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Alterations in meat nutrient composition in response to a partial replacement of corn with triticale in the broiler diet

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ABSTRACT

The study evaluated the effect of feeding triticale on proximate composition, amino acids (AA) profile and nutritional value of meat (breast and thigh) of broiler chickens. A 5-weeks trial (1-35d) was conducted on Cobb 500 broilers (n=400), allotted into two dietary groups with five replicates (40 birds/replicate) and fed control (C, corn-soybean meal) and triticale (T, corn-triticale-soybean meal) diets. Broilers meat's proximate composition and energy value showed no significant differences between treatments. Feeding triticale significantly increased ($P<0.05$) breast muscle concentrations of arginine, valine and phenylalanine, while the methionine and alanine decreased ($P<0.05$) and did not affect the total AA, essential, non-essential or flavor AA. Concerning the thigh muscle AA profile, partial corn replacing with triticale decreased ($P<0.05$) the content of isoleucine, leucine, serine and alanine, without altering the total AA, EAA, NEAA, flavor AA or their ratio. The limiting AA in both muscle types was leucine, irrespective of diet. The EAA index and net protein value revealed no significant differences between diets or muscle types. In conclusion, partial corn replacement in broilers' diets with triticale did not adversely affect the proximate meat composition and positively impacted broilers' nutritional meat protein quality.

Keywords: amino acids profile, broilers, meat composition, triticale

INTRODUCTION

Triticale (*genus X Triticosecale*), a hybrid obtained by crossing wheat (*Triticum aestivum* L.) and rye (*Secale cereale* L.), is considered an alternative cereal for monogastric feeding due to its comparable nutrients' value to corn or wheat (Korver et al., 2004; Leeson and Summers, 2005). Moreover, triticale crops, combining the wheat productivity with the rye hardiness, have higher adaptability to agronomic conditions (e.g. lower soil requirements), resistance to drought and disease, and higher productivity potential (Boros, 1999). Triticale crop is spread in Romania, the cultivated area with triticale in 2018 was 78887 ha, with a production yield of 4277 kg/ha (FAOSTAT, 2020). Thus, triticale was not used as a common cereal for broiler chicks feeds compound due to the chemical composition variability compared to the other grains and antinutritive factors contents (e.g. soluble non-starch polysaccharides, NSP) that reduced the nutrients digestibility (Pourreza et al., 2007; Bederska-Łojewska et al., 2017). Triticale has higher protein and lysine contents than the parental species, and energy values are lower than wheat and corn (Rachwał, 2010). According to Smulikowska and Rutkowski (2005), triticale could be used, without enzyme addition, up to 20% in broilers diets and up to 30–40% in laying hens diets while the use of enzyme increases the level from 20 to 40%.

There are previous research focused on evaluating triticale effects as a partial or total substitute of corn or wheat on broilers performance and carcass traits (Hermes and Johnson 2004; Korver et al., 2004; Osek et al. 2010; Mahbub et al., 2011; Osek et al., 2013; Djekic et al., 2015; Gheorghe et al., 2017), or performance, gastrointestinal traits, gut morphometry and blood chemistry (Józefiak et al., 2007; Santos et al., 2008; Zarghi and Golian, 2009; Ozek et al., 2012; Alijošius et al., 2016) or meat quality, e.g. physical-chemical and sensory parameters, and fatty acids profile (Osek et al., 2010; Osek et al., 2013), e.g. physical and sensory evaluation (Al-Hajo et al., 2013), e.g. color and sensory characteristics (Alijošius et al., 2018) with different results, but no study reported the amino acids composition of broiler meat. Therefore, the study aimed to evaluate the effect of triticale as a partial corn replacement on broiler chickens' meat muscle proximate composition, amino acids profile and nutritional protein quality.

MATERIALS AND METHODS

The institute's ethical committee (INCDBNA-Balotesti, Romania) approved all protocol procedures according to the EU legislation (Directive 2010/63/EU).

