Neem (Azadirachta indica) Leaf Powder as Phytogenic Feed Additives Improves the Production Performance, and Immune Organ Indices of Broiler Chickens

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Abstract | In recent years, incorporating phytogenic feed additives as an alternative to synthetic antibiotics is promising in poultry production. It is commonly added to poultry diets to boost immunity and improve production performance. This study was conducted to ascertain the potential of neem leaf powder (NLP) on broiler chickens’ production and cell-mediated immunity. Sixty Cobb broiler chickens were distributed to four experimental treatments containing graded levels of NLP, 0% (T₁), 2% (T₂), 4% (T₃), and 6% (T₄) and arranged in a Completely Randomized Design experimental set-up. Each treatment was replicated three times, with five birds in each replication. The bi-weekly body weight gain (BWG), average daily gain (ADG), voluntary feed intake (VFI), feed conversion ratio (FCR), cell-mediated immunity, and return above feed and chick costs were observed within 42 days experimental period. The results showed significant differences (p<0.05) from all parameters, except for the cell-mediated immunity. The feed intake of broiler chickens was significantly reduced, whereas birds without NLP got the highest feed intake. Although feed intake of broiler chickens fed with NLP was significantly reduced, the body weight gain was not affected. Also, the feed conversion ratio of broiler chickens fed with 4% NLP showed better than birds fed with 0%, 2%, and 6% NLP. In return above feed and chick cost, birds fed with 4% NLP is more profitable, and the income generated increases as high as 25.73% compared to the birds fed without NLP in the diet. In conclusion, 4% NLP could be incorporated into the diets of broiler chickens without fear of compromising growth and immunity responses.

Keywords | Neem leaf powder, feed additives, growth performance, weight gain, cell-mediated immunity

INTRODUCTION

Poultry production is one of the most dynamic and ever-expanding sectors in the world. It helps fill the gap between the requirement and availability of high-quality protein for human consumption (Alkhalf et al., 2010). However, producing good quality meat and eggs without harmful residues within a short time interval is challenging in the poultry industry (Uko and Kamalu, 2008). In 2019, the Philippines chicken industry increased up to 14.72% of the total value of the agriculture industry, equivalent to 25,941 billion compared to the third quarter of 2017 and 2018 accounted for 13.27% and 13.96%, respectively (PSA, 2019; Galang, 2019). The production volume grew from 8.48% to 465,150 metric tons during the third quarter of 2019 (PSA, 2019; Galang, 2019). This was brought by increased demand as consumers shifted from pork to chicken meat due to the African Swine Fever outbreak.
Thus, there is an increasing demand for chicken meat and its by-products in the market.

One of the significant constraints in poultry production is the feed cost, accounting for 70–80% of the production cost, especially in developing countries (Siahan et al., 2021; Zacaria and Ampode, 2021). Also, incorporating synthetic antibiotics into poultry diets to boost production performance and meet animal protein demand is a public health concern (Lagua and Ampode, 2021). Synthetic antibiotics as growth promoters are expensive and have an adverse effect on the consumer’s health due to the survival of the residues of antibiotics in the tissues of the birds (Ezzat et al., 2018). Hence, the ban on adding antibiotics into poultry diets has sought producers to utilize alternative natural feedstuffs to meet the demands (Hossain et al., 2012).

It was found out that phytogenic feed additives such as Curcuma longa, Moringa oleifera, Echinacea purpurea, Azadirachta indica, and other herbs boost production performance by the increasing growth rate, better feed conversion ratio, greater livability, enhanced immune stimulation, lower mortality in poultry and reduced total production costs (Kafi et al. 2017, Maass et al., 2005; Roth-Maier et al., 2005, Windisch et al., 2007).

Neem tree (Azadirachta indica) is considered a perennial tree under the mahogany Meliaceae family. It is a herbal plant that exhibits various beneficial pharmacological properties, including immunomodulatory effects in broilers (Jawah et al., 2013; Upadhyay et al., 1992). Several feeding trials using neem leaves in animal production were conducted, especially as an anti-helmintic agent. However, most studies used neem seeds as a protein source in animal feed (Aruwayo et al., 2011). The medicinal properties of this plant as antifungal, antiviral, antibacterial, and growth promoter manifested its significance without adverse effects on chickens (Ubu et al., 2019). However, neem leaf meal has anti-nutritional content such as sodium nimbolide, gallic acid, azadirachtin, and nimbidin, affecting nutrient utilization (Kharde and Soujanya, 2014).

Hence, its usage is drastically reduced in feeds to capture beneficial effects with less adverse effects. The addition of neem leaves into ruminant feeds also enhanced plants’ utilization and decreased the severe feed inadequacy during the dry season (Tiwary & Pandey, 2008).

