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EFFECT OF ALFALFA PROTEIN CONCENTRATE (APC) SUPPLEMENTATION TO FATTENER DIETS ON PERFORMANCE, CARCASS TRAITS AND MEAT QUALITY*

Krzysztof Pietrzak, Eugeniusz R. Grela♦

Institute of Animal Nutrition and Bromatology, University of Life Sciences in Lublin, Akademicka 13, 20-950 Lublin, Poland
♦Corresponding author: ergrela@interia.pl

Abstract

The aim of the study was to estimate the effects of different levels of alfalfa protein concentrate (APC) supplementation on pig performance, carcass value and meat quality. The experiment was conducted on 50 crossbred pigs (PL × PLW) × Duroc of 29.0±0.5 kg initial body weight, which were divided into four treatment groups. Control (C) group was fed standard mixtures, groups E15 and E30 were fed with 1.5% or 3.0% APC additive, respectively. In group E30P the animals received feed including 3.0% APC for 14 days, followed by 14 treatment-free days. The production results were based on AGD, FCR, FI. Some slaughter traits and carcass value were measured. Determinations were carried out for physicochemical properties of fresh and thermally treated *m. longissimus*. The tissue samples (backfat, *m. longissimus*, liver, heart) were collected to analyze the fatty acid profile and cholesterol content. The recorded ADG was by 4.8% higher and FCR by 4.3% lower in E30P group as compared to control. The loin eye area was larger than in group C by 7.0% and by 8.3% in groups E30 and E30P, respectively. Carcass meatiness was also higher in groups E30 and E30P as compared to control, by 5.0% and 5.6%, respectively. The higher (P≤0.05) TPA parameters (hardness, chewiness and gumminess), as compared to control, was characteristic of the meat from animals receiving 3.0% APC. The lowest cholesterol content in the tissue samples (backfat, *m. longissimus*, liver) was recorded in group E30 (1.01, 0.51, 3.19 mg/g, respectively). Better fatty acid composition in backfat and *m. longissimus* was observed after 3.0% APC introduction into fatteners diet.

Key words: APC, lucerne, pig, carcass, meat quality

In swine production, apart from high animal performance (meatiness, ADG and FCR), the emphasis is put on pork quality (Rossi et al., 2013) which depends on genetic background in 30% and in 70% on environmental factors (Karpiesiuk and

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Falkowski, 2009), primarily feeding (Pietrzak and Grela, 2015). Therefore, feed material quality, nutrient content and a type of feed additives are crucial for both animal health and quality of pork (Karwowska et al., 2012; Pettigrew and Esnaola, 2001; Hanczakowska et al., 2015). One of feed supplements is the studied additive (APC – alfalfa protein concentrate) which contains β-carotene, vitamins (A, C, D, E, K), minerals (Ca, P, Mg, K, Fe, Mg, Cu, Zn), coumarins, isoflavones, naphthoquinone, saponins, alkaloids, coumesterol and L-canavanine amino acid (Grela and Pietrzak, 2014; Kundan and Anupam, 2011; EFSA, 2009; Aziz et al., 2006), thereby APC improves quality of such products. In dairy cow nutrition addition of APC increases the level of α-linolenic acid in milk (Dang van et al., 2011), the inclusion of the APC in turkey diets acts as an antioxidant in the raw poultry meat (Karwowska et al., 2010), the addition of a protein concentrate from lucerne to a feed mixture for hens increases the color intensity of egg yolk (Grela et al., 2014 b), in pig nutrition APC improves carcass value, which is characterized by higher loin eye area, better meatiness and also higher levels of PUFA in pork (Pietrzak and Grela, 2013; Karwowska et al., 2012; Grela et al., 2008). Some studies (Nasir and Grashorn, 2010; Raeesi et al., 2010) indicate continuous or intermittent application of phytogenic feed additives, i.e. *Allium sativum* powder and *Echinacea purpurea* juice to promote production performance and appropriate feed nutrient metabolism.

The objective of the research was to determine the effect of two levels of APC (1.5% and 3.0%) administered continuously or intermittently on production performance, some carcass traits and meat quality.

Material and methods

Animals and feeding

All procedures used in this experiment were approved by the responsible Ethics Committee for Experiments on Animals. The trial was conducted on 50 crossbred (Polish Landrace × Polish Large White) × Duroc grower gilts and 50 barrows of 29.0±0.5 kg initial body weight, randomly allocated into four treatment groups. Control (C) group was fed standard diets without APC addition, group E15 received 1.5% APC supplementation and groups E30 and E30P the mixtures with 3.0% APC inclusion. Two following feeding systems were implemented, the first applied to groups C, E15 and E30 where the animals received the in-feed additive continuously, while the other was used in group E30P – the animals received feed including 3.0% APC for 14 days, followed by 14 treatment-free days. The chemical composition and concentration of active ingredients of APC has been reported by Grela and Pietrzak (2014). Fatteners received complete grower and finisher mixtures (Table 1) which were analyzed to evaluate content of basic nutrients and detergent fiber fraction according to the standard AOAC (Association of Official Analytical Chemists) procedures (2012). The diets were balanced in terms of contents of metabolizable energy, total protein, fatty acids digestible to the end of the small intestine as well as minerals and vitamins to cover the fatteners' nutrient requirement according to Grela

