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EFFECT OF KERINCI DUCK'S INTESTINAL PROBIOTIC (*Bacillus Circulans* And *Bacillus Sp*) AS FEED ADDITIVE ON BROILER PERFORMANCE

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ABSTRACT

The aim of this study was to determine the effectiveness of probiotics *Bacillus circulans* and *Bacillus sp* as feed additive on broiler performance. The study was assigned using completely randomised design with 5 treatments and 4 replications. The treatments were R-0 (Basal diet/RB without probiotics and antibiotics), R-1 (RB + 35 ppm of Oxytetracycline in the drinking water), R-2 (RB + 35 ppm of Virginiamycine in the ration), R-3 (RB + 5 cc of liquid probiotic per litre drinking water) and R-4 (RB + 5 g of probiotics powder per kg ration). The measured parameters were broiler performance, amount of *Bacillus circulans*, *Bacillus sp* and *Escherichia coli*, total cholesterol in meat and antibiotic residue in meat. Results of this study showed that there was no significant effect ($P>0.05$) on broiler performance (feed intake, body weight gain and feed conversion ratio). However, the treatments highly significant ($P<0.01$) have decreased total cholesterol in meat and amount of *Escherichia coli*. Amount of *Bacillus* and lactic acid bacteria were highly significant ($P<0.01$) increased by the treatments. Oxytetracycline residue was not detected in meat, whereas, virginiamycine was detected. It is concluded that probiotics can be used in the broiler ration as feed additive and effectively decreased total cholesterol of meat, amount of pathogenic bacteria in broiler and prevented the oxytetracycline residue accumulation.

Keywords: Antibiotics, Broiler Performance, Cholesterol.

INTRODUCTION

Probiotics is living microbes those can increase human and animal health by balance micro flora in digestive tract when it is consumed in lives condition and enough amounts (Fuller, 1992). Probiotics products generally is lactic acid bacteria (LAB) and some Bacillus. However, it is not all of lactic acid bacteria and Bacillus bacteria are considered probiotics.

One of probiotics products that was isolated from digestive tract of Kerinci local duck was *Bacillus circulans* and *Bacillus sp* those have characteristics such as produce protease enzyme, number of cells are 3.25×10^{11} CFU/mL and 3.65×10^{11} CFU/gram, pH was 5.4 (Manin et al. 2002). Both of these bacteria could survive at pH of 2 – 5, stored at room temperature for 6 months and survived on different level of pH in digestive tracts of Kerinci local ducks (Manin et al. 2003).

Probiotics balanced non pathogenic bacteria and eliminated pathogenic bacteria by competitive exclusion (Pascual et al. 1999). Feed probiotics also reduced using antibiotics (Conway and Wang, 2000). Besides, probiotics increased individual health without any adverse effects to the animal such as residual metabolite products and resistance (Lopez, 2000).

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Use antibiotics as feed additive in poultry industries resulted human (consumer) restlessness because antibiotics were used to therapy and mixed into the ration in the small amount. At the present time, antibiotics are used overdoses to promote animal growth. As a consequence, residual antibiotics detected higher than that of maximum residue limit in some animal products in the market such as meat and eggs. Continually misusing antibiotics also result resistance of those antibiotics. Rusiana and Iswarawanti (2004) in SEAMEO (Southeast Asian Minister of Education Organization) and Tromed RCCN (Tropical Medicine Regional Centre for Community Nutrition) seminar reported that 85% of broiler meat in JABOTABEK (Jakarta, Bogor, Tangerang and Bekasi) contained antibiotic residues of tylosin, penicillin, oxytetracycline and Kanamycine.

MATERIALS AND METHODS

This experiment were used 100 day. The basal ration consisted 54% of maize, 5% of rice bran, 20% of fish meal, 14% of soybean meal, 3% of coconut meal, 2% of vegetable oil and 1% of bone meal. The basal ration contained 21% of crude protein and 3100 kcal/kg of Metabolisable energy. Media used to count number of Bacillus circulans and Bacillus sp bacteria was bacto agar, yeast extract, peptone, glucose, pancreatic digest of casein, beef extract and $MnSO_4$. Media used to count number of Escherichia coli was Mac Conkey Agar (MCA).

Methods

This experiment was assigned to Completely Randomised Design (CRD) in 5 treatments and 4 replications. The treatments were:

R-0 : Basal ration/Basal diet without any addition of probiotics and antibiotics
R-1 : Basal ration with 35 ppm of oxytetracycline (OTC) was added on drinking water
R-2 : Basal ration was added by 35 ppm of Virginiamycine
R-3 : Basal ration with 5 cc of liquid probiotic was added per litre drinking water
R-4 : Basal ration was added by 5 g of probiotic powder per kg ration

Probiotics of Bacillus circulans and Bacillus sp concentration in the drinking water and in the ration were 10^{11} CFU/cc of liquid probiotics and 10^{11} CFU/gram of probiotics powder.

