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## PORK QUALITY AND BOAR TAINT AS IMPORTANT ISSUES IN MODERN SWINE PRODUCTION

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#### Abstract

The objective of this study was to analyze and criticize the published literature related to pork quality in male pigs, in modern swine production.

There are different types of quality and different factors that can influence the formation and assessment of quality. Meat from uncastrated males may expressed offensive odor and flavor, known as Boar taint, due to which it becomes less desirable for most of the market. Awareness about the side effects of the surgical castration is significantly increasing, while consumers are more interested in how animals are raised and produced.

To date, there are promising results from several modern on-line technologies, for boar taint detection at slaughterhouse, which are under development regarding cost efficiency, simplicity and analysis time. The sorting of boar carcasses is still based on the olfactory assessment of the animals` backfat. None of suggested alternatives for surgical castration guarantees the entire elimination of boar taint. However, further research is needed to establish a harmonized system, as a combination of different, currently available methods that would deal with the boar taint issue. Moreover, there is not yet any accepted official reference method available for the analysis of skatole and androstenone. This makes the difficulty in comparing the results between laboratories and different studies. Therewith, it is difficult to develop a harmonized system for entire elimination of boar taint. Within each European country, the percentage of carcasses with high boar taint levels and the results from consumers acceptance studies could be useful while developing an official reference method for skatole and androstenone analysis. It was encouraging to see that there is a market for tainted meat lovers which could be bigger if consumers were more informed about boar tainted products and their benefits.

Key words: Pork quality, Boar taint.

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#### 1 Introduction

During the process of globalization, changes in life style and changes in social values, we have witnessed that consumer awareness and demand for the high-quality meat has been significantly increased. After satisfied the need for a certain amount of meat, the focus shifts to the quality. The ultimate consumers of meat and meat products are becoming choosier when buying. Within different countries, customer requirements and preferences for quality meat differ. In developed countries, consumers increasingly prefer a meat rich in protein, vitamins, minerals, and poor in energy content (fat). While in developing countries, this trend is not yet so widespread. The meat industry is therefore daily confronted with more issues and challenges how to improve meat quality and fulfill all the requirements. Possibility to predict the quality of pork is one of the most crucial role in the business success of the manufacturer. Also, a lot of attention is being paid to issues related to sustainability of meat production and animal welfare. Especially in recent few years, surgical castration of male pigs has become the main issue of animal welfare concern. This happened because it has been scientifically proven that surgical castration is a painful procedure and undermines the welfare of animals. Non-castrated boars have better meat quality traits with higher content of muscle tissue and protein as compared to castrates. Problem of reporting an unpleasant odor and taste of non-castrated boars caused by accumulation of androstenone and skatole in adipose tissue gives an occasion for intensive research related to the occurrence of this defect. From centuries ago, surgical castration is used to avoid the development of aggressive behavior in male pigs during sexual maturation and to avoid the development of boar taint. Non-invasive and painless alternatives to castration are still being studied and are practiced in only a limited number of European countries (EU). Some of them use analgesia and anesthesia during and after castration to relieve pain while, in another surgical castration is performed even without any anesthesia. In a small number of countries male pigs are raised as entire male or they are immunocastrated. According to `` European Declaration on alternatives to surgical castration of pigs``, surgical castration of pigs should be abandoned by 1. January 2018 (Briyne et al., 2016). This only increases the pressure of the adoption of generally accepted alternative in eliminating boar taint. The importance of solving this problem lies in resolving at the same time animal welfare issues, issue of carcass quality and meat quality in non-castrated boars, off-flavors and consumers acceptance issue, environmental issue, production profitability issues and many other.

Finally, the aim of the study was to examine, present, analyze and criticize the published literature related to pork quality in male pigs. This work reviews: the importance of pork for human consumption, the pork quality, factors that may affect pork quality, quality measurement, defects and causes, boar taint as a major disadvantage, different approaches in combating boar taint, consumers` perception and acceptance of boar tainted meat, analytical tools to determine boar taint on the slaughter line and more. You will find the answers to the questions what are the challenges on the way to meet consumers' expectations, whether it is possible to eliminate boar taint or not, how can pork quality in male pigs be improved, all through literature overview, discussion, personal evaluation and suggestions for further investigation.

#### 2 Literature overview

#### 2.1 Production and consumption of pork

Among some other indicators, the importance of pork can be shown through the world pork production. Demand for pork grows along with incomes rise in developing countries while, in developed countries remains more stable or grows very slowly. According to results from 2015, production and consumption of pork occupies leading position in European Union (EU) countries and second position in the World. In EU-28 (28-member states of EU), pork consumption leads with 33.0 kg/capita followed by poultry with 22.7, beef 10.8 and sheep 1.8 kg/capita. While, world pork consumption with 12.5 kg/capita stands immediately after chicken with 13.5 kg/capita (OECD-FAO, 2016). As the exception in some countries there is no pork consumption due to cultural and religious limitations (Algeria, Bangladesh, Egypt, Ethiopia, Iran, Pakistan, Sudan & Turkey) (FAO, 2017).

In Norway, pork takes first place again with 40 % of the total meat production in 2015, compared to poultry and beef with 28 and 24 % respectively (Statistiks sentralbyrå Norway, 2015).

The world's largest pork producer and consumer is China with 51.850 (in 1.000 metric tons) followed by EU with 23.350 and United States with 11.307 in 2016. EU has further expanded their position as the leading global pork exporter becoming a key supplier to China and other Asian markets. Global pork trade continues to rise which only confirms the importance of pork for human consumption (USDA-FAS, 2016).

#### 2.2 The pork quality (the "pork quality" concept and different types of quality)

In recent years, during the rapid expansion of global trade, the meaning of pork quality has become more complex. According to Warriss (2010), there are just two types of quality. Functional quality that relates to the desirable characteristics that one product should possess and conformance quality as the product that exactly meets consumer's specifications. When most people talk about quality they usually mean on functional quality. The major components of meat quality include treats such as: quantity of saleable product, ratio of fat to lean, muscle size and shape, fat texture and color, amount of marbling (intramuscular fat) in lean, color and water holding capacity (WHC) of lean, chemical composition of lean, texture and tenderness, juiciness, flavor, nutritional value, chemical safety, microbiological safety and acceptable husbandry of animals.

In the past, the main purpose for rearing pigs was more for lard which they produced then for their lean meat. They were selected to grow large and fat because of lard. Nowadays, modern pigs are relatively lean having been selected over the years to have less and less fat and larger muscles. This means that definition of pork quality varies over time (Warriss, 2010).

The value given to the factors of quality can differ from livestock producers to processors and final consumers. Sensory quality of fresh pork is a major determinant of consumer choices around the world, with different preferences among countries (Ngapo, Martin & Dransfield, 2007a, 2007b). Nevertheless, nutritional quality, sensory (eating) quality, technological quality, hygienic/toxicological quality and ethical quality are all included in the "pork quality" concept. (Rosenvold & Andersen, 2003a). In other words, the pork quality is used to describe the overall characteristics of pork meat. Further, we are going to explain each type of quality included in the "pork quality" concept.

At first, when we talk about nutritional quality, it is known that the nutritional value plays a key role in choosing different types of food for well balanced and quality diet. The nutritional quality provides information about the content of protein and amino acid composition, fat (the fatty acid profile), vitamins and minerals. As a very concentrated source of protein with a high biological value, meat contains all the essential amino acids in composition which matches with our own proteins. Also, meat gives an important source of vitamins (B1 (thiamine), B3 (niacin), B2 (riboflavin), B12 (cobalamin), B6, A (retinol)), minerals (iron, copper, zinc and selenium) and lipids. In the diet, meat only does not provide carbohydrate, fiber, vitamin K and vitamin C (Warriss, 2010). Although meat is one of the most nutritional foods, consumers may consider the high content of fat and cholesterol in meat as undesirable and unhealthy which explains why there is much interest in manipulating its fatty acid composition (Wood et al., 2003) and produce healthier meat. Thus, the goal is to achieve a higher ratio of polyunsaturated to saturated fatty acids (PUFA) (in pigs, to increase n-3 PUFA). Compared with beef and lamb, pork has the best P: S ratio while, beef and lamb have better n-6: n-3 ratios (Warriss,

2010). The fatty acid profile of monogastric meat animal species is relatively easy to alter by proper feeding. Dietary fatty acids are absorbed directly and unchanged from the intestine of non-ruminant species (Ellis et al., 1999; Enser et al., 2000) and deposited in both muscle and fatty tissues, making pork an excellent delivery system for fat of a healthy composition for human consumption (Morel at al., 2006).

The hygienic and toxicological quality is about food safety which is always more important than sensory quality or eating quality. The meat should be free from substances like growth hormones, antibiotics and other medicines, pathogenic microorganisms and the environmental contaminants, heavy metals and other toxins. One of the ways of ensuring food safety is tracing and following all foodstuffs, food producing animals and other substances intended for use as foods, through all stages of the food chain (Olsson & Brandt 2015). The demand for traceability increases due to various food scandals related to food borne diseases such as "mad cow disease" (BSE), Salmonella, E. coli, Listeria monocytogenes in salmon, Campylobacter in chicken, Yersinia enterocolitis in pigs, Toxoplasma and Scrapie in sheep, due to the environmental contaminants such as dioxin, cadmium, radioactive material and other.

Sensory quality shows qualitative traits of meat that can be assessed by using our senses related to shape, color, taste, smell, tenderness/hardness and juiciness. Color is the main determinant of meat appearance when consumers need to purchase and evaluate its acceptability. Also, appearance of meat and its technological characteristics are close related. Pale, soft, exudative (PSE) and dark, firm, dry (DFD) meat are examples of undesirable appearance (Warriss, 2010).

Eating quality or palatability includes texture, juiciness and flavor/odor. Tender and juicy meat is preferable with exception in some countries where consumers prefer chewy meat (African countries). Flavor and odor are closely related. Flavor depends on water-soluble constituents while odor depends on fat-soluble constituents. When abnormal odor is present, either from spoilage or from Boar taint that can override all other quality traits (Warriss, 2010). Moreover, storage conditions and temperature are very important because meat can spoil due to inadequate storage conditions. The method of preparation and cooking affects sensory quality and eating quality of meat.

Technological quality represents quality evaluation of the raw material related to further processing (smoking, drying, manufacturing of bacon and other). Among the most important

there are: water holding capacity (WHC) as the main determinant of technological value, pH (potential of hydrogen), lipids (saturated, unsaturated, poly-unsaturated), content of connective tissue and antioxidant-status.

Ethical quality refers to the need that meat should come from animals which have been bred, reared, handled and slaughtered in ways that promote their welfare and in systems which are sustainable and environmentally friendly (Warriss, 2010). Consumers value such type of meat as a more ecological compared with the commercially produced pork, beef or chicken.

## 2.3 The most important indicators of pork quality (post-mortem changes, rigor mortis, pH, water holding capacity, color and intramuscular fat - marbling)

The formation of meat starts after slaughter of animal when blood flow, oxygen flow and nutrients flow stop connection with muscles. Then, in anaerobic conditions, the glycogen reserves in the cell serve as the last source of energy. When this source is spent, lactic acid starts to accumulate because it cannot be removed by blood system. This leads to gradually acidification of muscles (pH goes down), denaturation of proteins and transformation of muscle to meat (Warriss, 2010).

When muscle enter rigor mortis, its length becomes fixed because it cannot be stretched, carcass becomes stiffer and fat firmer as it cools. The strength of rigor mortis determines the tenderness/hardness of meat. Once rigor occurs, the ``softening`` of meat is only possible by enzymatic reaction during the "ageing" of meat. Therefore, there are different recommended ``ageing`` times (from slaughter to meat consumption) for different types of meat. For example, to achieve 80% of maximum tenderness (measured at 1 °C degree), for chicken meat it takes 8 hours after slaughter, for beef 10 days and for pork 4.2 days (Warriss, 2010). Rigor takes different times to develop in different species. In pork, according to Savelli et al. (2005), rigor mortis starts from 15 minutes to an hour and ends in six hours. The time of onset of rigor mortis is determined only by the availability of adenosine triphosphate (ATP) and not by the pH value. If the animal has been exhausted before slaughter, it is possible to have rigor even with relatively high pH in the muscle (known as alkaline rigor). The speed of rigor development can be reduced if the carcass is cooled quicker or it can be increased if glycogen is depleted by longer preslaughter animal stress. Factors that affect the level of glycogen in animal before

slaughter, also affect indirectly the rate of rigor occurrence (Warriss, 2010). If an animal passes through stress immediately before slaughter, the degradation of glycogen becomes more intensive. This continues post mortem with increasing the concentration of lactic acid and formation of a pale, soft and exudative meat. On the other hand, when stress persisting over a long period prior to slaughter, glycogen reserves are exhausted and formation of lactic acid is insufficient. Then meat becomes dark, firm and dry, and prone to failure because, the higher pH value of meat does not inhibit the growth of microorganisms (Dalmau et al., 2009).