et al. (2009). The animals from each group were divided into three pens: two pens with 5 gilts and 5 barrows each and third pen with 2 gilts and 3 barrows (in groups C and E15) and 3 gilts and 2 barrows (in groups E30 and E30P). The animals were kept in the fattening unit with slatted flooring (1 m^2 per animal), free access to drinkers and automatic feeders (feeding *ad libitum*). The zoohygienic conditions were the same for all treatment groups: light – 8 hours a day, with a minimum intensity 40 lx; temperature – 18°C; relative moisture – 70%; ventilation – 80 m^3 air per hour per animal. During the experiment pigs were weighed individually at the start of trial, at the end of grower period (about 70 kg BW) and at the end of finisher period (about 110 kg BW). Feed intake was recorded in individual pens and fattening periods.

Table 1. Composition and nutritive value of growing-finishing pig diets

Item	Grower				Finisher			
	C	E15	E30	E30P	C	E15	E30	E30P
Ingredients (kg)								
barley	530	530	530	530	742	742	742	742
wheat	200	200	200	200	100	100	100	100
fish meal	50	50	50	50	20	20	20	20
soybean meal	160	145	130	130	80	65	50	50
APC	0	15	30	30	0	15	30	30
soybean oil	20	20	20	20	20	20	20	20
ground limestone	12	12	12	12	12	12	12	12
2-calcium phosphate	10	10	10	10	10	10	10	10
salt (NaCl)	4.0	4.0	4.0	4.0	4.0	4.0	4.0	4.0
vitamin-mineral premix[1)]	10	10	10	10	10	10	10	10
lysine HCl	3.8	3.8	3.8	3.8	1.9	1.9	1.9	1.9
methionine	0.2	0.2	0.2	0.2	0.1	0.1	0.1	0.1
Nutrients (g/kg DM)								
dry matter	893.4	893.3	892.9	893.1	892.5	892.5	892.4	892.6
crude protein	173.3	173.4	173.2	173.7	151.5	151.2	151.6	151.4
crude fiber	39.6	39.5	39.7	39.5	42.3	42.1	42.3	42.2
NDF	173.8	174.0	173.9	173.7	191.6	191.8	191.5	191.7
ADF	63.4	63.6	63.3	63.5	64.2	64.4	64.2	64.1
ADL	8.7	8.8	8.6	8.7	9.6	9.5	9.6	9.6
ether extract	38.4	38.8	38.3	38.6	37.3	37.5	37.4	37.3
lysine	13.2	13.3	13.4	13.4	8.2	8.3	8.2	8.2
methionine	3.5	3.4	3.5	3.3	2.5	2.5	2.4	2.5
Ca	9.6	9.6	9.5	9.6	8.2	8.1	8.2	8.3
P	7.2	7.3	7.3	7.2	6.3	6.4	6.4	6.4
ME (MJ/kg)[2)]	12.8	12.8	12.8	12.8	12.6	12.6	12.6	12.6

[1)] Vitamin-mineral premix: 5 g Ca ($CaCO_3$)/($Ca(H_2PO_4)$/(CaI_2); 1.3 g P ($Ca(H_2PO_4)$); 100 mg Fe ($Fe(SO_4) \times 7H_2O$); 100 mg Zn (ZnO); 23 mg Cu ($CuSO_4 \times 5H_2O$); 1.2 mg I (CaI_2); 0.3 mg Se (Na_2SeO_3); 8,000 IU vitamin A; 1,000 IU vitamin D_3; 60 IU vitamin E; 0.60 mg vitamin K; 4 mg riboflavin; 22 mg niacin; 15 mg pantothenic acid; 0.02 mg vitamin B_{12}; 750 mg choline.

[2)] ME – metabolizable energy according to the equation of Kirchgessner and Roth (1983).

