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# EFFECT OF FEED WITHDRAWAL AND PROGUT ON BROILER PERFORMANCE, CARCASSES TRAITS AND BLOOD PARAMETERS

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## ABSTRACT

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### Keywords

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This study evaluated the effect of feed withdrawal without/ with feed additive (Progut®) on broiler chickens' performance, carcass traits and some blood parameters, in a factorial arrangement (4 feed restriction×2 levels of feed additive). Three hundred twenty-one-day-old broiler chicks (Cobb 500) were distributed into 8 treatments with four replicates. In the first week all chicks fed starter basal diet, however during the second week, broiler chicks were exposed to feed restriction by feed withdrawal time (0, 6, 9, 12 hours/day). Broiler chicks exposed to feed withdrawal had low body weight gain and feed consumption at 2 weeks of age. Feed withdrawal had no effect on final weight, body weight change, total feed intake, total feed conversion, economic efficiency, carcass traits and serum blood composition of 42-day-old-broiler chicks. Feed withdrawal and feed additive in broiler diets decreased total microflora counts and *E. coli* and enhanced *lacto Bacillus*, amylase and chemo trypsin in broiler guts. Feed additive (Progut®) decreased total feed consumption, improved total feed conversion and increased economic efficiency of broilers. Feed additive increased the level of  $\beta$  globulin and decreased the level of  $\alpha$  globulin in serum blood of broiler chicks. The obtained results suggested that feed withdrawal during the second week of age had not effect on broiler performance, carcass traits and serum blood characteristics and improved *lacto Bacillus*, amylase and chemo trypsin. Feed additive (Progut®) in broiler diets improved growth performance and decreased total bacteria counts, *E. coli* and increased *lacto bacillus* counts in broiler guts.

**Contribution/Originality:** Feed withdrawal reduces feed consumption, feed costs and abdominal fat, improves feed utilization and benefits from compensatory growth. Yeast improves health of the bird, reduces using antibiotics and improves the quality of the carcass. Feed withdrawal was combined with yeast product to improve growth performance of broilers and their carcasses.

## 1. INTRODUCTION

Rising feed costs drive the interest in interventions such as feed restriction to improve feed efficiency and reduce abdominal fat content [1] reported there are 2 main methods to apply feed restriction, each with their own effect on production performance. Qualitative feed restriction is defined as limiting (specific) nutrient intake through dilution of the diet. Food deprivation is a commonly used management strategy in broilers, aiming to prevent excessive weight gain during growth and thereby to solve some health-related problems [2] while also preventing precocious fat deposition [3]. Feed restriction also presents some economic benefits [4].

Quantitative feed restriction is defined as reducing nutrient intake through reducing the amount of feed consumed. Following a period of feed restriction, broilers normally experience a period of rapid growth, called compensatory growth [5]. The extent to which broilers show compensatory growth depends on many factors such as environment, period and method of the applied restriction, strain, and sex [6]. In addition, [7] showed that chickens provided 80% of ad libitum intake from 8 to 16 d of age do not differ in body weight, feed conversion ratio, or fat content at 35 d of age compared to *ad libitum* fed chickens. Yeast (*Saccharomyces cerevisiae*) and yeast cell wall (YCW) have been reported to contain polysaccharides and thus may have the potentials to improve the performance and health of birds [8]. The growth-promoting effect and immunomodulatory potential of prebiotic yeast, YCW products and their cell wall contents such as  $\beta$ -glucan and  $\alpha$ -mannan in recent years are gaining research interest [9]. Live yeast (LY) is one of the most potential microorganism derived products that can be used in animal ration as a dietary supplement [10] as well as a potential alternative to feed antibiotics in animals [11]. Mannan oligosaccharide, derived from the outer cell wall of yeast (*Saccharomyces cerevisiae*), can be applied as a growth promoter as well as a possible alternative to antibiotics in broiler diets [12]. Dietary supplementation with whole yeast and yeast cell walls at 1.5–2 g/kg level could improve growth performance and meat yield in broilers [13]. Progut® has successfully passed through different types of trials: In vitro trials: In laboratory trials Progut® have shown its ability to prevent *E. coli* attachment to gut mucus, to modify intestinal microbiotic and stimulate immunity. Progut® has consistently demonstrated beneficial effects on intestinal microbiotic and immunity with different animal species. Progut® in poultry feeds has led to improved vitality, feed utilization, better productivity and growth. Better intestinal health and immunity improves performance and profitability in poultry production.

Therefore, this study was performed to investigate the effect of feed restriction and feed additive (Progut®) on broiler growth performance, carcass traits and serum blood characteristics.

## 2. MATERIALS AND MEHTODES

This experiment was done at Fac. of Agric., Mans. Univ., throughout September and October 2021. This experiment was conducted to evaluate the effects of four-time feed withdrawal at the second week of age (0, 6, 9, 12 hours/day) with or without feed additive (Progut®) on broiler performance, carcass traits and some blood parameters.

