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Responses of Modern Broiler Chicks to Stocking Density, Green Tea, Commercial Multi Enzymes and Their Interactions on Productive Performance, Carcass Characteristics, Liver Composition and Plasma Constituents

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Abstract: The objective of this work was to study the responses of modern broiler chicks to stocking density, green tea, commercial Multi enzymes (Optizyme® p-5) and their interaction on productive performance, carcass characteristics, chemical composition of liver and plasma constituents. Commercial breeds (Lohman) of broiler chicks were housed at different densities of 10, 14 and 18 bird/m^2 during the period from 7 to 43 days of age. Chicks within each density were fed four experimental diets; one of them was used as a control diet. In the other three diets the control diet was supplemented with green tea (5g/kg feed) or Multi enzymes (1 g /kg feed), or mixture of both. Therefore, there were 12 experimental treatments in which each treatment was replicated four times. Results could be summarized as follows: Stocking density had no significant effect on growth of broiler chicks, feed intake and feed conversion ratio (FCR) during 7-43 days of age, except for increasing stocking density of 18 bird/m^2 during only the period from 22 to 43 days of age while reduced growth and feed intake significantly. Addition of green tea, Multi enzymes or their mixture had no significant effect on growth, feed intake and FCR of broiler chicks. There were significant interactions among stocking density and feed additives. The best dressing percentage was obtained from group stocked at 14 bird/m^2 and fed the control diet. Also, hind part weight and % were the highest of birds stocked at 10/m^2 and fed diet supplemented with Multi enzymes or those stocked at 14 bird/m^2 and fed the control diet. There was significant interaction between stocking density and feed additives on spleen as absolute or relative weight, showing that the highest weight and % of spleen were obtained of groups fed mixture of Multi enzymes and green tea and stocked at 10 bird/m^2, and groups fed diets supplemented with Multi enzymes and stocked at either 14 or 18 bird/m^2. Dressing % and hind part weight (g) and (%) decreased significantly as a result of increasing stocking density from 14 to 18 bird/m^2, but they were not significantly different from those stocked at 10 bird/m^2. Multi enzymes improved absolute weight (g) of front part compared to the control diet, while addition of green tea, Multi enzymes and their mixture improved front part % significantly compared to the control diet. A mixture of green tea and Multi enzymes significantly increased liver%, while Multi enzymes decreased significantly absolute and heart % compared to the control group. A mixture of green tea and Multi enzymes significantly decreased intestinal weight and % compared with the control diet without or with green tea. Stocking density had no significant effect on chemical composition of liver and plasma constituents of broiler chicks as well as liver functions as judged by plasma ALT and AST activities during the experimental period. Green tea had no significant effect on chemical composition of liver and plasma total protein and its fraction, total lipids and cholesterol and plasma AST and ALT activities. In conclusion broiler chicks could be stocked up to 14 bird/m^2 without adverse effect on growth performance, while addition of green tea, Multi enzymes or their mixture had no enhancing effect on growth performance of broiler stocked at different densities.

Key words: Broiler chicks, stocking density, feed additives, growth performance, chemical constituents

Introduction

Increasing stocking density of broilers is a management practice used for reducing costs associated with labor, housing, fuel and equipments. However, crowding of broilers can lead to reductions in performance (Shanawany, 1988). Broiler performance and health can be influenced by very high stocking density (Weaver *et al*., 1982; Webster, 1990), thereby it is important to assure that adequate floor space is available for each bird (Al-Homidan, 2001).

Parkhurst *et al*. (1977); Beremski (1987); Mizubuti *et al*. (1994) concluded that there was no significant difference in broiler performance due to stocking density. However, Buckland *et al*. (1971); Proudfoot *et al*. (1979); Weaver *et al*. (1982); Reiter and Bessei (2000) suggested there was a linear increase in growth with decreasing stocking density at 5 wk of age, respectively. The decrease in growth performance resulted from increasing stocking density could be attributed to the increase in stress resulting from competition for feed and water, increase of house temperature, microbial activity, and ammonia production.

On the other hand, there are possible ways to alleviate the stress resulting from increasing stocking density

such as assuring adequate feeding and watering space, improve the ventilation and house environment. Nowadays, non-conventional growth enhances are utilized in broiler nutrition to improve nutrient digestibility, control of pathogenic microorganisms, facilitate a favorable intestinal microbial balance, and enhancing absorption of calorigenic nutrients across the gut wall through increasing its absorption capacity (Nelson *et al*., 1963; Attia *et al*., 1997; Al-Harthi, 2002; El-Husseiny *et al*., 2002; El-Deek *et al*., 2003). In this regard, Tollba (2003) and Tollba and Hassan (2003) found that black cumin, garlic, thyme or fennel as natural feed additives to broiler diets under normal and high temperature conditions improved growth, FCR, and decreased mortality rate.