Carcass and meat measurements

The fatteners were slaughtered in accordance with the technology currently employed in meat industry, using the electrical stunning. Their right side carcasses were subjected to shortened slaughter analysis with the standard SKURTCh method (Różycki and Tyra, 2010). The heart and liver were weighed, whereas samples for the laboratory evaluation were collected from the liver, *longissimus* muscle, backfat and whole heart. The *longissimus* muscle (loin) samples were taken near the last thoracic and first lumbar vertebra, while backfat samples over the shoulder blade cutting out a lobe of 5 cm width and 10 cm length from a forequarter cut. Immediately after collection, the samples were stored at –20°C. Total fat of backfat, liver, heart and the *m*. *longissimus* for fatty acid analysis were extracted with a chloroform/methanol mixture according to the Folch et al. (1957) method. A percentage of fatty acid methyl esters was estimated using the gas chromatography procedure on a Varian CP-3800 chromatograph. The chromatograph operating conditions for fatty acid separation were as follows: the capillary column CP WAX 52CB DF 0.25 mm of 60 m length, gas carrier – helium, flow rate 1.4 ml/min, column temperature 120°C gradually increasing by 2°C/min up to 210°C, determination time 127 min, feeder temperature 160°C, detector temperature 160°C, other gases – hydrogen and oxygen. Cholesterol content in tissues was measured using the colorimetric method of Rhee et al. (1982). Lipid quality indices, i.e. atherogenic index (AI) and thrombogenic index (TI) were calculated according to the Ulbricht and Southgate (1991) equations. Hypocholesterolemic/Hypercholesterolemic ratio (h/H) was obtained according to Fernández et al. (2007). Analyses were conducted for physicochemical properties of loin eye area, namely in the samples of *longissimus* muscle from the last thoracic and first lumbar vertebra. The muscle samples were measured for pH_1 after 45 min and for pH_2 after 24 h post slaughter, and specific electrical conductivity (EC) with a PQM I-Kombi apparatus as well as subjected to color analysis according to CIE L*a*b* system (CIE, 1976) using Minolta CR-310 apparatus. Water binding capacity (WBC) was determined according to Grau and Hamm (1952). Drip loss percentage was estimated based on the difference between the weight of a muscle sample (packed in a foil bag) before and after the 60-min heat treatment in a water bath at 70°C. Analyses of the thermally-treated meat samples (after determination of drip loss) included the measurements of color (in CIE L*a*b* system) and texture parameters. A shear force test was performed with the use of ZwickRoell B0.5 testing machine in order to determine the maximal shear force and energy, and texture profile analysis (TPA) was conducted to determine: hardness, elasticity, gumminess and chewiness of meat samples. Cylindrical samples (20 × 25mm) were subjected to double compression till half their height using TA.XT.plus texture analyzer equipped in a P/10 attachment and an HDP/90 platform. Head speed was 10 mm/min. The shear force was measured using a Warner-Bratzler type shearing element. The maximum shear force was read out at head shift speed of 50 mm/min. The analyses were conducted on cubicoid samples (20 × 20 mm in cross-section). The samples were cut crosswise. Every measurement was performed in three replications, and results were presented as the mean replicate values.

Statistical analysis

Results were analyzed statistically with Statistica v.6.1 (2003). Mean value and standard deviation were computed for all analyzed variables. One-way analysis of variance ANOVA was carried out and the significance of differences between mean values determined with Duncan's test at significance levels of $P \leq 0.05$.

Results

Growth performance and carcass quality

The results of fatteners production and some carcass traits are presented in Table 2. The highest ADG of fatteners was recorded for group E30P and it was 4.8% higher as compared to control (C). Daily FI was similar for all the treatment groups, though FCR varied. The lowest FCR was reported for E30P group and it was lower by 4.3% than in groups C and E30. Cold dressing yield and meat content of ham were at a similar level in all groups, notably loin eye area and carcass meatiness differed statistically between the groups. The highest carcass meatiness was determined in the fatteners from groups E30 and E30P and it was higher by 5.0 and 5.6% respectively as compared to control. Similarly loin eye area was statistically higher by 7.0 and 8.3% in these groups, respectively as against the control. The fatteners from E30P group had the thinnest backfat and it was 6.2% less than in animals from the control group. Inclusion of 3.0% APC into fatteners diet in E30 group has significantly increased the weight of kidneys and liver, by 9.8 and 7.6% on average, as compared to control with no effect on heart weight.

Table 2. Growth performance and carcass quality traits and weight of some organs of fatteners

Item	Feeding groups				SEM
	C	E15	E30	E30P	
1	2	3	4	5	6
Initial body weight (kg)	30.4	30.5	30.6	30.5	0.62
Slaughter body weight (kg)	108.9	111.2	110.1	112.8	1.25
Average daily gains (g/d)	872 a	897 ab	883 ab	914 b	29.6
Daily feed intake (kg)	2.23	2.25	2.26	2.24	0.14
Feed conversion ratio (kg/kg)	2.56 a	2.51 ab	2.56 a	2.45 b	0.17
Cold dressing yield (%)	78.4	78.2	78.3	78.1	1.35
Meat of ham (%)	79.2	79.5	79.2	79.4	1.08
Loin eye area (cm^2)	44.4 a	46.3 ab	47.5 b	48.1 b	0.82
Meatiness of carcass (%)	55.3 a	55.9 a	60.3 b	60.9 b	0.91
Backfat thickness (cm)					
shoulder	3.12 a	3.05 ab	2.98 ab	2.89 b	0.084
midback	1.67 a	1.59 ab	1.60 ab	1.56 b	0.062
rump I	1.81 a	1.81 a	1.74 ab	1.71 b	0.053
rump II	1.31	1.29	1.24	1.25	0.034
rump III	1.71 a	1.69 a	1.63 ab	1.61 b	0.072
rump (3 measurements)	1.61 a	1.59 a	1.54 ab	1.52 b	0.055
average from 5 measurements	1.92 a	1.88 ab	1.84 ab	1.80 b	0.063

Table 2 – contd.

