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Effects of Dietary Probiotic, Prebiotic and Enzyme Mixture Supplementation on Performance, Carcase, Organs, Ileal pH and Viscosity of Broilers

Karma Yeme İlave Edilen Probiyotik, Prebiyotik ve Enzim Karışımının Etlik Piliçlerde Performans, Karkas, Organlar, İleal pH ve Viskozitesi Üzerine Etkileri

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Key Words:

Broiler, Probiotic, Prebiotic, Enzyme, Performance.

ABSTRACT

Objective: The experiment was conducted to determine the individual and combined effects of probiotic, prebiotic and enzyme on performance, carcase, organs, intestinal pH and viscosity of broilers.

Material and Methods: A total of seven hundred day-old male Ross-308 broiler chicks were individually weighted and distributed into 28 floor pens with 25 chicks per pen in from 0 to 42 d of age. Seven starter and grower diets were formulated to provide a similar nutrient profile with the exception of using experimental feed additives (1 g per kg) or a combined addition of these additives (0.5 g+0.5 g per kg). The diets were supplemented with: no feed additives (control), probiotic, prebiotic, enzyme, probiotic+prebiotic, probiotic+enzyme mixture, prebiotic+enzyme mixture.

Results: Probiotic, probiotic+prebiotic and probiotic+enzyme supplementation significantly increased body weights at 42 days (P<0.05). There were no differences in feed intake, feed conversion ratio, relative weights of the proventriculus, gizzard, duodenum, jejunum, ileum, colon, cecum liver, pancreas, spleen, heart and bursa of broilers among the treatments during the experiment (P>0.05). Intestinal pH and viscosity were decreased by adding feed additives when compared with the control.

Conclusion: The synergistic effects between probiotics, prebiotics and enzymes should be further investigated, and suitable combinations and levels should be determined in practice.

Anahtar Kelimeler:

Etlik piliç, Probiyotik, Prebiyotik, Enzim, Performans.

ÖZ

Amaç: Deneme, probiyotik, prebiyotik ve enzimin etlik piliçlerde performans, karkas, organlar, bağırsak pH'ı ve viskozitesi üzerine tek başına ve kombine etkilerini belirlemek amacıyla yürütülmüştür.

Materyal ve Metot: Bir günlük yaşta 700 adet erkek Ross-308 etlik civciv, bireysel olarak tartılmış ve her bölmede 25 civciv olacak şekilde 28 gruba dağıtılarak 0-42. günler arasında barındırılmıştır. Denemede kullanılan katkı maddeleri dışında benzer besin madde profiline sahip ve kullanılan yem katkı maddesinin tek başına (1 g/kg) veya kombine şekilde (0.5 g/kg+0.5 g/kg) ilave edildiği yedi adet başlatma ve büyütme yemi formülasyonu oluşturulmuştur. Yemler, katkı maddesi içermeyen (kontrol), prebiyotik, enzim, probiyotik+prebiyotik, probiyotik+enzim ve prebiyotik+enzim içerecek şekilde hazırlanmıştır.

Bulgular: Probiyotik, probiyotik+prebiyotik ve probiyotik+enzim ilavesi 42. gün canlı ağırlığını önemli derecede artırmıştır (P<0.05). Deneme süresince, gruplar arasında yem tüketimleri, yemden yararlanma oranları ile bezel mide, taşlık, duedonum, jejunum, ileum, kalın bağırsak, kör bağırsak, karaciğer, pancreas, dalak, kalp ve bursanın oransal ağırlıkları bakımından bir farklılık saptanmamıştır (P>0.05). Yem katkısı ilavesinin bağırsak pH'ı ve viskozitesi üzerine önemli bir etkisi belirlenmiştir. Kontrol grubu ile karşılaştırıldığında deneme katkılarının ilavesi ile bağırsak pH'ı ve viskozitesi azalmıştır.

Sonuç: Probiyotikler, prebiyotikler ve enzimler arasındaki sinerjik etkiler daha fazla araştırılmalı, pratikte uygun kombinasyonlar ve seviyeler belirlenmelidir.

