Short Note

Fatty acid composition of wild boar (Sus scrofa scrofa) meat compared to commercial hybrid and crossbreed Mora Romagnola swine. A preliminary study

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The increasing population of wild boar has made this wild game species acquire commercial interest. Consumers appreciate this type of meat for its high nutritional value.
In our research, we have compared fat composition of different types of meat. The loin of 9 hunted wild boars, 9 intensively kept commercial swines and 14 crossbreed Mora Romagnola pigs, kept in the open, have been sampled. Moreover, we have collected samples from ham and shoulder of wild boars, to identify which cut could have the most favourable characteristics. All animals came from Bologna District. A.O.A.C. Official Method 996.06 was used to determine lipid composition. Finally, statistical analysis was performed.
According to our results, wild boar meat had a significantly lowest content of fatty acids, highest PFA:SFA ratio and highest content of ω-3 and EPA. Crossbreed Mora Romagnola registered the highest content of fatty acids, a low value of PUFA:SFA ratio, like the hybrid pigs, but the value of ω-6:ω-3 ratio was closer to that of wild boar.
This preliminary research suggests that the wild boar meat has the most favourable fatty acids composition, and that crossbreed Mora Romagnola pigs could be considered in a middle position between wild boar and commercial swine.
Future researches are needed to deepen these aspects.

Keywords: Wild boar, Mora Romagnola, commercial pig, fatty acids, *longissimus* muscle.

1. INTRODUCTION

Over the last 50 years, the wild boar (*Sus scrofa scrofa*) population has increased throughout Italy [1, 2]. The increasing population density of these animals makes them a major agricultural pest. However, the wild boar has also acquired great importance as wild game species [1]. Consumers like wild game meat because of its high nutritional value and special taste. Therefore, wild boar has become an important source of food, and consumer interest in this species has increased [3, 4].
In order to ensure the safety and quality of wild game meat and products, a specific European legislation regulates their processing and marketing. Meat of wild game animals must be handled in approved game handling establishments, which have specific operational and structural requirements, in accordance with Regulation (EC) 178/2002, 852/2004 and 853/2004. These establishments should ensure that animal-by-products are handled and disposed of in accordance with Regulation (EC) 1069/2009. Reg. (EC) 853/2004 and Reg. (EC) 854/2004, define hygienic processing parameters, inspection procedures and training of food business operators and competent authorities. The traditional

domestic preparation of wild game foodstuffs is to cook them slowly (e.g. sauces or stewed) or to cure them. These kinds of preparations significantly reduce the health risks associated with eating this type of meat [5, 6].

In human diet, meat is an important source of nutrients such as proteins, fats, vitamins and minerals. Breeding conditions, the diet and animal genetics are the main determiners of nutritional composition [7].

Regarding fat composition, the diet of the swine influences the fatty acids composition of their adipose and muscle tissues. They are monogastric animals and not ruminants, thus during the digestion process fatty acid double bonds are not hydrogenated [8]. Therefore, saturated (SFA), monounsaturated (MUFA) and polyunsaturated (PUFA) fatty acids, which are present in their diet, deposit in their tissues without any change in fatty acids double bonds [9]. Wild boars are omnivorous and their diet is 90-95% composed of vegetable food (plants, roots, seeds, grains and fruits) and the remaining 5-10% is composed of animals (small mammals and birds, eggs, insects, earthworms, snails, carrions and frogs) [10, 11, 12]. In contrast, domestic pigs have complete formulated diets [7]. Previous studies have shown how in Italy wild boar meat contains more ω-3 than the meat of animals with complete diets [13]. Moreover, wild boar meat is characterised by high content of palmitic (16:0), stearic (18:0), oleic (18:1) and linoleic (18:2) acids. However, oleic and linoleic acids levels change depending on food availability in a certain habitat [14].