This experiment was designed in factorial arrangement (4×2). Three hundred twenty unsexed one-day-old Cobb-500 broiler chicks were randomly assigned to eight experimental treatments with four replicates each (10 birds in each replicate). All chicks were fed the starter (0-3 weeks of age) and grower-finisher diets (3-6 weeks of age). The experimental diets were offered to chicks *ad libitum* during the experimental period (six weeks of age) except during the second week of age which feed withdrawal was done. The experimental treatments 1, 3, 5 and 7 fed the basal diets without additive, however Treatments 2, 4, 6 and 8 fed basal diets supplements with 1g Progut/kg in starter diet and 0.5g/kg in grower-finisher diet. Diets were formulated using feedstuffs analyses tables of NRC National Research Council [14]. Table 1 shows feed component and calculated analysis of the experimental diets.

Each 3 kg premix contains: Vit. A, 10000000 IU; vit. D<sub>3</sub>, 2000000 IU; vit. E, 10000 mg; vit. K, 1000 mg; vit. B<sub>1</sub>, 1000 mg; vit. B<sub>2</sub>, 5000 mg; vit. B<sub>6</sub>, 1500 mg; vit. B<sub>12</sub>, 10 mg; folic acid, 1000 mg; biotin, 50 mg; pantothenic acid, 10000 mg; nicotinic, 30000 mg; Fe, 30000 mg; Mn, 60000mg; Zn, 50000 mg; I, 300mg; Co, 100mg; Cu, 4000 mg; Se, 100 mg and CaCO<sub>3</sub> up to 3000 g.

Feed additive (Progut®) was new generation yeast product.

Dose of broilers: Starter: 1 kg/T; finisher: 0.5 kg/T.

Table 1. Composition of experimental diets of broiler chicks.

Ingredients	Starter diet	Grower-Finisher diet
Yellow corn	57.5	65.2
Soybean meal	25.6	21.0
Corn gluten	10.5	8.0
Sun-flower meal oil	1.5	1.0
Di-Calcium Phosphate	1.8	1.8
Limestone	2	2
Salt	0.3	0.3
Premix	0.3	0.3
Methionine	0.1	0.1
Lysine	0.4	0.3
Total	100	100
Calculated analysis (As Fed Basis: [14])		
Metabolizable energy (ME), kcal/kg	3050	3061
Crude protein (CP), %	23.1	20.1
Ether extract (EE), %	4.15	3.85
Crude fiber (CF), %	3.19	3.01
Total Phosphorus %	0.72	0.70
Available Phosphorus%	0.47	0.46
Calcium, %	1.22	1.21
Methionine, %	0.52	0.46
Methionine + Cystine,%	0.91	0.81
Lysine, %	1.62	1.05

### 2.1. Chicken Growth Performance

Chicken's live body weight (LBW) and feed intake (FI) were weekly recorded at replicate basis. Body weight gain (BWG) and feed conversion ratio (FCR) were calculated. The cumulative means of LBW, FI, BWG, and FCR were calculated for the whole experimental period (0-42 days of age).

### 2.2. Carcass traits of Broiler Chicks

At the end of feeding trial (42 days of age), 5 chicks were randomly chosen from each treatment to perform slaughter test. Feed was withdrawn for 8 h. before slaughtering. Individual LBW of birds was recorded immediately before slaughtering. Carcass traits were recorded. Procedures for cleaning out were performed on the hot carcasses. Weights of carcass yield (CY) and edible organs were determined and expressed as a percentage of live body weight at slaughter.

### 2.3. Serum Blood Parameters

Five chicks from each treatment at 42 days of age were chosen to collect 5 serum blood samples. Blood serum was separated by centrifugation process at 3000 rpm for 15 minutes. Serum concentrations of total protein, Albumin, Globulin, Total lipids, Triglycerides, cholesterol, were measured by commercial kits (commercial kits: Spectrum Diagnostic kits S.A.E., Egyptian company of biotechnology, 2016).

### 2.4. Estimation of Humoral Immune Response

(IgG) Immunoglobulin G, (IgM) Immunoglobulin M, (IgA) Immunoglobulin A were measured by commercial kits in blood serum of chicks (commercial kits: Spectrum Diagnostic kits S.A.E., Egyptian company of biotechnology, 2016).

## 2.5. Statistical Analysis

Statistical processing of results was performed by using two-way analysis of variance of the general linear model (GLM) procedure of the SAS [15]. The significant differences between treatment means were separated by Tukey's Multiple Range-test ( $P < 0.05$ ). The following statistical model was used:  $Y_{ij} = \mu + F_i + A_j + FA_{ij} + e_{ij}$ . Where:  $Y_{ij}$  = observed traits;  $\mu$  = the overall mean;  $F_i$  = effect of feed restriction;  $i = (0, 1, 2 \text{ and } 3)$ ;  $T_j$  = effect of additive (Progut®);  $j = (1 \text{ and } 2)$ ;  $ET_{ij}$  = effect of interaction between feed restriction and additive Progut®;  $e_{ij}$  = experimental random error.

