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Conference Paper

Implementation of Meniran Extract (*Phyllanthus Niruri Linn*) on the Performance of Broiler Chickens Infected by *Mycoplasma gallisepticum* Caused Chronic Respiratory Disease

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Abstract

Background: *Chronic respiratory disease* (CRD) of chicken is a disease that has great economic losses in poultry industry in the world. The losses are mainly due to the decrease of body weight gain, feed efficiencies, hatchabilities and increase conversion of the feed, of embryo mortality. The main causative agent of *Chronic Respiratory Disease* (CRD) is *Mycoplasma gallisepticum*. *Mycoplasma gallisepticum* attacks the respiratory tract, especially in young broiler chickens with age ranged 3-5 weeks. CRD treatment usually uses macrolide antibiotics, because it has proven effective to inhibit protein synthesis. However, it is not recommended to continuously given because the chicken can be resistant to the medicine and leave a harmful residue to consumers. The development of herbal medicine utilization currently is mostly implemented for the treatment of diseases that infected livestock. *Meniran* plants (*Phyllanthus niruri Linn*) is one of the plants that can be used as prevention and alternative treatment caused by *Chronic Respiratory Disease* (CRD). *Meniran* (*Phyllanthus niruri Linn*) has the content of bioactive compounds that have antibacterial activity, including terpenoids, alkaloids, flavonoids, saponins, and tannins. The purpose of this study is to test and evaluate the effectiveness of *Meniran* extract (*Phyllanthus Niruri Linn*) on the performance of broiler chickens infected by *Chronic Respiratory Disease* (CRD), seen from the feed conversion.

Methods: The subject used in this study was 30 broiler chickens of Lohmann strains, the dose of *Mycoplasma gallisepticum* infection was 10^8 CFU/ml and also prepared meniran extract. This study used experimental method using completely randomized design with 5 treatments and 5 replicates. This study used meniran therapy dosage in each broilers with P1 = 60%/1ml/kgBB (body weight), P2 = 62,5%/ml/kgBB (body weight) and P3 = 65%/ml/kgBB (body weight) and PO(-) treatment without infected and without therapy, PO(+) treatment with infected and without therapy. The data was analyzed using analysis of variance and tested further by Duncan test.

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Results: The results showed that the addition of *meniran* extract could give different effect in the feed conversion. The treatment of P0 (+) was different with P0(-), P1, P2 and P3.

Conclusion: *Meniran* extract (*Phyllanthus Niruri Linn*) with a dose of 60% could improve the performance of broiler chickens infected by *Chronic Respiratory Disease* (CRD).

Keywords: *Meniran*, *Mycoplasma galisepticum*, *Chronic Respiratory Disease* (CRD), performance of Broiler Chickens, Feed Conversion.

1. Introduction

Broiler chickens is one of the cheap protein compared with cow or goat's beef. The advantage of broiler chickens is its fast growing, to make it quick sold before it reached 5 weeks with an average weight of 1,5 kilograms. Broiler chickens is very efficient in converting feed into meat. Feed is a very influential factor in determining the success of chicken maintenance, especially broiler chickens. Feed costs reach 60-70% of total production costs and improve protein efficiency. The use of local feed ingredients becomes an alternative to reduce production costs. The local feed ingredients that are used must have functions of feed like nutrition needed by livestock, cheap and easy to get. The main nutrients that has important function to the broiler growth are protein, energy (carbohydrates and fats), vitamins, minerals and water [16].

One of the obstacles that often delays the development of poultry populations is the outbreak of various pathogenic diseases. *Chronic respiratory disease* (CRD) in chickens is a pathogen endemic disease that causes great losses in the poultry industry not only in Indonesia, but also in the world [6].

Beside causes great losses in economic from upstream to downstream, *Chronic respiratory disease* (CRD) also causes suppression against immunosuppressive. It causes the body failing to obtain immunity from vaccinations [4]. In addition, infected chickens become carriers so the area where the farm is located becomes an endemic area. Treatment, prevention and control of *Chronic respiratory disease* (CRD) in both breeders and commercial chickens have been frequently conducted, but the prevalence of CRD still occur [11]. Infected chickens' price are usually cheaper, but carcasses that show *airsacculitis* are still acceptable after disposing its internal organs and cleaned washing

(personal observation). It shows that hygiene problems still become concerned of Indonesia. *Chronic respiratory disease* (CRD) in chickens is a disease that causes great losses in the poultry industry in the world. In Indonesia, the losses reach hundreds of billions rupiah per year [18].

