

# Effects of formulating diets with digestible amino acids and enzyme supplementation on the chemical composition of breast muscles in two broiler genotypes

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## ABSTRACT

The experiment was carried out on ISA 215 and Ross 308 broilers. Within each genotype, two hundred 21-day-old chickens were allocated to 4 groups of 50 birds (5 replicates of 5 males and 5 females per group). From days 22 to 42 of life the birds of both crosses were given two barley-based diets containing a high proportion of rapeseed meal and a mix of legume seeds. The formulation of the diet was based on total (TAA) or digestible amino acids (DAA), and diets were prepared with or without supplementing a commercial enzyme preparation containing  $\beta$ -glucanase, cellulase, xylanase,  $\alpha$ -amylase and protease.

Compared with ISA, Ross birds had lower ( $P<0.01$ ) body weight (BW) gains, but greater breast muscle yield (151 vs 162 g/kg BW;  $P<0.01$ ) and lower fat content in this muscle (10.6 vs 9.9 g/kg;  $P<0.01$ ). In ISA broilers, there was an increase in the level of muscle fat in the birds receiving the DAA diet. The significant interaction showed that the level of C18:0 acid increased in ISA females fed the DAA diet. The enzyme supplement increased ( $P\leq 0.05$ ) the crude protein (CP) content and reduced ( $P\leq 0.01$ ) the proportion of fat in ISA broilers (particularly in females;  $P<0.05$ ). The muscle lipids of ISA chickens fed the enzyme-supplemented diet contained more C18:1 (particularly males;  $P<0.05$ ) and total monounsaturated FA ( $P\leq 0.01$ ). Ross chickens responded to the formulation based on DAA with an increase in the CP and dry matter content in the breast muscle ( $P\leq 0.01$ ). The tendency towards reduced fat levels in the Ross birds fed the DAA diet was significant for females (interaction;  $P<0.05$ ). In contrast to ISA, in Ross birds added enzymes did not significantly affect the proportions of the analysed muscle components.

**KEY WORDS:** digestible amino acids, feed enzymes, breast muscle, chemical composition, genotype, broiler chickens

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## INTRODUCTION

Nutritional factors play an important role in modifying the chemical composition of chicken meat and its value as a food product, which is determined mainly by the quantitative and qualitative proportion of lipids (Mieczkowska and Smulikowska, 2005). It has also been shown that the deposition of breast muscles, the most valuable part of chicken meat, and the level of chemical components in these muscles vary according to bird genotype and sex (Gornowicz and Dziadek, 2001).

In an effort to maximize the use of amino acids (AA) for muscle formation in broiler chickens it is particularly important that the levels of AA in diets be formulated precisely with regard to their availability from individual feed ingredients. In evaluating availability of a protein-bound AA, determining the degree to which it is released from feedstuffs is of special importance. By taking account of only that portion of the total amount of an amino acid (TAA) that has been digested, it is possible to predict with a high probability the amount of AA that will be in a form suitable for absorption and utilization for tissue anabolism. Some earlier studies showed that the use of digestible amino acid (DAA) values in the formulation of diets containing sources of amino acids that are less well digested than those in soyabean meal (e.g., rapeseed meal, peas or faba beans) has a favourable effect on the quality of broiler carcasses in terms of increased muscling and reduced fatness (Perttilä et al., 2002; Szczurek, 2003b). No reports have revealed whether, and to what extent, formulation based on DAA values affects the chemical composition of muscles, especially when chickens are fed diets rich in components of lower AA digestibility. In an earlier study from our laboratory, feeding broilers during days 22-42 of age diets formulated according to the content of digestible AA in feed ingredients improved utilization of dietary nitrogen (Szczurek, 2003a). It was therefore assumed that the amount of crude protein ( $N \times 6.25$ ) deposited in chicken muscles may vary depending on the amino acid system used to formulate the composition of the grower diet.