1	2	3	4	5	6
Weight of heart (g)	324	325	327	326	10.3
Weight of kidney (g)	163.4 a	168.3 ab	179.4 b	165.7 ab	8.24
Weight of liver (kg)	1.71 a	1.77 ab	1.84 b	1.79 ab	0.126

a, b – values in rows with different letters differ significantly (P≤0.05).

Meat quality

There were no statistically significant differences observed in pH values measured 45 min and 24 h after slaughter and in electrical conductance between the groups (Table 3). The analysis of L* and a* values did not demonstrate significant differences between the *longissimus* muscle samples, while yellowness (b*) and chroma value (C*) proved to be more intensive in loin of fatteners with 3.0% APC dietary incorporation as compared to control. Measurements of meat water holding capacity (WHC) did not show statistically significant differences for any of the traits under study. Assessment of the chosen traits of m. *longissimus* samples treated thermally did not exhibit any significant differences between the CIE Lab parameters, apart from chroma value (C*) which, like raw meat, was by 8.0 and 6.7% higher in the samples taken from fatteners from groups E30 and E30P, as compared to control (Table 4). Free drip amount remained at a similar level for all the groups. The feed additive investigated did not affect meat tenderness as the Warner-Bratzler test showed that shear force value did not differ significantly between the groups. The analysis of texture profile (TPA) has highlighted significant effect of the APC dietary supplement on meat quality improving meat hardness, cohesiveness and chewiness of the analyzed loin (Table 4).

Table 3. Meat quality indices of *longissimus* muscle after slaughter

Item	Feeding groups				SEM
	C	E15	E30	E30P	
pH_1 45 min after slaughter	6.23	6.25	6.21	6.22	0.121
pH_2 24 h after slaughter	5.50	5.52	5.53	5.51	0.112
Electrical conductivity mS cm^{-1}	16.8	16.9	17.2	17.0	0.231
Meat color CIE:					
lightness L*	51.75	51.94	52.08	51.97	1.25
redness a*	19.94	19.96	20.56	20.48	0.641
yellowness b*	1.42 a	1.52 ab	1.69 b	1.66 b	0.126
color chroma C*	19.83 a	20.56 ab	21.41 b	21.16 b	0.279
hue h°	4.5	4.6	4.8	4.7	0.134
Water holding capacity:					
G-H (cm^2)	7.89	8.03	8.14	8.05	0.45
G-H (mg)	75.12	76.32	77.35	76.49	1.48
M/T × 100	19.23	20.45	21.23	20.93	3.14

a, b – values in rows with different letters differ significantly (P≤0.05).
G-H – water holding capacity according to Grau and Hamm; M/T – filter paper press method.

Table 4. Meat quality indices of *longissimus* muscle after heat treatment

Item	Feeding groups				SEM
	C	E15	E30	E30P	
Meat color CIE:					
lightness L*	67.22	67.61	68.24	68.02	0.521
redness a*	11.47	11.56	11.71	11.63	0.182
yellowness b*	3.61	3.74	3.78	3.76	0.112
color chroma C*	11.21 a	11.85 ab	12.45 b	12.32 b	0.163
hue h°	17.2	17.5	17.8	17.7	0.634
Drip loss (%)	29.15	28.45	28.02	28.17	0.351
Warner-Bratzler test:					
shear force (N)	61.5	60.4	59.7	60.6	3.16
shear energy (J)	0.25	0.23	0.21	0.24	0.021
Texture profile analysis:					
hardness (N)	121.3 b	105.8 a	96.9 a	102.3 a	15.8
springiness (mm)	0.54	0.55	0.52	0.51	0.021
chewiness (N × mm)	21.24 b	18.78 ab	15.74 a	16.21 a	5.26
gumminess (N)	35.27 b	31.28 ab	28.34 a	29.12 a	5.69

a, b – values in rows with different letters differ significantly (P≤0.05).

