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Effect of addition of dry distilled rose petals in the diet on the meat quality in entire male pigs

Влияние на добавянето на сух дестилиран розов цвят в дажбата върху качеството на месото при некастрирани мъжки прасета

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ABSTRACT

The objective of this study was to assess the effect of addition of phytonutrients from dry distilled rose petals (DDRP) in the feed on the meat quality and boar taint of entire male pigs. A total of 30 male 146 days old pigs from Danube white breed with an average body weight of 67 kg were allocated to one of the three groups – control (castrated male pigs - C), entire male pigs (EM), and EM fed with addition of 5 g of DDRP in 1 kg of feed (EM+R). At the age of 186 days and average live weight of 109 kg, the pigs were slaughtered and samples of m. Longissimus lumborum (LL) were subjected to analysis of meat quality characteristics (pH 45 min, pH 24 h, water holding capacity (WHC), L*a*b*c* colour space), proximate composition of the meat, fatty acid composition, androstenon and skatole content. The results showed that sex category had much more influence on the investigated traits than DDRP. The two groups EM pigs presented lower backfat content (P<0.001), lower intramuscular fat content (P<0.001) and higher lean meat percentage (P<0.01) in LL muscle, compared to C group. An increase in the n-6 and n-3 polyunsaturated fatty acids proportion and especially the content of alpha-linolenic acid, and intensity of meat colouring in the intact males, in comparison to castrated males, were found. The boar taint detected in the meat showed a trend of decreasing in the group consuming dry distilled rose petals.

Keywords: m. Longissimus lumborum, lean meat percentage, intramuscular fat content, fatty acid composition, meat colour, boar taint

РЕЗЮМЕ

Целта на това проучване беше да се оцени ефектът от добавянето на фитонутриенти от сух дестилиран розови цвят (СДРЦ) във фуража върху качеството на месото и миризмата на нерез от цели мъжки прасета. Общо 30 мъжки прасета на възраст 146 дни от Дунавска бяла порода със средно телесно тегло 67 кг бяха разпределени в една от трите групи - контролна (кастрирани мъжки прасета - КМП), некастрирани мъжки прасета (НМП) и НМП, хранени с добавяне на 5 g СДРЦ в 1 kg фураж (НМП+Р). На възраст от 186 дни и средно живо тегло 109 кг прасетата бяха заклани, бяха взети проби от m. Longissimus lumborum (LL) и бяха подложени на анализ на характеристиките на качеството на месото (pH 45 минути, pH 24 часа, капацитет за задържане на вода (WHC), L*a*b*c* цветово пространство), приблизителен състав на месото, състав на мастните киселини, съдържание на андростенон и скатол. Резултатите показват, че половата категория е имала много по-голямо влияние върху изследваните признаци от СДРЦ. Двете групи НМП имаха по-малка дебелина на гръбната

сланина (P<0,001), по-ниско съдържание на интрамускулни мазнини (P<0,001) и по-висок процент постно месо (P<0,01) в LL мускул, в сравнение с групата КМП. Установено беше увеличение на пропорцията на n-6 и n-3 полиненаситени мастни киселини и особено на съдържанието на алфа-линоленова киселина и интензивността на оцветяване на месото при некастрираните прасета в сравнение с кастрираните. Миризмата на нерез, открита в месото, показва тенденция към намаляване в групата, консумирала сух дестилиран розов цвят.

Ключови думи: m. Longissimus lumborum, процент постно месо, интрамускулно съдържание на мазнини, състав на мастни киселини, цвят на месото, миризма на нерез

INTRODUCTION

Smell and taste are one of the most important quality attributes of meat. In pork, the quality might be negatively affected by androstenone and skatole that are responsible for the so-called boar taint in intact male animals. Androstenone is a testicular steroid, whereas skatole is a by-product of intestinal bacterial fermentation in the hind gut. Boar taint is usually eliminated through castration; however, the surgical castration is negative from the animal welfare point of view (Lundström et al., 2009; Tuyttens et al., 2012; Tomasevic et al., 2020). Hence, alternatives and economically viable methods should be explored and applied to minimize or completely eliminate the boar taint appearance in pork from entire male pigs. A possible solution might be the development of specific nutritional strategies as well as inclusion of ingredients containing bioactive compounds. A wide range of ingredients aimed to influence the synthesis of skatole and androstenone have been identified in literature including tannin extracts, raw potato starch, organic acids, fructooligosachhrides, inulin from chicory root, etc. (Ivanova and Stoyanchev, 2019).