The use of different combinations of carbohydrase enzymes ( $\beta$ -glucanase, cellulase, xylanase, hemicellulase and  $\alpha$ -amylase) in barley- and/or wheat-based diets containing such ingredients as lupins or peas and rape seed products, significantly improves the degradation of non-starch polysaccharides (NSP) and the digestibility of starch, crude protein and amino acids in broilers (Cowieson et al., 2003; Meng et al., 2005; Orda et al., 2006). Considering this, it seems possible to expect some changes in the levels of protein, fat and/or mineral components in chicken meat when birds are fed enzyme-supplemented diets. Therefore, it is surprising that there are few reports in the literature demonstrating the effects of feed enzymes on the retention of basic chemical constituents in broiler muscles. Because of interactions between diet ingredients, different enzymatic activities

of preparations used, and the sex of birds, prior research has produced inconclusive results. Kondzielska and Pisarski (2000) showed a significant reduction in intramuscular fat content in breast muscles after supplementation of barley-based diets with  $\beta$ -glucanase. Wojnowska and Gornowicz (2006) reported that the use of xylanase from *Bacillus subtilis* in a wheat-and-maize-based diet brought about a significant increase in fat content and a tendency towards increased crude protein retention.

Based on data obtained in an experiment with chickens representing two commercial broiler crosses, the presented work aimed at determining and comparing changes in the proportions of basic chemical constituents and composition of fatty acids in the breast muscles as caused by formulation of multi-ingredient diets on the basis of total or digestible amino acids and by supplementation of these diets with an enzyme complex (primarily carbohydrases). Within each genotype, sex interactions were used to evaluate the reaction of chickens to the dietary factors applied.

## MATERIAL AND METHODS

The study protocol and bird handling procedures were reviewed and approved by the Local Ethics Committee for Animal Experimentation.

### *Diets and animals*

Using locally grown cereals and plant protein sources, two grower-type diets were formulated to contain high proportions of barley, rapeseed meal and legume seeds (Table 1).

In the TAA diet, the levels of total amino acids followed values suggested in Poultry Feeding Standards (1996). The digestible amino acids (DAA) diet was formulated to reach the values obtained by converting TAA levels into DAA levels using the methodology and estimates proposed by Dalibard and Paillard (1995). Averaged true faecal digestibility coefficients derived from tables issued by Rhone-Poulenc (1993) and Ajinomoto Heartland LLC ([www.lysine.com/new/tecpoul2.htm](http://www.lysine.com/new/tecpoul2.htm)) were used to calculate the DAA contents in individual feed ingredients.

Each diet was prepared with or without an enzyme preparation (1 g/kg on top of feed). According to the manufacturer's specification (Hameco Agro B.V.) the preparation contained:  $\beta$ -glucanase, 400; cellulase, 600; xylanase, 400;  $\alpha$ -amylase, 15; protease, 14 units/g.

The concurrent introduction of different protein sources enabled the experimental diets to be formulated with high accuracy according to the levels of 9 essential amino acids while avoiding excessive amounts of some AA and the need

Table 1. Composition of experimental grower diet formulated on a total (TAA) or digestible (DAA) amino acid basis, g/kg

Item	Diet	
	TAA	DAA
<i>Feed ingredients</i>		
barley (10.9% CP)	321.2	316.1
maize (9.7% CP)	130.0	130.0
rapeseed meal (34.0% CP)	150.0	150.0
field pea seeds var. Piast (19.0 % CP)	150.0	150.0
lupin seeds var. Polonez (31.1% CP)	60.0	60.0
faba beans var. Nadwiślański (26.2% CP)	60.0	50.0
soyabean meal (45.5% CP)	5.0	20.0
DL-methionine (99%)	1.8	1.9
constant components <sup>1</sup>	122.0	122.0
<i>Calculated or analysed</i>		
metabolizable energy, MJ/kg <sup>2</sup>	12.23	12.22
crude protein <sup>2</sup>	164.80	168.30
Ca <sup>3</sup>	9.70	9.70
P available <sup>3</sup>	4.40	4.40
<i>Amino acids<sup>4</sup></i>		
total		
lysine	9.6	9.8
methionine	4.3	4.4
methionine + cystine	7.8	8.0
tryptophan	1.8	1.9
threonine	6.7	6.9
arginine	10.2	11.7
valine	8.3	8.8
leucine	13.2	13.9
isoleucine	7.1	7.7
digestible		
lysine	8.0	8.2
methionine	3.9	4.0
methionine + cystine	6.5	6.8
tryptophan	1.5	1.6
threonine	5.5	5.7
arginine	8.9	9.2
valine	6.7	7.0
leucine	11.7	12.0
isoleucine	5.9	6.3