Nonetheless, the speed and intensity of pH decreasing value have a major impact on color, flavor, water holding capacity, juiciness and tenderness. If the pH value drops very quickly, the meat will be characterized as pale, soft and exudative while, very slow and incomplete decline in pH value leads to a dark, firm and dry meat. Normal meat quality implies a moderate rate of pH decline, but also a complete drop in pH value when it reaches the isoelectric point of the myosin (at 5. 4) (Hofmann, 1994).

Water holding capacity is a measurement of meat ability to retain water during cooking, grinding or pressing. Water holding capacity determines juiciness of meat and depends primarily on the pH value. Therefore, it is important to alleviate stress before slaughter because it leads to reducing the pH value in the muscles while the animal is still alive (Henckel et al., 2000), which adversely affects the ability of meat to retain water.

The color plays a key role at a time when consumers evaluate the freshness of meat. Since it is the first visible quality trait, color score can help determining changes in the quality of meat. In general, meat color is dependent on species, age and muscle type. The normal color of fresh pork is pinkish-red but, it happens that different effects can cause deviations from these colors. The most important factors that might affect the color of fresh pork are: the content of pigments and their oxidative status, the content of intramuscular fat tissue and speed of post-mortem glycolysis.

The myoglobin (Mb), as the most important pigment of meat, has the function of storing and delivering oxygen in muscles. The rate of myoglobin oxidation determines meat color stability (Faustman, Sun, Mancini, & Suman, 2010). Thus, during exposure to oxygen, pinkish-red color of fresh pork can pass either into pink (Mb oxidized to oximyoglobin-MbO2) or brown

(oxidation up to metmyoglobin- MetMb) color. Brown color is not desirable for consumers because it is linked to meat which is not fresh.

The speed of post-mortem glycolysis, as well as muscle-specific, affect the rate of discoloration in meat. The red, oxidative (type I) muscle fibers have lower glycogen content and are more susceptible to the development of dark, firm and dry meat as opposed to white, glycolytic (type II) fibers which have greater reserves of glycogen however, fast glycolysis and therefore more inclined to pale, soft and exudative meat (Dalmau et al., 2009).

Intramuscular fat or marbling is an accumulation of adipose tissue between the muscle fibers or between the muscle bundles in the connective tissue. Intramuscular fat contributes to the improvement of meat quality by affecting the flavor, juiciness, tenderness and visual characteristics of meat (Katsamuta, 2011). Intramuscular fat is positively correlated with percentage of red muscle fiber and negatively correlated with white muscle fiber in muscle (Hwang et al., 2010). Beef, pork, lamb, and poultry have different flavor due to the variation of the flavor precursors generally in the fat between and within species. If there is a lack of marbling in meat, there is a lack of juiciness too, as a major quality issue in pork. There are different recommendations for the optimal content of intramuscular fat from different authors. According to Wood (1990), the optimal value of intramuscular fat is minimum 1%, which is the most proper for the United Kingdom trade. While, Pisula & Florowski (2009) claim that for optimum flavor, value of intramuscular fat should range from 2.5% up to 3%. However, meat acceptability by the consumer can be reduced if the fat content is greater than 3.5% (Meisinger, 2002). The quantity of intramuscular fat is affected by many factors including animal breed, slaughter weight (Park et al., 2002), feeding strategy (Du, Yin, & Zhu, 2010), and growth rate (Smith et al., 2009). Intramuscular fat increases with advancing age when the major stages of muscle growth have been completed. Deposition of intramuscular fat is highly heritable and is positively correlated with general body fatness in the animal (Hwang et al., 2010).

# 2.4 Factors that may affect pork quality (breeding, genetics, feeding, production systems, handling before and after slaughter, cooling, storage and packing)

The requirements for higher pork quality are becoming higher and higher. From all the factors, genetic base represents the key criterion when determining production targets. The genetic influence on the pork quality includes the differences between breeds and differences between the pigs within the same breed (Rosenvold & Andersen, 2003a). The genetics determines the 20-30% of the quality of meat, and the rest of the quality is determined by other factors (Kocwin-Podsiadło, 2002). To meet market demands, genetic breeding has brought to a gradual reduction of fat content in meat and, a higher proportion of muscle tissue. This adversely affected the quality of pork, especially the flavor and juiciness. Since every breed has own production characteristics, by crossing certain breeding lines desired production goals can be achieved. Thus, Hampshire is often used in crosses for the production of fattening pigs with very low percentage of pale, soft and exudative/dark, firm and dry meat, Pietrain for good conformation, Large White for good fertility, good maternal characteristics and good pork quality, and Duroc, as a terminal breed, for increase of intermuscular fat in muscle tissue.

The feeding is another significant factor for pork quality. To produce a healthier meat, the quality of pork can be altered by different feeding sources. This refers primarily to the fatty acids profile where it is desirable to achieve the higher ratio of polyunsaturated to saturated fatty acids (P: S) and a more favorable balance between n-6 and n-3 polyunsaturated fatty acids (n-6: n-3). It is known that the fatty acids profile affects the technological meat quality by influencing on fat tissue firmness (hardness), shelf life (lipid and pigment oxidation) and flavor. The effect of fatty acids on firmness is due to the different melting points of the different fatty acids, the effect on shelf life is due to the suitability of unsaturated fatty acids to oxidize, develop rancidity and color changing and the effect on meat flavor is due to the production of volatile, odorous, lipid oxidation products combined with Maillard reaction products during cooking. Because of this, a-tocopherol (vitamin E) have been used to delay lipid and color oxidation and to extend shelf life (Wood et al., 2003). Several studies have shown that ruminant meat, especially from animals that have consumed grass (contains high levels of linolenic acid (18: 3)), have better n-6: n-3 ratio compared with pork where, linoleic acid (18: 2) is higher (due to the high content of 18:2 in the cereal-based diets) causing an undesirably high n-6: n-3

ratio and a higher P: S ratio. Although 18:2 is at a higher level, the fatty acids in pork are relatively unsaturated (more suitable for oxidation processes) which is similar with poultry meat (Enser, 1999). It is interesting that the United States pig industry has used this similarity to label pork as the "other white meat" (Wood et al., 2003). Also, there are different opinions about supplementing pig diets with linoleic acid (18:3) (rapeseed oil, linseed) to lower the n-6: n-3 ratio. According to one group of authors, there is no effect on meat quality (Enser et al., 2000; Leskanich et al., 1997) while another group claim that some negative effects on odor and flavor can occur, especially by preparation treatment (Myer et al., 1992; Shackelford et al., 1990). Enser et al. (2000) and Sheard et al. (2000) showed that the n-6: n-3 ratio in pork could be reduced on less than 4 by feeding crushed whole linseed, with no detectable negative effects on meat quality.

Nonetheless, feeding pigs based on additional sucrose and other carbohydrates several days prior to slaughter can significantly increase the glycogen level which reflects positively on pH value of the mature meat and prevent appearance of dark, firm and dry meat (Gardner & Cooper, 1979). Some researchers have shown that addition of magnesium (Mg) prior slaughter can reduce stress response and enhance pork quality (D'Souza et al., 1998b). Diet deficient in protein and amino acids increases marbling, which is desirable because intramuscular fat improves tenderness, juiciness, flavor and taste of the meat (Rosenvold & Anderson, 2003a).

Pigs reared in different production systems respond differently on stress prior to slaughter, which influence the quality of meat (Lebret, 2008). Terlouw et al. (2004) have shown that pigs which are grown extensively are less aggressive when mixed with other unknown pigs, prior to slaughter, compared with pigs which are grown conventionally. This positively affect the pork quality because the level of injury is smaller, the level of glycogen higher in the muscles ante and post mortem and pH lower. Furthermore, extensively and organically grown pigs have a higher yield of meat and a higher content of intramuscular fat compared to the pigs which have been intensively reared (Danielsen et al., 2000; Sundrum et al., 2000).

It is important to treat animals properly in the period prior to slaughter because improper treatment increases the amount of stress and consequently affects the pork quality deterioration. The stress accelerates the normal post-mortem changes, pH decline and glycogen degradation by increasing the hormones adrenaline, noradrenaline and cortisol, glucose concentrations, lactate content, free fatty acids, the ketone body and urea in blood, consequently influencing the deterioration of the pork quality. This involves a lot of activities starting at farm, continuing

during transport and in a slaughterhouse. Some of the stress factors include: high temperature and changes in the speed of the vehicle, noise and overcrowding of animals, deprivation of food and water, injuries, exhaustion, the distortion of the social group and mixing with unknown animals, changes in the environment, and others (Andersen et al., 2005; Adzitey, 2011). Every animal experiences stress differently depending on genetics, gender, age and previous experience. The most sensitive to stress are meaty pigs (Hampshire and Pietrain), female and young animals (Adzitey, 2011).

One of the most important factor for the pork quality is the beginning of the cooling of carcasses that should start within 30 minutes after stunning. The start time of cooling carcasses and cooling intensity are important for the quality of meat from a hygienic and technological aspects. The temperature should be lowered as soon as possible to a value that prevent or slow the reproduction of microorganisms and further development of biochemical processes in the meat (Pisula & Florowski, 2009). From the moment of cooling down, storage, proper care and handling up to the packing plant, packaging and further distribution to the end consumer, have an important role in ensuring the preservation of a high pork quality. According to Grandin (1994) the final pork quality depends 50% on the producer and the same, 50% on the packer.

#### 2.5 Quality measurement

The pork quality measurement can be determined by subjective human visual analysis, by laboratory methods or by rapid, computerized online methods, with a lower or higher degree of accuracy.

Measuring muscle pH and temperature (because temperature affect the pH value) early post mortem are commonly used to identify and predict potential meat quality problems. According to Dalmau et al., (2009) the measurements are usually performed 45 minutes and 24 hours after slaughter, with two to three times repetition to get the average value. During the measurement 45 minutes after slaughter, optimum pH value is larger than 6.1 while, the temperature of the carcass is 37 ° C to 39 ° C. The measurement 24 hours after slaughter gives optimum value for pH between 5.6 and 5.9 and for the temperature of the carcass, 4 ° C.

To determine a muscle's water holding capacity, percentage of lean tissue drip loss is commonly used as the most reliable method (the loss in the meat sample weight due to drip and evaporation, divided by the original sample weight and multiplied by 100) (NPPC, 2000). Evaluation of pork color acceptability can be carried over subjective human analysis or computerized vision analysis. Most consumers object to a fresh pork color when is too pale or too dark. They assume that pale pork is often tough and dry after cooking, while darker pork has been in the display case for a longer period or it was obtained from older animals. Subjective human color analysis has a certain degree of error because humans are prone to tiredness and distraction when they can accept poor quality cuts or reject good cuts. Computerized color analysis can identify and sort acceptable and unacceptable colored cuts without these shortcomings (NPPC, 2000). There are color standards which are used as references such as the Japanese color scale (1 to 6) or the NPPC (2000) color standards (1 to 6) (Morgan & Forrest, 1997). Classification of meat color can also be performed with spectrophotometers (analysis of light reflectance) in a three-dimensional coordinate system where, the CIE L\*, a\*, b\* color scale was designed to represent the human perception of color. The a\* scale is a measure of the relative intensity of red and green, b\* considers the intensity of the colors blue and yellow and L\* represents the overall lightness or darkness (Morgan et al., 1997).

There are also on-line technologies such as: The PH-STAR<sup>™</sup> pistol, The Meatcheck<sup>™</sup>, Fiber optic probes (FOP), real-time ultrasound (RTU) and others, which are capable of rapid, on-line pork quality measurement.

#### 2.6 Boar taint as a major disadvantage of pork quality nowadays

After the appearance and color as the first obvious pork quality traits, odor and flavor are crucial when consumers generate the final decision about the acceptability and desirability of pork. Since ancient times it is known that meat from uncastrated male pigs (boars) may have expressed offensive odor and flavor (Boar taint), due to which it becomes less desirable for the market. Because consumers become increasingly aware about the side effects of the surgical castration, about the importance of animal welfare, more interested in how animals are raised and produced, an increasing number of countries began with raising non-castrated pigs as an

alternative method to avoiding surgical intervention. Consequently, boar taint becomes a major disadvantage of pork quality nowadays.

Boar taint is an unpleasant, offensive odor and flavor that occurs when pork (from entire males) is frying or cooking. This offensive odor is primarily caused by high presence of androstenone (5α-androst-16-en-3-one) and skatole (3-methylindole) even, other compounds such as indole, androstenols and p-cresol may also contribute. Skatole is a product of microbial breakdown of L-tryptophan in the intestine of the pig and androstenone is a steroid hormone produced in the testis after onset of puberty. Scatole has been characterized as having an offensive faecal-like odour, while androstenone is often described as having a urine-like odour. When these compounds cannot be metabolized by the liver (insufficient clearance), they accumulate in the adipose tissue (Bonneau, 1998). More than 10 % of the sexually mature, non-castrated male pigs carry boar tainted meat (meat with above 1.0 µg androstenone / mg fat and/or 0.25 µg skatole / mg fat) (Bonneau et al., 1992). The most commonly used threshold values to categorize tainted meat for androstenone are 0.5 and 1.0 µg/g of adipose tissue (Claus et al., 1994; Font i Furnols et al., 2003); and for skatole 0.10 and 0.20 µg/g of adipose tissue (Bonneau et al., 1992; Claus et al., 1994; Font i Furnols et al., 2003). Androstenone and skatole level increases with sexual maturity of boars and they can be also found in barrows and gilts but in low concentrations.

