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Broilers Serum Cholesterol and Glutamic Oxaloacetic Transaminase and Their Relation to Antibiotic in Feed and Medication Programs in Four Broiler Producers in Semarang Region-Central Java, Indonesia

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Abstract: This study was carried out to investigate the use of antibiotic in feed and medication programs and their effects on broilers Serum Cholesterol and Glutamic Oxaloacetic Transaminase (SGOT). The broilers were sampled randomly from four small-scale broiler producers (BP1, BP2, BP3, BP4) in Semarang region-Central Java, Indonesia. Observed data comprising medication programs and the types of feed ingredients in the commercial complete feed were obtained from medication program brochures and feed labels respectively. Four unsexed broilers from each poultry producer were randomly sampled at selling point in poultry slaughterhouse. Broilers market body weight and abdominal fat percentage were measured on the spot. Blood was taken to obtain the serum samples for determination of serum cholesterol and SGOT. An independent experimental study (E1) was also carried out approximately the same time as the field study using the same feed as in BP-4 and raised under similar condition as in the broiler producers. However, no medication program was employed. The results showed that all feeds from four different feed companies (FC1, FC2, FC3, FC4) used by the four BPs respectively contained antibiotics. Antibiotics were also administered in medication program via drinking water. The market age was found to be 34-35 days old and market weight was in average 1760.38±54.14 g with no significant difference among producers. This market weight had no significant difference to E1. There was no significant difference in broilers abdominal fat percentage and SGOT among BPs. However, these abdominal fat and SGOT were significantly higher than E1. There is a significant difference in serum cholesterol of broilers among BPs, where BP-4 had the highest (145.2±6.59 mg dl^{-1}) and BP-3 had the lowest serum cholesterol level (117.53±9.76 mg dl^{-1}). These serum cholesterol and SGOT levels were significantly higher than those in E1 or in those published results that used in-feed antibiotic and no-medication (E2) or no antibiotic in feed nor medication (E3). These results suggested that antibiotic in feed and medication program affect lipid and hepatic metabolism of broilers which is reflected by an increase in serum cholesterol and SGOT.

Key words: Broiler, cholesterol, SGOT, antibiotic, feed, medication

Introduction

Antibiotics as feed additives have been used since chlortetracycline was found in 1940 (Spring, 1999). Its continued use is due to promotion of growth and feed efficiency. The action of in feed-antibiotic is partly mediated by a decrease in the number of competitive pathogenic bacteria in the gut and hence increases the utilization of nutrients (Cook, 2000; Dibner and Richards, 2005). However, after 50 years of its use, it has created global market pressure for in feed-antibiotics bans due to the possible link between antibiotic residues in animal products and the emergence of resistance microorganism associated with particular use of antibiotics (Salyers, 1999). In spite of this ban, the poultry industries especially in hot-humid climate like Indonesia have faced difficulties in adopting it. This is mainly due to high environmental stress especially in small-scale poultry producers who use an open cage house. Antibiotics remain an effective feed additive in these poultry production settings as it can suppress mortality, support maximal growth and hence profitable. The following field research was carried out to study the use of antibiotics in feed and medication program by broiler producers in Semarang regions, Central Java. The broiler producers have been using the commercial feeds produced by local well known feed companies. The antibiotic use was also studied for its effect on the concentration of serum cholesterol and the activity of Serum Glutamic Oxaloacetic Transaminase (SGOT). These biochemical information would provide additional weights in considering such antibiotics use in commercial broiler production system.

