MEAT QUALITY, CHEMICAL AND FATTY ACIDS COMPOSITION AND OXIDATIVE STABILITY OF PORK FROM ENTIRE MALES, SURGICAL CASTRATES AND GILTS AFTER BETAINE SUPPLEMENTATION TO DIET

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Abstract: This study was conducted to assess the effect of sex and betaine supplemented diet on chemical composition, cholesterol content, meat quality, fatty acids composition and oxidative stability of pork from entire males, surgical castrates and gilts. A total of forty-two pigs – (entire males - EM, surgical castrates – SC, and gilts - G, each of 14) progeny of Landrace sows and Hampshire x Pietrain boars were involved in the trial. Pigs were allocated to the control and experimental groups (each of 21 pigs – 7 EM, 7 SC and 7 G). Control pigs received standard diet without any supplement whereas experimental ones were fed the same diet with supplement of betaine (1.25 g.kg⁻¹ of feed) for thirty days prior to slaughter. Castrates had significantly higher intramuscular fat and cholesterol content (P<0.05) than entire males and gilts. Also, they had greater content of vaccenic, arachidonic (P<0.05), oleic, eicosanoic, and total monounsaturated fatty acids (P<0.01). Contrary, entire males had the highest level of linolenic, linoleic, total polyunsaturated and n-6 fatty acids (P<0.05). Sex of pigs did not have any effect on meat quality and oxidative stability of pork. Betaine supplementation increased cholesterol content in castrates compared to other two sexes (P<0.05). Drip loss value was reduced in group of entire males (P<0.05) and oxidative stability of muscle was improved in all three groups (P<0.05). Fatty acids profile was not influenced by betaine treatment. Interactions between sex and betaine supplementation were observed for cholesterol concentration, drip loss value, oleic, linolenic, total polyunsaturated and n-6 fatty acids as well as oxidative stability after 30 and 120 min. of incubation.

Key words: betaine, entire males, pork quality, fatty acids, oxidative capacity
Introduction

Surgical castration of piglets has been widespread procedure performed routinely in almost all European Union member states. The main reason for implementing this practice is to avoid the unpleasant odour so-called boar taint arising from adolescence of uncastrated males and releasing in the heat treatment of their meat (Bonneau, 1998). Recently, growing demands of animal rights organizations as well as public initiatives for livestock welfare put considerable pressure on union’s governing authorities towards to stopping the surgical castration of pigs in the EU after 2020. Therefore, rearing entire males might become the main practice in EU pig husbandry. It is well-known that entire males have several advantages compared to castrates, such as faster growth, better feed conversion, and higher lean meat content in the carcass. However, some studies have suggested several problems regarding meat quality of entire males, other than boar taint – higher incidence of DFD (dry-firm-dark) meat (EFSA, 2004), worse toughness (Pauly et al., 2008), lower ultimate pH (Aluwé et al., 2013), less intramuscular fat content (Skrlep et al., 2010, 2012), softer fat due to higher proportion of unsaturated fatty acids (Pauly et al., 2009), higher drip loss (Aluwé et al., 2013).

Sex differences in meat quality, fat content and fatty acid composition of fat tissue are well-known in the literature. Generally, entire males have lower fat content compared to castrates and females based on different hormonal status. Differences in fatty acid composition between entire males, castrates and gilts are mainly related to changes in the rate of subcutaneous fat accumulation in the carcass (Wood et al., 2008; Pauly et al., 2009; Grela et al., 2013; Mackay et al., 2013). Meta-analysis of Pauly et al. (2012) describing studies from 1990 until 2010 confirmed that entire males have a greater amount of polyunsaturated fatty acids (PUFA) than castrates but not gilts. They also have a lower content of saturated (SFA) and similar content of monounsaturated (MUFA) fatty acids, higher PUFA/SFA and lower n-6/n-3 ratios compared to castrates (Pauly et al., 2009; Grela et al., 2013). At the same time, analyses showed that entire males have a higher capacity for protein deposition in the body and higher turn-over (Batorek et al., 2012; Pauly et al., 2012; Trefan et al., 2013). Fat deposition and fatty acid profile in pigs are affected not only by hormones but also by genetics (Canovas et al., 2009; Cho et al., 2011), which results in different enzymes activity (Doran et al., 2006; Missotten et al., 2009; Mackay et al., 2013), dietary energy and protein intake (Wood et al., 2004), and dietary fat composition as well (Missotten et al., 2009; Benz et al., 2011).