#### 2.6.1 Metabolic pathways of skatole and androstenone

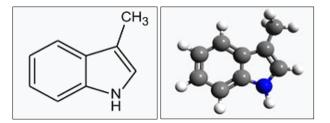


Figure 1. The chemical structure of skatole (Available at: <u>https://en.wikipedia.org/wiki/Skatole</u>).

#### 2.6.1.1 Skatole metabolism

Skatole occurs in the intestine of the pig because of microbial breakdown of L-tryptophan amino acid. As it can be seen from the graph below (Figure 2), after the absorption in the gut

wall, metabolite of L-tryptophan goes to the liver through the blood stream. If the degree of synthesis of skatole is greater than the clearance of the liver, that leads to its accumulation in the adipose tissue (also accumulation of boar taint). Otherwise, skatole gets metabolized by the liver and excreted through the excretory system (kidney or bile) (Babol et al., 1998a). Skatole metabolism is conducted by two phases in the liver even though, the second phase is not fully elucidated (Parkinson, 2001) and should be an area for further investigation. The major metabolites from the first phase of skatole metabolism are 3-OH-3-methylindolenine, 3methyloxindole and 3-OH-3-methyloxindole (Babol et al., 1998a). It has been shown that from metabolizing enzymes, the most important for skatole clearance are enzymes from cytochrome P450 (CYP) family such as CYP2A19 and 2E1 but, CYP1A2, 2C33 and 3A can also metabolize skatole, as well as aldehyde oxidase (Babol et al., 1998a). Some studies have been performed where high CYP2A19 and 2E1 enzymatic activity is in negative correlation with skatole concentrations (Zamaratskaia et al., 2006). Moreover, CYP1A, 2A and 2E expression and activity have been shown to be low in entire male pigs compared to females (Zamaratskaia et al., 2006) and surgically castrated pigs (Zamaratskaia et al., 2009). Thus, to find a solution for the boar taint problem, there is a need to gain more knowledge about the regulation and activity of these CYPs enzymes.

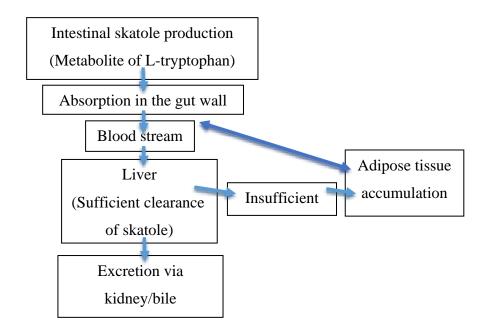


Figure 2. Skatole metabolism (Zamaratskaia & Squires 2009).

#### 2.6.1.2 Androstenone metabolism

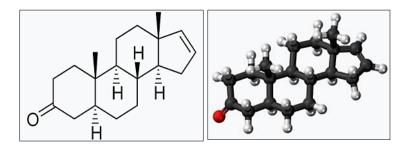


Figure 3. The chemical structure of androstenone (Available at: <u>https://en.wikipedia.org/wiki/Androstenone</u>).

Androstenone is produced in the Leydig cells of the testis. From there, via the blood stream, it enters either into the salivary glands, or into the liver where becomes metabolized or, in case of insufficient liver clearance, androstenone accumulates in the adipose tissue (Figure 4). Metabolic processes are controlled by expression of many genes that encode enzymes involved in liver metabolism (Zamaratskaia & Squires, 2009). The liver metabolism is conducted by two phases. In the first phase, enzyme 3α-hydroxysteroid dehydrogenase (HSD) is responsible to convert and rostenone to  $3\alpha$ -and rostenol while enzyme  $3\beta$ -HSD converts and rostenone to  $3\beta$ androstenol. Researches have shown that the major part of androstenone is converted to 3βandrostenol (Sinclair et al., 2005). Also, it has been shown that low hepatic  $3\beta$ -HSD expression and activity is associated with high concentrations of androstenone (Nicolau-Solano et al., 2007). However, the manipulation with porcine  $3\beta$ -HSD gene is still not fully elucidated (Dong et al., 2012). In the second phase, metabolizing enzymes (sulfotransferase- SULT2A1, SULT2B1 and uridine diphosphate glucuronosyltransferase-UGT) convert further the androstenols to sulfoconjugated and glucuronidated liver metabolites, which are finally excreted through the excretory system. Next to this, Sinclair et al. (2006) have found that the hepatic expression and activity of SULT2B1 enzyme is breed dependent. According to the same authors, for SULT2A1 enzyme, it has been shown that its higher rate in pigs was accompanied with low androstenone concentrations. Again, there are still some gaps in knowledge linked to the SULT2A1 gene manipulation and molecular cloning.

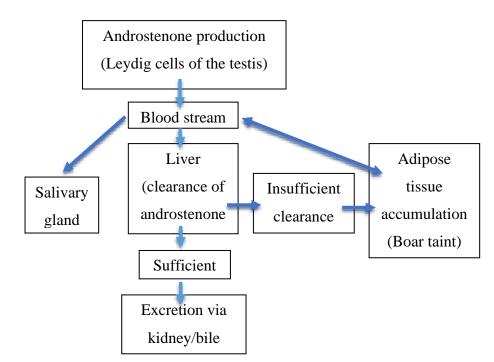


Figure 4. Androstenone metabolism (Zamaratskaia & Squires 2009).

#### 2.6.2 Factors that affect the level of androstenone and skatole

#### 2.6.2.1 Factors that affect the level of androstenone

There are many factors that may affect the level of androstenone. Among the most important are genetics, breed, gender, sexual maturity, age, live weight, liver metabolism, breeding, weather seasons and daylight (Zamaratskaia, 2004).

The genetic base has a significant influence in regulating the intensity of androstenone synthesis. Pigs with a higher share of adipose tissue have a higher level of boar tainted meat. According to Migdal et al. (2009), the highest concentration of androstenone belongs to Duroc pigs while Large White have higher concentration of this compound compared with Landrace pigs (the lowest androstenone carrier). With genetic selection, occurrence of androstenone can be reduced but mostly in parallel with the reduction of the reproductive capacity and growth rate. Androstenone and skatole concentration increases with the age and live weight and depends on the individual ability of an animal for its synthesis (Bonneau, 1987).

It has been proven that different weather seasons may influence on androstenone level in adipose tissue, blood and seminal plasma of boars. From October to December, the concentration of androstenone is five times higher than in other periods of a year. The reduction of daylight (or artificial lighting) leads to the increase in the concentration of androstenone (Claus et al., 1983).

Walker (1978) states that if the aim of production is slaughtering of large weight pigs, it is not recommended to keep together boars with gilts during breeding. When they are accommodated together, it may lead to an increased level of androstenone in adipose tissue.

Unlike skatole, it has not been established that diet can affect androstenone level (Lundström et al., 1988).

#### 2.6.2.2 Factors that affect the level of skatole

Skatole concentration depends on genetics, age, gender, diet and breeding (Zamaratskaia, 2004; Lundström et al., 1988). The genetic influence on the skatole level is confirmed by various concentrations of skatole between different breeds (Babol et al., 2004). According to Friis (1993), skatole metabolism and excretion are faster and more effective in female animals which explains the higher level of skatole in the adipose tissue of males. Skatole concentration is in the positive correlation with body mass and age (Walstra et al., 1999).

The main source of tryptophan (for the production of skatole) arises from debris- from the intestinal mucosa, during the replacement process of the intestinal epithelium. Therefore, the main source of skatole does not come from diet (Claus et al., 1994), although it may depend on it (Jensen & Jensen, 1998). It has been shown that high energy diet increases the level of skatole (Claus et al., 2003). Moreover, a diet rich in a protein with a low digestibility stimulates the production of skatole, whereas a large amount of indigestible carbohydrate (starch from raw potato), which bypass the digestion in the small intestine, decreases its production (Jensen et al., 1995). According to Jensen & Jensen (1998), it has been observed that fructooligosaccharides, lupines and raw potato starch have the best effect on the reduction of skatole production. Beside commercial feed, boars fed with raw potato starch, two weeks before slaughter, have shown significantly lower level of skatole in dorsal fat, compared with control group (Zamaratskaia et al., 2003). Also, it has been indicated that inulin from chicory roots, as indigestible oligosaccharide, may affect skatole reduction (Hansen et al., 2006). Gibson & Roberfroid (1995) have classified inulin as a prebiotic due to good health effects on the colon microflora of boars. Inulin reduces the formation of skatole in colon by changing the microbial fermentation (Rideou et al., 2004). According to Claus et al. (2003), skatole level

can be reduced using casein as a protein source in diet. Deprivation of food 26 hours before slaughter may reduce the level of skatole production but, at the same time, it may increase the level of androstenone (Kjeldsen, 1993).

Breeding also influences on skatole production. Thus, free (ad libitum) feeding regime affects higher skatole deposition in adipose tissue compared with restricted feeding regime (Øverland et al., 1995), while free water (and/or wet feed) access leads to lower skatole level compared with dry feeding system (Kjeldsen, 1993). Pens with high amount of feces and urine and high density of animals per m2 may also increase skatole level, especially in summer period (Hansen et al., 1993).

#### 2.7 The surgical castration of male pigs

Surgical castration has been performed usually in the first week of piglets` life, to avoid unpleasant and offensive odor (Boar taint), to prevent undesirable sexual and aggressive behavior of male pigs (Lealiifano et al., 2009). According to European legislation, it was allowed to perform the surgical castration up to 7 days after birth. It has been thought that newborn piglets do not feel the pain due to immaturity and underdevelopment of the nerve structure but, research has disproved this belief (Fitzgerald, 1994). Therefore, surgical castration is a painful and stressful procedure (Earley & Crowe, 2002), which confirms intensive advertising, increased levels of stress hormones and modified behavior of young piglets. The increased level of vocalization during surgical castration is also one of the indicators of pain (Taylor et al., 2001). Changes in behavior as a result from this intervention last for hours or days (Hay et al., 2003). The response to physical castration is similar in individuals aged from 3 to 17 days (Taylor et al., 2001). It should be emphasized that this procedure has been performed for centuries without any pain relief. In recent years, with changing social values, surgical castration is becoming significant animal welfare concern. Confirmation that castration without anesthesia is a painful and stressful experience that undermines the welfare of the animals (Prunier et al., 2006) has been the reason for the abandonment of this procedure in many countries, especially in Europe (Bonneau, 1998). Moreover, surgical castration has another disadvantage such as reduced growth rate, lower feed conversion rate and reduced production performances - fattier carcasses (Bonneau & Squires, 2004).

Performing surgical castration without anesthesia was banned in Norway from 2002 and in Switzerland from 2009. The UK and Ireland have been completely abolished castration while in other European countries it is still partially performed with/without the use of analgesics and/or anesthetics.

According to the ``European Declaration on alternatives to surgical castration of pigs`` (2010), the first step in combating practices castration began 01. 01. 2012, when it was ordered the using of anesthetic and/or analgesic while performing physical castration. The second step requires that the practice of surgical castration should be completely banned from 01. 01. 2018 in European Union member states. In September 2015, The Federation of Veterinarians of Europe together with The European Commission started to collect and analyze results from 24 European countries to determine the progress in achieving the Declaration goals. The results have shown that approximately one third of male pigs (36%) were raised as entire males, the majority (61%) were still surgically castrated and just 3% of male pigs were immunocastrated (Figure 5). This research indicates that achieving the goals of the Declaration of 2018 is still far from being met (Briyne et al., 2016). However, if the boar taint problem is not solved by January 2018, the Brussels declaration could produce a decrease in pork acceptability and consumption, at least for sensitive consumers (Font-i-Furnols et al., 2016).

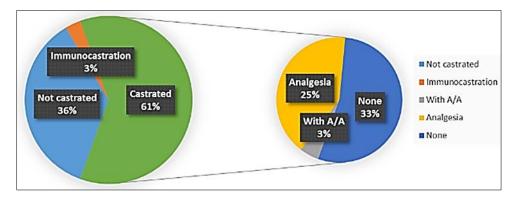


Figure 5. Castration methods in European countries from 2015 (Briyne et al., 2016).

#### 2.8 The main advantages and disadvantages of the production of noncastrated boars

Among others, the main advantages of non-castrated boars are: improved feed conversion rate up to 9%, higher growth rate up to 14% and higher lean meat content of the carcasses for up to 20%, compared with castrates (Babol & Squires 1995; Bonneau & Squires, 2004).

