

COMPARISON BETWEEN YOGHURT POWDER (AS PROBIOTIC) WITH IMPORTED PROBIOTIC AGAINST PATHOGENIC BACTERIA IN THE SMALL INTESTINE OF BROILERS

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ABSTRACT : The research has been conducted to evaluate the number of benefits bacteria (total bacteria, lactic acid bacteria and *Escherichia coli*) in the gastrointestinal tract of broilers fed diets supplemented with yoghurt powder loaded with wheat bran, yellow corn and lentil. 210 one day old broilers Ross 308 were used in this study. There were seven experimental diets namely, each containing three replications (10 chicks in each replicate). The control treatment was supplemented with a basal diet without any addition, while the T2, T3, T4 treatments were fed diets supplemented with 3% of lentil, yellow corn and wheat bran loaded on the yoghurt powder (30 mg/kg diet), respectively. Also, T5, T6, T7 treatments were fed diets supplemented with 0.05% of imported probiotics, which is Labazyme, Biolac and Interzym (0.5 mg/kg diets), respectively. At 35 days of age, the lactic acid bacteria significantly increased ($P<0.05$) in broiler fed wheat bran, labazym, yellow corn and lentil respectively compared to the control and other treatments. But the number of total bacteria significantly decreased ($P<0.05$) in wheat bran, yellow corn, lentil and labazym treatments compared to the control and other imported probiotics treatments. On the other hand, *Escherichia coli* in the broiler gut significantly decreased ($P<0.05$) in the wheat bran, lentil and yellow corn compared to the control and other treatments, respectively. However, the pH more decreased in treatment (4) compared with control and other treatments especially in jejunum and ileum of the intestinal tract of broilers.

Key words : Broiler, yoghurt, wheat bran, yellow corn, lentil, lactic acid bacteria and *Escherichia coli*.

INTRODUCTION

At the moment, attention focused on replacing imported feeds with alternative feeds at affordable prices as the production of poultry feed approaches 70 percent of total production costs (Abd El-Gawad *et al*, 2014). Increasing evidence shows pathogenic bacteria regulate and colonize the gastrointestinal tract in poultry and causes significant losses in the poultry industry. *Escherichia coli* (Ballou *et al*, 2016) is an impotent pathogenic bacterium. *Escherichia coli* is a pathogenic bacterium that affects the health of humans and livestock. Livestock is the carrier of these bacteria, where they reside in the intestinal tract and then contained in human-eaten meat (van den Bogaard *et al*, 2001).

All animals, including avian species have a digestive system has a dynamic property that adjust itself according to current circumstances and physiological requirements. The comparatively acidic (pH) of the birds gastrointestinal tract (GIT) also depends on certain factors like poultry nutrition, nutrients form and more specifically (GIT) content of microflora. A mutual correlation exists between content microflora and pH with

nutrients and microflora (Sarraf *et al*, 1985). In specified (GIT) areas the pH level is a factor that determines a particular population of microbial. Additionally, pH level influences absorption value of maximal nutrients and the digestibility. The pathogens mostly grow near (7) pH or slightly higher. On the other hand, beneficial microorganisms survive in pH (5.8-6.2) and competition with pathogens, so reducing pH improves the absorption of nutrients (Boling *et al*, 2001).

Previous studies reported that fermentation foods have decreased crude fiber content but increased crude protein content, so yogurt is fermented probiotic products that have an excellent anti bactericidal effect on foodborne pathogens (Yesillik *et al*, 2011). It also contains a large number of lactic acid bacteria that function as probiotics in chickens ' intestines which control microflora and works to exclude pathogenic bacteria from the digestive system.

Wheat bran is rich in B vitamins and minerals. Bran is made up of more than half water-insoluble fiber, bran made up of fiber components (53%). While the chemical composition of wheat bran fiber is complex, it primarily includes cellulose and pentosans that closely linked to

proteins. Each protein and carbohydrate represent 16% of total bran dry matter, but the mineral content is rather high (7.2%). This explains a branch's very high protein and carbohydrate rates (Cornell, 2003).

