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A comparison study of probiotic, postbiotic and prebiotic on performance and meat quality of broilers

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ABSTRACT

Using of antibiotics it has started nearly eighty years ago since 1940 the purpose of using for increasing the immunity against microbes, diseases and to enhance growth in poultry production. The development in bacteria to become resistant against drugs results from using antibiotics for long period it may transformed to humans as well. "Biotic" feed additives, such as probiotics, prebiotics, and postbiotics, are one potential option for increasing poultry development and health. More attention has recently been paid to modern dietary solutions, especially probiotics, postbiotics and prebiotics. A300 one day old Ross 300 chicks distributed randomly into five treatment three replicates twenty birds in each one. The treatment groups include: negative control (a basic diet), positive control (a basic diet) + oxytetracycline 0.05%, T1= a basic diet + 0.3% lactobacillus plantarum (postbiotic), T2= a basic diet + 0.3% Bacillus licheniformis (probiotic), T3= a basic diet + 0.3% Fructo-oligosaccharides (prebiotic). As a result of adding the natural additives to the feed, Birds fed T3 gained more live body weight and had significantly higher ($p<0.05$) than negative and positive control groups at the same time had lower ($p<0.05$) FCR. Moreover, carcass weight and bursa of fabricius were higher ($p<0.05$) in birds that were fed with T1 when compared with negative control. The group of birds that fed with T1, T2 and T3 decreased ($p<0.05$) drip loss and coocking loss. These natural supplements can be added to enhance growth performance and meat quality in chicken production.

INTRODUCTION

Antibiotics widely used for over eighty years the purpose of using it was for increase the immunity against microbes, diseases and also used to enhance growth in poultry production. The development in bacteria to become resistant against drugs results from using antibiotics for long period it may transformed to humans as well (Tania *et al.*, 2018). World health organization noted public health concern because of the use of antibiotics within animal's diet. In 2006 European Union banned using antibiotic in the animal diet as promoters of growth (Castanon, 2007). United States of America in 2009 urges antibiotics must be forbidden as growth enhancers (Al-Khalaifa *et al.*, 2019). Biotic" feed additives, such as probiotics, prebiotics, and postbiotics, are one potential option for increasing poultry development and health. More attention has recently been paid to modern dietary solutions, especially probiotics, postbiotics and prebiotics. The International

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Scientific Association for Probiotic and Prebiotic defines probiotic as live bacteria it has beneficial health advantages to the host when it provided in appropriate proportions (Yeşilyurt *et al.,* 2021). When probiotics used as dietary supplements it leads to balance in the bacteria which exists in the intestine of the host (Hussein *et al.,* 2020). Utilizing probiotics improves intestinal resistance and morphology, as well as encouraging the host's metabolic function by reducing the risk of disease caused by harmful microorganisms (Caly *et al.,* 2015). One type of beneficial bacteria which is *Lactobacillus* used as probiotic for decades because of the beneficial impacts on performance, antioxidant capacity and increasing immunity of host (Gao *et al.,* 2017). Moreover, posbiotics are byproducts or metabolic products of bacteria or microorganism which has beneficial effect within the host (Zendeboodi *et al.,* 2020). Using Postbiotics are mostly associated with resistance actions, such as increasing the stability of the intestinal mucosal barrier and removing dangerous microbes with antimicrobial substances by enhancing immunity system of host (De Marco et al., 2018). In study which done by Kareem *et al* (2021) demonstrated that by using postbiotic and inulin together combined in dietary of broiler poultry it leaded to improvement in poultry growth performance and immunity. Definition of prebiotics is components of feed which is non-digestible extracted from sugars such as Fructo-oligosaccharides (FOS), galacto-oligosaccharides (GOS), and trans-galacto-oligosaccharides (TOS) (Sobolewska *et al.,* 2017). probiotics are basically fueled by prebiotics and after that postbiotics are delivered, they are essentially the "remains" or byproducts of probiotics. Antimicrobial metabolites found in postbiotics, such as natural acids and bacteriocins, can reduce the pH of the digestive system and limit the proliferation of infections in animal feed and gut (Aguilar-Toalá *et al.,* 2018). However, no study has been conducted a comparison between postbiotics, probiotics and prebiotics. Thus, the aim of this study is to make a comparison between probiotic postbiotic and prebiotic to determine which is most suitable for using as alternative for antibiotic in poultry production and meat quality characteristics.