Materials and Methods

Field study: This study was carried out during the broilers production time from October to December 2006, in four broiler producers (BPs) selected randomly and assigned as BP-1, BP-2, BP-3, BP-4. The four broiler producers were located in Semarang region near by Semarang City, Central Java, Indonesia. Each of the

Table 1: Feed Ingredients Used by Commercial Feed Company

Feed Company-1	Feed Company-2	Feed Company-3	Feed Company-4
	Corn	Corn Yellow	Corn
	Rice meal	Soybean Meal	Rice Meal
Not written on the label	Corn Gluten	Canola meal	Fish Meal
	Pollard	Fish Meal	Soybean Meal
	Meat Bone Meal	Meat Meal	Coconut Meal
	Fish Meal	Rice Meal	Meat Bone Meal
	Soybean Meal	Pollard	Pollard
	Groundnut Meal	Vitamins	Groundnut Meal
	Cereals Meal	Minerals	Leaf Meal
	Vegetable Oil	Antioxidants	Canola
	Calsium		Vitamins
	Phosphate		Calsium
	Methionine		Phosphor
	Lysine		Trace minerals
	Minerals		
	Vitamins		
Antibiotic present but not specifically named and written	Bambermysine Diclazuril	Contains coccidiostat	Zinc Bacitracin Monensin

Source : Feed Label from each Feed Companies

Table 2: Nutrients content of each commercial feed written on the feed label

Nutrients	FC-1for BP-1	FC-2 for BP-2	FC-3 for BP-3	FC-4 for BP-4
	---------- % ----------			
Moisture	12	13	14	13
Crude Protein	19-21	21-23	21	19-23
Crude Fat	4	4	4	5
Crude Fibre	4.5-5	5	4	5
Ash	6.5	6.5	6.5	7
Ca	0.9-1	0.9-1.2	0.9-1.1	0.9
P	0.7-0.9	0.7-0.9	0.7-0.9	0.6

Source : Feed Label of each Feed Company

four broiler producers have been using a particular commercial-complete feed from four different Commercial Feed Companies assigned as FC-1, FC-2, FC-3, FC-4 respectively. Each commercial feed from each feed companies was provided with medication program sheets as a guide to maximize broilers production. Data collection consisted of: 1) medication program obtained from medication program sheet in each BPs, 2) the types of feed ingredients in the commercial feed were obtained from feed labels in each BPs, 3) market age and body weight, 4) abdominal fat percentage, 5) serum total cholesterol and 6) Serum Glutamic Oxaloacetic Transaminase (SGOT). Four unsexed broilers from each BPs were taken randomly at selling point in poultry slaughterhouse. Broilers market body weight and abdominal fat percentage was measured on the spot using 2 kg capacity balance. Blood was taken to obtain the serum samples for determination of serum cholesterol and SGOT. Serum cholesterol was measured using "cholesterol-oxidase para-aminophenazone" (CHOD-PAP) "enzimatic colorimetric" method (Kit from Human Gesellschaft, Germany). The activity of Serum Glutamate Oxsaloacetat Transaminase (SGOT) was measured using Kinetic method according to International Federation of Clinical Chemistry (IFCC) recommendation (Kit from Human Gesellschaft, Germany). Abdominal fat was obtained by separating the abdominal fat from the carcass. All data were analyzed using ANOVA and Duncan's multiple range test was used when means were significant ($p<0.05$).

Experimental study with no medication (E1): This experiment was carried out approximately the same time as the field study. A total of 50 CP-707 DOC unsexed broilers with initial body weight 46.35±4.66 g were used in this experiment. On day 10, broilers were allocated randomly into 4 replicates with 10 chickens per replicate. The birds were given *ad libitum* commercial feed as used by BP-4 and raised under similar condition as that in small-scale broiler producers at the Faculty facilities. The birds received ND vaccines at 4 and 21 days old as a routine vaccination schedules as in BPs. Birds were given free access to drinking water. No medication program was employed. At day 35 one bird of each replicate was sampled randomly and the blood was collected via brachial vein to obtain serum samples for determination of total cholesterol and SGOT.

Results

Antibiotics in feed: Table 1. showed that the four types of commercial feeds employed by the four broiler producers contained antibiotics.