Corn is one of the most critical feedstuffs used to feed poultry and is an important energy source (Ali, 2019). Lentil is a source of protein imported (22-29%) and feed for poultry birds. Adding lentil is an excellent source of micronutrients and phytochemicals from cottages (Rochfort and Panozzo, 2007). Lentil contains high amounts of antioxidant ingredients such as condensed tannins and phenolic acids (Xu and Chang, 2010). The objective of this study was to investigate the effect of feeds such as wheat bran, yellow corn and lentil loaded on yogurt powder and pH of GIT content on the number of lactic acid bacteria and to study adverse effects on pathogenic bacteria such as *Escherichia coli*.

MATERIALS AND METHODS

Experimental design

A total of 210 broiler chicks (ROSS 308) one day old, weighing rate (40 g/chicks) were divided randomly to seven experimental groups (n=30). Each group contains three replicates (10 birds/replicate). This study used the powder of yoghurt in amount 9 gm mixing with 992 gm of lentil, yellow corn and wheat bran powder. Then 3 gm from this mixing per kg feed was added to the basal diet.

The broiler chicks were randomly allotted to one of the seven experimental diets namely, the first treatment a concentrate mixture without any addition (control T₁). The second treatment (T₂) was fed concentrate mixture +3 gm/kg from the powder of yoghurt and lentil. The third treatment (T₃) was fed concentrate mixture + 3 gm/kg from powder yoghurt and yellow corn. The fourth treatment (T₄) was fed concentrate mixture + 3gm/kg from powder yoghurt and wheat bran. The fifth treatment (T₅) was fed concentrate mixture + 0.5 gm/kg from imported probiotic (Labazym). The sixth treatment (T₆) was fed concentrate mixture + 0.5 gm/kg from imported probiotic (Biolac). The seventh treatment (T₇) was fed concentrate mixture + 0.5 gm/kg from imported probiotic (Interzym).

The experimental diets formulated to meet the nutritional requirements of commercial broilers during starter stage (1-21 days) and the finisher stage (22-35 days) (Table 1) according to NRC (1994).

The chickens were slaughtered after 35 days and the intestinal contents of 6 birds were collected from each treatment and placed in plastic containers and closed tightly. Then the total bacteria were determined, one ml

Table 1 : Chemical composition of dietary treatments (starter and finisher) diets.

Ingredient (%)	Starter diet (1-21d)	Finisher diet (21- 35 d)
Yellow corn	42.75	41.75
Wheat	15.0	22.0
Soybean meal (44%)	34.0	27.0
Broiler protein concentrate (48%)	5.0	5.0
Vegetable oil	0.8	2.3
Premix	0.2	-
Limestone	1.5	1.7
Common salt	0.25	0.25
Total	100	100
Calculated composition		
ME(Kcal/Kg diet)	3010	3174
Crud protein	23.10	20.14
Calorie: protein ratio	130.30	157.60
Calcium (%)	0.93	0.99
Phosphorus available (%)	0.42	0.51
Lysine (%)	1.35	1.17
Methionine+ Cystine(%)	0.89	0.83

* ME: Metabolizable energy

from mixed (yellow corn, lentil and wheat bran addition to yoghurt powder) to 9 ml from sterile peptone water. Then mix with nutrient agar in petri dishes. Whenever the count of *E. coli* same to the previous one, but mixed with macconky agar and put in the incubator at 37°C for 24 hours by using the pour plate method. Colonies have been expressed and counted as colony forming unit (CFU). When enumerating lactic acid bacteria was used, 1 ml from mixing material added to 9 ml sterile peptone water after this put in the petri dishes with MRS agar, also incubated for 48 hours and then are counted by the bacteria counting device. In the experiment was determined the pH of the content of the small intestine (jejunum, ileum) of broilers by use Senso Direct pH 200 made from Lovibond company.

Statistical analysis

Review of all treatments in Table 2 was performed on data using Completely Randomized Design (CRD) using qi square but table (3) using SPSS Software (2012) ANOVA in one way.

RESULTS AND DISCUSSION

The effects of dietary supplementation on pH are shown in Table 2. Chickens fed T₃, T₄ and T₅ diet had significantly decreased pH in jejunum and ileum part of the small intestine in chickens compared with those fed other diets and control diet.

This observation could be attributed to the possible initiation by the LAB of the development of other

Table 2 : showed the effect of yogurt loaded with some legume and cereal (lentil, yellow corn and wheat bran) and compared with imported probiotics (labazym, biolac and interzym) on the ph of ileum and jujenum of broiler chickens.

