doi: 10.17221/8441-VETMED

Levels of nitrogenous substances and amino acids in bodies of Ross 308 hybrid cocks and hens over the course of rearing

E. Strakova, P. Suchy, P. Navratil, I. Herzig, M. Machacek

University of Veterinary and Pharmaceutical Sciences, Brno, Czech Republic

ABSTRACT: Over the course of Ross 308 hybrid broiler chick cock and hen rearing, whole chicken bodies including feathers were monitored for changes in the levels of nitrogenous substances, essential amino acids (EAA) and non-essential amino acids (NEAA). At five-day intervals (Days 1, 5, 9, 15, 20, 26, 30, 35 and 40 of rearing), randomly selected chickens were slaughtered after 24 h of fasting. Over the course of rearing, N-substance levels ranged from 629.1 to 429.0 g/kg dry matter in hen bodies and from 616.0 to 477.3 g/kg dry matter in cock bodies. N-substance levels were statistically significantly different in the two sexes on rearing Days 35 and 40 ($P \leq 0.01$). The levels of all amino acids (AA) in dry body matter of chickens up to 15 days of age dropped significantly over time, while in the period from Day 20 to Day 40, some AA levels increased or reached the baseline. With most AA, the levels were lower in hens than in cocks over the course of the trial. At the end of the trial (Days 35 and 40), statistically significant differences between the sexes were found for most EAA ($P \leq 0.01$; $P \leq 0.05$). NEAA, except for Ser and Ala, were also significantly higher in cocks on Days 35 and 40 ($P \leq 0.01$; $P \leq 0.05$). Among the monitored EAA, the dry matter of the bodies of hens exhibited the highest levels of Leu, 8.70% on average, followed by Val 6.54%, Lys 5.26%, Ile 5.25%, Thr 4.84%, Phe 4.30%, Tyr 2.51% and Met 2.21%. Leu was also the most abundant in the cock bodies, 8.42% on average, followed by Val 6.30%, Lys 5.36%, Ile 5.06%, Thr 4.57%, Phe 4.45%, Tyr 2.88% and Met 2.17%. Knowledge of the levels of nitrogenous substances, EAA and NEAA in the whole bodies of broiler chickens including feathers will help to determine optimal rearing conditions.

Keywords: broiler; nitrogenous substance; amino acid; essential; non-essential

Knowledge of the amino acid (AA) levels in the body and feathers of broiler chickens is useful for determining suitable rearing conditions (Saunders et al. 1977; Stilborn et al. 1997). AA levels in the body are relatively constant; however, minor differences have been described, such as between the sexes and between chickens of different ages. According to Baker (1997), the need for AA is affected by a number of factors, such as protein levels, energy levels and the presence of protease inhibitors; environmental factors, such as crowding density, space around the feeders, heat and cold; changes to the state of health; genetic factors such as sex, and inclination towards leanness or fattiness of the meat.

There are numerous data in the literature about the need for N-substances and AA in the feed (such as NRC 1994; Dozier et al. 2008). Poultry need 22 AA for body protein development, part of which may be synthesised by the birds (non-essential amino acids, NEAA), while essential amino acids (EAA) cannot be synthesised in the required amounts (Applegate and Angel 2008). Of the EAA, lysine (Lys) and threonine (Thr) cannot be produced by the animals at all, since they do not have the transaminases needed for their synthesis. Essential AA for the organism also include those amino acids that, although synthesised by the body, are not produced in sufficient amounts. These include tryptophan (Try), histidine (His),

Supported by the Ministry of Education, Youth and Sports of the Czech Republic (Grant No. MSM 6215712402).

doi: 10.17221/8441-VETMED

phenylalanine (Phe), leucine (Leu), isoleucine (Ile), methionine (Met), valine (Val) and arginine (Arg). Their synthesis is more a theoretical rather than a practical problem, since the fodder does not include the required keto-acids needed for their synthesis. The fodder therefore must supply the full needs for all EAA (Zelenka et al. 2007).

Semi-essential amino acids may be synthesised in the organism but only from one of the essential amino acids, e.g. cysteine from methionine, tyrosine from phenylalanine. While the need for phenylalanine may only be met by phenylalanine, the need for tyrosine may be compensated for with either tyrosine or phenylalanine. When there is insufficient cysteine (Cys) in the fodder, the animal can create it from methionine, but methionine cannot be replaced with cysteine (Zelenka et al. 2007).

Individual NEAA can be developed from other non-essential or essential amino acids, but synthesis from essential AA is not usually biologically and economically beneficial. This for example applies to cysteine (Cys). The proportion between the nitrogen content of essential and non-essential amino acids in the feed mix should be approximately 1 : 1 (Zelenka et al. 2007).

Literature data concerning AA levels in chicken bodies and feathers are rare and heterogeneous (Stilborn et al. 1997; Stilborn et al. 2010), with no monitoring having been done with the broiler hybrids currently used in practice. The genetic potential of broiler chickens has increased considerably in recent years and this higher yield has also resulted in changes in their needs for amino acids (Salehifar et al. 2012). That is why this study, monitoring the progress of the rearing of Ross 308 hybrid chickens with regard to sex, was aimed at determining the changes in nitrogenous substances, EAA and NEAA levels in the whole bodies including feathers of the test animals.

