The effect of herbal drink on the levels of high density lipoprotein and low density lipoprotein of broiler chicken

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Abstract. The objective of this research was to evaluate the effect of herbal drinks from mixture extract of basil leaves, lime leaves and lemongrass leaves on the levels of high-density lipoprotein (HDL) and low-density lipoprotein (LDL) of broiler chickens. The research used 120 birds of DOC. The basal diet was formulated to contain 3000 Kcal/kg Metabolizable Energy (ME) and 22% crude protein (CP). Experimental design used was completely Randomized Designed with 6 treatments and 4 replicates. The treatments were as follows: P0: control (without herbal drink), P1: basal diet + 10% herbal drink in drinking water, T2: basal diet + 20% herbal drink in drinking water, P3: basal diet + 30% herbal drink in drinking water, P4 : basal diet + 40% herbal drink in drinking water and P5 : basal diet + 50% herbal drink in drinking water. The parameters observed were HDL and LDL. The data were analyzed with analysis of variance and if any significant difference were analyzed Duncan’s multiple range tests. The result showed that the treatment of herbal drinks significantly (p <0.05) on the levels of HDL and LDL of broiler chickens. Conclusion of the research was herbal drinks from mixture extract of basil leaves, lime leaves and lemongrass leaves increase the level of HDL and decrease the level of LDL of broiler chickens.

1. Introduction
The use of antibiotics growth promoter (AGP) in poultry farming has been aimed at preventing the disease so that it can increase chicken growth [1]. The problems of the used antibiotics were antibiotic resistance and antibiotic residues in meat products that have had a negative impact on human health. Another problem of the product of broiler chicken was the cholesterol content in the meat. The cholesterol content has been potentially leading to diseases such as stroke and heart disease. Therefore it is necessary the alternatives of natural feed additives from local resources which are safe for consumers, by utilizing herbal drinks from mixture extract of basil leaves, lime leaves and lemongrass leaves.

The mixture extract of basil leaves, lime leaves, and lemongrass leaves was an herb that has the potential of antibacterial activity against Salmonella thyphi and Staphylococcus aerus [2]. The previous study showed that utilizing a mixture extracts of basil leaves, lime leaves, and lemongrass leaves as quail drinks up to a concentration of 10% improved the performance of quail and the blood profil of quail [3,4]. Siregar et al. [5] reported that the herbal drink up to a concentration of 50% in drinking water for broilers increased the growth of broiler chickens. Based on that potential, herbal drinks can act as a substitute for antibiotics which are able to increase the level of high-density lipoprotein (HDL), and decrease the level of low-density lipoprotein (LDL) of broiler chickens. So far, the little information about the research on the use of herbal drinks from a mixture extracts of basil leaves, lime leaves, and lemongrass leaves as a substitute for AGP of broiler chickens to increasing the level of high-density lipoprotein (HDL) and reducing the level of low-density lipoprotein (LDL) of broiler chickens. So that, it is necessary to conduct the research utilized of herbal drinks from a mixture of extracts of basil leaves, lime leaves, and lemongrass leaves which can increase HDL levels and reduce LDL levels in broiler chicken blood. The use of herbal
Drinks from a mixture of extracts of basil leaves, lime leaves, and lemongrass leaves was also very safe to be applied, cheap and easy to find in the farmer’s surrounding environment. The objective of this research was to evaluate the effect of herbal drinks from mixture extract of basil leaves, lime leaves and lemongrass leaves on the levels of high-density lipoprotein (HDL) and low-density lipoprotein (LDL) of broiler chickens.

2. Materials and Methods
The research used 120 birds of day old chicks (DOC). The treatments were as follows: P0: control (without herbal drink), P1: basal diet + 10% herbal drink in drinking water, P2: basal diet + 20% herbal drink in drinking water, P3: basal diet + 30% herbal drink in drinking water, P4: basal diet + 40% herbal drink in drinking water and P5: basal diet + 50% herbal drink in drinking water. The basal diet was formulated to contain 3000 kcal/kg of metabolizable energy and 22% of crude protein. The composition of the basal diet presented in Table 1.

| Ingredient      | Composition (%) |
|-----------------|-----------------|
| Rice bran       | 6.00            |
| Corn            | 39.50           |
| Soybean meal    | 34.00           |
| Coconut meal    | 5.00            |
| Coconut oil     | 3.00            |
| Fish meal       | 10.00           |
| Premix          | 1.50            |
| Salt            | 1.00            |
| **Amount**      | **100.00**      |
| Crude protein (%)| 22.06           |
| ME (kcal/kg)    | 3131.07         |
| Crude fat (%)   | 4.96            |
| Crude fiber (%) | 5.11            |
| Phosphor (%)    | 0.49            |
| Calcium (%)     | 1.48            |

The research was conducted in two steps. The first step of the research was herbal drink preparation. Herbal drinks derive from a mixture extract of basil leaves, lime leaves and lemongrass leaves and brown sugar. The herbal drink is made by weighing the basil leaves, lime leaves, and lemongrass leaves with the same ratio, then washed, thinly sliced, and blended. The mixture of the three leaves was put in a 1000 ml glass, closed for 24 hours, filtered then added with brown sugar as much as 5% of the extract. The extract was ready to be given to the chicken according to the treatment.

