COMPARISON OF INTRAMUSCULAR FAT QUALITY BETWEEN PIGLETS OF BÍSARO AND INDUSTRIAL BREEDS – A PRELIMINARY STUDY

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Abstract

Selection for lean meat quality has led to a decrease in total fat contents compromising on meat palatability. Preliminary studies showed higher total intramuscular fat content in autochthones Bísaro *breed when compared to piglets of industrial breed. Analysis of intramuscular fatty acid profile of the semimembranosus/semitendinosus muscles showed that* Bísaro *piglets had a significantly lower content of saturated fatty acid and higher levels of unsaturated fatty acids. The monounsaturated fatty acid (C18:1) content was significantly higher than that in industrial breed. Further studies are required as the breeding of animals, beneficial to human health, is important.*

Key words: *piglets, total fatty acid, fatty acid profile, autochthones breed, industrial breed*

1. INTRODUCTION

A recent article in the Wall Street Journal (Ames 2009) refers to the consumption of roasted piglet meat in the Central Region of Portugal as being anywhere near to 3000 suckling pigs per day as per information of the Gastronomic Brotherhood of the Bairrada Piglet. According to the National Institute of Statistics for Portugal (INE 2014), the total number of piglets slaughtered in Portugal in the Central region was 280,000 animals for the year 2013, the highest when compared to other regions of Portugal. In 2011, the "Leitão da Bairrada" was selected as the one of the 7 Gastronomic Wonders of Portugal and therefore holds an important position economically and gastronomically for the region.

There are three main autochthones breeds of pigs in Portugal, the Alentejo pig, the Bísaro pig and Spotted pig of Alcobaça. The Alentejo pig is adapted more to the South and Spotted of Alcobaça are found in the Central region, with the Bísaro pigs being found in Central and Northern regions of Portugal.

The breeding of the Bísaro pigs is directed towards the recovery of the breed that is classified as almost extinct, the production of meat for the industry of local ham and sausage products and also for the production of piglets for the preparation of the famous roasted piglet "Leitão da Bairrada".

During the old days the typical "Leitão da Bairrada" was defined as a roasted pigling obtained from piglets from the locally bred Bísaro pigs. These piglets are slow growing and usually take a longer time to reach weaning age, approximately 45 days for weaning, and thereafter to reach the ideal live weight for slaughter of 10 to 12kgs (ANCSUB 2013). This led to a greater turn to the use of industrial crossbred breeds to produce faster growing piglets to meet the demand, the deficit of which culminated into the importation of live piglets or frozen piglet carcasses into Portugal. However, the use of these piglets has led to alterations in the quality of the final product and it has been shown that the breed of animal has a great influence on the quality of the meat. A recent study by Franco, Vasquez & Lorenzo (2014) on growth performance, carcass and meat quality of a local Spanish Celtic breed and their crosses with Landrace and Duroc breeds, showed better growth performance for cross breeds but with a lower content of intramuscular fatty acids. Meat obtained from breeds with greater production of lean meat often demonstrates a negative correlation for meat flavor (Newcom et al. 2004; Mörlein et al. 2007). Mörlein et al. (2007) stated that within the commercial breeds studied, those that had a higher intramuscular fat had more favorable meat quality traits. In an earlier study Furman et al. (2010) on comparing fatty acids of the Slovenian local pig breed and that of the different

types of commercial breed, found that there were differences in the profile of fatty acids between these animals. Pugliese & Sirtori (2012) showed that the indigenous breeds are gaining more and more importance in the food chain when compared to improved breeds, primarily due to better quality of meat. Petrović et al. (2014) compared the profile of fatty acids in the local pig breeds Mangalitsa and Moravka found in the region of Serbia. They observed significant differences between breeds in the quantity of saturated fatty acid and monounsaturated fatty acids.

Ludke & López (1999) have shown that lipids are important for maintaining health and several functions in the animal and human body. They are required by all living cells and tissues of the body, where they play structural and functional role in plasma membrane and synthesis of steroid hormones. Fats obtained from animal source can play an important role in these body functions. However, these authors caution on the consumption of animal fats for persons prone to high cholesterol levels.