One of the possibilities to influence pork quality, its composition and other parameters is the addition of various supplements (vitamins, minerals, trace elements, creatine, betaine, etc.) to the pigs’ nutrition (Lauridsen et al., 1999; Apple et al., 2001; Swigert et al., 2004; Lahučký et al., 2007; Su et al., 2013;
Betaine is known as a product of choline degradation. It occurs naturally in many tissues where it plays a role in various metabolic processes as a methyl donor (e.g. formation of methionine). Other studies have suggested that betaine may influence the efficiency of energy utilization in pigs. Some reports have shown positive effect of dietary betaine supplementation on pork quality such as pH (Matthews et al., 2001a, 2001b; Hur et al., 2007), colour (Yang et al., 2009; Su et al., 2013); whereas other studies have shown no effect on sensory properties (Øverland et al., 1999), subjective marbling and firmness-wetness, or even though, negative effect on subjective color of the loin muscle (Matthews et al., 1998). Recently, some studies reported positive impact of dietary betaine supplementation on reduction of fat deposition in pig carcasses (Huang et al., 2006, 2008; Sales, 2011). This decrease is associated with the increasing rate of lipolysis and decreasing rate of lipogenesis. Both of these biochemical processes are regulated by many enzymes (Huang et al., 2006, 2009).

Since there is a lot of studies in the literature related to the performance, pork quality and fat composition of castrated or female pigs but less of uncastrated males, the aim of this study was to evaluate the effect of betaine supplementation on chemical composition, meat quality, fatty acid profile and oxidative capacity of pork in entire males and to compare it to the surgical castrates and gilts.

**Materials and Methods**

Totally forty-two pigs (entire males - EM, surgical castrates – SC, and gilts - G, each of 14) – crosses between Landrace sows and Hampshire x Pietrain boars were involved in the experiment. Animals were housed in the pens at experimental farm of Research Institute for Animal Production (RIAP). They were located in test station at 20 – 25 kg live weight for timely adaptation to the new space and feed. Experiment started at 30 kg of live weight (age of 80 ± 7 days). Pigs were divided to the control and experimental groups (each of 21 pigs – 7 EM, 7 SC and 7 G). Control group received the standard diet (Table 1) without any supplement. Experimental group was fed standard diet with the same composition as in control group but with supplement of betaine (1.25 g.kg⁻¹ of feed) for thirty days prior to slaughter. Pigs were allowed free access to drinking water during whole experiment. Feed was provided on *ad libitum* basis. Pigs were regularly weighed in weekly intervals.
Table 1. Composition and nutritive value of diets

| Ingredients, % | Control | Betaine | Chemical analysis | Control | Betaine |
|---------------|---------|---------|-------------------|---------|---------|
| Barley        | 42.7    | 42.7    | Dry matter, %     | 86.30   | 86.30   |
| Wheat         | 21.0    | 21.0    | Crude protein, %  | 16.84   | 16.84   |
| Oat           | 15.0    | 15.0    | Crude fat, %      | 2.43    | 2.43    |
| Soybean meal  | 12.0    | 12.0    | Crude fibre, %    | 4.86    | 4.86    |
| Wheat brans   | 2.0     | 2.0     | Ash, %            | 4.56    | 4.56    |
| Meat and bone meal | 2.0     | 2.0     | N-free extract    | 57.68   | 57.68   |
| Fodder yeast  | 1.7     | 1.7     | Metabolizable energy (MJ) | 12.31 | 12.31 |
| Mineral supplement | 2.5   | 2.5   | Lysine, g.kg⁻¹     | 8.64    | 8.64    |
| Biofactor supplement | 0.6 | 0.6 |                         |         |         |
| Fodder salt   | 0.5     | 0.5     |                           |         |         |
| Betaine       | -       | 0.125   |                           |         |         |

