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Effect of adding soy lecithin, xylanase and their combination to low energy diet on carcass traits, meat quality, and some blood parameters of broiler chicks.

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Abstract

This study investigated the synergism relationship between lecithin (L) and xylanase (Xyl.) enzyme when applied to low dietary metabolizable energy (ME) on broilers carcass, meat quality and some blood parameters. 160 chicks randomly were allocated to 5 treatments, each of 32 birds (4 replicates of 8 birds each). The 1st group; chicks were fed basal diet without studied supplementation (control). In 2nd group, chicks were fed low ME diet (-150Kcal / kg lower than **NRC,1994** recommendation) and labeled as NC. In 3rd, 4th and 5th groups, chicks were fed NC supplied with 0.5g lecithin (NC + 0.5g L), 0.2g xylanase (NC + 0.2g Xyl) and 0.5g lecithin + 0.2g xylanase (NC +0.5g L + 0.2g Xyl), respectively. The growth trail lasted 35 days. The obtained results show that group of NC + 0.2g Xyl recorded significant increase in carcass %. The lowest meat total saturated fatty acids, total cholesterol, LDL and triglyceride were achieved for group of NC + 0.5g lecithin. Moreover, both NC + 0.5g L and NC +0.5g L + 0.2g Xyl recorded a significant decrease in plasma total cholesterol, triglycerides and LDL also, increase in plasma HDL compared to control.

In conclusion, addition of 0.5g lecithin alone or in a combination with 0.2g xylanase to broilers fed low-energy diet is more effective in carcass, meat quality and blood plasma lipid profile.

Keywords: Broilers, meat quality, soy lecithin, xylanase, carcass traits.

Introduction

Oils are widely supplemented in poultry feed to meet the high energy demand of fast growing broilers **(Patterson *et al.,* 2012)**. An increase in the fat content of the diet had increase the energy value of food, but may also alter the blood lipid profile, depending on the type of fat used in broiler diets **(Dubey, 2009).** Hence increase broilers meat total lipids **(Saleh *et al.,* 2019).**

Soybean lecithin (SL) is a by-product from the soybean oil (SO) refinement process represent an economic alternative and permit giving an added value to residual products **(van Nieuwenhuyzen and Tomás, 2008)** , which is extracted from the SO degumming process, is mainly composed of polar lipids (> 60%), especially of phospholipids (PL), but also contains an important amount of neutral lipids (30%-40%), as triacylglycerols (TAG) and free fatty acids (FFA) **(Bueschelberger *et al.,* 2015).** Furthermore, SL represents a good source of phosphorus, choline and energy for broiler chickens **(Mateos *et al.,* 2012; Borsatti *et al.,* 2018)** and its combination with other fats and oils could be interesting in order to exploit positive synergies on lipid utilization **(Ravindran *et al.,* 2016).** In this respect, **Siyal *et al.* (2017)** showed that broilers fed diets supplemented with 0.1% soy lecithin had significant decrease in serum cholesterol, triglyceride and low density lipoprotein concentrations (LDL) compared to control and 0.05% soy lecithin. Also, **Mu and Kitts (2018)** found that broilers fed diets contained 50 and 100% soy lecithin achieved significantly (P<0.05) higher polyunsaturated fatty acids (PUFAs) especially linoleic and α-linolenic acids in breast and leg muscles, respectively compared to broiler fed 0 and 25% soy lecithin.

Exogenous carbohydrases such xylanase has been reported to improve energy utilization and the performance of broilers **(Olukosi and Adeola 2008 ;Cowieson *et al.,* 2010 and Williams *et al.,* 2014)** by hydrolysis of cell wall arabinoxylans and improve the access of endogenous digestive enzymes to cell contents **(Kocher *et al.,* 2003; Meng *et al.,* 2005 and Omar *et al.,* 2008).**

Selim *et al.* (2019) found that adding a combination of xylanase and protease to broilers diet had a significant (P ≤ 0.05) increase in dressing % and a reduction in edible parts and abdominal fat compared to control group. Moreover, **Saleh *et al.* (2019)** indicated that broilers fed low-energy diet supplied with mix of Xylanase and Arabinofuranosidase enzymes had significantly (p < 0.05) increased muscle total lipids content compared to other groups fed control or low-energy diets without enzyme supplementation. Therefore, the principal objective of the current research was to investigate the effects of lecithin, xylanase enzyme and their combination on carcass characteristics, meat quality and blood parameters in broilers fed low energy diet.

