AMINO ACID APPARENT DIGESTIBILITY ASSESSMENT FROM LAYING HENS DIETS SUPPLEMENTED WITH OIL INDUSTRY BYPRODUCTS

Iulia Varzaru[1*], Tatiana Panaite[2], Arabela Untea[2], Gabriela Cornescu[1], Mariana Ropota[2], Il. Van[1]

[1]University of Agronomic Science and Veterinary Medicine, Bucharest, Romania
[2]National Research Development Institute for Animal Biology and Nutrition – Balotesti, Romania

Abstract

An experiment was conducted on 114 laying hens in order to determine the amino acid apparent digestibility coefficients from diets supplemented with camelina meal, linseed meal and fenugreek seeds. The hens were assigned to 3 groups (C, E1, E2) and housed in metabolic cages (2 hens / cage, 38 hens / group). The control group received a conventional diet for laying hens with the age of 59 weeks. The experimental diets were enriched in PUFA by using 5% linseed meal and 2 % camelina meal (E1); 3% linseed meal, 3 % camelina meal and 1 % fenugreek seed (E2). During the final week of the experiment (for 5 days), the ingesta, the feed leftovers and the droppings were daily recorded with accuracy for each cage, in order to perform the amino acid balance. The digestibility coefficients of the dietary essential amino acids decreased significantly when compared with the control: for lysine 86.85 ± 2.74 % in C, 83.76 ± 3.83 % in E1, 81.34 ± 2.29% in E2; for methionine 87.30 ± 1.43 % in C, 84.48 ± 2.71 in E1, 80.83 ± 1.51% in E2; for cystine 87.30 ± 1.43 in C, 86.66 ± 2.11 % in E1, 86.13 % for E2. The amino acid analysis of laying hen eggs revealed a significant decrease (P≤0.05) of lysine, methionine and cystine in the albumen from E2, and a significant decrease (P≤0.05) of methionine in the yolk from E2, when compared with the control. This fact is correlated with the amino acid digestibility coefficients. The results highlight the decreasing effects of laying hens diets supplemented with oily byproducts on amino acid digestibility.

Key words: amino acid, digestibility, oily byproducts, laying hens

INTRODUCTION

The hen's egg is a complex biological and chemical entity. In recent years, the lipid composition of chicken egg has been an area of primary consumer concern, due to the connection between specific dietary lipids and the development of coronary heart disease and some forms of cancer [12].

The feeding value of the egg can be influenced by the formulation and composition of layer diets [6]. Hen egg enrichment in PUFA, compared to the standard eggs, can be done by feeding the layers diets which include vegetal oils (linen, canola, safflower), oleaginous seeds (linen, sunflower, canola) or by-products (wheat bran) [4].

The increasing prices of the high quality conventional forage sources challenged the scientists to identify low-economic value plants that can be used as sources of PUFA, protein. Most large companies no longer consider residues as waste, but as a raw material for other processes. By-product feedstuffs, which contain little economical value as edible foods for human consumption have become dietary sources of PUFA in animal nutrition.

Camelina (Camelina sativa L.) is an oilseed-producing plant in the family Brassicaceae (Cruciferae). If the oil from camelina becomes a viable substrate for biodiesel production, the marketing and beneficial use of the resultant meal would further increase the value of the crop [5]. Fenugreek (Trigonella foenum-gracecum. L) seeds are reported to have restorative and nutritive properties and are shown to

*Corresponding author: iulia_maros@yahoo.com
The manuscript was received: 12.12.2014
Accepted for publication: 26.03.2015

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stimulate digestive processes [10]. Moreover, fenugreek seeds contains antioxidants and protects cellular structures from oxidative damage [3]. Linseeds produce a vegetable oil known as flaxseed or linseed oil, which is one of the oldest commercial oils, rich in omega-3 fatty acids.

One of the problems which arises when PUFA-rich feeds are used is how is influenced the availability of other nutrients from layers diet. Knowledge of digestibility coefficients for individual amino acids in feed ingredients enables the formulation of diets more closely to the requirements of the bird [8].

The objective of the study was to determine the amino acid apparent digestibility coefficients from diets supplemented with camelina meal, linseed meal (oily byproducts) and fenugreek seeds.