INTROCUTION

Antibiotic growth promoters have been banned by European Union since 2006 Jan 01, to avoid antimicrobial resistance. Consequently, the poultry industry must develop alternatives to antibiotic growth promotants used to maintain efficient poultry production. Since then, feed additives like probiotics, prebiotics and enzymes have taken on interest broiler production.

Probiotics are single or mixed culture of living microorganisms which when administrated in adequate numbers apply health benefits for the host by improving intestinal microbial balance, increase of colonization resistance against pathogens and improving the immune responses (Kabir, 2009; Brisbin et al., 2010; Cencic and Chingwaru, 2010; Das et al., 2012). The species of microorganisms currently being used in probiotic preparations are varied, and lactic acid bacteria, i.e., Lactobacillus bulgaricus, Lactobacillus acidophilus, Lactobacillus casei, Lactobacillus helveticus, Lactobacillus lactis, Lactobacillus salivarius, Lactobacillus plantarum, Streptococcus thermophilus, Enterococcus faecium, Enterococcus faecalis, Bifidobacterium spp., are the most common type of bacteria used as probiotics (Khaksefidi and Rahimi, 2005; Kabir, 2009). Many beneficial effects of probiotics were suggested, such as improved immune system, modification of gut microbiota, reduced inflammatory reactions, decreased ammonia and urea excretion, lower serum cholesterol, and improved mineral adsorption; however probiotics may have not directly positive impact on performance parameters and production profitability (Ferreira et al., 2011). Few of experiment have shown improvements in growth performance, decreased mortality and morbidity or increased resistance to colonization by pathogens associated with feeding prebiotics. Numerical improvements in performance may be economically important on large-scale production farms (Patterson and Burkholder, 2003).

Prebiotics are non-digestible feed ingredients that beneficially affect the host by selectively altering the composition and metabolism of the gut microbiota (Huyghebaert et al., 2011; Das et al., 2012). Prebiotics may provide energy for the growth of endogenous favourable bacteria in the gut, such as bifidobacteria and lactobacilli, thus improving the host microbial balance (Das et al., 2012). Although prebiotics may have a similar mechanism as probiotic in supporting the gut health of chicken (Huyghebaert et al., 2011), it may have more benefits than probiotics, because prebiotics stimulate commensal bacteria which have adapted to the environment of gastrointestinal tract (Adil and Magray, 2012; Alloui et al., 2013). Prebiotics have been reported to improve defence and reduce mortality of bird preventing the spread of pathogens (Ganguly, 2013). The most common prebiotics used in poultry are oligosaccharides, including inulin, fructooligosaccharides, mannanoligosaccharides, galactooligosaccharides, soya-oligosaccharides, xylooligosaccharides, pyrodextrins, isomaltooligosaccharides and lactulose (Huyghebaert et al., 2011; Kim et al., 2011; Alloui et al., 2013).

In combination, prebiotics and probiotic bacteria create symbiotics, which can provide furthermore benefits than probiotics or prebiotics alone.

The use of enzymes in poultry feed has increased dramatically in the last decade. Various exogenous enzymes including -glucanase, xylanase, amylase, α-galactosidase, protease, lipase, phytase, etc. have been supplemented in poultry diets for decades (Adeola and Cowieson, 2011; Bedford and Cowieson, 2012). To reach their potential in improving the nutritive value of feedstuffs, enzymes have to be biologically active when reaching the gastrointestinal tract. The structure of enzyme is critical to its activity. The structure of enzymes can be altered by exposure to heat, extremes of pH, or certain organic solvents such that activity can be decreased or completely abolished (Spring et al., 1996). The beneficial effects of some feed enzymes for improving nutrient availability and bird performance are well-established (Bedford and Morgan, 1996). Enzyme supplementation breaks polymeric chains into smaller pieces, reduces the gut viscosity, and hence improves the nutritive value of feedstuffs (Smits and Annison, 1996).

Therefore, the objectives of this study were to evaluate the individual and combined effects of three dietary feed additives, probiotic, prebiotic and enzyme, on performance, organs, intestinal pH and viscosity.

