Effectiveness of Gambir Solution (UncariagambirRoxb) on the Performances of Broiler Chickens were Infected by Escherichia coli.

Dora Villa Manik, Ma'ruf Tafsin, Armyn Hakim Daulay, Nurzainah Ginting, Hasnudi.

Animal Production Program Study, Faculty of Agriculture, University of Sumatera Utara, Medan 20155, Indonesia

E-mail: doravillamanik20@gmail.com

Abstract. DORA VILLA MANIK, 2018. "Effectiveness of Gambir Solution (UncariagambirRoxb) on the Performances of Broiler Chickens Infected by Escherichia coli".Supervised by MA'RUF TAFSIN and ARMYN HAKIM DAULAY.

Gambir has the potential as an antibacterial which is traditionally used as a drug for diarrhea. This study examines the effectiveness of gambir solution on performance which includes feed consumption, body weight gain, feed conversion and income over feed cost (IOFC) in broiler chickens infected with E. coli. It was held in November - December 2017 at the Laboratory of Biology, Animal Husbandry Study Program, Faculty of Agriculture University of Sumatera Utara, using a completely randomized design (CRD) with 6 treatments and 3 replications. The treatment consisted of PO_A = without infection and without treatment, PO_B = infection with E. coli and P1, P2, P3, respectively E. coli infection + Gambir solution 2%, 4%, 6% and P4 = E. coli + infection Tetracycline antibiotics. The results showed that the administration of gambir solution at a dose of 2% - 6% was significantly (P<0.05) effective in improving the performances of broiler chickens infected with E.coli and able to compensate for the use of commercial antibiotics in the form of tetracycline. The highest IOFC is in the 4% gambir solution. **Keywords**: Gambir, Escherichia coli, and broiler performance.

1. Introduction

Colibasilosis is an infectious disease in poultry caused by pathogenic Escherichia coli bacteria as primary or secondary agents. Under normal conditions E. coli is present in the digestive tract of the chicken. Approximately 10-15% of all E. coli found in healthy chicken intestines are classified as pathogenic serotypes. Cases of colibasilosis have been reported in various countries in the world. In Indonesia, this disease is found in broilers and laying hens in various regions. Gordon and Poernomo [1] reported two cases of cholicepticemia from 455 samples (0.4%) examined at the Veterinary Research Institute (Balitvet) for one year (March 1973 to February 1974). The results of Wiedosari and Wahyuwardani's research [2] in broiler disease case studies in Sukabumi and Bogor districts showed that the most common diseases were colibasilosis (22.2%), ascites (12.5%), gumboro (12.5%) , Newcastle disease (ND) (10%), Salmonella pullorum (10%), and necrotic enteritis (7.5%). The disease mainly occurs in chickens aged 11-21 days (57.5%) and occurs in the rainy season (60%). Along with increasing bacterial resistance must be balanced with the discovery of new drugs. This prompted the discovery of new antibacterial sources that are more effective against micropatogen infections, cheaper, have smaller side effects, and are available in large quantities so that resistance can be overcome. Indonesia as a tropical country has biodiversity that has potential as a source of phenolic compounds. It is known that flavonoids from some plants in Indonesia have proven antibacterial activity. Among the plants in question is Gambir (UncariagambirRoxb). Gambir is a thick extract obtained from the processing of the leaves and stems of the gambir plant (UncariagambierRoxb) which is deposited, dried, and printed in various forms. Traditionally, this plant is used as a medicine for diarrhea / dysentery, as a mixture in shading to strengthen teeth,

tanneries, dyes, medicinal burns, and mouthwash in throat pain. The ability of gambir as a medicinal plant is caused by the presence of bioactive components. The main phytochemical component in gambir leaves is a flavonoid in the form of catechins about 40% Hayani [3]. According to Lucida, et al. [4] the main phytochemicals in gambier plants are found in the leaves in the form of flavonoids (50% catechins). The high flavonoid in gambir leaves is thought to have the potential as an antibacterial that can inhibit the growth of E. coli bacteria.