Green tea leaves (Camellia sinensis) containing antioxidative tea catechins consisting of various polyphenols (Miura *et al*., 2001; Varilek *et al*., 2001). Tea catechins have a variety of pharmacologic effects, i.e. antioxidative (Lin *et al*., 1996), antimutagenic (Jain *et al*., 1989), anticarcinogenic (Sano *et al*., 1999), and anti-inflammatory effects (Varilek *et al*., 2001). Recently Miura *et al*. (2001) reported that ingestion of green tea improved growth of mice and did not affect plasma cholesterol and triglyceride concentrations. However, plasma peroxides were reduced indicating that *in vivo* oxidative state is improved by tea ingestion. Moreover, aortic cholesterol and triglyceride contents were 27 and 50% lower in tea group than in the control group, respectively.

Multi enzymes have been found to increase availability of protein/amino acid, ME and minerals for broilers (Makled, 1993; Jeroch *et al*., 1995; Bedford, 1996; Attia *et al*, 2001; Aulrich and Flachowsky, 2001; Brufau *et al*., 2002). In this concern, Osman and Tanios (1983) reported that amylase levels in the proximal parts of the intestine of broilers and laying hens was sharply decreased upon exposed to heat stress. However, the response in maltase was insignificant. In this regard, Zanella *et al*. (1999) found that Avizyme 1500® improved the overall CP digestibility by 2.9%, and improved growth by 1.9 and FCR by 2.2% respectively of broilers fed corn soybean diet, indicating that enzyme supplementation completely compensated for the reduced energy of the diet. Bedford (1996) concluded that exogenous enzymes could improve digestion for protein, starch and fat, by removing the antinutritional factors, which interfere with normal processes of digestion, or by digestion of fibre components that would otherwise pass undigested into the environment, and extend the use of enzyme to play a significant role in heath of the digestive tract.

The aim of this study was to investigate the responses of a modern broiler strain to different stocking densities and the impact of green tea or commercial Multi enzymes and their mixture as an agent for alleviating stress and the interaction between them on productive performance, digestibility of nutrients, carcass characteristics, and chemical composition of liver and plasma constituents.

Table 1: Composition and calculated analyses of the experimental diets

Ingredients, %	Starter-grower	Finisher
Yellow corn	54.00	65.70
Soybean meal (44%CP)	39.52	28.20
Limestone	0.92	0.92
Dicalcium phosphate	1.57	1.32
Vit+Min mix1	0.25	0.25
NaCl	0.25	0.25
DL-methionine	0.2	0.12
Commercial blend of oils	3.29	3.24
Total	100.0	100.0
Calculated values		
ME kcal/kg diet	2986	3121
Crude protein,%	22	18
Methionine,%	0.54	0.41
TSAA,%	0.91	0.73
Lysine,%	1.22	0.94
Ca,%	0.90	0.80
Available P, %	0.44	0.38

[1]Vitamins and minerals mixture provide per kilogram of diet: vitamin A (as all-trans-retinyl acetate); 12000 IU; vitamin E (all rac-α-tocopheryl acetate); 10 IU; k_3 3mg; Vit. D_3, 2200 ICU; riboflavin, 10 mg; Ca pantothenate, 10 mg; niacin, 20 mg; choline chloride, 500 mg; vitamin B_{12}, 10mμg; vitamin B_6, 1.5 mg; thiamine (as thiamine mononitrate); 2.2 mg; folic acid, 1 mg; D-biotin, 50mg. Trace mineral (milligrams per kilogram of diet) : Mn, 55; Zn, 50; Fe, 30; Cu, 10; Se, .1 and Ethoxyquin 3mg.

Materials and Methods

Birds, housing and management: This experiment was conducted at King Abdulaziz University; Faculty of Meteorology Environment and Arid land Agriculture. Unsexed Lohman broiler chicks were raised in floor pens under similar managerial and hygienic conditions. A mash starter-grower and finisher feed and water were offered *ad libitum* from tube feeders and automatic nipple drinkers, respectively with a twenty-four hours light program. Diets (Table 1) were formulated based on NRC (1994) tables of feedstuffs to meet nutrients requirements recommended for broilers. Chicks were randomly distributed into the experimental groups with keeping approximately similar initial live body weight. Number of dead birds was recorded throughout the experiment period from 7-43 days of age.

Factorial experimental design (3x4) was used to study the responses of modern broiler breeds to stocking density e.g. 10, 14 and 18 bird/m^2 and the efficacy of green tea, Multi enzymes mixture and their mixture to alleviate the stress resulted from increasing housing density. Chickens within each stocking density were fed four experimental diets; one of them was used as

Table 2: Effect of stocking density and addition of green tea, Multi enzymes or their mixture and their interaction on growth (g/bird/period) of broiler chicks from 7 to 43 days of age