Neem leaves have been found to have a higher crude protein concentration than any other non-leguminous tree leaves (Adjorlolo et al., 2016). Moreover, it has low fiber and is considered a suitable protein supplement for ruminants in poor-quality diets. Neem leaves were also discovered to be a useful dry season fodder species where ruminant feeding during the prolonged dry season is a significant concern (Adjorlolo et al., 2016). However, few studies about neem leaf powder as feed additives have reported no adverse effect of feeding to poultry and livestock. Thus, this study investigates the potential of Neem leaf powder for broiler chickens’ production and cell-mediated immunity.

MATERIALS AND METHODS

Birds, diets, and management
The study was carried out following the standard rearing of farm animals as stipulated in the Good Animal Husbandry Practices of the Philippines concerning animal farming, health, and welfare (PNS/BAFPS, 2008). A week before the arrival of the experimental birds, the brooding house was constructed at an elevated type made of bamboo slats, disinfected, and cleaned thoroughly. A total of 60 at one-day-old Cobb broiler chicks were purchased from a reliable Agrivet supply in Tacurong City, Philippines, and housed at Ala, Esperanza, Sultan Kudarat. During the brooding period, artificial light was provided for twenty-four (24) hours for fourteen (14) days to regulate the birds’ body temperature. The bulb was placed at the center of the brooding cage to allow the equal distribution of heat.

After the brooding period, the experimental birds were randomly distributed into four treatments and replicated three times, with five birds in every replication. All broilers were raised in a wire-floored pen, measuring 1x1 square meter per bird, fed ad libitum, and individual waterer and feeding trough was provided for each pen. The feeding trial lasted for 42 days with two feeding periods, the starter and the finisher phase. The experimental birds were given a starter ration from 15 to 28 days and gradually shifted to a finisher ration from 29 to 42 days. The formulated diets met the nutrient requirements based on the Philippine Recommends Livestock Feed Formulation (PCAARRD, 2000). The experimental diet was incorporated with graded levels of Neem Leaf Powder at 0%, 2%, 4%, and 6% of the starter and finisher phases (Table 1).

Collection and preparation of neem leaf powder
The fresh neem leaves were collected in the locality of Ala, Esperanza, Sultan Kudarat. The fresh leaves were washed thoroughly using clean water to remove dirt and other unwanted matters and air-dried for seven (7) days. The dried leaves were ground using an attrition mill, sieved through a 1 mm sieve to produce neem leaf powder, and stored in large plastic containers with tight-fitting lids until needed (Dumaup and Ampode, 2021). The neem leaf powder was subjected to the proximate analysis following the AOAC (2016) procedure, and the chemical analysis was used in formulating the experimental rations.
### Table 1: Composition and Chemical Analysis of Starter and Finisher Ration

| Ingredients                                      | T₁ (% as fed basis) | T₂ | T₃ | T₄ | T₁ (%) | T₂ | T₃ | T₄ |
|--------------------------------------------------|---------------------|-----|-----|-----|--------|-----|-----|-----|
| Ground Yellow Corn                               | 50.00               | 49.00 | 50.00 | 50.00 | 54.30 | 51.40 | 50.05 | 50.20 |
| Rice Bran D₁*                                     | 11.00               | 12.80 | 12.00 | 12.00 | 12.00 | 12.00 | 12.00 | 11.00 |
| Soybean (US)                                     | 20.00               | 18.00 | 14.00 | 20.00 | 14.00 | 15.00 | 14.00 | 14.00 |
| Fish Meal, 60%                                    | 7.00                | 7.20  | 7.00  | 6.00  | 8.00  | 8.00  | 8.00   | 8.00   |
| Copra Meal                                       | 10.00               | 10.00 | 7.00  | 5.00  | 10.00 | 10.00 | 10.00 | 10.00 |
| Neem Leaf Meal                                   | 0.00                | 2.00  | 4.00  | 6.00  | 0.00  | 2.00  | 4.00   | 6.00   |
| Dicalcium phosphate                              | 0.40                | 0.20  | 0.16  | 0.16  | 0.40  | 0.40  | 0.50   | 0.20   |
| Limestone                                        | 1.00                | 0.20  | 0.20  | 0.10  | 0.90  | 0.80  | 1.00   | 0.20   |
| Lysine HCL                                       | 0.10                | 0.10  | 0.20  | 0.20  | 0.05  | 0.05  | 0.10   | 0.05   |
| D-L Methionine                                   | 0.10                | 0.10  | 0.10  | 0.20  | 0.05  | 0.50  | 0.05   | 0.05   |
| L-Threonine                                      | 0.10                | 0.16  | 0.16  | 0.10  | 0.05  | 0.05  | 0.05   | 0.05   |
| Vit. Premix¹                                      | 0.20                | 0.20  | 0.20  | 0.20  | 0.20  | 0.20  | 0.20   | 0.20   |