Recently, much attention has been drawn towards the enzymatic system in the liver. Certain possibilities for reducing skatole and androstenone exist by influencing the liver enzymatic system and it might be particularly affected by secondary plant metabolites and flavonoids (Urbanová et al., 2016). From the literature it is established that tannins, water soluble polyphenols, could decrease boar taint perception (Bahelka et al., 2021). Authors found that dietary tannins supplementation (3 and/or 4%) increased several saturated (SFA), monounsaturated (MUFA), and polyunsaturated fatty acids (PUFA) in pork. Vlahova-Vangelova et al. (2020) also reported that the DDRP or dihydroquercetin supplementation increases the amount of unsaturated fatty acids with 1.2 - 4.3% of two muscles and backfat of pigs. Polyphenols could also increase the redness index of breast fillet in poultry (Pirgozliev et al., 2018).

Dry distilled rose petals (Rosa damascene; abbev. DDRP) are a by-product of rose oil production, for which Bulgaria is famous in the world. According to Dragoev et al. (2020), DDRP are rich in antioxidants with synergistic effect. The amount of distilled rose petals obtained after production of 1 kg rose oil is approximately 50 kg. Further, from the average production of 1500 kg rose oil per year, DDRP to be derived are 75 000 kg (Balev et al., 2015). As a by-product, DDRP are currently discarded and even pollute the environment. Instead, if its efficiency for elimination of boar taint in the meat from intact male pigs is proven, it may be an appropriate solution for the farmers. The addition of DDRP to feeds to reduce and eliminate boar taint in pork and to possibly improve its sensory and technological parameters could be considered as an innovation in pig nutrition practices. Hence, the aim of the study was to assess the potential of phytonutrients from DDRP in the feed to affect the quality characteristics of meat and boar taint in entire male pigs.

MATERIALS AND METHODS

This experiment was conducted in accordance with Art.14 of Part V. Breeding and Livestock Units from European Convention for the Protection of Vertebrate Animals used for Experimental and Other Scientific Purposes, Commission Recommendation 2007/526/EC and Council Regulation (EC) No1099/2009. The

experiment was approved by the Bulgarian Scientific Ethics Committee and the requirements of the Council Directive 2010/63/EC were met.

Experimental animals and housing

The trial was carried out in the experimental farm of the Agricultural Institute - Shumen, Bulgaria with male Danube White pigs, following the Regulation 21/14.12.2005, regarding the minimum requirements for animal protection and welfare in pig breeding. A total of 30 male pigs, equalized by origin, age, and weight were used in the experiment. Pigs were allocated to one of the three groups – control, castrated pigs (C); entire male pigs (EM) and entire male pigs with addition of 5 g of dried distilled rose petals (DDRP) in 1 kg of feed (EM+R). They were settled into pens providing comfortable conditions for the animals (1m^2/pig). Attached to each of the pens was an outside yard for walking (2m^2/pig). Pigs were placed in the pens when their average body weight was 30 kg and were monitored trough the growing phase of fattening. The monitoring was made by weighing of pigs every two weeks. The experiment started when the pigs reached 67 kg of live weight (146 days of age) and lasted until the pigs reached 109.03 kg live weight (186 days of age) – finisher phase of fattening.

Experimental diets

The animals from the three groups were fed standard grower and finisher feeds in pellets provided ad libitum. The components, chemical composition and energy values of basal diets are given in Table 1. During the experimental period, DDRP in amount of 5 g/kg feed was added to the feed of the pigs from EM+R group. The DDRP were supplied by Damascena rose oil distillery, located in Skobelevo village, municipality of Pavel Banya, Stara Zagora district, part of Bulattars Production Company Ltd (Sofia, Bulgaria). After pressing, petals were dried and ground to particle size < 0.4 mm. Samples of finisher feed and roses were taken and subjected to proximate analysis and analysis of fatty acid profile. Data are presented in Table 2.