<sup>1</sup> constant components, g/kg: rape seed oil 42, lard 42, dicalcium phosphate 20, limestone 10, NaCl 3 and trace minerals-vitamin premix 5 (provided per kg of diet, mg: retinyl palmitate 6.6, cholecalciferol 0.08, dl- $\alpha$ -tocopheryl acetate 40, menadion 2.25, thiamin 2.0, riboflavin 7.25, pyridoxine 4.25, cyanocobalamine 0.03, biotin 0.1, folic acid 1.0, nicotinic acid 40, calcium pantothenate 12, choline chloride 450, Fe 65, Zn 65, Mn 100, Cu 15, I 0.8, Se 0.25, Co 0.4, monensin sodium 100)

<sup>2</sup> calculated on the basis of chemical analysis of feed ingredients, energy value - using formulas of The European Table of Energy Values for Poultry Feedstuffs, 1989. 3rd Edition (WPSA, Wageningen)

<sup>3</sup> calculated according to the data given in Poultry Feeding Standards (1996)

<sup>4</sup> total amino acid values (TAA) are analysed, digestible amino acid values (DAA) are calculated on the basis of analysed amino acid composition of raw feedstuffs

to use crystalline L-valine, L-leucine and L-isoleucine. Both diets were supplemented with feed-grade crystalline DL-methionine considered 100% digestible. An exchange efficiency of 81.2% was also assumed for the use of DL-methionine to equalize total and digestible cystine content. The level of the other AA was adjusted by differentiating the proportions of soyabean meal and faba beans. The crude protein content, determined by the assumed amount of essential AA, and the proportion of dietary energy, were defined as the sum of the levels of particular ingredients. The calculated energy value of the diets was lower than that usually recommended for broilers older than 3 weeks, which provided an extra opportunity to improve energy utilization using enzymes.

One-day-old chickens of two commercial crosses: ISA 215 and Ross 308. (preliminary amount 240 per cross, sex ratio 1:1) with an average body weight of 39 and 38 g, respectively, were obtained from local hatcheries. The birds were raised in battery brooders under the same environmental conditions. Over the first 3 weeks, the chicks were fed *ad libitum* a maize-wheat-soyabean meal starter diet (12.85 MJ ME, 196 g crude protein, 11.5 g lysine, 8.3 g met+cys, 7.4 g threonine, 2.5 g total tryptophan per kg).

On day 21 of age, the chickens were weighed and birds with extreme body weights rejected. Birds selected for the experiment (200 ISA and 200 Ross broilers) were randomly allocated to experimental groups (50 birds per group) and placed in 87 × 90 × 37 cm wire cages. Within each genotype, four groups with 5 replicates (cages) of 5 males + 5 females were formed. From days 22 to 42 of age, chickens of both genotypes were fed experimental diets provided in mash form. Birds had free access to feed and water. During the experiment the temperature in the broiler house was maintained at 21°C and the lighting cycle was 20 h light:4 h dark.

### *Experimental procedures*

Body weight, feed intake and mortality were monitored throughout the experiment. At the end of the experiment, ten birds (5 females and 5 males with live body weights close to the group average) were chosen from each feeding group within a genotype and used for slaughter analysis. The birds were fasted 12 h, weighed and slaughtered by cervical dislocation. Carcasses were cooled overnight at 4°C and weighed. The breast muscles (*M. pectoralis major*) were excised, weighed and homogenized. After the samples were taken to determine dry matter, the homogenates were stored at -20°C for later analyses.