Cholesterol content and fatty acid composition

The APC feed supplement did not affect significantly crude fat content in *m. longissimus*, liver, heart or backfat (Table 5). However, 3.0% APC applied continuously (E30) and intermittently (E30P) has beneficially decreased the cholesterol level in loin (on average 26.1 and 24.6%, respectively) and backfat (on average 16.5 and 14.0%, respectively) as compared to control. As for liver, this effect was noticed solely in E30 group with no influence on the cholesterol level in the cardiac muscle of the studied animals. The fatteners diet with the 3.0% APC additive was favorable to reduce saturated fatty acid (SFA) concentration with the concurrent rise of polyunsaturated fatty acid (PUFA) content in fatteners backfat (Table 6). The ratio of *n-6* to *n-3* fatty acids, as compared to control, has been significantly lower in the experimental groups (by 7.2, 14.4 and 11.5% in E15, E30 and E30P, respectively). The atherogenic (AI) and thrombogenic (TI) indices in E30 and E30P were close in value and significantly lower than in control (C). The hypocholesterolemic/Hypercholesterolemic acid ratio (h/H) was found significantly higher by 18.1 and 14.9% in the groups E30 and E30P, as compared to control. Table 7 presents the fatty acid composition in fatteners loin. The groups E30 and E30P showed a significantly elevated level of monounsaturated fatty acids (MUFA) in response to the feed supplement by 3.7 and 3.6%, respectively, in comparison to C group, with no effect on the SFA and PUFA content or *n-6/n-3* fatty acid ratio. The statistically significant increase in the hypocholesterolemic/Hypercholesterolemic acid (h/H) ratio was observed in group E30, but the AI and TI values determined for *m. longissimus* were not modified by

APC. The values of traits obtained from evaluation of the hepatic lipid composition were highly varied (Table 8). The experimental agent contributed to the changed profile of SFA, MUFA and PUFA, its effect manifested most clearly in E30 group and was confirmed statistically. The *n-6*/*n-3* fatty acid ratio changed under the studied additive in E30 and E30P groups as against control. In these groups a significant decrease of the thrombogenic index (TI) was noted. Regarding the cardiac muscle (Table 9), no significant differences were reported between the fatty acid profile, AI, TI indices or h/H acid ratio in the experimental groups. The statistically significant differences were determined only for the *n-6*/*n-3* fatty acid ratio in E30 group and it was 7.0% lower as compared to control.

Table 5. Crude fat and cholesterol content in tissues of finishing pigs

Item	Feeding groups				SEM
	C	E15	E30	E30P	
Musculus longissimus:					
crude fat (%)	2.41	2.35	2.34	2.38	0.12
cholesterol (mg/g)	0.69 a	0.57 ab	0.51 b	0.52 b	0.05
Liver:					
crude fat (%)	5.98	5.85	5.92	5.89	0.17
cholesterol (mg/g)	3.45 a	3.32 ab	3.19 b	3.33 ab	0.19
Heart:					
crude fat (%)	3.64	3.54	3.53	3.55	0.11
cholesterol (mg/g)	1.32	1.29	1.26	1.27	0.07
Backfat:					
crude fat (%)	83.78	83.19	83.09	83.26	0.87
cholesterol (mg/g)	1.21 a	1.12 ab	1.01 b	1.04 b	0.04

a, b – values in rows with different letters differ significantly (P≤0.05).

Table 6. Fatty acid composition (% total fatty acids) in backfat of finishing pigs

Item	Feeding groups				SEM
	C	E15	E30	E30P	
SFA	41.97 b	41.07 ab	40.13 a	39.95 a	1.22
MUFA	45.81	45.96	46.66	46.51	1.37
PUFA	11.17 a	11.88 ab	12.35 b	12.32 b	0.39
Σ *n-6*/Σ *n-3*	12.35 c	11.46 b	10.57 a	10.93 a	0.18
AI	0.57 b	0.53 ab	0.48 a	0.49 a	0.02
TI	1.39 b	1.31 ab	1.19 a	1.22 a	0.18
h/H	0.94 a	0.99 ab	1.11 b	1.08 b	0.03

a, b, c – values in rows with different letters differ significantly (P≤0.05).

SFA – saturated fatty acids; MUFA – monounsaturated fatty acids; PUFA – polyunsaturated fatty acids; AI – atherogenic index; TI – thrombogenic index; h/H – hypocholesterolemic/Hypercholesterolemic ratio.

Table 7. Fatty acid composition (% total fatty acid) in fat of *longissimus* muscle

Item	Feeding groups				SEM
	C	E15	E30	E30P	
SFA	41.14	40.91	39.57	39.45	0.49
MUFA	48.27 a	48.86 ab	50.04 b	50.02 b	0.67
PUFA	9.52	9.83	10.02	9.92	0.18
Σ *n-6*/Σ *n-3*	16.97	16.47	16.03	16.31	0.71
AI	0.58	0.57	0.54	0.55	0.02
TI	1.32	1.28	1.23	1.25	0.04
h/H	1.82 a	1.89 ab	1.96 b	1.91 ab	0.06

a, b – values in rows with different letters differ significantly (P≤0.05).

SFA – saturated fatty acids; MUFA – monounsaturated fatty acids; PUFA – polyunsaturated fatty acids; AI – atherogenic index; TI – thrombogenic index; h/H – hypocholesterolemic/Hypercholesterolemic ratio.