According to the World Health Organization high consumption of fatty food can, cause serious health complications and therefore recommends in their Fact Sheet of 2015, a reduced consumption of fat to less than 30% of total energy with an increase in unsaturated fatty acid intake (WHO–Healthy diet Fact Sheet 394, 2015). Unsaturated fatty acids can therefore play an important part in the human diet especially with regards to the essential fatty acids needed by the human and animal bodies. The two most essential fatty acids needed by our bodies are the α-linolenic acid and the linoleic acid (Václavková & Bečková 2007).

Different studies were carried out to know the fatty acid profile of the adult Bísaro pigs. Pires da Costa et al. (2002) reported that the Bísaro breed of pigs have a good amount of intramuscular fat (marbling) with a good balance between the unsaturated and saturated fatty acids, favorable to the technological quality of meat. Later, Carvalho (2009) and Carvalho et al. (2011; 2013) conducted further studies on the profile of fatty acids in different muscles of the Bísaro pig breed, with animals of different age and weight groups and concluded that the meat from the Bísaro pig is healthy from a nutritional point of view, mainly because the levels of fatty acids of family Omega 3 and 6 present.

This preliminary study aims at studying the total fatty acid and the fatty acid profile in the piglets of the Portuguese Bísaro breed and comparing the results with that of the commercially sold industrial piglets.

2. MATERIALS AND METHODS

2.1. *Animals*

Figure 1: Bísaro female – ESAC

Figure 2: Bísaro piglets - ESAC

A total of 38 piglets were used for the study. Nineteen of these animals were of the Bísaro breed (Figure 1 & 2) and the other 19 of the common industrial crossbreeds (Landrace x Large White) and the Pietran male cross with (Landrace x Large White) females, generally considered as Hybrid animals. The Bísaro breed of animals were reared on the farm owned by the Agriculture College of Coimbra (Escola Superior Agrária de Coimbra-ESAC) and were reared under confined conditions until they are ready for slaughter.

The industrial crossbred animals were from different farms with the animals being reared under confined intensive system. The average carcass weight of the Bísaro piglets was 8.120kg ± 2.3 and that of the industrial breed was of 8kg ± 0.8. Even though the weights of the piglets are almost similar, the Bísaro piglets are ready for slaughter at an older age when compared to the industrial breed. For both groups of animals the commercially available concentrate feed for growing piglets was used. Water and feed was administered *ad-libitum.* The animals were slaughtered under similar conditions at the same slaughter house.

2.2. *Sampling process*

The meat samples were collected from the muscles *Semimembranosus*/S*emitendinosus* of the piglet. Soon after slaughter and approval by the Official Veterinarian, the carcasses were weighed and two pieces of meat (muscle) with approximately 20 g (each), were removed with the help of a scalpel (Figure 3).

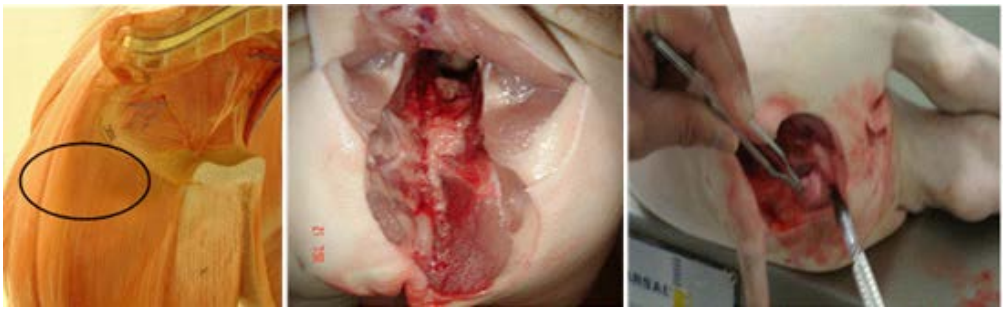

Figure 3: Obtaining muscle sample

A square-shaped cut was performed and about five centimetres deep. The samples were then placed in previously identified plastic bags suitable for food and placed in a thermal bag in ice.

On reaching the laboratory, each sample was subdivided and stored separately in order to facilitate its use at different times. The samples were frozen at a temperature of -80° C until further physicochemical analyses.

2.3. *Fat content and combined fatty acids analysis*

For analysis of the total fat content, extraction from the dried meat sample was carried out using the Soxhlet Extractor method as described in NP 1613 of 1979.