MATERIALS and METHODS

Animals and experimental design

A total of seven hundred day-old male Ross-308 broiler chicks were individually weighed and randomly distributed into 28 floor pens with 25 chicks per pen in Experiment. The birds were maintained in an open-sided broiler house and fed from 0 to 42 d of age. Each floor pen was furnished with wood shavings litter, a round feeders and a round drinker. Feed and water were consumed *ad libitum*. Temperature and relative humidity was maintained within the optimum range. Lighting was 23 h light and 1 h darkness.

Diets and feeding regimens

Seven starter and grower diets were formulated to provide a similar nutrient profile with the exception of using three feed additives (1 g per kg) or a combined addition of these additives (0.5+0.5 g per kg). Therefore, the arrangement of treatments were: 1) basal diets without additive (C), 2) basal diets+probiotic, 3) basal diets+prebiotic, 4) basal diets+enzyme mixture, 5) basal diets+combination of probiotic+prebiotic, 6) basal diets+combination of probiotic+enzyme mixture, 7) basal diets+combination of prebiotic+enzyme mixture. The probiotic preparation containing a multi-strain probiotic in dry white powder form (2×109 cfu/g) containing Streptococcus salivarius sub sp.Thermophilus, Lactobacillus (L) delbruckii sub sp. bulgaricus, L. acidophilus, L. plantarum, L.rhamnosus, Bifidobacterium bifidum, Enterococcus faecium, Candida pintoloppesii, and Asperigillus oryzae). The prebiotic preparation is comprised of Aspergillus meal. Aspergillus meal is derived from an active fermentation of a primary Aspergillus spp. It is the asporogenic mycelium contained in this totally dead product. A commercial multienzyme complex containing protease (7500 U/g), as well as of cellulase (500 U/g), amylase (350 U/g) and xylanase (350 U/g), endo-1,3; 1,4-beta glucanase (300 U/g), lipase (50 U/g) and -glucosidase (40 U/g), phytase (10 U/kg) activities.

All chicks were fed starter diets from 0 to 21 days of age and experimental grower diets from 22 to 42 days of age. Yellow maize-soybean based diets were utilized and all were formulated using linear programming to be isoenergetic and isonitrogenic. Starter and grower diets were fed *ad libitum* consumption. The ingredient and chemical composition of the basal starter and grower diets are presented in Table 1. The chemical analyses of basal diets were analyzed according to the procedures of AOAC (1980). ME values of diets were calculated using protein, ether extract, starch and sugar values (%).

Traits measured

At 21 and 42 days of age, all birds were weighed and feed intake of each replicate was determined. Mortality was recorded daily. Feed intake and feed conversion efficiency were adjusted for mortality. Fifty-six broilers (eight broilers each group) were sampled randomly at 42 d of age and slaughtered. Weights of the duodenum, jejunum, ileum, colon and cecum were recorded after ingesta was removed. The weights of these internal organs were expressed as a percentage of live body weight.

Digesta sample collected from the small intestine mixtured and homogenized then pH was determined by using pH-metre (Hanna 8314 model) and the viscosity of a 0.5 ml aliquot measured using a Brookfield Digital Viscometer (Model DV- II+PRO, Brookfield Engineering Laboratories, Stoughton, MA) maintained at 40 °C.

Statistical analysis

Data were subjected to ANOVA using General Linear Models (SPSS, 1997). The model included diet as main effects. Pen means served as the experimental unit for statistical analysis. Differences among means were tested using Duncan's multiple-range tests with a 5% level of probability was used.

Results

Average livability value was 98.87±0.80 for experiment and there were no treatment differences (P<0.05). The effect of dietary supplementation of probiotic, prebiotic, enzyme and their combination on body weights and body weight gains is presented in Table 2. The dietary treatments had significant effects on body weights and body weight gains of broilers at 42 days. Probiotic, probiotic+prebiotic and probiotic+enzyme supplementation significantly increased body weights at 42 days and body weight gains during the 0 to 42 days (P<0. 05). Body weights at 42 days and durig the periods 0 to 42 d, body weight gains of birds fed the diet containing prebiotic, enzyme and prebiotic+enzyme similar to control birds (P>0.05). The effect of dietary supplementation of probiotic, prebiotic, enzyme and their combination on feed intake and feed conversion ratio during the experiment is presented in Table 2. There were no differences in feed intake and feed conversion ratio between treatments during the experiment (P>0.05). Enzyme supplementation had significant effect positively on feed conversion ratio during the experiment. These results indicate that the addition of enzyme improved the feed conversion ratio but not statistically (P>0.05).