In the pre-agricultural human diet, the game species were the principal source of lipids. Modern humans genetically require the same lipid composition of food available in the pre-agricultural age. Dietary intake of essential fatty acids, such as ω-6 and ω-3, is fundamental for good health and the avoidance of chronic diseases because of their pro- and anti-inflammatory actions [15, 16]. However, modern diets are characterised by a high content of ω-6 polyunsaturated fatty acids (PUFA) and a lower content of ω-3, thus the ω-6:ω-3 ratio is very high. Indeed, cereal grains have become a fundamental component of human diet and, over the last fifty years, of domestic animals' diets too. Cereal grains are characterised by a high ω-6 and calories content, but a low ω-3 and antioxidant content as the vegetal oils (sunflower and rapeseed) used in animal feed. Cereals and meat products are the principal components of food in the modern human diet. This fact probably promotes the onset of cancer and cardiovascular, autoimmune, and inflammatory diseases [16].

The aim of this research is to compare the fat composition of wild boar meat with that of two different domestic pig breeds: intensively bred hybrid pigs and pigs crossbred with an Italian indigenous breed (*Mora Romagnola*), kept outdoor and in an organic farm. Mora Romagnola is an autochthonous Italian pig breed that is reared in Emilia Romagna. In 1953 its population reached 22,000 specimens, but with the arrival and spreading of the Large White one it declined, and the Mora Romagnola was close to extinction. In 1998 only 12 related individuals of this breed survived, and thanks to a project of WWF Italy and the University of Torino Mora Romagnola population was recovered and acquired a small production niche [17, 18]. However, this local breed is often crossed with the hybrid swine, in order to improve their productive performances [18]. The choice of including the crossbreed Mora Romagnola swine in the comparison is the real novelty of this study, indeed, to our knowledge, there aren't researches that compare the fatty acids composition of the meat of this local breed with that of wild boar and hybrid pig. Products of Mora Romagnola and its crossbreed have acquired a certain importance between the consumers, especially for salami production, so it is fundamental to give them as much information as possible on the quality of this meat. Moreover, the choice of analysing the fatty acids content of the different wild boar meat cuts can be considered an additional novelty of this research.

2. MATERIALS AND METHODS

2.1 SAMPLE COLLECTION

From October 2010 to March 2019, the carcasses of 9 wild boars (WB), 9 hybrid swines (HS) and 14 crossbreed Mora Romagnola pigs (MR) were sampled, collecting 500-gram samples from the loin, *Longissimus dorsi* muscle, from the shoulder and from the leg of the same animal. After their collection, all samples were immediately frozen at -20°C and maintained at this temperature until their delivery to the laboratory for analysis.

2.2 ANIMALS

The HS group comprised 9 female subjects: 5 aged 11 months and weighing 165-170 kg, and 4 were 15 months old and 200-230 kg. WB (5 females and 4 males) weighed 45-60 kg and they were all about 2 years old. Finally, the MR group counted 13 females and 1 male, aged 15-16 moths and weighing 120-130 kg. All animals came from the Emilia-Romagna region (Italy). WB were hunted in accordance with the National Wild Boar Control Plan and their carcasses were extracted in four different approved game handling establishments. WB were hunted without sex considerations as it usually happens, meanwhile for MR and HS quite all female subjects were selected, because the meat of the males is not appreciated by consumers.

Wild boars consume what they find in the territory, depending also on the different seasons. As previously