MATERIAL AND METHODS

Birds. The trial included 300 one-day sexed Ross 308 hybrid broiler chickens in two groups, 150 hens and 150 cocks.

Experimental design and treatments. The trial site was the accredited test stable of the Animal Nutrition Institute of the Veterinary and Pharmaceutical University in Brno. The chickens were kept in a pen on deep litter (wood shavings and saw-dust). Over the course of the rearing, the population density complied with the requirement for the optimum load of the surface, i.e. 17 birds per 1 m^2. The lighting mode was preset to 23 h of light and 1 h of darkness. Microclimatic conditions including ventilation were controlled automatically. The temperature of the bedding at the commencement of the trial was 34 °C. This was reduced daily by 0.3–0.4 °C, reaching 18 °C at the end of the trial. The state of health of the chickens was monitored on an ongoing basis.

Diets. Over the course of the rearing, the chickens of both sexes received identical fodder mixes, on Days 1 to 9 starter (BR1), on Days 10 to 30 grower (BR2) and on Days 31 to 40 finisher (BR3) (Table 1). The basic nutrient content and gross energy of the complex fodder mix fed during the trial corresponded to the recommended nutrient needs of the given hybrid breed (Anonymous 2007). The feeding space corresponded to the requirements specified in the technological procedures for Ross 308 (Anonymous 2012). Feed and drinking water were offered *ad libitum.*

Sample collection and preparation. On the first day of life of the chickens and then on Days 5, 9, 15, 20, 26, 30, 35 and 40, randomly selected 24-hour fasted chickens (not receiving feed, with access to water) were slaughtered. From Day 1 to Day 15 of life the bird bodies were analysed as a mixed sample, while from Day 20 to Day 40 individual bodies were analysed (Table 3).

The whole chicken bodies including feathers were homogenised in a K 120 F High Speed Cutter® (PSS Svidník, Slovak Republic) which cuts, grinds and mixes the processed material in three steps. The homogenisation product was weighed, dried and the chicken whole-body dry matter was calculated. Before the chemical analysis, the dried sample was homogenised using the Ultra Centrifugal Mill ZM 200® (RETSCH, Germany).

Sample analyses. Complete fodder mixes were analysed in terms of their basic nutrient content pursuant to AOAC (2003). Basic nutrient content (g/kg) and gross energy (MJ/kg) in the original matter and in the dry matter of complex fodders are shown in Table 1 and corresponded to the recommended nutrient needs for the given hybrid breed (Anonymous 2007).

The dry matter content in the homogenised samples was determined by weighing after drying at 105 °C. Nitrogen content was determined by the

doi: 10.17221/8441-VETMED

Table 1. Basic nutrient levels (g/kg) and gross energy (MJ/kg) in complex fodder mixes fed over the course of the trial

	Starter (BR1)		Grower (BR2)		Finisher (BR3)	
Water	124.4		111.9		111.7	
Dry matter	875.6	1000.0	888.1	1000.0	888.3	1000.0
N-substances (N × 6.25)	217.1	247.9	199.3	224.4	182.4	205.3
Fat	42.7	48.8	63.7	71.7	76.4	86.0
Fibre	26.8	30.6	23.4	26.3	21.3	24.0
NFE[1]	538.9	615.5	553.7	623.5	561.9	632.6
Starch	396.0	452.3	415.8	468.2	421.1	474.1
Organic matter	825.5	942.8	840.1	946.0	842.0	947.9
Ash	50.1	57.2	48.0	54.0	46.3	52.1
Ca	8.4	9.6	8.0	9.0	8.5	9.6
P	6.0	6.9	6.0	6.8	5.1	5.7
Mg	1.7	1.9	1.9	2.1	1.9	2.1
Gross energy	16.6	19.0	17.1	19.3	17.3	19.5

The premix of specifically active substances used by the producer contained: vitamin A 1 600 000 IU; vitamin D3 500 000 IU; alpha-tocopherol 10 000 mg; vitamin K3 300 mg; vitamin B1 800 mg; vitamin B2 1300 mg; vitamin B6 600 mg; vitamin B12 3 mg; biotin 30 mg; folic acid 500 mg; niacinamide 6000 mg; calcium pantothenate 2500 mg; betaine 50 000 mg; butylhydroxytoluene 3400 mg; propyl gallate 1200 mg; ethoxyquin 540 mg; ferrous sulphate monohydrate 10 000 mg; manganese oxide 16 000 mg; zinc oxide 16 000 mg; copper sulphate 1700 mg; potassium iodide 200 mg; sodium selenite 30 mg; cobalt sulphate 50 mg; phytase 50 000 FTU; glucanase 24 000 BGU; xylanase 1 100 000 EXU

[1]NFE = nitrogen-free extractives

Kjeldahl method using a Buchi analyser (by Centec Automatika, spol. s.r.o., Prague, Czech Republic) and nitrogenous substance content (crude protein) was calculated by multiplying the nitrogen levels by a coefficient of 6.25.

