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Impacts of Supplementation of Herbal Blend and Cinnamon Powder on Laying Performance and Egg Quality in Laying Hens

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ABSTRACT

Herbal plants are increasingly used in animal production as natural alternatives to enhance digestive health, improve animal performance and product quality, and reduce reliance on antibiotics. The present study aimed to assess the effects of a specific herbal mixture powder (HMP), composed of *Bidens pilosa* (80%), cinnamon (15%), and anise (5%), at different inclusion levels and to directly compare its impact with that of cinnamon powder (CP) on laying hen performance, egg quality, and egg nutritional content. This comparative study randomly assigned 360 laying hens to five dietary treatments with three replicates of 24 hens per group, and fed the experimental diets for 35 weeks. The treatment groups included the control group, which received the basal diet without supplementation (CG), basal diets supplemented with HMP at 1.0% (TG1), basal diets supplemented with HMP at 1.5% (TG2), basal diets supplemented with HMP at 2.0% (TG3), and a basal diet supplemented with 0.5% of CP (TG4). The present results demonstrated that dietary supplementation with 1.5% or 2.0% of HMP significantly improved laying rate and egg productivity, as indicated by reduced feed consumption per 10 eggs, without significantly affecting egg weight, yolk weight, yolk percentage, albumen weight, or albumen percentage compared to the control group. Additionally, HMP inclusion enhanced yolk pigmentation, eggshell weight and percentage, shell thickness, and Haugh units, although egg yolk crude protein content remained unchanged across treatment groups. The TG4 indicated improvements in certain egg quality traits, including higher haugh units and lower yolk fat and cholesterol levels than the control group. Supplementing with 1.5% and 2% of HMP notably decreased egg yolk cholesterol and fat levels compared to the control group, suggesting potential health benefits for consumers seeking lower-cholesterol eggs. The present study demonstrated that dietary supplementation with a multi-herbal powder mixture, particularly at 1.5 and 2.0%, significantly enhanced laying rate, egg production, and feed efficiency in laying hens. Additionally, it improved egg quality, including yolk color, eggshell thickness, and haugh units, and reduced egg yolk fat and cholesterol, providing a natural strategy for producing healthier eggs while potentially reducing reliance on synthetic additives.

Keywords: Cholesterol, Egg quality, Herbal supplementation, Laying hen

INTRODUCTION

Laying hens are frequently affected by a variety of infectious diseases, including pasteurellosis, paratyphoid, chronic respiratory disease, coccidiosis, infectious bronchitis, and infectious bursal disease (Serbessa et al., 2023). However, the widespread and inappropriate use of antimicrobials, including incorrect dosages and failure to observe withdrawal periods, has resulted in antibiotic residues accumulating in eggs and other poultry products, posing significant public health risks. Antibiotic residues in food may contribute to the development and dissemination of antimicrobial-resistant bacteria, a major global health threat (WHO, 2023). Several studies have documented the presence of antibiotic residues in commercial eggs, raising concerns regarding food safety (Yamaguchi et al., 2017; Diem et al., 2019). These concerns have accelerated interest in herbal feed additives, which are increasingly used as sustainable alternatives to antibiotics and natural growth promoters in poultry production worldwide, including in Vietnam (Windisch et al., 2008; Kikusato, 2021).

Phytogenic feed additives derived from specific medicinal plants such as cinnamon (*Cinnamomum* spp.), *Bidens pilosa (B. pilosa)*, and star anise (*Illicium verum*) have gained attention as dietary strategies to support gut health and productive performance in poultry. Cinnamon has been extensively studied in chickens and laying hens, with reported benefits on feed efficiency, serum lipid profile, and intestinal functions. These functions have been associated with cinnamon's antimicrobial and antioxidant properties, as well as its ability to enhance digestive activity and nutrient utilization (Al-Kassie, 2009; Shirzadegan, 2014; Ali et al., 2021).

Bidens pilosa has received comparatively limited attention in poultry nutrition; however, available studies in chickens indicated that dietary supplementation can improve growth performance and gut health, as reflected by enhanced intestinal morphology and modulation of gut microbiota (Chang et al., 2016; Memon et al., 2021). These effects are consistent with the high concentration of bioactive compounds in *B. pilosa*, including polyphenols, flavonoids, polysaccharides, and triterpenoids, which are linked to antioxidant activity and the preservation of intestinal integrity (Le et al., 2022).