Materials and Methods

This study was conducted at the Poultry Research Farm of Animal Production Department, Faculty of Agriculture at Moshtohor, Benha University.

Additives used in this study:-

1- Crude soy lecithin.

It is a commercial source of soy lecithin 100% which contains:-

Acetone insoluble Matter 62%, Acid value 30%, Peroxide value 3% and hexane insoluble Matter 0.3%. The recommended level for poultry is 500g / ton of finished feed.

2- Xylanase enzyme.

The activity until of xylanase is 20000 u / g. This enzyme produced by *Trichoderma reesei* and *Asperigullus niger*, it's media is mineral oil 0.5g, rice hulls 44.5g and calcium carbonate 35g. The recommended level for poultry is 200g / ton of finished feed.

Experimental diets and birds:-

One hundred and sixty 1-d old unsexed Cobb 500 broiler chicks were weighed and randomly distributed to five treatments of 4 replicates each (n=8).

The five groups were follows:-

The first (control) group, received the strain recommended metabolizable energy (3000, 3000 and 3100 Kcal during the starter, grower and finisher periods, respectively), while the 2nd group was fed low metabolizable energy diet (NC) less 150 Kcal than the previous recommendation levels, being 2850, 2850 and 2950 Kcal / kg diet during the same growth periods. Moreover, the third, fourth and fifth groups fed NC supplemented with 0.5g lecithin / kg diet, 0.2g xylanase / kg diet and 0.5g lecithin + 0.2g xylanase / kg diet, respectively. The experimental periods lasted 35 days of age. All diets were formulated according to **NRC (1994)** as listed in Table (1) during starter (1-10 days), grower (11-22 days) and finisher (23-35 days) periods of growth. Chicks were housed in floor pens and kept under the same managerial, hygienic and environmental conditions. Also, chicks were provided with free access to water, feed and received the same lighting and veterinary care program during the experimental period**.**

Table 1. Composition and calculated analysis of the basal and tested diets.

	Control diets			**-150kcal**		
	Starter 1-10d	Grower 11-22d	Finisher 23- 35d	Starter 1-10d	Grower 11-22d	Finisher 23- 35d
Yellow corn	53.60	56.80	62.75	54.60	58.75	64.00
Soybean meal (44%)	32.75	33.20	25.55	36.40	35.40	28.85
Corn gluten (60%)	7.00	3.00	4.55	4.35	1.20	2.00
DI-Calcium phosphate	1.90	1.90	1.90	1.90	1.90	1.90
Limestone	1.30	1.30	1.30	1.30	1.30	1.30
Soya oil crude	2.50	2.85	3.00	0.50	0.50	1.00
NaCl	0.30	0.30	0.30	0.30	0.30	0.30
Vitamins and minerals mix *	0.30	0.30	0.30	0.30	0.30	0.30
DL-Methionine	0.15	0.15	0.15	0.15	0.15	0.15
L-lysine HCL	0.10	0.10	0.10	0.10	0.10	0.10
Sodium bicarbonate	0.10	0.10	0.10	0.10	0.10	0.10
Total	100	100	100	100	100	100
Calculated analysis **						
Crude protein %	23.1	21.0	19.1	23.1	21.0	19.0
Metabolizable energy (Kcal / kg)	3003	3002	3100	2853	2853	2954
Crude Fiber %	3.76	3.79	3.40	4.00	3.96	3.62
Crude Fat %	5.18	5.57	5.87	3.22	3.29	3.91
Calcium %	1.01	1.01	0.99	1.02	1.02	1.00
Available phosphorus %	0.51	0.51	0.49	0.52	0.51	0.50
Lysine %	1.24	1.23	1.03	1.33	1.28	1.10
Methionine %	0.59	0.53	0.52	0.57	0.51	0.50
Meth. + Cyc. %	0.97	0.87	0.84	0.95	0.86	0.81
Sodium %	0.16	0.16	0.16	0.16	0.16	0.16

* Each 3kg of vitamins and minerals contains: Vitamin A 12,000,000 IU, Vitamin D3 5,000,000 IU, Vitamin E 80,000 mg, Vitamin K3 3000 mg, Vitamin B1 3000 mg, Vitamin B2 9000 mg, Vitamin B6 4000 mg, Vitamin B12 20 mg, Niacin 60,000 mg, Biotin 150 mg, Folic acid 2000 mg, Pantothenic acid 15,000 mg, Choline 500,000 mg, Manganese 100,000 mg, Zinc 100,000 mg, Iron 40,000 mg, Copper 15,000 mg, Iodine 1000 mg, Selenium 350 mg, and Cobalt 100 mg.