Results of experiment indicate that relative weights of the proventriculus, gizzard, duodenum, jejunum, ileum, colon, cecum liver, pancreas, spleen, heart and bursa of broilers at 42 d were not affected by dietary treatments in Table 3 (P>0.05). However, treatments tended slightly increase to weight of bursa of Fabricius (P>0.05).

In the present study, addition of probiotic, prebiotic, enzyme and their combination to diet did affect intestinal pH and viscosity in Table 4 (P<0.05). The highest ileal pH and viscosity were obtained from control. All treatments were similar and significantly decreased ileal pH and viscosity.

Discussion

In the present study, broilers given diets supplemented with probiotic, probiotic+prebiotic and probiotic+enzyme improved chicken growth performance. However, broilers given diets supplemented with prebiotic, enzyme prebiotic+enzyme tended to have a higher weight than broilers fed on control diet but not significantly. The results obtained from many studies, in which the effects of probiotic, prebiotic, enzyme or their combinations were investigated on growth performance in poultry, were not consistent. The effect of probiotic administration on the performance of broiler is variable. The results of the present experiment agree with the finding of Afsharmanesh and Sadaghi (2014); Mookiah et al. (2014); Zhang and Kim (2014); Lei et al. (2015) reported that dietary supplementation of probiotics improve broiler chicken growth rates. In many cases, the improvement in growth rate in the birds fed the diet containing probiotic was associated with increased feed intake (Abdel-Raheem et al., 2012; Landy and Kavyani, 2013; Lei et al., 2015) and improved feed efficiency or feed conversion ratio (Mountzouris et al., 2010; Hung et al., 2012; Fajardo et al., 2012; Shim et al., 2012; Zhang and Kim, 2014) compared with control birds. However, in our study, feed intake and feed conversion ratio were not affected by treatments. Furthermore, Khaksefidi and Rahimi (2005) reported that inclusion of probiotics in the diet resulted in improved performance of broiler compared to the control group in the 4th, 5th and 6th weeks. Also, Cao et al. (2013) reported that the growth rate of male broilers improved. Zhao et al. (2013) observed probiotic supplementation improved the average daily feed intake and average daily gain. In contrast, Fajardo et al. (2012) reported that no differences with probiotic supplementation in body weight gain of broilers. Moreover, Bai et al. (2013) reported no significant differences in growth performance during 22-42 days between broilers fed the probiotic diet and those fed control diet. In addition, Brzóska et al. (2012) reported that probiotics significantly reduced chicken mortality compared to the control group and feeding diets with bacteria to chickens did not increase body weight at 42 days of age or improve feed conversion compared to control chickens. Results of the present experiment indicate that the feed intake and feed conversion ratio were not significantly affected by treatments. The differences in the dose and nature of probiotics administrated and variation in the physiological state of the birds are likely the reasons (Huyghebaert et al., 2011).

In several studies, performance parameters in broilers have been evaluated with prebiotic supplementation. Body weight was reported to increase in the majority of studies (Yusrizal and Chen, 2003; Sims et al., 2004). Samarasinghe et al. (2003); Xu et al. (2003); Yusrizal and Chen (2003); Sims et al. (2004); Józefiak et al. (2008) and Yang et al. (2008) reported that body weight gain and feed conversion ratio were improved. Feed intake and feed conversion ratios generally improved with supplementation of prebiotics (Baurhoo et al., 2007; Samarasinghe et al., 2003; Xu et al., 2003; Yusrizal and Chen, 2003). Also, Mamiek (1993) and Tangendjaja (1993) observed improvements in body weights of broilers given Aspergillus meal. But, Baurhoo et al. (2009) reported that non-significant improvement in body weight. In some studies on broiler chickens, Aspergillus meal resulted in better body weight gain, feed intake and feed conversion ratio (Piray et al., 2007; Khaksar et al., 2008; Piray and Kermanshahi, 2008). Several studies have shown the beneficial effects of prebiotics on poultry performance (Spring et al., 2000; Xu et al., 2003;), but the publications about Aspergillus riginated prebiotics are almost limited. In this experiment, body weight is similar to control fed Aspergillus meal. Generally, in previous study body weight is improvement with Aspergillus meal (Khan et al., 2000; Piray et al., 2007; Khaksar et al., 2008). Navidshad et al. (2010) reported that in the study, Aspergillus meal caused no differences in feed intake among treatments. Similar results were reported by Khan et al. (2000) and Ghiyasi et al. (2007). However, there is positive reports, too (Piray et al., 2007). Aspergillus meal failed to improve feed conversion ratio in this research. Navidshad et al. (2010), which seems to be in agreement with the finding of Ghiyasi et al. (2007), but in disagreement with other authors (Khan et al., 2000; Piray et al.,2007; Khaksar et al., 2008). It is clear from the present study and published research that responses to prebiotic supplementation are inconsistent. A possible explanation for the differences among findings of different investigators may be related to the doses and type of prebiotic applied and diets composition.

