International Journal of Veterinary Science and Agriculture Research
Volume 5 Issue 6, November-December 2023
ISSN: 2582-4112, Available at www.ijvsar.com

Excretion of crude protein by turkeys after dietary intake of humic substances

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Keywords: *dry matter, humic substances, internal marker, nitrogen, poultry nutrition.*

Abstract: *The study aimed to examine the effects of the inclusion of humic substances (HS) to feed mixtures (FM) for turkeys (Hybrid Converter) on excretion of crude protein (CP) in faeces. The control and experimental groups of birds (n=40), aged 6 weeks, were provided with FM: a grower FM (CP 238.8 $g.kg^{-1}$) till 12 weeks, and a finisher FM (CP 195.7 $g.kg^{-1}$) till day 70 of the experiment. A supplement of 5 $g.kg^{-1}$ of HS (minimum of 650 $g.kg^{-1}$ humic acids and minimum of 50 $g.kg^{-1}$ fulvic acids in dry matter (DM)) was added to feed mixtures of experimental group. The excreta from both groups were sampled on days 27, 30, 34, 42 and 57. The chemical analyses of DM, crude protein (CP), ash, and ash insoluble in HCl were performed. The ratio of CP to ash insoluble in HCl was calculated. The results were assessed by the Student-Newman-Keuls test. In the case of ratio of CP excretion to ash insoluble in HCl, higher values were observed in the control group in all sampling periods with statistical differences on days 34 and 42 ($p<0.05$). The addition of HS into feed has a beneficial environmental impact on the reduction of CP excretion.*

I. INTRODUCTION

The production of broiler poultry is an important part of agriculture with the 105.26 million tons of meat products for the world market in 2023 [1]. The demand for poultry meat products has increased due to the increased demand of consumers to purchase meat with a low cholesterol content, which has resulted in an increase in the production of broiler poultry [2].

The fattening poultry excrete dust into the environment, which is created by peeling off the surface cells of the epithelium, from feed, skin and excrement. Therefore, dust pollution in objects is of organic and inorganic origin. Other pollution agents are microorganisms, namely non-pathogenic and pathogenic, NH_3, CH_4, N_2O, CO_2 and H_2O. The mentioned factors act on birds mechanically, immunosuppressively, allergenically or infectiously. Their effects can negatively affect the temperature and humidity of the air. The welfare effects of air pollutants such as ammonia have been observed at the behavioural and physiological levels. To reduce the produced NH3 emissions, it is necessary to know the principles of their formation. Nitrogen and nitrogenous substances are excreted in birds in the form of uric acid.

A large amount of ammonia is produced in the process of microbial mineralization of uric acid [3]. Ammonia causes respiratory diseases [3]. They are converted to ammonia by the activity of the enzyme urease, which is excreted in animal feces, within a few days. Under these circumstances, nitrogen is in the form of ammonium cation (NH_4^+) at acidic or neutral pH or in the form of ammonia (NH_3) at higher pH. It is possible to influence the concentration of ammonia in the environment of broiler poultry farms by changing the feed from the point of view of protein and amino acid content.

Humic substances (HS) are complex and heterogeneous mixtures of polydispersed materials that are formed by biochemical and chemical reactions during the decomposition and transformation of plant and microbial residues in the process of humification. The basic components of this process are plant lignin, its transformation products, polysaccharides, melanin, cutin, proteins, lipids, nucleic acids and fine coal particles [4]. Humic and fulvonic acids are two major components of HS that have similar functional structures and their general characteristics are similar to humus [5]. Fulvonic acids consist of many active functional groups: phenolic, hydroxyl, carboxylic and hydroquinone [6]. Gao et al. [7] found that fulvic acids can increase the proportion of beneficial bacteria in the gut, increase the secretion of digestive enzymes including lysozyme, protease, and acid-alkaline phosphatase, and improve intestinal digestion.

The aim of the experimental study was to evaluate the effects of dietary HS intake on nitrogen excretion measured as the ratio of nitrogenous substances to ash insoluble in HCl analyzed in the excreta of fattening turkeys.