Table 1. Ingredients, chemical composition and energy values of basal diet

Components	Grower (30-67 kg)	Finisher (67-109 kg)
formulations		
Maize, g/kg	150.00	130.00
Barley, g/kg	250.00	100.00
Wheat, g/kg	270.00	500.00
Wheat bran, g/kg	80.00	70.00
Bio-concentrate BC14[a*], g/kg	250.00	–
Bio-concentrate BC16[b], g/kg	–	200.00
Total:	1000.00	1000.00
Calculated chemical compositions		
Moisture, g/kg	171.00	157.00
Dry matter, g/kg	829.00	843.00
Organic substances, g/kg	782.20	799.10
Crude protein, g/kg	157.50	150.20
Crude fats, g/kg	28.10	24.20
Crude ash, g/kg	46.80	43.90
Neutral detergent fibers, g/kg	47.90	38.40
Lysine, g/100 g	0.80	0.72
Calcium, g/100 g	1.31	1.26
Phosphorus, g/100 g	0.85	0.31
Energy intake		
Digestible energy, MJ/kg forage	13.46	13.72
Exchangeable energy, MJ/kg forage	12.92	13.18

*Notice:

[a] The bio-concentrate BC14 contents: 312.10 g/kg crude protein, 10.70 g/kg crude fat, 153.00 g/kg crude ash, 38.10 g/kg crude fibers, 5.88 g/100 g lysine, 2.79 g/100 g methionine, 7.80 g/100 g calcium, 2.69 g/100 g phosphorus, 268 mg/kg copper as sulphate, 670 mg/kg dl-α-tocopherol, 93800 UI/kg vitamin A, 16080 UI/kg vitamin D3, 1975.845 kcal/kg total energy

[b] The bio-concentrate BC16 contents: 348.00 g/kg crude protein, 17.40 g/kg crude fat, 165.00 g/kg g crude ash, 108.30 g/kg crude fibers, 2.26 g/100 g lysine, 0.67 g/100 g methionine, i.e. 1.25 g/100 g methionine + cystine, 1.31 g/100 g threonine, 3.66 g/100 g calcium, 0.95 g/100 g phosphorus, i.e. 0.67 g/100 g absorbable phosphorus, 0.78 g/100 g sodium, 560.00 mg/kg iron, 545.00 mg/kg zinc, 195.00 mg/kg manganese, 100.00 mg/kg copper, 4.10 mg/kg iodine, 1.50 mg/kg selenium, 0.40 g/100g antioxidants, 320.00 mg/kg vitamin E, 32500 UI/kg vitamin A, 6000 UI/kg vitamin D3

Table 2. Chemical composition and fatty acid profile (% of fatty acid methyl esters - FAME) of the finisher diet and the DDRP (dried distilled rose petals)

Chemical components, %	Feed	DDRP
Crude protein	14.75	9.84
Fat	1.98	0.34
Ashes	2.83	3.35
Crude fibers	3.80	27.5
Nitrogen free extracts	58.45	47.52
Fatty acids, %		
C14:0	0.14	3.01
C15:0	0.10	-
C16:0	23.95	30.62
C16:1	0.96	2.04
C17:0	0.14	1.70
C18:0	2.30	13.06
C18:1	19.68	5.61
C18:2	48.80	23.41
C18:3	3.93	20.55

In vivo measurements

The *in vivo* measurements were done on the pigs from all groups using the ultrasound device Piglog 105. The following traits were recorded: 1. Thickness of the backfat between 3d and 4th lumbar vertebrae, 7 cm aside the midline (X1); 2. Backfat thickness between the 3d and 4th last rib (X2); 3. The thickness of LL muscle in X2 (X3). The lean meat percentage was calculated according to the following equation:

LM=63.8662-0.4465 X1-0.5096 X2+0.1281X3

Slaughtering procedure and sampling

The animals were slaughtered at an average live weight of 109.03 kg and 186 days of age in a certified slaughterhouse. The carcasses were skinned, eviscerated, the leaf fat was removed and split apart. The hot carcass weight was recorded for calculation of dressing percentage. The carcasses were cooled for 24 h at 4°C. Samples were taken from m. LL for analysis of meat quality and fatty acid profile.

Analysis of pH, colour and Water holding capacity (WHC) of meat

Value of pH, colour and WHC were measured as indicators of meat quality. The pH was measured in m. LL at 45 min and 24 h post mortem with portable pH meter equipped with glass electrode (Testo 205). The colour measurements were done at laboratory conditions at Trakia University 24 h post mortem on plain chops using the CIELAB method, expressed as lightness (L*), redness (a*), and yellowness (b*), with Chromameter CR-400 (Konica Minolta). Colour saturation (chroma, C*) and the hue angle h were also calculated. The pH and the colour measurements were done on three locations of the muscle and the results were averaged. Further the samples were minced in a meat grinder. Aliquots of 300 mg were used for determination of WHC, and the rest was vacuum-packed and stored at –20°C until analysis of proximate composition and fatty acid profile. All the determinations in the analyses below were done in triplicate per sample. WHC was determined as free water content (%) as described by Grau and Hamm (1952).

