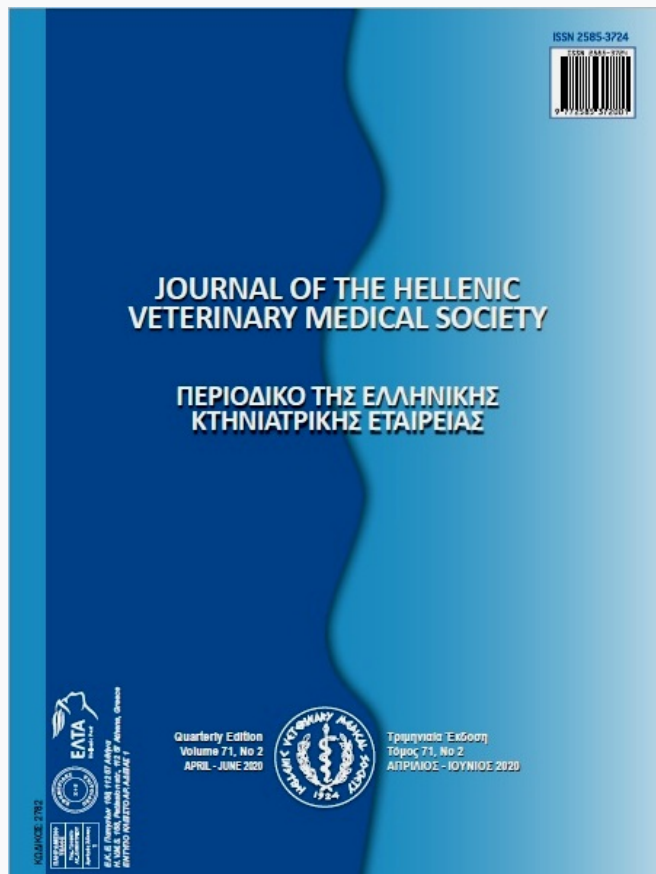


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Effects of the physical form of diet on growth performance, ascites and sudden death syndrome incidences in broiler chickens

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ABSTRACT: This experiment was conducted to determine the effects of the diet physical form, Mash (M), Pellet (P) and Extruded (E) on the growth performance, carcass characteristics and metabolic disorders ascites (AS) and as well as sudden death syndrome (SDS) in the broiler chickens. In this respect, feed intake (FI), weight gain (WG) and mortality were recorded throughout the experiment and biochemical parameters, hematology and carcass characteristics were tested at 35 and 42 days of age respectively. The results showed that with the increase of the average daily weight gain (ADWG) ($p<0.01$), the relative breast weight to the carcass weight ($p<0.05$), better feed conversion ratio ($p<0.01$), the lower relative cecum weight and gizzard to the carcass weight ($p<0.05$) were observed by applying the E diet form, as compared with the other treatments. A significant increase in the average daily feed intake (ADFI) was also observed by using the P diet form ($p<0.01$). Hematological parameters including hemoglobin (Hb), hematocrit (HCT), urea, uric acid, triglyceride, the ratio of low-density lipoprotein to high-density lipoprotein (LDL/HDL), very low-density lipoprotein (VLDL), enzyme activities of aspartate aminotransferase (AST) and alanine aminotransferase (ALT) of the blood serum were lowered by applying the M treatment, as compared with the other treatments ($p<0.05$). The results indicated that the performance and carcass characteristics were improved by the E and P diet forms; also, with raising the hematology parameters in these treatments, the mortality of ascites and SDS was increased.

Keywords: Ascites, Feed form, Metabolic disorder, Sudden death Syndrome

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INTRODUCTION

The physical forms of the diet mainly include three physical forms; mash, crumble and pellets (Banerjee, 1998). Various feed forms are the most important factors which directly influence the cost of mixed feed and production performance in the broiler (Abdollahi et al., 2013).