MATERIAL AND METHODS

Animals and Experimental design: This study was conducted at Poultry house/ Animal resources Dept./ College of Agricultural Engineering Sci/ Salahaddin University-Erbil/ Iraq. A commercial hatchery provided 300 chicks Ross 308 one day old. Chicks were randomly distributed into five treatment groups. Each group comprises three replicates, with twenty birds in each. The treatment groups include: negative control (Basal diet), positive control (Basal diet) + oxytetracycline 0.05%, T1= Basal diet + 0.3% *lactobacillus plantarum* (postbiotic), T2= Basal diet + 0.3% *Bacillus licheniformis* (probiotic), T3= Basal diet + 0.3% FOS (prebiotic). Water and feed will be offered from the day one *ad libitum* to the birds until 35 days of age, the feed was prepared by a private feed company that contains 3072, 3122 kcal/kg metabolizable energy, 22.5, 20 % crude protein starter (1 - 18 d) and grower (19 - 35 d) diets respectively. Vaccination program was consisting of Newcastle at 7 and 21 days and IB at 14 days of rearing.

Samples and data collection: Growth performance was measured weekly (body weight (BW), body weight gain (BWG), feed intake (FI) and FCR in the study.

Carcass Characteristics: At the 35th day, parts of body were cut and weighed individually and these include; legs, internal organs (liver, gizzard, heart, and spleen), abdominal fat, breast, thigh and drumstick. The conversion of internal organs and carcass parts to the percentage was by following formula: Cut yield% = [Weight of cut/Empty live body weight] ×100

Measurement of Meat Quality

Water Holding Capacity (WHC): Based on Honikel (1998) method, WHC was calculated by means of drip loss and cooking loss. Drip loss and cooking loss were measured as described by Kareem et al., (2015).

Meat shear force measurement: the texture analyzer (CT3^{TM}, USA) which was equipped with Volodkevitch bite jaw used to measure shear force on samples which was used for cooking loss. From each sample blocks of 1cm (height) x 1cm (width) x 2cm (length). The sample blocks was placed on Volodkevitch bite jaw to record shear force and the result was expressed as g.

Muscle pH measurement: A total of 1 g of muscle tissue was taken and mixed in 10 ml cold deionized water for 30 seconds. pH meter (PHS-3C, China) used was pre-calibrated with a pH 4.0 buffer and then with a pH 7.0 buffer.

Color Measurement: A Color Flex spectrophotometer (Shenzhen 3nh Technology Co., Ltd, China) used tin laboratory discover color of the meat values which was known as (L*, a*, b*, c* and h*), L* (lightness), a* (redness), b* (yellowness), c* (chroma), and h* (hue) values were recorded in triplicate for each sample and then averaged.

The experiment's statistical analysis followed a completely randomized design. The data were analyzed using (SAS) computer software version 9.4's General Linear Model (PROC GLM) (SAS Institute, Inc., 2014). While the data obtained for meat quality were analysed with sampling time repeated measure. The test were used in study was Duncan multiple range to compare the means of the treatment at the level of probability of 5% ($P< 0.05$).

RESULTS AND DISCUSSION

Table 1. presents the growth performance indices of birds which fed with postbiotics, probiotics and prebiotics. Except for T2, birds fed T3 gained more total weight and had a higher final body weight (P<0.05). While, there was no significant difference ($P>0.05$) between T2 and T1 in comparison with negative and positive groups. FI was higher ($P<0.05$) than positive group at the same time there was no significant differences ($P>0.05$) in comparison with other treatments when birds fed with T2. FCR was lower in birds fed with T3 compared with birds fed with negative control. There was no significant difference ($p>0.05$) in FCR between birds fed negative control, positive control, T1 and T2.