Calculated Analysis (%DM)

| Moisture                                         | 10.44               | 10.43 | 10.22 | 10.79 | 12.79 | 12.61 | 12.90 | 12.67 |
| Crude Protein                                    | 20.04               | 20.54 | 20.57 | 20.66 | 19.91 | 19.59 | 19.08 | 19.85 |
| ME (kcal/kg)²                                    | 2884                | 2864  | 2905  | 2926  | 2791  | 2887  | 2869  | 2870  |
| Ash                                             | 5.29                | 4.63  | 4.38  | 4.58  | 11.24 | 11.44 | 10.27 | 8.05  |
| Calcium                                          | 0.84                | 0.86  | 0.84  | 0.84  | 0.84  | 0.82  | 0.83   | 0.84   |
| Available Phosphorus                             | 0.48                | 0.49  | 0.45  | 0.47  | 0.43  | 0.45  | 0.46   | 0.42   |
| Lysine                                           | 1.13                | 1.11  | 1.13  | 1.13  | 1.13  | 1.06  | 1.10   | 1.14   | 1.09  |
| Methionine                                       | 0.45                | 0.46  | 0.44  | 0.47  | 0.44  | 0.48  | 0.50   | 0.51   |
| Meth + Cys                                       | 0.51                | 0.55  | 0.53  | 0.58  | 0.44  | 0.48  | 0.50   | 0.53   |
| L-Threonine                                      | 0.92                | 1.09  | 1.27  | 1.45  | 0.80  | 1.01  | 1.18   | 0.74   |
| Tryptophan                                       | 0.31                | 0.29  | 0.27  | 0.29  | 0.31  | 0.28  | 0.31   | 0.29   |

¹D₁ is a category of rice bran which has fine quality/texture.

¹Vitamin Mineral Premix: Vit. A 12000000iu, Vit D₃ 20000000iu, Vit. E 15000mg, Vit. K₃ 2000mg, Vit. C 10000 mg, Vit. B₁ 2000mg, Vit. B₂ 4000mg, Vit. B₆ 3000mg, Vit. B₁₂ 25000mg, Follic Acid 700mg, Pantothenic Acid 15000mg, Biotin 10000mg, Niacin 25000mg.

²Metabolizable Energy.

**Growth performance**

The initial weight (g/bird) was taken on the 15th day (right after brooding) and recorded at the start of the study. The final weight was determined at the end of the study (after 42 days) using a digital weighing scale maximum of 40kg with a difference of 5g. The birds’ final weight was subtracted from their initial weight to observe the weight gain, and the body weight gain (BWG) was measured every two (2) weeks to monitor their weight gain. The voluntary feed intake (VFI) was determined by offering a weighted amount of feed and subtracted by the feed refused every morning. The feed conversion ratio (FCR) was computed by comparing the amount of feed consumed to the broilers’ body weight gain.

**Cell-mediated immunity**

The broiler chickens were slaughtered following the guidelines of the Philippine National Standard (PNS/BAFS 103:2016) Code of Halal Slaughtering Practices for Poultry. Before slaughtering, the broiler chickens undergo fasting for twelve (12) hours, and birds nearest to the mean weight in every replication were slaughtered to determine the immune response by weighing the lymphoid organs, the bursa of Fabricius, and spleen. The bursa of Fabricius...
Table 2: Proximate composition of Neem Leaf Powder (NLP)

| Parameters Analysis (%) |
|-------------------------|
| Dry Matter 88.94 |
| Moisture Content 11.06 |
| Crude Protein 20.58 |
| Crude Fiber 14.13 |
| Ash 11.53 |

The analysis was performed in triplicate samples following the methods described by the AOAC (2016)

Table 3: Effects of Neem leaf powder on the growth performance of broiler chickens

| Parameters (days) | Treatments | T₃ (%) | T₄ (%) |
|------------------|------------|--------|--------|
| Body Weight (g)  |            |        |        |
| 15-28            | T1 0%      | 924.73±3.05  | 960.73±2.76 |
|                  | T2 2%      | 952.20±1.64  | 937.00±3.86 |
|                  | T3 4%      | 1706.00±5.72 | 1468.67±4.04 |
|                  | T4 6%      | 1158.67±1.23 | 1148.67±4.04 |
| Body Weight Gain (g) |                  |        |        |
| 15-28             | T1 0%      | 640.67±4.21  | 674.33±2.08 |
|                   | T2 2%      | 665.27±3.00  | 653.00±3.54 |
|                   | T3 4%      | 753.80±4.92  | 531.67±3.82 |
|                   | T4 6%      | 1184.67±50.31| 1184.67±50.31 |
| Daily Gain (g)    |            |        |        |
| 15-28             | T1 0%      | 45.76±0.30   | 48.17±0.14 |
|                   | T2 2%      | 47.52±0.21   | 46.64±0.25 |
|                   | T3 4%      | 53.84±0.35   | 37.98±0.27 |
|                   | T4 6%      | 42.31±1.79   | 42.31±1.79 |
| Feed Intake (g)   |            |        |        |
| 15-28             | T1 0%      | 740.93±6.26  | 736.53±3.61 |
|                   | T2 2%      | 719.33±5.91  | 742.60±4.93 |
|                   | T3 4%      | 1431.33±10.60| 1417.40±1.00 |
|                   | T4 6%      | 2160.00±4.61 | 2160.00±4.61 |
| Feed Conversion Ratio |          |        |        |
| 15-28             | T1 0%      | 1.15±0.01   | 1.09±0.01 |
|                   | T2 2%      | 1.08±0.01   | 1.11±0.01 |
|                   | T3 4%      | 1.90±0.02   | 2.67±0.01 |
|                   | T4 6%      | 1.52±0.08   | 1.83±0.08 |