Mash form diets can be easily prepared, requiring relatively less energy in the process of preparation (Banerjee, 1998). Mash feed is a form of complete feed that is milled and blended so the birds could not easily separate out the diet ingredients. Feed milled on a farm is typically a mash feed. Pelleted feed involves a finer ground feed processed with heat, pressure, moisture and the added ingredients to bind pellets together. A crumble is a pellet that has been broken down to the desired size, which is usually smaller than the pelleted feed. Making a pellet or crumble diet requires extra specialized processing and equipment and this could increase the processing diet costs (Jean and Trevidy, 2000). Several studies have reported that pellets increased the body weight, feed intake and feed efficiency (Munt et al., 1995; Asha Rajini et al., 1998b). Pelleted diet enhances feed intake and growth of animals probably related to reduce feed waste, decreases energy expended for consumption, improves palatability, and reduces dustiness of feed (Abdollahi et al., 2013). In recent years the extrusion technique is extensively used in animals feed because this process has many benefits such as high productivity, efficiency and high quality of the final product (Moritz et al., 2005). The extrusion process is a short-time by high temperature, starchy and/or proteinous feed or feed materials are cooked with the help of moisture, temperature and pressure. The results of this processing showed molecular transformation and chemical reactions within the processed feed or ingredients. The extrusion process also has positive impact in term of denaturing of harmful enzymes, inactivation of anti-nutritional factors, such as tannin, phytate, trypsin inhibitor, haemagglutinin and also sterilizes the final product of extrusion (Bjorck and Birkhed, 1984).

Ascites and sudden death syndromes are two types of the important metabolic disorders which caused by genetics, environmental and nutritional factors (Baghbanzadeh and Decuypere, 2008). The major factor regarding mortality is related to the fast growing birds in the poultry industry; these syndromes are related to the poor performance of the heart in fast growing broilers (Korte et al., 1999). Ascites is characterized by fluid accumulation in the abdominal cavity and

SDS is a sudden death in good conditions in a healthy bird without any apparent causes within 1-2 minutes, such that broilers flip over on their backs and die (Olkowski et al., 2008; Saki and Hemati, 2011).

Feed form is one the factors influencing the performance and growth rate in the broiler chickens. A direct relationship between the broiler growth rate (a high metabolic rate) with susceptibility to AS and SDS, especially when the birds are fully fed, has been reported (Proudfoot and Hulan, 1982; Olkowski et al., 2008; Siddiqui et al., 2009). Several researchers have shown that blood parameters change in the body with these syndromes (Kaul and Trangadia, 2003; Saki and Hemati, 2011). Our previous studies have revealed that blood profiles can be used as an indicator to determine and prevent the metabolic disorders, AS and SDS (Saki and Hemati, 2011). In the light of these issues, the objective of this study was to investigate the effects of the physical form of feed (M, P, and E diet forms) on the performance, carcass, and organ characteristics and to find out the relationship between agents to metabolic disorders, AS and SDS in the broiler chickens.

MATERIALS AND METHODS

Birds and housing

The experiment was carried out at Karaj-Iran. A total of 936 day-old broiler chicken (Ross, 308) with approximately 44.0 ± 2 g body weight was assigned randomly into 3 treatments: mash (M), pellet (P) and extruded (E), included 12 replicate pens per treatment. The study was conducted during 0–42 days of age. Broiler chickens were raised in floor pens (160×300 cm). Access to feed and water was *ad libitum*. The lighting schedule was 23h light/ 1h darkness at temperature 32° C the first day and then reduced by 3° C each week until third week and thereafter it was constant. The vaccination program was done according to local veterinarian suggestion.

Experimental diet

The ingredient chemical composition as well as the calculated composition of the diets during starter (0–14 d), grower (15–28 d) and finisher periods (29–42 d) are presented in Table 1.

The average length of feed in pellet and extruded was 1-1.5 mm at starter periods, 1.5-2.5 mm at grower periods and 2.5-3.5 mm at finisher periods. Diets were supplied at Beyza Feed Mill. Nutritional requirements were provided based on the standard recommendations (Ross, 2014).

Table 1. Composition and estimated nutrient value of diets at 0-42 days of age.

Ingredient, (%)/days	0-14	15-28	29-42
Corn	44.3	49.4	51
Soybean meal 44%	39.4	33.1	28.9
Wheat	8	10	14
Soybean oil	1.77	1.9	1.85
Other Nutrients*	6.53	5.6	4.25
Chemical composition			
Crude Protein	23.58	20.8	18.8
ME, (kcal/ kg)	2850	2920	2960
Crude Fiber	3.74	3.61	3.5
Ether Extra	3.89	4.17	4.14
Lysine	1.24	1.1	0.97
Methionine	0.61	0.55	0.49
Methionine+Cystine	0.9	0.82	0.74
Threonine	0.9	0.75	0.7
Ca	1.02	0.98	0.94
Available P	0.49	0.47	0.45

*Other Nutrients: Dicalcium phosphate, Salt, Fish meal, Vitamin and mineral premix.