Star anise (*Illicium verum*), rich in essential oil dominated by trans-anethole, has been evaluated as a phytogenic feed additive in poultry. Dietary supplementation of star anise powder or its essential oil significantly improves growth performance and enhances serum and liver antioxidant status in broiler chickens, as reflected by increased activities of antioxidant enzymes and reduced lipid peroxidation, which are attributed to bioactive compounds such as anethole, estragole, and anisaldehyde (Ding et al., 2017).

Comparative information on the effects of a defined herbal mixture containing cinnamon, *B. pilosa*, and star anise in laying hen diets remains limited. Therefore, the present study aimed to assess the effects of different inclusion levels of a herbal mixture powder (HMP) containing *B. pilosa*, cinnamon, and anise, compared with the effects of the cinnamon powder (CP), on laying performance, egg quality, and nutritional characteristics in laying hens.

MATERIALS AND METHODS

Ethical approval

The present study was conducted in accordance with the guidelines of the Council of the Faculty of Animal Science, Vietnam National University of Agriculture (Number: FAS/03/2024). Additionally, the chickens used in the experiment were raised under conventional poultry-farming conditions in Vietnam in accordance with the Vietnamese Livestock Law (No. 32/2018/QH14), and all experimental procedures were designed to minimize unnecessary suffering.

Animals, diets, and experimental design

The current study was conducted from April 2024 to December 2024 in Hai Duong Province, in the Red River Delta of Northern Vietnam. The study region had a sub-tropical climate with four distinct seasons (spring, summer, autumn, and winter). The average annual temperature was approximately 25.9°C, with the highest temperature recorded in June (30.9°C) and the lowest in January (18.4°C). The average annual relative humidity was 75.2%, ranging from 63% in December to 83% in February (NSO, 2025).

A total of 360 crossbred Egyptian laying hens, 30 weeks old with an average body weight of 1,700 g per chicken, were sourced from a local hatchery in Hai Duong, Vietnam. The feeding trial was carried out from 30 to 65 weeks of age. All chickens were vaccinated according to the routine immunization schedule. Vaccination against Marek's disease was administered at first day of age, infectious laryngotracheitis at 7 days of age, infectious bursal disease (Gumboro) at 28 days of age, and Newcastle disease at 45 days of age. These vaccines were supplied by Agriviet Veterinary Pharmaceutical Joint Stock Company, Vietnam. Vaccination against avian influenza (Amavet Veterinary Trading Joint Stock Company, Vietnam) was administered at 21 days of age. Vaccination against Newcastle disease (Agriviet Veterinary Pharmaceutical Joint Stock Company, Vietnam) was repeated every two months, and against avian influenza every six months.

All chickens were of the same breed and age and were raised under identical management conditions in ventilated, closed poultry houses equipped with fans and cooling systems, with ambient temperature and relative humidity not artificially controlled. A uniform lighting program of 16 hours of light per day at 8 lux was applied to all groups. Four hens were kept in each cage, measuring 40 × 65 × 38 cm (length × width × height). The cages were elevated 60 cm off the ground, and each had its own access door. The present study employed a comparative batch design. The chickens were randomly assigned to five treatment groups, with each group having three replicates of 24 hens (18 adjacent cages per replicate, with four hens per cage).