Table 8. Fatty acid composition (% total fatty acids) in fat of pig liver

Item	Feeding groups				SEM
	C	E15	E30	E30P	
SFA	42.39 b	41.79 ab	40.84 a	41.09 a	1.41
MUFA	18.74 a	19.04 ab	19.68 b	19.31 ab	0.52
PUFA	37.56 a	38.09 ab	39.43 b	38.24 ab	1.24
Σ *n-6*/Σ *n-3*	9.16 b	8.98 b	8.16 a	8.24 a	0.22
AI	0.34	0.33	0.32	0.32	0.02
TI	1.21 b	1.19 b	1.05 a	1.06 a	0.06
h/H	3.11	3.14	3.19	3.16	0.19

a, b – values in rows with different letters differ significantly (P≤0.05).

SFA – saturated fatty acids; MUFA – monounsaturated fatty acids; PUFA – polyunsaturated fatty acids; AI – atherogenic index; TI – thrombogenic index; h/H – hypocholesterolemic/Hypercholesterolemic ratio.

Table 9. Fatty acid composition (% total fatty acids) in fat of pig heart

Item	Feeding groups				SEM
	C	E15	E30	E30P	
SFA	32.29	32.21	32.02	32.13	0.84
MUFA	26.41	26.82	27.07	26.62	0.71
PUFA	40.43	40.47	40.38	40.43	1.05
Σ *n-6*/Σ *n-3*	8.84 b	8.66 ab	8.22 a	8.38 ab	0.23
AI	0.31	0.30	0.29	0.29	0.02
TI	0.77	0.75	0.71	0.72	0.04
h/H	3.56	3.62	3.73	3.69	0.12

a, b – values in rows with different letters differ significantly (P≤0.05).

SFA – saturated fatty acids; MUFA – monounsaturated fatty acids; PUFA – polyunsaturated fatty acids; AI – atherogenic index; TI – thrombogenic index; h/H – hypocholesterolemic/Hypercholesterolemic ratio.

Discussion

Introduction of the APC dietary supplement to fatteners diet has improved the ADG and FCR traits only in E30P group which received APC additive intermittently, every two weeks followed by 2 weeks of standard mixture supply. Beneficial effect of APC on ADG and FCR demonstrated in the present experiment was also indicated by other authors (Chen et al., 2014; Thacker and Haq, 2008; Wang et al., 2008 a) who studied feed supplementation with dried alfalfa and reported higher production effects in the final fattening stage. Positive impact of alfalfa demonstrated by improved carcass quality, i.e. higher meatiness and larger loin eye area was not found by Wang et al. (2008 a). Dietary inclusion of 3.0% APC supplied throughout the entire fattening period (E30 group) brought about higher weight of liver and kidneys which indicates enhanced metabolism in these organs in response to some active ingredients of APC (EFSA, 2009). The finding is consistent with the previous studies (Grela et al., 2008; Pietrzak and Grela, 2013) when after feeding pigs the diets with 1.5 or 3.0% APC, liver and kidneys weight increase was observed. Interestingly, APC additive administered at the same level but intermittently in E30P group did not contribute to higher liver and kidneys weight and moreover, allowed obtaining better animal performance than in E30 group. The experimental factor induced slight changes in color parameters prior to and post the thermal treatment, which was confirmed in other studies on alfalfa preparation use in fatteners (Karwowska et al., 2007; Karwowska, 2008) and turkeys (Karwowska et al., 2010). Likewise, alfalfa supplied as green forage did not change the *longissimus* muscle color (Wang et al., 2008 a; Karpiesiuk and Falkowski, 2009). Improvement of loin meat tenderness under APC effect was confirmed in the present experiment by the texture profile analysis. Similar tendency, but verified by the sensory characteristics, was reported by Karwowska et al. (2007). APC feed additive had positive influence on quality of meat and fat manifested by a reduced cholesterol level in these muscle tissues. Similar effect was observed when dried lucerne was incorporated into broiler chicken diets (Ponte et al., 2004). Saponins occurring in alfalfa were implicated in cholesterol level reduction, they prevent endogenous and exogenous cholesterol absorption through complex formation and enhanced secretion of steroids and bile acids and thus, intensify cholesterol metabolism in liver (Das et al., 2012; Soetan, 2008; Khaleel et al., 2005). The mechanism for the hypocholesterolemic activity of APC and lucerne saponins was confirmed by the values of pig blood serum biochemistry (Pietrzak and Grela, 2015; Wang et al., 2008 a, b). Fatty acid profile, the *n-6*/*n-3* fatty acid ratio, AI and TI index analyzed in the present experiment for muscle tissues were norm-referenced and did not deviate from the values reported by other authors (Grela et al., 2014 a; Razmaite and Švirmickas, 2012). As for *m. longissimus*, it was characterized by a higher MUFA level which corresponds to the results presented by Grela et al. (2008). However, the most noticeable effect of APC on fatty acid profile and other parameters was observed in fat of liver and backfat. Modification of the fatty acid composition is attributed to the fact that APC is a rich source of PUFA, especially *n-3* (EFSA, 2009; Grela and Pietrzak, 2014; Pezzi et al., 2005). The positive effect of APC was a decline of the atherogenic (AI) and thrombogenic (TI) index values in these tissues. Notably,

lower AI and TI values and higher hypocholesterolemic/Hypercholesterolemic acid (h/H) ratio in backfat are most desirable regarding consumer health (Razmaite and Svirmickas, 2012).