Efficient utilization of feed is essential for economic growth because feed accounts for around 60% to 70% of production costs (FAO, 2017). Therefore, in addition to the well-being of the animals, this way of production is justified from an economic standpoint of view. Also, efficient utilization of feed has less environmental consequences for the planet (OECD-FAO, 2016). Benefits of the meat quality from non-castrated boars are reflected mostly in a reduced proportion of adipose tissue for about 5%, in an increased content of unsaturated fatty acids and in a significant increase in the protein content due to muscle tissue increase (Babol & Squires 1995). Non-castrates deposit less fat because testosterone increases muscle growth and decreases intramuscular lipid deposition. The higher protein content in the meat of noncastrated pigs indicates on a higher nutritional value compared with castrates (Wood et al., 1986). In Europe and North America, a higher ratio muscle to fat is preferable. However, a minimum level of fat is necessary because fat content is associated with flavor development (Warriss, 2010). Miyahara et al. (2004) have found another desirable trait from non-castrates such as redder color and better water binding capacity. From another side, low fat content may give the firmer texture of pork, which is already firm due to years of intensive genetic selection according to the proportion of fat. Also, numerous studies have indicated that, even if boars have the same thickness of the fat as castrates, they will have softer fat because of C18: 2 fatty acids and a higher concentration of water (Wood et al., 1986). An important trait of fats is their resistance to oxidation which is dependent of the degree of saturation of their fatty acids (Warriss, 2010). Therefore, there are some recommendations to restrict unsaturated fatty acids in the diet of boars in the final fattening period.

The significant disadvantages are aggressive behavior and animal fights during the final phase of fattening (Bonneau & Squires, 2004). After reaching the full maturity, boars become aggressive, which often leads to the occurrence of injuries, bruises and skin abrasions. It is recommended to reduce the number of manipulations with animals. Also, animals should not interfere with unknown groups and they should be separated by sex, especially in the final

stages of fattening or prior slaughter. Certainly, the biggest disadvantage is the unpleasant odor of boars that occurs during the heat treatment of such produced meat.

## 2.9 Different approaches in combating boar taint as alternative to surgical castration (production of entire males, slaughter at lower weight and immunocastration)

Due to EU legislations and the ``European Declaration on alternatives to surgical castration of pigs``, surgical castration has recently been discontinued in an increasing number of European countries. These countries are currently producing a proportion of entire male pigs: Spain (around 80% of entire male pigs), The Netherlands (60%), Belgium (40%), France (10%) and Germany (10%) (Borrisser-Pairo et al., 2017). Among different approaches in combating boar taint, entire male production (non-castrated boars) has therefore become an attractive alternative to surgical castration. Compared to castrated pigs, entire male pig production gives leaner carcasses, less subcutaneous fat, a higher protein content (more profitable for farmers) (Lundström et al., 2009), better feed conversion ratio, higher growth rate (more economical production) and advantages within animal welfare issues. However, the occurrence of boar taint in entire male production remains a concern.

Therefore, some producers practiced slaughter of entire males at less weight gain or before the onset of puberty (Aldal et al., 2005). By slaughtering animals with less weight gain, boar taint is reduced but not removed completely (Aldal et al., 2005). In such case the main factor responsible for boar tainted meat is skatole (Bonneau et al., 1992).

Among some Australian producers, processors and retailers there is a belief that if entire male pigs are slaughtered below 90 kg live weight (LW), occurrence of boar taint is negligible. Therefore, there is no need for the implementation of surgical castration as well as for the implementation of immunocastration (D'Souza et al., 2011). On the other hand, it has been shown by the same authors that regardless of slaughter weight (73 to 115 kg LW), there is still a high proportion of pork samples exceeded the sensory threshold for androstenone and skatole.

The other alternative is immunocastration or vaccination, which gives opportunities for achieving similar performance results as entire males while eliminating boar taint. This method

uses the pig's natural immune system to prevent production of male hormones. Immunization against gonadotrophin releasing hormone (GnRH), by using Improvac® vaccine, is an effective way to eliminate boar taint and to reduce fail rates in pork from entire males. With this procedure, after the second immunization, skatole and androstenone levels in the subcutaneous fat become progressively reduced (Dunshea et al., 2001). The vaccine is a synthetic analogue of gonadotrophin releasing hormone coupled to a large carrier protein. Produced antibodies against gonadotrophin releasing hormone lead to testis regression and reduction in synthesis and accumulation of boar taint-causing androstenone (Zamaratskaia et al., 2008) and skatole (Zamaratskaia et al., 2012). Two vaccinations are needed for a full effect. The first one is intended at priming the animal immune system (administrated around 10 weeks of age) and, the second vaccine is intended when pigs reach their sexual maturity (about 4 to 6 weeks before slaughter), as the standard protocol recommended by vaccine producer. Before the second vaccination pigs behave like entire males while, after the second vaccination the pigs behave like physically castrated- barrows (Aluwe et al., 2016). As literature indicates, about one week after the second vaccination, feed intake (Weiler et al., 2013) and daily gain (Millet et al., 2011) increases and, fat deposition increases after 4 weeks (Dunshea et al., 2013).

Therefore, the timing of the second vaccination is crucial to find the balance between the advantages of entire males and barrows (Aluwe et al., 2016). This was the subject of research by the same authors, to evaluate the effect of the timing of the second vaccination (V2) on behavior, performance, carcass and meat quality, within the suggested standard timing of 4 and 6 weeks before slaughter. In total, 180 pigs (Pietrain x hybrid sow) were used in this experiment: 60 gilts, 60 male pigs vaccinated 6 weeks before slaughter (IM-6) and 60 male pigs vaccinated 4 weeks before slaughter (IM-4). All entire male pigs received the first vaccination (V1) at 14 weeks of age while, timing for the second vaccination was based on average pen weight of 75 kg for pigs vaccinated 6 weeks before slaughter and 85 kg for pigs vaccinated 4 weeks before slaughter. The pigs had free access to water and were fed ad libitum (with a three-phase feeding strategy). Pigs were sent to the slaughter house at comparable slaughter weight, when the average live weight of the pen was 117 kg (Figure 6).

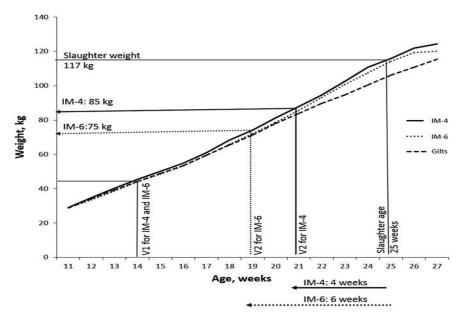


Figure 6. Experimental design (Aluwe et al., 2016).

Before the age of 20 weeks (before receiving the second vaccination), pigs vaccinated 6 weeks before and pigs vaccinated 4 weeks before slaughter showed significantly more aggressive behavior compared with gilts, as was expected. Pigs vaccinated 6 weeks before slaughter showed more sexual behavior than gilts. After the age of 20 weeks (after the second vaccination), aggressive behavior decreased for both groups. Between the pigs vaccinated 6 weeks before slaughter and gilts were no differences while, more aggressive behavior was still observed for pigs vaccinated 4 weeks before slaughter in comparison with gilts. Playing and aggressive behavior tended to reduce for pigs vaccinated 6 weeks before compared with pigs vaccinated 4 weeks before slaughter. In the last feeding phase, daily feed intake and daily gain were significantly lower for gilts compared with pigs vaccinated 6 weeks before and pigs vaccinated 6 weeks before slaughter. The earlier increase of feed intake was observed in pigs vaccinated 6 weeks before slaughter, feed intake was higher for pigs vaccinated 4 weeks before slaughter. During the last week before slaughter, feed intake was higher for pigs vaccinated 4 weeks before slaughter, feed intake was higher for pigs vaccinated 4 weeks before slaughter.

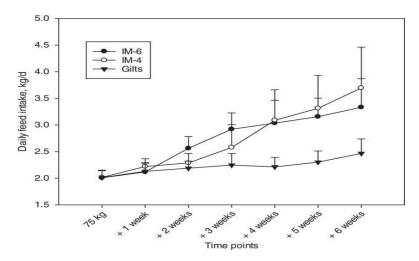


Figure 7. Weekly evolution of daily feed intake (Aluwe et al., 2016).

Carcass leanness and back fat measurements did not differ significantly between experimental groups. Dressing percentage was lowest for pigs vaccinated 4 weeks before, intermediate for pigs vaccinated 6 weeks before slaughter and the highest for gilts. The weight of gastrointestinal tract was lowest for gilts, intermediate for pigs vaccinated 6 weeks before and the highest for pigs vaccinated 4 weeks before slaughter. Testes weights did not differ significantly between pigs vaccinated 6 weeks before and pigs vaccinated 4 weeks before slaughter (Table 1).

Extending the time post second injection by only two weeks, daily feed intake, daily gain, feed conversion ratio, meat quality traits and palatability also did not differ significantly between pigs vaccinated 6 weeks before and pigs vaccinated 4 weeks before slaughter. Earlier vaccination increased dressing percentage and improved animal behavior in late finishing (Aluwe et al., 2016).

	IM-6	IM-4	Gilts	SEM	<i>P</i> -value
n	58	56	60		
Slaughter weight (kg)	119.7	119.8	117.7	0.6	0.164
Cold carcass weight (kg)	93.4	92.8	93.8	0.5	0.753
Fasting weight loss (%)	2.5 <sup>b</sup>	2.7 <sup>b</sup>	1.5 <sup>a</sup>	0.1	< 0.001
Dressing percentage (%)	78.0 <sup>b</sup>	77.4ª	79.7	0.1	< 0.001
Gastrointestinal tract (kg)	7.9 <sup>b</sup>	8.2 <sup>c</sup>	6.9 <sup>a</sup>	0.1	< 0.001
Testes weight (g)	278	260		11	0.421
Lean meat (%)	64.0 <sup>b</sup>	63.5 <sup>b</sup>	65.6ª	0.2	< 0.001
Fat thickness (mm)	8.5 <sup>b</sup>	8.7 <sup>b</sup>	6.8 <sup>a</sup>	0.2	< 0.001
Muscle thickness (mm)	67.1 <sup>b</sup>	67.5 <sup>ab</sup>	68.6ª	0.4	0.030

Table 1. Carcass traits of immunocastrates (IM-6 and IM-4) receiving V2 and gilts.

Source: Aluwe et al., 2016.

The objective of another study, which was carried out by the Dunshea et al. (2001), was also to evaluate the efficacy of Improvac vaccine in eliminating boar taint. Three hundred male pigs, 200 intact boars (boars treated with placebo or boars treated with Improvac) and 100 barrows, crossbred (Large White x Landrace) were used in experiment. Vaccines were administered 8 and 4 weeks before slaughter and slaughter age was 23 or 26 weeks. The concentration of boar taint in 100% of Improvac treated pigs were negligible or undetectable and not significantly different from those in barrows (male pigs castrated before puberty). While, 10 % of the control boars had high concentrations of both compounds. After the second vaccination, over the 4 weeks, Improvac treated boars grew more rapidly than control boars. Also, they were leaner and had better feed conversion efficiency.

At the time of the second vaccination, 85% of the treated boars and 2% of barrows had a serum testosterone concentration of > 2 ng/mL (as biologically significant). Within two weeks of the second vaccination, just 6 % of treated boars had testosterone concentration above 2 ng/ mL and at least 4 weeks after the second vaccine, 8 % of treated boars had testosterone concentration above 2 ng/mL. The highly significant reduction in testosterone concentration and the suppression in testicular growth, for at least 4 weeks of secondary vaccination, were confirmed by the approximately 50 % decrease in testes and bulbo-urethral gland weights and length at slaughter (Dunshea et al., 2001). In the Improvac treated pigs, the testosterone reduction did not have a detrimental effect on growth performance. Indeed, they had higher weight gain following the second dose and, over the 4 weeks, Improvac treated boars grew more rapidly than control boars. Also, they were leaner and had better feed conversion efficiency (Dunshea et al., 2001).

Although the efficacy of Improvac vaccine has been demonstrated many times, some researches showed that a small percentage of pigs may escape the immunocastration (Dunshea et al., 2001). That may happen either because of bad administration of the vaccine, a non-response to the vaccination in some pigs or due to some health problems (Font-i-Furnols et al., 2012). Also, there are some more disadvantages of imunocastraton such as price of the treatment, possible rejection of consumption due to the presence of hormones in such meat and possible risk of self-injection of the vaccine during the implementation of the treatment.

However, Font-i-Furnols et al., (2016) investigated the relationship between physical characteristics of the testes (volume and density) and boar taint compounds, as well as the

levels of boar taint compounds in entire (EM) and immunocastrated (IM) males during their growth. In their study, a total of 44 Pietrain  $\times$  (Landrace  $\times$  Duroc) pigs (4 entire males and 20 IM) were scanned by computed tomography, at several body weights (30, 70, 100 and 120kg), and after slaughtered to determine androstenone and skatole level. The first dose was applied at 12 weeks of age and the second vaccine at 18 weeks of age or 6 weeks before slaughter.