specified, wild boars are omnivores and their diet includes nuts, chestnuts, roots, tubers, rhizomes, wild fruits, mushrooms, walnuts, insects and invertebrates, small mammals and animal carcasses too.
The loin samples of the hybrid swine came from slaughterhouses in the Modena District.
Hybrid pigs were intensively bred and fed with a traditional diet, without any PUFA integration. The diet of industrial swine changes in accordance to their production periods. During the fattening period (70 kg – 160 kg), their diet is composed mainly by maize (45%), barley (19%), soft wheat middling (12.5%), soybean meal (10%), soft wheat bran (7%), sunflower meal (3%). During the last production phase, finished swines (140 kg - 170 kg) are fed principally with maize (60%), soybean meal (10.5%), soft wheat bran (10%), soft wheat middling (10%), barley (3.9%) and lard (1.3%).
The fourteen crossbreed Mora Romagnola pigs were raised in semi-wild conditions in two small breeding farms in the traditional fashion. The farmers provide 2-2.5 kilograms of pellet animal feed to animals for the entire year. The animal feed was composed of maize, wheat bran, wheat middling, barley meal, soybean and sunflower seeds cake, calcium carbonate and sodium chloride. Moreover, animals can integrate their own diet through what they found according to the seasons: in spring-summer, it is composed mainly of green grass, insects, earthworms, snails; during the autumn-winter, the integration can be made with acorns and chestnuts. The daily ratio is about 3 kg for adult pigs, but usually farmers increase it during the winter season, the end-phase of the production and for lactating sows. In particular, the finishing is conducted for 30 days in a pen, where the animal feed ratio is increased to 3.5 kilograms.

2.3 ANALYTICAL ANALYSIS

A.O.A.C. Official Method 996.06 "Fat (total, saturated and unsaturated) in food" was used to determine the lipid composition. Samples containing 100-200 mg of fat were hydrolysed with 10 mL HCl 8.3N at 80°C for 40 minutes with ethanol (2 mL) and pyrogallic acid, using triundecanoin ($C_{36}H_{68}O_6$) as internal standard with Vortex mixing every 10 minutes. At the end, the solution was cooled and extracted using a mixture of diethyl ether and light petroleum ether. Then the ether layer was evaporated using nitrogen stream on a steam bath. Derivatization was made at 100°C for 45 minutes in a hermetically closed vial using 2 mL boron trifluoride in 7% methanol and 1 mL toluene. After that 1.0 mL of hexane, 5.0 mL of water and 1.0 g of Na_2SO_4 were added, and the mixture was shaken one minute. After separation of the layers, the organic phase was injected in the gas chromatograph. The chromatographic conditions were as follow:
Supelco SP-2560 column (100 m × 0,25 mm × 0,20 µm)
- Injection volume: 2 µL (split 1:200)
- carrier gas: He 20 cm/sec
- Injector temperature: 250°C
- Detector (FID) temperature: 260°C
- hydrogen flow 40 mL/min., nitrogen flow 30 mL/min., air flow 240 mL/min.
- temperature program: 140°C for 4 min, 240°C at 4 °C/min, 240°C for 15 min

Fatty acids were identified by their relative retention time and quantified using the response factor for each acid and the conversion factor from fatty acid to triglyceride.
In order to obtain total fatty acids, all fatty acids, expressed as triglycerides, were summed. Similarly, SFA, MUFA and PUFA totals were calculated, summing their relative fatty acids. After that, ratios SFA/PUFA and ratio ω-6:ω-3 were considered. In addition, mean and standard deviation of all parameters were calculated for each group of animals.

2.4 STATISTICAL ANALYSIS

Fatty acids were compared as a percentage on the total fatty acids. Percentages are considered normally distributed and the comparison between groups was tested by One Way ANOVA test. When statistically significant differences were detected, a post hoc pairwise comparison across treatments was performed using Tukey's test. The statistical analyses were performed using the computer software program R. version 3.5.2 (Copyright © The R Foundation for Statistical Computing). Significance was established at $p < 0.05$.