Amino acid (AA) content was determined by acid hydrolysis in 6 N HCl at 110 °C for 24 h in an AAA 400 automatic amino acid analyser (by Ingos a.s., Prague, Czech Republic) on the basis of the color-generating reaction of the amino acids with an oxidization reagent – ninhydrin. Amino analysis was used to determine the pure protein level expressed by the sum of AA (Asp, Thr, Ser, Glu, Pro, Gly, Ala, Val, Met, Ile, Leu, Tyr, Phe, His, Lys and Arg). The amino acid content in starter (BR1), grower (BR2) and finisher (BR3) diets for broiler chickens expressed on a dry matter basis is presented in Table 2.

All presented results are recalculated to the chicken body dry matter (g/kg of dry matter) for more accurate reproducibility.

Statistical analysis. The results were processed using the statistical software Unistat CZ version 5.6 for Excel, where the mean values and their differences were assessed by multiple comparison with the help of the Tukey-HSD test, using 1% and 5% significance levels.

Table 2. The amino acid content in starter (BR1), grower (BR2) and finisher (BR3) diets for broiler chickens expressed on a dry matter basis (g/kg)

Amino acid	Starter (BR1)	Grower (BR2)	Finisher (BR3)
Asp	20.8	18.8	16.3
Thr	7.0	8.3	6.9
Ser	9.3	10.0	8.4
Glu	42.6	38.2	34.9
Pro	12.8	14.0	13.5
Gly	9.9	8.5	7.6
Ala	9.1	8.6	7.6
Val	10.9	9.5	8.6
Met	2.4	2.6	2.2
Ile	9.3	8.2	7.9
Leu	16.5	14.5	12.5
Tyr	6.9	6.3	5.6
Phe	10.9	9.6	9.2
His	6.3	5.1	4.6
Lys	13.4	12.2	10.9
Arg	15.9	15.0	13.3

doi: 10.17221/8441-VETMED

Table 3. Scheme of analysis of the bird bodies

Age (days)	Group 1 Ross 308 ♀	Group 2 Ross 308 ♂
1	30 birds, mixed sample	30 birds, mixed sample
5	20 birds, mixed sample	20 birds, mixed sample
9	10 birds, mixed sample	10 birds, mixed sample
15	5 birds, mixed sample	5 birds, mixed sample
20	10 birds, individual sample	10 birds, individual sample
26	10 birds, individual sample	10 birds, individual sample
30	10 birds, individual sample	10 birds, individual sample
35	10 birds, individual sample	10 birds, individual sample
40	10 birds, individual sample	10 birds, individual sample

Table 4. Comparison of live weight development in hens and cocks (kg)

Age (days)	Hens ♀	Cocks ♂
1	0.046	0.047
5	0.071	0.071
9	0.150	0.158
15	0.368	0.384
20	0.640	0.681
26	1.070	1.166
30	1.376	1.523
35	1.774	2.021
40	2.214	2.595

RESULTS

Dynamics of changes in live weight of broiler chickens over the course of rearing

Over the course of rearing, average values of chicken body live weights continually increased from 0.047 kg (Day 1) to 2.400 kg (Day 40); the average weight values are shown in Table 4. The chickens reached an average carcass weight of 2.4 kg at 40 days of age. Comparing the dynamics of growth in cocks and hens, it is evident that the cocks grew much faster, especially during the last third of the rearing period. From Day 9 of the rearing period, the dynamics of growth was more pronounced in cocks; their weight increased statistically more significantly than in hens ($P \leq 0.01$).

Dynamics of changes in dry matter content

Over the course of rearing, the composition of chicken whole-body dry matter underwent significant changes (Table 5). Following a moderate increase in levels between Days 1 and 5 of rearing, the dry matter levels then began to increase steadily. Over the course of the trial, dry matter levels were higher in hens than in cocks, with a statistically significant difference ($P \leq 0.01$) evident from Day 20 of rearing, i.e. in the period when the chickens were analysed individually.

Dynamics of changes in nitrogenous substance levels (crude protein)

N-substance levels in the dried chicken bodies were 629.1 g/kg dry matter in hens on Day 1 of the

Table 5. Dynamics of changes in composition of chicken dry body matter including feathers (g/kg)

Day of rearing	n	Dry matter	
		♀ ($\bar{x}$ ± SD)	♂ ($\bar{x}$ ± SD)
1	30	254.8	253.9
5	20	238.0	228.6
9	10	258.3	255.9
15	5	313.8	301.9
20	10	341.2 ± 5.92^{A}	331.3 ± 6.10^{B}
26	10	349.1 ± 3.85^{A}	330.2 ± 10.23^{B}
30	10	360.1 ± 12.34^{A}	341.6 ± 7.36^{B}
35	10	368.8 ± 11.89^{A}	348.5 ± 7.87^{B}
40	10	393.9 ± 13.12^{A}	359.2 ± 9.44^{B}

Different letters in the two columns denote statistical significance A : B ($P \leq 0.01$)

$\bar{x}$ = mean; SD = standard deviation

Table 6. Dynamics of changes in N-substance levels in chicken dry body matter (g/kg)