Treatments		Body weight gains		
Bird density/m^2	Additive	7-21 days	22-43 days	7-43 days
10	Control	218.8	1262.9	1781.7
	Green tea	484.7	1281.1	1765.8
	Enzyme	195.4	1276.3	1771.7
	Mixture	487.6	1210.6	1698.2
14	Control	510.8	1272.1	1782.9
	Green tea	471.7	1237.4	1709.1
	Enzyme	504.6	1217.4	1722.0
	Mixture	478.5	1300.9	1779.4
18	Control	522.9	1192.1	1715.0
	Green tea	479.1	1255.5	1734.6
	Enzyme	524.3	1233.2	1757.5
	Mixture	475.2	1183.6	1658.7
	P value	NS	NS	NS
	SEM	12.93	30.04	35.09
Effect of stocking density				
10		496.6	1257.7[a]	1754.4
14		491.4	1257.0[a]	1748.4
18		500.4	1216.1[b]	1716.5
P value		NS	0.04	NS
SEM		6.46	15.01	17.52
Effect of feed additives				
Control		517.9[a]	1235.7	1753.5
Green tea		478.0[b]	1255.6	1733.5
Enzyme		510.9[a]	1238.2	1749.1
Mixture		479.3[b]	1229.1	1708.4
P value		0.00001	NS	NS
SEM		7.46	17.33	20.24

a-b means within the same column within the same treatment not having similar superscripts are significantly different (P<0.05), NS = non significant at 0.05.

control diet. While in the other three diets the control diet was supplemented with green tea (5g/kg feed) or commercial Multi enzymes (Optizyme® p-5; 1 g /kg feed), or with mixture of green tea (5g/kg feed) and Multi enzymes (1 g/kg feed). Therefore, there were 12 experimental treatments in which each treatment was replicated four times. The experimental period lasted from 7 to 43 days of age.

Chicks were weighed at 7, 21 and 43 days of age, whereas feed intake and FCR were calculated at 21 and 43 days of age. At day 43 of age, five female broilers were slaughtered from each treatment to determine carcass % and yields, internal organs and chemical composition of liver. Liver samples were chemically analyzed for moisture, CP, total lipids, and ash according to AOAC (1990) and the values for liver composition were expressed on fresh basis, also cholesterol content as (mg/100g fresh weight) of liver.

Blood samples were collected from the slaughtered birds in heparinized tubes. Plasma was separated by centrifugation at 3000 rpm for 10 minutes and stored at -18°C until analysis. Blood samples were collected from four birds of each treatment. Concentration of plasma total protein and total lipid were determined using commercial kits (Roche Diagnostics GmbH, D-68298 Mannheim, Germany), albumin (Doumas *et al.*, 1977), cholesterol (Sigma Diagnostics, procedure No. 401) and ALT and AST were determined (Retiman and Frankel, 1957). Whilst plasma globulin was calculated by subtracting albumin from total plasma protein.

Statistical analysis: Data of each trial were analyzed using 3×4 factorial design of the GLM procedure of SAS® (SAS Institute, 1985). Duncan's New Multiple Range Test (Duncan, 1955) was used to test mean differences at P£0.05.

Results and Discussion

Results indicated that there was no significant effect of the interaction between stocking density and addition of green tea and Multi enzymes on growth, feed intake and FCR during any tested period. However, it is clear that

Table 3: Effect of stocking density and addition of green tea, Multi enzymes or their mixture and their interaction on feed intake (g /bird/period) of broiler chicks from 7 to 43 days of age

Treatments		Feed intake		
Bird density/m^2	Additive	7-21	22-43	7-43
10	Control	893.3	2829.3	3722.7
	Green tea	822.3	3005.3	3827.7
	Enzyme	832.0	2795.3	3627.3
	Mixture	830.7	2686.7	3517.3
14	Control	862.0	2754.7	3616.7
	Green tea	863.7	2547.0	3410.7
	Enzyme	843.0	2670.3	3513.3
	Mixture	955.3	2893.7	3489.0
18	Control	1050.7	2421.0	3471.7
	Green tea	834.7	2504.0	3338.7
	Enzyme	964.7	2534.0	3498.7
	Mixture	880.0	2527.3	3407.3
	P value	NS	NS	NS
	SEM	59.4	167.7	182.4
Effect of stocking density				
10		844.6	2829.2[a]	3673.8
14		881.0	2716.4[ab]	3597.4
18		932.5	2496.6[b]	3429.1
P value		NS	0.03	NS
SEM		29.7	83.9	91.2
Effect of feed additives				
Control		935.3	2668.3	3603.7
Green tea		840.2	2685.4	
3525.7Enzyme		879.9	2666.6	3546.4
Mixture		888.7	2702.6	3591.2
P value		NS	NS	NS
SEM		34.2	96.7	105.2

a-b means within the same column within the same treatment not having similar superscripts are significantly different (P<0.05), NS = non significant at 0.05

Multi enzymes addition to broiler stocked at 18 bird/m^2 improved growth and FCR by 2.5 and 1.7%, respectively. This may be due to the effect of Multi enzymes in improving nutrient availability (Bedford, 1996; Jeroch *et al.*, 1995) that could be beneficial for broilers under stocking stress and increasing competition for feeds.