3. RESULTS

The fat composition of loin, *Longissimus dorsi*, of wild boar, crossbreed Mora Romagnola and intensively bred hybrid swine were compared. On average, the results (g/100g of sample) showed that MR had the highest content of fatty acids (20.80 ± 6.74), followed by HS (15.87 ± 11.71) and finally by WB meat, which had significantly ($p < 0.01$) less amount of fatty acids (6.80 ± 5.58) than crossbreed Mora Romagnola loin. All three types of meat displayed a high monounsaturated fatty acids (MUFA) concentration. In HS and MR, the polyunsaturated fatty acids (PUFA) concentration was lower than that of saturated fatty acids (SFA). WB loin contained more PUFA than SFA (Fig. 1). Comparing the three groups, the MUFA and SFA concentration of HS and MR was significantly higher ($p < 0.05$) than that of WB meat, while the PUFA content did not differ significantly between the groups (Tab I).
In the samples of this study the PUFA:SFA ratio averaged for WB 1.12 ± 0.38, which is significantly higher ($p < 0.01$) than the values found in HS (0.39 ±

Table I - Mean percentage on total of fatty acids and standard deviation of each fatty acid in *M. Longissimus* muscle of Wild boar, Crossbreed Mora Romagnola and Hybrid pigs.
^{a,b}: Mean values of the same row with common letters do not present difference by statistical analysis (p>0.05). NA = not available.

	Wild boar (n=9)	**Crossbreed Mora Romagnola (n=14)**	**Hybrid pigs (n=9)**
Total fatty acids	6.80 (± 5.58) ^{a}	20.80 (± 6.74) ^{b}	15.87 (± 11.71) ^{a,b}
%ω-3 (g/100g)	0.28 (± 0.28) ^{a}	0.19 (± 0.10) ^{a}	0.14 (± 0.11) ^{a}
%ω-6 (g/100g)	1.85 (± 1.96) ^{a}	2.02 (± 0.60) ^{a}	1.92 (± 1.26) ^{a}
ω-6/ω-3	7.86 (± 6.80) ^{a}	12.09 (± 3.10) ^{a}	17.86 (± 6.18) ^{b}
PUFA/SFA	1.12 (± 0.38) ^{b}	0.28 (± 0.07) ^{a}	0.39 (± 0.09) ^{a}
SFA (g/100g)	1.80 (± 1.35) ^{a}	8.33 (± 2.97) ^{b}	6.15 (± 4.64) ^{b}
C12:0 (g/100g)	0.02 (± 0.01) ^{b}	0.01 (± 0.00) ^{a}	0.02 (± 0.01) ^{a,b}
C14:0 (g/100g)	0.25 (± 0.10) ^{b}	0.05 (± 0.04) ^{a}	0.21 (± 0.16) ^{a}
C16:0 (g/100g)	4.75 (± 1.85) ^{b}	1.04 (± 0.80) ^{a}	3.78 (± 2.87) ^{a}
C17:0 (g/100g)	0.07 (± 0.03) ^{b}	0.03 (± 0.02) ^{a}	0.05 (± 0.04) ^{a,b}
C18:0 (g/100g)	3.18 (± 1.07) ^{b}	0.67 (± 0.48) ^{a}	2.03 (± 1.53) ^{a}
C20:0 (g/100g)	0.04 (± 0.02) ^{b}	0.01 (± 0.01) ^{a}	0.03 (± 0.02) ^{a,b}
MUFA (g/100g)	2.70 (± 2.17) ^{a}	10.17 (± 3.36) ^{b}	7.61 (± 5.68) ^{b}
C16:1 (g/100g)	0.45 (± 0.18) ^{b}	0.07 (± 0.05) ^{a}	0.40 (± 0.29) ^{a}
C17:1 (g/100g)	0.04 (± 0.01) ^{a}	0.02 (± 0.01) ^{a}	0.05 (± 0.04) ^{a}
C18:1t (g/100g)	0.15 (± 0.17) ^{a}	0.07 (± 0.03) ^{a,b}	0.03 (± 0.03) ^{b}
C18:1ω9c (g/100g)	2.47 (± 2.02) ^{a}	8.85 (± 2.94) ^{b}	6.54 (± 4.92) ^{b}
C18:1ω11c (g/100g)	0.12 (± 0.08) ^{a}	0.65 (± 0.21) ^{b}	0.50 (± 0.35) ^{b}
C20:1 (g/100g)	0.04 (± 0.03) ^{a}	0.20 (± 0.07) ^{b}	0.13 (± 0.09) ^{b}
PUFA (g/100g)	2.15 (± 2.23) ^{a}	2.23 (± 0.66) ^{a}	2.07 (± 1.38) ^{a}
C18:2 (g/100g)	0.01 (± 0.00) ^{a}	0.01 (± 0.01) ^{a,b}	0.02 (± 0.01) ^{b}
C18:2ω6t (g/100g)	0.02 (± 0.02) ^{a}	0.02 (± 0.01) ^{a}	NA
C18:2ω6c (g/100g)	NA	NA	0.50 (± 0.17)
C18:3ω6 (g/100g)	1.72 (± 1.90) ^{a}	1.79 (± 0.56) ^{a}	2.70 (± 0.25) ^{a}
C20:2ω6 (g/100g)	NA	0.01 (± 0.00) ^{a}	0.02 (± 0.01) ^{a}
C20:3ω6 (g/100g)	0.04 (± 0.05) ^{a}	0.08 (± 0.02) ^{a}	0.08 (± 0.079) ^{a}
C20:4ω6 (g/100g)	0.01 (± 0.00) ^{a}	0.02 (± 0.01) ^{a,b}	0.05 (± 0.03) ^{b}
C22:2ω6c (g/100g)	0.08 (± 0.02) ^{a}	0.12 (± 0.05) ^{b}	0.09 (± 0.01) ^{a,b}
C22:4ω6c (g/100g)	NA	NA	0.01 (± 0.00)
C18:3ω3 (g/100g)	0.01 (± 0.01) ^{a}	0.03 (± 0.01) ^{b}	0.02 (± 0.01) ^{b}
C18:4ω3 (g/100g)	0.23 (± 0.28) ^{a}	0.13 (± 0.08) ^{a}	0.15 (± 0.02) ^{a}
C20:5ω3 (g/100g)	NA	0.02 (± 0.01) ^{a}	0.02 (± 0.00) ^{a}
C22:5ω3 (g/100g)	0.01 (± 0.01) ^{a}	0.01 (± 0.01) ^{a}	0.01 (± 0.01) ^{a}
C22:6ω3 (g/100g)	0.02 (± 0.01) ^{a}	0.02 (± 0.02) ^{a}	0.03 (± 0.00) ^{a}