Previous studies have shown that enzyme supplementation has the potential to improve the nutritive value of feedstuffs for broiler (Annison, 1992; Kırkpınar et al., 1996). Contrary to this, non-significant improvement in body weight was reported with enzyme supplementation (Omojola and Adesehinwa, 2007). To obtain the maximum benefit from the enzymes, the use of multiple enzymes is recommended as the combination of the enzymes may target different anti-nutritive compounds in the feedstuffs (Adeola and Cowieson, 2011). However, it should be noted that the beneficial effect of enzyme combination may be dependent on the diet composition (Meng et al., 2005). The results of Thorat et al. (2015) study indicate that multi-enzyme supplementation as growth promoter improved the growth performance of broilers significantly followed by prebiotics but not probiotics as compared to control group.

In the present study, feed intake and feed conversion ratio were not significantly affected by treatments during the trial. Similar observations were recorded by Baurhoo et al. (2009), Thorat et al. (2015) and Islam et al. (2010), who found no difference in feed intake of broilers when multi-enzymes or prebiotic into diet. In contrast to these results, Olukosi et al. (2007) and Woyengo et al. (2010) found improved feed conversion ratio in broilers receiving diet supplemented with multi-enzyme as compared to the control group. Thorat et al. (2015) showed that supplementation of multi-enzyme improved broiler feed conversion ratio but not probiotic or prebiotic.

Recent studies suggested that probiotics could be more effective when used with prebiotics (Mookiah et al., 2014). These results are consistent with our study. Symbiotics are relatively recent among additives used in poultry nutrition. Studies have suggested that performance can be further enhanced when using probiotics with together prebiotics. Some studies suggest that the effects of a combination of both feed additives are cumulative (Pijsel, 1996). Some researchers reported the importance and benefits of this kind of synergy between probiotics and prebiotics and the effectiveness in helping young animals to achieve better growth performance (Patterson and Burkholder, 2003). Abdel-Hafeez et al. (2017) was found that chicks fed diets supplemented with probiotic, prebiotic and symbiotic (with and without feed restriction) exhibited higher body weight and feed efficiency than chicks fed the control diets. However, the relative weights of liver, gizzard and proventriculus, small intestine and bursa of fabricius were found to be increased. Kabir et al. (2004) observed improvement in growth performance and immune response with probiotic in broilers. Researchers demonstrated that addition of probiotics increased the weight of the spleen and bursa of broilers. In the present study, relative weights of the proventriculus, gizzard, duodenum, jejunum, ileum, colon, cecum liver, pancreas, spleen, heart and bursa of broilers at 42 d were not affected by dietary treatments. These results agree with the finding of Park and Kim (2014) who observed that relative weights of liver, spleen and gizzard were not influenced by groups fed with probiotic. However, interestingly, birds fed diets containing probiotic showed an increase in the weight of bursa of Fabricius, as dietary levels of probiotic increased. In the present study, treatments tended slightly increase to weight of bursa of Fabricius. On the other hand, Sarangi et al. (2016) reported that the growth performance did not show any significant increase by the dietary inclusion of prebiotic, probiotic, and symbiotic compared with unsupplemented control in a commercial broiler chicken.