abcd Means+SD with different superscripts in the same row differ significantly. ** (P<0.01)

Table 4: Effects of Neem leaf powder on the immune organs of broiler chickens

| Parameters Treatments | T₁ (%) | T₂ (%) | T₃ (%) | T₄ (%) | P – value |
|-----------------------|--------|--------|--------|--------|-----------|
| Bursa weight, g       | 3.63   | 4.40   | 3.93   | 3.93   | 0.979 ns  |
| Spleen weight, g      | 0.83±11 | 0.67±0.57 | 0.83±0.6 | 0.63±0.6 | 0.020**   |
| Spleen Index          | 0.06±0.00 | 0.04±0.00 | 0.05±0.00 | 0.04±0.00 | 0.053 m   |
| Bursa Index           | 0.24±0.20 | 0.29±0.13 | 0.23±0.08 | 0.27±0.15 | 0.951 ns  |

Means+SD with different superscripts in the same row differ significantly; ns: not significant; **: P<0.01

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Table 5: Return above feed and chick cost of broiler chickens fed with graded levels of Neem leaf powder

| Parameters                              | Treatments | $T_1$ 0% | $T_2$ 2% | $T_3$ 4% | $T_4$ 6% |
|-----------------------------------------|------------|----------|----------|----------|----------|
| Final live weight, g                    |            | 1492.67  | 1492.67  | 1706.00  | 1468.67  |
| Price/kg live weight (PhP)              |            | 130.00   | 130.00   | 130.00   | 130.00   |
| Gross return/head (PhP)                 |            | 194.05   | 194.05   | 221.78   | 190.93   |
| Cost of DOC/head (PhP)                  |            | 30.00    | 30.00    | 30.00    | 30.00    |
| Feed Consumption (kg/head)              |            |          |          |          |          |
| Chick Booster Mash (kg)                 |            | 0.33     | 0.33     | 0.33     | 0.33     |
| Starter ration (kg)                     |            | 0.74     | 0.74     | 0.72     | 0.74     |
| Finisher ration (kg)                    |            | 1.63     | 1.41     | 1.43     | 1.42     |
| Price/kg of feed (kg)                   |            |          |          |          |          |
| CBM (kg)                                |            | 32.00    | 32.00    | 32.00    | 32.00    |
| Starter ration (kg)                     |            | 22.98    | 22.71    | 21.39    | 22.49    |
| Finisher ration (kg)                    |            | 22.47    | 22.95    | 21.93    | 21.70    |
| Total Feed Cost (PhP)                   |            | 10.56    | 10.56    | 10.56    | 10.56    |
| Chick Booster Mash (kg)                 |            | 17.01    | 16.81    | 15.40    | 16.64    |
| Starter ration (kg)                     |            | 36.63    | 32.36    | 31.36    | 30.81    |
| Finisher ration (kg)                    |            | 94.19    | 89.72    | 87.32    | 88.02    |
| RAFCC (PhP)*                            |            | 99.86    | 104.33   | 134.46   | 102.91   |

*All costs were shown in PhP (Philippine peso); 1 USD = 50.30 PhP

RESULTS

PROXIMATE COMPOSITION OF NLP
The chemical analysis of Neem leaf powder (NLP) exhibited 88.94% dry matter, 20.50% crude protein, 14.13% crude fiber, 11.06% moisture content, and 11.53% ash (Table 2).