Vitamin and trace elements supplied per kg of diet: vit A, 16000 IU; vit D3, 5000 IU; vit E, 60 mg; vit K3, 60 mg; vit B1, 2 mg; vit B2, 3.5 mg; niacin, 35mg; calcium pantothenate, 12.8 mg; vit B3, mg; vit B12, 0.017 mg; choline chloride, 1000 mg; folic acid, 1 mg; biotin, 0.2 mg; Mn, 85 mg; Zn: 70 mg; Cu: 18 mg; I, 1 mg; Co, 0.6 mg; Fe, 40 mg; Se, 0.07 mg; antioxidant, 100 mg.

Data collection

Feed intake, body weight and feed conversion ratio were measured weekly, as well as mortality was recorded daily and mortality was divided into three groups (Table 4).

The blood samples were taken at 35 days of age, due to a peak in blood and biochemical parameters at this age (Malan et al., 2007; Hosseini et al., 2014). Two birds in each replicate were selected and the blood samples were collected in two tubes; one tube including ethylenediamine tetra-acetic acid anticoagulation (EDTA) and other tubes without EDTA. The first tube was used to determine hematocrit (HCT) and hemoglobin (Hb) concentration by microhematocrit and cyanmethemoglobin methods respectively by kits Zist shimi (Zist shimi, Tehran, Iran). In the second tubes, obtain serum for the determine of glucose, triglyceride, cholesterol, uric acid, high-density lipoprotein (H.D.L), low-density lipoprotein (L.D.L), very low-density lipoprotein (VLDL), Ca, K were measured by diagnostic kits ParsAzmun (ParsAzmun Co, Tehran, Iran) and Spectrophotometer (Jenway Geno-

va MK3, UK). Enzymes activities ALT, AST, Alkaline Phosphatase (ALP) and lactate dehydrogenase (LDH) were considered by Vitros 350 autoanalyser (New York, USA; code 680-2153) and commercial kits (Vitros Chemistry Products, Ortho-Clinical Diagnostics, Johnson Company, New York, USA).

Broiler chickens were eviscerated and separated into different parts for assessment of carcass characteristics at 42 days of age. Weights were expressed as a percentage of the body weight, thus obtaining the relative weight of organs.

Statistical analysis

A completely randomized design (CRD) and the GLM procedure of SAS 9.1 software (SAS, 2009) by one-way analysis of variance (ANOVA) were used for each phase (1-14 d, 15-28 d and 29-42 d) and for the whole experimental period (1-42 d). Differences between treatments means were compared with Duncan's test. All significance was based on a P-value equal to 0.01 and 0.05. Mortality analyses were performed based on Chi-Square.

RESULTS

The effects of the diet physical form on the performance in the broiler chickens are shown in Table 2. The increase of the ADWG during different weeks ($P < 0.01$) and better FCR at these weeks ($P < 0.01$) have shown by the E diet form in the broiler chickens. Average daily feed intake was increased by the P diet form at 2- 5 weeks of age ($P < 0.01$). This indicated that the performance of the broiler chickens was strongly influenced by the physical form of the diet. No significant differences were observed in ADWG at 2-5 weeks of age between the P and E diet form in the broiler chickens (Table 2).

The effects of the feed form on hematology parameters and enzyme activities are presented in Table 3. There were no significant differences in the levels of cholesterol, H.D.L, L.D.L, ALP, LDH, Ca, K and glucose serum in broilers. The highest values of the hemoglobin and the hematocrit and the serum level of the urea, uric acid, triglyceride, ratio of the Low-density lipoprotein to the high-density lipoprotein (LDL/HDL), very low-density lipoprotein (VLDL), enzymes of the Aspartate aminotransferase (AST) and Alanine aminotransferase (ALT) ($p < 0.05$) were observed by the E and P treatments and also the lowest was shown by the M treatment (Table 3). In contrast, no significant differences were found between the E and P treatments.