II. Material and Methods

The experimental study was authorized by the Ethics Committee of the University of Veterinary Medicine and Pharmacy in Košice. 20 turkeys (Hybrid Converter, average weight 1726.8 g) from the age of 6 weeks were supplied from a commercial farm (Nádudvar, Hungary). The birds were randomly divided into 2 groups of 10 individuals each (control Co and experimental Exp) and marked with identification rings. Housing was carried out in 2 pens in an experimental poultry breeding hall with constant access to feed and water. The pens had an identical orientation. Two commercial feed mixtures (FM) (De Heus, JSCo, Czech Republic) were used for feeding during the experiment: a/ turkey midi forte (TMF, crude protein (CP) 238.8 $g.kg^{-1}$) from 1st day of the experiment up to 12 weeks of age, b/ turkey maxi (TM, CP 195.7 $g.kg^{-1}$) from 13 weeks of age to the 70th day of the experiment). The feed mixtures did not contain antibiotics and growth stimulators. FM contained the anticoccidial and histomonostatic sodium monensinate 70.0 $mg.kg^{-1}$.

The feed of experimental chickens was supplemented with products containing humic substances (HS) (Humac Ltd., Slovak Republic). Humac MycotoxiSorb was added to the feed of experimental group B in the amount of 5 $g.kg^{-1}$. The mentioned additive had the following parameters: particle size up to 200 µm, pH 5.8, maximum moisture 21%, humic min. 650 and fulvonic acids min. 50 $g.kg^{-1}$ in dry matter (DM). Food and water intake during the entire experiment was ad libitum. Turkey weights and feed consumption were measured during the experimental period at two-week intervals. Excrements were collected from both groups on a clean plastic pad for the analysis of DM, CP, ash and ash insoluble in HCl on day 27, 30, 34, 42 and 57.

Experimental diets and excrements were analyzed (Table 1) according to the methods of the Association of Official Analytical Chemists [8]. Determination of DM, CP, crude fat, starch and ash in both feed mixtures was carried out. Neutral detergent fiber was analyzed by the method of Van Soest et al. [9]. Atomic absorption spectrophotometry (AAS) was used to analyze the mineral composition of feeds [10] Phosphorus was quantified spectrophotometrically [11]. Determination of the proportion of ash insoluble in HCl in the feed mixture was carried out according to Daněk et al. [12]. The value of metabolizable energy (ME) was calculated by the formula according to Commission Regulation (EC) no. 152/2009 [13].

Data are expressed as mean ± standard deviation (SD) of individual values. The mean values of the results were compared by the Student-Newman-Keuls test.

Table 1 Analyzed content of nutrients in experimental diets

Analyzed nutrients ($g.kg^{-1}$)	Feed mixture	
	Turkey midi forte (TMF)	Turkey maxi (TM)
Dry matter	1000.0	1000.0
Crude protein	238.80	194.90
Crude fat	39.50	53.60
NDF [1]	146.60	167.40
ADF [2]	77.80	72.70
Starch	472.10	518.70
NSC [3]	592.50	643.0
Ash	78.50	62.40
Ca	11.40	12.30
P	6.90	6.70
Na	1,80	1.80
Cu	0.023	0.026
Zn	0.143	0.117
ME [4] ($MJ.kg^{-1}$)	13.67	14.10

[1-] neutral detergent fibre, [2-] acid detergent fibre, [3-] nonstructural carbohydrates, [4-] Metabolic energy (European Commission (2009)

III. Results and Discussion

The values of the CP excretion parameter, expressed as the ratio of excreted CP to ash insoluble in HCl, are shown in Table 2. A positive effect of HS intake on this parameter was observed in the experimental group of turkeys at all samplings periods (day 27, 30, 34, 42, 57) with statistically significant differences on the 34th and 42nd day of the experiment. A positive effect of HS intake on reduction of the excretion of CP in the experimental group of turkeys was observed.