The HMP was formulated to contain 80% *B. pilosa* powder, 15% cinnamon powder, and 5% star anise powder. The aerial parts of *B. pilosa*, cinnamon bark, and star anise fruits were purchased from local herbal markets in Hai Duong province, Vietnam, then ground separately through a 2-mm sieve and thoroughly mixed to obtain a homogeneous herbal mixture, which was then incorporated into the experimental diets. The experiment consisted of five dietary treatment groups. The control group was fed the basal diet without any supplementation (CG). Treatment group 1 received the basal diet supplemented with 1.0% of HMP (TG1), treatment group 2 received the basal diet supplemented with 1.5% of HMP (TG2), and treatment group 3 received the basal diet supplemented with 2.0% of HMP (TG3), and treatment group 4 (TG4) was fed the basal diet supplemented with 0.5% of CP, on a dry matter basis. The supplementation period lasted 35 weeks. Drinking water was provided *ad libitum* through nipple drinkers, and each hen received 110 g of the experimental diet daily, divided into two meals (06:00-07:00 and 13:00-14:00). The basal diet was formulated to meet the nutritional requirements for laying hens according to the Vietnamese National Standard (TCVN 2265:2020; VSQI, 2020). The basal diet and nutritional composition are provided in Tables 1 and 2.

Table 1. Basal diet of laying hens at the age of 30-65 weeks

Ingredients	Proportion (%)
Corn	56.23
Soybean meal	22.00
Meat and bone meal	2.90
Limestone powder	9.80
Rice bran	7.00
Premix*	0.28
Dicalcium phosphate (DCP)	0.64
DL - Methionine	0.25
L-Threonine	0.10
Salt powder (NaCl)	0.27
Natri bicarbonate ($NaHCO_3$)	0.15
Toxisorb	0.20
Choline Chloride	0.15
Phytase 5000	0.01
Antioxidant	0.02
Total	100

*Premix: 1000 g of this premix include Vitamin A 3,200,000 IU, Vitamin D3 1,000,000 IU, Vitamin E 6,000 mg, Vitamin K3 800 mg, Vitamin B1 600 mg, Vitamin B2 1,600 mg, Vitamin B6 1,200 mg, Vitamin B12 6 mg, Biotin 40 mg, Folic Acid 200 mg, Niacin 8,000 mg, Pantothenic Acid 3,200 mg, Iron (Fe) 16,200 mg, Copper 2,000 mg, Manganese 24,000 mg, Zinc 24,000 mg, Co 120 mg, Se 120 mg. Toxisorb is composed of 100% aluminosilicate, with major components, including SiO_2, Al_2O_3, and Fe_2O_3.

Sampling and analysis

Laying performance

From 30 to 65 weeks of age, the number of hens, egg production, and feed intake were recorded daily and summarized weekly to evaluate laying performance. Egg productivity was expressed as the number of eggs produced per hen per week and calculated by dividing the total weekly egg production for each treatment group by the group's average number of hens. Laying rate (%) was determined as the ratio of weekly egg production to the number of hens present. Feed consumption per 10 eggs (kg) was calculated as total feed intake in 1 week divided by the total number of eggs in one week, then multiplied by 10. Feed cost per 10 eggs was calculated by multiplying feed consumption per 10 eggs by the feed unit price, following the methods described by Bui et al. (2011). These measurements allowed a comprehensive assessment of production efficiency and feed utilization across the experimental groups.

Egg quality

At weeks 0, 4, and 8 of the experiment, 30 eggs per group were collected to evaluate egg quality parameters, including egg weight, yolk color, yolk weight, yolk proportion, albumen weight, albumen proportion, eggshell weight, eggshell proportion, eggshell thickness, and haugh units. Haugh units were calculated using the logarithmic formula described by Bui et al. (2011).

$$\text{Haugh units} = 100 \times \log (H - 1.7\, W^{0.37} + 7.57)$$

where H is the albumen height (mm), and W is the egg weight (g).

Additionally, at weeks 0, 4, and 8 of the experimental periods, six eggs per treatment group were collected to determine yolk chemical composition. Crude protein content was analyzed using the Kjeldahl method in accordance with ISO 20483:2006 (ISO, 2006), while total lipid content was determined following AOAC official method 925.32 (AOAC, 2002a). Cholesterol content in egg yolk was measured by gas chromatography with flame ionization detection (GC-FID) according to AOAC official method 994.10 (AOAC, 2002b). These analyses provided detailed information on the nutritional composition of eggs and allowed evaluation of potential effects of dietary treatments on egg quality and consumer-relevant health parameters.

Statistical analysis

Statistical analyses were performed using SAS 9.1 software. The Shapiro-Wilk test was used to verify the normality of the data. Differences among groups were evaluated by one-way ANOVA, and Duncan's multiple range test was applied to assess the significance of mean differences. A significance level of p-value less than 5% ($p < 0.05$) was used for all comparisons. Data were presented as mean ± standard error (SE).

