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Effects of Supplemental Humic Substances on Egg Production and Quality in Laying Hens

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Humic Substances의 급여가 산란계의 산란율과 난 특성에 미치는 영향

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ABSTRACT The effects of dietary humic substances (HS) on egg production and egg quality were studied using 252 (55-wk old) ISA brown laying hens. They were divided into 21 groups of 12 hens each and seven groups (experimental units) were assigned to 1) CON (basal diet), 2) HS5 (basal diet + 5% humic substances) or 3) HS10 (basal diet +10% humic substances) in a completely randomized block design. Hens had free access to diets and water for 6 wk. Egg production and egg quality were monitored over the 6-wk period. Results showed that 10% dietary HS decreased egg production and yolk diameter ($P<0.05$) compared to CON. Egg weight and yolk color were improved ($P<0.05$) in HS10 compared to CON. Egg shell breaking strength was increased significantly ($P<0.05$) when hens were fed HS5 diet compared to the others. There were no effects of treatments on egg shell thickness, yolk index, albumen height and Haugh unit. The results suggest that the dietary supplementation of HS at 5% or 10% decreases egg production, but HS at 5% can increase egg shell breaking strength. Hens fed 10% HS could increase egg weight and yolk color and decrease yolk diameter.

(Key word: humic substances, egg production, egg characteristics)

Introduction

Humic substances (HS) are defined as a series of relatively high-molecular-weight, yellow to black colored substances formed by secondary synthesis reaction (Stevenson, 1994). HS have many beneficial effects, such as antibacterial, antiviral and anti-inflammatory activity (Islam et al., 2005). During the past several years, humic substances have been used in poultry feed as an alternative additive to antibiotics (Onifade et al., 1999).

Remarkable changes in electrolyte balance and enhancements in immune potency of poultry in response to HS supplementation have been reported (Parks et al., 1986). Moreover, published articles showed that HS promote growth by altering partitioning of nutrient metabolism and reducing ammonia emission in poultry (Bailey et al., 1996; Parks, 1998; Shermer et al., 1998). To our knowledge, there are a limited number of articles dealing with dietary HS in laying hens. Therefore, the present study is to investigate the effect of HS supplementation on egg production and egg quality in laying hens.

Materials and Methods

1. Humic Substances

Humic substances make up a large portion of the dark matter in humus and are complex colloidal supramolecular mixtures hat have never been separated into pure components. HS consist of trace minerals (including iron, manganese, zinc and calcium), organic acids (including fulvic and humic acids) and others organic compounds.

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2. Animals and Diets

A total of 252 ISA brown laying hens (55-wk old) were blocked according to the cage location in a temperature and humidity-controlled room with auto-controlled light cycle of 14L: 10D. They were divided into 21 groups of 12 hens each and seven groups (experimental units) were assigned to 1) CON (basal diet), 2) HS5 (basal diet + 5% humic substances) or 3) HS10 (basal diet +10% humic substances) in a completely randomized block design. Hens had free access to diets and water for 6 wk and egg production and egg quality were monitored over the 6-wk period. The basal (control) diet (Table 1) was formulated to meet or exceed the nutrient requirements of laying hens recommended by the NRC (1994).

The diets of the first, second and third treatment were supplemented with 0 g/100 g, 5g/100 g and 10g/100 g humic substances, respectively. Humic substances were supplemented by replacing the same amount of wheat bran. Humic substances contained 6% moisture. 40% calcium humate, 10% fulvic acid, 18% trace minerals, 10% SiO_2 and 16% kaolin.

3. Sampling and Measurements

Eggs were collected daily and egg production was calculated on a hen-day basis. Eggs were individually weighed once a week. To determine the egg quality, 30 eggs were collected randomly from each treatment on saturdays, and all eggs except soft and broken eggs, were collected to measure eggshell breaking strength. Eggshell breaking strength (kg/cm^2) was measured using Compression Test Cell in Texture Test Systems (Model T2100C, Food Technology Corp., Rockville, MD). Shell thickness (mm) was determined in 3 different parts (upper end, lower end and middle) by using micrometer. Yolk height and albumen height were measured using vernier caliper. Albumen, egg shell and shell membranes were removed from each egg and yolk color was determined by using yolk color fan (Roche, Switzerland) according to the CIE standard colorimetric system. Yolk index (%) = (yolk height, mm/yolk diameter, mm)×100, and Haugh unit = 100×log (H +7.57−1.7 W0.37) were also calculated.