Day of rearing	n	N-substances in dry matter	
		♀ ($\bar{x}$ ± SD)	♂ ($\bar{x}$ ± SD)
1	30	629.1	616.0
5	20	632.8	630.8
9	10	571.8	563.5
15	5	495.2	511.8
20	10	480.6 ± 8.12	489.1 ± 20.23
26	10	480.6 ± 8.35	495.1 ± 22.45
30	10	484.6 ± 36.37	497.2 ± 18.67
35	10	455.1 ± 20.53^{A}	490.1 ± 23.29^{B}
40	10	429.0 ± 39.30^{A}	477.3 ± 13.78^{B}

Different letters in the two columns denote statistical significance A : B ($P \leq 0.01$)

$\bar{x}$ = mean; SD = standard deviation

doi: 10.17221/8441-VETMED

trial and 616.0 g/kg dry matter in cocks on the same day (Table 6). Up to 5 days of age, a slight increase was noticed in both sexes (632.8 and 630.8 g/kg dry matter, respectively) followed by a drop in the subsequent period from Day 9 to Day 20. This drop, with a slight deviation on Day 30, continued until the end of the monitored period. At the end of the rearing period, on Day 40, the values of N-substances in hens were in the range of 429.0 ± 39.30 g/kg dry matter, while in cocks they were in the range of 477.3 ± 13.78 g/kg dry matter, which in hens represented 67.8% and in cocks 75.7% of the values measured on Day 5. The differences between the N-substances in hens and in cocks were statistically significant on Days 35 and 40 ($P \leq 0.01$).

Table 7. EAA contents in broiler chicken bodies over the course of rearing (g/kg dry matter)

			Day of rearing								
			1	5	9	15	20	25	30	35	40
Lys	♀	$\bar{x}$	34.9	29.8	17.8	19.8	27.2	29.0^{A}	30.5	27.3^{A}	24.0
		± SD	–	–	–	–	3.083	2.653	2.182	2.520	4.015
	♂	$\bar{x}$	33.1	30.2	20.3	22.5	27.5	33.2^{B}	29.5	31.3^{B}	21.7
		± SD	–	–	–	–	1.885	1.709	3.427	1.839	3.119
Met	♀	$\bar{x}$	11.8	11.3	8.5	10.2	9.3	9.1	9.0	8.4^{a}	8.3^{a}
		± SD	–	–	–	–	0.932	0.910	1.279	0.944	0.944
	♂	$\bar{x}$	11.0	11.8	9.0	10.6	8.8	8.7	10.5	9.3^{b}	10.2^{b}
		± SD	–	–	–	–	0.684	1.337	2.362	0.986	1.926
Thr	♀	$\bar{x}$	24.3	26.5	22.8	18.8	17.7^{a}	18.4	18.7	18.8^{A}	17.1a
		± SD	–	–	–	–	1.271	0.905	1.529	0.960	1.425
	♂	$\bar{x}$	22.8	28.4	23.1	16.2	15.8^{b}	19.3	18.6	20.4^{B}	18.9b
		± SD	–	–	–	–	1.982	1.098	1.537	1.439	2.077
Leu	♀	$\bar{x}$	42.4	45.8	39.5	37.3	35.3^{a}	35.3	33.7	32.9^{a}	31.3^{A}
		± SD	–	–	–	–	3.323	1.348	2.637	2.686	2.735
	♂	$\bar{x}$	40.2	46.4	39.1	39.1	31.4^{b}	35.6	34.7	35.5^{b}	36.2^{B}
		± SD	–	–	–	–	2.715	1.757	2.943	1.785	1.747
Ile	♀	$\bar{x}$	25.5	26.9	24.8	22.3	21.3^{A}	20.8	20.2	19.7	18.8^{A}
		± SD	–	–	–	–	2.012	0.887	1.703	1.400	1.674
	♂	$\bar{x}$	23.2	27.6	23.5	24.5	18.4^{B}	21.2	20.4	20.8	21.9^{B}
		± SD	–	–	–	–	1.389	1.333	1.789	1.472	1.064
Val	♀	$\bar{x}$	32.6	34.9	30.6	26.5	25.0^{A}	24.8	23.6	23.9^{a}	22.3^{A}
		± SD	–	–	–	–	1.379	1.967	1.995	1.470	2.588
	♂	$\bar{x}$	30.7	35.9	29.7	27.5	22.2^{B}	25.1	24.1	25.4^{b}	26.3^{B}
		± SD	–	–	–	–	1.367	1.580	2.018	1.366	1.377
Tyr	♀	$\bar{x}$	18.8	18.1	5.4	8.0	11.0	11.3^{A}	14.1	14.6^{a}	13.3
		± SD	–	–	–	–	3.041	2.662	1.898	2.673	1.789
	♂	$\bar{x}$	18.5	19.7	10.9	8.9	12.3	15.1^{B}	15.6	16.8^{b}	12.9
		± SD	–	–	–	–	3.206	2.135	1.817	1.114	1.790
Phe	♀	$\bar{x}$	27.1	22.3	15.5	17.9	17.0^{A}	18.1	17.0	17.3	16.0^{A}
		± SD	–	–	–	–	1.169	1.233	1.365	1.080	1.577
	♂	$\bar{x}$	26.4	23.6	17.6	19.8	15.0^{B}	18.7	17.7	18.4	18.8^{B}
		± SD	–	–	–	–	1.272	1.254	1.299	1.259	1.850