There was no significant effect of stocking density on growth, feed intake and FCR of broilers during 7-21, 21-43 and 7-43 days of age, except FI during 22-43 days of age, where chicks were kept under stocking density of 18 chicks/m^2 consumed significantly less feed than those kept at 10 chicks/m^2 (Table 3). It was found that broilers stocked at 18 bird/m^2 showed lower growth than those stocked at 10 or 14 bird/m^2. They also consumed less feed than those stocked at 10 bird/m^2. While those stocked at 14 bird/m^2 showed intermediate feed intake. Perhaps, this may be due to increasing competition for feed as a result of increasing stocking density. Also, Grashorn and Kutritz (1991) indicated high densities above 20 birds/m^2 could reduce that feed intake. The present results are in partial agreement with those reported by Cravener *et al.* (1992) who found that birds housed at 0.07, 0.09 and 0.11 m^2 per bird had similar 7-wk body weight, all being significantly higher than birds housed at 0.05m^2 per bird. On the other hand, there were no significant differences in FCR values.

The decrease in growth performance resulted from increasing stocking density could be attributed to the increase in stress resulted from competition for feeds and water, increased house temperature, microbial activity, and ammonia production. In this regard, Buckland *et al.* (1971); Proudfoot *et al.* (1979); Weaver *et al.* (1982); Reiter and Bessei (2000) found a linear decrease in growth of broilers at 5 wks of age with increasing stocking density and this was mainly due to increasing temperature between and underneath the birds. This partially agrees with the decrease in growth showed during later age of 22-43 days rather than early age of 7-21 days (Table 2). On the other hand, Parkhurst *et al.* (1977); Beremski (1987); Mizubuti *et al.* (1994); Al-Homidan (2001) revealed that there was no significant difference in broiler performance due to stocking density.

Table 4: Effect of stocking density and addition of green tea, Multi enzymes and their mixture and their interaction on feed conversion ratio (g feed/g gain) of broiler chicks from 7 to 43 days of age

Treatments		Feed conversion ratio			Number of dead
Bird density/m^2 birds	Additive	7-21	22-43	7-43	
10	Control	1.727	2.242	2.091	1.0
	Green tea	1.699	2.335	2.160	3.0
	Enzyme	1.684	2.196	2.050	3.0
	Mixture	1.704	2.222	2.073	3.0
14	Control	1.690	2.165	2.028	2.0
	Green tea	1.833	2.074	2.001	1.0
	Enzyme	1.677	2.193	2.041	4.0
	Mixture	1.992	2.228	2.163	3.0
18	Control	2.003	2.034	2.025	3.0
	Green tea	1.743	1.998	1.926	2.0
	Enzyme	1.842	2.056	1.991	4.0
	Mixture	1.851	2.132	2.051	5.0
	P value	NS	NS	NS	NS
	SEM	0.116	0.127	0.095	0.50
Effect of stocking density					
10		1.704	2.249	2.094	4.0
14		1.798	2.165	2.058	3.0
18		1.860	2.055	1.998	5.0
P value		NS	NS	NS	NS
SEM		0.058	0.064	0.048	0.249
Effect of feed additives					
Control		1.807	2.147	2.048	2.0
Green tea		1.759	2.136	2.029	3.0
Enzyme		1.734	2.148	2.027	4.0
Mixture		1.849	2.194	2.096	5.0
P value		NS	NS	NS	NS
SEM		0.067	0.073	0.055	0.286

NS = non significant at 0.05.

It seems therefore that the responses to stocking density could be primary affected by feed and water spaces and environmental condition within the house.

Green tea, Multi enzymes or their mixture as independent variables had no significant effects on the overall growth, feed intake and FCR of broiler chicks (Table 2, 3 and 4). The lack of enhancement effect of these agents may indicate that lack of synergisms between their mode of action and the stress caused by stocking density. However, Tollba (2003); Tollba and Hassan (2003) found that black cumin, garlic, thyme or fennel as natural feed additives to broiler diets under normal and high temperature conditions improved growth, FCR, and decreased mortality rate.

In Literature, green tea catechins are known for their pharmacological effects such as antioxidative (Lin *et al*., 1996), antimutagenic (Jain *et al*., 1989), anticarcinogenic (Sano *et al*., 1999), and anti-inflammatory effects (Varilek *et al*., 2001). In this regard, Miura *et al*. (2001) reported that ingestion of green tea improved growth of mice and did not affect plasma cholesterol and triglyceride concentrations. However, plasma peroxides were reduced indicating that *in vivo* oxidative state is improved by tea ingestion. Moreover, aortic cholesterol and triglyceride contents were 27 and 50% lower in tea group than in the control group, respectively. On the other hand, green tea resulted in no enhancing impact on growth performance of broiler chicks housed at different densities.

Carcass characteristics: There were significant interaction between stocking density and feed additives on dressing percentage, hind part either as absolute weight (g) or (%), and abdominal fat percentage (Table 5). Results indicate that green tea, Multi enzymes and their mixture improved dressing percentage of broilers housed at 10/m^2 by 7.9, 6.4 and 5.1%, respectively. Meanwhile, these additives had no enhancing effect on dressing percentage of birds housed at 14/m^2. On the other hand, green tea and mixture of green tea and Multi enzymes did improve dressing percentage by 2.4 and 3.2% of birds housed at 18 m^2 compared to their respective control, respectively.