0.09) and MR (0.28 ± 0.07).

As shown in Table I, oleic acid (C18:1n9) was the most common fatty acid in HS and MR groups, which registered a significantly (p <0.05) highest value than WB. In HS and MR, palmitic acid (C16:0) was the second most common, followed by γ-linolenic acid (C18:3n6) and stearic (C18:0). In WB, the order was different: the first most common fatty acid was the palmitic acid, followed by the stearic, the oleic and finally the γ-linolenic acid. The γ-linolenic acid content did not differ significantly between the groups, whereas both the palmitic acid and the stearic acid were significantly higher (p <0.05) in WB than in the other two groups. Also, the oleic acid value significantly (p <0.05) differed in WB group, which registered the lowest average. Moreover, lauric acid (C12:0) was significantly higher (p <0.05) in WB loin than in MR, but did not differ compared to HS, whereas the myristic acid (C14:0) was significantly higher in WB rather than in both the other two groups. About the most important ω-3s, wild boar meat had a significantly lower content of alpha-linolenic acid (C18:3n3), but the docosapentaenoic (C22:5n3) and docosahexaenoic acids (C22:6n3) did not differ significantly (p>0.05) among the three groups. The ω-3 percentage found in wild boar meat was higher than in HS and MR, however it did not significantly differ. However, the ω-6:ω-3 ratio was significantly lower in WB and MR loin than in HS (p <0.05).