## 3. RESULTS AND DISCUSSION

### 3.1. Growth Performance

#### 3.1.1. Live Body Weight of Chicks

Table 2 showed final body weight (FBW) of chicks and total body weight gain were not affected by feed withdrawal or feed additive (Progut®) and the interaction between them. However, feed withdrawal and feed additive were enhanced numerically final body weight and body weight gain. Broiler chicks exposed feed withdrawal 12 hours/day during the 2<sup>nd</sup> week was decreased LBW compared with the group exposed feed withdrawal 6 hours/day. The interaction between feed withdrawal and feed additive did not affected in LBW at all the experimental periods and total weight gain. Our results agree with those of Van Der Klein, et al. [16] who showed that FBW on broiler chicks did not significantly affected by feed restriction. Jang, et al. [17] demonstrated that exposed broiler chicks to feed restriction had positive compensatory on growth at the total period. Khetani, et al. [6] reported that limited time feed of broiler chicks did not significantly affected on FBW compared with the control group. Tolcamp, et al. [18] found that FBW of broiler chicks was not different by used limited time feed due to feed restriction has associated with compensatory growth and enhanced FCR.

**Table 2.** Effects of feed withdrawal and feed additive (Progut®) on live body weight (kg) of broiler chick.

Treatments	Weeks of age							Total body weight gain, kg 0-6 wks
	0	1	2	3	4	5	6	
Feed withdrawal								
A1 (0 h)	0.046	0.143	0.352 <sup>ab</sup>	0.616	1.101	1.465	1.891	1.845
A2 (6 h)	0.045	0.146	0.358 <sup>a</sup>	0.641	1.080	1.511	1.977	1.932
A3 (9 h)	0.045	0.146	0.335 <sup>ab</sup>	0.618	1.048	1.499	1.986	1.940
A4 (12 h)	0.045	0.152	0.327 <sup>b</sup>	0.605	1.015	1.416	1.915	1.870
SEM	0.001	0.003	0.008	0.018	0.024	0.033	0.040	0.040
P value	0.127	0.123	0.027	0.553	0.095	0.188	0.288	0.283
Feed additive								
B1	0.045	0.149	0.333 <sup>b</sup>	0.646 <sup>a</sup>	1.041	1.445	1.902	1.86
B2(Progut)	0.045	0.144	0.353 <sup>a</sup>	0.594 <sup>b</sup>	1.082	1.500	1.982	1.94
SEM	0.001	0.002	0.005	0.013	0.017	0.023	0.029	0.029
P value	0.810	0.086	0.013	0.008	0.103	0.104	0.058	0.057
Interactions AB								
A1×B1	0.046	0.145	0.347	0.673	1.101	1.453	1.855	1.809
A1×B2	0.046	0.141	0.357	0.558	1.101	1.476	1.926	1.880
A2×B1	0.045	0.148	0.351	0.657	1.054	1.468	1.920	1.876
A2×B2	0.045	0.145	0.364	0.625	1.105	1.555	2.034	1.988
A3×B1	0.045	0.146	0.320	0.636	1.019	1.451	1.922	1.877
A3×B2	0.045	0.146	0.349	0.601	1.077	1.548	2.049	2.004
A4×B1	0.046	0.158	0.312	0.617	0.988	1.409	1.910	1.864
A4×B2	0.045	0.146	0.342	0.593	1.043	1.422	1.920	1.875
SEM	0.001	0.004	0.011	0.026	0.034	0.046	0.057	0.057
P value	0.569	0.453	0.686	0.269	0.812	0.726	0.738	0.741

**Note:** A1 = 0 feed withdrawal time at the 2<sup>nd</sup> week of age, A2 = 6 feed withdrawal time at the 2<sup>nd</sup> week of age, A3 = 9 feed withdrawal time at the 2<sup>nd</sup> week of age and A4 = 12 feed withdrawal time at the 2<sup>nd</sup> week of age.

B1 = without feed additive and B2 = with feed additive.

However, Zukiwsky, et al. [19] found that broiler exposed feed restriction had significantly increased FBW of broiler chicks compared with control group. Also, Abaseid, et al. [20] reported that feed withdrawal during 4<sup>th</sup> and 5<sup>th</sup> weeks of age had increased in FBW compared with control group. On the other hand, Orso, et al. [21] reported that FBW of broiler chicks was decreased due to feed restriction.

Our results agree with those of Aristides, et al. [22] who observed that broiler growth performance did not effect by used yeast in broiler diets. Yalçın, et al. [23] reported that broiler fed diets containing yeast was not affected on FBW of broiler chicks. However, He, et al. [24] found that used yeast in broiler diets was improved FBW compared with control group. Also, Sun, et al. [25] found that broiler FBW increased by supplemented yeast in broiler diets.