2. Research Materials and Methods

The study was conducted at the Laboratory of Biology, Animal Husbandry Study Program, Faculty of Agriculture, University of North Sumatra. This research was carried out for 2 months starting from November to December 2017. The tools used in this study included mortar porcelain, filter paper, funnel, analytic scales (digital), measuring cups, test tubes, erlenmeyer, pasteur pipettes, syringes, plastic bags, markers, counters, label paper, tissue, thermometers, plastic sheeting, experiment cages with a size of 50 cm x 100 cm x 100 cm as many as 18 plots of cages, 18 units of chicken feed and drink, and incandescent lamps (60 Watts) as many as 18 units as lighting and heating. The materials used in the study include 90 broiler chickens per day (DOC) CP707 strain from PT. Charoen Pokphand Jaya Farm, formalin, KMnO4, rodalon, vitachick, ND vaccine, ration, drinking water, distilled water, tetracycline antibiotics, gambier and E. coli bacterial isolates. This study was an experimental study using Completely Randomized Design (CRD) with 6 treatments and 3 replications namely 1 treatment without infection and without treatment, 1 E.coli infection treatment without treatment, 3 treatment of gambier solution and 1 commercial antibiotic treatment which was first broiler chicken infected with E. coli bacteria with a dose of 106 CFU / ml except treatment without infection. The treatment given is:

PO_A: Without E. coli infection and without treatment

PO_B: Infection with E. coli and without treatment

P1: E. coli infection + Gambir solution (UncariagambierRoxb) 2%

P2: E. coli infection + Gambir solution (UncariagambierRoxb) 4%

P3: E. coli infection + Gambir solution (UncariagambierRoxb) 6%

P4: E. coli infection + tetracycline antibiotics 0.1%

Research parameters are Feed Consumption, Body Weight Increase, Feed Conversion (FCR), Income Over Feed Cost (IOFC) and Drug Costs.

2.1 Feed Consumption

Feed consumption is calculated every day based on the difference between the amount of ration given and the remaining amount of ration.

2.2 Increased Body Weight

Body weight gain (PBB) is calculated based on the difference between the results of weighing the final body weight and the previous body weight per time interval weighing.

2.3 Feed Conversion Ratio (FCR)

Feed conversion is calculated by comparing the feed consumed with the body weight gain achieved each week.

2.4 Income Over Feed Cost (IOFC) and Drug Costs

Income over feed cost is the difference between the total revenue and the total ration cost used during the livestock enlargement effort. The cost of medicine is the amount of medicine spent during maintenance

3. Results and discussion

3.1 Feed consumption

Table 1. Average consumption of broiler chicken feed (g / head / week).

Weeks	Treatment						
weeks	P0 _A	PO _B	P1	P2	P3	P4	
Before infection							
Week 1 th	144,80	146,87	141,07	147,13	144,53	148,13	
Week ke-2 ^{tn}	310,73	312,13	308,07	312,73	307,60	309,87	
Average 1-2 weeks ^{tn}	227,77	229,50	224,57	229,93	226,07	229,00	
After infection							

After infection

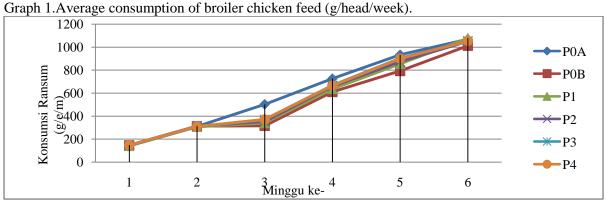
Week 3	502,67 ^a	316,00 ^d	339,73 ^{cd}	353,00 ^{bc}	370,33 ^b	371,07 ^b
Week 4	726,33 ^a	610,40 ^c	637,07 ^{bc}	659,47 ^b	667,13 ^b	664,93 ^b
Week 5	933,87 ^a	792,80 ^d	854,47 [°]	$874,40^{bc}$	903,00 ^{ab}	900,00 ^{ab}
Week 6	$1.068,73^{a}$	1.011,67 [°]	$1.078,87^{bc}$	$1.050,53^{ab}$	$1.054,47^{ab}$	$1.052,60^{ab}$
Average 3-6 weeks	807,90 ^a	682,72 ^d	715,03 °	734,35 ^{bc}	739,73 ^b	747,30 ^b