Intestinal viscosity is known to be a major factor limiting bird performance (Bedford and Morgan, 1996). In the present study, the highest ileal pH and viscosity were obtained from control. All treatments were similar and significantly decreased intestinal pH and viscosity. Narasimha et al., (2015) reported that viscosity in intestinal content was considerably reduced with enzyme or symbiotics (probiotic+prebiotic) addition to basal diet, corroborating with the present findings. Also, Owens et al. (2008) reported lower gut viscosity on different feed additives (Allzyme, Avilamycin, Avimos, Biomos, yeast extract, Avizyme, xylanase, Gustar alone or in combination). Contrary to this report, Kocher et al. (2002) reported that the enzymes had no effect on digesta viscosity in the jejunum or ileum. On the other hand Munj et al., (2010) studied the synergistic effect of prebiotics, probiotics and acidifier single or in combination in broiler chickens and observed no change in pH values measured among the experimental groups.

Table 1. The composition of basal starter and grower diets (g/kg)
Çizelge 1. *Başlangıç ve büyütme yemlerinin kompozisyonu (g/kg)*

Ingredient	Starter Diet (0-21 days)	Grower Diet (22-42 days)
Yellow maize	479.4	459.2
Wheat	30	127
Fullfat soybean	53.7	20
Soybean meal	177.9	224.7
Sunflower meal	200	121
Fish meal	15	15
Vegetable oil	2	2
Ground limestone	11.5	9
Dicalcium phosphate	16.3	12.6
Iodised sodium chloride	4.4	3
Lysin	5.3	2
DL-Methionine	1	1
Vitamin mixture[1]	2.5	2.5
Mineral mixture[2]	1	1
Analyzed composition (g/kg)		
Dry matter	887.1	883.6
Crude protein	220.2	200.3
Ether extract	57	66.12
Crude fiber	70.7	56.7
Crude ash	64.9	54.3
Total calcium	10.5	8.5
Calculated composition (g/kg)		
Available phosphorus	5	4.2
Sodium	2	1.5
Lysine	14.3	11.1
Methionine	5	4.5
Metabolizable energy (kcal/kg)	3000	3150.38

[1] Supplied mg/kg of diet: retinol acetate, 5.16; cholecalciferol, 0.0375; tocopheryl, 20; menadione, 5; thiamine, 3; riboflavin, 6; niacin, 25; calcium D-pantothenate, 12; pyridoxine, 5; cyanocobalamin, 0.03; folic acid, 1; D-biotin 0.05; choline chloride, 400; carophyll yellow, 25.

[2] Supplied mg/kg of diet: manganese, 80; iron, 60; zinc, 60; copper, 5; cobalt, 0.2; iodine, 1; selenium, 0.15; calcium carbonate, 447.

Conclusions

In conclusion, the results of the present study showed that probiotic, probiotic+prebiotic and probiotic+enzyme supplementation significantly increased body weights at 42 days. Probiotic, prebiotic and enzyme, alone or in combination, tended slightly increase to the relative weight of bursa of Fabricius. Intestinal pH and viscosity were decreased by adding probiotic, prebiotic and enzyme, alone or in combination when compared with the control. Synergistic effects could be exploited so as to maximise the performance. From reported findings, it can be concluded that the dietary addition of symbiotic resulted in enchanced production performance of broiler chickens. The synergistic effects between probiotics, prebiotics and enzymes should be further investigated, suitable commercial combinations and levels should be determined, and these results should be incorporated in practice.

Table 2. Effects of probiotic, prebiotic, enzyme and their combination on fattenning performance of broilers

Çizelge 2. *Probiyotik, prebiyotik, enzim ve kombinasyonlarının etlik piliçlerin besi performansı üzerine etkileri*

