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ROLE OF PROBIOTICS IN IMPROVING MICROBIAL LOAD OF BROILER CHICKEN CARCASSES

EL-DOSOKY, H.F.A.; SANYA, T. EL-GHAMRY and AZZA, E.A. HASSAN
Animal Health Research Institute, Mansoura branch
Email: nogoom.1980@gmail.com

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ABSTRACT

A total number of 90 one day old chicks were purchased from a poultry company at Mansoura city and divided into three groups (each of 30). All groups were prophylactically vaccinated according to the local routine vaccination programme. The first group (control group) was fed a basal diet (without probiotics), whereas 2nd&3rd groups were fed on the same basal diet supplemented with probiotics. At 42ndays of age, broilers were slaughtered, processed and bacteriologically examined for aerobic plate count *(APC),* enumeration of *Coliforms (MPN), E. coli* count, detection of *Salmonellae, Campylobacter jejuni* and *E. coli* where the log mean of *APC* in group 1, group2 and group3 were 5.5 ±0.9, 4.9 ±1 and 4.8 ±0.8 log_{10}cfu/ gm, the *MPN of Coliforms* were 3.4 ±0.9,3 ±0.9 and 2.7± 0.8 log_{10}cfu/ gm, *E. coli* count were 3.3 ±1, 3 ±0.8 and 2.9±0.7 log_{10} cfu/ gm with reduction percent from 40% to 30% and 33.3%, meanwhile The incidence of *Salmonellae* were reduced from 30% to 16.6% and 10% and *Campylobacter jejuni* incidence were reduced from 20% to 6.6% and 3.3% respectively. The experiment was triplicated and the results in group2 and group3 were reduced significantly (P<0.05).

Key words: *Probiotics, Microbial load, Broiler chicken*

INTRODUCTION

Probiotics are defined as viable microorganisms (bacteria or yeast) that exhibit a beneficial effect on the health of their host when they were ingested (Salminen *et al.,* 1998). The use of probiotics have started after the study reported by (Nurmi and Rantala, 1973) where the original objective was to control *Salmonella* infection and occupied the adhesion sites on intestinal epithelium, hence addition of probiotics have the objective of preventing the intestinal colonization of enteropathogenic bacteria in birds since probiotics and pathogenic bacteria compete for nutrients (Silva, 2000). Using of probiotics have been extensively studied world wide as a possible alternative for antibiotics by Jernigan and Kornegay (1985); Stavric and Kornegay (1995); Newman and Jacques (1995); England *et al.* (1996) and Pelicano *et al.* (2002). Jin *et al*. (1996) found that inclusion of probiotics (Lactobacilli and Bacillus subtilis) in the diet stimulated the favorable microbial balance in the gut leading to improving food efficiency and growth performance in broilers, therefore there is a world trend to reduce the usage of antibiotics in animal feed due to residues problems in the final product, most of broiler industry practitioners have been given a growth promoter as an additive in the ration Menten (2001 and 2002).

Therefore, the aim of this work was to evaluate the effect of probiotics as feed supplement on the bacteriological status of broiler chicken carcasses.

MATERIALS and METHODS

A total number of 90 one-day-old Cobb chicks were purchased from a poultry company at Mansoura city and divided into three groups. All groups were prophylactically vaccinated according to the local routine vaccination programme. Chicks were reared in three separated pens (3x2m) under good hygienic conditions.

The first group (control group) was fed on a basal diet (without growth promoters) which mixed first, placed in a clean unused sacks.

The second group was fed on the same basal diet supplemented with *Bacillus subtilis* spores $4x10^5$CFU/gm at 1.5 kgm/ton (Megalo, Amoun Man. Add. EL-obour city, Cairo, Egypt) added to the diet all over the period according to the manufacturer's instructions and thoroughly mixed, placed in a clean unused sacks.

The third group was fed on the same basal diet contained similar proportion of six strains of variable

organisms namely *Lactobacillus acidophilus* and *Lactobacillus casei, Bifidobacterium bifidum, Aspergillus oryzae, Streptococcus faecium* and *Torulopsis sps* added to the diet at 100 mg/kg diet all over the period of the experiment and thoroughly mixed, placed in a clean unused sacks and stored at room temperature.