### *Chemical analyses*

The basic chemical composition of the feed materials and breast muscles was determined according to AOAC (1990). The samples of feed were hydrolysed in 6

NHCl at 110°C for 22 h for amino acid analyses. For determination of methionine and cystine, preliminary oxidation with performic acid was carried out. Tryptophan was assayed following alkaline hydrolysis of samples in a barium hydroxide solution (110°C, 18 h). All amino acids were determined by HPLC using a Beckman 126AA System Gold analyser.

The fatty acid content of muscle lipids was analysed after extraction of samples according to Folch et al. (1957). Esters of fatty acids were determined using a Varian 3400 gas chromatograph with a CP-WAX 58 column, an 8200CX autosampler and Varian Star 4.5 software.

### *Statistical analysis*

The results were subjected to analysis of variance using the Statistica 5.1 computer package ([www.statsoft.pl](http://www.statsoft.pl)). The effects of feed formulation method (TAA vs DAA), enzyme supplementation of diets (no enzymes vs with enzymes) and sex on the chemical parameters of breast muscles were analysed separately for both genotypes in a three-factorial design and the significance of the main effects and interactions was determined. Differences between ISA and Ross broilers, in growth, some slaughter indices and chemical composition of breast muscles, were tested using the pooled t-test.

## RESULTS

Significant differences in body weight gains (BWG) associated with the genotype of chickens (ISA>Ross) were found as early as at 21 days of age, after the preliminary phase of feeding. Comparative analysis of the data for both sexes (Table 2) showed that ISA birds also had significantly higher daily BWG than Ross chickens during the experiment (days 22 to 42). Ross broilers had a similar dressing percentage to ISA birds but greater ( $P<0.01$ ) breast muscle yield per kg live body weight at slaughter. Compared with ISA, the muscles of Ross chickens contained on average less ( $P<0.01$ ) crude fat (CF) and similar amounts of dry matter (DM), crude protein (CP) and ash (CA), and total saturated (SFA), mono-unsaturated (MUFA) and polyunsaturated (PUFA) fatty acids in the lipids of the analysed tissue.

In the chickens of ISA genotype (Table 3), especially in females ( $AA \times S$ ;  $P<0.05$ ), the method of diet formulation (TAA vs DAA) resulted in an increased proportion of CF in the muscles of birds receiving the DAA diet. The use of this diet did not significantly affect the mean levels of other basic components or the proportions of the major fatty acids in the muscle lipids of ISA broilers of either sex. However, the  $AA \times S$  interaction ( $P<0.05$ ) showed a clear increase in the

Table 2. The comparison of some growth and post-slaughter parameters and chemical composition of breast muscles of broilers used in the experiment (overall means for both sexes)

Parameter	ISA 215	Ross 308	SEM
Initial (day 21) body weight, g	638 <sup>A</sup>	559 <sup>B</sup>	6.7
Daily weight gain during experiment, g	75.7 <sup>A</sup>	66.7 <sup>B</sup>	0.9
Dressing percentage, % of live body weight	74.4	74.2	0.2
Breast muscle yield, g/kg live body weight	151 <sup>A</sup>	162 <sup>B</sup>	1.6
<i>Basic chemical constituents of breast muscle, g/kg of raw tissue</i>			
dry matter	259	258	0.5
crude protein	237	237	0.5
crude fat	11.2 <sup>A</sup>	9.9 <sup>B</sup>	0.2
crude ash	10.6	11.1	0.1
<i>Fatty acids<sup>1</sup> in muscle lipid, mg/g of total fatty acids</i>			
∑SFA	336	341	2.5
∑MUFA	373	369	3.7
∑PUFA	290	289	2.7

<sup>1</sup> SFA - saturated fatty acids (C14:0 + C16:0 + C18:0)

MUFA - monounsaturated fatty acids (C16:1 + C18:1)

PUFA - polyunsaturated fatty acids (C18:2 + C18:3 + C20:4 + C20:5 + C22:6)

<sup>A,B</sup> means in a row with different superscripts are significantly different at P<0.01

level of C18:0 acid in females receiving the DAA diet. Ross chickens (Table 4), both males and females, responded to the formulation based on DAA values with a significant ( $P \leq 0.01$ ) increase in the proportion of DM and CP and a decrease in the fat-to-protein ratio ( $P \leq 0.05$ ). The tendency towards a reduced level of CF in the muscles of Ross birds fed the DAA diet was confirmed statistically in females (AA × S;  $P < 0.05$ ).