Tryptophan and cysteine were destroyed in the acid hydrolysis

Different letters in corresponding rows denote statistical significance A : B ($P \leq 0.01$); a : b ($P \leq 0.05$)

$\bar{x}$ = mean; SD = standard deviation

doi: 10.17221/8441-VETMED

Dynamics of changes in AA levels in chicken dry matter

In terms of EAA (Table 7), the analysis focused on Lys, Met, Thr, Leu, Ile, Val, Tyr and Phe. As for NEAA (Table 8), the analysis focused on Asp, Ser, Glu, Pro, Gly, Ala or semi-essential AA such as Arg and His. Tryptophan and cysteine were destroyed in the acid hydrolysis.

The absolutely highest values of most EAA and NEAA were measured in the chicken whole-body dry matter on Day 5. All EAA exhibited a considerable drop in their mean values on Days 9 and 15 compared to the previous period (Table 7). In

Table 8. NEAA values in broiler chicken bodies over the course of rearing (g/kg dry matter)

			Day of rearing								
			1	5	9	15	20	25	30	35	40
Arg	♀	$\bar{x}$	44.0	45.0	17.8	17.8	26.8	32.4	34.2	32.8^{A}	28.8
		± SD	–	–	–	–		5.979	2.373	3.294	3.771
	♂	$\bar{x}$	42.5	46.8	30.9	26.5	25.4	38.4	34.1	36.1^{B}	26.6
		± SD	–	–	–	–		2.926	2.993	1.813	4.188
His	♀	$\bar{x}$	13.3	10.9	10.1	11.5	14.6	13.6	13.3	13.3	11.8^{A}
		± SD	–	–	–	–	10.185	1.127	1.174	1.025	1.357
	♂	$\bar{x}$	13.8	11.4	10.2	11.6	9.6	14.0	13.1	13.8	13.4^{B}
		± SD	–	–	–	–	0.932	0.859	1.198	1.089	1.055
Asp	♀	$\bar{x}$	49.8	52.9	38.7	33.8	38.6^{a}	37.7^{A}	38.0	36.7^{A}	33.3
		± SD	–	–	–	–	1.251	1.998	1.774	1.834	3.834
	♂	$\bar{x}$	48.8	52.9	43.0	37.1	37.1^{b}	43.8^{B}	39.7	41.8^{B}	33.7
		± SD	–	–	–	–	1.753	2.520	2.655	2.014	3.791
Ser	♀	$\bar{x}$	31.4	30.3	21.3	16.6	17.9	19.2	20.3	20.2	18.4
		± SD	–	–	–	–	1.825	1.336	1.762	2.277	2.204
	♂	$\bar{x}$	30.3	28.9	21.1	17.6	15.7	20.5	19.3	22.3	19.8
		± SD	–	–	–	–	2.727	1.819	1.721	1.754	1.957
Glu	♀	$\bar{x}$	69.1	74.4	70.5	58.6	56.9^{A}	57.5	53.2	53.7^{A}	51.6^{A}
		± SD	–	–	–	–	3.262	6.733	2.586	8.801	5.594
	♂	$\bar{x}$	65.0	77.0	68.0	61.9	52.7^{B}	60.0	56.8	62.6^{B}	62.9^{B}
		± SD	–	–	–	–	1.878	4.830	5.114	4.049	4.507
Pro	♀	$\bar{x}$	37.3	41.6	32.5	18.8	23.8	28.4	25.0	25.0^{A}	22.2
		± SD	–	–	–	–	5.348	3.332	3.699	2.094	3.074
	♂	$\bar{x}$	33.5	42.4	33.6	28.5	28.5	29.0	27.3	28.1^{B}	24.5
		± SD	–	–	–	–	9.418	3.985	2.616	1.334	2.088
Gly	♀	$\bar{x}$	43.2	50.0	44.5	36.3	35.8	36.6	27.1	32.1^{A}	37.1
		± SD	–	–	–	–	1.819	3.161	2.346	13.115	13.983
	♂	$\bar{x}$	42.1	52.9	42.2	39.8	34.4	40.1	28.8	49.6^{B}	32.3
		± SD	–	–	–	–	1.557	8.275	7.417	16.494	2.357
Ala	♀	$\bar{x}$	33.4	39.5	34.5	31.2	29.0^{A}	29.3	28.0	24.7	18.0
		± SD	–	–	–	–	1.260	1.625	2.947	5.098	1.852
	♂	$\bar{x}$	33.1	39.8	31.7	32.5	26.8^{B}	27.1	26.6	20.8	17.9
		± SD	–	–	–	–	1.171	5.034	4.576	6.225	7.010

Tryptophan and cysteine were destroyed in the acid hydrolysis

Different letters in corresponding rows denote statistical significance A : B ($P \leq 0.01$); a : b ($P \leq 0.05$)

$\bar{x}$ = mean; SD = standard deviation

doi: 10.17221/8441-VETMED

the subsequent period most AA exhibited a slight decrease in their levels or their levels fluctuated. While on Day 1 higher values were recorded for hens, at the end of the trial period (Days 35 and 40), with the exception of Lys and Tyr, higher values were measured for cocks and the differences between the sexes were statistically significant for most EAA ($P \le 0.01$; $P \le 0.05$).