The distribution of androstenone and skatole levels according to live weights for immunocastrated and entire males is shown in Figure 8. In entire males androstenone content increased along with body weights, significantly between 70 and 120 kg while, the opposite was observed in immunocastrates due to administered second vaccine at 70 kg total body weight. The relationship between body weight and androstenone level is not very strong because androstenone and its variability increase at the same time with the weight of entire male pigs. Skatole and indole levels were significantly lower in immunocastrated than entire male pigs. Increased levels of skatole in adult entire males are related with the reduction of skatole metabolism in the liver due to inhibition of its metabolizing enzymes by testicular steroids (Doran et al., 2002; Zamaratskaia et al., 2008a).

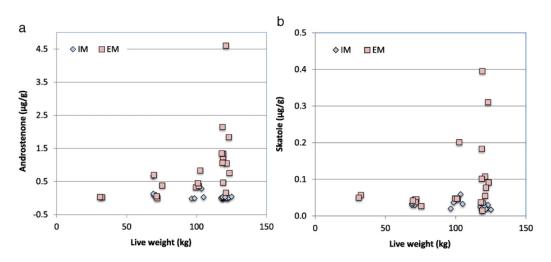


Figure 8. Distribution of androstenone (a) and skatole (b) content according to live weight for immunocastrated (IM) and entire male pigs (EM) (Font-i-Furnols et al., 2016).

Testes density which changes with growth was different in entire and immunocastrated males but, is not a reliable marker of the level of boar taint compounds. Testes volume of entire male pigs increased with total body weight, while testes volume of immunocastrated pigs decreased after 70 kg total body weight at the time when the second vaccine was administered. Testes to body volume ratio (Figure 9) is much better predictor for androstenone and could be a good tool at slaughter plants (as online method) to detect immunocastrated pigs with high boar taint compounds (Font-i-Furnols et al., 2016).

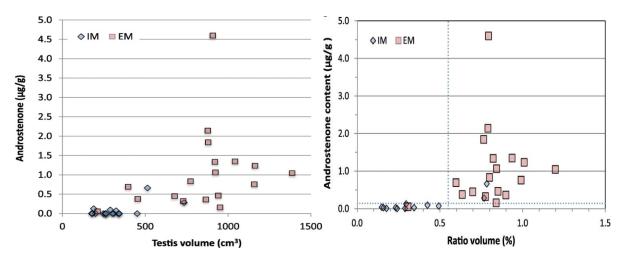


Figure 9. Relationship of androstenone with testes volume and with testes to body volume ratio (%), of IM and EM between 70 and 120 kg body weight (Font-i-Furnols et al., 2016).

The correlation between skatole content and testes volume was high, although the correlation between skatole and androstenone was moderate. Also, testes to body volume ratio was well correlated with skatole content (Figure 10). However, contrarily to androstenone, it could not be used to differentiate pigs according to its skatole level because an important number of pigs with low skatole levels had volume ratio values higher than 0.55%. Thus, volume ratio was more related with androstenone than skatole content (Font-i-Furnols et al., 2016).

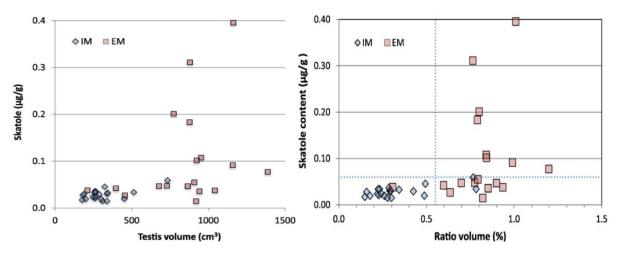


Figure 10. Relationship of skatole content of fat tissue with testes volume, and with testes to body volume ratio of IM and EM pigs, between 70 and 120 kg body weight (Font-i-Furnols et al., 2016).

Thus, a method to determine volume ratio online may provide a good tool at slaughter plants to detect pigs with high androstenone levels indicating non-effective immunocastration (Fonti-Furnols et al., 2016).

Further, Moore et al. (2017) explored and compared the pork quality of entire male pigs (EM) and pigs immunized against gonadotrophin releasing hormone (IC), related to body weight and feeding regime. Experiment was conducted on 64 Large White × Landrace × Duroc entire male and immunocastrated male pigs, at both light (64.8 kg) and heavy (106 kg) live weights and with two feeding regimes (ad libitum and restricted). The experiment has shown that there was no difference in final live weights between entire males and immunocastrated males. Feeding regime and initial weight had no effect on skatole neither on androstenone. Pigs fed ad libitum were just heavier than those fed the restricted diet. There was no difference in drip loss between entire and immunocastrated males when pigs were subjected to restrictive diet while, when they were fed ad libitum, immunocastrated males had 29% less drip loss than entire males. Light pigs had a greater drip loss and cook loss compared to heavy pigs. Pork from immunocastrated males had shown better quality grade than entire males.

Entire male pigs had significantly increased concentrations of skatole and androstenone than immunocastrated males, as expected. The threshold for skatole was exceeded by 31% of the heavy entire males and by 12.5% of the heavy immunocastrated males. The threshold level for androstenone was exceeded by 12.5% of heavy entire males. The sensory detection threshold for androstenone was not exceeded by light weight pigs while for skatole it was exceeded by 18.7%. Actually, 37.5% of the light entire male pigs fed ad libitum had skatole levels that exceeded the sensory threshold of  $0.2 \,\mu\text{g/g}$  which was a confirmation that the boar taint is still an issue at low carcass weights of entire males.

In the same experiment, it was shown that immunization against gonadotrophin releasing hormone is effective in eliminating boar taint and in reducing pork quality fail rates by approximately 10% compared to pork from entire males. Fail rates (score below 3) were lower 9.1% for quality grade and 12% for re-purchase intention in immunocastrated males (Table 2).

	Quality grade				Fail rate (% < 3)	P-value	
	1	2	3	4	5		
Entire male	5.6	24.2	37.1	27.4	5.6	29.8	0.007
IC <sup>a</sup> male	0.8	19.8	39.7	31.0	8.6	20.7	
	Re-purchase intention				Would not re-purchase (% < 3)		
	1	2	3	4	5	re purchase (w + 3)	
Entire male	14.5	24.2	24.2	21.0	16.1	38.7	0.001

 Table 2. Percentage of consumer scores for quality grade and re-purchase intention for entire males and IC males.

Source: Moore et al. (2017).

Another group of researchers, Pinna et al. (2015), were also investigated the effects of immunocastration but, in heavy male pigs. Italian heavy pig breeders were concerned about the appropriate vaccination scheme for their long-living pigs which were destined for the production of typical dry-cured ham. Such heavy pigs, slaughtered at 9–10 months of age and 160–170 kg live weight, were more susceptible to the onset of boar taint due to the increase of androstenone and skatole levels with age (Zamaratskaia et al., 2012). In this study, they were investigated the effects of two and three doses of immunization vs surgical castration on the quality traits of raw and dried hams. A total of 60 Landrace × Large White male pigs, with average live weight of  $165 \pm 10$  kg, were divided in three groups of 20 pigs (surgically castrated (SC) group, group with two (IC2) and group with three (IC3) doses of vaccine).

After completion of comparisons and researches, the results from green ham measurements showed that there were no differences between ham weights, although legs from immunocastrates appear to outweigh samples from surgically castrated pigs and thighs from the group with two doses of vaccine had greater weight loss after chilling than thighs from surgically castrated pigs. In this experiment, as a measure of fat unsaturation, an iodine number slightly above 70 was detected in all groups without differences. According to Barton Gade (1987), an iodine number above 70 is a marker of soft fat or highly unsaturated fats which are readily susceptible to oxidation and rancidity of the final product.

Further, the value for the final processing weight loss of dry-cured hams was highest in group with two doses and lowest in group with three-vaccine doses, as important for the ham industry from the financial point of view. Administering a third treatment at 4–5 weeks before slaughtering was beneficial for fresh ham traits such as: an adequate adipose ham covering, prevention from the excessive weight losses or too lean hams and preservation of the typical

ham shape by the end of maturation (Nanni Costa et al., 2008). Pigs with three-vaccine doses were always in the lowest class, according to concentration of skatole and androstenone while, pigs with two-dose vaccination in 18% of cases had more of skatole or androstenone levels (Table 4). Pigs with three-vaccine doses were the closest with those surgically castrated whose androstenone level was found more than 1  $\mu$ g/g in 2% of cases.

In Table 3, data from chemical analysis of belly adipose tissue, according to skatole and androstenone concentration, are arranged in classes starting from low class (below 100 ng/g (or < 0.1  $\mu$ g/g) for skatole and below 500 ng /g (or < 0.5  $\mu$ g/g) for androstenone) and upward. Thus, it was concluded that three-dose vaccine treatment may be a reliable alternative to surgical castration even when dealing with boar taint in the production of Italian dry-cured hams, obtained from heavy pigs in their full adulthood (Pinna et al., 2015).

Treatment group <sup>1</sup>		Androstenone (ng/g)					
	Skatole (ng/g)	<500	500-1000	>1000			
SC	<100	98	0	0			
	≥100	0	0	2			
IC2	<100	82	8	8			
	≥100	2	0	0			
IC3	<100	100	0	0			
	≥100	0	0	0			

Table 3. Skatole and androstenone content in SC and IC male pigs (belly fat).

<sup>1</sup> SC: surgically castrated; IC2: two-doses and IC3: three-doses of vaccine.

Source: Pinna et al. (2015).

The objective of another study, which was conducted out by Elsbernd et al. (2016), was to evaluate pork quality and sensory characteristics of fresh and frozen pork from gilts (G), physical castrates (PC), entire males (EM), and immunocastrates (IC). The survey included a total of 46 pigs, four treatments: G, PC, EM, and IC with a total of 11 or 12 pigs per treatment. All pigs were fed the same diet, ad libitum and housed individually. Vaccination was carried out at the 13th and 18th weeks of age. Pigs were slaughtered at heavier body weight of 145.0  $\pm$  1.3 kg. After aging for 10 days at 4 °C, loins were cut to make a roast which was vacuum packaged and frozen at -20 °C for a minimum of 14 days to a maximum of 30 days before analysis. The second half of the loin was cut into chops, weighted and used for fresh analysis. According to the obtained results, sexes did not differ in any of the pork quality characteristics (juiciness, chewiness, and tenderness) measured for each cut - chop or roast, or method - fresh or frozen. The only significant difference was marbling in frozen samples with physical

castrates having the most marbling and gilts and entire males having the least. Entire males had the highest scores among the sexes for boar odor in fresh chops and roasts and in frozen chops and roasts, compared with gilts, physical castrates and immunocastrates - not different from one another. Regardless of sex, for tenderness it was observed that chops were less tender and chewier than roasts, the frozen chops were the most tender and the least tender were the fresh chops. For chewiness, the results showed that the frozen chops were the least chewy and fresh chops were the chewiest. Off-flavor was also significantly different among cuts, with the chop having more off-flavor than the roast. Only juiciness was affected by freezing. The fresh chops were the juiciest and, frozen chops and/or roasts were the least juicy (Elsbernd et al., 2016).

Among European countries, information about the prevalence of boar taint and the consumer acceptability for tainted meat, may be useful for the European pig sector to find an adequate way to cope with it. There is one such study conducted by Borrisser-Pairó et al. (2016) in pigs from commercial farms in Spain. The objective of the study was to know the prevalence of boar taint in Spanish entire male production. A total of 903 samples of subcutaneous fat have been collected in 5 Spanish regions (Aragón, Catalonia, Castilla y León, Madrid/Castilla-La Mancha and Murcia), from 6 farms for each region. Samples destined for later chemical analysis for androstenone and skatole levels were at first selected by human nose methodology. Those classified as ``presence of boar taint`` were analyzed for androstenone by using the gas chromatography-mass spectrometry (GC-MS) and for skatole by high-performance liquid chromatography (HPLC). Detection limits of the panelists were 0.20  $\mu$ g/g of adipose tissue for androstenone and 0.05  $\mu$ g/g of adipose tissue for skatole. From the total number of samples, 10.2% showed values over the high-level threshold for androstenone and/or for skatole while 78.9% showed values below the low threshold. Carcass characteristics varied significantly between the different Spanish regions. Thus, Castilla y León was the region with the highest hot carcass weight, Catalonia with the lowest subcutaneous fat thickness while, the region with higher skatole concentration was Murcia. Genetic crossbreeds sampled were mainly Pietrain × (Landrace × Large White) (73, 4%) while, Duroc crossbreeds were only concentrated in Murcia (33%). It is known that Duroc accumulates more skatole compared with Pietrain (Xargay et al., 2010), which explains the higher skatole level in Murcia region. In such a way, it can be concluded that differences between carcass characteristics, androstenone and skatole levels, may be due to the different genetic crossbreeds used in the different regions.

According to Mathur et al. (2012), human nose can be useful to evaluate large numbers of samples but is not completely reliable method. Due to some false negatives which appearing, sensory assessment provided by trained people at slaughterhouses should be replaced with harmonized online method with higher accuracy and repetitiveness in boar taint detection. The main goal is to direct tainted meat for further processing where it can be masked and forwarded as a final product on the relevant market.