RESULTS

Laying performance

The inclusion of 1.0%, 1.5%, and 2.0% HMP in the diet significantly improved laying rate, egg productivity, and feed consumption for producing 10 eggs compared to the CG ($p < 0.05$), although the feed cost increased (Table 3). Notably, TG2 did not produce significant differences in laying rate (86.6%) or egg productivity (6.06 eggs/hen/week) compared to TG3 ($p > 0.05$). Furthermore, the feed cost per 10 eggs in TG2 (12,837 Vietnamese dong) was significantly lower than that in TG3 (13,162 Vietnamese dong; $p < 0.05$). The TG1 did not significantly alter the laying rate, egg productivity in hens, feed consumption per 10 eggs, or feed cost compared to the CP group ($p > 0.05$).

Egg quality

Dietary supplementation with HMP or CP did not significantly affect the egg weight, yolk weight, and percentage, or albumen weight and percentage ($p > 0.05$; Table 4). However, hens in TG2 showed significantly improved egg quality compared with the CG, as evidenced by greater eggshell weight (8.8 versus 8.3 g), increased shell thickness (34.6 versus 32.8 µm), and higher Haugh units (84.2 versus 80.7; $p < 0.05$). Similar findings were observed in TG3, which also showed a significantly higher yolk color than in the CG ($p < 0.05$). The Haugh units were significantly higher in TG4 than in CG ($p < 0.05$).

Egg chemical parameters

The chemical composition of the sampled eggs is presented in Table 5. Hens receiving diets supplemented with HMP or CP demonstrated significantly lower egg yolk cholesterol concentrations compared to the CG ($p < 0.05$). Specifically, TG3 had the lowest egg yolk cholesterol concentration, measured at 883 mg/100 g after eight weeks of feeding. Additionally, yolk fat content was significantly reduced in both the HMP and CP groups compared to the CG ($p < 0.05$). However, the supplementation of HMP and CP did not significantly affect the crude protein content in the egg yolk ($p > 0.05$).

Table 2. Nutritive composition of experimental diets

Composition \ Dietary treatment	CG	TG1	TG2	TG3	TG4	SEM
Crude protein (%)	16.2	16.2	16.2	16.1	16.1	0.05
Lipid (%)	3.5	3.5	3.5	3.4	3.4	0.08
Crude fiber (%)	4.0	4.1	4.1	4.2	4.1	0.03
Calcium (%)	4.0	4.1	4.1	4.1	4.1	0.04
Phosphorus (%)	0.5	0.5	0.5	0.5	0.50	0.02
Metabolizable energy (kcal kg^{-1})	2,801.7	2,808.2	2,809.3	2,802.2	2,804.3	47.26

CG: Control group (0.0% herbal mixture powder [HMP] or cinnamon powder), TG1: Treatment group with1.0% HMP; TG2: Treatment group with 1.5% HMP; TG3: Treatment group with 2.0% HMP; TG4: Treatment group with 0.5% cinnamon powder. SEM: Standard error of mean

Table 3. Effects of herbal mixture powder and cinnamon powder on laying performance and feed cost of crossbred Egyptian laying hens from 30 to 65 weeks of age

Items \ Dietary treatment	CG	TG1	TG2	TG3	TG4	SEM
Laying rate (%)	84.40^{c}	85.60^{b}	86.60^{ab}	87.70^{a}	85.50^{bc}	0.555
Egg production (egg/hen/week)	5.91^{c}	6.00^{b}	6.06^{ab}	6.14^{a}	5.99^{bc}	0.038
Feed for 10 eggs (kg)	1.30^{a}	1.29^{ab}	1.27^{b}	1.26^{c}	1.29^{ab}	0.008
Feed cost for 10 eggs (Vietnam Dong-VND)	11.708^{d}	12.512^{c}	12.837^{b}	13.162^{a}	11.825^{d}	83.56