4. Statistical Analysis

The data were subjected to analysis using a GLM procedure of SAS (SAS Institute, 1996) for the completely randomized experimental design. Differences between means were determined by Duncan's multiple range test (1955) at significance level of $P<0.05$.

Table 1. Diet composition (as-fed basis)

Ingreditents(%)	CON	HS5	HS10
Corn	50.36	50.36	45.36
Soybean meal	18.70	18.70	18.70
Wheat grain	10.00	10.00	10.00
Limestone	7.50	7.50	7.50
Wheat bran	10.00	5.00	–
Animal fat	4.44	4.44	4.44
Corn gluten meal	2.00	2.00	2.00
Tricalcium phosphate	1.40	1.40	1.40
Salt	0.30	0.30	0.30
DL-methionine	0.10	0.10	0.10
Mineral premix[1]	0.10	0.10	0.10
Vitamin premix[2]	0.10	0.10	0.10
Humic substance	–	5	10
Chemical composition[3]			
ME (kcal/kg)	2,904	2,904	2,904
Crude protein (%)	15.45	15.45	15.45
Lysine (%)	0.70	0.76	0.85
Methionine (%)	0.32	0.38	0.41
Calcium (%)	3.23	3.87	4.35
Phosphorus (%)	0.61	0.61	0.61
Available P (%)	0.35	0.35	0.35

[1] Provided per kg of premix: 25,000 mg Cu, 40,000 mg Fe, 60,000 mg Zn , 80,000 mg Mn, 1,500 mg I, 300 mg Co and 150 mg Se.

[2] Provided per kg of premix: 12,500,000 IU vitamin A, 2,500,000 IU vitamin D_3, 10,000 mg vitamin E, 2,000 mg vitamin K_3, 50 mg biotin, 500 mg folic acid, 35,000 mg niacin, 10,000 mg Ca pantothenate, 1,000 mg vitamin B_6, 5,000 mg vitamin B_2, 1,000 mg vitamin, B_1 and 15 mg vitamin B_{12}.

[3] Calculated values.

Results and Discussion

Table 2 shows the effect of dietary HS content on productivity of laying hens. Hens fed HS10 diet had lower (P<0.05) egg production than hens fed control or HS5 diets. Egg weight was higher in HS10 group than that in the CON or HS5 groups, showing no difference between the control or HS 5.

Table 3 shows the effect of diet on egg quality indicators (eggshell strength, eggshell thickness, yolk color, yolk height, yolk diameter, yolk index, albumen height and Haugh unit). Eggshell breaking strength was significantly increased (P<0.05) when diets supplemented with HS at level of 5%. Yolk color unit was significantlty increased (P<0.05) with the addition of 10% HS. However, yolk diameter was significantly reduced (P<0.05) in HS10 compared to the others.

In this study, we observed that HS had negative effect on egg production. These findings contradict the results of previous studies (Kucukersan et al., 2005; Yalcin et al., 2006), possibly because of differences such as feeding duration and dietary HS concentrations. When response to HS in egg production was observed, hens were fed HS concentrations for at least 0.1%, and at most 2% (Kucukersan et al., 2005; Yalcin et al., 2006). In our experiment, We began feeding in our experimental diets within 5% vs 10% HS, respectively. Kim et al. (2004) indicated that minerals in humic substances may affect animal performance.

Table 2. The effects of supplementation of humic substances on egg production of laying hens

Traits	CON[1]	HS5[1]	HS10[1]	SE[2]
Egg production				
0 week	84.78	83.79	82.96	1.74
6 week	79.18[a]	73.91[ab]	68.84[b]	2.03
Egg weight (g)				
0 week	65.19	65.17	65.92	0.79
6 week	63.55[b]	63.98[b]	67.46[a]	0.57

[1] Abbreviations CON, basal diet; HS5, basal diet + 5% humic substances; HS10, basal diet + 10% humic substances.

[2] Pooled standard error.