The treatment of herbal drink conducted for 30 d. Birds were randomly placed into 6 treatments and 4 replications with 5 birds per replicate. The birds were not given any vaccines and antibiotics. The parameters observed were HDL and LDL levels. The data was collected at the end of the experiment of 30 d of age, one of the broilers from each replicate was randomly selected. The blood sample was obtained from wing-vein and the blood was collected in an EDTA tube. The HDL and LDL levels were determined according to the method of Hassanuddin et al. [6].

The experimental design used was a completely randomized design (CRD) with 6 treatments and 4 replications. The data were analyzed with analysis of variance and if any significant difference were analyzed Duncan’s multiple range tests [7].
3. Results and Discussion
The average of high-density lipoprotein (HDL) and low-density lipoprotein (LDL) levels of broiler chickens that were treated with herbal drinks from a mixture extract of basil leaves, lime leaves, and lemongrass leaves were presented in Table 2. The results of the analysis of variance showed that the treatment of herbal drinks from a mixture extract of basil leaves, lime leaves, and lemongrass leaves had a significant effect ($p < 0.05$) on the levels of high-density lipoprotein (HDL) and low-density lipoprotein (LDL) in broiler chickens.

Table 2. The average of high-density lipoprotein (HDL) and low-density lipoprotein (LDL) in broiler chickens

| Treatment | High-Density Lipoprotein (HDL) (mg/dL) | Low-Density Lipoprotein (LDL) (mg/dL) |
|-----------|--------------------------------------|--------------------------------------|
| P0        | 57.50$^d$                            | 32.50$^e$                            |
| P1        | 58.50$^d$                            | 30.50$^d$                            |
| P2        | 63.00$^c$                            | 28.50$^c$                            |
| P3        | 66.50$^c$                            | 25.00$^d$                            |
| P4        | 68.50$^{ab}$                         | 20.50$^d$                            |
| P5        | 69.50$^a$                            | 18.00$^f$                            |

Means within a column with different superscripts are different at ($p < 0.05$)

3.1. High-Density Lipoprotein (HDL)
The results showed that HDL levels of broiler chickens that were treated with herbal drinks from a mixture extract of basil leaves, lime leaves, and lemongrass of 63.91 mg/dL with a range of HDL levels between 57.50 - 69.50 mg/dL. The HDL level in this study was in the normal standard range of broilers was 22 mg / dL and less than 130 mg / dL [8]. Rahmanetjad et al. [9] reported HDL levels of broiler chickens that supplementation with dried pomace tomatoes of 64.00 - 74.00 mg / dL. Haryanto et al. [10] stated that supplementation of banana peels at broiler chicken had the HDL levels of 13.47 - 34.73 mg / dL. Medah et al. [11] reported that range of the HDL levels of broiler chickens with supplementation of combination of ginger and lime extract of 65.88 to 84.10 mg / dL.

The high density lipoprotein (HDL) levels of broiler chickens was significantly increased ($p < 0.05$) were treated with herbal drinks from a mixture extract of basil leaves, lime leaves, and lemongrass leaves. The HDL levels was the highest in the P5 group (69.5 mg/dL) followed by P4 (68.50 mg/dL), P3 (66.50 mg/dL), P2 (63.00 mg/dL) and P1 (58.50 mg/dL). The lowest recorded in the P0 (57.50 mg/dL). This study indicated that treatment of herbal drink from a mixture extract of basil leaves, lime leaves, and lemongrass increases the HDL level of broiler chicken. Increasing HDL levels in this study was supported by decreasing data of cholesterol and triglyceride levels due to the supplementation of herbal drinks up to a concentration of 50% [12]. Ghaedi et al. [13], stated that the decrease of triglyceride and cholesterol has an effect on increasing HDL levels.

Similarly results with [9] reported that supplementation of dried pomace tomatoes increases the HDL levels of broiler chickens. Cao et al. [14] reported that the supplementation of fermented Ginkgo biloba leaves significantly increase HDL levels of broiler chickens. Puvaca et al. [15] reported that broiler chickens supplemented with garlic, black pepper, and red pepper significantly affect HDL levels. The contrary result to [16], stated that the supplementation of different herbal medicines had no effect on HDL levels of broiler chickens. Medah et al. [11], combination supplementation of ginger and lime did not affect HDL levels of broiler chickens.