Table 2. Average performance of broiler chicken by treatments at the different weeks

Variable	Dietary treatments *			SEM	P-Value
	Mash	Pellet	Extruded		
ADWG (g/d)					
1	13.81. ^c	17.05 ^b	17.71 ^a	0.115	0.0001
2	31.04 ^b	38.76 ^a	41.03 ^a	0.959	0.0018
3	52.77 ^b	74.20 ^a	75.66 ^a	1.094	0.0001
4	70.60 ^b	95.49 ^a	89.11 ^a	1.342	0.0001
5	85.80 ^b	95.55 ^a	93.64 ^a	1.168	0.0091
6	76.19 ^a	63.12 ^b	78.01 ^a	1.726	0.0059
ADFI (g/d)					
1	18.88	19.57	19.52	0.317	0.6171
2	44.04 ^b	53.16 ^a	51.48 ^a	0.719	0.0003
3	80.30 ^b	96.70 ^a	99.34 ^a	0.738	0.0001
4	116.52 ^c	144.92 ^a	136.97 ^b	1.341	0.0001
5	164.52 ^b	176.60 ^a	170.44 ^{ab}	1.567	0.0231
6	189.81	185.44	190.54	1.778	0.4676
FCR					
1	1.37 ^a	1.15 ^b	1.12 ^b	0.021	0.0004
2	1.24	1.18	1.11	0.021	0.0849
3	1.37 ^a	1.24 ^b	1.206 ^b	0.007	0.0001
4	1.48 ^a	1.36 ^b	1.33 ^b	0.008	0.0001
5	1.63 ^a	1.51 ^b	1.48 ^b	0.010	0.0001
6	1.84 ^a	1.82 ^a	1.70 ^b	0.018	0.0153

*The same superscript alphabets in the same row indicate a non-significant different at $p < 0.01$, $P < 0.05$.

(ADWG = Average Daily Weight Gain; ADFI= Average Daily Feed Intake; FCR= Feed Conversion Ratio)

*Treatments (n=3 Mash, Pellet, Extruded); replicates (Mash (n= 12), Pellet (n= 12), Extruded (n= 12), 26 bird in each pen

Table 3. Biochemical and hematology parameters and enzymes activities of blood broilers by treatments at 35 day of age

Variable	Dietary treatments*			SEM	P-Value
	Mash	Pellet	Extruded		
Hemoglobin, (g/dl)	11.825 ^b	13.475 ^a	12.725 ^a	0.157	0.0025
Hematocrit, (%)	32.450 ^b	36.400 ^a	34.625 ^{ab}	0.457	0.0107
Urea, (mg/dl)	2.333 ^b	2.458 ^{ab}	3.333 ^a	0.169	0.0057
Uric acid, (mg/dl)	4.900 ^b	7.083 ^a	6.533 ^a	0.241	0.2334
Cholesterol, (mg/dl)	111.667 ^a	115.667 ^a	122.500 ^a	2.496	0.2334
Triglyceride, (mg/dl)	102.17 ^b	131.50 ^{ab}	142.33 ^a	6.009	0.0409
H.D.L, (mg/dl)	82.500 ^a	75.500 ^a	76.167 ^a	1.811	0.2513
L.D.L, (mg/dl)	113.167 ^a	113.667 ^a	116.167 ^a	5.63	0.2122
LDL/HDL	1.371 ^b	1.505 ^{ab}	1.525 ^a	0.180	0.0311
VLDL, (mg/dl)	20.167 ^b	26.500 ^a	28.000 ^a	1.190	0.0387
AST, (IU/l)	272.17 ^b	481.83 ^a	503.67 ^a	26.708	0.0052
ALT, (IU/l)	11.000 ^b	22.667 ^a	27.000 ^a	1.933	0.0115
ALP, (IU/l)	91.390 ^a	93.017 ^a	95.132 ^a	18.195	0.7073
LDH, (IU/l)	1955.7 ^a	1975.5 ^a	2023.2 ^a	132.208	0.9773
Ca, (mg/dl)	10.50 ^a	10.87 ^a	11.32 ^a	0.152	0.119
K, (mmol/l)	5.28 ^a	5.38 ^a	5.66 ^a	0.100	0.2982
Glucose, (mg/dl)	241 ^a	253.33 ^a	248.167 ^a	2.388	0.1407

*The same superscript alphabets in the same row indicate a non-significant different at $P < 0.05$.

(HDL: High-density lipoprotein, LDL: Low-density lipoprotein, VLDL: Very low-density lipoprotein, AST:Aspartate aminotransferase, ALT: Alanine aminotransferase, ALP: Alkaline Phosphatase, LDH: Lactate dehydrogenase)

*Treatments (n=3 Mash, Pellet, Extruded); replicates (Mash (n= 12), Pellet (n= 12), Extruded (n= 12) with 26 bird in each pen.

There were significant differences in mortality among treatments ($p < 0.05$) (Table 4). The higher mortality percentage was found in the E as well as the P treatment and a lower mortality percentage was observed in the M treatment during the experiment. The rate of mortality was 5.45% during the experiment period. The higher mortality percentage of AS and SDS were appeared in the broiler chickens, which received the E and P diet form rather than the M diet form (Table 4).