.** According to **NRC (1994)**

Slaughtering and carcass characteristics:-

At 35 days of age, 1 bird per pen (4/treatment) was chosen for carrying out the slaughtering test. A total number of 20 birds slaughtered after 12h of feed withdrawal. After bleeding, and feather pecking, carcass, giblets including (liver, gizzard and heart), spleen and abdominal fat pad were removed and weighed and proportioned to live body weight as a percent.

Meat quality measurements:-

A total number of 20 samples of breast and thigh (from birds used previously in slaughtering test) were used monitor the effect on some physical and chemical traits of broiler meat. After chilling samples at 4°C for 24h, samples were lightly blotted using filter paper before reweighing. Drip loss percent was calculated as the percentage of the difference between weights before and after chilling for 24h and divided by the initial weight as described by **Saenmahayak *et al.* (2012).** Ultimate pH (pHu) was measured using pH meter, provided by a temperature control system, by probe method. The minimum depth to adopt was 1 cm after incision of the muscles as described by **Selim *et al* (2013).** Samples of thigh mixed with breast of the same bird were then stored at -20°C for 4 days before chemical measurements for total cholesterol (TC; mg/dl), Triglycerides (TG; mg/dl), Low density lipoprotein (LDL; mg/dl) and high-density lipoprotein (HDL; mg/dl) according to **Nasoetion *et al.* (2019).** Fraction of fatty acids were determined in meat by Gas Chromatography technique (GC) according to **Aura *et al.* (1995).**

Blood constituents:-

Blood samples were individually taken using heparinized syringes at the same time of slaughtering from 4 birds of each group. Blood samples were centrifuged immediately after collection at 2500 rpm for 20 min. and plasma were transferred into clean tubes then stored in a deep Freezer at approximately -20°C till the time of chemical analysis for Triglycerides (mg / dl) according to **(Soloni, 1971),** Total cholesterol (mg / dl), Low density lipoprotein (LDL; mg / dl) and High density lipoprotein (HDL; mg / dl) according to **Richmond (1973)** and Plasma liver enzymes (AST; u/l and ALT; u/l) were determine according to the method of **Reitman and Frankel (1957).**

Statistical Analysis:-

Data were statistically analyzed using ANOVA procedures of SAS **(SAS, 2004),** and significant differences between means was determined using Duncan's New Multiple-Rang test **(Duncan, 1955)** for all of experimental treatments.

The statistical model used for the trial was-:

$X_{ij} = \mu + T_i + e_{ij}$

Whereas:

X_{ij}= the observations

μ = Overall mean

T_i = Effect of treatment (i= 1,2,…..,5)

e_{ij} = Experimental error

Results and Discussion

Carcass traits

Effect of different treatments on carcass traits is presented in Table 2. Results show that group of NC + 0.2g xylanase recorded the highest carcass% without significant differences to others of negative control group (NC). While, the group of control and NC + 0.5g lecithin + 0.2g xylanase recorded the lowest values without significant differences between them. However, the rest of parameters did not show any significant among all groups. These results are supported by **Sarica *et al.* (2005)** who found that broilers fed on diets supplemented with antibiotic plus xylanase achieved significantly (P<0.05) higher hot and cold carcass yields than those fed diets supplemented with thyme, garlic, xylanase and garlic plus xylanase. Moreover, **Hajati (2010)** reported that supplementing corn-wheat-soybean diets with 500 mg kg−1 multi-enzyme significantly (p<0.05) increased broilers carcass at 44 days of age. While, the same group did not record any significant effect on heart, liver, gizzard, abdominal fat and spleen. Also, **Selim *et al.* (2019)** showed significant (P ≤ 0.05) increases in dressing percent due to xylanase and protease supplementation, while all carcass parts were not affected significantly (P > 0.05). On the other hand, **Hussein *et al.* (2019)** showed that broilers fed standard (3200 kcal/kg) and low energy diet (3000 kcal/kg) with or without enzyme mix supplementation had no significant differences in carcass traits (gizzard and liver).