Broilers and experimental diets

A 5-weeks trial (1-35 d) was conducted on four hundred one-day-old mixes sexed Cobb 500 broilers (42.36±1.22 g), procured from a commercial hatchery. They were individually weighed and allotted into two dietary groups with five replicates (40 birds/replicate). Chicks were reared in standard management-controlled conditions on wood shaving floor pens (2 x 1 m), fitted with feeders and nipple drinkers. A program of 23h of light and 1h of dark was used during the trial. The usual veterinary immunisation protocol for broilers diseases prevention (Marek's, Newcastle and Gumboro) was used.

Dietary treatment groups were control diet (C, corn-soybean meal) and triticale diet (T, corn-triticale-soybean meal), where triticale partially (50%) replace corn. The triticale was procured from a local crop producer and had a chemical composition of 89.15% dry matter, 11.60% crude protein, 1% crude fat, 3.83% crude fibre, 3000 kcal/kg metabolisable energy, and good essential amino acids (AA) profile (0.45% lysine, 0.57% sulphur AA, 0.50% threonine and 0.76% arginine).

The isocaloric and isonitrogenous diets were formulated according to the broiler's growth phases (starter, grower, finisher) to meet the nutritional requirements of hybrid broilers. The composition and nutrient content of diets are given in Table 1. During the trial, broilers had a*d libitum* access to feed and water.

Table 1. Composition and nutrients of three-phase broilers diets

Ingredients (%)	Starter (1-10 d)		Grower (11-22 d)		Finisher (23-35 d)	
	C	T	C	T	C	T
Corn	56.00	28.50	60.00	31.00	65.76	33.40
Triticale	0	28.50	0	31.00	0	33.40
Soybean meal	31.00	28.84	26.90	24.00	22.00	20.00
Corn gluten meal	5.00	5.00	4.60	4.60	3.60	3.60
Sunflower oil	2.80	3.98	3.50	4.41	4.00	4.98
Monocalcium phosphate	1.82	1.82	1.70	1.70	1.50	1.50
Calcium carbonate	1.47	1.47	1.39	1.39	1.27	1.27
Salt	0.30	0.30	0.30	0.30	0.30	0.30
DL-Methionine	0.15	0.13	0.20	0.18	0.19	0.16
L-Lysine HCl	0.40	0.40	0.35	0.36	0.32	0.33
Premix choline HCl	0.06	0.06	0.06	0.06	0.06	0.06
Premix vitamin-mineral[1]	1.00	1.00	1.00	1.00	1.00	1.00
Analysed composition (%)						
Metabolisable energy (MJ/kg)[2]	12.61	12.63	12.97	12.95	13.31	13.29
Dry matter	88.69	88.87	88.56	88.66	88.35	88.52
Crude protein	22.12	22.16	20.05	20.11	18.10	18.15
Lysine	1.32	1.32	1.19	1.19	1.05	1.05
Digestible lysine[3]	1.18	1.18	1.05	1.05	0.95	0.95
Methionine + cysteine	0.98	0.98	0.89	0.89	0.82	0.82
Digestible methionine + cysteine[3]	0.88	0.88	0.80	0.80	0.74	0.74
Calcium	0.90	0.91	0.85	0.85	0.76	0.77
Available phosphorus[3]	0.45	0.45	0.42	0.42	0.39	0.39
Crude fibre	3.59	3.98	3.40	3.75	4.10	4.23
Crude fat	5.70	6.00	6.40	6.59	6.20	6.52

Note: C, control; T, triticale. [1]Provided per kg diet: 4.47 mg vitamin A; 0.12 mg vitamin D3; 80 mg vitamin E; 4 mg vitamin K3; 4 mg vitamin B1; 9 mg vitamin B2; 4 mg vitamin B6; 0.020 mg vitamin B12; 15 mg vitamin

B5; 60 mg vitamin B3; 2 mg vitamin B9; 100 mg Mn; 100 mg Zn; 40 mg Fe; 15 mg Cu; 1.0 mg I; 0.30 mg Se; 0.25 mg Co; 60 mg lasalocid sodium (except finisher phase). [2]calculated values (NRC, 1994); [3]calculated.