Table (1): Effect of postbiotics, probiotics and prebiotics on BW, BWG, FI and FCR of broiler chicken at 35 day of age

Treatments	Initial BW	BW (g)	TWG (g)	FI (g)	FCR (g:g)
Negative control	38.66 ^{a}	2282.67 ^{b}	2244.00 ^{b}	3265.00$^{a\,b}$	1.46 ^{a}
Positive control	38.83 ^{a}	2264.33 ^{b}	2225.67 ^{b}	3176.67 ^{b}	1.43 ^{ab}
T1	37.83 ^{a}	2308.17 ^{b}	2270.33 ^{b}	3257.33 ^{ab}	1.44 ^{a}
T2	38.33 ^{a}	2348.50 ^{ab}	2310.33 ^{ab}	3273.00 ^{a}	1.42 ^{ab}
T3	38.87 ^{a}	2441.00 ^{a}	2402.00 ^{a}	3243.67 ^{ab}	1.35 ^{b}
SEM^{B}	0.001	0.018	0.019	0.014	0.012

^{ab}Means with various superscripts differ significantly (P<0.05) in the same column. ^{A}Negative control: a basic diet, positive control: a basic diet + oxytetracyclie, T1: postbiotic, T2: probiotic, T3: prebiotic ^{B}SEM: standard error of means (pooled).

Table 2. Presents the edible parts of birds provided diets with various additives. The gizzard weight of birds fed T1 was higher ($P<0.05$) than that of positive group birds. while there were no significant differences ($P>0.05$) in spleen and abdominal fat between treatment groups.

Table (2): Effect of postbiotics, probiotics and prebiotics on the percentage of edible parts %

Treatments	Heart	Liver	Gizzard	Spleen	Abdominal Fat	Bursa of Fabricius
Negative control	0.46 ^{a}	2.26 ^{a}	1.42 ^{ab}	0.10 ^{a}	0.65 ^{a}	0.10 ^{b}
Positive control	0.42 ^{ab}	2.28 ^{a}	1.31 ^{b}	0.08 ^{a}	0.74 ^{a}	0.12 ^{ab}
T1	0.40 ^{b}	2.18 ^{a}	1.66 ^{a}	0.10 ^{a}	0.73 ^{a}	0.15 ^{a}
T2	0.40 ^{b}	2.22 ^{a}	1.54 ^{ab}	0.08 ^{a}	0.80 ^{a}	0.13 ^{ab}
T3	0.40 ^{b}	2.44 ^{a}	1.54 ^{ab}	0.09 ^{a}	0.72 ^{a}	0.13 ^{ab}
SEM^{B}	0.008	0.049	0.047	0.004	0.034	0.007

^{ab}Means with various superscripts differ significantly (P<0.05) in the same column. ^{A}Negative control: a basic diet, positive control: a basic diet + oxytetracyclie, T1: postbiotic, T2: probiotic, T3: prebiotic ^{B}SEM: standard error of means (pooled).

Table 3. presents the relative carcass cuts of birds fed diets containing different additives. Birds served with the diets T1, T2 and T3 had higher (P<0.05) carcass weight than birds served with diets other treatments except negative control. There was no significant difference (P>0.05) between treatment groups in neck, wings and thigh with drumstick weight. Birds fed T2 and T3 had higher (P<0.05) breast weight than positive group birds (P>0.05) except T1 and negative control group birds. Furthermore, higher (P<0.05) back weight observed in the birds which were fed with the T3 than T1 except positive, negative control and T2 treatment groups.

Table (3): Effect of postbiotics, probiotics and prebiotics on the percentage of relative carcass cuts%

Treatments	Body weight	Carcass weight	Neck	Wings	Thigh with drumstick	Breast	Back
Negative control	2282 ^{b}	68.88 ^{ab}	3.68 ^{a}	7.00 ^{a}	20.40 ^{a}	25.77 ^{bc}	12.45 ^{b}
Positive control	2264 ^{b}	68.31 ^{b}	3.46 ^{a}	7.17 ^{a}	19.49 ^{a}	25.27 ^{c}	12.64 ^{b}
T1	2285 ^{b}	69.59 ^{a}	3.49 ^{a}	7.13 ^{a}	19.91 ^{a}	26.89 ^{ab}	11.72 ^{c}
T2	2348 ^{ab}	69.73 ^{a}	3.40 ^{a}	6.88 ^{a}	19.40 ^{a}	27.86 ^{a}	12.47 ^{b}
T3	2441 ^{a}	69.68 ^{a}	3.22 ^{a}	6.62 ^{a}	18.90 ^{a}	27.57 ^{a}	13.34 ^{a}
SEM^{B}	0.019	0.201	0.082	0.095	0.226	0.246	0.136

^{ab}Means with various superscripts differ significantly (P<0.05) in the same column. ^{A}Negative control: a basic diet, positive control: a basic diet + oxytetracyclie, T1: postbiotic, T2: probiotic, T3: prebiotic ^{B}SEM: standard error of means (pooled).