Parameters measured in this experiment were

1. Chicken performance (feed intake, body weight gain and feed conversion ratio)
2. Number of Lactic acid, *Bacillus sp* and *E. coli bacteria.*
3. Total cholesterol content in meat and residual antibiotics
4. Mortality rate

Statistical Analysis

Data were collected and tabulated using Excel Program and analysed using analysis of variance in Completely Randomised Design with 5 treatments and 4 replications. To determine the different effect among treatment groups was tested using Duncan's Multiple Range Test (Steel and Torrie, 1992).

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RESULTS AND DISCUSSIONS

Table 1 showed that antibiotics and probiotics not significantly influence (P>0.05) feed intake, body weight gain and feed conversion ratio of chicken, however, they significantly decreased (P<0.05) mortality rate. Similar effect of antibiotics and probiotics on feed intake, body weight gain and feed conversion ratio due to the nutrient content of each treatment ration were similar. Chicken fed similar diet (basal diet), thus, the nutrient intake among the chicken was also similar.

Chicken Performance

Tillman et al. (1986) reported that chicken feed consumption was vary depended on the feed quality especially nutrients (protein, lipids, crude fibre, etc.) and energy content. This study was different from Winarsih (2005) who found that offered probiotics *Bacillus sp* and antibiotics Zink bacitrasin significantly increased (P<0.05) broiler growth. This study found that antibiotics and probiotics significantly reduced (P<0.05) mortality rate of the chicken.

Table 2 showed that offered probiotics and antibiotics to the broiler were highly significant effect (P<0.01) on reduced the number of *Escherichia coli* bacteria in intestine of 42 days old chicken. Number of *E. coli* in intestine of chicken fed basal ration was 8.5965, whereas, in intestine of chicken fed basal ration with antibiotics were 6.2829 and 6.4711, in intestine of chicken fed basal ration with probiotics were 6.5590 and 6.6399, relatively.

Table 1. Average of Feed Intake (g/head/week), Body Weight Gain (g/head/week), Feed Conversion Ratio and Mortality Rate (%) of Chicken during the Experiment

Parameter	R-0 BR	R-1 BR + OTC	R-2 BR + Virgi	R-3 BR + LP	R-4 BR + PP
Feed Intake	418.29[a]	412.58[a]	411.03[a]	393.68[a]	378.17[a]
Final Body Weight	1243.51[a]	1201.42[a]	1247.73[a]	1240.98[a]	1201.71[a]
Weight Gain	210.57[a]	196.68[a]	198.68[a]	200.06[a]	191.05[a]
FCR	1.90[a]	1.99[a]	1.97[a]	1.91[a]	1.93[a]
Mortality	4.00[ab]	4.00[ab]	6.00[a]	0.00[a]	3.00[ab]

Note: different superscript on the same row means significantly different (P<0.05)
BR = basal ration, OTC = oxytetracycline, Virgi = Virginiamycine, LP = liquid probiotics, PP = powder probiotics, FCR = feed conversion ratio

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Table 2. Average of Number of Bacteria of Bacillus, Lactic Acid and Escherichia coli in Small Intestine of Forty Two-Day Old Chicken (transformation log x)

Treatment	Bacillus	Lactic Acid	Escherichia coli
R-0 (BR)	8.1466[a]	8.2736[b]	8.5965[a]
R-1 (BR + OTC)	7.5966[c]	7.7749[c]	6.2829[c]
R-2 (BR + Virgi)	7.4643[c]	7.6757[c]	6.4711[bc]
R-3 (BR + LP)	10.3028[a]	10.3382[a]	6.5590[b]
R-4 (BR + PP)	10.2708[a]	10.2893[a]	6.6399[b]

Note: different superscript on the same column means highly significant different (P<0.01)
BR = basal ration, OTC = oxytetracycline, Virgi = Virginiamycine, LP = liquid probiotics, PP = powder probiotics, FCR = feed conversion ratio

Result of the current study showed that added antibiotics and probiotics highly significant increased (P<0.01) number of Lactic acid bacteria. Fuller (1992); Jin et al. (1997) and Lopez (2002) reported that Bacillus bacteria could stimulate forming and have mutualism symbiosis with Lactic acid bacteria in the animal digestive tract when they were used as probiotics microbes. Similar report was stated by Jin et al. (1996) who added *Bacillus subtilis* and *Lactobacillus* into the broiler ration. Authors found that *Lactobacillus* population in the intestine increased whereas *E. coli* population decreased, relatively.