### 3.2. Feed Intake, Feed Conversion Ratio and Economic Efficiency of Broiler Chicks

Data in Table 3 showed broiler chicks exposed to feed withdrawal during the 2<sup>nd</sup> week (9, 12 hours/day) was decreased FI compared with the control group in this week. Feed additive was decreased FI for the 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> weeks of age and total feed intake. Where broiler chicks diets supplemented with feed additive (Progut®) were significantly improved total FCR compared with the control group. Feed additive was significantly increased economic efficiency compared with that of the control group. The interaction between feed withdrawal and feed additive did not affect in FI at all experimental periods, FCR and economic efficiency.

Our results partially agree with those of Shafiei, et al. [26] who found that used feed withdrawal time (6, 8, 10, 12 hours/day) had decreased FI of broiler chicks at 2<sup>nd</sup> week of age but did not affect in FCR. Van Der Klein, et al. [16] showed that FCR of broiler chicks did not significantly affected by feed restriction. Abaseid, et al. [20] reported that feed withdrawal during the 4<sup>th</sup> and 5<sup>th</sup> weeks of age had not effect on TFI compared with control group however; FCR was improved by feed withdrawal of broiler chicks. Novel, et al. [27] found that FCR of broiler chicks did not significantly affected by reared broiler under feed restriction at levels (50% and 25%).

On the contrary, Zukiwsky, et al. [19] found that broiler exposed feed restriction had significantly improved FCR of broiler chicks compared with control group. Orso, et al. [21] reported that FI of broiler chicks was decreased and FCR was improved compared with control by feed restriction. Butzen, et al. [7] determined that exposed broiler chicks to 20 % feed restriction at 8-16 day of age had improved FCR of broiler. Also, Romero, et al. [28] found that reared broiler under feed restriction had decreased FI on broiler. Urdaneta-Rincon and Leeson [29] reported that the improvement in economic efficiency by limited time feed on broiler chicks due to compensatory growth of broiler chickens and improved FCR. Lippens, et al. [30] showed that used feed withdrawal of broiler chicks improved FCR resulted improved in economic efficiency associated with decreased feed cost.

Our results agree with those of Ahiwe, et al. [13] who found that broiler fed yeast (1.5 to 2g/kg diets) was significant improved FCR. Ding, et al. [31] reported that yeast supplemented in broilers diets was improved FCR. Sousa, et al. [10] showed that FI did not affected by used yeast (6%) in broiler diets but improved broiler FCR. Also, Sun, et al. [25] observed that broiler FI decreased and improved FCR by supplemented yeast in broiler diets. Haldar, et al. [32] found that used yeast in broiler diets improved FCR in broiler. In this meaning, Zhang, et al. [33] and Spring, et al. [34] reported that the improvement in FCR in broiler fed yeast in diets due to improving the digestibility of nutrients resulted by live yeast cell wall could alter the gastrointestinal microorganism to beneficial organisms.

However, Cabuk, et al. [35] showed that broiler diets supplemented with yeast did not affected on FI and FCR compared with control group.

### 3.3. Carcass Traits

Table 4 showed the effects of feed withdrawal and feed additive on carcass traits of broiler chicks (Cobb 500). Carcass weight and parts percentages of broiler chicks were not affected by reared under feed withdrawal during

the second week of age. Feed additive in broiler diets was decreased on liver and goblets percentage compared with the control. Also, the interaction between feed withdrawal and feed additive were no significant differences in carcass parts percentages.

Our results agree with those of Zukiwsky, et al. [19] who observed that broiler exposed feed restriction had no effect on carcass parts percentage compared with control group. Sherif and Mansour [36] observed that broiler reared under feed withdrawal did not effected on carcass parts percentage. Van Der Klein, et al. [16] showed that carcass and parts percentage on broiler chicks did not significantly affected by feed restriction. David and Subalini [37] who reported that carcass and giblets percentages of broiler chickens did not affected by feed withdrawal (3, 5 and 7 hours daily).

**Table 3.** Effects of feed withdrawal and feed additive (Progut®) on feed intake (kg), total feed conversion ratio and economic efficiency (%) of broiler chick.