GROWTH PERFORMANCE
The broiler chickens fed with different levels of NLP showed a significant effect (p<0.05) on the final weight, bi-weekly and average daily weight gain, voluntary feed intake, and feed conversion ratio at 15-42 days (Table 3). Numerically, birds fed with 4% NLP ($T_3$) obtained the highest values among the parameters, including the final weight 1706.00±5.72, BWG (1411.0±71.44 g/bird), ADG (50.39±2.55 g/bird), and FCR with 1.52±0.08 compared to the birds without NLP in the diet. The values recorded for the cumulative mean of the final weight ranges from 1492.67±5.96 to 1706.00±5.72 g/bird, body weight gain 1184.67±50.31 to 1419.07±70.59 g/bird, average daily weight gain 42.31±1.79 to 50.68±2.52 g/bird, and mean daily feed intake was 2143.40±5.96 to 2366.27±16.58 g/bird. On the other hand, broiler chickens fed with 2% NLP ($T_2$) showed the least values from the BWG, ADG,
and VFI. The feed conversion ratio ranged from 1.52±0.08 (better) and 1.96±0.047, and it was significantly (p<0.05) different between dietary treatments where birds fed with 4% NLP (T1) had the lowest value. In the present study, a FCR of 1.52±0.08 (T1) means that the chickens gained 1 kilogram of weight for every 1.52±0.08 kilograms of feed intake (Table 3).

**Cell-Mediated Immunity**

The broiler chickens fed with graded levels of NLP had no significant differences (p>0.05) on the bursa weight, spleen and bursa indices of broiler chickens (Table 4). Although not significant, the numerical values of bursa weight and bursa index in birds fed with NLP are higher than those without NLP in the diet. On the other hand, the spleen weight (lymphoid organ) showed a significant difference (p<0.05) between each treatment where birds fed with 4% NLP (T3) got the highest value but still comparable to the birds fed with 0% NLP (T1) and 2% NLP (T4) in the diet. Moreover, the broiler chickens fed with 6% (T5) NLP got the lowest spleen weight but were statistically comparable to the birds with 2% (T2) NLP in the diet.

**Return and Above Feed and Chick Cost**

With the same amount of price per kilo (Php130/kilo), broiler chickens fed with 4% NLP (T3) got the highest final weight with 1706.00g/bird was a good asset for a higher market with 221.78Php gross income per chicken compared to the birds fed with 6% NLP (T5) with 190.93Php, and birds fed with 0% (T1) and 2% (T2) NLP with 194.05Php/bird (Table 5). As a whole, feed diets incorporated with NLP showed lower production costs than diets without NLP. With this result, broiler chickens fed with 4% (T3) NLP got the highest return above feed and chick cost amounting to 104.33 Php/bird than the birds without NLP (T1). 148.02 Php/bird (Table 5).

**DISCUSSION**

**Growth Performance**

In the present study, the inclusion of graded levels of NLP into the diet significantly improved (p<0.05) the growth performance of broiler chickens. However, the significant effect of NLP on the growth performance of broiler chickens is contrary to the findings of Deore et al. (2005), who reported that supplementation of neem oil in broilers resulted in poor performance in feed consumption and body weight, showing a dose-dependent adverse effect on the production performance. In addition, Shihab et al. (2017) reported that the inclusion of neem leaf powder in broiler diets had no significant (p=0.05) effect on the body weight and weight gains of broiler chickens. On the other hand, the result of the current study confirmed the investigation of Kharde and Soujanya (2014), who reported that male Venn Cobb broiler chickens fed with neem leaf powder had heavier weight gains than the birds without neem leaf powder in the diet. The improvement in weight gains might be due to anti-protozoal and immunostimulatory properties of neem leaves that help reduce the microbial load and improve broiler chickens’ performance (Wankar et al., 2009; Kharde and S. Soujanya, 2014).

In terms of voluntary feed intake, the data revealed that the inclusion of NLP significantly reduced (p<0.05) the feed intake of broiler chickens where birds fed without NLP (T1) has higher feed intake compared to broiler chickens with 4% NLP (T3) NLP got the highest feed intake compared to broiler chickens fed with 2%, 4%, and 6% NLP. This result is contrary to Landy et al. (2011), who reported that neem leaf powder at a rate of 7grams/kilo and 12 grams/kg to broiler diets at the age of 42 days had no significant effect on the feed intake of broiler chickens. However, the present study revealed that feed intake was significantly affected, where birds fed with NLP have lower feed intake than broiler chickens without NLP in the diet. Although birds fed with NLP have lower feed intakes, it was observed that the body weight gain of birds fed with 4% NLP (T3) is statistically heavier (p<0.05) compared to the birds with 0% NLP (T1) in the diet. This result may be due to the antimicrobial and anti-protozoal properties of neem leaf powder, which help reduce the microbial organisms of the birds as neem leaf might have suppressed the growth of harmful microorganisms. As a result, it creates a conducive environment for the active substances to aid digestion and improve production performance (Ketkar, 1976; Ezzat et al., 2018; Adeyemo and Akanmu, 2012; Kharde and Soujanya, 2014).