As with EAA, a drop in the mean values of most NEAA in the whole-body dry matter of 9- or 15-day-old chickens was observed (Table 8). Increased concentrations were noted in the subsequent periods which were maintained, with only minor deviations, at about the same levels until the end of the trial. On Day 1, NEAA levels were higher in hens than in cocks while on Day 35 and 40, with the exception of Ser and Ala, the levels were higher in cocks and a statistically significant difference was found between the sexes ($P \le 0.01$; $P \le 0.05$).

In the dry matter of homogenised chicken bodies including feathers, the highest EAA levels were observed for Leu, Lys and Val, while the lowest levels were measured for Met, Tyr and Thr. As for NEAA, the highest mean levels in the whole-body dry matter were measured for Glu, Asp and Gly. His exhibited the lowest dry matter level.

Leu was found to be the EAA with the highest level in the hen body dry matter, 8.70% on average, followed by Val 6.54%, Lys 5.26%, Ile 5.25%, Thr 4.84%, Phe 4.30%, Tyr 2.51% and Met 2.21%. As in the hens, Leu was also the EAA with the highest level in the cock dry body matter, 8.42% on average, followed by Val 6.30%, Lys 5.36%, Ile 5.06%, Thr 4.57%, Phe 4.45%, Tyr 2.88% and Met 2.17%.

As for the mean percentages of NEAA in the total sum of amino acids, the hen dry body matter contained the highest levels of Glu 14.37%, followed by Gly 9.15%, Asp 9.09%, Ala 7.32%, Pro 6.68%, Arg 6.29%, Ser 5.11% and His 2.43%. The mean percentages for cocks were, in descending order, Glu 13.98%, Asp 9.22%, Gly 9.01%, Arg 7.36%, Pro 7.00%, Ala 6.99%, Ser 4.92% and His 2.41%. Cocks exhibited a slightly higher percentage of NEAA (60.79%) than the hens (60.41%).

Changes in percentages of amino acids in chicken body protein

On the basis of the results of the amino analyses of chicken bodies, the percentages of the individual amino acids in the bulk protein of the whole-body dry matter of hens and cocks were calculated. Bulk protein was taken as the sum of the amino acids (Σ AA).

Leu, at 8.40%, exhibited the highest level of any of the EAA in the bulk protein (Σ AA) of hen dry body matter followed by Lys 6.87%, Val 5.96%, Ile 5.02%, Thr 4.52%, Phe 4.25%, Tyr 3.20% and Met 2.20%. Leu, at 8.29%, also constituted the largest percentage of any of the EAA in the bulk protein (Σ AA) of cock dry body matter followed by Lys 6.80%, Val 5.89%, Ile 4.91%, Thr 4.43%, Phe 4.22%, Tyr 3.46% and Met 2.27%.

DISCUSSION

The components of mixed fodder for poultry differ in the quality as well as quantity of their amino acid content. The amounts of the individual amino acids and their proportions determine the biological value of feed proteins. The highest value was observed for animal proteins whose availability in the animal body ranges between 70 and 95%. For financial reasons, however, they are replaced in animal nutrition with plant proteins with lower biological value, and availability ranging from 60 to 65%. To increase their availability, fodders for poultry are fortified with amino acids. The highest need for proteins, including amino acids, occurs in the early periods of poultry life, due to their high growth intensity and feather production. As the birds get older, their need decreases.

While a lot of information can be found in the literature about the need for AA and their supplementation in animal diet (Aletor et al. 2000; Mukhtar et al. 2007; Zelenka et al. 2007) as well as AA levels in the muscles of broiler chickens (Suchy et al. 2002; Strakova et al. 2006; Meluzzi et al. 2009; Bogosavljevic et al. 2010 and more), information about AA levels in the whole bodies of reared chickens is rare (Stilborn et al. 1997; Stilborn et al. 2010). There is a particular paucity of reports on broiler hybrids, which are now widely used in the poultry industry. In recent years the genetic potential of broiler chickens has increased considerably and their higher yield has resulted in different amino acid needs (Salehifar et al. 2012). As reported by Emmans (1989), the question of AA needs may be addressed by analysis of amino acid levels in the body and feather protein.

doi: 10.17221/8441-VETMED

Dynamics of changes in nitrogenous substance (crude protein) content

Changes in N-substance levels over the course of rearing were most pronounced between Day 5 and Day 15 of chicken age, when a substantial drop was recorded. In this period nutrient re-sorption from the yolk bag is terminated and the chickens develop new feathers. In the subsequent period until the end of the trial, the decrease in N-substance levels continued and statistically significant differences between the sexes were determined on Day 35 and Day 40.