Treatments	Feed intake/weeks of age							Total Feed conversion ratio	Economic efficiency
	1	2	3	4	5	6	TFI		
Feed withdrawal									
A1 (0 h)	0.122	0.299 <sup>a</sup>	0.490	0.744	0.927	1.083	3.663	1.991	129.125
A2 (6 h)	0.125	0.300 <sup>a</sup>	0.502	0.738	0.946	1.106	3.716	1.931	135.625
A3 (9 h)	0.120	0.265 <sup>b</sup>	0.504	0.753	0.966	1.154	3.761	1.943	134.250
A4 (12 h)	0.126	0.256 <sup>b</sup>	0.507	0.710	0.930	1.085	3.612	1.935	135.000
SEM	0.0045	0.0061	0.0094	0.0166	0.0145	0.0237	0.0496	0.0264	3.030
P value	0.767	0.001	0.596	0.315	0.227	0.152	0.195	0.365	0.427
Feed additive									
B1	0.125	0.301 <sup>a</sup>	0.524 <sup>a</sup>	0.775 <sup>a</sup>	0.954	1.102	3.779 <sup>a</sup>	2.041 <sup>a</sup>	125.313 <sup>b</sup>
B2(Progut)	0.122	0.259 <sup>b</sup>	0.477 <sup>b</sup>	0.698 <sup>b</sup>	0.931	1.112	3.597 <sup>b</sup>	1.859 <sup>b</sup>	141.688 <sup>a</sup>
SEM	0.0032	0.0043	0.0067	0.0118	0.0103	0.0168	0.0351	0.0187	2.143
P value	0.550	0.001	0.001	0.001	0.132	0.702	0.001	0.001	0.001
Interactions AB									
A1×B1	0.123	0.345	0.530	0.800	0.945	1.084	3.826	2.118	117.000
A1×B2	0.121	0.253	0.450	0.688	0.909	1.082	3.501	1.864	141.250
A2×B1	0.128	0.322	0.521	0.776	0.959	1.078	3.781	2.026	127.000
A2×B2	0.122	0.279	0.483	0.701	0.934	1.135	3.651	1.837	144.250
A3×B1	0.120	0.276	0.539	0.782	0.967	1.158	3.842	2.048	124.250
A3×B2	0.120	0.253	0.469	0.724	0.965	1.150	3.680	1.837	144.250
A4×B1	0.128	0.261	0.506	0.741	0.943	1.090	3.669	1.971	133.000
A4×B2	0.125	0.250	0.508	0.679	0.916	1.080	3.556	1.898	137.000
SEM	0.006	0.009	0.013	0.024	0.021	0.034	0.070	0.037	4.285
P value	0.974	0.004	0.021	0.656	0.860	0.711	0.4263	0.118	0.130

Note: a-b: Means within column with different superscripts are significantly different.

A1 – 0 feed withdrawal time at the 2<sup>nd</sup> week of age, A2 = 6 feed withdrawal time at the 2<sup>nd</sup> week of age, A3 = 9 feed withdrawal time at the 2<sup>nd</sup> week of age and A4 = 12 feed withdrawal time at the 2<sup>nd</sup> week of age.

B1 = without feed additive and B2 = with feed additive.

However, Shafiei, et al. [26] found that used feed withdrawal time at 8, 10 hours/day had increased carcass weight of broiler chicks compared with other groups. Abaseid, et al. [20] reported that feed withdrawal during the 4<sup>th</sup> and 5<sup>th</sup> weeks of age had increased in carcass of broiler chicks compared with control group.

Our results agree with those of Yalçın, et al. [23] who reported that broiler fed diets containing yeast was not affected on carcass and parts percentage. Fathi, et al. [38] reported that used yeast in broiler diets did not significant effected in carcass and giblet weight. Chumpawadee, et al. [39] found that carcass and parts percentage did not affected by used yeast in broiler diets. Waldroup, et al. [40] reported that carcass weight of broiler chicks did not affected by supplementation of broiler diet with bio-mos mannan oligosaccharide.

On the contrary, Ahiwe, et al. [13] found that broiler fed yeast (2g/kg diets) was significantly increased carcass percentage compared to control group. Yildirim, et al. [41] reported that used yeast in broiler diets did not significant effected in liver and gizzard weight.

## 3.4. Blood Parameters

Table 5, 6, 7 showed that the effects of feed withdrawal and feed additive (Progut®) on some serum blood characteristic and immunity of broiler chicks (Cobb 500). Serum blood characteristic of broiler chicks were not affected by reared under feed withdrawal during the second week of age. Feed additive supplemented on broiler diets had not significantly effect on serum blood characteristic, however significantly increased level of  $\beta$  globulin and decreased level of  $\alpha$  globulin in serum blood. Also, the interaction between feed withdrawal and feed additive were not significantly difference in serum blood characteristic.

Our results agree with those of Sherif and Mansour [36] who observed that broiler reared under feed withdrawal did not effected on plasma total protein, albumin, globulin, triglycerides and cholesterol of broiler chicks compared with the control group. Shafiei, et al. [26] found that plasma globulin; cholesterol and triglyceride values did not affected by used feed withdrawal with broiler chicks. Also, Xu, et al. [42] showed that the plasma total protein, albumin or globulin of broilers did not affected by reared broiler chicks under feed withdrawal. Afsharmanesh, et al. [43] found that broiler exposed of feed withdrawal did not effected on total cholesterol and triglycerides of blood broilers. Adeyemi, et al. [44] found that broiler exposed of feed withdrawal did not effected on serum concentrations of total protein, albumin and globulin.

**Table 4.** Effects of feed withdrawal and feed additive (Progut®) on carcass traits % of broiler chicks at 42 days of age.