*Composition declared by producer of feed mixture

The experiment was performed in accordance with Act on animal veterinary care No. 39/2007 of Slovak Republic and approved by Animal Care Committee of the Research Institute for Animal Production. Animals were slaughtered at 110 ± 5 kg live weight (age of 174 ± 11 days) at the experimental slaughterhouse of RIAP by electrical stunning (90 - 100 V, 0.9 - 1.0 A, 50 Hz) followed by exsanguination. Evisceration was completed about 20 min post mortem. Chilling of the carcasses (air temperature 2 - 4 °C, velocity 0.5 – 1.0 m.s⁻¹) started approximately 60 min after slaughter and was continued overnight. After 24-hours chilling of carcasses at 4 °C, the *longissimus dorsi* muscle (150 g of sample) was removed from the right side of carcass and sliced into chops 2.5 cm thick for further meat quality (colour, drip loss) analysis. One wrapped sample was stored in the dark for 5 days at 4 °C for the shear force and colour analysis. Forty-five minutes and 24 h after slaughter, pH values were measured in the loin muscle (*musculus longissimus dorsi* - LD) from the right side of carcasses between 13th and 14th rib using device METTLER TOLEDO (pH meter FiveGo™, Columbus, USA) with combined electrode. Colour was measured by spectrophotometer MINISCAN XE Plus (Hunter Associates Laboratory, Inc., Reston, USA). Drip loss was assessed 24 h after slaughter by Honikel method (*Honikel, 1998*). Four days after slaughter, the shear force was measured using TEXTURE ANALYSER TA-XT2i device (Stable Micro Systems Ltd, Surrey, UK). Approximately, thirty min. post mortem, *longissimus dorsi* muscle samples (100 g) between 13th and 14th rib were taken, wrapped into aluminium foils and imposed in liquid nitrogen for 5 days for oxidative stability analysis. The oxidative capacity of *longissimus muscle* homogenate was determined as thiobarbituric acid reactive substances (TBARS) according to Küchenmeister *et al.* (1999). TBARS values express as equivalents of malondialdehyde (MDA, nM.mg⁻¹ homogenate protein) which is breakdown product formed during peroxidation of lipids stimulated by Fe²⁺/ascorbate. To
stimulate lipid peroxidation, 3 ml of muscle homogenate was incubated in 0.1 mM ascorbate and 5 mM FeSO₄. From this, 0.5 ml was immediately removed and pipetted into 0.25 ml of 20% trichloroacetic acid in 100 mM KCl. The remaining homogenate was placed in a water bath at 37 °C and after 30, 60, and 120 min, 0.5 ml each of this incubated homogenate were transferred into the trichloroacetic acid. Then samples were centrifuged at 10 000 g for 10 min and 0.5 ml of the supernatants were mixed with 0.5 ml of thiobarbituric acid (0.67%) and boiled for 15 min in a water bath. The absorbance at 535 nm was determined immediately after cooling. All samples determined for chemical and fatty acid compositions as well as cholesterol determination were transported to the Chemical laboratory of the Slovak Agricultural University. Each sample (50 g) was homogenized and subsequently analysed by the Fourier Transform Infrared (FTIR) method (Carbonaro and Nucara, 2010) using the device Nicolet 6700 (IET Ltd., Illinois, USA).

Data from the experiment were analysed by a two-way ANOVA with fixed effects of treatment (betaine or none) and sex (entire males, castrates or gilts) as well as corresponding interactions between treatment and sex using procedure GLM of the statistical program SAS-STAT, version 9.1.3 (SAS Institute Inc., Cary, N.C., USA, 2002-2003). Basic statistics was done using MEANS procedure. The model used was:

\[ y_{ijk} = \mu + B_i + D_j + B*D_{ij} + e_{ijk} \]

where \( y_{ijk} \) – characteristic of trait selected, \( \mu \) – intercept, \( B_i \) – effect of sex (i = EM, SC, G), \( D_j \) – effect of diet (j = C, B), \( B*D_{ij} \) – two-way interaction effect sex x diet, \( e_{ijk} \) – random error (k = 1......nᵢⱼ). Data in tables are expressed as Least Square Means (LSM) ± standard error of the mean (SEM). Comparisons between groups were done by Scheffe’s test and differences were considered to be statistically significant at the level of \( P<0.05 \).