MATERIALS AND METHODS

Animal experiment

The experiment was conducted in an experimental hall on 114 Lohmann Brown hens (age 59 weeks), divided into 3 groups (C, E1, E2), housed in improved metabolic cages (2 hens / cage, 38 hens / group). During the experiment (5 weeks) the light regimen was 16 hours/day. Food and water were provided ad libitum. Diet formulation considered the nutritional requirements for laying hens (NRC, 1994). The structure of the control diet was: corn (32,67), rice (15%), wheat (15%), rapeseed meal (15%), soybean meal (10%), sunflower oil (1%). The control diet had 16.35 % crude protein, 5.44 % crude fat and 2710 kcal/ kg feed. The experimental diets differed from the control by inclusion of: 5% linseed meal and 2 % camelina meal (E1); 3% linseed meal, 3 % camelina meal and 1 % fenugreek seed (E2).

Raw materials and compound feed samples were collected and physico-chemical parameters were determined according to Regulation (EC) no. 152/2009.

During the final week of the experiment (for 5 days), the ingesta, the feed leftovers and the excreta were daily recorded with accuracy for each cage, in order to perform the amino acid balance.

Amino acid apparent digestibility coefficients were calculated based on chemical determination from ingesta and excreta samples, and daily data representing the feed consumption and the dropping amount.

The coefficient of apparent digestibility represents the ratio between the amount of absorbed amino acid and the amount of ingested amino acid (expressed as percent), where the amount of absorbed amino acid is the difference between ingested amino acid and excreted amino acid as droppings.

In the balance period, 18 eggs/group were collected in a randomized manner, and analyzed for amino acids and physical parameters.

Amino acids determination

Amino acids from compound feed, droppings, eggs, were determined by high performance liquid chromatography (HPLC) using a method according to the UE Regulation 152/2009, which was optimized and validated by Varzaru et al. (2013) [11].

Equipment. HPLC Finningan Surveyor Plus (Thermo-Electron Corporation, Waltham, MA); HyperSil BDS C18 column, with silica gel, dimensions 250 × 4.6 mm, particle size 5µm (Thermo-Electron Corporation, Waltham, MA); rotary evaporator Buchi (Zurich, Switzerland).

Chemicals. Reference materials: lysine, aspartic acid, alanine, leucine (Merck, Darmstadt, Germany), cysteic acid and methionine sulfone (Sigma, Deisenhofer, Germany). Reagents: hydrogen peroxide (30%), disodium phosphate, sodium citrate, phenol, hydrochloric acid (37%, d = 1.19 kg·L-1), sodium hydroxide, boric acid, sodium disulphite (all analytical reagent grade), methanol and acetonitrile (HPLC grade) (Sigma, Deisenhofer, Germany).

Plants origin

Fenugreek and camelina seed were grown on the experimental plots of the National Agricultural Research Development Institute INCDA-Fundulea, in the South East region of Romania and linseed meal was imported from Agrosom GmbH (Germany).

In order to observe the evolution of the amino acid intakes and elimination, as well as the existing correlation during the balance period, the statistic soft StatView was used.

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RESULTS AND DISCUSSION

The amino acid analysis of the compound feed revealed that the lysine concentration ranged between 0.878 – 0.902 % SU, cystine between 0.423 – 0.465 % SU and methionine between 0.431 – 0.502 % SU (table 1).

Amino acid digestibility coefficients were calculated based on ingested and excreted amino acid amounts, during the balance period (table 2).

Table 1 Amino acid concentrations (g % SU) in analyzed compund feed

Amino acid	C	E1	E2
aspartic acid	1.731	1.700	1.719
glutamic acid	4.229	4.385	4.532
serine	1.025	1.029	1.063
glycine	0.792	0.800	0.839
threonine	0.757	0.776	0.665
arginine	1.246	1.130	1.231
alanine	0.967	0.986	0.905
tirosine	0.538	0.570	0.548
valine	1.340	1.069	1.104
phenylalanine	0.952	0.963	0.944
isoleucine	0.655	0.652	0.657
leucine	1.529	1.500	1.534
lisine	0.878	0.901	0.902
cystine	0.423	0.463	0.465
methionine	0.502	0.472	0.431

In general, it can be noticed a decreasing of the amino acid apparent digestibility coefficients in the experimental groups (E1 and E2) compared with the control group. For example, lysine, methionine and cystine registered a significant decreasing ($P \leq 0.05$) of the digestibility for E1 and E2, when compared with C. The most pronounced significant reduction ($P \leq 0.05$) was observed in the group with supplemented fenugreek seed (E2), where it can be noticed also a significant decrease ($P \leq 0.05$) of threonine digestibility.