Treatments	Live weight (g)	Carcass %	liver%	Gizzard%	Heart%	giblet %	TEP %
Feed withdrawal							
A1 (0 h)	2340	71.249	2.557	1.401	0.561	4.519	75.767
A2 (6 h)	2386	72.091	2.134	1.379	0.569	4.082	76.173
A3 (9 h)	2264	72.305	2.294	1.216	0.596	4.106	76.411
A4 (12 h)	2250	72.054	2.356	1.233	0.579	4.168	76.222
SEM	63.1862	0.4407	0.1300	0.0769	0.0269	0.1766	0.4431
P value	0.390	0.360	0.165	0.215	0.808	0.283	0.772
Feed additive							
B1	2211 <sup>b</sup>	71.814	2.484 <sup>a</sup>	1.375	0.582	4.442 <sup>a</sup>	76.256
B2(Progut)	2409 <sup>a</sup>	72.035	2.186 <sup>b</sup>	1.239	0.570	3.996 <sup>b</sup>	76.031
SEM	44.6794	0.3116	0.0919	0.0543	0.0190	0.1249	0.3133
P value	0.004	0.621	0.029	0.086	0.659	0.017	0.6146
Interactions AB							
A1×B1	2274	71.332	2.736	1.566	0.551	4.853	76.185
A1×B2	2406	71.165	2.378	1.237	0.570	4.185	75.349
A2×B1	2212	72.172	2.322	1.544	0.568	4.434	76.605
A2×B2	2560	72.010	1.946	1.214	0.570	3.730	75.740
A3×B1	2228	71.851	2.412	1.224	0.597	4.233	76.084
A3×B2	2300	72.759	2.176	1.208	0.595	3.980	76.738
A4×B1	2130	71.902	2.467	1.168	0.612	4.247	76.149
A4×B2	2370	72.206	2.246	1.298	0.545	4.089	76.295
SEM	89.359	0.623	0.184	0.109	0.038	0.2497	0.627
P value	0.438	0.802	0.962	0.101	0.691	0.601	0.5478

Note: a-b: Means within column with different superscripts are significantly different.

A1 = 0 feed withdrawal time at the 2<sup>nd</sup> week of age, A2 = 6 feed withdrawal time at the 2<sup>nd</sup> week of age, A3 = 9 feed withdrawal time at the 2<sup>nd</sup> week of age and A4 = 12 feed withdrawal time at the 2<sup>nd</sup> week of age.

B1 = without feed additive and B2 = with feed additive.

TEP = total edible parts.

Our results agree with those of He, et al. [24] who reported that used yeast in broiler diets did not effected in serum total protein, globulin, albumin and increased in LgG value. Sun, et al. [25] reported that broiler fed yeast in their diet increased serum IgA and IgG and not effected on total protein. Yalçın, et al. [23] reported that yeast supplemented in broiler diets did not effected in serum total protein, cholesterol and triglyceride. Fathi, et al. [38] reported that used yeast in broiler diets (1, 1.25, 1.5 g/kg) did not significant effected in IgG and IgM at 21 day of age compared with control group.

On the contrary, Ding, et al. [31] found that addition yeast in broiler diets enhanced serum IgG in broilers indicate to improvement serum immunity. Orso, et al. [21] reported that IgY of serum was increased due to reared broiler chicks under feed restriction. Cotter, et al. [45] showed that the mannanoligosaccharides of live yeast has a role in improving the immune response by enhancing immunoglobulin production in poultry.

### 3.5. Total Micro Flora Counts and Some Enzymes

Table 8 showed the effects of feed withdrawal and feed additive (Progut®) on total micro flora counts and some enzymes of broiler chicks (Cobb 500). Feed withdrawal and feed additive used in broiler diets had significantly decreased on total micro flora counts and *E coil* and enhanced *lacto Bacillus*, amylase and chymotrypsin. Interaction between feed withdrawal and feed additive Progut® were not significantly difference in total micro flora counts and some enzymes of broiler chicks.

