# Novel Combination of *Andrographis paniculata* and *Phyllanthus niruri* to Improve Performance of Laying Hens Infected with *Escherichia coli*

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## Abstract

The study aims to learn the efficacy combination of *Phyllanthus niruri* and *Androgaphis paniculata* to improve the performance of laying hens infected with *Escherichia coli* pathogen. Fifty laying hens of Isa Brown strain were randomly divided into 5 treatments, each treatment was divided into 10 replications (n=10). Treatment P0- (control group without infected), P0+ (hens group infected with Avian Pathogenic's *Escherichia coli* without given extract), P1 (hens group infected with Avian Pathogenic's *Escherichia coli* with 10% *Phyllanthus niruri* extract and 30% *Androgaphis paniculata*), P2 (hens group infected with Avian Pathogenic's *Escherichia coli* with 20% *Phyllanthus niruri* extract and 20% *Androgaphis paniculata*) and P3 (hens group infected with Avian Pathogenic's *Escherichia coli* with 30% *Phyllanthus niruri* extract and 10% *Androgaphis paniculata*). Performances observed were feed consumption, Hen Day Production, eggs weight and feed conversion. Data analyzed by ANOVA and tested with the F test. The feed consumption showed in P1 was different from P3, P2, P0+ and P0-, P1 was different from P2 but not with P3, P0+, and P0- showed significantly different in each treatment. In P3 showed no differences with all treatments. The eggs weight showed different in P0+ for all treatments, while the other treatments in P1, P2, P3 and P0- showed no differences. The feed conversion showed in P0+ was different for all treatments, while other treatments showed no difference. P0+ treatment compared to (P0-, P1, P2) was significantly different, P0+ treatment compared to P3 treatment was not significantly different, between P3 and P1 treatment was not significantly different, and between P1 and P2 with P0- was not significantly different, also between P3 with P2 and P0- treatments was significantly different. Supply of *P. niruri* extract. and *A. paniculata* in laying hens can improve the performance of laying hens infected with *Escherichia coli* pathogen.

***Keywords:*** *Phyllanthus niruri, Andrographis paniculata, performance of laying hens, Escherichia coli*

## Introduction

The Laying hens are susceptible to colibacillosis which is caused by avian pathogenic *Escherichia coli* (APEC) as a primary or secondary agent. Colibacillosis causes growth problems, decreased production, increased number of abandoned chickens, decreased quality of carcasses and eggs, and supports the emergence of complex diseases of the respiratory, digestive and reproductive tracts which are quite difficult to treat[1]. The use of antibiotics in APEC really needs to pay attention toward the different sensitivity characteristics of Escherichia coli serotypes, some Escherichia coli serotypes are resistant to several antibiotics[2].

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Safe handling of bacterial diseases is to use medicinal plants. Indonesia as a tropical country has wealth of plants that have potential to become medicine. *Phyllanthus niruri* and *Andrographis paniculata* are plants that can be used as prevention and alternative treatment for APEC[3]. *P. niruri* is a plant that belongs to the genus *Phyllanthus* known as *P. niruri* which has antibacterial activity against APEC. *P. niruri* contains several chemical substances such as lignin, flavonoid, alkaloid, terpenoid, saponin and tannin[4]. *A. paniculata*'s constituent active compounds are lactone, tannin, saponin, alkaloid, flavonoid, and andrographolide which can increase immunity. The content of Andrographolide in *A. paniculata* can interfere the transfer pathways of viral and bacterial genetic material so it is effective against infectious agents[5].

This study aims to learn the efficacy combination of *P. niruri* and *A. paniculata* to improve the performance of laying hens (in the form of feed consumption, Hen Day Production, egg production, and feed conversion) infected with *Escherichia coli*.