These losses include decreasing egg production, fertility and hatchability in the range of 8 - 30%, embryo mortality 5 - 20%, mortality of chickens' infant 5-10%, weight gain obstructed 8 - 25%, and increased feed conversion [6]. Younger chickens are more susceptible to infection than adults, similar with roosters that are more vulnerable than hens [11]. Although the disease is pathogenic endemic and harmful to the poultry industry but until today CRD is less considered in Indonesia because the disease does not cause a large mortality epidemic. Currently, CRD is categorized in economic diseases, and has not been taken into account the impact that causes endemicity and immunosuppressive which has a great economic losses. Treatment of *Chronic Respiratory Disease* (CRD) is often conducted. However, until today CRD is still widespread throughout the world. It is known that *Mycoplasma gallisepticum* has no cell wall so penicillin and its derivatives are resistant and thus cannot be implemented for CRD treatment because of its antibiotic performance on the cell wall. Macrolid antibiotics such as tiamulin, tylosin, lincomycin, oxytetracyclin and enrofloxacin are usually implemented for CRD treatment, because it has performance to delay protein synthesis. However, it is not recommended to do continuous treatment with the same medicine, as it may leave harmful residues to the consumer of chicken products and may cause resistance [18].

The development of herbal medicine utilization currently is mostly implemented for the treatment of diseases that infected livestock. *Meniran* plants (*Phyllanthus niruri Linn*) is one of the plants that can be used as prevention and alternative treatment caused by CRD disease. *Meniran* (*Phyllanthus niruri Linn*) has the content of bioactive compounds that have antibacterial activity, including *terpenoids*, *alkaloids*, *flavonoids*, *saponins*, and *tannins*. *Lignans* are used as antioxidants in foods. In addition *lignans* are chemicals that are active in certain herbs as well. *Lignans* can be extracted with acetone or ethanol and often deposited as insoluble potassium salts [20]. *Flavonoids* are compounds of phenol, therefore the color changes when added bases or ammonia. Generally *flavonoids* are present in plants bound to sugar as glycosides and aglycans. *Flavonoids* are a combination of glycosides and present in all vascular plants. In vascular plants, some derivatives of *flavonoids* are found and only present in certain organs of plants such as those found in roots, stems, leaves, flowers, seeds, and bark. *Flavonoids* have function of which is to increase chicken growth and as an alternative to antibiotics in chicken farms [12]. *Tannin* uses as an anti-bacterial, where it lies within the plant

apart from cytoplasmic enzymes and proteins, but equation reactions can occur if the tissue is damaged [22].

⁴ The purpose of this study is to test and evaluate the effectiveness of *Meniran Extract* (*Phyllanthus Niruri Linn*) on the performance of broiler chickens infected by *Chronic Respiratory Disease* (CRD), seen from the performance of broiler chickens.

2. Methods

At the age of 15 days, male broiler chickens are randomly selected by lottery as much as 25 chickens to be adapted to the battery cage. At the age of 21 days the chickens infected by *Mycoplasma gallisepticum* bacteria as much as 1 ml/kgBB (body weight) intraperitoneally. *Meniran* extract given to the chickens at the age of 29 days, orally as much as 1ml/day/kgBB (body weight) for 6 days until reached 35 days. At age of 35 to 42 days it was then observed the effect of the added *meniran* extract. Making extract meniran ¹ with ethanol solven: *Meniran* plants that have been dried milled to obtain powder. Pollen meniran 1 kg extracted using maceration method by immersion in a solution of ethanol 96% as much as five liters for 3 x 24 hours. Stirring is done twice, morning and afternoon. Maceration process is performed three times. The results of the marinade in the form of filtrate is then filtered to further evaporated using a rotary evaporator which will yield a concentrated plant extracts meniran [9] and meniran application in broiler chicken.