For analysis of the profile of fatty acids, approximately 1g of crude meat was weighed and placed into a 15mL glass tube. To this tube were added 5 mL of heptane and 1 mL of methanol and the sample was fully macerated using a glass rod. After 2 minutes of resting, the supernatant was collected and transferred to another glass tube and 0.5mL of methanolic sodium methoxide (2M) was added and agitated for 4 minutes using a vortex. Following the stirring process 2 mL of the supernatant were collected and stored in Eppendorf's at temperatures of -18° C until further analysis.

The area relative percentage of combined fatty acids was determined by gas chromatography, using a chromatograph (HP 6890 series) equipped with a split/splitless injector (split ratio=1/20), a flame ionization detector (FID) and a capillary column (FFAP, Phenomenex, USA, 30 m x 0.32 mm, 0.25 µm phase thickness). The column oven program was: initial temperature of 60ºC for 4 minutes, followed by a temperature increasing ramp at 12ºC/min to 220ºC and then isotherm for 15 minutes. Fatty acids retention times were compared with those from authentic olive oil sample, used as reference.

2.4. *Statistical analysis*

The statistical analyses were conducted using the program ASSISTAT VERSION 7.7 BETA (2014). Making a descriptive analysis followed by ANOVA using the t test for a level of significance of 95%.

3. RESULTS AND DISCUSSION

The percentage of total fat and fatty acid profile between Bísaro piglets (BP) and Industrial piglets (IP) are presented in Table 1. On analyzing the data it is noted that there is a significant difference ($P<0.01$) between the BP and the IP in the percentage of total fatty acid, saturated fatty acid and monounsaturated fatty acid. The percentage of total intramuscular fat content was 2.4% for BP and 1.4% IP, with saturated fatty acid (SFA) being 31.9% for BP and 35.8% for IP and the monounsaturated fat (MUFA) being 45.3% in BP and 41.3% in IP. With reference to the profile of polyunsaturated fat (PUFA) there was no statistical difference between Bísaro (20.0%) piglets and the Industrial piglets (19.1%).

Table 1: Total Fatty acid and Fatty acid profile in *semimembranosus/ semitendinosus* muscles of Bísaro breed and commercial industrial piglets [1]

	TF (%)	Σ SFA (%)	Σ MUFA (%)	Σ PUFA (%)
Bísaro	2.4±1.4 [a]	31.9±4.1 [b]	45.3±3.8 [a]	20.0±2.8 [a]
Industrial	1.4±0.4 [b]	35.8±2.3 [a]	41.3±3.5 [b]	19.1±3.9 [a]

[1] Means with identical letters do not differ by the t test ($P<0.01$) of probability

Our results are similar to those found by Furman et al. (2010) when these authors studied the fatty acid profile differences between the local Slovenia breed, Krškopolje pigs, and the commercial fatteners. From their study if we take into consideration the total fatty acid (TF) percentage of the Krškopolje pigs (1.96%) and the TF percentage commercial meaty group (1.4%), the results are almost similar to ours. In our case, the animals were of a younger age group (varying between 40 and 60 days of age at slaughter) and a significantly higher percentage of TF was observed in the Bísaro piglets (2.4%) when compared to the commercial piglets (1.4%) which in case of the Bísaro breed is well within the recommended value of 2-2.5% for a better meat sensory quality. The ideal concentration of intramuscular fat has been estimated to be 2 to 3% (cited by Burkett 2009) However, a higher average quantity of total intramuscular fat (TF) was observed in the muscle *longissimus thoracis et lumborum* of Bísaro pigs in study conducted by Carvalho (2009) and Carvalho et al. (2011) in Bísaro pigs showing that the TF had an average value of 4.89g/100g with the minimum value of 2.4 and the maximum of 9.4g/100g.

The importance of the profile of fatty acids for human health has been well documented by many authors and the trend has been to breed animals with low saturated fatty acid content and higher monounsaturated and polyunsaturated fatty acids. De Smet, Raes & Demeyer (2004) in their review have mentioned that genetic variability has a great influence on fat deposition and intramuscular fat content and have stated that variation in fat content has an influence on the fatty acid composition, independent of the species, breed and dietary factors. Further these authors cited that the content of saturated fatty acid (SFA) and monounsaturated fatty acid (MUFA) increases with increased fatness than the polyunsaturated fatty acids (PUFA), leading to an imbalance in the PUFA/SFA ratio. However, from results obtained in this study, the quest for leaner meat has led to that, in spite of the total fat content being lower in the industrial piglets, the saturated fatty acids content was significantly higher (35.8%) when compared to the local Bísaro breed (31.9%) and on comparing the content of MUFA a significantly higher content was observed in the Bísaro piglets (45.3%) than in the industrial piglets (41.3%) with no significant difference in the PUFA content. Excess of polyunsaturated fatty acid has been implicated in its effect on the shelf-life of the product as high values leads to greater oxidation of the product and rancidity (Gunstone 2008).

