	–

Means in the same columns with different superscript letters are significantly different

S significant at $p<0.05$
LSD least significant difference
NS non-significant difference

utilisation or due to the summation of anabolic properties and oedema between muscle fibres, as reported previously (17). Another suggestion for the increase in body weight is the increase in deposition of body fat as a result of the administration of oestrogenic compounds. This data is in agreement with previous reports (34, 35, 41, 44, 56). It is reported that oestrogen increases the deposition of abdominal and liver fat, with lesser increase in other organs. Previous reports (26, 38, 49) indicate that administering exogenous oestrogen dramatically increases body weight, fatty liver and abdominal fat. The authors attributed their results to the hypervolemia due to increased water retention by the kidney, in combination with the friability of the liver due to increased liver lipid. Moreover, Thayer *et al.* (52) reported that a slight excess in weight of the oestrogenated chicken is the result of increased abdominal fat and not actual growth and it also improves the grade of the carcass in broilers, roosters and cocks. An increased gain in live weight in diethylstilbestrol-treated chickens was found to be due primarily to increased fat production, energy consumption and, to a lesser extent, to preferential synthesis of fat at the expense of protein tissue (27). Under the influence of oestrogen, total tissue gain increased and was composed of greater

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fat gain and lower protein gain. Furthermore, Nesheim (40) stated that oestrogens markedly stimulate food intake and, at times, weight gain as well, and that there is a marked increase in weight gain among chickens fed oestrogenic compounds.

In regard to the biochemical studies conducted during our survey, severe changes in the hepatic and renal function tests were observed. The proteinogram of treated chickens revealed hypoproteinemia, hypoalbuminemia and hypoglobulinemia at 30 days post administration. Our results may be attributed to disturbances of metabolism by the liver, in addition to the cumulative effect of hormones on the kidneys which leads to proteinuria (albuminuria). The hypoproteinemia and hypoalbuminemia may be caused by long-standing albuminuria due to glomerulonephropathy or hepatic damage due to the administration of hormones (23, 24). Moreover, Nesheim (40) stated that there seems to be a particular stimulation of lipogenesis of oestrogenic compounds to the extent that protein synthesis is depressed. Morgan (39) recorded a significant decrease in the plasma albumin in two non-laying hens injected subcutaneously with 17β-oestradiol dissolved in propylene glycol (20 mg/kg body weight) at two doses with three-day intervals. Very similar results were obtained by other authors (57) in roosters given oestrogen (0.005 mg/kg body weight) daily for 12 days and in chickens that received a combination of oestrogen and progesterone for 35 days (37). It has been suggested that the decrease in hepatic albumin synthesis after hormone treatment is due to an oestrogen-mediated decrease in the content of albumin mRNA.

Our study revealed a significant increase in the serum AST and ALT activities at 30 days post administration of Nordette® or LutoFolone®. The increased activities have been associated with hepatocellular damage (7, 9). The elevation of enzymes concurs with results of other workers (13) who attributed this increase to hepatocellular damage induced by the oestrogen and progesterone combination. This explanation was confirmed by Ganong (14) who reported an increase in the serum activity of transaminases after hormonal therapy (oestrogen and progesterone). The increase in ALT activity is a result of intrahepatic cholestasis by oestrogenic compounds administered for long periods. This leads to an alteration in the cell membrane permeability and enables the escape of the enzyme into serum at abnormally high levels (3). An evaluation of enzymes provides a clue to liver cell disorders reported earlier (1). On the other hand, the results of several studies have described the appearance of hepatocellular carcinoma in women using oral contraceptive steroids for prolonged periods of time (25). Although stopping the administration of hormones, the enzyme activities remained significantly increased and indicated liver damage.