The method in this study was experimental method composed based on *Rancangan Acak Lengkap (RAL)* / Completely Randomized Design with 5 treatments and 5 replications. The treatments that were tested listed as follows:

1. Group Po (-): A group of chickens without given any treatment (negative control).
2. Group Po (+): A group of chickens infected with *Mycoplasma galisepticum* without given *meniran* (positive control).
3. Group P1: A group of chickens infected with *Mycoplasma galisepticum* and given *meniran* extract at a dose of 60%.
4. Group P2: A group of chickens infected with *Mycoplasma galisepticum* and given *meniran* extract at a dose of 62,5%.
5. Group P3: A group of chickens infected with *Mycoplasma galisepticum* and given *meniran* extract at a dose of 65%.

¹¹ The variables observed in this study were feed consumption, weight gain (*PBB*) and feed conversion. The data obtained were analyzed using analysis of variance and if there were differences between treatments tested by Duncan's multiple-range test.

3. Results

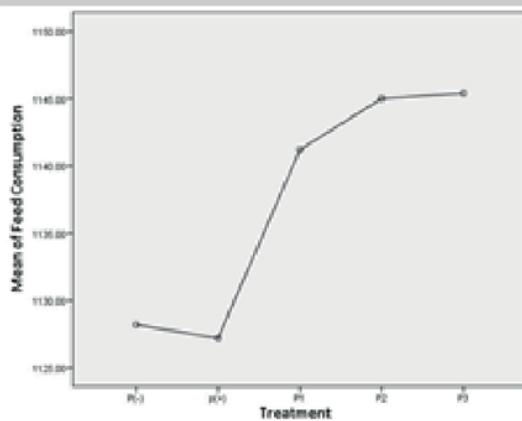
The observation results on the effect of *meniran* extract addition towards the performance of broiler chickens production that infected by *Mycoplasma galisepticum* were as follows:

3.1. Feed Consumption

Mean value of feed consumption among treatments.

Treatment	Mean value of \pm SD (deviation standard)
$P_{0(-)}$	1128.2 ^a \pm 39.902
$P_{0(+)}$	1127.2 ^a \pm 2.949
P_1	1141.2 ^a \pm 2.949
P_2	1145.0 ^a \pm 1.870
P_3	1145.4 ^a \pm 2.073

² Description: Different superscript on the same column showed significant value ($p < 0.05$)



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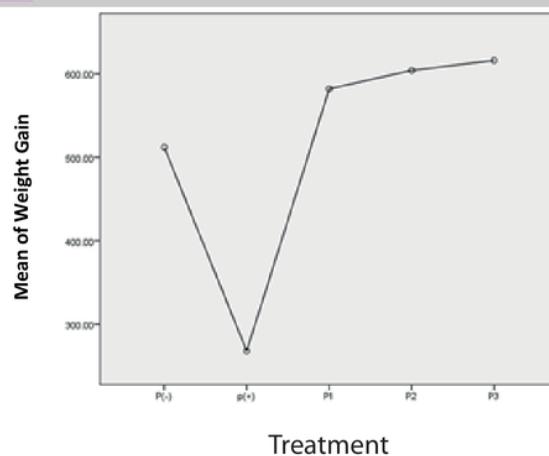
Based on ANOVA test, can be concluded that among treatments there were no significant difference in the feed consumption.

3.2. Weight gain

Mean of weight gain of broiler chickens among treatments.

Treatment	Mean value of \pm SD (deviation standard)
P ₀₍₋₎	512.0 ^b \pm 39.902
P ₀₍₊₎	268.0 ^a \pm 2.949
P ₁	582.0 ^{bc} \pm 2.949
P ₂	604.0 ^c \pm 1.870
P ₃	616.0 ^c \pm 2.073