The nutrient composition of the four commercial feed used by broiler producers i.e. moisture, fat, protein, crude fibre, ash, Calcium and Phosphor contents were similar. However, none of the feed label informed the metabolic energy content of the feed (Table 2).

Antibiotics in medication: Table 3 showed the medication program employed by the four broiler producers. The data showed that each of the four

Table 3: Medication compounds and schedule used by Broiler Producers

No.	Broiler Producers	Medication schedule	Administer on day-	Dosage
1.	BP-1	Antibiotic A	1, 2, 3, 5	10, 15, 20, 30 ml
		Vitamins	every day	-
		Vitamins	every day	-
		Antibiotic B	12-14, 17-19	40 gr
		Antibiotic C	21-24	80 gr
2.	BP-2	Vitamins	1-5, 19-21, 28-30	-
		Vitamins	13-15	-
		Antibiotic D	1-3	20 gr/4L drinking water
		Antibiotic E	15, 16, 17	90 gr/23-29L drinking water
		Antibiotic F	25, 26	-
3.	BP-3	Antibiotic G	1-3	1 cc/2 L drinking water
		Vitamins	5-6, 11-12, 14, 18, 20	1 g/10 L drinking water
		Antibiotic H	8-10, 21-23	1 g/2 L drinking water
		Antibiotic I	15-17	1 g/ L drinking water
		Growth-promoter	25-27	1 g/4 L drinking water
		Vitamin C	28-31	1 g/4 L drinking water
4.	BP-4	Antibiotic J	1-3	60 ml/150 L drinking water
		Vitamins	5-12, 10-12, 25-30	20-100g / 20-150L drinking water
		Antibiotic K	15-18	30 ml/60 L drinking water

Source : Medication sheet used in each BP

Table 4: Market Weight, Abdominal Fat, Serum Cholesterol, and SGOT of broilers from four different commercial broilers producers (BP) and Experiment with no medication

	Treatments				
Variables	BP_1	BP_2	BP_3	BP_4	E_1
Final Market Weight (g)	1733±45.13	1772±46.49	1725±55.99	1813±27.69	1698.63±70.69
Abdominal Fat (%)	$3.2±0.31^{a}$	$3.19±0.23^{a}$	$3.1±0.58^{a}$	$3.15±0.43^{a}$	$1.82±0.47^{b}$
Serum cholesterol (mg/dl)	$129.08±5.80^{bc}$	$136.95±1.55^{ab}$	$117.53±9.76^{c}$	$145.2±6.59^{a}$	$122.60±14.38^{c}$
SGOT (U/L)	$272.6±33.96^{a}$	$272.13±35.88^{a}$	$261.73±63.77^{a}$	$285.05±30.39^{a}$	$145.50±12.37^{b}$

Means within rows with no common superscript differ significantly ($p<0.05$)

medication programs recommended different antibiotics with different name of trade marks. All antibiotics in medication programs were administered via drinking water. In addition to antibiotic, additional vitamins were also administered via drinking water.

Final market age and weight: The final or market age was found to be 34-35 days. The market weight of broilers from the four producers ranged from 1.7-1.8 kg (Table 4). There is no significant different in this market weight among broilers from the four BPs ($p>0.05$). These market weights also showed no significant different to E1 (1.7 kg) ($p>0.05$).

Abdominal fat percentage: The abdominal fat percentage of broilers from the four broiler producers ranged from 3.1-3.2% (Table 4). There is no significant different in the percentage of abdominal fat among broilers from different producers ($p>0.05$). However, these abdominal fat percentage from the four BPs were significantly higher that those in E1 (1.82%).

Serum cholesterol of broilers: Serum cholesterol of broilers from the four poultry producers showed a value of 129.08 mg dl^{-1} for BP-1, 136.95 for BP-2, 117.53 for BP-3 and 145.2 for BP-4 (Table 4). There is a significant difference in these broilers serum cholesterol between broilers producers where BP-3 as the lowest and BP-4 as the highest ($p<0.05$). Broilers serum cholesterol level in BP1 and BP3 were not significantly different than E1, however serum cholesterol level in BP2 and BP4 were significantly higher than E1.