The increase of HDL level in this study because the herbal drink contains flavonoid, tannin and saponin. Guilamoe et al. [17], reported that flavonoids increasing HDL levels by increasing the levels of Apolipoprotein A1 as a lecithin cholesterol acyltransferase cofactor and as a lipoprotein receptor-ligand through inhibition of LDL oxidation in the body. The flavonoids have increased the excretion of cholesterol in the digestive system and inhibit the absorption of exogenous and endogenous cholesterol [18]. Tannins play a role in inhibiting the reduction of HMG-CoA for cholesterol synthesis.
and acetyl CoA acetyltransferase in the cholesterol esterification process [10]. HDL is a lipoprotein that is responsible for transporting lipids from the periphery to the liver [19]. Saponins play a role in reduced synthesis of cholesterol by inhibiting the activity of reductase and increases the excretion of bile salts by converting cholesterol to bile salts. The absorption of cholesterol and bile salts inhibited through the formation of micelle [10]. Hermier [20] states that HDL promotes the release of cholesterol from peripheral tissues and transports cholesterol to the liver for catabolism.

3.2. Low Density Lipoprotein (LDL)
The average LDL level of broiler chickens was treated with herbal drinks from a mixture extract of basil leaves, lime leaves, and lemongrass leaves of 25.92 mg/dL. This result was at the normal standard range for broilers that were less than 130 mg/dL [8]. Rahmatetjad et al. [9], HDL levels of broiler chickens given dried pomace tomato supplementation ranged from 49.00 - 69.20 mg/dL. Supplementation of fermented Ginkgo biloba leaves resulted in LDL levels of broiler chickens between 35.45 - 40.52 mg/dL [14]. Puvaca et al. [15], the LDL levels of broiler chickens supplemented with garlic, black pepper, and red pepper ranged from 8.30 - 36.70 mg/dL. Murray et al. [19], LDL in the body has a function to the main carrier of cholesterol from the liver to body tissues. LDL will circulate cholesterol to tissue cells. The excess LDL will be brought back by HDL to the liver which will then be excreted as bile acids.

The results showed that the treatment of herbal drinks from a mixture extract of basil leaves, lime leaves, and lemongrass leaves had a significant effect (p<0.05) on the low-density lipoprotein (LDL) levels of broiler chickens. The mean LDL level in this study respectively P0 32.50 mg/dL, P1 30.50 mg/dL, P2 28.50 mg/dL, P3 25.50 mg/dL, P4 20.50 mg/dL and P5 18.00 mg/dL. The results showed that the lowest LDL level at the P5 group and the highest LDL level at the control group. This result was supported by the data of cholesterol also decreasing along with the supplementation of herbal drink up to 50% [12]. LDL levels were strongly influenced by cholesterol levels [21]. Musa et al. [22], states that cholesterol levels have a positive correlation with LDL levels in the blood.

The result showed that the treatment of herbal drink decreased LDL levels compared to the control. Choi et al. [23] reported that supplementing with onion flour and α-tocopherol were able to reduce LDL levels of broiler chickens. Cao et al. [14] reported that supplementation of fermented Ginkgo biloba leaf significantly reduces LDL levels of broiler chickens. Garlic, black pepper, and red pepper significantly lowered LDL levels of broiler chicken [15]. Rostami et al. [24] stated that supplementation of *Scrophularia striata* and *Ferulago angulata* significantly reduced LDL levels in broiler chicken blood. A contrary result, [16] reported that supplementation of different herbal medicines did not affect the LDL level of broiler chickens. Medah et al. [11], supplementation of combination of ginger and lime did not affect HDL levels of broiler chickens.

Herbal drink from a mixture extract of basil leaves, lime leaves, and lemongrass leaves contain saponin, tannin, flavonoid, and phenolic compound. Saponin and tannin was able to inhibit cholesterol absorption by increases of cholesterol excretion by feces [25]. Further explained that, the process of inhibiting cholesterol by inhibiting the incorporation of cholesterol into micelles so that it was difficult to be absorbed in the intestine. Tannin inhibiting the reductase of HMG-CoA so influenced the reduction of cholesterol synthesis in the liver [26]. Flavonoids play an important role in hypocholesterolaemic mechanisms and hypolipidemic activity [15]. The active compounds in herbal drink such as eugenol, limonene, E. Caryophyllene, β-Caryophyllene, methyl eugenol, flavonoids, limonoids, coumarins, glycerolipids, and α-tocopherol [27, 28] act as an anti-tumor, anti-inflammatory, anticancer, and antioxidant [29, 30]. Khanahmadi and Janfeshan [31] reported that herbs contains phenolic components such as α-pinene, bornyl acetate, cis-oicinene, and β-pinene of act as antioxidants. Faustman and Wang [32], stated that α-tocopherol was an antioxidant that is very effective in protecting fatty acids from oxidation caused by free radicals. The antioxidant activity of herbal drinks reduced the production of very low density lipoprotein (VLDL) in the liver as a precursor of LDL [33].
4. Conclusion

Conclusion of the research was herbal drinks from mixture extract of basil leaves, lime leaves, and lemongrass leaves increase the level of HDL and decrease the level of LDL of broiler chickens.

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