Table 2: Effect of different treatments on carcass traits.

Treatments	Carcass traits %						
	Carcass %	Liver %	Gizzard%	Heart %	Giblets%	Spleen%	Abdominal fat%
Control	74.79^{c}	2.11	1.40	0.53	4.04	0.04	0.42
Negative control, NC	76.57^{ab}	1.98	1.42	0.46	3.86	0.09	0.29
NC + 0.5 g lecithin	75.15^{bc}	2.21	1.45	0.49	4.15	0.06	0.22
NC + 0.2 g xylanase	78.13^{a}	2.09	1.45	0.48	4.02	0.07	0.28
NC + 0.5 g lecithin + 0.2 g xylanase	74.71^{c}	2.02	1.43	0.55	4.00	0.08	0.28
±S.E	±0.52	±0.12	±0.09	±0.05	±0.20	±0.02	±0.08
Sig.	***	NS	NS	NS	NS	NS	NS

a, b….c means having different letters at the same column are significantly (P<0.05) different.
NS= not significant ***= (P<0.001)

The improvement carcass percent in NC + 0.2g xylanase group may be due to that xylanase could release more energy, hence increase carcass% **(Selim *et al.*, 2018)**. Whereas, other studies show that enzyme supplementation may improve the growth rate of broilers **(Khan *et al.*, 2006)** by enhancing the absorption of starch, amino acids **(Ziggers, 2006** and **Serena *et al.*, 2009)**.

Physical meat quality

Table 3 represents the effect of lecithin, xylanase enzyme supplementation and their interaction on Drip loss and pHu (physical meat quality measurements). The determined drip loss% of different broiler cuts showed significant reduction values in group of NC + 0.5g lecithin for both thigh and breast meat with a reduction percentage for thigh by 26.1% and 48% compared to control and NC, respectively. Moreover, a decrease % for breast was 30.1% and 71% compared to control and NC, respectively. Regarding to pHu values, it is worthy to note that none of the treatments show any significant varation between them. These results agree with **Zakaria *et al.* (2010)** who recorded that the addition of mixture enzymes did not significantly impact pH meat quality traits of broiler. Also, **Selim et al.(2016)** found that broilers fed diets less 100 or 150 Kcal / kg diet than standard diet had significant increase in drip loss by 36 and 46.4 %, respectively. In the present study, the reduction in drip loss% as a result of soy-lecithin may be due to that it contains phospholipids which increases the radical-scaving activity of vitamin E, then protecting the cell from oxidation **(Koya and Terao, 1995).** Then increase the ability of breast and thigh meat for processing, moreover, improve water holding capacity **(Barbut, 1993 and Zhang and Savage, 2010).**

Table 3. Effect of different treatments on physical meat quality.

Treatments	Drip loss%		pHu	
	Thigh	Breast	Thigh	Breast
Control	0.88[ab]	1.03[b]	6.39	6.14
Negative control, NC	1.25[a]	2.48[a]	6.29	6.13
NC + 0.5 g lecithin	0.65[b]	0.72[b]	6.49	6.15
NC + 0.2 g xylanase	1.04[ab]	2.36[a]	6.45	6.13
NC + 0.5 g lecithin **+** 0.2 g xylanase	1.12[a]	1.01[b]	6.36	6.46
±S.E	±0.12	±0.41	±0.24	±0.16
Sig.	*	**	NS	NS

a, b….d means having different letters at the same column are significantly ($P<0.05$) different.
NS= not significant *= ($P<0.05$) **= ($P<0.01$)