#### 2.10 Consumers` acceptance of boar tainted meat

Many consumer studies have been carried out to detect the acceptability of pork from entire male pigs. Thus, the objectives of one study, which was carried out by Panella-Riera et al. (2016), were to evaluate sensory acceptability of meat from entire male pigs in two consumer studies: Spain and United Kingdom, France and Italy, and to identify potential niche markets for meat with different boar taint levels. A total of 558 consumers from the different European countries participated in two consumer tests. They were required to eat pork on a regular basis and were stratified by age and sex. Test 1 was performed in Spain (n = 126) and United Kingdom (n = 146), and test 2 was performed in France (n = 139) and Italy (n = 140). Each test had 3 types of pork: meat from gilts (FE), low boar tainted meat (LBT), and a third type was medium boar tainted meat (MBT) or high boar tainted meat (HBT). After defrosted and cut, samples were cooked and salted before consumer's evaluation.

According to Panella-Riera et al. (2016), the acceptability of boar tainted pork can depend on the habit of eating meat from castrated or entire male pigs, procedures for preparing the meat and the ability of consumers to perceive androstenone and/or skatole. Thus, different habits of European consumers, regarding pork consumption, show different acceptability of boar tainted pork. For example, in Spain, United Kingdom and Italy over than 90% of consumers ate fresh pork more than 2 times per week while, this percentage was considerably lower in France (around 35%). In all countries, women were more responsible for buying fresh pork and for cooking at home than men. An exception was observed in France where percentages were very similar, 49.6% women and 50.4% men cooked at home. The most consumed product was dry cured ham in Spain and Italy, cooked ham in France and sliced bacon in United Kingdom. Due to practical reasons, meat was collected in Spain and France and after chemical analysis of androstenone and skatole levels, was distributed to two other countries.

The results were presented considering consumers from the two countries together because there was no significant interaction between the boar taint level and the country. In consumer test 1 (where consumers are used to eat boar meat), meat from gilts and medium tainted was more favorable than low boar tainted meat. The strongest odor was observed in medium tainted meat and gilts, and the highest strength of taste was observed in medium tainted compared to low boar tainted meat. In consumer test 2 (where consumers are not used to meat from entire mails), high boar tainted meat was rated worse than meat from gilts and low tainted meat was scored in between. Three main clusters were identified based on 'How delicious do you find this meat?': 1) Pork lovers (49.5–62.1% of the participants), 2) Boar meat lovers (12.4–21.7%), 3) Reject boar tainted meat (16.2–20.0%).

According to the obtained results, there was a group of consumers which refuse meat with boar taint, but there was also a group that prefer meat with medium or high boar tainted meat, identifying a niche for such meat. Therefore, it is important to obtain knowledge about consumer response towards boar tainted meat to reduce the risk of possible claims.

With the aim of decreasing the fail rate, the objective of another experiment was to determine how to improve consumer acceptability of pork and pork consistency by developing a cut x cooking method-based system. The experiment was conducted by Channon et al. (2016), where it was investigated how the gender (entire male, female and castrate), ageing period (2 or 7 days), cooking method and endpoint temperature (70 or 75 °C) affected the perception of consumers, while judging the eating quality attributes (flavor, tenderness and juiciness) of different roasted, stir fried or grilled cuts (from loin, shoulder and silverside). A total of 60 pigs, females, entire males and castrates (Large White x Landrace) participated in the experiment. Males destined to the castrate treatment were castrated at 1 day of age and all animals were slaughtered at 21-22 weeks of age. In sensory evaluation, it was involved 480 consumers between 18 and 65 years old, who eat pork and have no experience in the meat industry. They evaluated the eating quality attributes of the pork samples using a continuous 0-100-line scale (0 for `dislike extremely` to 100 for `like extremely`). Also, they graded the samples for quality into one of the categories: 1) Unsatisfactory, 2) Below average, 3) Average, 4) Above average and 5) Excellent. Each sample was also rated on a five-point scale for repurchase intention.

Androstenone and skatole concentrations in subcutaneous fat from entire male pigs were higher than for castrates and females (20% of entire males had androstenone levels exceeded 1  $\mu$ g/g

and 15% of entire males had both high, androstenone (exceeding 1  $\mu$ g/g) and skatole (exceeding 0.2  $\mu$ g/g)). Pork from entire male pigs had lower scores for juiciness, flavor, overall liking and quality grade than castrates and females. The fail rate of entire males was 23.0% across all treatments (3.9% and 5.3% higher than from females and castrates respectively). Dressing percentage of entire male carcasses was 2% lower compared with female and castrate carcasses. Loins (Musculus longissimus thoracis et lumborum) from entire males had a higher cooking loss in comparison with females and castrates.

Cooking loin steaks to an endpoint temperature of 75 °C reduced overall liking scores by 5.6 units than cooking to 70 °C compared with the average scores of the other cut type x cooking method combinations. Although it was claimed that neither ageing period nor endpoint temperature influenced any of the eating quality traits evaluated in this study, results show otherwise. Thus, juiciness was better from females aged for 7 days than for 2 days, from entire males also aged for 7 days juiciness was reduced by 5.3 units compared to 2-day aged pork. Loin steaks had the lowest juiciness scores followed by silverside (Musculus biceps femoris) roast, silverside stir fry, loin stir fry and roast, shoulder roast (Musculus triceps brachii) and shoulder stir-fry (Musculus supraspinatus). Also, from all the cut type x cooking methods, aroma, juiciness and flavor of the pork loin steak was influenced by final endpoint temperature, with lower scores for all when cooked to 75 °C compared with 70 °C. The cut type x cooking method also influenced tenderness and flavor. Hence, roast and stir fry cuts from the shoulder were rated highest for tenderness, followed by loin and silverside. Silverside roasts had the lowest flavor scores. Average overall liking and quality grade scores across cooking method (roast and stir fry) were highest for pork from shoulders, followed by loin and silverside.

In this experiment, as was expecting, pork from castrates was juicier and had higher flavor liking scores, overall liking, re-purchase intention and quality grade scores compared with entire males (females were in the middle). Intramuscular fat levels in both the pork loin and silverside from castrated males averaged 1% higher compared with entire males (Channon et al., 2016). Pork from castrates was more acceptable and the average fail rate for quality grade was lower than from entire males. However, castrated males, as already mentioned, have less efficient feed conversion ratio, fattier carcasses, lower growth rate and animal welfare issues, compared with entire males.

In addition to the research conducted by Channon et al. (2016), which showed that different meat cuts from the same carcass with different cooking methods can have different intensities of aroma, flavor, tenderness and juiciness, different meat cuts from the same carcass can also have different intensities of boar taint (Aaslyng et al., 2015).

Therefore, knowledge of the distribution of skatole and androstenone in the pig carcass and the corresponding sensory characteristic is significant to establish reliable sorting and optimized use of the boar tainted carcasses.

Relating to the above, Meinert et al. (2017) have shown the correlation between the concentrations of skatole and androstenone in the neck fat versus different pork cuts (from the same animal) and the perceived sensory characteristics of the cooked meat. The neck fat and the following meat cuts: neck (m. longissimus thoracis), loin, topside (m. semimembranosus), tenderloin (psoas major) and eye of round (m. semitendinosus) were collected from fourteen entire male pigs (with androstenone up to  $3.03 \ \mu g / g$  and skatole level up to  $0.70 \ \mu g / g$ ) for analysis. For sensory evaluation were used nine female trained sensory assessors. The loins were cut into 2 cm chops and fried until a core temperature of approximately 72°C was reached, the topside was cut into 8 mm thick schnitzels and fried until the same core temperature (70-72 °C) while, neck, silverside and tenderloin were cooked as whole roasts until 68–70 °C was reached. The content of skatole, indole and androstenone were analyzed from the neck fat of the 14 entire male pigs. The concentrations were distributed from 0.04 up to 0.7  $\mu g / g$  skatole and from 0.25 up to 3.03  $\mu g / g$  for androstenone (Table 4).

Pig no.	Skatole	Indole	Androstenone		
1	0.04	0.03	0.7		
2	0.04	0.03	3.0		
3	0.05	0.04	2.1		
4	0.06	0.03	1.5		
5	0.09	0.03	0.4		
6	0.12	0.03	0.5		
6 7	0.15	0.03	0.8		
8	0.26	0.05	1.7		
9	0.26	0.09	0.3		
10	0.35	0.13	0.8		
11	0.37	0.10	2.2		
12	0.40	0.03	0.3		
13	0.47	0.04	0.4		
14	0.70	0.09	1.7		

Table 4. Concentrations of skatole, indole and androstenone ( $\mu g / g$ ) in the neck fat from the 14 entire male pigs (the pigs are numbered according to increasing skatole concentrations).

Source: Meinert et al. (2017).

The concentration of skatole in the meat was noticeably lower than in the neck fat for most of the meat cuts (as low as 5% of the concentration in the neck fat), and for some of the samples

below the limit of quantification (LOQ) (Table 5). The concentration of androstenone in the meat was even lower than that of skatole and for many of the pork cuts it could not be quantified (<LOQ).

Pig no.	Neck fat	Neck	Loin	Tenderloin	Eye of round	Topside
Castrate <sup>a</sup>	-	-	<loq< td=""><td><loq< td=""><td><loq< td=""><td><loq< td=""></loq<></td></loq<></td></loq<></td></loq<>	<loq< td=""><td><loq< td=""><td><loq< td=""></loq<></td></loq<></td></loq<>	<loq< td=""><td><loq< td=""></loq<></td></loq<>	<loq< td=""></loq<>
1	0.04	<loq< td=""><td><loq< td=""><td><loq< td=""><td><loq< td=""><td><loq< td=""></loq<></td></loq<></td></loq<></td></loq<></td></loq<>	<loq< td=""><td><loq< td=""><td><loq< td=""><td><loq< td=""></loq<></td></loq<></td></loq<></td></loq<>	<loq< td=""><td><loq< td=""><td><loq< td=""></loq<></td></loq<></td></loq<>	<loq< td=""><td><loq< td=""></loq<></td></loq<>	<loq< td=""></loq<>
2	0.04	<loq< td=""><td><loq< td=""><td><loq< td=""><td>-</td><td>-</td></loq<></td></loq<></td></loq<>	<loq< td=""><td><loq< td=""><td>-</td><td>-</td></loq<></td></loq<>	<loq< td=""><td>-</td><td>-</td></loq<>	-	-
3	0.05	<loq< td=""><td><loq< td=""><td><loq< td=""><td>-</td><td><loq< td=""></loq<></td></loq<></td></loq<></td></loq<>	<loq< td=""><td><loq< td=""><td>-</td><td><loq< td=""></loq<></td></loq<></td></loq<>	<loq< td=""><td>-</td><td><loq< td=""></loq<></td></loq<>	-	<loq< td=""></loq<>
4	0.06	<loq< td=""><td><loq< td=""><td><loq< td=""><td><loq< td=""><td>-</td></loq<></td></loq<></td></loq<></td></loq<>	<loq< td=""><td><loq< td=""><td><loq< td=""><td>-</td></loq<></td></loq<></td></loq<>	<loq< td=""><td><loq< td=""><td>-</td></loq<></td></loq<>	<loq< td=""><td>-</td></loq<>	-
5	0.09	<loq< td=""><td><loq< td=""><td><loq< td=""><td><loq< td=""><td><loq< td=""></loq<></td></loq<></td></loq<></td></loq<></td></loq<>	<loq< td=""><td><loq< td=""><td><loq< td=""><td><loq< td=""></loq<></td></loq<></td></loq<></td></loq<>	<loq< td=""><td><loq< td=""><td><loq< td=""></loq<></td></loq<></td></loq<>	<loq< td=""><td><loq< td=""></loq<></td></loq<>	<loq< td=""></loq<>
6	0.12	0.012	0.011	0.013	0.013	0.008
7	0.15	0.009	0.004	0.009	0.011	<loq< td=""></loq<>
8	0.26	0.011	0.014	0.011	0.014	<loq< td=""></loq<>
9	0.26	0.012	0.009	0.012	0.012	0.008
10	0.35	0.013	0.013	0.013	0.018	0.013
11	0.37	0.017	0.019	0.015	0.028	0.012
12	0.40	0.020	0.015	0.018	0.100	0.018
13	0.47	0.022	0.016	0.020	0.027	0.017
14	0.70	0.027	0.022	0.031	0.033	0.019

Table 5. Concentrations of skatole in the meat cuts ( $\mu g / g$ ) (the concentrations of skatole in the neck fat are shown for comparison).

Source: Meinert et al. (2017).

In this study, a strong link between skatole in the neck fat and in the meat cuts was confirmed. The highest correlation was in tenderloin (0, 95) and the lowest in topside (0, 82) (Table 6). Therefore, the concentrations of skatole in a neck fat samples may be suitable for predicting the concentrations of skatole in the selected muscle cuts. While, the same correlation for androstenone could not be calculated because the quantified amount of androstenone was too low for statistical analysis.