The enzyme supplement increased the proportion of CP ( $P \leq 0.05$ ) and reduced the proportion of CF ( $P \leq 0.01$ ) in the muscles of ISA chickens (Table 3). The fat-to-protein ratio was also decreased. The reduction in fat content was particularly evident in females (E × S;  $P < 0.05$ ). The muscle lipids of ISA chickens receiving enzyme-supplemented diets contained significantly more C18:1 acid and total MUFA. The level of C18:1 acid underwent substantial modification in males (E × S;  $P < 0.05$ ). In Ross broilers (Table 4), the enzymes used did not significantly affect the proportions of the analysed chemical components in breast muscles. No enzyme supplement × sex interaction was found in the birds of this genotype.

Table 3. The proximate chemical composition (g/kg of raw tissue) and the major fatty acid content (mg/g of total fatty acids) of ISA 215 broiler breast muscle lipid depending on the dietary formulation based on a total (TAA) vs digestible (DAA) amino acids, the absence (-) or presence (+) of feed enzymes in the diet and sex of birds (main effect means, three-factorial ANOVA)

Parameters	Main effects						Interactions					
	formulation method (AA)		enzymes (E)		sex (S)		SEM	AA × E		E × S		AA × E × S
	TAA	DAA	-	+	male	female		AA × S	E × S			
<i>Basic constituents</i>												
dry matter	258	259	258	259	257 <sup>a</sup>	260 <sup>b</sup>	0.7	ns	ns	ns	ns	ns
crude protein	236	237	235 <sup>a</sup>	238 <sup>b</sup>	236	237	0.7	ns	ns	ns	ns	ns
crude fat	10.8 <sup>a</sup>	11.5 <sup>b</sup>	12.0 <sup>A</sup>	10.3 <sup>B</sup>	10.5 <sup>a</sup>	11.8 <sup>b</sup>	0.3	ns	*	*	ns	ns
crude ash	10.7	10.4	10.5	10.6	10.5	10.6	0.2	ns	ns	ns	ns	ns
fat:protein (×100)	4.6	4.8	5.1 <sup>A</sup>	4.3 <sup>B</sup>	4.5	4.9	0.1	ns	ns	*	*	*
<i>Major fatty acids</i>												
C16:0	229	229	233	225	230	228	2.6	ns	ns	ns	ns	ns
C18:0	95	98	97	96	92 <sup>a</sup>	101 <sup>b</sup>	2.3	ns	*	ns	ns	ns
C18:1	359	364	348 <sup>A</sup>	375 <sup>B</sup>	371	352	5.4	ns	ns	*	ns	ns
C18:2	196	206	204	198	200	202	2.7	ns	ns	ns	ns	ns
ΣSFA <sup>1</sup>	334	338	340	332	333	339	4.0	ns	ns	ns	ns	ns
ΣMUFA <sup>1</sup>	370	376	360 <sup>A</sup>	387 <sup>B</sup>	385 <sup>a</sup>	361 <sup>b</sup>	5.8	ns	ns	ns	ns	ns
ΣPUFA <sup>1</sup>	285	296	297	284	287	294	4.5	ns	ns	ns	ns	ns

<sup>1</sup> for explanations see Table 2  
 within main effects means in a row with different letters differ significantly: <sup>a,b</sup> - P≤0.05; <sup>A,B</sup> - P≤0.01  
 interactions: ns - non significant, \* - P<0.05



Table 4. The proximate chemical composition (g/kg of raw tissue) and the major fatty acid content (mg/g of total fatty acids) of Ross 308 broiler breast muscle lipid depending on the dietary formulation based on a total (TAA) or digestible (DAA) amino acids, the absence (-) or presence (+) of feed enzymes in the diet and sex of birds (main effect means, three-factorial ANOVA)