Chemical meat quality

Results of chemical meat quality parameters of broiler chicks fed diet supplemented with lecithin, xylanase or lecithin + xylanase are shown in Table (4). All tested groups decreased meat total saturated fatty acids compared to control and NC. On the other side, total poly-unsaturated fatty acids were significantly increased in these treated groups than others fed either control or NC diets. In respect of chemical meat quality, triglycerides, total cholesterol and LDL levels were significantly reduced in groups fed NC diet and supplemented with either 0.5g / kg lecithin, 0.2g / kg xylanase or the combination between them compared with control and NC. While, HDL values were significantly increased. These obtained results are in partial agreement with, **Mu and Kitts (2018)** who studied the effect of replacing 2.5% tallow with graded levels of soy-lecithin (0, 25%, 50% and 100%) on broilers meat composition of fatty acids and, found that chicks fed diets contained 50 and 100% soy lecithin achieved significantly ($P<0.05$) higher polyunsaturated fatty acids (PUFAs) in breast and leg muscles, respectively compared to broiler fed 0 and 25% soy lecithin. Moreover, the same authors found no effect in saturated fatty acids with feeding lecithin in both breast and leg meat. Also, **Dubey (2009)** reported that broilers fed diet contained 2, 2.5 and 3% crude soy lecithin (CSL) reduce total cholesterol in breast and thigh compared to control, 1% and 1.5% CSL. While, the chemical quality measurements of broiler meat including LDL and HDL did not changed by supplementing low-energy diet (-100 and -150 Kcal / kg diet) with a single dose of xylanase **Selim *et al.* (2015)** or pectinase **(Selim *et al.*, 2016).**

In current study, lecithin acts as an emulsifier which could distribute oil droplets evently in the emulsion and enhance the absorption and digestion of lipids so decrease lipid in meat **(Rovers, 2014).** Moreover, regarding to lecithin antioxidant properties, it makes synergistic action with α-tocopherol to stabilize PUFAs against lipid oxidation **(Koga and Terao, 1995).**

Table 4. Effect of different treatments on chemical meat quality.

Treatments	Chemical meat quality						
	TSFA (mg/100g fat)	TMUSFA (mg/100g fat)	TPUSFA (mg/100g fat)	Triglycerides (mg/dl)	Cholesterol (mg/dl)	HDL (mg/dl)	LDL (mg/dl)
Control	2089.67[a]	1792.67	890.33[d]	61.13[a]	143.23[a]	87.36[ab]	39.20[a]
Negative control, NC	1997.67[b]	1842.00	931.33[c]	60.50[a]	139.83[b]	86.27[b]	37.57[b]
NC + 0.5 g lecithin	1511.00[e]	2049.00	1138.00[a]	51.57[c]	128.43[d]	91.25[a]	34.33[c]
NC + 0.2 g xylanase	1592.00[d]	1995.00	1127.00[a]	55.90[b]	137.80[b]	86.62[b]	35.37[c]
NC + 0.5 g lecithin + 0.2 g xylanase	1814.00[c]	1608.00	995.00[b]	52.33[c]	132.67[c]	90.68[a]	35.23[c]
±S.E	±7.54	±132.10	±5.58	±0.82	±1.08	±1.20	±0.40
Sig.	***	NS	***	***	***	*	***

TSFA=total saturated fatty acids , TMUSFA= total mono-unsaturated fatty acids, TPUSFA= total poly-unsaturated fatty acids
a, b….e means having different letters at the same column are significantly (P<0.05) different.
NS= not significant *= (P<0.05) ***= (P<0.001)

Blood constituents

Results in Table 5, show no significant effect on plasma AST and ALT values. These results are in harmony with **Saleh *et al.* (2019)** who found that liver function indicators (AST, ALT) were not significantly affected by adding xylanase to broilers fed low-energy diet and illustrated that xylanase might be safe in poultry rations without negative effects on vital organ functions (liver). Also, **Rahnama *et al.* (2019)** with supplementation lecithin in broilers diet. Regarding to plasma lipid profile, all groups of NC and others fed NC added with different feed additives recorded significantly lower total cholesterol, triglycerides and LDL than the control. While the opposite situation was recorded for HDL, which all tested groups recorded higher HDL values than control.

These obtained results agree with recent research of, **Liu et al. (2020)** who found that adding 61.80% and 97.16% de-oiled lecithin to broilers diets resulted in significant increase in serum HDL and decrease serum LDL compared to control. They related this improvement to the better emulsification of supplementation of lecithin, so it can effectively and fully use of fat. Also, blood serum HDL/C and LDL/C contents support the lipid metabolism.

Table 5. .Effect of different treatments on plasma blood parameters.