In conclusion, APC application improved production effects, yet it contributed only to a small extent to changes of meat color and texture. Content of bioactive compounds, mainly saponins, had the potential to beneficially affect lipid metabolism and caused low cholesterol levels and favorable fatty acid profile in the tissues under study. This effect was most clearly observable in response to 3.0% APC supplementation to pig diets, however its continuous supply increased liver and kidneys weight. Higher weight of the organs may be a manifestation of physical effect of some APC compounds on organism, therefore the 3.0% dietary additive to fatteners diet is recommended to be used intermittently.

References

AOAC (2012). Official methods of analysis of AOAC International. 19th ed., Latimer G.W. (ed.). Gaithersburg, Maryland, USA.

Aziz A.B., Grossman S., Budowski I., Ascarielli I., Bondi A. (2006). Antioxidant properties of lucerne extracts. Food Agricult., 19: 605–608.

Chen L., Gao L., Zhang H. (2014). Effect of graded levels of fiber from alfalfa meal on nutrient digestibility and flow of fattening pigs. J. Integr. Agr., 13: 1746–1752.

Dang van Q.C., Bejarano L., Mignolet E., Coulmier D., Froidmont E., Larondelle Y., Focant M. (2011). Effectiveness of extruded rapeseed associated with an alfalfa protein concentrate in enhancing the bovine milk fatty acid composition. J. Dairy Sci., 94: 4005–4015.

Das T.K., Banerjee D., Chakraborty D., Pakhira M.C., Shrivastava B., Kuhad R.C. (2012). Saponin: Role in animal system. Vet. World., 5: 248–254.

EFSA (2009). Opinion on the safety of "Alfalfa protein concentrate" as food. EFSA J., 997: 1–19.

Fernández M., Ordóñez J.A., Cambero I., Santos C., Pin C., de la Hoz L. (2007). Fatty acid compositions of selected varieties of Spanish dry ham related to their nutritional implications. Food Chemistry, 101: 107–112.

Folch J., Lees M., Stanley G.H.S. (1957). A simple method for the isolation and purification of total lipids from animal tissue. J. Biol. Chem., 226: 497–509.

Grau R., Hamm R. (1952). Eine einfache Methode zur Bestimmung der Wasserbindung in Fleisch. Fleischwirtschaft, 4: 295–297.

Grela E.R., Pietrzak K. (2014). Production technology, chemical composition and use of alfalfa protein-xanthophyll concentrate as dietary supplement. J. Food Process. Technol., 5: 373–377.

Grela E.R., Semeniuk W., Florek M. (2008). Effects of protein-xanthophyll (PX) concentrate of alfalfa additive to crude protein-reduced diets on nitrogen excretion, growth performance and meat quality of pigs. J. Cent. Eur. Agr., 8: 669–676.

Grela E.R., Pastuszak J., Bloch U. (2009). Handbook of modern pig feeding. Recommendation for practice (in Polish). SRRiL Progress, Lublin.

Grela E.R., Hanczakowska E., Kowalczuk-Vasilev E. (2014 a). Correlations between cholesterol content, fatty acid composition and health lipid indices in fat of chosen tissues and organs of finishing pigs. Pol. J. Vet. Sci., 17: 535–537.

Grela E.R., Ognik K., Czech A., Matras J. (2014 b). Quality assessment of eggs from laying hens fed a mixture with lucerne protein concentrate. J. Anim. Feed Sci., 23: 236–243.

Hanczakowska E., Świątkiewicz M., Grela E.R. (2015). Effect of dietary inclusion of a herbal extract mixture and different oils on pig performance and meat quality. Meat Sci., 108: 61–66.

Karpiesiuk K., Falkowski J. (2009). The efect of feeding and housing conditions of growing-finishing pigs on pork quality. Pol. J. Natur. Sci., 24: 198–206.