## Materials and Methods

The fifty laying hens of Isa Brown strain were randomly divided into 5 treatments, each treatment was divided into 10 replications (n=10). Treatment P0- (control group without infected), P0+ (hens group infected with Avian Pathogenic's *Escherichia coli* without given extract), P1 (hens group infected with APEC with 10% *P. niruri* extract and 30% *A. paniculata* , P2 (hens group infected with APEC with 20% *P. niruri* extract and 20% *A. paniculata*) and P3 (hens group infected with APEC with 30% *P. niruri* extract and 10% *A. paniculata*). Performances observed were feed consumption, Hen Day Production, egg production and feed conversion. Data analyzed by ANOVA and tested with the F-test.

## Results and Discussion

**Table 1. Mean of Feed Consumption (gram/day) and Standard Deviation in Layer Hens Infected by *Escherichia coli* with *Phyllanthus niruri* and *Andrographis paniculata* Extracts Therapy.**

| Treatment | Mean ± SD |
|---|---|
| P0 (-) | 120.80ab ± 6.52117 |
| P0 (+) | 118.99ab ± 1.39208 |
| P1 | 114.05a ± 2.19062 |
| P2 | 122.62b ± 5.35530 |
| P3 | 118.05ab ± 4.80858 |

* Different superscripts in the same column show significant values ($p$ <0.05)

Based on ANOVA statistical analysis, there was significant difference in feed consumption ($p$ <0.05), then continued with Duncan test with a significant level of 5% to compare the differences obtained in other treatments. The results of Duncan test showed that P1 was different from P3, P2, P0+ and P0-, P1 was different from P2 but not with P3, P0+, and P0- showed significantly different in each treatment. P3 showed no differences with all treatments.

**Tabel 2. Mean of Egg Production (gram/day) and Standard Deviation in Layer Hens Infected by *Escherichia coli* with *Phyllanthus niruri* and *Andrographis paniculata* Extracts Therapy.**

| Treatment | Mean ± SD |
|---|---|
| P0- | 61.81b ± 2.79 |
| P0+ | 57.22a ± 0.76 |
| P1 | 61.05b± 3.05 |
| P2 | 64.04b ± 2.34 |
| P3 | 63.20a ± 2.26 |

*Note: [a,b,c,d,e]Different superscripts in the one column show significant values ($p$ <0.05)

Based on ANOVA statistical analysis, there was significant difference in feed consumption (p <0.05), then continued with Duncan test with a significant level of 5% to compare the differences obtained in other treatments. The results of Duncan test showed different in P0+ for all treatments, while the other treatments in P1, P2, P3 and P0- showed no differences.

**Table 3. Mean of Hen Day Production and Standard Deviation in Layer Hens Infected by *Escherichia coli* with *Phyllanthus niruri* and *Andrographis paniculata* Extracts Therapy.**

| Treatment | Mean Hen Day Production (%) ± SD |
|---|---|
| P0- | 98,25b ± 3,5 |
| P0+ | 87,75a ± 3,5 |
| P1 | 94,75ab ± 3,5 |
| P2 | 96,50b ± 4,04 |
| P3 | 91,25ab ± 6,70 |

*Different superscripts in the same column show significant values ($p < 0.05$).

Based on ANOVA statistical analysis, there was significant difference in feed consumption ($p$ <0.05), then continued with Duncan test with a significant level of 5% to compare the differences obtained in other treatments. The results of Duncan test showed P0+ was different from P0- and P2 was not different from P1 and P3. P0- was different from P0+ and showed no differences with P1, P2, P3. P1 showed no differences with all treatments. P2 was different from P0+ and showed no difference with the other treatments. P3 showed no differences with all treatments.

**Tabel 4. Mean of Feed Conversion and Standard Deviation in Layer Hens Infected by *Escherichia coli* with *Phyllanthus niruri* and *Andrographis paniculata* Extracts Therapy.**

| Treatment | Mean ± SD |
|---|---|
| P0- | 1.95a ± 0.027 |
| P0+ | 2.07b ± 0.034 |
| P1 | 1.87a ± 0.055 |
| P2 | 1.91a ± 0.056 |
| P3 | 1.87a ± 0.067 |

*Note: [a,b, c,d] Different superscripts in the one column show significant values ($p$ <0.05).