Table 4. Number and rate of mortality in broiler chicken by treatments at 0-42 days of age

Mortality/ Treatments	Mash		Pellet		Extruded		Total		
	No	(%)	No	(%)	No	(%)	No	(%)	
Ascites	0	0	12	1.28	9	0.96	21	2.24	
SDS	2	0.21	3	0.32	4	0.43	9	0.96	
Others	8	0.85	2	0.21	11	1.18	21	2.24	
Total	10	1/07	17	1.82	24	2.56	51	5.45	
p-value								0.0471	

*The same superscript alphabets in the same row indicate a non-significant different at $P < 0.05$.

*Treatments (n=3 Mash, Pellet, Extruded); replicates (Mash (n= 12), Pellet (n= 12), Extruded (n= 12) 26 bird in each pen

Number: 936 day-old broiler chicken; Number (No), Percent (%)

The relative breast weight to the carcass was lower by the M diet form than the E and P treatments ($p < 0.05$). The greater relative weight of the cecum and gizzard to the carcass weight were indicated than the E and P treatments at 42 days of age ($p < 0.05$) (Table 5). No significant differences were monitored in carcass percentage, lung, abdominal fat, liver, intestine length and tibia bone yields in respect to all treatments.

Table 5. Average relative weights (%) of the viscera of the broilers by treatments at 42 day of age

Variable	Dietary treatments			SEM	P-Value
	Mash	Pellet	Extruded		
Carcass yield, %	68.49 ^a	71.33 ^a	71.38 ^a	2.429	0.0928
Breast, %	23.10 ^b	25.39 ^a	26.19 ^a	0.376	0.0118
Lung, %	0.46 ^a	0.36 ^a	0.44 ^a	0.022	0.1775
Liver, %	2.48 ^a	2.53 ^a	2.65 ^a	0.059	0.4918
Gizzard, %	1.84 ^a	1.21 ^b	0.84 ^c	0.061	0.0001
Abdominal fat, %	0.93 ^a	1.22 ^a	0.95 ^a	0.058	0.1136
Length of intestine, %/g	7.29 ^a	6.82 ^a	7.02 ^a	0.2221	0.107
Cecum, %	0.63 ^a	0.56 ^a	0.46 ^b	0.018	0.0054
Tibia bone, %	0.93 ^a	0.91 ^a	0.85 ^a	0.040	0.7032

*The same superscript alphabets in the same row indicate a non-significant different at $P < 0.01$, $P < 0.05$.

DISCUSSION

With the increase of the average daily weight gain, a better FCR was found by applying E and P treatments, in comparison to the M diet form. The average daily feed intake was increased by the P diet form rather than the M one in the broiler chickens.

Atapattu et al. (2005) have found that broiler chickens spend more and less time in feeding and resting by M and P diet respectively. Therefore, better utilization of energy and nutrients in the case of P diets, may be increased. Nir et al. (1996) have also indicated a higher weight gain in those broiler chickens fed with an extruded soybean meal rather than those fed with a non-extruded soybean meal. Also, Asha Rajini et al. (1998b) have reported a heavier body weight in the chickens fed with the P form than the M diet.

Hamm and Stephenson (1959) have indicated that P diets could give a greater feed intake than the M form diet. Asha Rajini et al. (1998b) have stated that P diet could have a better feed efficiency than the M diet. Munt et al. (1995) observed that broilers fed by the P diet showed a better growth performance in comparison to those fed by the M diet. Increased body weight, feed intake and improved the feed: the gain ratio was shown by feeding the P, as compared with those birds fed by the M diets.

Many researchers have indicated the positive effect of E on the performance of the chicken (Moritz et al., 2005; Marsman et al., 1997). They have attributed this better performance to improved gelatinization, intake, digestibility, removal of antinutritional factors and the good quality of the extruded feed or ingredients. The different performance of poultry and nutrient digestibility could be mainly due to various processing techniques and extrusion conditions. To obtain the maximum nutrient digestibility and ensure the best performance of the poultry processing techniques and extrusion conditions should be maintained at the standard levels.