Table 4. presents drip loss, cooking loss, tenderness, pH & color of *pectoralis major* muscle from birds fed with the diets containing different additives. The meat from birds fed with T1, T2 and T3 diets by comparing it with meat from birds fed with negative control diets had lower (P<0.05) drip loss. Furthermore, birds fed with T1 and T3 diets had significantly lower (P<0.05) drip loss than positive control. However, no significant (P>0.05) difference observed among T1, T2 and T3 in. There was significant differences (P>0.05) observed among T1, T2 and T3 in cooking loss comparing with the negative control and positive control, however no significant differences (P>0.05) were recorded for shear force of *Pectoralis major* muscle of broiler chickens among the treatments.

Table (4): Effect of postbiotics, probiotics and prebiotics on drip loss, cooking loss, tenderness, pH and color

Treatments	Drip loss	Cooking loss	Tenderness	pH	L*	a*	b*
Negative control	4.30 ^{a}	25.65 ^{a}	333.08 ^{a}	6.09 ^{a}	51.76 ^{a}	10.52 ^{a}	9.05 ^{ab}
Positive control	4.16 ^{ab}	26.56 ^{a}	332.25 ^{a}	6.07 ^{a}	53.30 ^{a}	11.24 ^{a}	8.26 ^{b}
T1	2.91 ^{c}	20.02 ^{b}	332.41 ^{a}	5.87 ^{b}	47.28 ^{a}	10.80 ^{a}	10.72 ^{a}
T2	3.26 ^{bc}	21.89 ^{b}	333.25 ^{a}	5.96 ^{ab}	48.07 ^{a}	9.66 ^{a}	9.17 ^{ab}
T3	2.93 ^{c}	20.54 ^{b}	333.83 ^{a}	6.02 ^{a}	46.77 ^{a}	10.24 ^{a}	9.94 ^{ab}
SEM^{B}	0.17	0.69	0.34	0.02	1.08	0.59	0.32

^{ab}Means with various superscripts differ significantly (P<0.05) in the same column. ^{A}Negative control: a basic diet, positive control: a basic diet + oxytetracyclie, T1: postbiotic, T2: probiotic, T3: prebiotic ^{B}SEM: standard error of means (pooled). L*=lightness, b*=yellowness, a*=redness.

The results of pH and color values (L* a* b*) of *Pectoralis major* muscle in birds which were fed with postbiotic, probiotic and prebiotic shown in Table 5. The pH value at day 0 was significantly lower in the *Pectoralis major* muscle of broiler chickens fed of T1 compared to the *Pectoralis major* muscle from birds fed the positive and negative control diets and T2. There was no significant difference (P>0.05) in lightness and redness among all treatments. A significant decrease in b* (yellowness) (P<0.05) was found in the positive control in comparison with other treatments. While there were no significant differences (P>0.05) between the remaining treatments. In this study, using supplement postbiotic, prebiotic and probiotic lead to enhanced final body

weight of broiler chickens. It has been found using probiotics improves microorganisms balance within host animals intestinal and probiotics leads to stimulating growth and activity of certain type of bacteria in the colon of the host animal. The enhancement in the intestinal absorption which in result enhances the performance it has been assumed (Al-Khalaifa *et al.*, 2019). Similarly, in study which was done by Nabizadeh, (2012) supplementation of 1% inulin as prebiotic lead to enhancement in total body weight and body weight gain while there was no significant effect on the feed intake in broiler birds. In the study which was done by Kareem *et al.* (2021) using postbiotics had beneficial effect on improving growth of birds. Using postbiotics which may decrease reduplication of pathogenic microorganisms in the gut because postbiotic have properties of both bactericidal and bacteriostatic properties. By supplementation of probiotic and postbiotic microbiota will be balanced that is necessary for early development of the intestine could be lead to higher feed intake then improving performance.