It is clear that green tea, and Multi enzymes improved

Table 5: Effect of stocking density and addition of green tea, Multi enzymes and their mixture and their interaction on carcass characteristics of female broiler chicks slaughtered at 43 days of age

Treatments		Carcass weight (g)	Dressing, %	Front part, g	Front part, %	Hind part, g	Hind part, %	Wings weight, g	Wings, %	Abdominal fat, g	Abdominal fat,%
Bird density/m^2	Additive										
10	Control	1131.5	60.6	607.0	32.4	524.5	28.2	81.78	4.47	10.23	0.55
	Green tea	1237.8	65.4	656.8	34.7	581.0	30.7	77.00	4.08	26.03	1.43
	Enzyme	1306.3	64.5	689.8	34.6	616.5	30.9	83.00	4.16	18.2	0.91
	Mixture	1119.5	63.7	608.8	34.6	510.8	29.1	75.63	4.31	9.90	0.57
14	Control	1271.5	65.9	619.5	32.1	652.0	33.8	73.00	3.81	13.45	0.68
	Green tea	1143.8	65.1	616.0	35.1	527.8	30.1	70.05	4.00	18.25	1.04
	Enzyme	1261.5	63.2	701.5	35.1	560.0	28.1	82.70	4.14	10.30	0.51
	Mixture	1207.8	65.2	643.8	34.8	564.0	30.4	72.90	3.94	11.25	0.61
18	Control	1137.5	62.1	602.5	32.8	535.0	29.2	72.13	3.94	9.78	0.53
	Green tea	1173.8	63.6	641.5	34.8	532.3	28.8	73.20	3.97	7.50	0.40
	Enzyme	1138.8	61.4	616.5	33.2	522.3	28.1	75.90	4.09	14.18	0.77
	Mixture	1149.5	64.1	635.3	35.4	514.5	28.7	73.83	4.13	14.43	0.80
	P value	NS	0.06	NS	NS	0.02	0.001	NS	NS	NS	0.008
	SEM	55.8	1.15	33.0	0.86	27.06	0.81	3.24	0.20	2.85	0.16
Effect of stocking density											
10		1198.8	63.8^{ab}	640.6	34.1	558.2^{ab}	29.7^{ab}	79.4^{a}	4.26	16.1a	0.87
14		1221.1	64.9^{a}	645.2	34.3	575.9^{a}	30.6^{a}	74.7^{b}	3.97	13.3ab	0.71
18		1149.9	62.8^{b}	623.9	34.1	526.0^{b}	28.7^{b}	73.8^{b}	4.03	11.5b	0.62
P value		NS	0.05	NS	NS	0.04	0.008	0.04	NS	NS	NS
SEM		27.9	0.58	16.50	0.43	13.53	0.41	1.62	0.10	1.43	0.08
Effect of feed additives											
Control		1180.2	62.8	609.7^{b}	32.4^{b}	570.5	30.4	75.6^{ab}	4.07	11.15^{b}	0.59^{b}
Green tea		1185.1	64.7	638.1^{ab}	34.9^{a}	547.0	29.9	73.4^{b}	4.02	17.26^{a}	0.96^{a}
Enzyme		1235.5	63.3	669.3^{a}	34.3^{a}	566.3	29.1	80.5^{a}	4.13	14.23^{ab}	0.73^{ab}
Mixture		1159.0	64.4	629.3^{ab}	35.0^{a}	529.8	29.4	74.1^{b}	4.13	11.86^{b}	0.66^{b}
P value		NS	NS	NS	0.003	NS	NS	0.05	NS	0.05	0.04
SEM		32.3	0.66	19.1	0.50	15.64	0.47	1.87	0.12	1.65	0.09

a-b means within the same column within the same treatment not having similar superscripts are significantly different (P<0.05), NS = non significant at 0.05

absolute weight (g) of hind part of chickens housed at 10/m^2 by 10.8, and 17.5%, respectively. While green tea, Multi enzymes and their mixture improved relative weight of hind part by 8.9, 9.6 and 3.2%, respectively. Meanwhile, green tea, Multi enzymes and their mixture did not improve weight of hind part (g) or (%), but on the contrary even though unexplained negative effect were noticed in birds housed at 14 or 18/m^2 (Table 5).

The interaction between stocking density and feed additives indicated that green tea increased abdominal fat % by 160 and 52.9% of birds housed at 10 and 14 /m^2 compared to their respective controls, respectively. While, it was decreased by 24.5% of birds housed at 18/m^2 compared to its respective control (Table 5). Meanwhile, Multi enzymes increased abdominal fat % of birds stocked at 10 and 18/m^2 by 65.5 and 45.3%, respectively. While it was decreased by 25% of groups stocked at 14/m^2. On the other hand, mixture of green tea and Multi enzymes increased abdominal fat % of group stocked at 18/m^2 by 50.9%, while it had a neglect able impact on groups stocked at 10 and 14/m^2.

There were significant effect of stocking density on dressing percentage, hind part weight (g) and (%) in which these criteria were decreased when broilers were stocked at 18/m^2 compared to those of birds stocked at 14/m^2. On the other hand, broilers stocked at 10//m^2 showed intermediate dressing percentage and hind part weight (g) and (%). These results agree with those reported by Cravener *et al.* (1992) who found stocking density at 0.05 m^2/bird decreased carcass weight significantly compared to those stocked at 0.07, 0.09 and 0.11 m^2 per bird.