In order to establish which wild boar meat cut had the most favourable fatty acid composition, the shoulder, ham and loin fatty acid composition of hunted wild boar

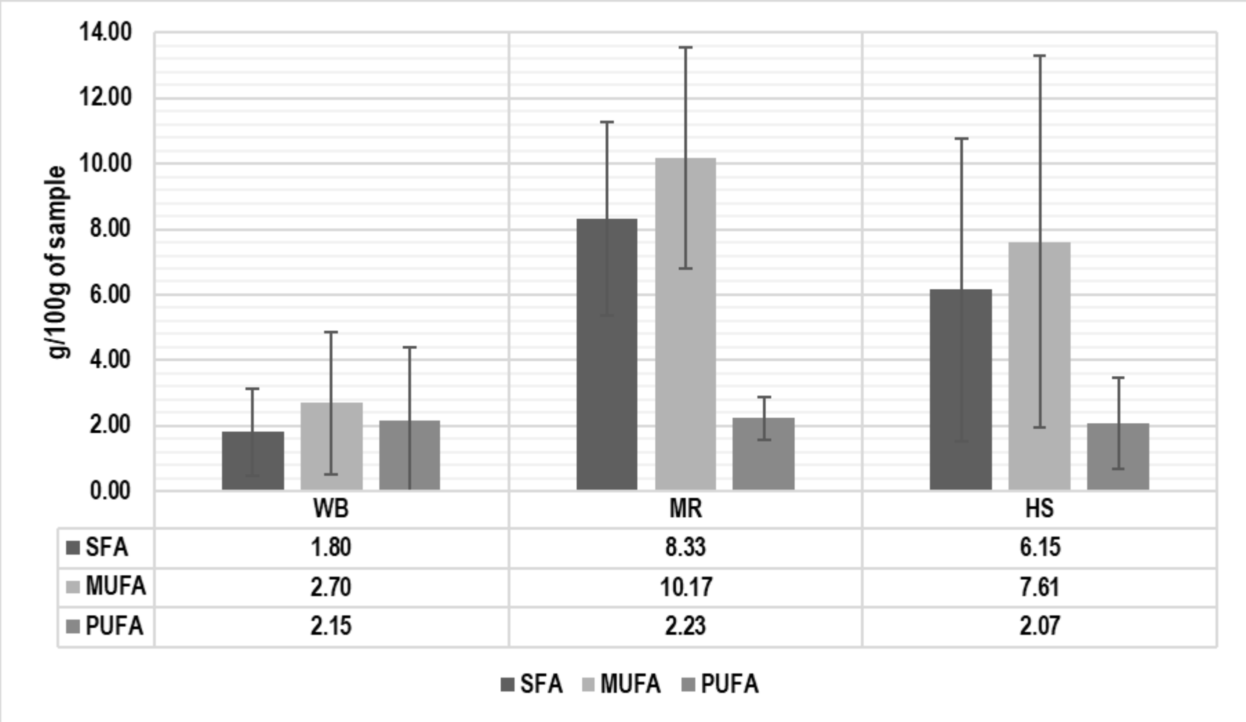

Figure 1 - Mean and standard deviation of fatty acids composition (SFA=saturated fatty acids, MUFA= monounsaturated fatty acids and PUFA= polyunsaturated fatty acids) of *Longissimus dorsi* muscle in Wild boar (WB), Crossbreed Mora Romagnola (MR) and Hybrid pigs (HS).