The cumulative feed conversion ratio (FCR) of broiler chickens fed with 4% NLP (T3) got the lowest (better) FCR compared to other treatments. It should be noted that FCR measures the efficiency of poultry and livestock in converting animal feed into the desired output. The FCR indicates that the lower the value, the more efficient the birds convert feed to live weight (Ampode et al., 2020). In this study, broiler chickens fed with 4% NLP have the lowest FCR of 1.52±0.08 than those fed with 0%, 2%, and 6% NLP. This result is similar to Kharde and Soujanya (2014), who reported that supplementation of NLP in broiler chicken diets significantly improved the feed conversion ratio. The higher body weight gains might be due to growth-promoting and antimicrobial properties of neem leaves that helped reduce birds’ microbial load and improved feed efficiency (Jong et al. 2009; Wankar et al. 2009).

**Cell-Mediated Immunity**

In avian species, adaptive immunity encompasses both humoral and cell-mediated immune responses (Erf, 2004). The humoral or antibody-mediated immune responses ef-
fectively combat the extracellular antigens. On the other hand, cell-mediated immunity is focused on eliminating intracellular antigens that have infiltrated cells, such as viral proteins and proteins originating from neoplastic cell transformation (Erf, 2004; Eladia and Ampode, 2021).

In the present study, the indicators of cell-mediated immunity, i.e., bursa and spleen indices, were not significantly affected (p>0.05) when NLP was incorporated in the diet. However, a significant difference (p<0.05) was observed in the spleen weight. The spleen weight of broiler chickens fed with 4% NLP (T₃) is higher but statistically comparable to the birds fed with 0% (T₁) and 2% (T₂) NLP and birds fed with 6% NLP (T₄) got the lowest spleen weight but statistically comparable to the birds fed with 2% (T₂) NLP. The spleen and bursa indices are indicators of cell-mediated immunity, which means that the higher the immunity index, the stronger the broiler chickens’ immune response (Fu Chang et al., 2004; Dumaup and Ampode, 2020).

Many studies reported that neem leaf powder plays a vital role in strengthening the immune system (Zahid et al., 2013, Al-Samarrai, 2012, Talpur and Ikhuwanuddin, 2013). However, the cell-mediated immunity of the current study fed with NLP was not significantly affected. Although not significant, it was observed that the body weight gains of broiler chickens fed with NLP are significantly higher than birds without NLP in the diet. This might be attributed to neem leaf powder in the diet, which increased humoral and cell-mediated immune responses (Sadekar et al. 1998). At the same time, it killed or slowed down the growth of many organisms such as bacteria, viruses, and fungus, which boost the production performance of broiler chickens (Sadekar et al., 1998).

Moreover, the findings of Jawad et al. (2013) reported that neem leaf powder had good immunomodulatory effects against Newcastle disease (ND) and infectious bursal diseases (IBD) as indicated by the serum antibody titers. Also, it showed higher mean antibody titer values against ND compared to the negative control group (Jawad et al., 2013). Hence, the variations of the findings might be due to the level of NLP incorporated in the diet, breeds or strain of the experimental animals, and the quality of the neem leaf powder used in the study.

CONCLUSION

The feed intake of broiler chickens fed with graded levels of NLP was significantly reduced, where birds without NLP in the diet got the highest feed intake. The body weight gain was significantly improved in broiler chickens fed with 4% NLP as compared to other treatments. Also, the feed conversion ratio of broiler chickens fed with 4% NLP showed better than birds fed with 0%, 2%, and 6% NLP. Moreover, no significant difference was observed in the cell-mediated immunity, and no mortality of birds was recorded. The return above feed and chick cost of broiler chickens fed with 4% NLP is more profitable, and the income generated increases as high as 25.73% compared to the birds fed without NLP in the diet. However, a future digestibility study using a large population of experimental animals is recommended to assess the nutrient flow and retention from the digestive sites. In conclusion, 4% NLP could be incorporated into the diets of broiler chickens without fear of compromising growth and immunity responses.

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NOVELTY STATEMENT

Several studies using phytogenic feed additives or supplements to broiler chickens were conducted. However, the information utilizing neem leaf powder as phytogenic feed additives to broiler chickens and investigating its potential on the production performance, economic traits, and cell-mediated immunity are scarce.

AUTHORS CONTRIBUTION

Both authors contributed equally to this work.

CONFLICT OF INTEREST

The authors declared no conflict of interest.
REFERENCES

• Adeyemo GO, Akanmu AM (2012). Effects of Neem (Azadirachta indica) and Pawpaw (Carica papaya) leaves supplementation on performance and carcass characteristics of broilers. Int. J. Curr. Res. 4(12): 268-271.

• Adjorolfo LK, Timbong-Jones E, Badau S, Adogla-Bessa T (2016). Potential contribution of Neem (Azadirachta indica) leaves to dry season feeding of ruminants in West Africa. Livest. Res. Rural. Dev. 28(5): 1-10

• Alkhalf A, Alhaj M, Al-homidan I (2010). Influence of probiotic supplementation on blood parameters and growth performance in broiler chickens. Saudi J. Biol. Sci. 17(3): 219-225. https://doi.org/10.1016/j.sjbs.2010.04.005

• Al-Samarrai G, Singh H, Syarhabil M (2012). Evaluating eco-friendly botanicals (natural plant extracts) as alternatives to synthetic fungicides. Ann. Agric. Environ. Med. 19:673–676.