**Table 5.** Effects of feed withdrawal and feed additive (Progut®) on serum blood parameters of broiler chicks.

Treatments	Total protein g/dl	Albumin g/dl	Globulin g/dl	ALB/GLB R	$\alpha$ globulin g/dl	$\beta$ globulin g/dl	$\gamma$ globulin g/dl
Feed withdrawal							
A1 (0 h)	5.580	2.550	3.030	1.620	0.980	0.760	5.580
A2 (6 h)	5.520	2.630	2.890	1.650	1.000	0.810	5.520
A3 (9 h)	5.480	2.520	2.960	1.620	1.020	0.840	5.480
A4 (12 h)	5.710	2.530	3.180	1.610	0.930	0.830	5.710
SEM	0.102	0.094	0.135	0.038	0.039	0.034	0.102
P value	0.417	0.836	0.480	0.893	0.419	0.377	0.417
Feed additive							
B1	5.540	2.555	2.985	1.610	1.030 <sup>a</sup>	0.775 <sup>b</sup>	5.540
B2 (Progut)	5.605	2.560	3.045	1.640	0.935 <sup>b</sup>	0.845 <sup>a</sup>	5.605
SEM	0.072	0.066	0.095	0.027	0.028	0.024	0.072
P value	0.528	0.958	0.660	0.441	0.021	0.051	0.528
Interactions AB							
A1×B1	5.720	2.600	3.120	1.620	1.080	0.740	5.720
A1×B2	5.440	2.500	2.940	1.620	0.880	0.780	5.440
A2×B1	5.420	2.600	2.820	1.620	1.000	0.780	5.420
A2×B2	5.620	2.660	2.960	1.680	1.000	0.840	5.620
A3×B1	5.400	2.560	2.840	1.660	1.120	0.780	5.400
A3×B2	5.560	2.480	3.080	1.580	0.920	0.900	5.560
A4×B1	5.620	2.460	3.160	1.540	0.920	0.800	5.620
A4×B2	5.800	2.600	3.200	1.680	0.940	0.860	5.800
SEM	0.144	0.132	0.191	0.054	0.055	0.049	0.144
P value	0.297	0.771	0.724	0.242	0.086	0.859	0.297

Note: a-b: Means within column with different superscripts are significantly different.

A1 = 0 feed withdrawal time at the 2<sup>nd</sup> week of age, A2 = 6 feed withdrawal time at the 2<sup>nd</sup> week of age, A3 = 9 feed withdrawal time at the 2<sup>nd</sup> week of age and A4 = 12 feed withdrawal time at the 2<sup>nd</sup> week of age.

B1 = without feed additive and B2 = with feed additive.

**Table 6.** Effects of feed withdrawal and feed additive (Progut®) on serum blood parameters of broiler chicks.

Treatments	Total lipids mg/dl	Triglycerides mg/dl	Cholesterol mg/dl
Feed withdrawal			
A1 (0 h)	5.870	179.600	211.200
A2 (6 h)	5.750	178.100	206.500
A3 (9 h)	5.780	173.000	202.100
A4 (12 h)	5.530	178.800	210.200
SEM	0.258	2.751	3.237
P value	0.816	0.335	0.203
Feed additive			
B1	5.920	176.900	206.700
B2 (Progut)	5.545	177.850	208.300
SEM	0.182	1.946	2.289
P value	0.156	0.732	0.625



Treatments	Total lipids mg/dl	Triglycerides mg/dl	Cholesterol mg/dl
Interactions AB			
A1×B1	6.000	180.800	212.000
A1×B2	5.740	178.400	210.400
A2×B1	6.400	176.200	205.000
A2×B2	5.100	180.000	208.000
A3×B1	5.860	178.200	199.600
A3×B2	5.700	167.800	204.600
A4×B1	5.420	172.400	210.200
A4×B2	5.640	185.200	210.200
SEM	0.365	3.891	4.578
P value	0.211	0.037	0.889

Note: A1 = 0 feed withdrawal time at the 2<sup>nd</sup> week of age, A2 = 6 feed withdrawal time at the 2<sup>nd</sup> week of age, A3 = 9 feed withdrawal time at the 2<sup>nd</sup> week of age and A4 = 12 feed withdrawal time at the 2<sup>nd</sup> week of age.  
B1 = without feed additive and B2 = with feed additive.

**Table 7.** Effects of feed withdrawal and feed additive (Progut®) on immunity serum blood parameters of broiler chicks.

Treatments	IgG mg/100ml	IgM mg/100ml	IgA mg/100ml
Feed withdrawal			
A1 (0 h)	9.975	2.540	0.825
A2 (6 h)	10.030	2.540	0.814
A3 (9 h)	9.958	2.511	0.842
A4 (12 h)	9.999	2.528	0.818
SEM	0.040	0.040	0.015
P value	0.614	0.949	0.585
Feed additive			
B1	9.976	2.507	0.816
B2 (Progut)	10.005	2.553	0.834
SEM	0.028	0.028	0.011
P value	0.474	0.253	0.260
Interactions AB			
A1×B1	9.982	2.558	0.824
A1×B2	9.968	2.522	0.826
A2×B1	9.984	2.528	0.804
A2×B2	10.076	2.552	0.824
A3×B1	9.980	2.464	0.850
A3×B2	9.936	2.558	0.834
A4×B1	9.958	2.476	0.786
A4×B2	10.040	2.580	0.850
SEM	0.057	0.056	0.022
P value	0.545	0.574	0.303

Note: IgG= Immunoglobulin G; IgM= Immunoglobulin M; IgA= Immunoglobulin A.

A1 = 0 feed withdrawal time at the 2<sup>nd</sup> week of age, A2 = 6 feed withdrawal time at the 2<sup>nd</sup> week of age, A3 = 9 feed withdrawal time at the 2<sup>nd</sup> week of age and A4 = 12 feed withdrawal time at the 2<sup>nd</sup> week of age.  
B1 = without feed additive and B2 = with feed additive.