Description: Different letters on one line indicate that the numbers are significantly different (P < 0.05). tn: not significantly different (P> 0.05)

Table 1 shows that the average consumption of broiler chicken rations before E.coli was not significantly different (P> 0.05). This is because the palatability of the feed given is as good and broiler chickens are in good health so that they consume rations with amounts not much different. According to Anggorodi [5] (1990) states that in consuming feed is influenced by several factors, namely animal health, ration palatability, livestock activity, ration energy and ration protein levels.

Table 1 shows that the treatment after infection with E. coli had a significant effect (P <0.05) on the average feed consumption. Duncan's test results showed that the average feed consumption in the P0A treatment was significantly different from the P0B of 807.90 g and 682.72 g, respectively. This means that feed consumption decreases in broiler chickens that have been infected with E. coli. Where chickens infected with E.coli cause diarrhea to make the chicken lazy to move, sleepy and have no appetite so that the amount of ration consumption decreases. This is in accordance with Akoso's statement [6] (1993), which states that chickens infected with colibasilosis generally show clinical signs; skinny, dull fur, decreased appetite, and moodiness. Its growth is disrupted, diarrhea, dirty hair or sticky stool around its buttocks.

Table 1 shows that the treatment of E. coli after being treated at 3 weeks with a solution of gambier and tetracycline gave an average feed consumption (average 3-6 weeks) which was better than untreated treatment, where Duncan's further test showed that POB is significantly different from P1, P2, P3 and P4 while P1 is significantly different from P3 and P4 but not significantly different from P2. The highest average was found in P4 treatment of 747.30 g and followed by P3, P2, P1 and the lowest in P0B treatment at 682.72 g. This shows that the administration of gambir and tetracycline solutions in E.coli-infected treatments was able to improve the average ration consumption compared to the treatment that was not given. This is related to the level of health of broiler chickens after being infected and then treated with different doses. According to Anggorodi [5] (1990) which states that feed consumption is influenced by animal health, palatability, ration quality and administration procedures.



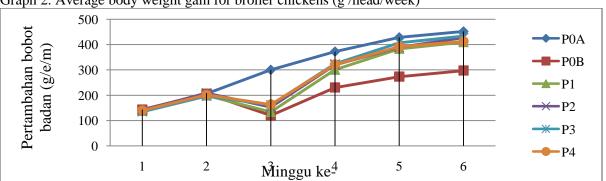
Graph 1 above shows that the average feed consumption at week 3 to week 6 with the administration of gambir solution in the treatment of P2 (4%) and P3 (6%) can compensate for the graphic pattern in the treatment of P4 (tetracycline) in broiler chickens infected with E. coli, respectively 734.35 g, 739.73 g and 747.30 g. This is because the solution of gambier contains catechins which is a potential polyphenol compound as an antibacterial where its properties are able to treat diarrhea in broiler chickens. According to Hayani [3] (2003), which states that the ability of gambier as a medicinal plant is caused by the presence of bioactive components. The main phytochemical component in gambir leaves is a flavonoid in the form of catechins around 40%. According to Lucida et al. [4] (2007) the main phytochemicals in gambier plants are found in the leaves in the form of flavonoids (50% catechins). The high flavonoid in gambir leaves is thought to have the potential as an antibacterial that can inhibit the growth of E. coli bacteria.