Parameters	Main effects						Interactions				
	formulation method (AA)		enzymes (E)		sex (S)		SEM	AA×E	AA×S	E×S	AA×E×S
	TAA	DAA	-	+	male	female					
<i>Basic constituents</i>											
dry matter	256 <sup>A</sup>	260 <sup>B</sup>	258	258	257	259	0.7	ns	ns	ns	ns
crude protein	235 <sup>A</sup>	239 <sup>B</sup>	237	237	237	237	0.8	ns	ns	ns	ns
crude fat	10.2	9.7	10.3	9.6	9.3 <sup>a</sup>	10.6 <sup>b</sup>	0.3	ns	*	ns	ns
crude ash	11.1	11.0	11.1	11.0	11.1	11.0	0.1	ns	ns	ns	ns
fat:protein (×100)	4.4 <sup>a</sup>	4.0 <sup>b</sup>	4.3	4.1	3.9 <sup>A</sup>	4.5 <sup>B</sup>	0.1	ns	ns	ns	ns
<i>Major fatty acids</i>											
C16:0	242	239	238	243	237	244	1.8	ns	ns	ns	ns
C18:0	92	89	88	93	89	92	1.5	ns	ns	ns	ns
C18:1	360	352	355	357	365 <sup>a</sup>	347 <sup>b</sup>	4.2	ns	ns	ns	ns
C18:2	205	209	210	204	207	207	1.7	ns	ns	ns	ns
ΣSFA <sup>1</sup>	344	338	339	343	335 <sup>a</sup>	347 <sup>b</sup>	3.0	ns	ns	ns	ns
ΣMUFA <sup>1</sup>	375	364	367	372	379 <sup>a</sup>	360 <sup>b</sup>	4.7	ns	ns	ns	ns
ΣPUFA <sup>1</sup>	287	292	291	288	290	287	2.9	ns	ns	ns	ns

<sup>1</sup>for explanations see Table 2  
within main effects means in a row with different letters differ significantly: <sup>a,b</sup>-P≤0.05, <sup>A,B</sup>-P≤0.01; interactions: ns - non significant, \*-P<0.05

## DISCUSSION

The results of many experiments testing the productive value of broilers show that ISA 215 birds are characterized by a high rate of growth, while Ross 308 chickens have a high breast muscle-to-body weight ratio. As expected, the yield of breast muscles (BM) in relation to live body weight was higher in Ross than in ISA broilers. The results of comparative studies indicate that BM yield is determined by the increased dimensions of muscle fibres and this trait is significantly correlated with the total amount of muscle fibres found in BM (Scheuermann et al., 2004). The genotype-related significant differences in the levels of basic chemical components in BM concerned the level of lipids (ISA > Ross). These observations are consistent with the findings of Gornowicz and Dziadek (2001) and support their classification of the commercial crosses of broiler chickens used in Poland into groups with a higher (including ISA 215) and lower (including Ross 308) proportion of crude fat in BM.

The diets used in the experiment contained major protein components for which the values of AA digestibility and bioavailability do not differ markedly (Parsons, 1996). Although some discrepancies between the actual and calculated (using published coefficients) amounts of digestible AA cannot be excluded, this allows the conclusion that the supply level of AA available to chickens for tissue synthesis was higher in the DAA diet. An indirect confirmation of this conjecture was the significant AA  $\times$  S interaction found in Ross females, indicating a decrease in the proportion of fat in BM when the DAA diet was fed. Many studies have shown that an increased AA-to-energy ratio in the diet reduces fat deposition in the bodies of chickens. Unlike in ISA chickens, the significant modifications in the analysed chemical parameters of Ross male and female muscles observed under the influence of the formulation method also included an increased level of CP. This resulted in an increase in DM content when the diet formulated according to digestible AA was used. This effect was comparable with the beneficial impact of the increasing AA supply on crude protein retention in the bodies of growing chickens (Eits et al., 2002). The fact that a similar phenomenon did not occur in ISA chickens is no surprise in the light of the study by Tesseraud et al. (2001). These authors showed that genetic factors modify CP metabolism and retention in the BM of chickens, whereas broilers characterized by a high yield of these muscles are more sensitive to changes in the supply of AA, lysine in particular.