The chicken fed on the aforementioned diets from 1st to 42nd day of age. The chicken was allowed to have free access to a starter diet with crude protein (21.5%) during the first three weeks and then to a finisher diet with crude protein (19.5%) during the second three weeks and free access of water. The experiment was repeated three times. There was a complete separation between different groups, the house was locked at all times to prevent unauthorized entry. Prior to initiating this study the house was thoroughly cleaned, sanitized, disinfected and new wood shavings litter were placed. At 42nd of age, broilers were slaughtered and processed then placed on ice till bacterial examination. All equipments were cleaned, sanitized using commercial sanitizer after each time.

Bacteriological Examination:
muscle and skin tissues (25gm) from each chicken were homogenized with 225ml of 0.1% peptone water in a stomacher for 2.5 minutes at 3000 rpm followed by ten fold six serial dilution in 0.1% peptone water. Each of the prepared samples were examined for enumeration of its bacterial content as follows:

1- Aerobic plate count according to APHA (2001)

2- Enumeration of Coliforms according to FDA (2005)

3- Escherchia coli counts according to FDA (2005)

4- Isolation and identification of Salmonellae and Campylobacter jejuni were done according to the techniques recommended by FDA (2005) and Smibert (1984) respectively.

RESULTS

Table1: Illustrates statistical analytical results of Aerobic plate count, Coliforms (MPN) and E. coli of the examined broiler carcasses fed on probiotics expressed as log cfu/gm (n=30).

Microbial count Logcfu/gm ±S.E.	Group1 (control group)	Group2 (treated feed)	Group3 (treated feed)
Aerobic plate count	5.5 ±0.9	4.9 ±1*	4.8 ±0.8 *
MPN of Coliforms	3.4 ±0.9	3 ±0.9 *	2.7±0.8 *
E. coli count	3.3 ±1	3 ±0.8*	2.9±0.7 *

N.B. APC=aerobic plate count, MPN=most probable number of Coliforms, *Means the results were significantly different ($P<0.05$).

Table 2: Shows the incidence of the tested bacteria in the examined +ve samples.

Microbial incidence	Group1 control group		Group2 (treated feed)		Group3 (treated feed)	
	No	%	No	%	No	%
E. coli	12	40	9	30	10	33.3
Salmonellae	9	30	5	16.6	3	10
Campylobacter jejuni	6	20	2	6.6	1	3.3

DISCUSSION

Probiotics were used to get rid of abnormalities in the gastrointestinal tract produced by stress and therefore normalize the gut activity (Kutlu and Gorgulu, 2001), hence the achieved results gave a profile about the effect of some probiotics on microbial content of broiler carcasses. The results in Table (1) showed that the mean of APC in group 1 (control group), group 2 and group 3 were 5.5 ±0.9, 4.9 ±1 and 4.8 ±0.8 $\log_{10}$cfu/ gm respectively, the results were reduced significantly (P<0.05) and in accordance with Fritts *et al*. (2000) who achieved reduction in APC, from 4.34 to 4.17 $\log_{10}$cfu/gm in the examined broiler chicken carcasses, Ali (2010) APC reduced from 6.35±0.72 to 4.81±0.25 $\log_{10}$ cfu/gm, after addition of *bacillus subtilis spores* to the chicken diet and Khaksefidi and Rahimi (2005) after using a basal diet contain *Lactobacillus acidophilus* and *Lactobacillus casei, Bifidobacterium bifidum, Aspergillus oryzae, Streptococcus faecium* and *Torulopsis sps*. The results were reduced significantly (P<0.05) in chicken fed on diet supplemented with probiotics as compared with control group.

Table (1) declared that the mean MPN of *Coliforms* were 3.4 ±0.9,3 ±0.9 and 2.7±0.8 $\log_{10}$cfu/gm respectively, where the results were reduced significantly (P<0.05) in agree with *Fritts et al. (2000)* who achieved *Coliforms* reduction from 2.37 to 2.12 $\log_{10}$cfu/ml in the examined broiler chicken carcasses, Khaksefidi and Rahimi (2005) from 2.52 to1.55 log cfu /ml and so Ali (2010) from 3.50±0.20 to 2.40±0.20 $\log_{10}$cfu /gm.

The obtained results of *E. coli* count in Tables (1&2) were significantly reduced (P<0.05) where the counts were 3.3 ±1, 3 ±0.8 and 2.9±0.7 $\log_{10}$ cfu/gm with reduction percent from 40% to 30% and 33.3%respectively as those obtained by Fritts *et al*. (2000) who reported tha*t E. coli* reduced from 2.58 *t*o 2.27 $\log_{10}$cfu/gm after probiotics addition to the diet, also Ali (2010) mentioned that *E. coli* incidence reduced from 40% to 33.3%.