Weeks	Treatment							
vv eeks	PO _A	PO _B	P1	P2	P3	P4		
Before infection								
Week 1 th	139,27	143,07	138,43	140,53	134,47	139,57		
Week ke-2 ^{tn}	206,00	205,67	199,00	208,33	196,67	201,33		
Average 1-2 weeks	172,63	174,37	168,72	174,43	165,57	170,45		
After infection								
Week 3	300,13 ^a	119,67 ^c	134,00 ^{bc}	152,67 ^{bc}	161,67 ^{bc}	163,13 ^b		
Week 4	372,27 ^a	230,13 ^c	300,67 ^b	319,33 ^{ab}	$324,00^{ab}$	321,20 ^{ab}		
Week 5	428,53 ^a	273,20 ^b	383,33 ^a	389,00 ^a	407,00 ^a	392,00 ^a		
Week 6	451,93 ^a	297,07 ^b	409,67 ^a	427,00 ^a	433,33 ^a	412,13 ^a		
Average 3-6 weeks	388,22 ^a	230,02 °	306,92 ^b	322,00 ^b	331,50 ^b	322,12 ^b		

3.2 Weight gain Table 2. Average body weight gain for broiler chickens (g / head / week)

Description: Different letters on one line indicate that the numbers are significantly different (P <0.05). tn: not significantly different (P>0.05)

The results of analysis of variance showed that broiler chickens before E.coli infection did not have a significant effect (P > 0.05) on the average body weight gain. This is because the average previous feed consumption shows the same amount so that the rate of chicken body weight gain is also good and one of the factors that influence the body weight gain rate is feed consumption. This is in accordance with Kartadisastra's statement [7] (1994), which states that chicken body weight will be determined by the amount of feed consumption, the greater the body weight of the chicken the more the amount of feed consumption. Table 2 shows that the average body weight gain of broilers after E.coli infection had a significant effect (P <0.05). Duncan's follow-up test showed that POA was significantly different from POB, which were 388.22 g and 230.02 g, respectively. This means E.coli infection can inhibit weight gain in broiler chickens. This is consistent with the statement of Pierard, et al [8] (1990) which states that in infected broiler chickens from E. coli bacteria it is very bad. This disease can cause death during the maintenance period to gain weight when harvested chicken does not reach the standard. Average body weight gain after E.coli infection showed that POB treatment was significantly different from P1, P2, P3 and P4. The highest average is found in treatment P3 that is equal to 331.50 g and followed successively in treatment P4, P2, P1 and the lowest in treatment POB is 230.02 g. This means that the administration of gambir and tetracycline solutions can increase the average body weight gain of broilers infected with E.coli. This is suspected because gambier can be used as a diarrhea drug to fight pathogenic bacteria in the intestine so as to optimize the absorption of food substances in the digestive tract in broiler chickens. According to Rahmawati, et al. [9] (2013), which states that gambir stimulates the release of bile gums which helps smooth the digestive process in the stomach and intestines. Other functions are as a mixture of drugs, as a medicine for burns, headache medication, diarrhea medication, dysentery medicine, mouthwash, canker sores, and skin pain medication.



Graph 2. Average body weight gain for broiler chickens (g /head/week)

Graph 2 shows that the average body weight gain at week 3 to week 6 with the administration of gambier solution in treatment P1 (2%), P2 (4%), and P3 (6%) is seen giving the same graph pattern with the treatment chart pattern P4 (tetracycline) in broiler chickens infected with E.coli. This means that gambir is able to compensate for tetracycline in increasing the average body weight gain of broilers infected with E.coli.