Unlike in Ross, feeding a diet formulated on DAA basis increased the level of lipids in the muscles of ISA females, especially stearic acid (AA  $\times$  S interaction; Table 3). Assuming that the amount of AA available to chickens was higher when the DAA diet was fed and considering that, depending on the performance parameter, the AA requirements of females are usually lower compared with males (Garcia et al., 2006; Lumpkins et al., 2007), the above relationship could result

from the increased retention of fat derived from endogenous synthesis. It may be speculated that this fat could form during liponeogenesis from carbon residues of AA that were overabundant in relation to the needs of the birds of this sex. The lack of a similar reaction in Ross females can be linked to the higher nutrient requirements of Ross broilers compared with the amount of nutrients, including both forms of AA supplied in the feed. This was confirmed when the performance parameters of both chicken genotypes were compared in an earlier paper (Szczyrek, 2004).

The genotype of birds was a factor that determined changes in the chemical composition of muscles under the influence of the enzyme supplement. In ISA broilers, supplementation of diets with enzymes reduced the amount of CF, especially in females ( $E \times S$  interaction). A similar reaction to a multi-enzyme supplement in Starbro females (with higher fatness than males) was reported by Kondzielska and Pisarski (2000) who used barley diets supplemented with a mixture of vegetable oil and lard. In the case of Ross birds, with the same sex-related differences in CF level as those in ISA, the effect of supplemental enzymes was not significant. Improvements in fat digestibility and retention in the presence of exogenous carbohydrases were reported for Starbro males (Smulikowska and Mieczkowska, 1996). Therefore it seems that with a certain composition and the high level of fat in the experimental diets, the different reactions of birds of the same sex to the enzyme preparation used were determined by genotype. Differences resulting from bird genotype were also noticeable in the reaction of birds to the enzyme supplement with regard to CP content. Only ISA chickens responded to the diminishing of the adverse effect of water-soluble and insoluble NSP by enzymes (e.g.,  $\beta$ -glucans derived from barley or cellulose from rapeseed meal) and the expected increase in the pool of AA absorbed from the small intestine. This was likely due to the increased retention of protein in the rapidly increasing body weight of these birds, in response to the higher supply and/or favourable changes in the proportion of AA reaching the blood stream (Tesseraud et al., 2001). It is worth noting that some authors observed a significant improvement in the retention of nitrogen compounds in broiler carcasses under the influence of the enzyme complex (cellulase, xylanase,  $\alpha$ -amylase) added to a wheat-based diet with a high proportion of peas (Cowieson et al., 2003). The increased total MUFA (significant in ISA broilers) resulted from the increased proportion of oleic acid. Assuming an improvement in nutrient absorption conditions in the small intestine in the presence of NSP-degrading enzymes, this was possibly related to the better utilization of lupin seed fat, which is rich in this acid (Mieczkowska and Smulikowska, 2005). The increased *de novo* synthesis of C18:1 acid due to the raised supply of energetic substrates (glucose) and activity of cellular desaturase cannot be ruled out. This increase could result from the activity of the preparation's exogenous amylase, which assists the hydrolysis of starch derived from barley and maize.

The present results showed an inconsistent reaction of broiler chickens of two commercial genotypes to the systems of amino acid balancing and enzyme supplementation used in a low-energy grower diet based on barley and containing rapeseed meal and a mix of legume seeds as the main protein sources. Ross 308 broilers, characterized by greater breast muscle yield, reacted to diet formulation with digestible amino acid values with an increase in the crude protein and dry matter content of muscles. In ISA 215 chickens, with a lower yield of these muscles, the fat content increased. Faster-growing ISA broilers responded to the multi-enzyme supplement with increased deposition of crude protein and a reduced level of fat in the analysed tissue. The different reactions to the formulation with total or digestible amino acids and enzyme supplementation, with regard to the crude fat content of breast muscles and the proportion of oleic and stearic acids in this fat, could result from differences in the needs for amino acids associated with the sex and origin of the experimental birds.

The above relationships concerning the evaluated nutritional factors can be used in modelling the chemical composition, and thus the technological quality and dietetic value of the meat of broiler chickens representing different genotypes.

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