Moritz et al. (2005) observed that the E process of corn led to an increase in the body weight of broiler chickens when they were in 0 to 3 weeks of their age. The improved growth performance of the broilers fed by the extruded (E) SBM has been lighted by Marsman et al. (1997). In modern feed milling operations, E can be considered as the basic process to enhance the profitability of the feed. The E process can be useful in terms of the enhanced nutritional

value and efficiency of ingredients and feed, depending upon many factors such as the structure and chemical composition of the ingredients, processing conditions and the machinery used in processing (Rahman et al., 2015).

The nutritional value and, digestibility of feed ingredients, therefore, could influence the performance of the poultry based on the variations in temperature, moisture, screw speed, pressure, time along with extruded material chemical composition and structure. To attain the maximum results from the extrusion processing techniques, all conditions should be maintained at optimum levels (Lin et al., 1997). In general, heating improves the digestibility of proteins by inactivating enzyme inhibitors and denaturing the proteins that may expose new sites for enzyme attacks (Camire et al., 1990). Conditioning time can change gelatinization degree of starch. So, the degree of gelatinization can affect the growth rate and body weight in the poultry.

Several studies have also shown that AS and SDS could cause high mortality in the broilers which are in 3 to 5 weeks of their age (Gardiner et al., 1988; Wide-man, 2001). Broilers in the M treatment had a significantly lower mortality rate due to ascites and SDS.

Genetic selection for growth and feed conversion ratio increased the final body weight and performance (Decuypere et al., 2005; Olkowski et al., 2008); this was accompanied by the insufficiency of oxygen supply to the tissues in rapidly growing broiler chickens (Huchzermeyer, 2012; Wideman et al., 2013; Hasanpur et al., 2015). This could be as a result of the enhanced cardiac output and the reduced oxygen content of blood, the increased production of red blood cells and hematocrit value; these were consistent with the results of the current study (Decuypere et al., 2005).

Feed form is one of the effective factors on the performance and growth rate in the broilers. Feeding by the P diet form, as compared to the M one, increased body metabolism, hence raising the incidence of SDS and ascites in the broiler chickens; so it could be identified as the major causes of mortality related to the fast growth (Proudfoot and Hulan, 1982; Kaul and Trandgia, 2003; Druyan et al., 2009).

In the broilers susceptible to AS and SDS syndromes, the biochemical conditions, hematological parameters and enzymes, and hormones were

changed. The P and E diet forms increased the feed intake and growth rate due to high metabolic demand; this was as a result of the increase in hemoglobin, hematocrit and enzymes such as AST, ALT and ALP values (Schindhelm et al., 2006). Broiler chickens fed with E and P diet forms showed increased hematological parameters, as well as higher serum levels including urea, uric acid, triglyceride, as well as the rise of the ratio of LDL/HDL, VLDL, AST and ALT, as compared with those fed by the M diet form. The changes in enzymes activities and lipid metabolism could be related to the metabolic abnormalities and the high metabolic demand in the liver. This could lead to hepatocellular degeneration and destruction of the liver cells, which, in turn, result in the leakage of these enzymes into the blood stream; so, liver damage could occur (Hung et al., 2008; Saki and Hemati, 2011; Parmar et al., 2012; Shen et al., 2014; Senanayake et al., 2015). There is, however, little information regarding the changes in biochemical parameters and enzymes activities that can cause AS and SDS in different forms of feed.

In this experiment, carcass characteristics such as the relative breast weight were increased and the relative weight of cecum and, gizzard was decreased by E and P diet forms. The results, therefore, showed that feeding the mash form had a limited effect on carcass characteristics.

Amerah et al. (2007) also observed that the weight of the breast was significantly increased by the P diet than the M diet form in broiler chickens; this was in agreement with our study. Ahmed and Abbas (2013) indicating that the decrease in the relative weight of gizzard could be realized by the P diet rather than the M one in chickens. Gizzard weight was influenced by the diet form and size, resulting in a higher weight and yield in the broilers fed by M, as compared to the P diets. The gizzard function might have been affected by the M diet, resulting in a significant muscle development. Detailed results are reported in the literature (Lopez and Baiao, 2004), demonstrating that the gizzard relative weight of the broilers fed by the M diets was greater than of those fed by the P feeds, which was in agreement with the current study.

CONCLUSION

The results of this study, therefore, showed that the diet form played an important role in the feed efficiency, carcass characteristics and mortality of the broiler

chickens. The P and E diet forms increased the performance and carcass characteristics; so the increase in these parameters was associated with the mortality of ascites and SDS in the broiler chickens. However, the association between mortality (AS and SDS) and changes in hematology and enzymatic parameters needs to be further investigated in the future studies.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

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