Proximate composition

The moisture, protein and ash contents of m. LL, feeds and rose petals were determined according to AOAC, (2004).

Fatty acid analysis and cholesterol

The fatty acid composition was determined according to the method of Bligh and Dyer [10] with slight modifications as described by Vargas–Ramella et al. (2020). The fatty acids were transesterified according to the procedure previously described by Domínguez et al. (2015), with some modifications. Fatty acids are presented as percentage of the total amount of the fatty acid methyl esters (FAME) identified (Christie, 1982).

Detection of boar taint

Boar taint detection was estimated on the slaughter line, by a trained person applying "hot knife" technique as described by Aluwé et al. (2012). The odour was scored from 0 to 3 points, with 0 defined as neutral undetectable

and 3 as strong boar taint.

HPLC analysis of androstenone and skatole

Detection and quantification of androstenone and scatole were done by the HPLC method described by Hansen-Moller (1994) with slight modification of Ampuero Kragten et al. (2011). Back fat samples from each pig carcass were collected at slaughterhouse and then melted in microwave. Methanol extraction from 0.5 g melted fat were performed and androstenone and skatole analysed by HPLC Surveyor 1100 (Thermo scientific) with a fluorescence detector set on 346/521 nm in extraction and emission. An ODS 2 (Hypersil) reverse phase column was used. The calibration solutions and each of the samples were measured three times.

Statistical evaluation

The statistical analysis was performed using JMP v.7 software package. Each pig was used as an experimental unit. The normality of distribution for the data and homogeneity of variances were checked by Shapiro-Wilk and Brown-Forsythe tests. The differences between groups were evaluated through one-way ANOVA procedure and Tukey post hoc comparison at P<0.05.

RESULTS

In vivo measurements of pigs

As presented in Table 3, the live weight and the hot carcass weight of the pigs were not affected by the treatment, however significant differences were observed in regard to the dressing percentage (P<0.0001). The C group had higher dressing percentage when compared to the groups of EM, whereas the latter showed similar values of this trait (65.50% and 63.68%, respectively for EM and EM+R). Furthermore, considerable differences among groups were found concerning the backfat thickness at X1 (P<0.0001) and LL thickness (P=0.026). Both EM and EM+R groups showed considerably lower backfat thickness at X1 in comparison to the C group well as lower thickness of LL muscle. It could be noticed that in regard to these parameters the EM+R group had intermediate position, however, no significant differences due to rose petals supplementation were detected. The discrepancies in the above-mentioned parameters were associated with significant differences in the lean meat (P=0.006). The highest backfat thickness measured at X1 in C group corresponded to the lowest lean meat percentage in comparison to the EM and EM+R groups.

Meat quality attributes

The pH, WHC and instrumental colour measurements of m. LL are presented in Table 4.

The values of pH 45 differed significantly among the groups (P=0.012), being the higher in the C pigs than in the animals from EM+R group. The same discrepancies were not observed for the pH measured 24 h post mortem. The colour parameters also did not present considerable differences among groups except for the b* (P=0.030)

Table 3. *In vivo* measurements of carcass quality in castrated (C), entire male (EM) and entire male pigs consuming DDRP (EM+R)

Item	Control group C	Experimental group EM	Experimental group EM+R	SEM	Significance
Live weight, kg	107.90	109.40	109.80	5.62	0.964
Hot carcass weight, kg	76.96	71.60	70.40	3.85	0.450
Dressing %	71.27^{a*}	65.50^{b}	63.68^{b}	2.62	<0.0001
Bakfat thickness X_1, mm	20.00^{a}	11.80^{b}	13.30^{b}	3.76	<0.0001
Backfat thickness X_2, mm	13.60	11.70	11.10	2.85	0.142
m.LL thickness, mm	51.20^{a}	43.70^{b}	45.50^{ab}	6.04	0.026
Lean meat, %	54.40^{a}	58.23^{b}	58.05^{b}	2.71	0.006

Notice: Values connected with different letters are significantly different (P<0.05)

and C* values (P=0.034). The C group presented lower b* values as well as lower colour saturation (8.93) than EM+R pigs.

The chemical composition of meat (Table 5) showed significant differences between groups only in regard to the content of fat (P=0.0005) and ashes (P=0.0045). Intramuscular fat (IMF) content was significantly higher in C pigs (2.42%) that in the entire males and the entire males receiving rose petals in the feed. The same was observed in ash content being higher in C group compared to EM and EM+R. No discrepancies in moisture, protein and total cholesterol content were observed between the three groups.