Weeks			Т	reatment		
weeks	P0 _A	PO_B	P1	P2	P3	P4
Before infection						
Week 1 th	1,04	1,03	1,02	1,05	1,08	1,06
Week ke-2 th	1,51	1,53	1,55	1,50	1,56	1,54
Average 1-2 weeks ^{tn}	1,28	1,28	1,29	1,28	1,32	1,30
After infection						
Week 3	1,68 ^b	2,73 ^a	2,54 ^a	$2,35^{ab}$ $2,08^{ab}$	2,33 ^{ab}	$2,33^{ab}$ 2,07 ab
Week 4	1,97 ^b	2,80 ^a	2,15 ^{ab}	$2,08^{ab}$	$2,06^{ab}$	2,07 ^{ab}
Week 5	2,19 ^b	2,98 ^a	2,28 ^b	2,25 ^b	2,22 ^b	2,30 ^b
Week 6	2,42 ^b	3,62 ^a	2,55 ^b	2,47 ^b	2,44 ^b	2,56 ^b
Average 3-6 weeks	2,05 °	3,03 ^a	2,38 ^b	2,29 ^{bc}	2,26 ^{bc}	2,31 ^b

3.3 Feed Conversion Ratio

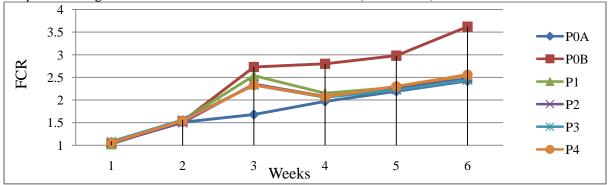
Table 3. Average feed conversion for broiler chickens (tails / week)

Description: Different letters on one line indicate that the numbers are significantly different (P <0.05). tn: not significantly different (P>0.05)

Table 3 shows that the average feed conversion before E.coli infection did not have a significant effect (P > 0.05) and the average feed conversion rate was very good (below 2). This is because the treatment of POA, POB, P1, P2, P3 and P4 is as good as digesting the nutritional value of the ration so that the feed conversion is not much different. According to Anggorodi [10] (1985) feed conversion is influenced by several factors such as age of livestock, nation, nutritional content of rations, conditions of temperature and condition of livestock, management and use of good seeds. According to Rasyaf [11] (1994), states that the smaller the conversion of rations means that the ration is more efficient, but if the conversion of the ration is enlarged, then there is waste of feed.

Table 3 shows that the E.coli-infected treatment had a significant effect (P < 0.05) on the average feed conversion. Duncan's test results showed that the POA treatment was significantly different from the treatment of POB respectively of 2.05 and 3.03. This means that E.coli infection in broiler chickens increases the average feed conversion. This is because the chicken's digestive tract is disrupted so that it cannot function optimally to maximize the utilization of nutritional ration values so that the increase in the ratio between feed consumption and body weight gain of broiler chickens. This is in accordance with Tabbu's statement [12] (2000) which states that the intestine that contains the most germs is the jejunum, ileum and cecum. Even though the chemical digestion process takes place in the small intestine. This is in accordance with the statement of Denbow [13] (2000) which states that trhenium has an important role in absorbing nutrients, amino acids, vitamins, and monosaccharides. Nutrient absorption by the duodenum, jejunum, and ileum is transferred into the blood circulation and lymph circulation throughout the body. for

Average feed conversion on the week after E.coli infection showed that P0B treatment was significantly different from P1, P2, P3 and P4. The highest feed conversion rate was found in P0B treatment at 3.03 and the lowest in P3 at 2.26. This shows that broiler chickens treated with gambir solution (P1, P2, P3) and tetracycline (P4) have a better ability to digest nutritional values from rations compared to untreated treatment (P4). According to Lacy and Vest [14] (2000), which states that some of the main factors that influence feed conversion are genetic, ration quality, disease, temperature, cage sanitation, ventilation, treatment, and cage management. Factor of rationing, lighting also plays a role in influencing feed conversion, ration travel rate in the digestive tract, physical form of ration and ration nutrition composition.