Sample collection

At the end of the trial (35 d), six birds per treatment group were selected, euthanised by cervical dislocation, bleeding, defeathered, and eviscerated for muscle sampling. After the breast (*Pectoralis major*) and thigh (*Biceps femoris*) muscles were manually removed, the samples were grounded (Maxigel TC-121, Italy), homogenised, packed and frozen at –20˚C until chemical analyses.

Proximate analyses

The chemical composition of dietary components and feeds samples was determined following OJEU (2009) standards as follows: ISO 6496:2001 for dry matter, ISO 5983-2:2009 AOAC 2001.11 for crude protein, ISO 6492:2001 for crude fat, ISO 6865:2002 crude fibre, ISO 2171:2010 for ash, ISO 6490-2:1983 for calcium, and phosphorus by spectrophotometry method.

The breast and thigh muscle samples' proximate composition were determined according to (OJEU, 2009) standards: ISO 1442:2010 for dry matter, ISO 937:2007 for protein, ISO 1444:2008 for fat and ISO 936:2009 for ash. The meat energy value was estimated based on the content and physical equivalents of caloric amounts of fat (9.45 kcal/g) and protein (5.65 kcal/g).

Amino acids determination

As previously stated by Vărzaru et al. (2013), the AA profile was analysed by high-performance liquid chromatography utilising an HPLC Surveyor Plus Thermo Electron and HyperSil BDS C18 column dimensions 250mm x 4.6mm x 5m (Thermo Electron, Massachusetts, United States). In brief, the process involves acid hydrolysis to liberate AA from protein molecules, followed by sulphur AA oxidation with performic acid. The lysine, methionine, threonine, arginine, valine, phenylalanine, isoleucine, leucine, aspartic acid, glutamic acid, serine, glycine, alanine, tyrosine, and cysteine were AA identified and expressed as g/100 g. The essential AA (EAA), non-essential AA (NEAA) and their ratio were calculated. The flavour AA was calculated as the sum of aspartic acid, glutamic acid, glycine, alanine, and arginine (Liu et al., 2015).

Nutritional parameters calculation

AA score (AAS) was calculated by FAO (2013) reference pattern protein of the adult [lysine, 4.8; methionine + cysteine, 2.3; isoleucine, 3; leucine, 6.1; phenylalanine + tyrosine, 4.1; threonine, 2.5; valine, 4.0 (g/100g of protein)], following the WHO/FAO (2002) equation. The AA with the lowest AAS is considered the limiting AA.

$$\text{AAS (\%)} = \frac{\text{g of AA in 100 g of a test protein}}{\text{g of AA in 100 g of requirement pattern}} \times 100$$

EAA index (EAAI) was calculated by WHO/FAO (2002) as the geometric mean of the EAA ratio in the protein sample to their respective values in the reference pattern protein, following the equation:

$$EAAI = 100 \text{ x} \sqrt[n]{\frac{Lys_p}{Lys_s} \text{ x} \ldots \text{x} \frac{Val_p}{Val_s}}$$

where: *p* – protein sample; *s* – standard protein; *n* – number of amino acids (the pairs of methionine + cysteine and phenylalanine + tyrosine counted as one).

Net protein value (NPV) was estimated as follow:

$$NPV\,(\%) = \frac{\text{the lowest AAS \%}}{\text{protein \%}} \text{ x } 100$$

Statistical analysis

Data results were analysed by one-way analysis of variance (ANOVA) using GLM (SPSS software v.20, 2011). The results are shown as means and SEM (standard error of the mean). Statistical significance between means was reported at P<0.05.