Similar results were also presented by Furman et al. (2010), where they noted that the local Krškopolje pigs had a significantly higher MUFA (48.5%) content than the meaty or normal commercial breed of pigs, with significantly lower SFA (33.8%) content when compared to all commercial breed types. However a partial difference was observed from our study, in that they observed a higher quantity of PUFA in the meaty commercial breed (22.56%) as compared to the local breed (17.61%), whereas in the normal and fatty types of the commercial breed the results were similar to ours, that is, no significant differences were observed in the quantity of PUFA between breeds. Though, in our case the local Bísaro breed had a higher quantity of PUFA (20%) when compared to the industrial pigs (19.1%). Petrović et al. (2014) mentioned that the breeds of pigs influenced the total fatty acids, quantity of saturated fatty acids and monounsaturated fatty acids but not the polyunsaturated fatty acids when in their study these authors compared two local indigenous breeds. However they found very low levels of PUFA (4.1%) when compared to our study or that of Furman et al. (2010), whereas their report of a higher MUFA (56.41%) content compared to that of SFA (39.4%) are in keeping with our findings.

The recent FAO report of Expert Consultation in 2010 recommends that apart from defining the broad groups of fatty acids in a diet, it could be even more beneficial to know the individual fatty acids within each group as they may have unique biological properties and health effects that could improve human health. In our study it was noted that within the saturated fatty acids, the most common SFA found were the C 14:0, C16:0, C18:0 & C20:0, in the MUFA group they were C16:1 & C18:1 and in the PUFA group were C18:2 & C18:3. On comparing the fatty acid profiles of the two different breeds (Table 2; Figures 4 & 5), it was noted that the following quantities of fatty acids were present. SFA C16:0 (palmitic acid 23.62% in BP and 26.16% in IP), C18:0 (6.64% stearic acid in BP and 8.43% in IP), C14:0 (myristic acid 1.18% in BP and 1.49% in IP) and C20:0 (arachidic acid 1.04% in BP and 0.6% in IP). Of all these SFA it is considered that the myristic acid is the most prejudicial to human health as it increases the blood cholesterol level (Tholstrup, Vessby & Sandstrom 2003). From our results we can observe that in comparison to IP (1.49%), the BP had a lower quantity (1.18%) of myristic acid, thereby having a positive dietary effect. On the other hand the MUFA values for the two different breeds were C18:1 (oleic acid 41.92% for BP and 37.38% for IP), clearly showing that the significantly higher ($P<0.01$) quantity of oleic acid in the local Bísaro piglet can be a positive factor for human health as it has been shown that oleic acid, also known as omega 9, can help attenuate the effects of saturated fatty acids (Carluccio et al. 1999). The FAO Expert Consultation report (2010) mentions that C18:1n-9 is the major MUFA found in the Western dietary sources. In the C16:1 variety of MUFA the BP showed 3.99% of palmitoleic acid and 3.70% in IP. In contrast, though no significant difference was observed between groups for PUFA, the industrial piglets showed higher levels of linolenic acid (1.14% C18:3) when compared to 0.55% in BP. The fatty acid C18:2 (linoleic acid) was 19.48% in BP and 19.44% in IP. Similar results for the composition of fat in Bísaro pigs was reported by Pires da Costa et al. (2002) who demonstrated that there exists a good balance in unsaturated/saturated fatty acids ratio (neutral lipids (LN) 1.23 ± 0.28; polar lipids (LP) 2.00 ± 0.21), with predominance of the monounsaturated Oleic (C18: 1-LN 45.6; 24.5 LP), which suggests a good quality of fat in this breed of pigs. In the study by Carvalho et al. (2013) in Bísaro pigs they observed a similar trend for the quantities of SFA, MUFA and PUFA. On analyzing the fatty acid profile in *bíceps femoris* muscle they found an average of 35.1% for SFA, 44.43% for MUFA and 20.69% for PUFA. Though no concrete evidence exists, it has been shown that replacement of SFA by MUFA could result in a reduced risk of coronary heart disease (CHD) as there is strong evidence that increasing MUFA in diet reduces the low density lipoproteins and increases the high density lipoproteins. There is convincing evidence that increasing PUFA availability in human diets could prevent CHD (FAO 2010). All these combined results show that the fatty acid content from the local Bísaro breed of pigs could bring some benefits for the human diet when consumed in moderation.