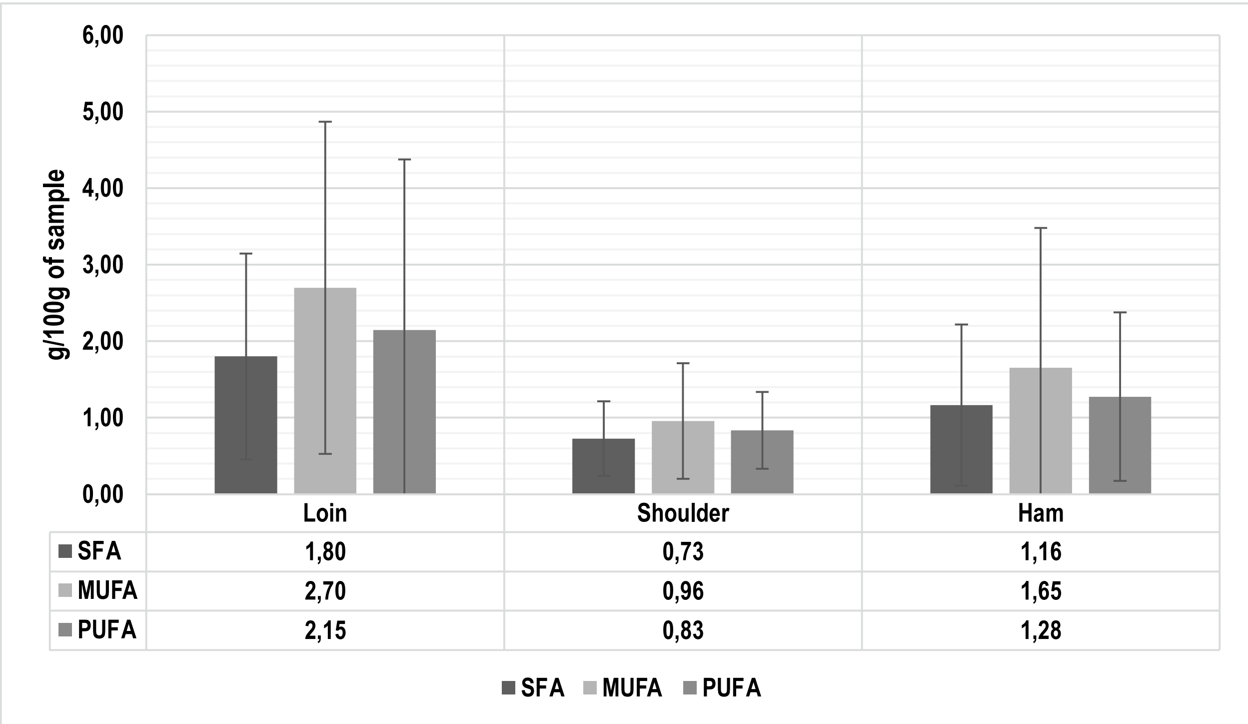

Figure 2 - Comparison of fatty acids composition average (SFA=saturated fatty acids, MUFA= monounsaturated fatty acids and PUFA= polyunsaturated fatty acids) of Loin, Shoulder and of Ham wild boar and standard deviation.

were compared. The loin had a higher content of fatty acids than shoulder and ham, but all meat cuts were richer in MUFA, followed by PUFA and SFA (Fig. 2). Comparing all parameters, no statistically significant difference (p>0.05) was found among the three cuts of WB meat.

4. DISCUSSION

The aim of this preliminary research was to compare the fatty acid composition of the loin of wild boar, Mora Romagnola and hybrid swine, in order to understand if the composition of the crossbreed Mora Romagnola, kept in semi-wild condition, could be more similar to that of wild boar or to that of the intensively bred hybrid pigs. On average, the results showed that MR had the highest content of fatty acids, whereas the WB meat had a significantly less amount of fatty acids rather than the MR. Mora Romagnola and its crossbreed are characterised by high content of fat, because are prone to the adipogenesis, and their meat results as marbled [17, 18].

One of the important parameters that should be considered in the evaluation of the quality of meat is the SFA: MUFA: PUFA and PUFA:SFA ratio. The American Heart Association has underlined that the ideal SFA: MUFA:PUFA should be 1:1:1, in order to obtain the best LDL/HDL ratio (cardiovascular risk marker) [19]. Moreover, the British Department of Health (1994) [20] and World Health Organisation (2003) [21] recommend a PUFA:SFA ratio above 0.4, in order to reduce the risk of coronary diseases in human beings.