It has been reported that type of postbiotic *L. plantrum* has inhibitory effect on pathogenic microorganisms (Choe *et al.*, 2013; Kareem *et al.*, 2014). Results of study could be related to postbiotics leads to enhancing gut health and growth performance by decreasing the population of pathogenic microorganism in the gut. In addition the study which is done by Kareem *et al.* (2016) using combination of postbiotic and inulin as dietary supplement lead to enhancing growth performance and diet efficiency in poultry bird using postbiotic leads to significant difference in final body weight compared with positive and negative control. In contrast, in a study was done by Rahimi and Khaksefidi. (2006) no significant differences has been observed in bird growth performance under heat-stress conditions between two groups of probiotic and antibiotic. In this study, the T3 birds had the lowest FCR by comparing to negative control birds. This observation in agreement with the study of Kridtayopas *et al.* (2019) who reported prebiotic supplementation improved FCR under high stocking density conditions ($P < 0.05$).

Dietary supplementation of prebiotic had effect on carcass yields such as carcass weight in broiler chickens. Supplementation of prebiotics leads to improve weight gain and feed intake maybe because of this the carcass weight improved as well. In agreement with Rocha et al. (2010) who found the birds fed with mixture of probiotics lead to increasing breast yield at the age of 43 days, while without mixture of probiotics lead to decrease in yield of carcass. In contrast Sahin *et al.* (2008) and Chumpawadee *et al.* (2008) they found that no significant effect on yields of carcass in birds by using probiotic, prebiotic and synbiotic. In another study which was done by Nunes *et al.* (2012) found no significant effect on thigh yield by using growth promoters. Also in contrast Humam *et al.* (2019) who assumed using postbiotic as a fed additive lead carcass weight to significantly increase in birds fed with postbiotic compared with positive and negative control.

One of the most essential qualitative features in consumer's final decision is tenderness on a particular cut of poultry muscle (Fletcher, 2002). The birds which fed with postbiotic, prebiotic and probiotic had lower drip loss which support the findings of study was done by Kareem *et al.* (2015) who reported that the effect of "postbiotic and inulin" supplementation on drip loss, cooking loss and shear force in chicken breast meat are shown postbiotic and inulin additive groups a significant difference was observed in drip loss compared with control groups. In agreement with Ali (2010) who reported that bird fed with probiotic had lower cooking loss than control birds. Conversely, in another study which was done by Khalafalla *et al.* (2011) who found that probiotic and prebiotic had no effect on cooking loss of broiler breast meat. Zhou *et al.* (2010) also reported that basal diet supplemented with probiotic, *B. coagulans* ZJU0616, had beneficial effects on the shear force of chicken meat. Study found that using probiotics lead to converting fat in the meat into the favorable fat, which leaded to participating tenderness in the meat. Mixing *Clostridium butyricum* which is type of probiotic in the diet leaded to enhancement in tenderness in the broiler birds. In contrast to traditional basal diet, using probiotic *Lactobacillus* in diet of broiler leaded higher appearance, texture and overall acceptability (Yang *et al.*, 2010). According to Hossain *et al.* (2012) it was observed that pH values at day 0 significantly decreased with treatment contains postbiotic and prbiotic compared to control groups except treatment contains prebiotic. Likewise, one of the important indicators for meat quality is muscle pH, breast meat having a pH of 6.0 at 15-30 min is