As reported by Stilborn et al. (2010), protein levels in broiler chicken bodies (without feathers) were significantly affected by age and sex. In the period from Day 0 to Day 42 of bird age, these researchers noted both age-dependent drops in protein levels and lower values in hens, which is in full agreement with the observations reported here. A higher reduction in N-substance levels in hens than in cocks reflects their earlier maturation and higher fat deposit rates (Stilborn et al. 2010).

Dynamics of changes in AA levels in chicken dry body matter

As with the N-substances, a significant drop in all EAA levels was noted on Day 9 and Day 15 of chicken age. Regarding Lys, Met and Phe, a subsequent increase was observed on Day 15 of the rearing period. From Day 20 to the end of the trial, no significant changes in EAA values were observed. Sklan and Noy (2005) monitored the AA composition of broiler bodies (without feathers) between Day 0 and Day 42 and observed that after the first 14 days of age the composition changed only negligibly, which is in complete agreement with the results of this study. Comparisons of the levels of the individual EAA in the dry body matter with regard to the sex of the reared chickens at the end of the trial period (Day 35 and Day 40) revealed higher levels in cocks, with the exception of Lys and Tyr, with statistically significant differences found for most EAA.

Stilborn et al. (2010) also reported a significant decrease in the levels of all monitored AA in the period between Day 0 and Day 14 of chicken age and in the following period (Day 42 to 112) a moderate increase, except for Phe where the drop was permanent. Sex was another factor affecting some AA, both EAA and NEAA. Differences between the sexes were only observed in later rearing periods and were ascribed to different maturation rates. In our trial we recorded statistically significant differences between the sexes already on Days 35 and 40. This may reflect the genetic potential of modern hybrids.

The trends described for EAA can also be generally applied to NEAA and correlated with data published by Stilborn et al. (2010). Minor differences may be explained by the fact that in our trial we analysed whole chicken bodies including feathers while Stilborn et al. (2010) only analysed chicken bodies without feathers. In another work (Stilborn et al. 1997), dealing with the effect of age on AA content in broiler chicken feathers, the same authors documented that age also plays a significant role in AA levels in feathers, especially between Days 14 and 42 of bird age. Kreuzer et al. (1988) also reported that AA levels in birds change with age, especially in the early growth period (Weeks 0 to 2). The amino acids Pro, Lys, His and Cys were specifically affected by feather changes, and changes in connective tissues and muscles and their proportions in the body. In contrast, Nitsan et al. (1981) who studied the AA composition of the body, skin and feathers of goslings from hatching to Day 42 of age, did not notice any effect of age. They found that gosling feather proteins are characterised by higher levels of Cys, Ser, Pro, Val and Tyr compared to the body and skin proteins. Similarly, Pellett and Kaba (1972) reported that AA composition is relatively constant in rapidly or relatively rapidly growing animals.

Changes in amino acid percentages in chicken dry body matter and protein

The percentage that EAA constituted of the total AA before Day 15 was slightly lower in cock dry body matter (39.21%) than in hen dry body matter (39.59%). From Day 20 to Day 40 of chicken age, this difference was slightly reduced. The measured percentage of essential amino acids out of the total AA in cocks was 40.26%, with the same value in hens being 40.42%.

Comparing the two sexes between rearing Days 20 and 40, large differences were found in the percentage of Tyr, the dry body matter of cocks contained

doi: 10.17221/8441-VETMED

0.25% more of this amino acid on average, while Met also exhibited a higher mean value. On the other hand, the dry body matter contained higher percentages of Thr, Leu, Ile, Val, Lys and Phe. In the case of the semi-essential and non-essential amino acids, higher values were found in cocks for Asp, Glu, Pro and Gly. The dry body matter of hens contained higher percentages of Agr, His, Ser and most of all Ala.

Age plays a more significant role in the levels of individual and total EAA than the hybrid type or sex of broiler chickens. Total NEAA as a proportion of total EAA increases with broiler age, which points to the feather composition changes that occur at this time (Stilborn et al. 1997).

The body proteins include 22 amino acids, all essential for the organism. Poultry need nitrogenous substances in quantities ensuring sufficient amounts of all essential amino acids and also of semi-essential and non-essential amino acids or substances needed for their synthesis. There is a long-term trend to not increase or even decrease nitrogenous substance content in fodder, although animals from modern genetic stock grow more intensely and therefore take up more nitrogenous substances from the fodder (Hussein et al. 2001). If the use of industrially made amino acids maintains a good balance of the individual essential amino acids, then the mixture can contain less nitrogenous substances. Such mixed fodder is usually cheaper and yet does not decrease the yield of the animals. The organism then secretes less excess amino acids, nitrogen content in the droppings decreases and thus the environmental burden is reduced.

REFERENCES

Anonymous (2007): Ross 308. Broiler Nutrition Specification. Accessed May 2014. http://en.aviagen.com/assets/Tech_Center/Ross_Broiler/Ross_308_Broiler_Nutrition_Spec.pdf.