**Results and Discussion**

Sex of pigs had significant effect on IMF and cholesterol content. Both of these parameters were the highest (\( P<0.05 \)) in surgical castrates compared to entire males and gilts (Table 2). The same results regarding IMF have been reported in other studies (Škrlep et al., 2010; Gispert et al., 2010). Differences in IMF and cholesterol concentrations are associated with hormonal activity of sex steroids which influence lipid synthesis and metabolism. Surgical castration eliminates the effect of these steroids resulting in higher fatness of carcass including higher content of IMF. In general, betaine supplementation increased cholesterol content in experimental pigs compared to the control. This increase was due to significant enhancement in group of betaine supplemented surgical castrates which is showed
by interaction of sex and diet (Table 6), whereas entire males and gilts of control or supplemented groups had similar levels of cholesterol. This is in agreement with findings of Albuquerque et al. (2017) and Li et al. (2017) reporting increased cholesterol concentration in castrates or gilts, respectively. Matthews et al. (2001b) found higher plasma cholesterol concentration after 0.125% of betaine supplementation to the pigs' diet. Martins et al. (2010) reported increased cholesterol content in subcutaneous dorsal fat deposition but not in the musculus semimembranosus. Authors suggested that betaine affects cholesterol metabolism in the liver by stimulating lipid mobilisation and enhancing hepatic lipoprotein secretion.

Table 2. Effects of sex and betaine treatment on chemical composition and cholesterol content of longissimus dorsi muscle

| Trait                  | Sex (LSM) | Signif. | Diet (LSM) | Signif. | SEM | Sex x B |
|------------------------|-----------|---------|------------|---------|-----|---------|
| Total water, %         |           |         |            |         |     |         |
| EM                     | 74.4      | ns      | 74.1       | ns      | 0.3 | ns      |
| SC                     | 73.8      |         | 74.2       |         |     |         |
| G                      | 74.3      |         |            |         |     |         |
| Total protein, %       |           |         |            |         |     |         |
| EM                     | 21.6      | ns      | 21.7       | ns      | 0.2 | ns      |
| SC                     | 21.5      |         | 21.6       |         |     |         |
| G                      | 21.8      |         |            |         |     |         |
| Intramuscular fat, %   |           |         |            |         |     |         |
| EM                     | 2.6a      | *       | 3.0        | ns      | 0.2 | ns      |
| SC                     | 3.5b      |         | 3.1        |         |     |         |
| G                      | 2.8a      |         |            |         |     |         |
| Cholesterol, %         |           |         |            |         |     |         |
| EM                     | 0.38a     |         | 0.41a      | *       | 0.2 | *       |
| SC                     | 0.58b     |         | 0.51b      |         |     |         |
| G                      | 0.36a     |         |            |         |     |         |

a,b Different letters within row mean significant differences at * = P<0.05, EM – entire males, SC – surgical castrates, G – gilts, C – control group, B – betaine supplemented group, LSM – least square mean, SEM – standard error of means, ns = not significant (P≥0.05)

Sex did not affected quality traits of longissimus dorsi muscle. There was only tendency (P = 0.054) of paler meat of entire males compared to castrates measured 24 h after slaughter but not after 5 days (Table 3).