Table 2: Fatty acid profile (%) in *semimembranosus/semitendinosus* muscles of the Bísaro & Industrial piglets [1]

Fatty acid	Bísaro	Industrial
C14:0	1.18±0.5[a]	1.49±0.8[a]
C16:0	23.62±3.1[b]	26.16±2.6[a]
C16:1	3.99±1.4[a]	3.7±1.7[a]
C18:0	6.64±1.6[b]	8.43±1.8[a]
C18:1	41.92±4.2[a]	37.38±3.3[b]
C18:2	19.48±3.1[a]	19.44±4.2[a]
C18:3	0.34±0.3[a]	0.54±0.6[a]
C20:0	0.38±0.6[a]	0.09±0.2[a]

[1] Means with identical letters do not differ by the t test (P<0.05) of probability

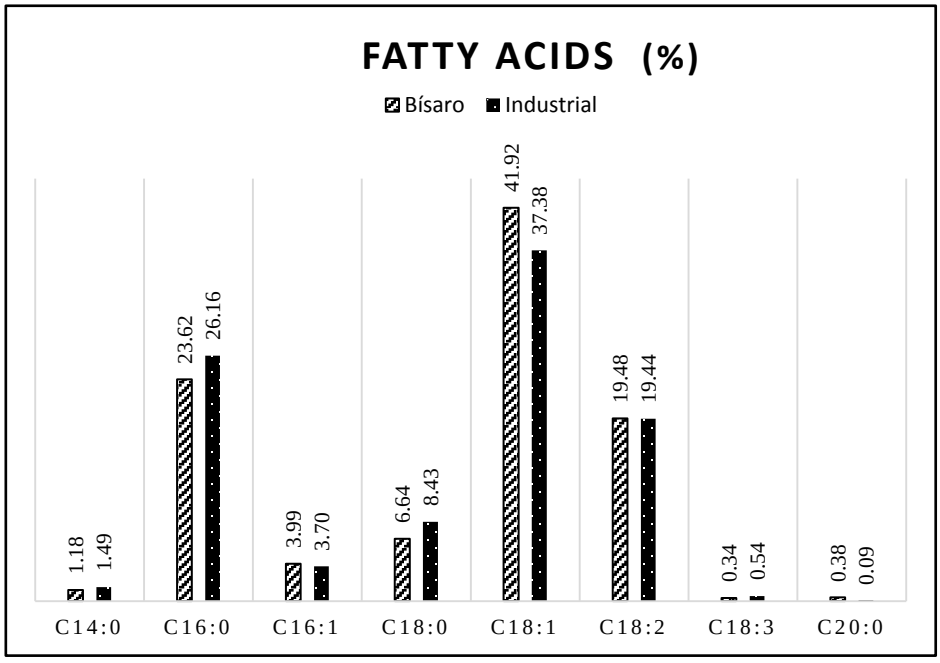

Figure 4: Fatty acid profile of intramuscular fat of piglets.