^{a,b,c,d} Means in the same row not sharing a common superscript letter, differ significantly at $p < 0.05$. CG: Control group (0.0% herbal mixture powder [HMP] or cinnamon powder), TG1: Treatment group with1.0% HMP, TG2: Treatment group with 1.5% HMP, TG3: Treatment group with 2.0% HMP, TG4: Treatment group with 0.5% cinnamon powder. SEM: Standard error of mean

Table 4. The effects of dietary supplementation of herbal mixture powder and cinnamon powder on egg quality of crossbred Egyptian laying hens

Items \ Dietary treatment	CG	TG1	TG2	TG3	TG4	SEM
Egg weight (g)	65.9	66.7	66.9	67.3	66.3	0.56
Egg yolk color	13.0[b]	13.1[b]	13.3[ab]	13.6[a]	13.2[ab]	0.14
Egg yolk weight (g)	18.0	18.2	18.3	18.5	18.2	0.25
Egg yolk rate (%)	27.3	27.2	27.4	27.6	27.4	0.33
Albumen weight (g)	39.6	40.0	39.8	39.6	39.7	0.49
Albumen rate (%)	60.1	60.0	59.3	58.8	59.9	0.43
Eggshell weight (g)	8.3[b]	8.5[b]	8.8[a]	9.1[a]	8.4[b]	0.14
Eggshell rate (%)	12.6[b]	12.8[b]	13.2[a]	13.6[a]	12.7[b]	0.22
Eggshell thickness (mm)	32.8[b]	32.9[b]	34.6[a]	35.2[a]	34.0[ab]	0.57
Haugh units (Hu)	80.7[c]	82.2[b]	84.2[a]	85.2[a]	82.2[b]	0.74

[a,b,c] Means in the same row not sharing a common superscript letter differ significantly at $p < 0.05$. CG: Control group (0.0% herbal mixture powder [HMP] or cinnamon powder), TG1: Treatment group with1.0% HMP, TG2: Treatment group with 1.5% HMP, TG3: Treatment group with 2.0% HMP, TG4: Treatment group with 0.5% cinnamon powder. SEM: Standard error of mean

Table 5. The effects of dietary supplementation of herbal mixture powder and cinnamon powder on chemical parameters of egg yolk

Items	Dietary treatment	CG	TG1	TG2	TG3	TG4	SEM
Crude protein (g/100g)	0 week	16.3	16.2	16.4	16.4	16.4	0.13
	4 weeks	16.5	16.1	16.3	16.5	16.3	0.68
	8 weeks	16.2	16.5	16.3	16.4	16.4	0.17
Fat (g/100g)	0 week	28.3	28.8	28.6	28,9	28.5	0.23
	4 weeks	28.3[a]	27.8[ab]	27.5[ab]	27.3[b]	27.6[ab]	0.30
	8 weeks	28.3[a]	27.4[b]	27.0[bc]	26.6[c]	27.0[bc]	0.20
Cholesterol (mg/100g)	0 week	1053	1052[I]	1084[I]	1082[I]	1074[I]	23.9
	4 weeks	1054	1025[I]	1006[II]	968[II]	989[II]	31.8
	8 weeks	1054[a]	934[bII]	931[bII]	883[bIII]	940[bII]	26.5

[a,b,c] Means in the same row not sharing a common superscript letter differ significantly at $p < 0.05$. [I; II; III]: Means in the same column not sharing a common superscript differ significantly at $p < 0.05$. CG: Control group (0.0% herbal mixture powder [HMP] or cinnamon powder), TG1: Treatment group with1.0% HMP, TG2: Treatment group with 1.5% HMP, TG3: Treatment group with 2.0% HMP, TG4: Treatment group with 0.5% cinnamon powder. SEM: Standard error of mean

DISCUSSION

Laying performance

Dietary supplementation with the herbal mixture composed of *B. pilosa* L., *Cinnamomum verum*, and *Illicium verum* at inclusion levels of 1.5% or 2.0% considerably improved laying rate and egg productivity, while reducing feed conversion ratio (feed intake per 10 eggs). The present results indicated that this plant-based formulation positively influenced feed efficiency and overall laying performance. Similar findings have been reported in previous studies using phytogenic feed additives in laying hens. Abo Ghanima et al. (2020) reported that dietary supplementation with 0.3 mg rosemary essential oil or 300 mg cinnamon essential oil improved feed utilization efficiency and egg production in ISA laying hens. Similarly, Gerzilov et al. (2015) observed that supplementation with a herbal mixture containing garlic, cinnamon, yarrow, rosemary, thyme, basil, and oregano improved feed conversion ratio by 3.37% and egg production by 1.79%. These findings support the present results and suggest that phytogenic feed additives may enhance laying performance by improving digestive efficiency and nutrient utilization.