Table 3. Effects of sex and betaine treatment on pork quality

| Trait               | Sex (LSM) | Signif. | Diet (LSM) | Signif. | SEM | Sex x B |
|---------------------|-----------|---------|------------|---------|-----|---------|
| pH45                |           |         |            |         |     |         |
| EM                  | 6.36      | ns      | 6.42       | ns      | 0.28| ns      |
| SC                  | 6.42      |         | 6.29       |         |     |         |
| G                   | 6.33      |         |            |         |     |         |
| pH24                |           |         |            |         |     |         |
| EM                  | 5.72      | ns      | 5.68       | ns      | 0.08| ns      |
| SC                  | 5.59      |         | 5.52       |         |     |         |
| G                   | 5.61      |         |            |         |     |         |
| Colour L’           |           |         |            |         |     |         |
| EM                  | 50.68a    | 0.054   | 49.97      | ns      | 0.69| ns      |
| SC                  | 49.81b    |         | 49.37      |         |     |         |
| G                   | 50.03     |         |            |         |     |         |
| Colour a’           |           |         |            |         |     |         |
| EM                  | 1.75      | ns      | 1.86       | ns      | 0.16| ns      |
| SC                  | 1.86      |         | 1.94       |         |     |         |
| G                   | 1.85      |         |            |         |     |         |
| Colour b’           |           |         |            |         |     |         |
| EM                  | 7.66      | ns      | 7.75       | ns      | 0.25| ns      |
| SC                  | 7.86      |         | 7.78       |         |     |         |
| G                   | 7.84      |         |            |         |     |         |
| Colour L’           |           |         |            |         |     |         |
| EM                  | 51.67     | ns      | 51.56      | ns      | 0.72| ns      |
| SC                  | 52.06     |         | 51.37      |         |     |         |
| G                   | 51.82     |         |            |         |     |         |
| Colour a’           |           |         |            |         |     |         |
| EM                  | 2.76      | ns      | 2.65       | ns      | 0.32| ns      |
| SC                  | 2.70      |         | 2.61       |         |     |         |
| G                   | 2.70      |         |            |         |     |         |
| Colour b’           |           |         |            |         |     |         |
| EM                  | 8.28      | ns      | 8.48       | ns      | 0.31| ns      |
| SC                  | 8.47      |         | 8.53       |         |     |         |
| G                   | 8.38      |         |            |         |     |         |
| Drip loss, %         |           |         |            |         |     |         |
| EM                  | 4.11      | ns      | 4.21a      | *       | 0.33| *       |
| SC                  | 3.87      |         | 3.83b      |         |     |         |
| G                   | 4.32      |         |            |         |     |         |
| Shear force, kg      |           |         |            |         |     |         |
| EM                  | 5.36      | ns      | 5.20       | ns      | 0.25| ns      |
| SC                  | 5.07      |         | 4.88       |         |     |         |
| G                   | 5.08      |         |            |         |     |         |

a,b Different letters within row mean significant differences at * = P<0.05, EM – entire males, SC – surgical castrates, G – gilts, C – control group, B – betaine supplemented group, LSM – least square
Betaine supplementation in present study had no significant effect on pH (45 min and 24 h), colour (24 h and 5 days) and shear force values. Some other studies reported a higher initial or ultimate pH after betaine supplementation (Matthews et al., 2001a, 2001b; Hur et al., 2007). This increase of pH may indicate that rate of pH decline after slaughter is slower in pigs fed betaine supplement. As known, meat pH is closely related to meat colour. This trait was not also affected by betaine supplementation. Similar results considering pork colour were reported by other authors (Matthews et al., 2001a; Feng et al., 2006). Positive effect of betaine on meat colour was found by Yang et al. (2009). In contrary, some studies reported reducing colour darkness (Matthews et al., 2001b). These discrepancies in experiments may be due to different genotypes and muscle types of used pigs. Betaine supplementation in our study significantly (P<0.01) decreased drip loss. Again, it was due to significant lowering only in one sex – entire males, which is presented by interaction between sex and betaine (Table 6). Similar results - decreasing by 11% after betaine administration – were found by Matthews et al. (2001a). Possible effect of betaine can be explained by stabilisation of cell membranes against fluid loss from muscle.