RESULTS AND DISCUSSION

Feeding triticale as partial corn replacement in broilers diet had no significant effects (P>0.05) on overall period (1-35 days) performance variables (body weight gain 1866 *vs* 1895 g/broiler; feed intake 3340 *vs* 3322 g/broiler; feed conversion ratio 1.79 *vs* 1.75 g feed/g gain), carcasses traits (carcass yield 70.20 *vs* 70.71%; breast 23.25 *vs* 25.10%; thigh 19.32 *vs* 19.61%) and muscle pH_{24h} (breast 5.86 *vs* 5.91; thigh 5.97 *vs* 5.95) compared to control (Gheorghe et al., 2017).

Chemical composition of broilers meat

The meat chemical composition of broilers depends on genotype, slaughter age, sex, anatomical region, rearing system, and nutrition (Bogosavljević-Bošković et al., 2010). The raw poultry meat basic nutrients are proteins (18.4-23.4%), lipids (1.3-6.0%) and minerals (0.8-1.2%) (Culioli et al. 2003). The fat content showed a higher variability according to the muscles type being higher in the thigh than the breast (Castellini et al., 2002).

Broilers meat's proximate composition and energy value have shown no significant differences between dietary treatments (P>0.05; Table 2). Our results agree with international food composition references (Anses-Ciqual, 2020; USDA, 2016).

Osek et al., (2010) studied the triticale effects on Ross 308 broilers diets as partial or total corn and/or a wheat substitution on performance, carcass, physicochemical and organoleptic meat traits. Regarding the meat chemical composition, these authors reported that breast muscle had a high ash content as an effect of feeding broilers with a corn-wheat diet, as well as corn-triticale diet, while the ash content was decreased as an effect of feeding wheat-triticale (P<0.05). The same authors found that wheat-triticale or triticale diets mainly decreased the leg muscle protein. In contrast, corn-triticale, wheat-triticale, or triticale diets decreased the leg muscle dry matter content (P<0.05). Osek et al., (2010) concluded that fed triticale as partial or total cereals substitution in broilers decreased performance, but improved meat quality (e.g., fatty acids profile), providing consumers health benefits.

Table 2. Proximate composition (g/100 g) and energy value (kcal/100 g) of broilers meat[1]

Item	C	T	SEM	*P*-value*
Breast muscle				
Dry matter	25.13	25.10	0.26	*0. 986*
Crude protein	22.66	22.58	0.23	*0. 886*
Crude fat	1.27	1.25	0.02	*0. 999*
Ash	1.18	1.22	0.01	*0. 092*
Energy value	140	139	1.50	*0.898*
Thigh muscle				
Dry matter	24.60	25.42	0.42	*0.160*
Crude protein	20.29	20.54	0.28	*0.218*
Crude fat	3.26	3.74	0.14	*0.088*
Ash	1.08	1.11	0.02	*0.715*
Energy value	143	151	2.75	*0.117*

Note: [1]Means of 12 samples/group; SEM, standard error of the mean; *Not significant (P>0.05).

A study by Kokoszynski et al., (2018) in pheasants have been shown that partial dilution of finisher basal diet (71-112 d) with triticale (whole grain) had no adverse effect on productivity but affected the carcass traits negatively meanwhile meat quality in terms of physicochemical and fatty acid composition varied.

No significant changes in the meat chemical composition of Cobb 500 broilers have been reported by Gheorghe et al., (2021) in research concerning the partial substitution of corn with sorghum or sorghum-peas.

Amino acids profile of broilers meat

Broilers muscle tissue is the major source of protein and AA. Evaluating the AA profile of muscle provides an essential tool to determine the flavour and potential nutritional value of meat and is necessary for nutritionally health-conscious consumers (Kim et al., 2013).

Table 3 presents the amino acids composition of broilers breast muscle. Our results showed that dietary triticale increase significantly the levels of arginine (P=0.019), valine (P<0.0001), phenylalanine (P=0.012), while the methionine (P=0.011) and alanine (P=0.004) decrease in breast muscle. There was no significant effect on the total AA, EAA, NEAA or flavour AA of broilers breast muscle (P>0.05). The EAA/total AA or EAA/NEAA ratio do not differ between groups.