The current findings highlighted the potential of HMP at 1.5% and 2.0% to enhance egg production and improve feed efficiency. The improved laying performance and feed utilization observed in the present study might be attributed to bioactive components of the herbal additives, including cinnamaldehyde in cinnamon, polyphenols in *B. pilosa*, and anethole in star anise, which have been associated with enhanced digestive efficiency, intestinal function, and metabolic stability in poultry (Al-Kassie, 2009; Chang et al., 2016; Ding et al., 2017).

Egg quality

Dietary supplementation with 1.5% or 2% of HMP notably enhanced egg quality parameters, including yolk color, eggshell weight, eggshell thickness, eggshell percentage, and Haugh units. Yolk color is a key consumer-preference trait influenced by dietary pigments, with evidence suggesting that 20-60% of dietary pigment compounds are transferred to the yolk (Li et al., 2016). Thickened eggshells improve structural integrity, reducing breakage during handling, transportation, and storage (Ketta and Tůmová, 2016). Superior albumen quality was achieved in the HMP-supplemented groups, as evidenced by Haugh unit values above 72 that qualify the eggs as AA quality according to USDA standards. These improvements might be associated with herbal polysaccharides and antioxidant compounds, including flavonoids, tannins, and saponins, which inhibit protein oxidation and maintain albumen integrity (Yu et al., 2023).

Egg lipid and cholesterol content

All herbal-supplemented groups exhibited substantial reductions in egg yolk fat and cholesterol levels. Cinnamon, in particular, has been reported to inhibit lipid peroxidation, reduce hepatic lipogenesis, and lower HMG-CoA reductase activity, thereby decreasing cholesterol deposition in eggs (Sharma et al., 2009; Shirzadegan et al., 2014). The current findings are consistent with previous studies indicating that dietary supplementation with phytogenic feed additives can reduce egg yolk cholesterol in laying hens, which has been attributed to flavonoids and phenolic compounds that inhibit intestinal cholesterol absorption and biosynthesis (Li et al., 2016; Dilawar et al., 2021). Producing low-cholesterol eggs is relevant for human health and meets growing consumer demand for functional foods.

CONCLUSION

Dietary supplementation with 1.5% or 2.0% of HMP improved laying rate and egg productivity, reduced feed consumption per 10 eggs, and enhanced key egg quality parameters, including yolk color, eggshell weight, eggshell thickness, and Haugh units. Moreover, HMP supplementation effectively reduced egg yolk lipid and cholesterol content, demonstrating its potential for producing healthier eggs that meet consumer demands. Future studies should explore the effects of *B. pilosa*, *Cinnamomum verum*, and *Illicium verum* in essential oil form and identify the specific bioactive compounds responsible for the observed improvements in egg quality and cholesterol reduction.

DECLARATIONS

Authors' contributions

Huong Quynh Vu authored the original text, contributed to the experimental design, and carried out the experiments. Duy Van Nguyen contributed to the experimental design, conducted the experiments, and performed the statistical analysis. Ton Dinh Vu contributed to the design of the experiment, reviewed and approved the final edition, and analysed data for publication. All authors have read and approved the final edition of the manuscript before publication.

Competing interests

The authors declared no conflicts of interest.

Ethical considerations

Ethical issues, including plagiarism, consent to publish, misconduct, data fabrication and/or falsification, double publication and/or submission, and redundancy, have been checked by all the authors. The authors did not use any AI tools for writing or revising the text of this article, nor for any stage of preparing the present study or analyzing the data.

Availability of data and materials

The data obtained in the present study are available upon reasonable request from the corresponding author.

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