[a,b] Means in the same row not sharing the same superscripts differs(P<0.05).

Adequate mineral concentrations and balance between minerals are also important items. Thus, excessive amounts of one mineral may interfere with utilization of one or more

Table 3. The effects of supplementation of humic substances on egg production of laying hens

Traits	CON[1]	HS5[1]	HS10[1]	SE[2]
Eggshell breaking (kg/cm^2)				
0 week	3.61	3.71	3.77	0.12
6 week	3.09[b]	3.43[a]	2.84[b]	0.08
Eggshell thickness (mm)				
0 week	0.32	0.32	0.33	0.01
6 week	0.30	0.31	0.30	0.01
Yolk color unit				
0 week	7.54	7.68	7.55	0.12
6 week	7.25[b]	7.31[ab]	7.45[a]	0.05
Egg yolk index[3]				
0 week	0.47	0.46	0.47	0.01
6 week	0.44	0.45	0.45	0.01
Yolk diameter(cm)				
0 week	3.76	3.80	3.75	0.03
6 week	3.96[a]	3.98[a]	3.91[b]	0.02
Yolk height(cm)				
0 week	1.76	1.73	1.74	0.02
6 week	1.75	1.77	1.75	0.01
Albumen height(cm)				
0 week	0.84	0.83	0.82	0.03
6 week	0.83	0.82	0.80	0.02
Haugh uint[4]				
0 week	89.30	89.46	89.64	1.43
6 week	89.77	88.50	87.70	1.65

[1] Abbreviations CON, basal diet; HS5, basal diet + 5% humic substances; HS10, basal diet +10% humic substances.

[2] Pooled standard error.

[a,b] Means in the same row not sharing the same superscripts differs(P<0.05).

[3] Yolk index=yolk height/yolk diameter.

[4] Haugh unit = 100×log (H+7.57−1.7 W0.37).

other elements (Church, 1991). Supplementary calcium generally needed in many animal diets because of the demands for most minerals for skeletal growth, lactation, or egg production (Keshavarz and Nakajima, 1993). As the recommendation of the NRC (1994) for commercial laying hens, calcium level was 3.25 g/h/d in our experimental diet. However, calcium concentration was still high in HS additive. With added HS 10% to diet, egg production decreased significantly. Harms and Waldroup (1971) indicated that excess dietary calcium may cause reduced egg production and extra feed consumption. More severe toxicity can occur when excess calcium is ingested over long periods. Very high levels of calcium can result in appetite loss, nausea, vomiting, abdominal pain, and even coma (Isbister, 2003). Egg weight was increased with diets added 10% HS. This response in egg weight due to various HS intake levels is consistent with other reported research (Kucukersan et al., 2005; Yalcin et al., 2006). Further investigation is needed to directly evaluate the effect of different HS contents on laying hens.

The eggshell consists of about 94 to 97% calcium carbonate. Calcium which is major macro-mineral involved shell formation is increased with shell breaking. However, the hens have the ability to absorb from the digestive system and to mobilize Ca from the medullar bones (Keshavarz and Nakajima, 1993). Humic acid inhibits the growth of pathogenic bacteria and moulds, thus it decreases the levels of mycotoxins. They also improve protein digestibility and calcium utilization (Islam et al., 2005).

Yolk color is dependent on the diet of the hen; if the diet contains yellow/orange plant pigments known as xanthophylls, then they are deposited in the yolk, coloring it. This result is not consistent with the published research of Yoruk et al. (2004) who reported that addition of humate had no effect on yolk color. At the moment, It is not clear if the HS was influenced the yolk color. Possible mechanisms have not yet been identified. Further investigation is needed to substantiate this observation. The results suggests that the dietary supplementation of HS at 5% or 10% decreases egg production, but HS at 5% can increase egg shell breaking strength. HS at 10% can increased egg weight and yolk color and decrease yolk diameter. Further research with laying hens is needed to clarify the mechanism of HS at different dietary levels.