Results showed that birds stocked at 10/m^2 had significantly higher abdominal fat (%) than those stocked at 18/m^2, meanwhile those stocked at 14/m^2 showed intermediate value. Cravener *et al.* (1992) found similar results. In brief, the changes in dressing (%), hind part weight (g) and (%) and abdominal fat (%) as result of changes in stocking density may reflect the impact of the stress resulted from increasing stocking density on nutrient metabolism and/or portioning.

Data in Table 5 showed that feed additives as an independent variable had significant effect on front part weight (g) and (%), and wings weight (g) as well as abdominal fat weight (g) and (%). Results indicate that Multi enzymes increased significantly front weight (g) and (%) compared to the control group. This may indicate an increase in nutrient availability due to Multi

Table 6: Effect of stocking density and addition of green tea, Multi enzymes and their mixture and their interaction on body organs of female broiler chicks slaughtered at 43 days of age

Treatments		Liver weight, g	Liver, %	Heart weight, g	Heart, %	Gizzard weight, g	Gizzard, %	Spleen weight g	Spleen, %	Pancreas, g	Pancreas, %	Intestinal g	Intestinal %
Bird density/m^2	Additive												
10	Control	37.4	2.05	9.95	0.55	37.15	2.05	2.23	0.123	5.08	0.280	99.25	5.37
	Green tea	43.3	2.28	11.18	0.59	34.53	1.83	2.00	0.108	5.60	0.294	104.2	35.51
	Enzyme	39.7	1.99	8.23	0.41	35.93	1.80	1.15	0.058	5.45	0.274	89.8	34.52
	Mixture	40.0	2.28	10.35	0.59	35.13	2.01	3.03	0.173	4.78	0.275	84.55	4.83
14	Control	39.3	2.05	12.25	0.64	36.90	1.92	2.30	0.120	5.65	0.295	102.9	55.38
	Green tea	36.4	2.07	8.65	0.49	34.88	1.98	2.23	0.126	3.98	0.227	88.28	5.02
	Enzyme	41.7	2.08	10.35	0.52	36.25	1.81	3.00	0.151	5.05	0.253	94.95	4.75
	Mixture	46.2	2.49	8.60	0.46	36.98	2.00	1.65	0.089	5.30	0.287	80.50	4.38
18	Control	38.6	2.09	10.23	0.55	36.55	2.01	2.65	0.145	3.93	0.215	87.15	4.80
	Green tea	36.3	1.97	9.78	0.53	31.85	1.72	2.23	0.120	5.28	0.287	95.48	5.19
	Enzyme	37.0	1.99	8.88	0.48	39.85	2.15	2.83	0.151	6.13	0.328	93.98	5.07
	Mixture	40.8	2.27	8.23	0.46	31.33	1.74	2.10	0.118	4.33	0.240	76.58	4.26
	P value	NS	NS	0.04	NS	NS	NS	0.0001	0.0001	NS	NS	NS	NS
	SEM	2.86	0.14	0.82	0.045	2.11	0.132	0.255	0.015	0.59	0.032	5.81	0.332
Effect of stocking density													
10		40.1	2.15	9.93	0.54	35.7	1.92	2.10	0.12	5.23	0.281	94.5	5.05
14		40.9	2.17	9.96	0.53	36.3	1.93	2.29	0.12	4.99	0.266	91.7	4.88
18		38.2	2.08	9.28	0.51	34.9	1.91	2.45	0.13	4.91	0.267	88.2	4.83
P value		NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
SEM		1.44	0.07	0.41	0.023	1.05	0.066	0.128	0.075	0.29	0.016	2.91	0.166
Effect of feed additives													
	Control	38.4	2.07b	10.81a	0.58a	36.9	1.99	2.39	0.129	4.88	0.263	96.5^{a}	5.18^{a}
	Green tea	38.7	2.11b	9.87ab	0.54ab	33.8	1.85	2.15	0.118	4.95	0.269	96.0^{a}	5.24^{a}
	Enzyme	39.4	2.02b	9.15b	0.47 b	37.3	1.92	2.33	0.120	5.54	0.285	92.9^{a}	4.78^{ab}
	Mixture	42.3	2.35a	9.06b	0.51ab	34.5	1.92	2.26	0.127	4.80	0.267	80.5^{b}	4.49^{b}
	P value	NS	0.03	0.04	0.04	NS	NS	NS	NS	NS	NS	0.006	0.03
	SEM	1.65	0.08	0.47	0.026	1.22	0.076	0.147	0.087	0.34	0.018	3.36	0.192

a-b means within the same column within the same treatment not having similar superscripts are significantly different ($P<0.05$), NS = non significant at 0.05.