Sex of pigs had considerable effect on several fatty acids concentrations in our study. Castrates had higher content of oleic and total MUFA than entire males (P<0.01); vaccenic and arachidonic acids than gilts (P<0.05); and eicosenoic acid (P<0.01) than both, entire males and gilts (Tab 4). On the other hand, entire males had higher content of linolenic acid than castrates (P<0.05); linoleic, total PUFA and n-6 fatty acids (P<0.05) than both, castrates and gilts. The same results in regard of oleic and linolenic acids were observed in study of Grela et al. (2013). Cai et al. (2010) also reported higher concentrations of PUFA in meat of entire males compared to castrates. Greater amount of PUFA and lower SFA in entire males compared to castrates (or gilts) have been reported in other studies (Jaturasitha et al., 2006; Pauly et al., 2009, 2012). These differences depend on different fatty acid composition of two basic lipid fractions – phospholipids, which are part of cell membranes (relatively constant with increasing fatness of body), and neutral lipids (triacylglycerols), which are accumulated in fat depots and their proportion increases with increasing body fatness (De Smet et al., 2004; Wood et al., 2008). Content of PUFA is higher in phospholipids (35-48%) than in triacylglycerols (5-14%) (Cameron et al., 2000) and increases slower than SFA and MUFA (contained in neutral lipids) during growth and fattening, which results in lower relative proportion of PUFA in pig body (Riley et al., 2000). Generally, carcass fatness/leanness has been found to be strongly associated with fatty acid composition. Increased backfat thickness has negative relationship to the degree of unsaturation (Wood et al., 2008). Recently, research in this area showed that also
other factors such as genes (Gunawan et al., 2013), sex steroids and enzymes involving in lipid synthesis and metabolism (Hallenstvedt et al., 2012; Corominas et al., 2013; Mackay et al., 2013), and nutrition (Missotten et al., 2009; Benz et al., 2011) as well, may all affect fat deposition and fatty acid composition of pig muscles and adipose tissue. Supplementation of betaine in the presented study had no effect on fatty acids profile of longissimus dorsi muscle. The same result was reported in the study of Madeira et al. (2015). Albuquerque et al. (2017) also did not find any influence of betaine on SFA, MUFA, PUFA and n-6/n-3 and PUFA/SFA ratios, as well. In contrary, study of Madeira et al. (2016) found decreasing concentrations of vaccenic, palmitoleic and total MUFA after betaine addition to the diet of entire male pigs. Interactions between sex and betaine in present study was observed for oleic, linolenic, total PUFA and n-6 fatty acids (Table 6). Differences within sex for control and supplemented groups were small and insignificant (P>0.05).

Table 4. Effects of sex and betaine supplementation on fatty acid composition of intramuscular fat (% of total fatty acids) in the longissimus dorsi muscle

| Trait          | Sex (LSM) | Signif. | Diet (LSM) | Signif. | SEM | Sex x B |
|----------------|-----------|---------|------------|---------|-----|---------|
|                | EM        | SC      | G          | C       | B   | SEM     | Sex x B |
| Myristic C14:0 | 1.31      | 1.28    | 1.26       | ns      | 1.28| 1.28    | ns       |
| Palmitic C16:0 | 24.50     | 24.57   | 24.46      | ns      | 24.51| 24.54   | 0.04     |
| Steric C18:0   | 11.30     | 11.28   | 11.31      | ns      | 11.29| 11.31   | 0.07     |
| Total SFA      | 39.39     | 39.42   | 39.40      | ns      | 39.41| 39.40   | 0.38     |
| Oleic C18:1n-9 | 44.66a    | 47.42b  | 45.80ab    | **      | 46.07| 45.98   | ns       |
| Eicosenoic C20:1 | 0.61a     | 0.69b   | 0.60ab     | **      | 0.64| 0.66    | ns       |
| Vaccenic C18:1t11 | 4.42ab   | 4.50a   | 4.39b      | *       | 4.45| 4.46    | ns       |
| Total MUFA     | 49.71a    | 52.63b  | 51.26ab    | **      | 51.32| 51.58   | 0.86     |
| Arachidon C20:4n-6 | 1.38ab   | 1.46a   | 1.34b      | *       | 1.40| 1.41    | ns       |
| Linolenic C18:3n-3 | 2.07a     | 1.92b   | 2.05a      | *       | 1.97| 2.01    | 0.03     |
| Linoleic C18:2n-6 | 9.36a    | 8.33b   | 8.60b      | *       | 8.89| 8.96    | 0.69     |
| Total PUFA     | 14.83a    | 13.35b  | 13.49b     | *       | 13.81| 13.72   | 0.78     |
| n-3 FA         | 2.95      | 2.88    | 2.85       | ns      | 2.91| 2.89    | 0.12     |
| n-6 FA         | 11.88a    | 10.47b  | 10.65b     | *       | 11.08| 11.14   | 0.56     |
| n-6/n-3        | 4.05      | 3.64    | 3.76       | ns      | 3.86| 3.85    | 0.43     |
| PUFA/SFA       | 0.37      | 0.34    | 0.34       | ns      | 0.34| 0.35    | 0.13     |