 Table 6. Pearson's correlation coefficient between the concentrations of skatole measured in

 the neck fat and in the meat cuts from entire males.

Correlation	Skatole			
Neck	0.96 (P < 0.0001)			
Eye of round	0.86 (P = 0.0003)			
Tenderloin	0.95 (P < 0.0001)			
Topside	0.82 (P = 0.001)			
Loin	0.91 (P < 0.0001)			

However, the intensity of boar taint related flavor in the cooked meat cuts was generally highest in the pigs with the highest concentration of either skatole or androstenone in the neck fat with some exceptions. For example, the pigs numerated with numbers 9., 10. and 12. had a very low amount of skatole and androstenone in the neck fat and an intense boar taint flavor in the cooked meat (Table 7).

This observation leads to the conclusion that much lower concentrations of skatole and androstenone can be detected in the cooked meat compared to the established thresholds for skatole and androstenone in fat (Meinert et al., 2017).

Table 7. The scores for manure flavor (skatole) and for urine flavor (androstenone) in the cooked meat cuts, respectively (the concentrations of skatole (S) and androstenone (A) in the neck fat ( $\mu g / g$ )).

Pig no.	S	А	Neck	Loin	Tenderloin	Topside	Eye of r	Pig no.	S	Α	Neck	Loin	Tenderloin	Topside	Eye of r
Castrate	-	-	2.2 <sup>cde</sup>	2.5 <sup>cde</sup>	1.2 <sup>c</sup>	0.6 <sup>f</sup>	0.2 <sup>c</sup>	Castrate	-	-	1.9 <sup>cd</sup>	1.9 <sup>de</sup>	1.2 <sup>b</sup>	0.8 <sup>d</sup>	0.8 <sup>c</sup>
1	0.04	0.68	1.9 <sup>de</sup>	2.0 <sup>e</sup>	2.0 <sup>abc</sup>	3.2 <sup>bcdef</sup>	2.3 <sup>bc</sup>	12	0.40	0.25	6.5 <sup>abc</sup>	5.3 <sup>abcd</sup>	5.5 <sup>ab</sup>	5.7 <sup>abc</sup>	7.3 <sup>ab</sup>
2	0.04	3.03			3.4 <sup>abc</sup>	2.2 <sup>def</sup>	-	9	0.26	0.34	1.5 <sup>d</sup>	3.9 <sup>cde</sup>	1.9 <sup>b</sup>	2.3 <sup>cd</sup>	0.9 <sup>c</sup>
3	0.05	2.09	4.5 <sup>bcde</sup>		4.5 <sup>abc</sup>	5.3 <sup>abcd</sup>	-	5	0.09	0.36	1.6 <sup>d</sup>	3.4 <sup>cde</sup>	1.8 <sup>b</sup>	0.6 <sup>d</sup>	1.7 <sup>bc</sup>
4	0.06	1.51	5.3 <sup>bcd</sup>	3.5 <sup>bcde</sup>	2.9 <sup>abc</sup>	3.3 <sup>bcdef</sup>	1.0 <sup>c</sup>	13	0.47	0.43	4.5 <sup>cd</sup>	4.4 <sup>abcde</sup>	3.5 <sup>ab</sup>	3.2 <sup>cd</sup>	5.8 <sup>abc</sup>
5	0.09	0.36	1.3 <sup>e</sup>	2.1 <sup>de</sup>	2.1 <sup>c</sup>	1.2 <sup>ef</sup>	2.0 <sup>bc</sup>	6	0.12	0.48	3.6 <sup>cd</sup>	0.0 <sup>e</sup>	4.8 <sup>ab</sup>	5.2 <sup>abcd</sup>	4.0 <sup>abc</sup>
6	0.12	0.48	3.7 <sup>bcde</sup>			4.9 <sup>bcde</sup>	5.3 <sup>abc</sup>	1	0.04	0.68	2.2 <sup>cd</sup>	1.6 <sup>de</sup>	2.1 <sup>b</sup>	3.6 <sup>cd</sup>	1.9 <sup>bc</sup>
7	0.15	0.82	4.7 <sup>bcde</sup>	3.3 <sup>bcde</sup>	4.4 <sup>abc</sup>	4.9 <sup>bcde</sup>	2.4 <sup>bc</sup>	10	0.35	0.78	3.9 <sup>cd</sup>	6.6 <sup>abc</sup>	3.0 <sup>b</sup>	3.0 <sup>cd</sup>	3.3 <sup>abc</sup>
8	0.26	1.73	6.1 <sup>abc</sup>	6.7 <sup>ab</sup>	5.3 <sup>abc</sup>	5.9 <sup>abcd</sup>	6.8 <sup>ab</sup>	7	0.15	0.82	3.9 <sup>cd</sup>	4.1 <sup>bcde</sup>	5.8 <sup>ab</sup>	4.5 <sup>bcd</sup>	2.3 <sup>abc</sup>
9	0.26	0.34		5.5 <sup>abcd</sup>	2.6 <sup>abc</sup>	3.3 <sup>bcdef</sup>	0.2 <sup>c</sup>	4	0.06	1.51	5.4 <sup>abc</sup>	4.9 <sup>abcd</sup>	4,2 <sup>ab</sup>	4.1 <sup>bcd</sup>	1.0 <sup>c</sup>
10	0.35	0.78	5.8 <sup>abcd</sup>	6.1 <sup>abc</sup>	2.7 <sup>abc</sup>	2.8 <sup>cdef</sup>	4.7 <sup>abc</sup>	14	0.70	1.72	9.8 <sup>ab</sup>	8.5 <sup>ab</sup>	8.3 <sup>a</sup>	<b>9.4</b> <sup>a</sup>	<b>8.1</b> <sup>a</sup>
11	0.37	2.18	7.8 <sup>ab</sup>	<b>7.9</b> <sup>a</sup>	6.6 <sup>ab</sup>	6.9 <sup>ab</sup>	6.6 <sup>ab</sup>	8	0.26	1.73	6.0 <sup>abc</sup>	6.9 <sup>abc</sup>	6.6 <sup>ab</sup>	5.7 <sup>abc</sup>	5.3 <sup>abc</sup>
12	0.40	0.25	<b>7.6</b> <sup>a</sup>	8.0 <sup>a</sup>	6.0 <sup>abc</sup>	6.4 <sup>abc</sup>	9.3 <sup>a</sup>	3	0.05	2.09	4.8 <sup>bc</sup>	6.9 <sup>abc</sup>	4.0 <sup>ab</sup>	4.9 <sup>abcd</sup>	-
13	0.47	0.43	4.3 <sup>bcde</sup>	6.5 <sup>abc</sup>	3.7 <sup>abc</sup>	3.6 <sup>bcdef</sup>	6.2 <sup>ab</sup>	11	0.37	2.18	9.3 <sup>a</sup>	8.9 <sup>a</sup>	5.7 <sup>ab</sup>	8.2 <sup>ab</sup>	6.2 <sup>abc</sup>
14	0.70	1.72	<b>9.9</b> <sup>a</sup>	<b>8.6</b> <sup>a</sup>	<b>8.3</b> <sup>a</sup>	<b>9.0</b> <sup>a</sup>	<b>9.5</b> <sup>a</sup>	2	0.04	3.03	4.2 <sup>cd</sup>	5.4 <sup>abcd</sup>	4.2 <sup>ab</sup>	4.1 <sup>bcd</sup>	-

Source: Meinert et al. (2017).

In another study, conducted out by Aaslyng et al. (2016), the consumers response towards meat from different parts of the carcass, originating from castrates and entire male pigs, was again in the correlation with skatole and androstenone content in the back fat. The overall aim was to assess consumer liking for boneless chops (m. longissimus thoracis et lumborum) and schnitzels (m. semimembranosus), with androstenone content up to 9.4  $\mu$ g /g and skatole content up to 0.92  $\mu$ g /g (determined by chemical and human nose analysis from the back fat). A total of 64 entire males and 25 castrates were selected for the analysis from a Danish commercial slaughterhouse in 2013. Castrates were included as a control group. The thresholds for skatole and androstenone were chosen in a high range to obtain as large variation as possible (between 'below 0.05  $\mu$ g /g' and 'above 2  $\mu$ g /g' for androstenone and 'below 0.1  $\mu$ g /g' and 'above 0.4  $\mu$ g /g' for skatole). Chops were sliced on 2 cm thick and schnitzels on 8 mm thick. Both were fried to a core temperature of approximately 70–72 °C. In total, 181 consumers were participated for the chops assessment and 164 consumers for the schnitzels assessment, aged between 18 and 70 (all of them ate pork at least once a month).

These two studies were performed with two different groups of consumers and cannot be compared directly. However, the meat was assessed in sessions from up to 11 consumers per each session. They were asked to evaluate general liking with emphasis on odor and flavor. Moreover, a trained nine assessors, all female, aged 41 to 68, with up to 20 years of experience in assessing pork, were used for sensory descriptive analysis (performed on one sample from each animal).

Based on the obtained results, there was no difference in juiciness between chops and schnitzels from castrates and entire males. The most important attribute for the consumers when they assessed the chops was tenderness. For the schnitzels, the effect of tenderness was smaller and insignificant. The chops from castrates were more tender in comparison with chops from the entire males. Independently of the androstenone and skatole content, consumers generally preferred meat from castrates. When investigated the effect of androstenone and skatole on liking/disliking, it has been shown that androstenone was mainly responsible for the very few negative reactions (very strong dislike), while skatole was more responsible for the negative reactions in general. Androstenone had a greater effect on flavor than on odor, while skatole had a similar effect on both, odor and flavor. Skatole had a slightly greater effect on the chops than androstenone, while androstenone had a greater effect than skatole in the schnitzels. For the trained assessors, it was easier to classify an entire male from the flavor than from the odor in both chops and schnitzels. But, when it comes to determining a sorting threshold, this study has shown that the variation in consumer liking was relatively large and, it would be optimal if both skatole and androstenone were considered (Aaslyng et al., 2016).

Therewith, consumers` acceptance of pork with different levels of boar taint related off-flavors, within a meat alone and within a meal context, was investigated by Meier-Dinkel et al. in 2016. The main research questions were: whether consumer liking of boar tainted meat is linearly related to trained assessors` evaluation of back fat odor and whether consumer liking of boar tainted chops is moderated by meal presentation context? For the experiment, boar chops were chosen according to the olfactory ``fat score`` (based on the backfat detection of off-odors caused by androstenone and skatole, assessed by trained experts) from 24 boars, slaughtered at an average weight of 90 kg. In total, 37 consumers were participated in consumer acceptance

test (female 51% and male 49%, aged from 18 to 65 years, required to consume pork at least once per week). Chops were fried to a core temperature of 69 °C and after salt and pepper were added, were immediately served on the preheated plates (for meat presentation). For the meal presentation, chops were served together with string beans, potato croquettes and commercial gravy on the preheated plates as well. Each consumer evaluated 3 pure pork chops on one day and 3 meals on another day. All liking scores (for overall liking, appearance, odor, flavor, tenderness, juiciness and aftertaste liking) were recorded on 9-point scale (from 1 for extremely dislike to 9 for extremely like). Obtained results have shown that the main effect of meal context on overall liking scores was not significant. The overall liking did not differ: score for pure meat was 6.3 and score for meal meat was also 6.3 (Table 8). Further, no significant effects of context and context by fat score interactions were found for odor, flavor and after taste liking. Also, tenderness liking was not affected by any factor. The trained experts` fat score predicts consumer liking (high fat scores imply lower consumer liking), which was confirmation that the fat score was positively correlated with the androstenone and the skatole content. Therefore, sorting of carcasses by olfactory assessment appears reasonable (Meier-Dinkel et al., 2016).

	- /						
	Pure pre	esentation	Meal context				
	Mean	SD	Mean	SD			
Expected liking	6.0	1.7	6.7	1.4			
Overall liking	<mark>6.3</mark>	1.7	<mark>6.3</mark>	1.9			
Appearance liking	5.8	1.9	6.2	1.8			
Odor liking	6.5	1.5	6.4	1.6			
Flavor liking	6.5	1.5	6.5	1.7			
Texture liking	5.9	1.8	5.9	2.2			
Juiciness liking	6.7	1.4	6.3	1.8			
Aftertaste liking	6.2	1.5	6.4	1.5			

 Table 8. Mean scores and standard deviation of the various likings depending on the presentation context (pure meat and within meal).

Source: Meier-Dinkel et al. (2016).

The literature suggests that the negative impact of boar taint in consumer acceptability would be less in processed pork than in fresh meat because processing may decrease negative perceptions of androstenone and skatole. Thus, Lunde et al. (2008) indicated that strong food flavor additives like oregano extracts and liquid smoke affected the perception of boar taint. Aaslyng et al. (2015) stated that unpleasant odors or flavors can be masked in cooked meat products (bacon and pork belly roll), where smoking or spices are used in the preparation.