Graph 3. Average feed conversion ratio for broiler chickens (tails / week)

Graph 3 shows that the 4th week feed conversion average in treatment P1, P2, and P3 with a dose of 2%, 4% and 6% gambier solution decreased following the P4 chart pattern. This means that the treatment carried out during the third week was successful in improving the average feed conversion rate for broilers infected with E.coli. From Graph 3, it can be seen that P2 and P3 are able to match the POA treatment chart pattern without infection, meaning that P2 and P3 are capable of being as efficient as POA in digesting the nutritional value of rations. This is because the solution of gambir is known as an antibacterial which is also known to have high levels of natural antioxidants so that it can be efficient in digesting the nutritional value of rations. This source of natural antioxidants is thought to be able to reduce oxidative stress in chickens. According to Aditya and Putri [15] (2016), which states that the gambier plant (UncariagambirRoxb) is a plant containing derivatives of polyphenol compounds, namely catechins, tannins, epicatechin, querselinepigallocatechin and several other derivative compounds. One of the natural antioxidants is catechins which are polyphenols which have the potential as antioxidants and antibacterials. Most catechins are found in gambier plants, so gambier plants are known as antioxidants and antibacterials. According to Pambayun, et al [16] (2008), which states that the ability of gambir as an antibacterial is caused by polyphenols which are easily bound to other organic compounds, especially proteins. The formation of complex compounds causes the function and role of these compounds to decrease and even cause bacterial cell leakage and death.

Treatment	Final Weight (kg / head)	Income (Rp / head)	Feed consumption (kg / head)	Feed costs (Rp/ekor)	IOFC (Rp)	Drug costs (Rp)	IOFC (Rp/head) and Drug costs (Rp/head)
PO _A	1,94	36.863,80	3,69	28.870,25	7.993,55	-	7.993,55
P0 _B	1,31	24.922,93	3,19	24.976,66	-53,72	-	-53,72
P1	1,61	30.533,00	3,31	25.911,56	4.621,44	100	4.521,44
P2	1,68	31.882,00	3,40	26.600,60	5.281,40	200	5.081,40
P3	1,70	32.255,67	3,45	26.990,53	5.265,13	300	4.965,14
P4	1,67	31.757,87	3,45	26.991,58	4.766,29	250	4.516,29

3.4 Income Over Feed Cost (IOFC) and Drug Costs
Table 4. Average income over feed cost (IOFC) and drug costs for six weeks of research (Rp /head)

- - -

Description: The selling price of chicken is Rp. 19.000/Kg

Feed prices Rp.7830/Kg

Gambir Price IDR 12.000/Chunk (12 g)

Prices of tetracycline Rp. 2500/pack (10 g)

Table 4 shows that the values differ in IOFC broiler chickens. The highest average is found in the POA treatment of Rp. 7,993,55 followed by P2, P3, P4, P1 and the lowest was in the POB treatment of Rp. 53,72, which means that E.coli infected broilers reduced the IOFC value to reach the level of loss in POB treatment. This is related to the decreasing weight of livestock causing the selling price to decrease. In this study the best IOFC figures were found in the control treatment without infection (POA), because POA had the highest body weight compared to the infected treatment (POB, P1, P2, P3 and P4). The decline in income was due to the sale price of broiler chickens not based on the recorder but based on body weight, so that broiler chickens with treatment P0A, P0B, P1, P2, P3 and P4 gave different prices. This was supported by Rasyaf [17] (2003), which stated that Income Over Feed Cost was influenced by feed consumption, weight gain, feed costs and selling price per tail.

Table 4 shows that the treatment of E.coli infected with gambir solution (P1, P2, P3) and tetracycin (P4) showed higher IOFC prices compared to the infected treatment (P0B) without treatment. The average IOFC which has been reduced by the cost of drugs in the treatment of P1, P2, P3 and P4 respectively is Rp. 4,521.44; Rp. 5,081,40; Rp. 4,965.14 and Rp. 4,516.29. The highest average was found in P2 treatment followed by P3, P4, P1 and P0B at the level of loss. This is related to the conversion value of each treatment, where the lower conversion value is the higher the IOFC value of broiler chicken. The conversion of P0B ration was significantly different from P1, P2, P3, P4 (table 3. weeks after infection). This is in accordance with Rasyaf's statement [17] (2003), which states that in relation to the technical grip of production, it can be assumed that the more efficient the chicken is in turning food into meat, which means that the conversion of the ration is very good, the better the IOFC value.