described as being of high quality (Alvarado *et al.,* 2007; Fernandez *et al.,* 1994). The pH values observed for all additives in this study were around (6.0). This lower pH values observed for prebiotic in agreement with the findings of type of prebiotics used did not influenced pH 24, also this study in agreement with the study of Kareem *et al.* (2015) the pH value at 0 day was significantly decreased with all postbiotic and inulin additives compared with the control groups. Study in agreement with the study of Maiorano *et al.* (2012). While in disagreement with Sang-Oh and Byung- Sung (2011) reported that using prebiotic in the dietary lead to decreasing in pH of broiler meat. Quite the reverse, Pelicano *et al.* (2003) did not observed significant differences in muscle pH between control birds and those fed probiotics. Color of meat is critical parameter affecting customer decisions (Tavaniello *et al.,* 2018). Color of the meat used as important indicator of quality of the meat of broiler. Age, sex, nutrition, processing and pH effect on the color of the meat in broilers (Ozturk *et al.,* 2012). In this study birds fed positive control, T2 diet had lower yellowness than birds who fed with negative control. However, no significant difference has been observed in lightness among all treatment groups. In broilers normal color of breast meat has an L* of approximately equal 55 while L* values more than 60 leads to be pale appearing (Schilling *et al.,* 2008; Van Laack *et al*., 2000). Moreover, L* value the results of the current study were within the standard range ($46 \leq L^* \leq 51$). L* (lightness) is a breast meat color scale in which the color of the breast meat is described as dark, normal, or pale when the L* indication reaches the point < 50, ($50 \leq L^* \leq 56$) or > 56, respectively (Petracci *et al.,* 2004). This study is contrast with study done by Zhao *et al.* (2012) found that using prebiotics in the dietary significantly affects the redness ($a*$) of the fillets.

CONCLUSION

This study aimed comparison among probiotic postbiotic and prebiotic to determine which is most suitable for using as alternative for antibiotic in poultry production and meat quality characteristics. Dietary supplementation with postbiotic, probiotic and prebiotic have enhanced growth performance, carcass quality, and meat quality in broilers. These natural supplements can be added to enhance growth performance and meat quality in chicken production.

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دراسة مقارنة بينوبوستبيوتيك و بروبيوتيك و بريبيوتيك على الأداء و جودة اللحم في فروج اللحم

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الكلمات المفتاحية:
بوستبيوتيك ، بريبيوتيك ، بروبيوتيك ، أداء النمو ، جودة اللحم.

الخلاصة

استخدمت المضادات الحيوية منذ ما يقرب من ثمانين عامًا منذ أربعينيات القرن الماضي، وكان الغرض من استخدامها زيادة المناعة ضد الميكروبات والأمراض، كما تستخدم لتعزيز النمو في إنتاج الدواجن. إن تطور البكتيريا لتصبح مقاومة للأدوية ناتج عن استخدام المضادات الحيوية لفترة طويلة وقد تتحول إلى الإنسان أيضًا. حيث تعتبر إضافات الأعلاف "الحيوية"، مثل البروبيوتيك ووالبوستبايوتك والبريبايوتك، أحد الخيارات المحتملة لزيادة نمو الدواجن وصحتها. تم الاهتمام مؤخرًا للحلول الغذائية الحديثة، وخاصة البروبيوتيك والبريبايوتكس والبريبايوتكس. جلب 300 فرخة (Ross- 308) بعمر يوم واحد من مفقس أهلي. حيث تم توزيع الأفراخ إلى خمس معاملات عشوائيا. تكونت كل مجموعة من ثلاث مكررات، في كل منها عشرين طائرًا. تشمل المعاملات: محموعة السيطرة السالبة (العليقة الأساسية+ أوكسي تتراسيكلين بتركيز 0.05٪)، محموعة السيطرة الموجبة (العليقة الأساسية+ ٪ Lactobacillus)، T1 = العليقة الأساسية + 0.3٪ (postbiotic Lactobacillus plantarum، T2 = (العليقة الأساسية + 0.3٪ Bacillus licheniformis (بروبيوتيك)، T3 العليقة الأساسية + 0.3٪ فركتو اوليكوسكرايد (بريبايوتك). كانت نتيجة الإضافات الطبيعية إلى علف فروج اللحم ارتفاع معنوي ($p < 0.05$) في وزن الجسم في المعاملة T3 مقارنة بمحموعة السيطرة السالبة والموجبة، في نفس الوقت تحسنت معامل التحويل الغذائي FCR معنويا ($P < 0.05$) ، علاوة على ذلك كانت وزن الذبيحة والنسبة المئوية لجراب فابريشيا أعلى في الطيور التي تم تغذيتها في T1 مقارنة مع السيطرة السالبة. مجموعة الطيور التي تغذت على T1 و T2 و T3 انخفضت ($P < 0.05$) من فقدان التنقيط وفقدان الطفو اثناء الطبخ. يمكن إضافة هذه المكملات الطبيعية لتحسين أداء النمو وجودة اللحم في الدجاج .