The serum total cholesterol level increased with the administration of Nordette® or LutoFolone®. Such an increase may be attributed to the hepatic cholestasis induced by oestrogenic compounds which leads to regurgitation of all cholesterol contents to the blood, resulting in hypercholesterolemia. This result revealed that the oestrogenic compounds were hepatotoxic as demonstrated in chickens in some reports (17, 49). In addition, hyperlipemia (19, 26) with increased serum triglycerides and cholesterol (42) was produced by oestrogen. On the contrary, previous observations (32) reported that the fall in total plasma cholesterol was attributable to a marked increase in the number of low-density lipoprotein (LDL) receptors in the liver, with consequent rapid clearance of lipoproteins from plasma. Other authors (36) reported that a daily subcutaneous administration of 17β-ethinyl oestradiol in a dose of 5 mg/kg to rabbits for 10 days resulted in a significant decrease in the plasma cholesterol level in comparison to that of controls. The difference may be attributed to the dose, species, treatment, duration and route of administration of sex steroids. Although the preparation used in this study may be considered oestrogen-dominant, there could be some synergic interaction between the components of the oestrogen and progesterone combination. For this reason, the combination

of oestrogen and progesterone used over extended periods of time caused an increase in total cholesterol levels. This observation concurs with previous reports (55).

The increased serum CK in treated chickens may be attributed to muscle damage and other tissue damage which leads to an alteration in cell membrane permeability and enables the escape of the enzyme in the serum in abnormal high amounts (3). The serum CK activity is not elevated unless the material is highly irritating (9) and this was in line with the dangerous effect of oestrogens on the muscles. These results were in accordance with other authors (8, 24, 37).

Evaluation of the renal function in chickens treated with Nordette® or LutoFolone® revealed significant changes only at 45 days of age but insignificant changes at 60 days. In our study, the serum creatinine and uric acid levels were significantly increased in comparison to controls, but the increase was higher with Nordette®- than with LutoFolone®-treated chicken. Increased serum values of creatinine and uric acid were usually associated with increased oestrogen levels in the blood (55). The prominent rise of creatinine and uric acid in serum without efficient excretion provides further confirmation regarding the harmful effects on the kidney of repeated injections of both oestrogens and progesterone hormones (8). All changes in the renal parameters associated with renal damage were most probably due to salt and water retention accompanying the administration of oestrogenic compounds. Hyperuricemia may be due to either a decrease in the rate of tubular excretion accompanying the renal damage or to an increase in uric acid production due to the catabolism of body tissues (9, 13, 54). The combined use of two hormones enhanced hepatic and renal disturbances (13, 58). At 15 days after the administration of drugs, a slight improvement in kidney function was observed.

In regard to the evaluation of hormonal residues in the tissues of the chicken, our study revealed a significant increase in the concentration of oestrogen in the liver and muscles at 30 days post administration of both Nordette® or LutoFolone®. Oestrogen residues in the liver samples were higher than that in muscle. Similar findings have been obtained by other workers (2, 45). However, insignificant changes were observed in progesterone residues in the liver and muscle tissues. This indicates that the oestrogen was dominant and that it affected the tissues significantly. Similar findings and comments have been reported by others (30, 31, 50). In regard to the determination of oestrogen and progesterone in the liver and muscles post ceasing the administration of Nordette® or LutoFolone® for 15 days, the same results were obtained but with lower levels than those observed at 30 days post administration. This remaining level of oestrogen in tissue is considered to be a risk factor that entails a special risk to the animals (45) and probably also to the consumer. It is worth noting that some countries strictly prohibit the application of sex hormones-like activities as anabolic agents and, in particular, the use of oestrogens (28, 29).

Conclusion

It can be concluded that both Nordette® and LutoFolone®, each containing oestrogen and progesterone, improved body performance, leaving hormonal residues in tissues and altered liver and kidney functions. Therefore, the use of these hormones in chickens as anabolic agents by some poultry growers creates a special risk to chickens and probably to the consumer. It is recommended that the laws and regulations concerning the use of these anabolic agents, particularly oestrogen in chickens, should be modified to prohibit their use.

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