4. Conclusion

Administration of gambir solution at a dose of 2% - 6% effectively improves performance (feed consumption, body weight gain, feed conversion and IOFC) broiler chickens infected with E. coli and able to compensate for the use of commercial antibiotics in the form of tetracycline. While the highest IOFC number is at 4% dose, that is Rp. 5,081.40 per head.

References

- Gordon, W.A.M. And Sri Poernomo. 1976. A Poultry Disease Index In Bogor. Analysis Of Poultry Disease Examination At The Poultry Diagnostic Laboratory, Lpph Bogor Between March 1973 And February 1974. Hemerazoa, 69: 36–39.
- [2] Wiedosari and Wahyuwardani. 2012. Case Study of Broiler Chicken in Sukabumi and Bogor District Pathology Laboratory Center for Veterinary Research, Bogor.
- [3] Hayani E. 2003. Analysis of Catechin Levels from Gambir with Various Methods. Agricultural Engineering Bulletin 8: 31-32.
- [4] Lucida H, Bachtiar A, and PutriWa. 2006. Formulation of Oral Antiseptic Preparations from GambirCatechins [Thesis]. Bogor: Faculty of Mathematics and Natural Sciences, Bogor Agricultural University.
- [5] Anggorodi, R.1990. General Animal Food Science. PT. Gramedia. Jakarta.
- [6] Akoso, B.T. 1993. Poultry Health Manual.First Edition.Kanisius.Yogyakarta.
- [7] Kartadisastra, H. R. 1994. Management of Chicken Feed.Kanisius.Yogyakarta.
- [8] Pierard, D., Van Etterijck, R. Brenaert. J., Moriau, L &Louwers, S. 1990. Results of Screening for Verocitotoxin-producing Escherichia coli in Belgium. European Journal of Clinical Microbiology and Infectious Disease 9, 198-201.
- [9] Rahmawati, N. Bakhtiar, A. Prima Putra, D. Isolation of Catechins from Gambir (UncariaGambir (Hunter). Roxb) for Pharmaceutical and Cosmetic Preparations. Indonesian Pharmaceutical Research Journal: 2012.
- [10] Anggorodi, R. 1985. General Animal Food Science.Gramedia Main Library, Jakarta. Anggorodi, R.1995. Various Poultry Nutrition.Gramedia Main Library. Jakarta.
- [11] Rasyaf, M. 1994. Farming broilers. Spreaders. Jakarta.

[12] Tabbu, C.R. 2000. Colibasilosis.Chicken disease and prevention.Kanisius, Jogyakarta. Vol. 1, Hal: 31-51.

- [13] Denbow DM. 2000. Gastrointestinal anatomy and physiology. Inside: Whittow JC, editor. Sturkie'sAvian physiology. 5th Ed. London: Academic Pr. pp. 299-325
- [14] Lacy, M. and L.R. Vest. 2000. Improving Feed Conversion in Broilers: A Guide For Growers.
- [15] Aditya. M., and Putri R.A. 2016.Benefits of Gambir (UncariaGambirRoxb) As an Antioxidant.Faculty of Medicine, University of Lampung.
- [16] Pambayun, R., Pangawikan, A.D., Widowati, T.W., Hamzah, B., Santoso, U., Yudono, B. and Gardjito, M. (2008). Utility of gambircatechin for tofu preservation. The 14th World Food Congress, International Union Food Science and Technology (IUFOST), Sanghai, China.
- [17] Rasyaf, M. 2003. Broiler Farming.Self-helpers. Jakarta.