Table 3. Average of Total Cholesterol Content in the Chicken Meat (mg/dl)

Treatment	Replication-				Average
	I	II	III	IV	
R-0 (BR)	96.00	73.00	107.00	79.00	88.750[a]
R-1 (BR + OTC)	23.00	24.00	30.00	33.00	27.500[b]
R-2 (BR + Virgi)	36.00	30.00	40.00	30.00	34.000[b]
R-3 (BR + LP)	34.00	42.00	23.00	24.00	30.750[b]
R-4 (BR + PP)	32.00	24.00	27.00	28.00	27.750[b]

Note: different superscript on the same column means significant different (P<0.05)
BR = basal ration, OTC = oxytetracycline, Virgi = Virginiamycine, LP = liquid probiotics, PP = powder probiotics, FCR = feed conversion ratio

Antibiotics Residue in Meat

Table 4. Antibiotics Residue in Meat (ppm)

Replication	Oxytetracycline	Virginiamycin
I	0.00	0.00
II	0.00	0.38
III	0.00	0.00
IV	0.00	0.90

Total Cholesterol in Meat

Table 3 showed that mixed antibiotics or probiotics either into the ration or in drinking water of chicken were highly significant influenced (P<0.01) total cholesterol content in meat. Cholesterol content was higher in meat of chicken either fed ration containing antibiotics (34.000 mg/dl) or probiotics (27.750 mg/dl) or drunken water containing antibiotics (27.500 mg/dl) or probiotics (30.750 mg/dl) than that of chicken

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fed only basal ration (88.750 mg/dl) for 42 days. It means that antibiotics reduced 62.22% and probiotics reduced 64%, of total cholesterol content, relatively. Reduce cholesterol content due to that probiotics Bacillus circulans and Bacillus sp could increase intestinal homeostasis that were possible to cholesterol destruction or degradation into cholat acid in the liver, thus, cholesterol content in the meat decreased (Gruenewald, 1989; Fuller, 1992).

Table 4 showed that 35 ppm of oxytetracycline in drinking water offered to the chicken for 42 days did not cause any residue oxytetracycline accumulation in meat. It was different effect with 35 ppm of virginiamycine when it was mixed into the ration of chicken and offered to the chicken for 42 days. Virginiamycine residue was detected in meat approximately 0.38 and 0.90 ppm.

Antibiotics usually used in animal production not only for therapy and disease prevention but also to promote animal growth. However, continually offered antibiotics might have adverse effect in animal such as residue accumulation in animal products and animal resistance to certain antibiotics (Pascual et al. 1999; Barton and Hart, 2001; Lee et al. 2001).

Detectable virginiamycne residue in this experiment indicated that either level of virginiamycine in chicken ration has to be reduced or virginiamycine have to be withdrawal several days before slaughter. In contrary to oxytetracycline that mixed into drinking water as 35 ml/L was not resulted residue accumulation. It might be due to that offered oxytetracycline to the chicken through drinking water reduced the oxytetracycline activity.

Result of the current study was contrary to Rusiana and Iswarawanti (2004) and Rusdi (2004) report. The first authors found that 85% of broiler meat contained oxytetracycline residue. The other author found that broiler meat sample from slaughter house and slaughter place in West Java contained 0.1107 ppm in average. This level was higher than that of maximum residue limit recommended by WHO (1999) and SNI (2000) that is 0.1 ppm.

Mortality Rate

Mortality rate calculated in the present study was higher than 17%. Table 1 showed that among all treatment groups, probiotics could decrease number of chicken died. It is similar as Pascual et al. (1999) who found that probiotics could balance non pathogenic bacteria and eliminate pathogenic bacteria through competitive exclusion. Probiotics could also reduce using antibiotics (Conway and Wang, 2000) and improve animal health. It was different with antibiotics; probiotics would not cause any residual accumulation and antibiotic resistance (Lopez, 2000). High chicken mortality might be due to that some chicken in all treatment groups were infected by Marek's diseases.

CONCLUSION

It is concluded that:

1. Probiotics could be used as feed additive. It was not significant on improve broiler performance but significantly reduce cholesterol content in meat and number of Escherichia coli.
2. Oxytetracycline residue was undetectable in meat, whereas, virginiamycine residue was detectable.
3. Liquid probiotics could reduce mortality rate of chicken.

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