The incidence results in table (2) declared that using of probiotics reduce *Salmonellae* from 30% to 16.6% and 10% respectively these results were similarly to those recorded by Caramori, (2001) who used probiotics in flocks challenged with *Salmonella enteritidis*, Fritts *et al.* (2000) obtained 40% reduction in *Salmonella spp.,* Maruta *et al.* (1996), Khaksefidi and Rahimi (2005) and Caramori *et al. (2005)* reported 60% reduction of experimentally infected broilers with *Salmonella enteritidis* and Ali (2010) recorded that probiotics feeding reduce *Salmonellae* incidence from 33.3% to 20%.

The incidence results of *Campylobacter jejuni* (C. jejuni) in table (2) after probiotics feeding in chicken carcasses were reduced from 20% to 6.6% and 3.3% respectively similarly to Fritts *et al.* (2000) who reported reduction from 3.43 to 2.85 $\log_{10}$cfu/ml, while Maruta *et al.* (1996) and Khaksefidi and Rahimi (2005) found reduction from 3.04 to 2.67 log cfu /ml respectively and Ali (2010) obtained reduction from 16.6% to non detected.

The bacteriocidal effect of probiotics were probably accompanied with production of antibodies this confirmed the hypothesis of Chaveerach *et al*. (2004) and the reduction of microbial load may be due to production of different antimicrobial components by probiotics such as organic acids, hydrogen peroxides, carbon peroxides, diacetyl, low molecular weight antimicrobial substances, bacteriocins and adhesion inhibitors. Some bacteriocins produced by specific probiotics strains can fulfill a role in the inhibition of common broiler pathogens, this agree with Andreatti and Sampalo, (1999), Avonts and De Vuyst (2001) and Meurman (2005) and stimulate intestinal immunity (Ouwerhand *et al.,* 1999) in addition to the results reported by Chiang and Hsieh (1995) and Takahashi *et al*. (2005) about broilers fed probiotic-supplemented diet showed better carcass yield and meat quality when compared to the broilers fed the unsupplemented diet.

In conclusion, probiotics addition to chicken diet as a feed supplement were recommended to improve the growth, microbial status and carcass yield of broiler chicken carcasses.

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دور البروبيوتك في تحسين الحمل البكتيري لذبائح بداري التسمين

حاتم فتحي احمد الدسوقي ، سنية طه الغمري ، عزة السيد علي حسان
Email: nogoom.1980@gmail.com

اشتملت الدراسة على عدد تسعين كتكوت عمر يوم تم تجميعها من احدى شركات الدواجن بمدينة المنصورة حيث تم تقسيمها إلى ثلاث مجموعات (ثلاثون كتكوت لكل مجموعة) وتربيتها وتغذيتها منفصلة علي عليقة أساسية للمجموعة الأولي أما المجموعة الثانية والثالثة تم تغذيتها علي العليقة الأساسية مضافا عليها البروبيوتك وبعد مرور٤٢ يوم تم ذبح كل مجموعة منفصلة داخل المجزر وبعد انتهاء مراحل الإعداد والتجهيز قبل التغليف مباشرة تم اخذ العينات للفحص البكتيري لمعرفة العد الكلي للميكروبات الهوائية والعد الكلي لميكروب الايشريشيا كولاي والعد الاحتمالي للميكروبات القولونية ومعرفة مدي تواجد ميكروبات السالمونيلا والكامبيلوباكتر جيجيوناي حيث أظهرت النتائج بعد تكرار التجربة ثلاث مرات وجود تأثير واضح للبروبيوتك علي الإقلال من تواجد البكتيريا السابقة ، حيث إن متوسط العد الكلي للميكروبات الهوائية كان ٥,٥±٠,٩، ٤,٩± ١ و ٤,٨±٠,٨ / جم، متوسط العد الكلي لميكروب الايشريشيا كولاي كان ٣,٣± ١، ٣±٠,٨ و٢,٩±٠,٧ / جم والعد الاحتمالي للميكروبات القولونية كان ٣,٤±٠,٩، ٣±٠,٩، و٢,٧±٠,٨ /جم علي الترتيب للمجموعات الثلاث بينما تم عزل ميكروب السالمونيلا والكامبيلوباكتر جيجيوناي من العينات السابقة بنسبة اقل من المجموعة الأولي مما سبق يتضح أن إضافة البروبيوتك له تأثير واضح علي الإقلال من تواجد البكتيريا وتحسين صفات لحوم الدواجن.