적 요

본 연구는 humic substances의 급여가 산란계의 생산성과 난 특성에 미치는 영향을 알아보고자 실시하였다. 55주령 ISA brown 갈색계 252수를 공시하였고, 6주간 시험을 실시하였다. 처리구는 1) CON (basal diet), 2) HS5 (basal diet added humic substances 5%), and 3) HS10 (basal diet added humic sudstances 10%)으로 처리구당 7반복 반복당 12수씩 완전 임의 배치하였다. 전 시험 기간 동안 humic substances을 10% 첨가한 처리구는 난 생산성과 난황 직경이 대조구와 비교하여 유의적으로 감소하였다(P<0.05). 난황색은 HS10 처리구가 CON 처리구와 비교하여 높게 나타났다(P<0.05). 개시시 난중은 처리구간에 차이가 없었다(P<0.05). 난각 강도에서는 HS5 처리구가 다른 처리구들과 비교하여 유의적으로 높게 나타났다(P<0.05). 결론적으로, 산란계 사료내 HS 5% 또는 10%의 첨가는 난황색 및 난각 강도를 향상시킬 수 있을 것으로 사료된다.

(색인어: Humic substances, 산란율, 난 특성, 산란계)

References

Bailey CA, White KE, Donke LS 1996 Evaluation of Menefee Humate™ on the performance of broilers. Poult Sci 75 (Suppl.1):84(Abstr).

Church DC 1991 Livestock Feeds and Feeding. 3rd ed Prentice Hall Englewood Cliffs New Jersey.

Duncan DB 1955 Multiple range and multiple F test. Biometrics 11:1.

Harms RH, Waldroup PW 1971 The effect of high dietary calcium on theperformance of laying hens. Poult Sci 50: 967-969.

Islam KMS, Schuhmacher A, Gropp JM 2005 Humic acid substances in animal agriculture. Pakistan J Nutr 4:126-134.

Isbister GK 2002 Delayed asystolic cardiac arrest after diltiazem overdose; resuscitation with high dose intravenous calcium. The Emergency Medicine Journal 19:355-357.

Kim SW, Hulbert LE, Rachuonyo HA, Mcglone JJ 2004 Relative availability of iron in mined humic substance for weaning pigs. Asian-Aust J Anim Sci 17:1266-1270.

Kucukersan S, Kucukersan K, Colpan I, Goncuoglu E, Reisli

Z, Yesilbag D 2005 The effects of humic acid on egg production and egg traits of laying hen. Veterinary Medicine Czech 50:406-410.

Keshavarz K, Nakajima S 1993 Re-evaluation of calcium and phosphorus requirements of laying hens for optimum performance and eggshell quality. Poult Sci 72:144-153.

NRC 1994 Nutrient Requirements for Poultry. (9th Rev ed) National Academy Press Washington DC.

Onifade AA, Babatunde GM, Odunsi AA, Olorede BR, Muna E 1999 Comparison of the supplemental effects of *Saccharomyces cerevisiae* and antibiotic in low-protein and high-fiber diets fed to broiler chickens. Arch Tierernahr 52: 29-32.

Parks CW 1998 The use of Menefee Humate™ in typical and low-crude protein diets for turkey and in the bioremediation of petroleum contaminated soil amend with poultry litter as a co-substrate and nutrient source. Master Thesis North Carolina State University Raleigh NC.

Parks C, Ferket PR, Thomas LN, Grimes JL 1986 Growth performance and immunity of turkeys fed high and low crude protein diets supplemented with menefee humate. Poult Sci 75:138-143.

SAS 1996 SAS User's Guide.Version 8.02. SAS Institute. Inc Cary. NC. USA.

Stevenson FJ 1994 Humic Chemistry Genesis Composition Reaction 2nd ed John Wiley and Sons Wiley : 480-512.

Shermer CL, Maciorowski KG, Bailey CA, Byers FM, Ricke S 1998 Caecal metabolites and microbial populations in chickens consuming diets containing a mined humate compound. J Sci Food Agri 77:479-486.

Yalcin S, Ergun A, Ozsoy B, Yalcin S, Erol H, Onbasilar I 2006 The effects of dietary supplementation of L-carnitine and humic substances on performance, egg traits and blood parameters in laying hens. Asian-Aust J Anim Sci 19: 1478-1483.

Yoruk MA, Gul M, Hayirli A, Macit M 2004 The effects of supplementation of humate and probiotic on egg production and quality parameters during the late laying period in hens. Poult Sci 83:84-88.