According to Tang et al. [14], it is possible to regulate nitrogen metabolism by the treatment with fulvic acids, which are components of HS, and increase the utilization of nitrogenous compounds in the intestine of poultry and thus reduce the elimination of ammonia through excreta. Ammonia is produced in the poultry intestine by the deamination of amino acids and the hydrolysis of urea.

If the ammonia content of the intestine is reduced, then the compensatory effect of ammonia on the intestinal cells is alleviated, resulting in an improvement of the intestinal barrier and intestinal histomorphology [15].

Decrease of ammonia was observed in the experiments Ji et al. [16] as well, when the dietary addition of HS for pigs decreased emission by 3 to 18%.

HS were shown to inhibit urease activity. Therefore, the direct application of HS to manure also reduced beef feedlot ammonia emissions [17].

IV. Conclusion

Dietary intake of HS at an application dose of 5 $g.kg^{-1}$ reduced the excretion of nitrogenous substances measured as the ratio of CP excretion to ash insoluble in HCl in the case of all samplings. It points to the potentially positive impact of these additives on the environment.

Table 2 Content of dry matter, nitrogenous substances, ash and ash insoluble in HCl analyzed in excrements of turkeys after intake of humic substances (mean ± SD, n = 8)

Sampling (day)	Dry matter $[g.kg^{-1}]$		CP $[g.kg^{-1}]$		Ash $[g.kg^{-1}]$		Ash insoluble in HCl $[g.kg^{-1}]$		CP / Ash insoluble in HCl	
	Co	Exp	Co	Exp	Co	Exp	Co	Exp	Co	Exp
27	266.95 ^{a} ± 34.53	340.26 ^{a} ± 17.22	5.92 ^{a} ± 3.30	8.58 ^{a} ± 4.15	45.96 ^{a} ± 10.17	68.16 ^{b} ± 9.28	1.88 ^{a} ± 1.86	2.50 ^{a} ± 0.80	7.70 ^{a} ± 1.71	3.72 ^{a} ± 2.34
30	237.84 ^{a} ± 27.08	259.20 ^{a} ± 18.03	4.16 ^{a} ± 0.49	4.81 ^{a} ± 2.40	32.47 ^{a} ± 13.08	43.89 ^{b} ± 7.15	1.11 ^{a} ± 0.60	2.05 ^{b} ± 0.29	5.79 ^{a} ± 4.92	2.47 ^{a} ± 1.53
34	302.71 ^{a} ± 16.89	284.48 ^{a} ± 24.50	18.14 ^{a} ± 13.21	6.39 ^{b} ± 3.92	49.99 ^{a} ± 1.69	45.06 ^{b} ± 4.35	1.85 ^{a} ± 0.27	2.09 ^{a} ± 0.41	10.22 ^{a} ± 8.23	3.21 ^{b} ± 2.26
42	206.54 ^{a} ± 13.07	234.16 ^{b} ± 14.82	4.04 ^{a} ± 0.45	4.85 ^{b} ± 1.14	28.31 ^{a} ± 6.80	38.02 ^{b} ± 1.35	1.15 ^{a} ± 0.25	1.89 ^{b} ± 0.36	4.87 ^{a} ± 0.81	2.68 ^{b} ± 0.99
57	210.22 ^{a} ± 13.99	205.47 ^{a} ± 16.66	3.25 ^{a} ± 1.23	2.92 ^{a} ± 0.73	23.45 ^{a} ± 5.17	28.75 ^{b} ± 1.70	1.00 ^{a} ± 0.50	1.19 ^{a} ± 0.07	4.48 ^{a} ± 3.44	2.46 ^{a} ± 0.62

Means with different indexes are significantly different (^{a,b} P < 0,05)

Acknowledgements

The experimental study and its evaluation were financed by the grants VEGA-1/0402/20 - The influence of additives in the nutrition of monogastric animals on production health, production, product quality and the environment as well as KEGA-006ZUVLF-4/2022.

References

[1] P. Maharjan, D.A. Martinez, J. Weil, C. Suesuttajit, C. Umberson, G. Mullenix, K.M. Hilton, K.M., A. Beitia, C.N. Coon, Physiological growth trend of current meat broilers and dietary protein and energy management approaches for sustainable broiler production, *Animal, 15(Suppl 1),* 2021, 100284.