2 Description: Different superscript on the same column showed significant value ($p<0.05$)



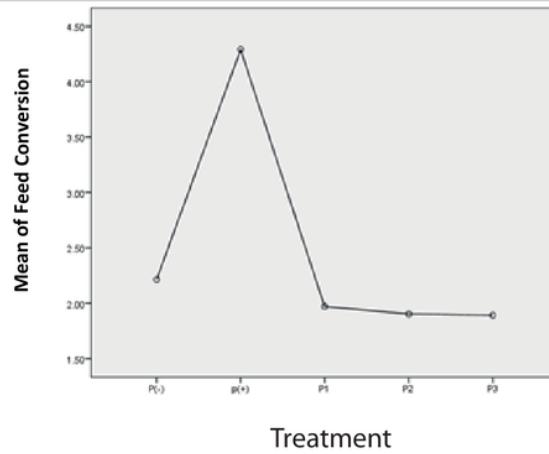
Based on the results of statistics analysis using ANOVA test, can be concluded that among treatments there were significant difference in weight gain ($p<0.05$), thus it continued using Duncan test with 5% significant level to determine the weight gain differences in each treatment. The Duncan test result showed that P₀₍₊₎ was different with P₀₍₋₎, P₁, P₂ and P₃. P_{0 (-)} showed real significant different with P_{0 (+)}, P₂ and P₃. P₁, P₂ and P₃ were not significantly different among treatments.

² 3.3. Feed Conversion

Mean of feed conversion of broiler chickens among treatments.

Treatment	Mean value of \pm SD (deviation standard)
P ₀₍₋₎	2.2148 ^a \pm .21025
P ₀₍₊₎	4.2904 ^b \pm .69998
P ₁	1.9710 ^a \pm .15893
P ₂	1.9030 ^a \pm .13032
P ₃	1.8928 ^a \pm .27954

² Description: Different superscript on the same column showed significant value ($p<0.05$)



Based on the results of statistics analysis using ANOVA test, can be concluded that among treatments there were significant difference in feed conversion ($p<0.05$), thus it continued using Duncan test with 5% significant level to determine the feed conversion differences in each treatment. The Duncan test result showed that P₀₍₊₎ showed the highest feed conversion result with real significant value of P₀₍₋₎, P₁, P₂ and P₃.

2. Discussion

Based on the results of this study showed that the group with the smallest dose treatment had shown influence to the increase of weight gain and decrease of feed conversion, so it can be concluded that *meniran* extract has influence to increase weight gain and decrease feed conversion in broiler chickens infected by *Mycoplasma gallicepicum* bacteria. Increased weight gain and decreased feed conversion showed good performance. In chickens infected by *Mycoplasma gallicepicum* bacteria or *Chronic Respiratory Disease* will decrease weight gain and increase feed conversion, caused by decreasing appetite to cause decreasing feed consumption and hampering weight gain [4].

Weight gain showed that the feed consumed by chickens was quite efficient and widely used for growth. If the chickens consumed a lot of food but the weight gain was low then it was suspected the food absorption in the chickens' digestive tract was not perfect. It could also be caused of the chickens were sick, besides other factors like species, temperature and food quality. This weight gain was in line with feed consumption which showed real significant influence differences. Based on the results of this study showed that although the food consumed decreased but there was increased weight in broiler chickens. Feed conversion is the ratio between feed consumption and weight gain in each week during one production period. Feed conversion involves chickens' growth and feed consumption. Rapid growth with low feed consumption showed high feeding efficiency. Feed conversion is one indicator of business success for breeders. Comparison of feed consumption and weight gain will result in a number as the basis of economic calculation.

Currently, CRD is included in the category of economic diseases, not yet counted for the endemicity and immunosuppressive impacts that have a great economic loss [18]. The mechanism of infection is that *Mycoplasma gallicepicum* enters through the nasal cavity and then attaches to epithelial receptors called *sialoglycoprotein (Patron recognition receptors sites)* mediated by adhesin and protein called *bleb (Pathogen associate molecular patrons)* located at the end of the mycoplasma cell organ [6]. Furthermore, mycoplasma cells penetrate and destroy the epithelial mucosa while multiplying. With the mediation of epithelial cilium movement and bleb, mycoplasmic cells move toward the abdominal air membrane pouch [11]. Inflammation occurring in epithelial tissue is not a result of mycoplasma toxin but is more due to immune responses from hosts in the form of inflammatory [18]. This inflammation causes inhibition to the development of *T helper 1 cell (Th1 cell)* so that the cytotoxic of *T cell* becomes inactive resulting in pathogen infection becoming persistent [8]. Another impact is an increase in the production of *Tumor necrosis factor α (TNFα)* which results in a decreased *Th2 cell*

response, resulting in neutralizing antibody responses to bacterial or viruses infections also decreasing drastically [5]. This condition concludes mycoplasma infection causes immunosuppression against infected chickens with *Mycoplasma gallisepticum*.