Our results showed how the WB registered the best SFA:MUFA:PUFA ratio. WB presented a lipidic profile characterised by 1:1.5:1.2 ratio, while MR and HS recorded similar ratios, respectively 3.7:4.6:1 and 3:3.7:1. Regarding PUFA:SFA, a high value of this ratio had been already found in hunted wild boars in Lithuania (0.43-0.53) [22] and in Portugal (0.52-0.60) [23]. In a previous study by Barbani et al. (2011) [24], the PUFA:SFA ratio found in intensively bred pigs was lower than that for wild boar meat (0.3-0.5 vs 0.7-0.8). Our results confirm what is reported in above mentioned works, indeed the PUFA: SFA value was significantly highest in WB than in HS, and the crossbreed Mora Romagnola registered the lowest value but it did not differ significantly compared to that of HS.

The ω-6:ω-3 ratio is another important parameter, useful in the evaluation of the nutritional quality of meat. It should be under 4.0, to prevent the onset of many diseases, such as cancer, cardiovascular disease or inflammatory and autoimmune disease, caused by a high content of ω-6 PUFA in the diet [16]. While Marisco et al. (2007) [25], found reared wild boar meat richer in ω-3 than that of domestic pigs, our results did not underline any significant difference between the three groups. However, in this study the ω-6:ω-3 ratio in MR and WB were significantly lower than in HS, even if all three groups registered values above 4.0. Similarly, in works of Skewes et al. (2009) [26] and Barbani et al. (2011) [24] the ω-6:ω-3 ratio for WB meat was between 6.0 and 8.5. Wood et al. (2004) [27] reported high ω-6:ω-3 ratio values (9.60-11.48) in the *Longissimus dorsi* muscle of different breeds of domestic pigs. Though values of PUFA:SFA ratio observed in MR meat was closer to that observed in hybrid pigs, the ω-6:ω-3 ratio was closer to that of wild boar. Therefore, crossbreed Mora Romagnola could be considered in a middle position between wild boar and commercial swine. Differently, other Italian breeds reared outdoors (Casertana and Nero Siciliana) were found closer to the hybrid swine [28, 29]. The features shared by the autochthonous Italian breeds, included the Mora Romagnola, and the hybrid pigs depends probably on their genetic. Indeed, the local breeds are often crossed with the commercial swine, like the Mora Romagnola considered in this study, in order to improve their productive performances [18]. Moreover, the observed crossbreed Mora Romagnola underwent a finishing process of 30 days that probably influences their lipidic profile, which, as previously specified, highly depends on their diet.

Fatty acids composition and ω-6:ω-3 ratios are highly influenced by diet composition [23, 30, 31]. In our research, the palmitic and the stearic values of WB meat were significantly higher than those of MR and HS, but the oleic acid value was significantly lower. This result only partially confirms what reported by Sales and Kotrba in 2013 [14], where also the oleic acid was one of the principal fatty acids found in wild boar meat. The high variability found in WB could be determined by seasonality and the differences caused by a change of food availability and diet composition, as suggested in researches of Dimatteo et al. (2003) [32] and Zochowska-Kujawska et al. (2010) [33]. Further investigations are necessary to verify this hypothesis.

Barbani et al. (2011) [24] observed a higher content of fatty acids in the shoulder than in the leg of hunted wild boars. In that case, both cuts of meat were richer in SFA than in PUFA. The different cut of wild boar meat in this research did not differ significantly.

5. CONCLUSIONS

The results of this research suggest that wild boar meat present favourable characteristic of fatty acids composition, high PUFA:SFA ratio and low ω-6:ω-3 ratio. The meat of crossbreed Mora Romagnola pigs kept in a semi wild condition shows intermediate features between the wild boar and the hybrid swine.

This work intends being only a preliminary study and all these aspects should be investigated further in future researches, considering also other aspects that can influence the quality of fat composition, such as the gender and age of the animals, and the seasonality of the samples' collection.

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