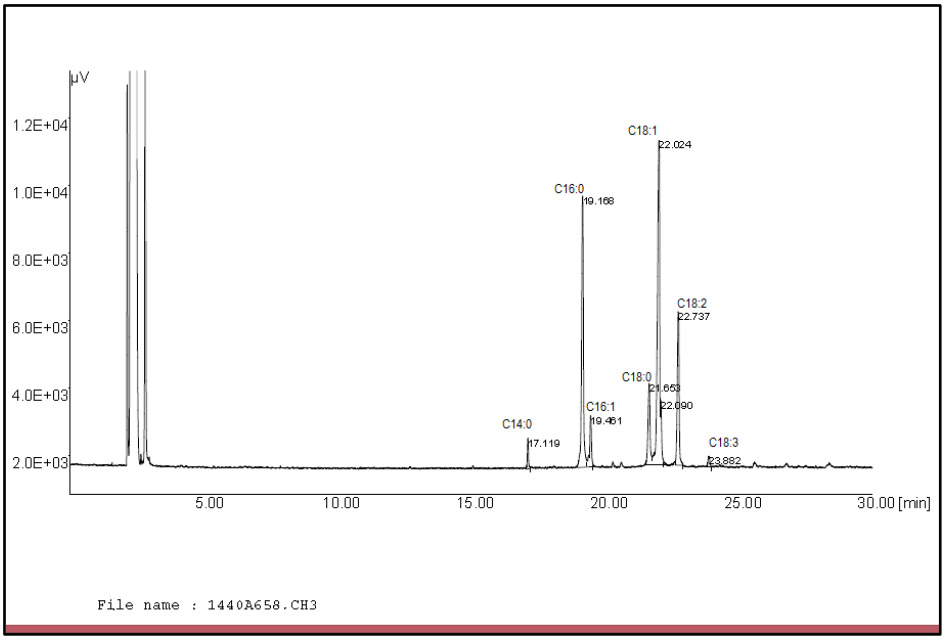

Figure 5: Chromatogram of fatty acid profile of intramuscular fat of piglets.

In table 3 are shown the profile of fatty acids in the *semimembranosus/semitendinosus* muscles of the Bísaro male and female piglets, it was observed that there was no influence of sex within the fatty acid group profiles of the piglets. Carvalho (2009) in the study of the influence of gender on different groups of fatty acids, reported a non-significant differences (P>0.05) in the different muscle tissues analyzed. In the *semimembranosus* muscle this author noted that the saturated fat present in male was 36.21 % and in females 34.72%. The percentage of monounsaturated fat was 37.00% in males and 41.40% in females and the polyunsaturated fat was 26.80% in males and 24.26% in females. No significant difference was noted (P>0.05). The results obtained by this author after the analysis of different age and weight groups of Bísaro were similar to our findings in piglets.

Table 3: Fatty acid profile in *semimembranosus/semitendinosus* muscles of the Bisaro & Industrial piglets - Differences between Male and Female piglets[1]

	TF (%)	Σ SFA (%)	Σ MUFA (%)	Σ PUFA (%)
Male	2.84±1.8 [a]	30.43±3,8 [a]	45,80±3,4 [a]	20.83±2.9 [a]
Female	2.30±1,2 [a]	32.78±4,6 [a]	45.03±4.5 [a]	18.69±2.3 [a]

[1] Means with identical letters do not differ by the t test (P<0.01) of probability

4. CONCLUSIONS

Meat has been a part of the human diet for centuries and the type of meat consumed has evolved with time. As human health is considered as being of prime importance in today's health conscious world emphasis is being placed on prevention of alteration in blood biochemical parameters, such as high cholesterol levels, which can be detrimental to human health. The production of food that is beneficial to the human health is one of the priorities of today's research goals. The findings in this work meet the priorities of the consumer in what concerns quality, palatability and health. It is noted that the fatty acid composition in the muscles of the indigenous Bísaro piglet and the commercially bred industrial piglets is different. The meat of the Bísaro piglets seems to be more beneficial from a nutritional point

of view due to higher levels of monounsaturated and polyunsaturated fatty acids, being these essential to human diet, and on the other hand presenting lower levels of saturated fatty acids when compared to the industrial breed. Apart from this, the roasted pigling "Leitão da Bairrada" is considered to be one of Portugal most praised gastronomic delicacy and therefore, as mentioned by the Gastronomic Brotherhood of the Bairrada Piglet, the importance of promoting the breeding of these Bísaro pigs and the registration of the product as a Certified Product from the Bísaro piglet. Furthermore, studies envisaging the valorization of the genetic resources are aligned with the regional smart specialization strategy and, due to the impact of "Leitão da Bairrada" on the regional economy, it is expected that the Bísaro breed will recover its former importance. This could also help in the recovery of the breed that is almost in extinction and create awareness to the general public of the interest in consuming quality food from the local breed. In many countries in Europe, the breeding of pigs with meat characteristics important from the consumer point of view in so far as the organoleptic quality and benefits for human health is concerned, is currently gaining more interest.

It is hoped that this study will be one of many to show the importance of the recovery of the Bísaro piglet production and the importance of registration of the product "Leitão da Bairrada" as a certified national gastronomic product produced with Bísaro piglets.

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