• Ampode KM, Galgo SJ, Lapurga IG (2020). Pinto peanut Meal: It’s Potential as Dietary Supplement for Philippine Mallard Ducks. Int. J. Biosci., 16(5): 319-326. http://dx.doi.org/10.12692/ijb.16.5.319-326

• AOAC (2016). Official Methods of Analysis of the Association of the Official Analytical Chemists. 20th ed. Washington, D.C., USA.

• Arowayo A, Maigandi SA, Malami BS, Daneji AI (2011). Haematological and Biochemical Parameters of Uda Lambs Fed Graded Levels of Alkali-Treated Neem Kernel Cake. NJBAS., 19(2):277-284.

• Deore UB, Ingle RS, Waghmare SP, Pathak VP, Joshi MV (2005). Clinicopathological investigations in broilers given different levels of neem oil supplementation in feed. J. Bombay Vet. Coll., 13(1-2): 110-111.

• Dumaup HJ, Ampode KM (2020). Inclusion of Water Hyacinth Meal in Broiler Chicken Diets: Potential on the Production Performance and Cell-mediated Immunity. Int. J. Biosci., 17(6):469-479. http://dx.doi.org/10.12692/ijb.17.6.469-479.

• Eladia RE, Ampode KMB (2021). Moringa (Moringa oleifera Lam.) Pod meal: nutrient analysis and its effect on the growth performance and cell-mediated immunity of broiler chickens. J. Anim. Health Prod. 9(2): 170-177. http://dx.doi.org/10.17582/jahp.2019/9.2.170.177

• Erf GF (2004). Cell-mediated immunity in poultry. Poul. Sci., 83(4): 580–589. https://doi.org/10.1093/ps/83.4.580.

• Ezzat HN, Abood SS, Jawad HS (2018). A review on the effects of neem (Azadirachta indica) as feed additive in poultry production. J. Entomol. Zool. Stud., 6(1): 1331–1333

• Fu-Chang L, Qiu-Xia L, Xiuling Z (2004). Comparative studies on growth performance, nutrient digestibility, immunity index and protease activities between weaning-2 month and 2-3 month New Zealand rabbits. In Proceeding: World Rabbit Congress, Puebla, Mexico, 885-890.

• Galang VM (2019). Poultry industry maintains 2019 growth target. Business World. Accessed: https://tinyurl.com/4m3x4y8nu

• Hossain ME, Kim GM, Lee SK, Yang CJ (2012). Growth Performance, Meat Yield, Oxidative Stability, and Fatty Acid Composition of Meat from Broilers Fed Diets Supplemented with a Medicinal Plant and Probiotics. Asian-Aust. J. Anim. Sci. 25(8): 1159 – 1168. https://doi.org/10.5715/ajas.2012.12090

• Jawad Z, Younus M, Mutti-ur R, Maqbool A, Munir R, Muhammad K, Korejo RA, Qazi IH (2013). Effect of neem leaves (Azadirachta indica) on immunity of commercial broilers against new castle disease and infectious bursal disease. Afr. J. Agric. Res., 8(36): 4596-4603. https://doi.org/10.5897/AJAR2013.7852

• Kafi A, Uddin MN, Uddin MJ, Khan MMH, Haque ME (2017). Effect of Dietary Supplementation of Turmeric (Curcuma longa), Ginger (Zingiber officinale) and their Combination as Feed Additives on Feed Intake, Growth Performance and Economics of Broiler. Int. J. Poult. Sci., 16: 257-265. https://doi.org/10.3923/ijps.2017.257.265

• Ketkar CM (1976). Utilization of Neem (Azadirachta indica Juss.) and its by-products. Final Tech. Rep. Directorate of Non-Edible Oils and Soap Industry, Khadi and Village Industries Commission, Hyderabad, India.

• Kharde KR, Soujanya S (2014). Effect of garlic and neem leaf powder supplementation on growth performance and carcass traits in broilers. Vet. World. 7(10): 799-802. https://doi.org/10.14020/vetworld.2014.799-802

• Lagua EB, Ampode KMB (2021). Turmeric Powder: Potential alternative to antibiotics in broiler chicken diets. J. Anim. Health Prod. 9(3): 243-253. http://dx.doi.org/10.17582/journal.jahp/2021/9.3.243.253

• Landy N, Ghalamkari GH, Tooghany M (2011). Performance, carcass characteristics, and immunity in broiler chickens fed dietary neem (Azadirachta indica) as alternative for an antibiotic growth promoter. Livest. Sci., 142(1):305-309. https://doi.org/10.1016/j.livsci.2011.08.017

• Latif IK, Majed HM, Sahar H (2014). Determine the weight of thymus, bursa of Fabricius and spleen and its ratio to body weight in some diseases of broilers. MRVSA., 3(1): 8-14.