a,b Different letters within row mean significant differences at * = P<0.05, ** = P<0.01, EM – entire males, SC – surgical castrates, G – gilts, C – control group, B – betaine supplemented group, LSM – least square mean, SEM – standard error of means, SFA – saturated fatty acids, MUFA – monounsaturated fatty acids, PUFA – polyunsaturated fatty acids, ns = not significant (P≥0.05)

An oxidative effect of sex and betaine supplementation is shown in Table 5. Sex of pigs did not affect oxidative stability of pigs muscle. As known, higher content of PUFA and lower of SFA (expressed as PUFA/SFA ratio) is beneficial from the human health point of view. On the other hand, fat containing a higher
amount of PUFA is softer and more liable to oxidative deterioration. Results suggest significant reducing of peroxidation – lower values of MDA production after incubation with Fe²⁺/ascorbate mixture – in pigs fed betaine. This result is in accordance with the study of Su et al. (2013). Tendency of reducing the lipid oxidation (insignificant differences) in pork stored for 13 days were only outlined in the study of Hur et al. (2007). Significant interactions between sex and diet were found after 30 and 120 min incubation in the all three groups - entire males, gilts (P<0.05) and castrates (P<0.05, 0.01 resp.). So, positive effect of betaine supplementation on stabilization of cell membranes against oxidation was manifested in whole set as well as within each sex of pigs.

Table 5. Effects of sex and betaine supplementation on oxidative stability of longissimus dorsi muscle

| Trait          | EM (LSM) | SC (LSM) | G (LSM) | Signif. | Diet (LSM) | Signif. | SEM | Sex x B |
|----------------|----------|----------|---------|---------|------------|---------|-----|---------|
| TBARS (0 min)  | 0.06     | 0.05     | 0.06    | ns      | 0.06       | 0.06    | ns  | ns      |
| TBARS (30 min) | 0.24     | 0.23     | 0.25    | ns      | 0.28       | 0.21    | *   | 0.02    |
| TBARS (60 min) | 0.30     | 0.32     | 0.29    | ns      | 0.35       | 0.27    | *   | 0.02    |
| TBARS (120 min)| 0.38     | 0.39     | 0.36    | ns      | 0.43       | 0.30    | *   | 0.02    |

a,b Different letters within row mean significant differences at * = P<0.05, ** = P<0.01, EM – entire males, SC – surgical castrates, G – gilts, C – control group, B – betaine supplemented group, LSM – least square mean, SEM – standard error of means, TBARS - thiobarbituric acid reactive substances (nmol/mg protein), ns = not significant (P≥0.05)

Table 6. Interactive effect of sex and betaine supplementation on cholesterol content, fatty acids and oxidative stability of longissimus dorsi muscle

| Trait          | EM (LSM) | SC (LSM) | G (LSM) | SEM |
|----------------|----------|----------|---------|-----|
| Cholesterol, % | 0.36     | 0.39     | 0.46    | 0.38 |
| Drip loss, %   | 4.48     | 3.65     | 3.95    | 4.38 |
| Oleic C18:1n-9 | 44.61    | 44.68    | 47.46   | 45.83|
| Linolenic C18:3n-3 | 2.06     | 2.08     | 1.90    | 2.06 |
| Total PUFA     | 14.76    | 14.91    | 13.44   | 13.53|
| n-6 FA         | 11.39    | 11.99    | 10.39   | 10.72|
| TBARS (30 min) | 0.28     | 0.21     | 0.27    | 0.28 |
| TBARS (120 min)| 0.42     | 0.32     | 0.45    | 0.42 |

a,b Different letters with row mean significant differences at P<0.05, A,B Different letters with row means significant differences at P<0.01, EM – entire males, SC – surgical castrates, G – gilts, C – control group, B – betaine supplemented group, LSM – least square mean, SEM – standard error of means, SFA – saturated fatty acids, MUFA – monounsaturated fatty acids, PUFA – polyunsaturated fatty acids, TBARS - thiobarbituric acid reactive substances, ns = not significant (P≥0.05)
Conclusion