Fatty acid profile of m. LL

A total of 15 fatty acids were detected in the LL muscle (Table 6). The major saturated fatty acids were C16:0 and C18:0. The latter tended to differ among groups (P=0.094), showing higher content in C group compared to EM and EM+R groups. The opposite trend was observed in regard to the percentage of C17:0, where both groups of entire males displayed higher content of this fatty acid in m. LL that C group. Gradual increase in the percent of C15:0 was observed showing the highest content in EM+R group, however the differences between groups were not significant. The content of C14:0 remained unaffected by castration or inclusion of rose petals in the diet.

Table 4. Quality attributes of m. LL in castrated, entire male and entire male pigs consuming DDRP

Item	Control group C	Experimental group EM	Experimental group EM+R	SEM	Significance
pH 45 min.	6.14^{a*}	5.87^{b}	5.93^{ab}	0.19	0.012
pH 24 h	5.46	5.44	5.45	0.05	0.422
WHC, %	23.17	22.76	24.44	2.91	0.417
L*	51.48	52.84	50.74	2.68	0.225
a*	6.50	6.51	7.41	1.17	0.158
b*	6.05^{a}	6.87^{ab}	7.40^{b}	1.06	0.030
C*	8.93^{a}	9.55^{ab}	10.53^{b}	1.30	0.034
h*	0.75	0.81	0.78	0.09	0.491

*Notice: Values connected with different letters are significantly different (P<0.05)

Table 5. Proximate composition of m. LL in castrated, entire male and entire male pigs consuming DDRP

Item	Control group C	Experimental group EM	Experimental group EM+R	SEM	Significance
Moisture, %	74.49	76.05	75.43	1.85	0.186
Protein, %	21.82	21.29	21.59	1.59	0.762
IMF,%	2.42^{a*}	1.57^{b}	1.84^{b}	0.43	0.0005
Ashes, %	1.27^{a}	1.09^{b}	1.14^{b}	0.12	0.0045
Cholesterol, mg/100 g	41.96	38.54	38.66	4.04	0.146

*Notice: Values connected with different letters are significantly different (P<0.05)

Table 6. Fatty acid profile of m. LL in castrated, entire male and entire male pigs consuming DDRP

Item	Control group C	Experimental group EM	Experimental group EM+R	SEM	Significance
C14:0	1.44	1.40	1.54	0.30	0.577
C15:0	0.52	0.80	1.22	0.81	0.206
C16:0	26.30	25.73	27.85	3.14	0.353
C16:1	4.18	3.83	4.22	0.69	0.439
C17:0	0.31	0.62	0.62	0.33	0.091
C18:0	11.55	10.16	10.27	1.44	0.094
C18:1n-9	47.27^{a*}	43.35^{ab}	39.94^{b}	4.85	0.014
C18:2n-6	5.99^{a}	9.74^{b}	10.21^{b}	2.15	0.0006
C18:3n-3	0.18^{a}	0.26^{ab}	0.33^{b}	0.10	0.019
C20:2n-6	0.18^{a}	0.30^{b}	0.22^{ab}	0.09	0.045
C20:3n-6	0.22^{a}	0.36^{b}	0.37^{b}	0.09	0.005
C20:4n-6	1.51^{a}	2.79^{b}	2.59^{b}	0.72	0.002
C20:5n-3	0.08	0.16	0.20	0.11	0.107
C22:5n-3	0.20^{a}	0.35^{b}	0.32^{ab}	0.11	0.026
C22:6n-3	0.07	0.15	0.10	0.09	0.251

*Notice: Values connected with different letters are significantly different (P<0.05)

The content of the major monounsaturated fatty acid (MUFA), C18:1n-9 differed significantly among groups (P=0.014). Its maximum value was observed in the C group and minimum in EM+R group. The three examined groups differed considerably in regard to the proportion of C18:2n-6 (P=0.0006) and C18:3n-3 (P=0.019). The m. LL of castrated pigs had lower content of C18:2n-6 and C18:3n-3 in comparison to the entire males from both groups. The same significant differences were observed in the percentage of the n-6 polyunsaturated fatty acids (PUFA), (C20:2n-6, C20:3n-6 and C20:4n-6). On the other hand, only C22:5n-3 from the n-3 long chain PUFA displayed significant differences among groups (P=0.026), with lower percentage in C group when compared to the EM and EM+R groups. The changes in the total content of MUFA, PUFA, n-6 and n-3 PUFA corresponded with those described for the individual FA (Table 7). The C group had the highest content of MUFA and lowest of PUFA, n-3 and n-6. The lipid nutritional indices did not differ among the groups, except P/S (P=0.0006), being lowest in C pigs compared to EM and EM+R.