Our results agree partially with Lunedo, et al. [46] who found that used feed restriction on broiler chicks reduced *Enterococcus* and *Enter bacteriaceae* enhanced *Lactobacillus* counts. Shafiei, et al. [26] found that used feed withdrawal time at 8, 10 hours/day had not effected on *E. coli* count however, *Lactobacilli* count was enhanced by exposed broiler chicks 12 h feed withdrawal. Also, Siegerstetter, et al. [47] reported that broiler exposed high feed restriction improved *lactobacillus* and decreased *Escherichia/Shigella*. Yan, et al. [48] reported that broiler exposed feed restriction had increased beneficial macrobiotic indicate to improved digestion in digestive tract.

Our results agree partially with Ahiwe, et al. [13] who found that broiler fed yeast (1.5 to 2g/kg diets) was significantly enhanced trypsin and chymotrypsin of birds. Ogbuewu, et al. [49] found that account pathogenic bacteria decreased in gut intestine due to increased acidic in intestine by used yeast in broiler diets. Yalçın, et al. [23] reported that broiler fed diets containing yeast decreased *E. coli* colonization and increased total aerobic bacteria in jejunum and ileum of the broilers.

**Table 8.** Effects of feed withdrawal and feed additive (Progut®) on total micro flora counts in broiler intestine and some enzymes of broiler chicks.

Treatments	Total Bacteria count	E. Coli	Lacto bacillus.	Amylase	Lipase	Trypsin	Chemo-trypsin
Feed withdrawal							
A1 (0 h)	12.875 <sup>a</sup>	6.580 <sup>a</sup>	6.105 <sup>c</sup>	3.032 <sup>c</sup>	10.050	27.413 <sup>a</sup>	18.948 <sup>b</sup>
A2 (6 h)	11.088 <sup>c</sup>	5.422 <sup>b</sup>	6.653 <sup>a</sup>	3.248 <sup>a</sup>	11.215	25.305 <sup>b</sup>	18.463 <sup>b</sup>
A3 (9 h)	11.782 <sup>b</sup>	5.522 <sup>b</sup>	6.542 <sup>ab</sup>	3.180 <sup>b</sup>	12.392	27.592 <sup>a</sup>	20.665 <sup>a</sup>
A4 (12 h)	10.912 <sup>c</sup>	5.367 <sup>b</sup>	6.402 <sup>b</sup>	3.042 <sup>c</sup>	14.058	26.997 <sup>a</sup>	21.117 <sup>a</sup>
SEM	0.059	0.065	0.035	0.015	1.082	0.413	0.132
P value	0.001	0.001	0.001	0.001	0.097	0.005	0.001
Feed additive							
B1	12.957 <sup>a</sup>	6.915 <sup>a</sup>	4.868 <sup>b</sup>	3.116	12.224	26.154 <sup>b</sup>	19.212 <sup>b</sup>
B2 (Progut)	10.372 <sup>b</sup>	4.530 <sup>b</sup>	7.983 <sup>a</sup>	3.135	11.633	27.499 <sup>a</sup>	20.385 <sup>a</sup>
SEM	0.042	0.046	0.024	0.011	0.765	0.292	0.093
P value	0.001	0.001	0.001	0.218	0.593	0.005	0.001
Interactions AB							
A1×B1	14.800	8.170	4.280	3.200	10.130	27.223	19.133
A1×B2	10.950	4.990	7.930	2.863	9.970	27.603	18.763
A2×B1	12.563	6.520	5.193	3.113	10.650	24.020	16.873
A2×B2	9.613	4.323	8.113	3.383	11.780	26.590	20.053
A3×B1	12.713	6.520	5.153	3.150	11.730	27.040	19.940
A3×B2	10.850	4.523	7.930	3.210	13.053	28.143	21.390
A4×B1	11.750	6.450	4.843	3.000	16.387	26.333	20.900
A4×B2	10.073	4.283	7.960	3.083	11.730	27.660	21.333
SEM	0.084	0.091	0.049	0.021	1.531	0.584	0.187
P value	0.001	0.001	0.001	0.001	0.215	0.336	0.001

**Note:** a-c: Means within column with different superscripts are significantly different.

A1 = 0 feed withdrawal time at the 2<sup>nd</sup> week of age, A2 = 6 feed withdrawal time at the 2<sup>nd</sup> week of age, A3 = 9 feed withdrawal time at the 2<sup>nd</sup> week of age and A4 = 12 feed withdrawal time at the 2<sup>nd</sup> week of age.

B1 = without feed additive and B2 = with feed additive.

#### 4. CONCLUSION

The obtained results suggested that feed withdrawal used during the second week had not effect on broiler performance, carcass traits and serum blood characteristics and improved *lacto Bacillus*, amylase and chymotrypsin. Feed additive (Progut®) in broiler diets improved broiler growth performance and improved intestinal health.

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