Table 2 Apparent digestibility coefficients (%) for analyzed amino acids

Amino acid	C	E1	E2
aspartic acid	83.74 ± 2.55 ^{c}	81.13 ± 3.04	79.50 ± 2.43 ^{a}
glutamic acid	85.41 ± 2.42 ^{c}	83.50 ± 2.72	82.49 ± 1.72 ^{a}
serine	89.91 ± 1.66	88.98 ± 1.63	88.40 ± 1.17
glycine	71.20 ± 3.21	68.19 ± 4.99	67.92 ± 3.31
threonine	78.68 ± 3.11 ^{c}	77.42 ± 3.53 ^{c}	70.79 ± 3.37 $^{a, b}$
arginine	90.42 ± 1.78 $^{b, c}$	86.99 ± 2.50 ^{a}	85.82 ± 1.84 ^{a}
alanine	77.90 ± 4.34 $^{b, c}$	72.95 ± 5.15 ^{a}	68.86 ± 2.13 ^{a}
tirosine	86.12 ± 2.89 ^{c}	83.65 ± 3.02	80.81 ± 2.20 ^{a}
valine	84.98 ± 2.41 $^{b, c}$	79.11 ± 3.35 ^{a}	78.41 ± 1.93 ^{a}
phenylalanine	85.93 ± 1.81	85.67 ± 2.65	83.93 ± 1.66
isoleucine	79.07 ± 3.30 ^{c}	77.20 ± 3.24	75.37 ± 2.57 ^{a}
leucine	86.49 ± 2.16 ^{c}	84.65 ± 2.88	83.05 ±1.79 ^{a}
lisine	86.85 ± 2.74 $^{b, c}$	83.76 ± 3.83 ^{a}	81.34 ± 2.29 ^{a}
cystine	87.21 ± 1.40	86.66 ± 2.11	86.13 ± 0.67
methionine	87.30 ± 1.43 $^{b, c}$	84.48 ± 2.71 $^{a, c}$	80.83 ± 1.51 $^{a, b}$

Where: ^{a}significantly different ($p \leq 0.05$) from C; ^{b}significantly different ($p \leq 0.05$) from E1; ^{c}significantly different ($p \leq 0.05$) from E2.

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Albumen samples analysis showed a significant reduction (P≤0.05) of lysine concentration in E2 group compared with C group. Methionine and cystine content in albumen lowered significatly (P≤0.05) in E1 and E2 compared with C (table 3).

The reduction of the amino acid concentrations in albumen samples from E1 and E2 groups compared with C group, is correlated with the reduction of the amino acid apparent digestibility coefficients in the two groups.

The amino acid profile determination in yolk samples revealed a significant decreased (P≤0.05) of methionine concentration in E2 group, compared with C and E1 groups (table 3).