Karwowska M. (2008). Effect of applying alfalfa extract to the diet of pigs on meat colour. ŻNTJ, 60: 282–288.
Karwowska M., Dolatowski Z.J., Grela E. (2007). The effect of dietary supplementation with extracted alfalfa meal on oxidation stability of cooked ham. Pol. J. Food Nutr. Sci., 57: 271−274.
Karwowska M., Stadnik J., Dolatowski Z.J., Grela E.R. (2010). Effect of protein-xanthophylls (PX) concentrate of alfalfa supplementation on physico-chemical properties of turkey breast and thigh muscles during ageing. Meat Sci., 86: 486–490.
Karwowska M., Dolatowski Z.J., Grela E.R. (2012). Influence of dietary supplementation with protein-xanthophylls (PX) concentrate of alfalfa investigations on pig meat quality. Fleischwirtschaft. Inter., 27: 88–91.
Khaleel A.E., Gad M.Z., El-Maraghy S.A., Hifnawy M.S., Abdel-Satter E. (2005). Study of hypocholesterolemic and antiatherosclerotic properties of *Medicago sativa* cultivated in Egypt. J. Food Drug Anal., 13: 212–218.
Kirchgessner M., Roth F.X. (1983). Schätzgleichungen zur Ermittlung des energetischen Futterwertes von Mischfuttermitteln für Schweine. J. Anim. Physiol. Anim. Nutr., 50: 270–275.
Kundan S.B., Anupam S. (2011). Phytochemical and pharmacological potential of *Medicago sativa*. A review. Pharm. Biol., 49: 211–220.
Nasir Z., Grashorn M.A. (2010). Effects of intermittent application of different *Echinacea purpurea* juices on broiler performance and some blood parameters. Arch. Geflügelk., 74: 36– 42.
Pettigrew J.E., Esnaola M.A. (2001). Swine nutrition and pork quality: A review. J. Anim. Sci., 79: 316–342.
Pezzi P., Martino G., Simone N., Fusaro I., Giammarco M., Lambertini L. (2005). Feeding dehydrated alfalfa increases polyunsaturated fatty acids concentration in Marchigiana beef muscle. Ital. J. Anim. Sci., 4: 269–271.
Pietrzak K., Grela E.R. (2013). The influence of protein-xanthophyll concentrate of alfalfa on growth performance and carcass value of growing-finishing pig. Ann. UMCS, sect. EE, 31: 10–17.
Pietrzak K., Grela E.R. (2015). Influence of alfalfa protein concentrate dietary supplementation on blood parameters of growing-finishing pigs. Bull. Vet. Inst. Pulawy, 59: 393–399.
Ponte P.I.P., Mendes I., Quaresma M., Aguiar M.N.M., Lemos J.P.C., Ferreira L.M.A., Soares M.A.C., Alfaia C.M., Prates J.A.M., Fontes C.M.G.A. (2004). Cholesterol levels and sensory characteristics of meat from broilers consuming moderate to high levels of alfalfa. Poultry Sci., 83: 810–814.
Raeesi M., Hoeini-Aliabad S.A., Roofchaee A., Zare Shahneh A., Pirali S. (2010). Effect of periodically use of garlic (*Allium sativum*) powder on performance and carcass characteristics in broiler chickens. WASET, 68: 1213–1219.
Razmaitė V., Švirmickas G.J. (2012). Comparison of fatty acid composition in different pig tissues. Vet. Med. Zoot., 58: 77–82.
Rhee K.S., Dutson T.R., Smith G.C., Hostetler R.L., Reiser R. (1982). Cholesterol content of raw and cooked beef longissimus muscles with different degrees of marbling. J. Food. Sci., 47: 716–719.
Rossi R., Pastorelli G., Cannata S., Tavaniello S., Maiorano G., Corino C. (2013). Effect of long term dietary supplementation with plant extract on carcass characteristics meat quality and oxidative stability in pork. Meat Sci., 95: 542–548.
Różycki M., Tyra M. (2010). Metodyka oceny wartości tucznej i rzeźnej przeprowadzana w Stacjach Kontroli Użytkowości Rzeźnej Trzody Chlewnej (SKURTCh). Stan Hodowli i Wyniki Oceny Świń (in Polish). IZ, Kraków, Poland, 28: 94–105.
Soetan K.O. (2008). Pharmacological and other beneficial effects of antinutritional factors in plants – a review. Afr. J. Biotechnol., 25: 4713–4721.
Thacker P.A., Haq I. (2008). Nutrient digestibility, performance and carcass traits of growing-finishing pigs fed diets containing graded levels of dehydrated lucerne meal. J. Sci. Food Agr., 88: 2019–2025.
Ulbricht T.L.V, Southgate D.A.T. (1991). Coronary disease seven dietary factors. Lancet, 338: 985–992.
Wang C., Xu X., Yang Y., Lian H., Wang Y., Zhang C. (2008 a). Study on effects of alfalfa meal on carcass quality and serum items of fattening pigs. Sci. Agric. Sinica, 41: 1554–1559.

Wang C.Z., Li D.F., Yan X.B., Wang Y.H., Guo Y.X., Jiang Y.B. (2008 b). Effects of alfalfa meal on production performance and serum of fattening pigs. Acta Prataculturae Sinica., 6: 71–77.

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