Treatment	Jujenum	Ileum
T1	6.1 ^b ± 0.260	6.18 ^b ± 0.213
T2	5.75 ^b ± 0.164	5.85 ^c ±0.104
T3	5.55 ^c ± 0.266	5.56 ^c ±0.233
T4	5.05 ^c ± 0.151	5.15 ^f ±0.104
T5	5.03 ^c ± 0.236	5.38 ^f ± 0.248
T6	6.11 ^{ab} ± 0.213	6.25 ^b ± 0.258
T7	6.56 ^a ±0.108	6.61 ^a ±0.194
LSD	0.366	0.283

Deferent small letters means significantly between treatments.

antimicrobial metabolites, which have had a major impact on the overall count of bacterial and increased inhibition pathogenic bacteria. Although, bacteria of Lactic acid (LAB) and other bioactive components that inhibit these species. Some of these bioactive components might be bacteriocins that are antimicrobial protein components that are inhibitory to susceptible organisms (Ogunbanwo *et al*, 2003) and function by the membrane destroying (Savadago *et al*, 2006).

However, there is a reciprocal connection between the quality of PH and microflora with micro flora and nutrient (Sarra *et al*, 1985).

Into particular gastrointestinal tract areas, the level of pH is a factor that determines specific microbial community and also influences absorption value of most nutrients and the digestibility. Mostly, the grow of pathogens at 7 pH or slightly higher. In comparison, advantageous microorganisms reside in pH (5.8-6.2) and interact with pathogene (Ferd, 1974), thus reducing pH in organic acids (as yoghurt) also increases the absorption of nutrients (Boling *et al*, 2001).

The effect of dietary supplementation on a number of bacteria total bacteria, lactic acid bacteria and *Escherichia coli* (Table 3). Significantly decreased ($P < 0.05$) in a number of total bacteria in treatment (T_6 , T_5 , T_4 , T_2 and T_3) compared with control and T_7 treatments. This observation could be due to the decreased pH for food supplemented compared with other diets. The reduction in the population of total bacteria in treatment (T_6 , T_5 , T_4 , T_2 and T_3). The current observation is in tandem with the findings of Savadago *et al* (2006), who observed that PH for food supplemented is decreased and it is responsible for the inhibition of total bacteria.

The LAB in diets not only decreases intestinal pH by going production of organic acid but also prevents enteropathogens from colonizing through bacteriocin

Table 3 : Effect of wheat bran, yellow corn and lentil mixed with powdered yoghurt and imported probiotic (labazym, biolac and interzym) on total bacteria ,lactic acid bacteria and *E. coli* bacteria.

Treatment	Total bacteria ×10 ⁹	Lactic acid bacteria ×10 ⁸	<i>E. coli</i> ×10 ⁶
T ₁	2.84 ^a	1.92 ^d	74.75 ^a
T ₂	1.39 ^c	2.06 ^c	8.5 ^c
T ₃	1.98 ^b	2.97 ^c	42.5 ^b
T ₄	1.28 ^d	9.51 ^a	4.0 ^e
T ₅	1.26 ^d	4.21 ^b	31.5 ^c
T ₆	0.67 ^e	0.55 ^e	45.50 ^b
T ₇	2.42 ^a	0.50 ^e	66.9 ^a
X ²	9.81 [*]	22.95 ^{**}	125.78 ^{**}

T = treatment, X² = mean, * = mean average between all type of bacteria, small latters mean significantly effect between experimental dietary treatments.

production and competitive exclusion, antagonistic activity. Besides, the administration of feed contains yogurt raises the chicken gut LAB population (Savvidou *et al*, 2009; Sun *et al*, 2013). The lower pH in diets acidifies the higher gastrointestinal tract, thus improving a gizzard's barrier function against pathogens. As found in the current study, yogurt in broiler diets also creates unfavorable environments for the proliferation of certain enteropathogens, such as *Escherichia coli* (Liang *et al*, 2012). As well as, the result showed significantly decrease ($p < 0.05$) in a number of *E. coli* in T_2 compared with other treatments except for T_4 , this effect due to lectin material which found in the lentil was acting as an inhibitor to pathogenic organism such as *E. coli* (Sindhu *et al*, 2013).

In summary, the present study showed that use yoghurt powder (as probiotic) in the diet had beneficial effects on reduced the concentration of pH in jejunum and ileum part of the small intestine in chickens compared with other diet and reducing the population of total bacteria in broiler chickens.

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