Table 3. Amino acids profile of broilers breast muscle[1]

Item (g/100 g)	C	T	SEM	*P*-value
Essential AA (EAA)				
Lysine	7.40	7.70	0.10	*0.169*
Methionine	2.30	2.10	0.05	*0.011*
Threonine	5.49	5.62	0.10	*0.548*
Arginine	5.08	5.33	0.06	*0.019*
Valine	4.78	5.05	0.05	*0.0001*
Phenylalanine	4.15	4.31	0.03	*0.012*
Isoleucine	4.33	4.41	0.04	*0.359*
Leucine	5.88	5.81	0.04	*0.480*
Non-essential AA (NEAA)				
Aspartic acid	9.40	9.42	0.06	*0.909*
Glutamic acid	13.39	13.32	0.07	*0.646*
Serine	3.89	3.90	0.02	*0.967*
Glycine	5.28	5.17	0.06	*0.350*
Alanine	4.52	4.34	0.04	*0.004*
Tyrosine	3.15	3.24	0.04	*0.289*
Cysteine	0.72	0.71	0.01	*0.767*
Total AA	79.76	80.39	0.30	*0.340*
EAA	39.40	40.29	0.25	*0.072*
EAA/Total AA ratio	0.49	0.50	0.001	*0.064*
NEAA	40.36	40.09	0.15	*0.405*
EAA/NEAA ratio	0.98	1.00	0.01	*0.065*
Flavor AA	37.67	37.56	0.17	*0.768*

Note: [1]Means of 6 samples/group; SEM, standard error of the mean.
Flavor AA = Aspartic acid + Glutamic acid + Glycine + Alanine + Arginine (Liu et al., 2015).
Means differ significantly at P<0.05.

Concerning the AA profile of broilers thigh muscle (Table 4), we noticed that feeding triticale diet, the content of isoleucine (P<0.0001), leucine (P=0.01), serine (P=0.012) and alanine (P=0.026) decreased, with no negative impact on the total AA, EAA, NEAA, flavour AA or their ratio (P>0.05).

No publications have been found regarding the effect of feeding triticale on muscle AA profiles of broilers for comparison. In our recent work, we also noticed that even some individual AA showed variation in the broilers' meat muscle, the total AA, essential or flavor-related AA contents were not significantly affected by feeding sorghum or sorghum-peas (Gheorghe et al., 2021).

Table 4. Amino acids profile of broilers thigh muscle[1]

Item (g/100 g)	C	T	SEM	*P*-value
Essential AA (EAA)				
Lysine	6.68	6.91	0.09	*0.216*
Methionine	1.55	1.46	0.02	*0.044*
Threonine	5.36	5.16	0.06	*0.129*
Arginine	5.36	5.01	0.12	*0.153*
Valine	4.20	4.46	0.07	*0.062*
Phenylalanine	3.94	3.87	0.05	*0.532*
Isoleucine	4.15	3.85	0.07	*0.010*
Leucine	5.59	5.29	0.06	*0.0001*
Non-essential AA (NEAA)				
Aspartic acid	8.86	8.60	0.09	*0.079*
Glutamic acid	13.21	12.98	0.12	*0.375*
Serine	3.94	3.70	0.06	*0.012*
Glycine	5.22	5.07	0.07	*0.322*
Alanine	3.71	3.60	0.04	*0.026*
Tyrosine	3.12	3.07	0.07	*0.724*
Cysteine	0.65	0.63	0.01	*0.212*
Total AA	75.55	73.67	0.58	*0.070*
EAA	36.83	36.02	0.25	*0.101*
EAA/Total AA ratio	0.48	0.49	0.001	*0.282*
NEAA	38.72	37.65	0.35	*0.068*
EAA/NEAA ratio	0.95	0.96	0.004	*0.281*
Flavor AA	36.36	35.26	0.31	*0.068*

Note: [1]Means of 6 samples/group; SEM, standard error of the mean.
Means differ significantly at P<0.05.