Based on ANOVA statistical analysis, there was significant difference in feed consumption ($p$ <0.05), then continued with Duncan test with a significant level of 5% to compare the differences obtained in other treatments. The results of Duncan test showed P0+ was different for all treatments, while other treatments showed no difference.

*P. niruri* and *A. paniculata* contains tannin, flavonoid, saponin dan alkaloid compounds[6]. *P. niruri* contains terpenoid, flavonoid, alkaloid, saponin and tannin compounds. According to Gunawan immune cells to increase the immune system[7]. Flavonoids inhibit the function of cell membrane by interfering with the peptidoglycan constituent components in bacterial cells so that the cell wall layer is not formed completely which causes cell death[8]. Alkaloids in *P. niruri* are alkaline compounds containing nitrogen atoms that function as antimicrobial, antimalarial, antidiarrheal and antidiabetic. Alkaloids work by destroying the peptidoglycan constituent components in the bacterial cell wall and inhibiting the synthesis of nucleic acids, thereby inhibiting the energy metabolism of bacterial cells[9]. Saponins are antimicrobial, these compounds can reduce the surface tension of the cell walls which causes the cell walls to lysis and eventually bacterial death[10]. Tannin compounds, which are compounds of *P. niruri*, have mechanism of action to inhibit and kill bacteria that react with bacterial cell membrane and destroy or inactivate the function of genetic material in bacterial cells[11]. The mechanism of tannin compounds is that they enter into the cell walls of bacteria that have been lysed due to the action of saponin and flavonoid compounds so tannin compounds can easily enter the bacterial cell wall and coagulate the protoplasm of bacterial cells. Tannins also have target on cell wall polypeptides so the formation of the cell walls becomes imperfect which causes bacterial cells to become lysed due to osmotic or physical pressure, so the bacterial cells will die. The improvement in egg production is also influenced by terpenoids which act as antibacterial inhibitors of *E. coli*[12]. *A. paniculata* has an active component, namely andrographolide which has an antibacterial effect against various microbes by damaging the bacterial cell membrane resulting in inhibition of specific enzyme biosynthesis and enhancing the immune system[13]. Tannins are included polyphenol compounds that can inhibit bacterial cell adhesion, inhibit enzymes and disrupt protein transport in the cell layer so bacterial cells become lysed due to osmotic pressure and physical pressure, while flavonoid compounds can inhibit bacterial growth by damaging the arrangement of the plasma membrane and cause changes in the permeability of the bacterial cell wall at low concentrations[14]. Saponins have antibacterial mechanism by reducing the surface tension of the bacterial cell walls so that they interfere with the survival of the bacteria. Alkaloid compounds function as antibacterials and have mechanism of destroying the components of peptidoglycan in bacterial cells so the bacterial cell wall is not formed completely and causes cell death[15]. Supply of *A. paniculata* and *P. niruri* extracts combination causes healing in layer hens infected with APEC and results in returning to the normal process of egg formation and egg production. Supply of combination dose 20%:20% of *P. niruri* and *A. paniculata* extracts improves the performance of laying hens.

On the other hand, Indonesia is an archipelago with approximately 17,508 islands and is covered by tropical rain forest, seasonal forest, swamp, subalpine shrub vegetation, coastal vegetation, and mountain vegetation. With its reflective mixture of Asian and Australian native species, Indonesia is stated to possess the second largest biodiversity in the world, with around 40,000 endemic plant species including 6,000 medicinal plants. Consequently, Indonesia is rich in medicinal plants which were used by its population traditionally from generation to generation in curing. Therefore, natural resources in Indonesia are very supportive for the use of herbal medicine-based therapies in in vitro or in vivo tests[16,17,18,19,20].

## Conclusion

In conclusion, from the result of this study, it can be concluded that *P. niruri* linn and *A. paniculata* combination improves the performance of laying hens infected with *E. coli*.

**Conflict of Interest:** The authors declare that they have no conflict of interest.

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