Treatments	Liver enzymes		Lipid profile			
	AST u/l	**ALT u/l**	**Total cholesterol (mg/dl)**	**Triglycerides (mg/dl)**	**HDL (mg/dl)**	**LDL (mg/dl)**
Control	16.67	27.00	219.00[a]	44.33[a]	135.33[c]	53.67[a]
Negative control, NC	18.33	26.67	197.33[ab]	29.67[bc]	145.00[b]	37.33[b]
NC + 0.5 g lecithin	17.33	27.33	179.33[b]	29.33[bc]	146.87[b]	24.67[c]
NC + 0.2 g xylanase	16.67	25.67	190.67[b]	34.67[b]	146.67[b]	39.00[b]
NC + 0.5 g lecithin + 0.2 g xylanase	19.00	27.33	189.00[b]	27.00[c]	157.80[a]	23.33[c]
±S.E	±1.19	±1.62	±7.27	±1.72	±2.60	±4.00
Sig.	NS	NS	*	***	***	***

a, b….c means having different letters at the same column are significantly (P<0.05) different.
NS= not significant *= (P<0.05) ***= (P<0.001)

On contrary, **Dubey (2009)** found that chicks fed diet containing different levels of crude soy lecithin (0, 1, 1.5, 2, 2.5 and 3%) had no significant effect on serum cholesterol, HDL and ALT. Also, these results are coincident with the findings of **Saleh *et al.* (2019)** who reported that plasma HDL was significantly increased and total cholesterol decreased in broilers fed low-energy diet supplied with xylanase compared to control, such this improvement might be due to enhance the fat digestibility hence improve its absorbtion.

Conclusion

In conclusion, broiler chicks fed low energy diet (-150 Kcal / kg diet) supplemented with single dose of 0.5g lecithin, 0.2g xylanase or combination between them enhanced carcass relative weight, increased blood plasma HDL concentration, decreased plasma LDL, total cholesterol and triglycerides and improved meat quality.

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تأثير اضافة ليسيثين الصويا والزيلانيز والتداخل بينهم للعلائق المنخفضة فى الطاقة على صفات الذبيحة وجودة الذبيحة وجودة اللحم وبعض مقاييس الدم لدجاج التسمين

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تهدف هذه الدراسة الى التحقق من علاقة التآزر بين الليسيثين وإنزيم الزيلانيز عند اضافتهم لعلائق منخفضة الطاقة على خواص وجودة الذبيحة لدجاج التسمين. استخدم فى هذة الدراسة160 كتكوت موزعة عشوائياً على 5 معاملات ؛ بكل منها 32 طائر (4 مكررات بكل منها 8 طيور). تغذت كتاكيت المجموعة الأولى على عليقة اساسية بدون اى اضافات واعتبرت كنترول. تغذت كتاكيت المجموعة الثانية على عليقة منخفضة الطاقة (–150 كيلوكالوري / كجم أقل من الموصى به) واعتبرت كنترول سالب. تغذت كتاكيت المجموعات الثالثة والرابعة والخامسة على عليقة الكنترول السالب مضاف إليها 0,5 جم ليسيثين/كجم و 0,2 جم زيلانيز /كجم و 0,5 جم ليسيثين + 0,2 جم زيلانيز / كجم ، على التوالي. استمرت الدراسة 35 يومًا من العمر. أظهرت النتائج المتحصل عليها أن مجموعة المغذاة على الكنترول السالب +0,2جم زيلانيز / كجم سجلت زيادة معنوية في الاوزان النسبية للذبيحة. حققت المجموعة المغذاة على الكنترول السالب +0,5 جم ليسيثين /كجم أقل نسبة من الأحماض الدهنية المشبعة الكلية والكولستيرول الكلى والكوليسترول منخفض الكثافة والدهون الثلاثية. علاوة على ذلك ، سجل كل من المجموع المغذاة على الكنترول السالب +0,5جم ليسيثين أو الكنترول السالب + 0,5جم ليسيثين +0,2جم زيلانيز انخفاضًا معنويًا في الكوليسترول الكلي ، والدهون الثلاثية والكولستيرول منخفض الكثافة في بلازما الدم ، وزيادة في كولستيرول عالى الكثافة في بلازما الدم مقارنة بالكنترول.

الخلاصة:– إضافة 0,5 جم من الليسيثين بمفرده أو مع 0,2 جم زيلانيز إلى علائق دجاج التسمين منخفضة الطاقة يكون أكثر فاعلية على الذبيحة وجودة اللحوم ودهون بلازما الدم.