The present results showed that irrespective of dietary treatments, the AA profile of both muscle-types contained higher glutamic acid, followed by aspartic acid, lysine, leucine, threonine and arginine, and lower content of sulphury AA. Comparable findings were reported in broiler chicks (Gálvez et al., 2020; Gheorghe et al., 2021; Vargas-Ramella et al., 2021) or in turkey meat (Gálvez et al., 2018).

On the other hand, no adverse effect of diets or muscle type was found on the EAA/total AA ratio (> 40%) or EAA/NEAA ratio (> 60%), these results agree with other reports (Liu et al., 2015; Kim et al., 2017).

Nutritional quality of broiler meat

The AAS estimates the richness of individual EAA in food or feed ingredients based on a reference pattern protein, whereas the EAAI estimates the protein quality by the geometrical mean value of EAA related to a reference protein (WHO/FAO, 2002). It is considered that the EAAI is more linked with the biological protein quality than the AAS.

As can be seen in Table 5, breast muscle of broilers fed triticale have higher AAS of aromatic AA (P=0.036) and valine (P<0.0001), while sulphury AA had lower AAS (P=0.024) compared to control. The limiting AA in breast muscle was leucine (~96%), with no significant differences between dietary treatments (P>0.05). The results of the thigh muscle of broilers fed triticale (Table 5) have shown an increased AAS of valine (P=0.042) and a decreased AAS of leucine (P<0.0001) and isoleucine (P=0.01) compared to control. The thigh muscle limiting AA was leucine (~89%).

Table 5. Amino acids score (AAS), essential AA index (EAAI) of essential AA and net protein value (NPV) from meat muscle protein of broilers[1]

Item (%)	Reference protein[2] (g/100 g)	C	T	SEM	*P*-value
Breast muscle					
AAS					
Lysine	4.80	154	160	2.13	*0.169*
Sulphur AA[3]	2.30	131	121	2.46	*0.024*
Leucine	6.10	96	95	0.70	*0.480*
Isoleucine	3.00	144	147	1.38	*0.359*
Aromatic AA[4]	4.10	178	184	1.60	*0.036*
Threonine	2.50	220	225	3.81	*0.548*
Valine	4.00	120	126	1.36	*0.0001*
EAAI		144	146	0.66	*0.234*
NPV		21.83	21.52	0.37	*0.711*
Thigh muscle					
AAS					
Lysine	4.80	139	144	1.89	*0.216*
Sulphur AA[3]	2.30	96	91	1.38	*0.084*
Leucine	6.10	92	87	0.98	*0.0001*
Isoleucine	3.00	138	128	2.26	*0.010*
Aromatic AA[4]	4.10	172	169	2.95	*0.628*
Threonine	2.50	214	207	2.55	*0.129*
Valine	4.00	105	111	1.81	*0.042*
EAAI		131	128	1.04	*0.236*
NPV		18.15	17.82	0.22	*0.488*

Note: [1]Means of 12 samples/group; SEM, standard error of the mean; [2]FAO, (2013).
[3]Methionine + Cysteine; [4]Phenylalanine + Tyrosine. Means differ significantly at P<0.05.

We noticed that threonine had the highest AAS in both types of meat muscle irrespective of dietary treatments, followed by aromatic AA, lysine, and isoleucine.

In our research, EAAI and NPV results (Table 5) revealed no significant differences between dietary treatments or muscle types, confirming that the biological protein quality of the meat was not adversely affected.

CONCLUSION

In conclusion, triticale use in broilers' diets did not cause significant changes in proximate meat composition but positively altered broilers' meat protein quality in terms of amino acids profile, essential amino acids index and net protein value. Therefore, triticale represents an alternative to partial corn replacement in broilers' diets, mainly in climate change conditions.

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