Detection of boar taint

The presence of the boar taint in the three groups is shown in Figure 1.

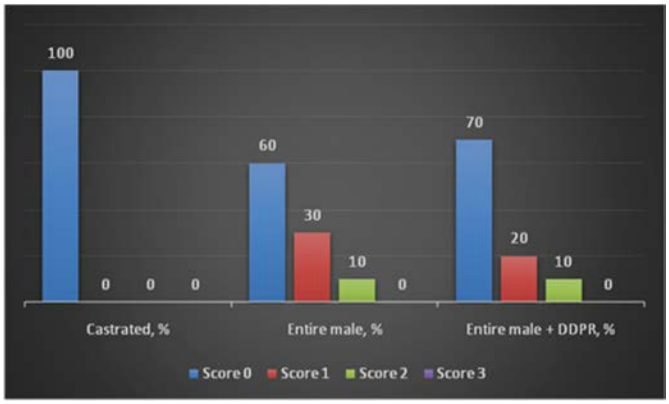

Figure 1. Human nose score for detection of boar taint in the slaughterhouse based on hot knife method with 0 to 3 score (from lack of taint to strong boar taint)

Table 7. Total amounts of saturated, mono- and polyunsaturated fatty acids (%) and lipid nutritional indices of m. LL in castrated, entire male and entire male pigs consuming DDRP

Item	Control group C	Experimental group EM	Experimental group EM+R	SEM	Significance
SFA	40.12	38.71	41.50	3.06	0.177
MUFA	51.45	47.18	44.16	4.63	0.010
PUFA	8.43	14.11	14.34	3.10	0.0005
n-3	0.53[a*]	0.92[b]	0.95[b]	0.25	0.003
n-6	7.90[a]	13.19[b]	13.39[b]	2.93	0.0007
P/S	0.21[a]	0.36[b]	0.35[b]	0.07	0.0006

*Notice: Values connected with different letters are significantly different (P<0.05)

The addition of distilled rose petals decreased the percentage of the pigs with boar taint determined in the meat. While no boar taint was detected in 60% of the pigs from EM group, this percentage was augmented to 70% in EM+R group consuming rose petals included in the feed. Subsequently, 30% of the EM pigs had score 2 for boar taint, while this percent was lower (20%) in the EM+R group. Also, 10% of both groups of entire males showed slight odour of boar taint, although a strong odour with score 3 was not detected at a whole.

The data obtained from the HPLC measurements show that only single samples were found with detectable high amounts of both androstenone and skatole. There were, however, samples that showed increased content of only one of both boar taint compounds. The mean value in fat of which no boar odour was detected was 0.65 µg/g for androstenone and 0.14 µg/g for skatole (Table 8). In carcasses with a low degree of boar taint, only skatole at a concentration of 0.16 µg/g was detected, and in carcasses with a clear boar odour, high androstenone values (above 0.94 µg/g) detected and skatole (above 0.18 µg/g) or the skatole itself is in high quantities (above 0.28 µg/g). Simple statistics showed an average content of androstenone in EM pigs was higher by 26.72% from EM+R (0.928 µg/g in EM vs. 0.68 µg/g in EM+R). There were significant differences (P<0.05) between the groups C and EM or EM+R for androstenone, but for skatole the statistically significant difference was only between C and EM. For both components in EM and EM+R groups ANOVA resulted in P>0.05. The DDRP did not affect the content of skatole (0.124 µg/g in EM vs. 0.123 µg/g in EM+R). As a whole, there was a slight difference between the values of the two methods of detection of boar taint, probably due to the lack of training of the observers.

Table 8. Concentrations of androstenone and skatole, based on HPLC method of detection of boar taint in castrated, entire male and entire male pigs consuming DDRP

Pig group	Human score boar taint	Androstenone µg/g	Skatole µg/g
Control group C			
1	0	na*	0.04
2	0	na	na
3	0	0.08	0.11
4	0	na	0.03
5	0	0.06	0.06
6	0	na	0.09
7	0	na	0.07
8	0	na	na
9	0	na	na
10	0	0.005	0.02
Average	-	0.015	0.042
Experimental group EM			
11	2	1.38	0.18
12	0	0.48	0.16
13	0	0.65	0.08
14	2	2.12	0.21
15	1	0.74	0.19
16	0	na	0.06
17	0	0.63	na
18	2	1.46	0.22
19	0	0.98	0.14
20	0	0.84	na
Average	-	0.928	0.124
Experimental group EM + R			
21	0	0.78	0.17
22	0	0.62	0.05
23	1	0.91	0.28
24	0	0.35	0.09
25	0	0.68	na
26	0	na	0.08
27	0	0.63	0.11
28	2	0.94	0.21
29	2	1.24	0.18
30	0	0.65	0.06
Average	-	0.680	0.123