Table 3 Amino acid concentrations in albumen and yolk samples

Amino acid	C	E1	E2
	albumen, g % SU		
ac aspartic	7.300 ± 0.32	7.261 ± 0.28	7.209 ± 0.19
ac glutamic	11.610 ± 0.20	11.739 ±0.28	11.680 ± 0.28
serina	5.724 ± 0.37	5.804 ± 0.25	5.509 ± 0.16
glicina	3.244 ± 0.16[b]	3.027 ± 0.11 [a, c]	3.227 ± 0.18[b]
treonina	4.039 ± 0.32	3.964 ± 0.19	4.003 ± 0.21
arginina	5.319 ±0.30[b]	4.941 ± 0.38[a]	5.289 ± 0.11
alanina	5.902 ± 0.21	5.667 ± 0.19	5.707 ± 0.19
tirozina	3.528 ± 0.31	3.539 ± 0.28	3.534 ± 0.17
valina	6.703 ± 0.37	6.895 ± 0.32	6.843 ± 0.14
fenilalanina	5.925 ±0.43[b]	5.505 ± 0.29[a]	5.523 ± 0.23
izoleucina	5.113 ± 0.38[c]	5.033 ±0.29	4.699 ± 0.11[a]
leucina	7.504 ± 0.23[c]	7.746 ± 0.28	7.861 ± 0.22[a]
lizina	5.663 ± 0.38 [c]	5.437 ± 0.30	5.197 ± 0.19 [a]
cistina	1.974 ± 0.16 [b, c]	1.569 ±0.13 [a]	1.672 ± 0.16 [a]
metionina	3.483 ± 0.23 [b, c]	3.008 ± 0.22 [a]	2.935 ± 0.25 [a]
	yolk, g % SU		
ac aspartic	3.231 ± 0.18	3.098 ± 0.08	3.085 ± 0.25
ac glutamic	4.506 ± 0.15	4.435 ± 0.12	4.517 ± 0.15
serina	2.413 ± 0.11	2.391 ± 0.10	2.405 ± 0.20
glicina	0.918 ± 0.05	0.858 ± 0.04	0.904 ± 0.06
treonina	1.794 ± 0.06 [b]	1.684 ± 0.09 [a]	1.700 ± 0.11
arginina	2.732 ± 0.17 [b]	2.545 ± 0.10 [a]	2.494 ± 0.07
alanina	1.840 ± 0.12	1.775 ± 0.09	1.744 ± 0.06
tirozina	1.576 ± 0.04	1.573 ± 0.05	1.558 ± 0.05
valina	1.992 ± 0.08	2.017 ± 0.05	2.059 ± 0.12
fenilalanina	1.469 ± 0..09 [b, c]	1.307 ± 0.10 [a]	1.280 ± 0.06 [a]
izoleucina	1.698 ± 0.09	1.634 ± 0.09	1.648 ± 0.07
leucina	2.968 ± 0.18	2.831 ± 0.14	2.783 ± 0.15
lizina	2.517 ± 0.15	2.624 ± 0.10	2.610 ± 0.09
cistina	0.585 ± 0.02	0.589 ± 0.03	0.550 ± 0.04
metionina	0.621 ± 0.02 [c]	0.598 ± 0.05 [c]	0.545 ± 0.04 [a, b]

Where: [a]significantly different (p≤ 0.05) from C; [b]significantly different (p≤ 0.05) from E1;
[c]significantly different (p≤ 0.05) from E2

A reduction with 4.16 % of lysine concentration in the albumen from E1, and with 8.97 % in the albumen from E2, was observed.

In albumen, methionine decreased with 25.81 % in E1 and 18.06 % in E2, while cystine registered a reduction with 15.79 % in E1 and 18.67 % in E2 (fig. 1).

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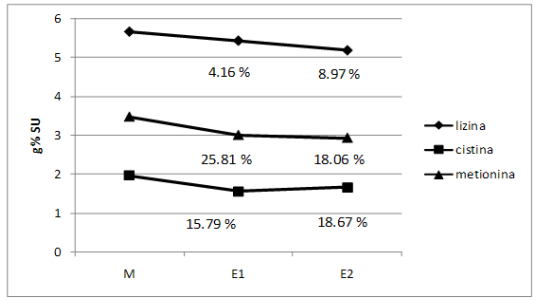

Fig. 1 The decreasing of essential amino acid concentrations in albumen samples

Other previous research regarding the nutrient digestibility from camelina and linseed meal have yielded similar results. Acamovic et al. (1999) [1] studied the nutritive value of camelina meal in poultry nutrition. Although the protein content is high, the researchers observed that camelina has a nutritive disadvantage like most crops from Brassica Family – the glucosinolates content and their degradation products. Other studies with mice have shown that the inclusion of camelina meal has reduced performance [7], although amino acid profiles are reasonable [9]. However, Acamovic et al. (1999) [1] showed low digestibility coefficients of nitrogen and dry matter.

Aziza et al. (2013)v[2] investigated the effect of camelina and linseed meal inclusion in the laying hens diet, on the apparent digestibility of crude protein and fatty acids. The researchers observed a significant decreased of the crude protein digestibility from the supplemented feed.

CONCLUSIONS

Laying hens diet enriched in PUFA by the inclusion of 5% linseed meal and 2 % camelina meal (E1), and 3% linseed meal, 3% camelina meal and 1 % fenugreek seed (E2), significantly decreased ($P \leq 0.05$) amino acid apparent digestibility coefficients, when compared with the control group (C).

The amino acid analysis of laying hen eggs revealed a significant decrease ($P \leq 0.05$) of lysine, methionine and cystine in the albumen from the group with 3% linseed meal, 3 % camelina meal and 1 % fenugreek seed (E2), and a significant decrease of methionine in the yolk of the same group, when compared with the control. This fact is correlated with the amino acid digestibility coefficients.

ACKNOWLEDGEMENTS

This paper was published under the frame of European Social Found, Human Resources Development Programme 2007-2013, project no. POSDRU/159/1.5/S/132765.

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