• Maass N, Bauer J, Paulicks BR, Bohmer BM, Roth-Maier DA (2005). Efficiency of Echinacea purpurea on performance and immune status in pigs. J. Anim. Physiol. Anim. Nutr. 89:244–252. https://doi.org/10.1111/j.1439-0396.2005.00501.x

• Philippine Council for Agriculture, Aquatic and Natural Resources Research and Development (PCAARRD). (2000). The Philippines Recommends for Livestock Feed Formulation. (64): 36-81

• Philippine National Standard (PNS 103), 2016. Code of Halal Slaughtering Practices for Poultry. Bureau of Agriculture and Fisheries Standards, pp 1-27

• PSA (2019). Philippine Statistics Authority. Chicken Situation Report. PSA.gov.ph- Chicken Inventory: Accessed: https://tinyurl.com/46cajj44

• PNS/BAFPS. 2008. Code of Good Animal Husbandry Practices. Philippine National Standard/Bureau of Agriculture and Fisheries Product Standards 60, 1-14.

• Roth-Maier DA, Bohmer BM, Maass N, Damme K, Paulicks BR (2005). Efficiency of Echinacea purpurea on performance of broilers and layers. Arch. Geflügelk. 69:123–127

• Sadekar RD, Kolte AV, Barmase BS, Desvi VF (1998). Immunopotentiating effects of Azadirachta indica (Neem) dry leaves powder in broilers, naturally infected with IB virus. Indian. J. Exp. Biol. 36(11):1151-3.

• Shihab IM, Al-Zuhariy MTB, Absallam SM, Mutar SS (2017). Impact of supplementation Neem powder (Azadirachta indica) to diet broiler in immunological, physiological and productive traits. Adv. Environ. Biol., 11(3): 44-51.

• Shahaan VNS, Wahyuni TH, Dalay AH, Lubis SR (2021). Utilization of Golden Snail Flour 548 (GSF) on Ration of Quail. Jurnal Peternakan Integratif. 8(3): 195–202

• Sun Jong, Y., Byoung KA, Chang Won K (2009). Effects of dietary garlic powder on growth performance and mRNA
expression of hepatic HMG-CoA reductase in broiler chickens. J. Anim. Sci. Technol., 51(4): 307-314.

• Tapur AD, Ikhwanuddin M (2013) Azadirachta indica (neem) leaf dietary effects on the immunity response and disease resistance of Asian seabass, Lates calcarifer challenged with Vibrio harveyi. Fish Shellfish Immunol., 34(1): 254-264. http://dx.doi.org/10.1016/j.fsi.2012.11.003

• Tiwary MK, Pandey A (2010). Feeding neem (Azadirachta indica) products to small ruminants as anthelmintics. Food Sci Tech Lett., 1(1): 10.

• Ubua JA, Ozung PO, Inagu PG (2019). Dietary Inclusion of Neem (Azadirachta indica) Leaf Meal Can Influence Growth Performance and Carcass Characteristics of Broiler Chickens. Asian J. Biol. Sci., 12: 180-186. http://dx.doi.org/10.3923/ajbs.2019.180.186

• Upadhyay SN, Dhawan S, Garg S, Talwar GP (1992). Immunomodulatory effects of neem (Azadirachta indica) oil. Int. J. Immunopharmacol. 14(7):1187–93. doi: 10.1016/0192-0561(92)90054-o.

• Uko OJ, Kamalu TN (2008). Trend of food consumption and efficiency of broiler production with raw or heat-treated neem kernels. Archivos de Zootecnia. 57 (220): 489–496

• Wankar AK, Shirbhate RN, Bahiram KB, Dhenge SA, Jasutkar RA (2009). Effect of neem leaf powder supplementation on growth in broilers. Vet. World., 2(10): 396–397.

• Windisch W, Schedle K, Plitzner C, Kroismayr A (2007). Use of phytogenic products as feed additives for swine and poultry. J. Anim. Sci. (86) 140-148. http://dx.doi.org/10.2527/jas.2007-0459

• Zacaria A, Ampode KM (2021). Turmeric (Curcuma longa Linn.) as Phytogenic Dietary Supplements for the Production Performance and Egg Quality Traits of Laying Japanese Quail. J. Anim. Health Prod. In press.

• Zahid J, Younus M, Rehman M, Maqbool A, Munir R, Muhammad K, Korejo RA, Qazi IH (2013). Effect of neem leaves (Azadirachta indica) on immunity of commercial broilers against Newcastle disease and infectious bursal disease. Afr. J. Agric. Res. 8(37): 4596–4603.