[2] I. Dinev, D. Kanakov, I. Kalkanov, S. Nikolov, S. Denev, Comparative pathomorphologic studies on the incidence of fractures associated with leg skeletal pathology in commercial broiler chickens, *Avian Diseases*, *63*, 2019, 641-650.

[3] S.E. Bauer, K. Tsigaridis, R. Miller, Significant atmospheric aerosol pollution caused by world food cultivation, *Geophysical Research Letters*, *43*, 2016, 5394-5400.

[4] O. Bezuglova, Molecular structure of humus acids in soils, *Journal of Plant Nutrition and Soil Science, 182*, 2019, 676-682.

[5] A.Z. Muhammad, A. Sonita, W. Deana, W. Mia, Preparation of Fe3O4-chitosan hybrid nanoparticles used for humic acid adsorption, *Environmental Nanotechnology, Monitoring & Management*, *6*, 2016, 64-75.

[6] G.R. Aiken, D.M. McKnight, R.L. Wershaw, P. MacCarthy, Humic substances in soil, sediment, and water, *Soil Science*, *142*, 1986, 323.

[7] Y. Gao, J. He, Z. He, Z. Li, B. Zhao, Z. Chu, Effects of fulvic acid on growth performance and intestinal health of juvenile loach Paramisgurnus dabryanus (Sauvage), *Fish & Shellfish Immunology, 62*, 2017, 47-56.

[8] P. Cuniff, Official Methods of Analysis of AOAC. 16th edn., in *Association of Official Analytical Chemists* (Ed.), (Arlington: AOAC, 1995).

[9] P.J. Van Soest, J.B. Robertson, B.A. Lewis, Methods for dietary fibre, neutral detergent fibre and nonstarch polysaccharides in relation to animal nutrition, *Journal of Dairy Science, 74(10)*, 1991, 3583-3597.

[10] J.C. Van Loon, *Analytical Atomic Absorption Spectroscopy: Selected Methods*, in Academic Press (Ed.), 1st edition, (New York, 1980) 348 pages.

[11] L.H.M. Carvalho, T. De Koe, P.B. Tavares, An improved molybdenum blue method for simultaneous determination of inorganic phosphate and arsenate, *Ecotoxicology and Environmental Restoration*, *1*, 1998, 13-19.

[12] P. Daněk, A. Paseka, J. Smola, J. Ondráček, R. Bečková, M. Rozkot, Influence of lecithin emulsifier on the utilisation of nutrients and growth of piglets after weaning, *Czech Journal of Animal Science, 50(10)*, 2005, 459-465.

[13] European Commission, Commission Regulation (EC) No 152/2009 of 37 January 2009 laying down the methods of sampling and analysis for the official control of feed, *Official Journal of European Union, 54*, 2009, 1-130.

[14] C. Tang, W. Kong, H. Wang, H. Liu, L. Shi, V.A. Uyanga, J. Zhao, X. Wang, H. Lin, H. Jiao, Effects of fulvic acids on gut barrier, microbial composition, fecal ammonia emission, and growth performance in broiler chickens, *Journal of Applied Poultry Research, 32*, 2023, 100322.

[15] H. Ichikawa, T. Sakata, Stimulation of epithelial cell proliferation of isolated distal colon of rats by continuous colonic infusion of ammonia or short-chain fatty acids is nonadditive, *Journal of Nutrition, 128*, 1998, 843-847.

[16] F. Ji., J.J. McGlone, S.W. Kim, Effects of dietary humic substances on pig growth performance, carcass characteristics, and ammonia emissions, *Journal of Animal Science, 84*, 2006, 2482-2490.

[17] Y. Shi, D.B. Parker, N.A. Cole, B.W.Auvermann, J.E. Mehlhorn, Surface amendments to minimize ammonia emissions from beef cattle feedlots, *Transactions of the American Society of Agricultural Engineers*, *44*, 2001, 677-382.