*Notice: na – below limit of detection

DISCUSSION

The results of the study showed that the differences in the carcass and meat quality traits were attributed to the sex of the animals rather than the addition of the distilled rose petals in the diet. The fat deposition and subsequently the carcass fatness is one of the key issues in most pig breeding programs, since it indirectly affects the feeding efficiency and determines the carcass value and consumers' acceptance of pork (Fontanesi et al., 2012). The thickness of the backfat and the content of lean meat in this study were affected mainly by castration and not by the addition of rose petals in the diet of the pigs. The entire males from both groups presented lower backfat thickness, when compared to the castrated pigs, which has been demonstrated in other studies (Bahelka et al., 2015, Batorek et al., 2012, Škrlep et al., 2019a). On the other hand, Teye (2009) did not observe any difference between castrated and entire male pigs in regard to this trait in Meishan x Large white pigs, but the research was done with hybrid pigs from completely different origin. The thinner backfat of the intact males in this study was associated with higher percentage of lean meat, showing advantage of the latter in the lean meat deposition when compared to the castrates. Such results have been reported in meta-analytical studies by Batorek et al. (2012), Pauly et al. (2012), Trefan et al. (2013) and Poulsen Hautrup et al. (2018). The higher lean meat content and less fat deposition of entire males compared to castrates are due to differences in their energy and nutrient metabolism (Pauly et al., 2008), triggered by the anabolic effect of the gonadal steroids. Both groups of intact males had lower pH 45 min measured in m. LL in comparison to the castrated animals. The values were within the range of 5.87-5.93 indicating no presence of PSE meat (Velazco, 2001). The rate of the pH decline was not affected by the sex or treatment and the values of the pH 24 varied within 5.44-5.46. Similar values were reported by Škrlep et al. (2012, 2020) for entire males and surgically castrated pigs. The values of pH 24 corresponded to relatively higher values of L* the muscles of the three groups (51.01-53.84). Significant differences were detected for b* and C* parameters in the LL muscle. Both b* and C* were higher in EM and EM+R groups, however EM+R presented the highest values when compared to the C group. Also, the value of the a* tended to be higher in the entire males receiving rose petals. It could be suggested that in addition to the sex, bioactive compounds of the phytonutrients might affect the colour of the meat. In a previous study, Vlahova-Vangelova et al. (2020) observed changes in the colour of pig muscles as a result of addition of two doses of distilled rose petals residue to the feed. The authors reported higher b* values in m. LD and m. SM, measured 24 h post mortem in pigs supplemented with distilled rose petals which is in line with our results. However, they observed higher values of L* but lower a* in the supplemented animals. The higher a* and b* parameters could be due to the accumulation of carotenoids in the muscle and the antioxidant activity of the rose supplement that could protect myoglobin from oxidation. According to Dragoev et al. (2020), while assessing the by-products from rose oil production for application in animal nutrition, thirteen glycosides of kaempferol, ten glycosides of quercetin, six glycosides of gallic acid and the two flavonol aglycones have been identified in dry rose petals. These polyphenols possess significant antioxidant activity.

The chemical composition of m. LL was characterized by higher IMF content in the castrated pigs in comparison to both groups of entire males. This corresponded to the higher lean meat percentage reported for EM group. Higher IMF content attributed to castration has been reported also by Gispert et al. (2010), Trefan et al. (2013) and Škrlep et al. (2019b, 2020). Intramuscular fat is an important quality trait, closely associated with mat sensory qualities such as flavour, tenderness and juiciness. The recommended values for optimum sensory quality of pork differ substantially from 1% (Wood, 1990) to 2.0-4.0% in USA (Meisinger, 2002). According to Fortin et al. (2005), the minimum IMF level, necessary to ensure pleasing eating experience is 1.5%. In our study, the levels of IMF in the entire males varied between 1.54 -1.84%, indicating IMF content suitable for consumers who are looking for healthier low fat meat as well as no negative effect on the sensory traits. It was due to the origin of the

pigs, the Danube white breed, which is characterized by good quality of the meat, staying for a longer period well preserved as a national genetic pool.