Serum Glutamic Oxaloacetic Transaminase of Broilers: The activity of SGOT of broilers from the four broiler producers ranged from 261.73-285.05 U/L (Table 4). There is no significant different in these SGOT values among broilers from different producers ($p>0.05$). However these are significantly higher compared to E1 (145.50 U/L).

Comparison of broilers serum cholesterol and SGOT from four broiler producers with published experimental data not employing medication with the same production setting: The results of serum cholesterol and SGOT from the four broiler producers and E1 were compared to published in-feed antibiotic experiments consisting of: 1) with in-feed antibiotic and no medication with different types of feed ingredients (E_2), or 2) with no antibiotic in feed nor medication (E_3) (Table 5) (Indriani and Murwani, 2005; Shofianingtyas and Murwani, 2005).

Table 5: Comparison of Field Data with Other Published Experimental Research

	Variables	
Treatment	Serum cholesterol (mg/dl)	SGOT activity (U/L)
BP-1	129.08±5.80^{bc}	272.60±33.96^{a}
BP-2	136.95±1.55^{ab}	272.13±35.88^{a}
BP-3	117.53±9.76^{c}	261.73±63.77^{a}
BP-4	145.20±6.59^{a}	285.05±30.39^{a}
E1	122.60±14.38^{c}	145.50±12.37^{b}
E2	86.35±5.00^{d}	173.10±27.00^{b}
E3	98.33±13.67^{d}	180.18±32.50^{b}

Means within columns with no common superscript differ significantly ($p<0.05$), E1: Experimental research using the same open cage house as small-scale commercial poultry production system. E2 and E3: Published experimental research using the same open cage house as small-scale commercial poultry production system. Feed ingredients used in E1: commercial feed as in BP4 (in this experiment); in E2: local yellow corn, mung bean, soybean meal, rice meal, fish meal, shell powder, vitamins and mineral mix free antibiotics (self made)+in-feed antibiotic chlortetracycline 100 ppm; in E3: as in E2 with no in-feed antibiotic. Feed and water were given ad libitum (Indriani and Murwani, 2005; Shofianingtyas and Murwani, 2005)

Statistical analyses showed that serum cholesterol in E2 and E3 are significantly lower than E1 and BP-1 to BP-4 ($p<0.05$), while E1 was similar to BP-1 and BP-3 ($p>0.05$). The activity of SGOT in E1, E2 and E3 were similar ($p>0.05$) but significantly lower than BP-1 to BP-4 ($p<0.05$) (Table 5).

Discussion

It is clear from the data above that all of the commercial feed used by the four samples of broiler producers contained antibiotics. The medication program used by each of broiler producer also showed an intensive use of antibiotic since the arrival of day old chick broilers. The types of antibiotic use in each medication program are different and this was presumably to avoid accumulation or residue in broilers' various organs.

Market weights of broilers from each of the four broiler producers were similar ranging from 1.7-1.8 kg. This indicates the average market weights that are acceptable in Semarang region. The percentage of abdominal fat was also similar among broilers from the four broiler producers reaching 3.1-3.2%. This percentage of abdominal fat is commonly found in broilers raised under intensive rearing using commercial complete feed, but this value is higher than the average value of 2.3% found by Leenstra *et al.* (1986) or Yang *et al.* (2003). Broilers fed the same feed as in BP4 but no medication in E1 has lower percentage of abdominal fat (1.82%). This difference between E1 and BP4 might be due to the different management in BP4 which used antibiotics in medication program. There has been no reports on the effect of medication on broilers abdominal fat deposition so far.