Treatments	Body weights (g)		Body weights gain (g)			Feed intake (g)			Feed conversion ratio		
	21 d	42 d	0-21 d	22-42 d	0-42 d	0-21 d	22-42 d	0-42 d	0-21 d	22-42 d	0-42 d
Control	645^{b}	2122^{c}	605^{b}	1477	2082^{c}	1088	2954	4042	1.80	2.00	1.94
PRO	657^{b}	2174^{ba}	617^{b}	1517	2134^{ba}	1075	2985	4060	1.74	1.97	1.91
PRE	626^{c}	2142^{bc}	586^{c}	1516	2102^{bc}	1048	2968	4016	1.79	1.96	1.91
E	644^{b}	2155^{bc}	604^{b}	1511	2115^{bc}	1017	2883	3900	1.68	1.91	1.84
PRO+PRE	673^{a}	2218^{a}	633^{a}	1546	2179^{a}	1085	3052	4137	1.71	1.98	1.90
PRO+E	654^{b}	2173^{ba}	614^{b}	1519	2133^{ba}	1088	2992	4080	1.77	1.97	1.91
PRE+E	546^{b}	2165^{bc}	606^{b}	1519	2125^{bc}	1052	2872	3924	1.74	1.89	1.85
SEM	45.14	16.50	6.37	16.22	18.66	48.00	99.76	142.53	0.02	0.05	0.03
P Value	0.0001	0.0014	0.0001	0.1165	0.0015	0.8921	0.7873	0.8907	0.1523	0.5266	0.1291

PRO Probiotic, PRE Prebiotic, E Enzyme, ^{a,b} Means within a column in each variable with no common superscript differ significantly (P≤0.05), SEM Standard error of means, Feed conversion ratio: feed intake/weight gain.

Table 3. Effects of probiotic, prebiotic, enzyme and their combination on relative weights of organ[1] of broilers

Çizelge 3. *Probiyotik, prebiyotik, enzim ve kombinasyonlarının etlik piliçlerin nispi organ[1] ağırlıkları üzerine etkileri*

Treatments	Prov.	Giz.	Due.	Jej.	İleum	Colon	Cecum	Liver	Panc.	Spleen	Heart	Bursa
Control	0.38	1.35	0.73	2.21	0.28	0.15	0.36	2.65	0.26	0.26	0.41	0.11
PRO	0.37	1.28	0.80	2.58	0.28	0.16	0.41	2.99	0.29	0.29	0.41	0.16
PRE	0.34	1.36	0.77	2.69	0.31	0.16	0.41	2.71	0.27	0.27	0.39	0.15
E	0.38	1.45	0.80	2.54	0.30	0.16	0.43	2.83	0.27	0.27	0.38	0.14
PRO+PRE	0.38	1.35	0.74	2.48	0.30	0.15	0.39	2.85	0.30	0.30	0.38	0.16
PRO+E	0.36	1.37	0.81	2.62	0.29	0.16	0.42	2.74	0.27	0.27	0.38	0.15
PRE+E	0.38	1.37	0.77	2.52	0.29	0.16	0.40	2.73	0.28	0.28	0.40	0.15
SEM	0.01	0.05	0.03	0.16	0.02	0.01	0.02	0.12	0.02	0.01	0.02	0.02
P Value	0.648	0.243	0.564	0.170	0.644	0.978	0.166	0.296	0.647	0.647	0.521	0.162

[1]Organ weight, g/100 g of body weight. Prov. Proventriculus, Giz. Gizzard, Due. Duedonum, Jej. Jejenum, Panc. Pancreas, Bursa Bursa of Fabricius, PRO Probiotic, PRE Prebiotic, E Enzyme, SEM Standard error of means.

Table 4. Effects of probiotic, prebiotic, enzyme and their combination on intestinal pH and viscosity of broilers
Çizelge 4. *Probiyotik, prebiyotik, enzim ve kombinasyonlarının etlik piliçlerin bağırsak pH ve viskozitesi üzerine etkileri*

Treatments	pH	Viscosity
Control	6.37[a]	1.23[a]
Probiotic	6.20[b]	1.12[b]
Prebiotic	6.26[b]	1.13[b]
Enzyme	6.18[b]	1.11[b]
Probiotic+Prebiotic	6.22[b]	1.11[b]
Probiotic+Enzyme	6.20[b]	1.13[b]
Prebiotic+Enzyme	6.20[b]	1.12[b]
SEM	0.03	0.02
P Value	0.045	0.021

[a,b] Means within a column in each variable with no common superscript differ significantly (P≤0.05). SEM Standard error of means.

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