Table 7: Effect of stocking density and addition of green tea, Multi enzymes and their mixture and their interaction on chemical composition of liver of female broiler chicks slaughtered at 43 d of age

Treatments		Moisture, %	CP, %	EE, %	Cholesterol mg/100 g	Ash, %
Bird density/m^2	Additive					
10	Control	74.2	68.7	21.6	66.2	6.23
	Green tea	74.2	67.6	22.8	64.1	6.00
	Enzyme	74.5	67.6	22.6	63.8	6.20
	Mixture	73.8	67.4	23.0	65.9	5.86
14	Control	73.7	68.5	21.7	66.8	6.12
	Green tea	74.0	68.6	21.7	63.2	6.09
	Enzyme	74.7	67.7	22.6	65.9	5.94
	Mixture	73.9	67.3	23.1	64.5	6.15
18	Control	74.2	67.5	22.4	68.3	6.27
	Green tea	74.5	68.0	22.9	66.4	6.35
	Enzyme	74.4	68.6	21.5	62.2	6.02
	Mixture	74.6	67.8	22.7	59.9	5.92
	P value	NS	NS	NS	NS	NS
	SEM	0.24	0.41	0.39	0.55	0.243
Effect of stocking density						
10		74.1	67.8	22.5	65.0	6.07
14		74.1	68.0	22.3	65.1	6.07
18		74.4	68.0	22.4	64.2	6.14
P value		NS	NS	NS	NS	NS
SEM		0.12	0.21	0.19	0.28	0.122
Effect of feed additives						
	Control	74.0^{b}	68.3	22.9^{a}	67.1	6.21
	Green tea	74.2^{ab}	68.1	22.5^{ab}	64.6	6.15
	Enzyme	74.5^{a}	68.0	22.2^{b}	64.0	6.05
	Mixture	74.1^{ab}	67.5	22.9^{a}	63.4	6.00
	P value	0.08	NS	0.02	NS	NS
	SEM	0.14	0.24	0.23	0.34	0.141

a-b means within the same column within the same treatment not having similar superscripts are significantly different P<0.05), NS = non significant at 0.05

enzymes addition (Makled, 1993; Jeroch *et al*., 1995) Meanwhile, green tea and mixture of green tea and Multi enzymes improved significantly front weight (%) compared to the control group, with no significant difference between different feed additives in % and weight (g) of front part. It was observed that wings weight (g) of Multi enzymes supplemented group was significantly higher than that of those supplemented with green tea or mixture of green tea and Multi enzymes, which in turn showed no significant difference between them. On the other hand, wings weight (g) of feed additives supplemented groups was similar to that of the control group. Results showed that green tea supplementation resulted in unexplained significant increase in weight (g) and (%) of abdominal fat compared to the control group and those supplemented with mixture of green tea and Multi enzymes (Table 5). On the other hand, abdominal fat weight (g) and (%) of group fed Multi enzymes supplemented diet were not significantly different from those of the other experimental groups.

Internal organs: Results displayed in Table 6 indicate that there were no significant interaction between stocking density and feed additives in the most of the studied traits except for a significant interaction between stocking density and feed additives on heart weight (g) and spleen weight (g) and (%). Results revealed that green tea and its mixture with Multi enzymes increased heart weight (g) of broilers housed at 10/m^2 by 12.4 and 4.0%, respectively. Meanwhile; green tea, Multi enzymes or their mixture decreased heart weight (g) of groups stocked at 14 or 18 bird/m^2.

Results indicate that spleen weight (g) and (%) were decreased considerably due to Multi enzymes addition to groups stocked at 10/m^2, while the contrary was shown when Multi enzymes added to group stocked at 14/m^2, while, Multi enzymes had negligible impact on group stocked at 18/m^2. Green tea addition resulted in a considerably decrease in spleen weight (g) and (%) of broilers stocked at 10 and 18/m^2, while it had negligible impact on group stocked at 14/m^2. A mixture of green tea and Multi enzymes increased spleen weight

Table 8: Effect of stocking density and addition of green tea, Multi enzymes and their and Multi enzymes and their interaction on plasma constituents of female broiler chicks slaughtered at 43 d of age

Treatments		Total Protein	Albumin	Globulin	Total lipid	Cholesterol	AST	ALT
Bird density/m^2	Additive	(g/dl)	(g/dl)	(g/dl)	(g/l)	(mg/dl)	(u/dl)	(u/dl)
10	Control	4.85	2.43	2.42	7.10	170.8	22.5	11.7
	Green tea	5.26	2.64	2.62	7.11	164.5	23.2	11.7
	Enzyme	5.23	2.52	2.71	7.19	167.7	21.1	11.9
	Mixture	4.93	2.40	2.52	7.07	162.2	23.2	11.4
14	Control	5.02	2.60	2.42	7.37	168.2	23.2	11.1
	Green tea	5.32	2.65	2.67	7.26	171.6	21.9	11.6
	Enzyme	4.81	2.42	2.39	7.33	167.1	22.4	11.6
	Mixture	5.11	2.53	2.58	7.03	156.9	21.6	12.7
18	Control	4.64	2.33	2.30	7.18	163.7	21.4	11.8
	Green tea	5.02	2.47	2.55	7.13	171.6	?23.1	11.8
	Enzyme	4.76	2.45	2.31	7.07	165.4	22.6	11.7
	Mixture	5.12	2.51	2.61	6.78	164.4	21.4	11.8
	P value	NS	NS	NS	NS	NS	0.01	NS
	SEM	0.275	0.118	0.186	0.303	7.49	0.56	0.42
Effect of stocking density								
10		5.07	2.50	2.57	7.12	166.3	22.5	11.6
14		5.06	2.55	2.51	7.25	165.9	22.3	11.8
18		4.88	2.44	2.44	7.04	166.3	22.1	11.8
P value		NS	NS	NS	NS	NS	NS	NS
SEM		0.138	0.059	0.093	0.152	3.75	0.28	0.21
Effect of feed additives								
	Control	4.83	2.45	2.38	7.22	7.22	22.4	11.5
	Green tea	5.20	2.58	2.61	7.17	7.17	2.7	11.7
	Enzyme	4.93	2.46	2.47	7.20	7.20	22.0	11.7
	Mixture	5.05	2.48	2.57	6.96	6.96	22.1	12.0
	P value	NS	NS	NS	NS	NS	NS	NS
	SEM	0.159	0.068	0.108	0.176	4.33	0.31	0.25