The fatty acid profile of meat is of crucial importance for its healthy quality. The differences in the fatty acid profile of m. LL observed in this study were mostly attributed to the sex of the pigs. Both groups of entire males displayed decreased amounts of MUFA determined by the lower C18:1 n-9 proportion. The lower content of the latter corresponded with the diminished IMF in EM and EM+R pigs. On the other hand, SFA remained unaffected in the three groups, except with the tendency towards lower content of C18:0 in EM and EM+R. It is known that extremely high amounts of SFA (primarily fatty acids C12:0, C14:0, and C16:0), may contribute to heart disease by raising plasma low density lipoproteins (Fernandez et al., 2005, Cutrignelli et al., 2008). In this study no significant changes were observed in the content of C14:0 and C16:0, indicating no negative effect of sex or treatment for the healthy value of pork.

The proportion of n-3 and n-6 fatty acids were substantially increased in the entire males due mainly to the higher content of C18:2 and C18:3, although no differences in feed intake were observed. The trend towards elevated contents of these two fatty acids that could be noticed in EM+R group might be attributed to the indirect influence of rose petals. For example, polyphenols of *Rosa damascena* have been indicated to interfere with lipid metabolism through inhibiting the activities of pancreatic lipase and HMG COA reductase (Cholamhoseinian et al., 2012, 2010). It can be seen that fatty acid profile of EM+R pigs presents considerable amounts of C18:2n-6 and C18:3n-3 (Table 6.). Their changes were associated with significantly increased amounts of C20:2, C20:3, C20:4, C22:5, as well as trends toward higher contents of C20:5 and C22:6. Similar effect of sex on the fatty acid profile was reported by Cai et al. (2010), Grela et al. (2013) and in meta-analytical research of Pauly et al. (2012). Although the high content of PUFA is undesirable because of their adverse effect on the storage stability and texture in the processed meat products, their health benefits, especially those of n-3 PUFA have been described in numerous studies (Siriwardhana et al., 2012; Veselinović et al., 2017; Kones and Rumana, 2017).

Pork normally contains high levels of C18:2n-6 which is associated with unfavourably high n-6/n-3 ratio. Levels of n-6/n-3 are inversely related to the risk of cancer (Luu et al., 2018) and cardiovascular diseases (Okuyama et al., 2000). According to Wood et al. (2003), n-6/n-3 ratio should not exceed 4, however, in this study, it varies within 14.75-15.14. Furthermore, the P/S ratio should be higher than 0.4, however the values observed in this study were within the range of 0.21-0.36, indicating that the effect of sex and treatment including rose petals are not enough to manipulate the fatty acid profile of pork in regard to favourable values of n-6/n-3 and P/S. The increased amount of PUFA in the entire males that we observed was not sufficient to affect the atherogenic and thrombogenic indices. Their values were lower from those reported by Caldara et al. (2018), who similar to us did not observe any difference in these traits attributed to the sex of the animals.

The presence of the distilled rose petals in the diet of the pigs led to decrease in the boar taint. It could be suggested that the bioactive compounds in the DDRP, especially flavonoids, affect the liver enzymatic system reducing androstenone and skatole. Several studies demonstrated that dietary flavonoids are potent inhibitors of cytochrome P450 enzymes (CYP450) and membrane transporters (Wahajuddin et al., 2013; Mustapić et al., 2018; Bojić et al., 2019). Further in an in vitro study of Ekstrand et al. (2015), it was revealed that selected flavonoids, particularly myricetin, isorhamnetin, and quercetin may affect the activities of porcine CYP1A, CYP3A, and CYP2E1. However, this mechanism of action requires further elucidation.

CONCLUSIONS

The results of the study showed that most of the meat quality traits were affected by the sex of the pigs, rather than the added phytonutrients in the diet. The entire

males presented lower backfat content, intramuscular fat content and higher lean meat percentage in LL muscle, compared to the control group. The fatty acid profile of LL muscle in the intact males was affected by the addition of DDRP showing increase in the n-6 and n-3 polyunsaturated fatty acids proportion and especially the content of alpha-linolenic acid, in comparison to castrated males. The addition of the distilled rose petals in the diet affected the meat colour in the direction of increasing the intensity of meat colouring. Furthermore, the boar taint detected in the meat showed a trend of decreasing in the group consuming dry distilled rose petals. For this reason, more research about the effect of rose petals on meat quality and boar taint are desirable.

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CONFLICTS OF INTEREST

The authors declare no conflict of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, or in the decision to publish the results.

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