The significant difference of serum cholesterol in broilers among BPs with BP1 and BP3 were not significantly different than E1 and BP2 and BP4 were significantly higher than E1 may signify the effect of different types of feed ingredients in complete feed by each FCs and the use of antibiotic in medication program by BPs. This possibility is corroborated by experimental study E1 and 2 other published in-feed antibiotic experiments from the same author (Indriani and Murwani, 2005). The use of the same commercial feed as in BP-4 in experiment E1 but raised under no medication program has resulted in a steep decrease in serum cholesterol concentration. These cholesterol values indicated that antibiotic use in medication alone can increase broilers serum cholesterol significantly (BP4 compared to E1). How antibiotic in medication program lead to a change in cholesterol metabolism is interesting. No other studies had been carried out to specifically address this in broilers. Antibiotic use in feed or medication should decrease microbial load which lowers immune stimulation. While immune stimulation is associated with altered lipid metabolism indicated by increase in serum cholesterol, trigliceride level and adipose deposition (Humphrey *et al.*, 2002; Khovidhunkit *et al.*, 2004), antibiotic use in feed and medication associated with low or absent of immune stimulation therefore should not increase cholesterol level. This possibility is in agreement with an experiment in rats carried out by Mukherjee *et al.* (1970), who showed that administration of chlortetracyclin at therapeutic level (35 ppm) lowered cholesterol biosynthesis. However, this is not the case in these field findings. The possibility is that in the absence of immune stimulation, there is no energy needs for mounting immune response. The available energy for growth (presumably in the form of pool acetyl CoA) therefore might be in excess and converted to adipose accretion and cholesterol synthesis reflected by higher level of abdominal fat and serum cholesterol respectively. This possibility merits further studies.

The effect of antibiotic in feed is discussed by comparing cholesterol level in E1 to published results of broilers in E2 (with in-feed antibiotic and no medication) and E3 (in which no antibiotic in feed nor medication was employed and the types of feed ingredients were different than E1) (Table 5) as experimental study employing the same commercial feed CF-4 as in BP4 but with no in-feed antibiotic is not possible. The data showed that cholesterol levels of broilers in E2 and E3 are lower than E1, but E2 and E3 are similar. Antibiotic in feed appeared to have no effect to serum cholesterol. The difference between E1 and E2 in which both use in-feed antibiotic may be due to different rasio of metabolic energy to protein (EM:P). Therefore the effect of antibiotics in feed remains elusive. Comparing field and experimental data with published results E2 and E3 which used no antibiotics and no MBM in the feed

ingredients, there is an indication that broilers raised with no antibiotic have lower serum cholesterol. However there is also a possible contribution on the use of MBM in the commercial feeds to cholesterol level in all of the broilers from BPs. MBM contains a considerable amount of lipid including cholesterol and this may contribute to elevation of serum cholesterol in broilers. This possibility is in line with findings by Cheng and Yu (1997) who found that total serum cholesterol of Wistar rats that received antibiotic Bacitracin-Neomicyn Sulphate (0.7g $100g^{-1}$) together with dietary cholesterol were significantly higher than rats receiving the same antibiotic but with no dietary cholesterol. They also concluded that antibiotic alters lipid metabolism in rats and the effect are most pronounced in those also fed cholesterol. Therefore elevated level of serum cholesterol in BPs appeared to be due not only to antibiotic in medication and feed but also to the presence of cholesterol in the feed ingredients.

The activity of SGOT broilers among the four broiler producers were not different significantly but they were significantly higher compared to the activity of SGOT from the three experimental studies mentioned earlier where no medication was administered (Table 5). The administration of antibiotics in feed and drinking water appeared to increase hepatic work load associated with the metabolism of incoming antibiotics as well as lipid and hence higher SGOT.

Conclusion: The use of antibiotic in-feed and medication in broilers production at four small-scale poultry producers in Semarang region showed an intensive use. This intensive use resulted in high level of total serum cholesterol and SGOT. These results showed for the first time the effect of antibiotic in feed and medication on several biochemical parameters and productivity of broilers raised under local commercial broiler production setting.

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