Anonymous (2012): Ross 308. Broiler Performance Objectives. Accessed May 2014. http://en.aviagen.com/assets/Tech_Center/Ross_Broiler/Ross308BroilerPerfObj2012R1.pdf.

Aletor VA, Hamid II, Niess E, Pfeffer E (2000): Low-protein amino acid- supplemented diets in broiler chickens: Effects on performance, carcass characteristics, whole-body composition and efficiencies of nutrient utilization. Journal of the Science of Food and Agriculture 80, 547–554.

AOAC, Association of Official Analytical Chemists (2003): Official Methods of Analysis. 23rd ed. International Association of Analytical Chemists, Washington, DC, USA.

Applegate TJ, Angel R (2008): Protein and amino acid requirements for poultry. Animal Sciences. Accessed February 2015. http://puyallup.wsu.edu/dairy/nutrient-management/data/publications/Protein%20and%20amino%20acid%20for%20poultry-final.pdf.

Baker DH (1997): Ideal amino acid profiles for swine and poultry and their application in feed formulation. Biokyowa Technical Review 9. Biokyowa, Chesterfield, MO.

Bogosavljevic S, Pavlovski Z, Petrovic MD, Doskovic V, Rakonjac S (2010): Broiler meat quality: Proteins and lipids of muscle tissue. African Journal of Biotechnology 9, 9177–9182.

Dozier WA, Kidd MT, Corzo A (2008): Dietary amino acid responses of broiler chickens. Journal of Applied Poultry Research 17, 157–167.

Emmans GC (1989): Computer simulation in poultry nutrition. In: Proc. 3rd European Symposium Poultry Nutrition, Peebles, Scotland, 91–104.

Hussein AS, Cantor AH, Pescatore AJ, Gates RS, Burnham MJ, Ford MJ, Paton ND (2001): Effect of low protein diets with amino acid supplementation on broiler growth. Journal of Applied Poultry Research 10, 354–362.

Kreuzer M, Kirggessner M, Steinhart H (1988): Effect of the level of energy and protein supply, respectively of age on the amino acid composition of the broiler chicks. Archiv fur Geflugelkunde 52, 133–141.

Meluzzi A, Sirri F, Castellini C, Roncarati A, Melotti P, Franchini A (2009): Influence of genotype and feeding on chemical composition of organic chicken meat. Italian Journal of Animal Science 8, 2S, 766–768.

Mukhtar AM, Mekkawi A, El Tigani M (2007): The effect of feeding increasing levels of synthetic lysine and methionine in broiler chicks. Research Journal of Animal and Veterinary Sciences 2, 18–20.

Nitsan Z, Dvorin A, Nir I (1981): Composition and amino acid content of carcass, skin, and feathers of the growing gosling. British Poultry Science 22, 79–84.

NRC (1994): National Research Council. Nutrient Requirements of Poultry. 9th rev. ed. National Academies Press, Washington, DC. 173 pp.

Pellett PL, Kaba H (1972): Carcass amino acids of the rat under condition of determination of net protein utilization. Journal of Nutrition 102, 61–68.

Salehifar E, Shivazad M, Foroudi F, Chamani M, Kashani RB (2012): Reevaluation of digestible amino acid requirements of male and female broilers based on different ideal amino acids rations in starter period. Livestock Science 147, 154–158.

doi: 10.17221/8441-VETMED

Saunders AJ, Wessels JPH, Gous RM (1977): Carcass amino acid composition and utilization of dietary amino acids by chickens. South African Journal of Animal Science 7, 111–115.

Sklan D, Noy Y (2005): Direct determination of optimal amino acid intake for maintenance and growth in broilers. Poultry Science 84, 412–418.

Stilborn H L, Moran ET, Gous RM, Harrison MD (1997): Effect of age on feather amino acid content in two broiler strain crosses and sexes. Journal of Applied Poultry Research 6, 205–209.

Stilborn HL, Moran ET, Gous RM, Harrison MD (2010): Influence of age on carcass (feather-free) amino acid for two broiler strain crosses and sexes. Journal of Applied Poultry Research 19, 13–23.

Strakova E, Suchy P, Vitula F, Vecerek V (2006): Differences in the amino acid composition of muscles from pheasant and broiler chickens. Archive fur Tierzucht – Archives of Animal Breeding, Dummerstorf 49, 508–514.

Suchy P, Jelinek P, Strakova E, Hucl J (2002): Chemical composition of muscles of hybrid broiler chickens during prolonged feeding. Czech Journal of Animal Science 47, 511–518.

Zelenka J, Heger J, Zeman L (2007): Recommended Nutrient Content in Poultry Diets and Nutritive Value of Feeds for Poultry (in Czech). 1st ed. Mendel University of Agriculture and Forestry, Brno. 30 pp.

Received: 2015–03–07
Accepted after corrections: 2015–08–20

Corresponding Author:

Prof. Ing. Eva Strakova, Ph.D., University of Veterinary and Pharmaceutical Sciences Brno, Faculty of Veterinary Hygiene and Ecology, Department of Animal Husbandry and Animal Hygiene, Palackeho tr. 1/3, 612 42 Brno, Czech Republic
E-mail: strakovae@vfu.cz