This study used *meniran* herbal plants to help improve the conversion of chicken feed infected by *Chronic Respiratory Disease*. In general, the benefits of using herbs for humans and animals is to increase the immune system (immunomodulator), disease prevention and health restoration [5]. *Meniran* acts as an appetite enhancer with its chemical content possessed [22]. In this study, allegedly giving herbs could stabilize the health conditions of chickens and improve the efficiency of chickens' feed. According to [8], *meniran* plants (*Phylanthus niruri Linn*) have immunomodulatory benefits, such as medicine that can restore and repair the immune system whose function is disturbed or to suppress the excessive function, besides increasing the immune [20]. The content of compounds in *meniran* extract such as *alkaloids* and *tannins* are capable of providing inhibitory activity against microbes [24]. In the *meniran* extract (*P. niruri L.*), there are active compounds of the *phenol class* (*flavonoids*), *alkaloids*, *saponins*, and *tannins*. *Flavonoids* are active *phenol* compounds. *Phenol* compounds and *phenolic* compounds derivatives can also cause denaturation of proteins present in cell walls thus can destroy the structure and change the permeability mechanisms of microsomes, lysosomes, and cell walls [17].

Alkaloids compounds are a group of active compounds derived from plants. The *alkaloids* performed as antibacterial by destroying the peptidoglycan component on the *bacterial* cell, so that the cell wall layer will not completely formed and caused *cell* mortality [20]. Secondary metabolite compounds of *saponins* are substances that can interact with bacterial cells then the bacterial cell wall will become lysis or break. Similarly, the compound of *tannin* which is a secondary metabolite compounds that in low concentration can inhibit the growth of bacteria by coagulated bacterial protoplasm [11] stated that the speed and efficiency of bacterial damage by antibacterial compounds was affected by temperature, *pH*, time, concentration and the presence of other organic components. According to 12) bacteria inhibited its growth by destroying cell walls, changing cell permeability, altering protein molecules and nucleic acids, coagulating protoplasm. *Alkaloids* are the largest group of active compounds of plants. *Phenol* compounds derived from plants have the ability to form complexes with proteins through hydrogen bonds that can inhibit the formation of proteins and nucleic acids. The work mechanism of *alkaloids* compounds in inhibiting the growth of bacteria is similar with the work mechanism of chloramphenicol antibiotics by inhibiting the formation of protein synthesis to interfere the bacterial metabolism. Mentioned that *phenol* compounds also contained -OH groups that could dissolve lipids in cell walls

to interfere and affect the integrity of cytoplasmic membranes and inhibit ATP-ase bonding in cell membranes and cause cell lysis. *Saponin* was a secondary metabolite compound that was widely present in nature and antimicrobial.

According to [20], *saponin* compounds when interacting with bacterial cell wall then the wall would break or lysis. *Saponin* would disrupt the surface tension of the cell wall, then when the surface tension interrupted, antibacterial substances would easily be able to enter cells and disrupting the metabolism to cause the bacteria's mortality. According to Masduki (1996), *tannins* had an antibacterial role by binding proteins so the formation of cell walls would be delayed. The delayed mechanism of *tannins* was through lysis bacterial walls caused by *saponins* and *flavonoids*, causing the *tannins* to easily enter into bacterial cells and coagulate the cell protoplasm. *Flavonoids* functioned to destroy the composition of cells and changed the permeability mechanism of the bacteria's cell wall, while *tannins* could suppress the development of fungus by inhibiting the formation of new cells so that the disruption of cell division that caused the growth of the fungus became abnormal. *Saponins* performed to disrupt the stability of the fungus' cell membrane so it became lysis [24].

5. Conclusion

Based on the results of this study, there are two conclusions as follows:

1. Giving *meniran* extract at a dose of 60% has influenced the increase of weight gain and decrease of feed conversion in broiler chickens infected by *Mycoplasma gallisepticum* bacteria.
2. Addition of *meniran* extract can improve the performance of broiler chickens infected by *Mycoplasma gallisepticum* bacteria caused Chronic Respiratory Disease (CRD)

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