[a-b] means within the same column within the same treatment not having similar superscripts are significantly different (P<0.05), NS = non significant at 0.05

(g) and (%) of group stocked at 10/m^2, while the contrary was shown of groups stocked at 14 or 18/m^2.

Irrespective of feed additives, there was no significant effect of stocking density on the internal organs, showing that internal organs were developed normally under different densities for rearing broilers.

Feed additives as an independent variable had significant effect on liver (%), heart weight (g) and (%), and intestinal weight (g) and (%). Results revealed that mixture of green tea and Multi enzymes increased significantly liver (%) compared to the control diet without or with green tea or Multi enzymes (Table 6). The increase in liver (%) may indicate fat accumulation due to addition of mixture of green tea and Multi enzymes.

Multi enzymes supplementation resulted in significantly smaller heart weight (g) and (%) than that of the control group. Also, only heart weight (g) of group fed mixture of green tea and Multi enzymes was smaller than that of the control group. On the other hand, green tea supplemented group exhibited intermediate heart weight (g) and (%).

Regardless of stocking density, results showed that intestinal weight (g) and (%) were significantly affected by feed additives. It is obvious that, mixture of green tea and Multi enzymes significantly decreased intestinal weight (%) compared to the control group without or with green tea or Multi enzymes supplementation, and intestinal weight (%) compared to the control group without or with green tea supplementation. The decrease in intestinal weight and percentage of group fed diet supplemented with mixture of green tea and Multi enzymes may explain the insignificant decrease in growth of this group and may indicate an alternation in the intestinal function, Pancreas weight (g) and (%) were not significantly affected (Table 6).

Chemical composition of liver: Obviously, there were no negative effect of stocking density on percentage moisture, protein, ether extract, ash and cholesterol (mg/100 g) of liver, nor were there significant interaction between stocking density and feed additives on these criteria (Table 7).

Feed additives as independent variable had significant effects on percentage moisture and ether extract of liver. Results indicated that Multi enzymes addition increased percentage moisture and decreased ether extract of liver compared with the control group. The increase in ether extract of liver was not connected with either liver weight or percentage nor with abdominal fat weight or percentage. It should be mentioned however, that green tea had no significant effect on percentage moisture, CP, EE, cholesterol and ash of liver (Table 7).

Blood plasma constituents: There were no significant impacts of stocking density and feed additives or their interaction on the most of the plasma constituents except for plasma AST activity (Table 8). Results indicated that green tea addition to the diet for birds stocked at 10 and 18/m^2 increased the activity of plasma AST compared to the control group, while the opposite trend was shown when it was added to broilers stocked at 14/m^2. Enzyme addition decreased plasma AST activity of group housed at 10 and 14 birds/m^2, however the effect was more pronounced of group housed at 10/ m^2 than those housed at 14 chicks/m^2 (6.2 vs.3.4%). On the other hand, enzyme addition to broilers housed at 18/m^2 increased plasma AST activity by 5.6%.

Addition of the mixture of green tea and Multi enzymes to broilers housed at 10 birds/m^2 increased plasma AST activity , whilst decreased it of group stocked at 14 birds/ m^2, and this essentially parallel the effect of green tea on plasma AST activity of these groups (Table 8).

The effect of green tea on the activity of plasma AST could be attributed to its antioxidative (Lin et al., 1996).

The present results agree with those reported by Miura *et al.* (2001) who reported that ingestion of green tea did not affect plasma cholesterol and triglyceride concentrations. However, plasma peroxides were reduced indicating that *in vivo* oxidative state is improved by tea ingestion. Moreover, aortic cholesterol and triglyceride contents were 27 and 50% lower in tea group than in the control group, respectively. Also, Attia *et al.* (2001) found that Multi enzymes addition to broilers diet had no adverse effects on plasma constituents.

It is clear that stocking density and different types of additives used herein had no negative effects on plasma constituents of broiler chicks and liver functions as judged by plasma ALT and AST during the experimental period.

In conclusion broilers chicks could be stocked up to 14 bird/m^2 without adverse effect on growth performance, while addition of green tea, Multi enzymes or their mixture had no enhancing effect on growth performance of broiler stocked at different densities herein.

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