In conclusion, sex of pigs had significant effect on IMF and cholesterol content as well as some monounsaturated and polyunsaturated fatty acids. Surgical castrates had highest IMF and cholesterol concentrations. Their IMF was formed significantly in greater amount of MUFA compared to gilts and entire males which had highest content of linoleic, linolenic, total PUFA and n-6 fatty acids. From the healthy human nutrition perspective, higher proportion of linolenic and total PUFA as well as less IMF and cholesterol in meat from entire males could be beneficial after an anticipated ban of surgical castration of piglets in the EU countries. However, higher content of n-6 PUFA, especially linoleic acid, could be problematic. Betaine supplementation significantly increased cholesterol content in surgical castrates, decreased drip loss in entire males and improved oxidative stability of pork in all three groups. Lower drip loss and better oxidative stability, especially in entire males, may be beneficial for meat industry, wholesale and retail, however further research is needed to evaluate different doses and time of supplementation of betaine.

Kvalitet mesa, hemijski sastav, profil masnih kiselina i oksidativna stabilnost svinjetine od nekastriranih mužjaka, hirurških kastrata i nazimica nakon dodavanja betaina u ishrani

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Rezime

Ova studija je sprovedena da bi se procenio uticaj pola i dodatka betaina u ishrani na hemijski sastav, sadržaj holesterola, kvalitet mesa, sastav masnih kiselina i oksidativnu stabilnost svinjskog mesa nekastriranih nerastova, hirurških kastrata i nazimica. U ispitivanje je bilo uključeno ukupno četrdeset i dva grla (nekastrirani nerastovi - EM, hirurški kastrati - SC, i nazimice - G, svaki po 14), potomci landras svinja i hempšir x pijetren nerastova. Svinje su raspoređene u kontrolne i eksperimentalne grupe (svaka po 21 grlo - 7 EM, 7 SC i 7 G). Kontrolne svinje su trideset dana pre klanja dobijale standardnu ishranu bez ikakvih dodataka, dok su eksperimentalne hranjene istom hranom sa dodatkom betaina (1,25 g.kg⁻¹ hrane). Kastrati su imali znatno viši sadržaj intramuscularne masti i holesterola (P<0,05) u odnosu na nekastrirane nerastove i nazimice. Takođe, imali su veći sadržaj
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vakcenične, arahidonske (P<0,05), oleinske, eikosanojske i ukupnih mononezasićenih masnih kiselina (P <0,01). Suprotno tome, nekastrirani nerastovi su imali najviši nivo linolenske, linolne, ukupnih polinezasićenih i n-6 masnih kiselina (P<0,05). Pol svinja nije imao uticaj na kvalitet mesa i oksidativnu stabilnost svinjskog mesa. Dodatak betaina povećao je sadržaj holesterola u kastratima u poređenju sa ostala dva pola (P<0,05). Vrednost kala ceđenja smanjena je u grupi nekastriranih nerastova (P<0,05), a oksidaciona stabilnost mišića poboljšana je u sve tri grupe (P<0,05). Tretman betainom nije uticao na profil masnih kiselina. Primećene su interakcije između pola i dodavanja betaina kod koncentracije holesterola, vrednosti kala ceđenja, oleinske, linolenske, ukupnih polinezasićenih i n-6 masnih kiselina, kao i oksidativnu stabilnost posle 30 i 120 min. inkubacije.

Ključne reči: betain, nekastrirani mužjaci, kvalitet mesa svinja, masne kiseline, oksidativni kapacitet

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