Martinez et al. (2016) evaluated also the use of smoking and/or spices for their ability to mask boar taint in frankfurters (cooked and smoked sausage of pork, with or without casing; hot dog), manufactured from entire pigs. In their study five frankfurter types were considered: control (prepared according to a standard recipe - lean meat 50%, pork fat 25%, ice/water 25%, potato starch 2,5% and other additives); smoked (standard recipe + smoking by sawdust); flavoring (JBT-200) + smoked (standard recipe + higher amount of commercial flavoring + smoking); spicy (standard recipe + spices and herbs mixture - white pepper, mustard seed, paprika, nutmeg mace, coriander seed, small cardamom fruit and sweet marjoram herb), and spicy + smoked type of frankfurter. Raw material for frankfurter manufacturing were obtained from thirteen carcasses with high androstenone and low skatole levels, purchased from Spanish slaughterhouses. From these, lean meat and fat were randomly divided into batches in such a way that all batches included lean and fat from all selected pigs. Androstenone analysis was performed using the gas chromatography-mass spectrometry technique (GC-MS) and skatole analysis by high-performance liquid chromatography (HPLC). The five types of frankfurter sausages were sensory analyzed by a trained sensory panel consisted of ten panelists with long experience in sensory evaluation. Each panelist evaluated 6 samples per type of frankfurter. The evaluated attributes were ranked according to the different odors (``androstenone odor``, ``frankfurter odor`` and ``other odors``) and flavors (``androstenone flavor``, `` androstenone aftertaste``, ``frankfurter flavor`` and ``other flavors``).

Obtained results have shown significant differences for androstenone and skatole content among frankfurter types, even though meat and fat from the same pigs were used in all batches. The androstenone value in ``flavoring + smoking`` frankfurter was the highest (1.157  $\mu$ g/g in fat tissue) while, the highest skatole level was found in ``control`` frankfurters (0.129  $\mu$ g/g in fat tissue, still below the sensory threshold). Average values for androstenone in frankfurters (1.018  $\mu$ g/g in fat tissue) were lower than those reported from the neck fat samples. This may be due to the manufacturing process because several authors indicated that androstenone and skatole are volatile compounds and therefore can be evaporated during processing or heating (Bonneau et al., 1992). Regarding odor attributes control frankfurters presented the highest "androstenone odour" with 3.1 score (0 = not perceivable; 10 = extremely perceivable) while, for masked frankfurters the "androstenone odour" score varied from 0.0 to 0.9. These results indicate the effectiveness of evaluated strategies to mask boar taint odour. Smoked and flavouring + smoked frankfurters had lower scores for "androstenone odour" compared with spicy frankfurters, which indicate that smoking was more effective than spices in masking androstenone odour. From all the masking strategies, only the combined use of spices and smoking could eliminate the "androstenone odour", "androstenone flavour" and "androstenone aftertaste" (score was 0.0). Nonetheless, the difference between spicy + smoked frankfurters and the other masked frankfurters was small (0.4-0.6 points). According to Kenji & Mitsuo (2002), spices have deodorizing effect due to isothiocyanate compounds (components of mustard for example) which stimulate mucus secretion in the nasal cavity and paralyse sensory function.

Even though all masking strategies from this study could decrease boar taint perception in frankfurters, only the use of spices together with a smoking process caused the androstenone to be imperceptible (Martinez et al., 2016).

Another group of authors, Borrisser-Pairo et al. (2017), have investigated the acceptability of meat from castrated and entire pigs cooked with different cooking methods, and the consumer sensitivity to androstenone. The pigs used in their study were crossbreeds Pietrain x (Landrace x Large White), slaughtered at approximately 100 kg live weight. Carcasses were selected according to high levels of boar taint by human nose methodology. Androstenone analysis was performed by the gas chromatography-mass spectrometry (GC-MC) and skatole analysis by high-performance liquid chromatography (HPLC). The selected carcasses had high levels of androstenone (from 2.02 to 2.30  $\mu$ g/g in fat tissue) and low/medium levels of skatole (from 0.05 to  $0.14 \,\mu\text{g/g}$  in fat tissue). Carcasses from castrated pigs were selected as control samples. To mask boar taint, two cooking methods were used: 1) vacuum cooking (known as sous-vide) and 2) fried/breaded with garlic and parsley. During vacuum cooking, loin chop samples were put in polyethylene bags, sealed with vacuum-sealing machine and placed in a water bath at 75 °C until the core temperature of 72 °C. For the second cooking method, 50 g of breadcrumbs, 8 g of white garlic, 8 g of parsley and salt were minced in a blender. Each sample was flipped 6 times in the mixture before placing in the fry pan (until the core temperature of 83 °C). The gender ratio of the 150 consumers was balanced and 2/3 were regular pork consumers. Consumers had to answer on: 1) do you smell anything, 2) can you score the intensity of smell

and 3) do you like this smell? After had answered they were classified on "low sensitive", "middle sensitive" and "high sensitive" consumers. They had to evaluate smell and flavor acceptability of each sample from castrated and entire pigs, for both cooking methods.

Results have shown that 30% of them were insensitive or low sensitive to androstenone, 32% middle sensitive and 37% high sensitive, with no differences between genders. However, when they were classified in 2 groups ("insensitive/middle/low" or "high sensitive"), women's sensitivity to androstenone tended to be higher than men. From those consumers that were sensitive to androstenone, 61,5% disliked the odor and the rest found it neutral or liked it. It was confirmed that the percentage of consumers who disliked androstenone increased with the sensitivity.

Regarding acceptability of flavor, results were similar for meat from castrated and entire males, except for the high sensitive consumers which preferred a little more meat from castrated pigs than from entire males, with no significant difference between cooking methods. Regarding smell acceptability, results showed that high sensitive consumers preferred castrated meat in the sous-vide although in the fried/breaded with garlic and parsley cooking method the result was opposite - they preferred the smell of entire male pig meat.

Regarding the results for overall liking of the meat, both cooking methods had similar liking scores for the castrated and entire male samples. Therefore, both methods were useful to mask the off-flavor from boar tainted meat. Anywise, the fried/breaded with garlic and parsley cooking method seemed to be more effective in improving the acceptability, even in high sensitive consumers (Borrisser-Pairo et al., 2017).

According to previous studies, different cooking methods can, more or less, mask boar taint and increase acceptability of tainted meat, even among high sensitive consumers. The use of herbs and aromatic plants can also be effective in masking boar taint odor. However, consumers studies are necessary to assess and confirm sensory acceptability of pork products obtained from boars.

### 2.11 Analytical tools to determine boar taint on the slaughter line



Photo 1. The olfactory assessment of the animals` backfat (Source: http://boars2018.com/2013/07/french-experiment-detecting-boar-taint/).

Because none of suggested alternatives for surgical castration guarantees the entire elimination of boar taint, developing methods to assess boar taint on the slaughter line to sort out tainted carcasses is needed (Haugen et al., 2012).

So far, the only method that has been taken for online use in slaughterhouses is a colorimetric method with possibility to measure the sum of both- skatole and indole. A Colorimetric method was developed in Denmark and introduced in Danish slaughterhouses from 1991. During this method, the fat samples are physically removed and transferred to the automated analyzer on the slaughter line. The method is based on solvent extraction of fat followed by addition of reagent and spectrophotometric fluorescence measurement of skatole and indole. The results are used for later sorting of carcasses down the production line with a capacity of 200 samples per hour (Mortensen & Sørensen, 1984). The benefits of this method are rapidity and simplicity but, it does not provide information about the androstenone and is not applicable for slaughterhouses of high capacity in Europe where, up to 1000 carcasses may need to be analyzed in an hour (Haugen et al., 2012).

Radioimmunological measurements are fast and sensitive but, they cannot be used for routine on-line analysis because they use radioisotopes (Andresen, 1975).

Chemical gas sensor arrays, so-called "electronic noses", have been suggested to have a potential for rapid objective sorting of boar tainted pig carcasses. The results may show significant correlation between the sensor readings and levels of skatole and androstenone or

sensory attributes related to boar odor and flavor. The technique requires training and calibration against sensory data or content of skatole and androstenone in the analyzed samples (Haugen, 2006).

Direct mass spectrometry (MS) has also been applied to measure boar taint. The direct MS technique is based on transfer of the gas phase of a sample into the ion-source of the mass spectrometer, followed by ionization and mass fragmentation of the molecules. This technique can be combined with different sampling methods and most used are headspace or pyrolysis (Ampuero & Bee, 2006).

Another technique that is under development for boar taint detection is gas phase Fourier Transform Infra-Red (FTIR) combined with photo acoustic spectroscopy (PAS) (Haugen et al., 2008).

Recent work has demonstrated that androstenone, skatole and indole can be separated and detected within 10 seconds by use of ultra-fast GC. Commercial ultra-fast gas chromatographic instruments have become available, enabling significantly shorter analysis times than conventional gas chromatographs and which may have a potential for the analysis of the boar substances. However, sampling is the critical stage because it is necessary to carry out a classical cleanup step to isolate the boar taint compounds prior to GC analysis. (Haugen et al., 2008).

Biosensing, using trained insects is another interesting method that has been investigated as a potential future rapid method for sorting boar carcasses. The use of trained insects (solitary parasitic wasps) is based on classical conditioning and the feeding behavioral response is applied as a positive recognition of the learned odor. These insects have shown that they are able to recognize skatole, indole and androstenone (Haugen et al., 2008).

Nevertheless, these methods do not yet seem to fulfil all the industrial method specifications regarding cost efficiency, simplicity and analysis time. To date, the sorting of boar carcasses is based on the olfactory assessment of the animals` backfat (Mathur et al., 2012) as no technical method copes with the time constraints of the slaughter process (Haugen, Brunius & Zamaratskaia, 2012). Generally, methodologies for assessing the boar-tainted carcasses consist of sampling the neck fat and evaluating the odor by human nose methodology (Trautmann et al., 2016) or performing chemical analyses of the content of androstenone and skatole in the

fat sample (Meinert et al., 2013). Traditionally the suitability of the whole carcass for consumption is assessed based on the neck sample even though recent research has shown a higher concentration of skatole and indole in the belly fat (Wesoly, Stefanski & Weiler, 2016). Carcass neck fat is usually heated with a hot iron at the slaughter line when trained assessors evaluate the odor of the fat to detect boar taint. Therefore, there is always a possibility for human distraction and wrong evaluation. In an optimal case, the sensory evaluation of the back fat allows the prediction of consumer liking or rejection.

More reliable sorting at the slaughter-line should provide doubtless directing selected tainted carcasses to the appropriate market. Consequently, there is still a huge need for reliable on-line technologies, capable of rapid, cost-effective and easy to use boar taint detection.

# 3 Discussion part

The aim of the present work was to examine how can pork quality of male pigs be improved in modern swine production.

It was shown, at the beginning of this work, how the value given to the factors of quality can differ from livestock producers to processors and final consumers. However, in the "pork quality" concept which describes the overall characteristics of pork meat, the hygienic and toxicological quality always come in front, regardless of the sensory quality represents a major determinant of consumer choices. To ensure food safety and quality of the product, the application of Herd Health Surveillance Programs (HHSP) at the level of livestock production, a system of Hazard Analysis and Critical Control Points (HACCP) at the level of manufacturing, meet the criteria of safety and health safety, and ISO standards, whose application is not mandatory but highly desirable, guarantee the quality of the product. Good Hygiene Practice (GHP), Good Manufacturing Practice (GPP) and Good Laboratory Practice (GLP) are also useful procedure and can provide the basis for a system such as ISO 9001 and HACCP (Acamovic & Kljajic, 2003).

Further, it was reported how the carcass characteristics and meat quality depend on a variety of different factors as well as how the meat from uncastrated male pigs may have expressed offensive odor and flavor, known as boar taint. As it was explained, boar taint becomes a major disadvantage of a modern swine production in the moment when consumers` awareness grew

about the side effects of the surgical castration and the importance of how animals are grown and produced.

Raising non-castrated male pigs is regarded as an alternative method to avoiding surgical intervention. Thus, one group of authors, D'Souza et al. (2011), have shown that there is no need for the implementation of surgical castration as well as for the implementation of immunocastration if entire male pigs are slaughtered below 90 kg live weight because the occurrence of boar taint is in that case negligible. The same authors have shown that regardless of slaughter weight, there is still a high proportion of pork samples exceeded the sensory threshold for androstenone and skatole, which was not in agreement with the previous results. Dunshea et al. (2001) have shown that a small percentage of pigs may escape the immunocastration either because of bad administration of the vaccine or a non-response to the vaccination. Contrarily, the same authors, Dunshea et al. (2001), claimed how the concentration of boar taint in 100% of Improvac treated pigs were negligible or undetectable. According to Moore et al. (2017), it was also confirmed that Improvac vaccine is effective in eliminating boar taint and reducing pork quality fail rates, but only by approximately 10% compared to pork from entire males.

As the standard protocol, it was recommended by vaccine producer that two vaccinations are needed for a full effect. However, in one study, conducted out by Pinna et al. (2015), pigs with three-vaccine doses were the closest with those surgically castrated. Therefore, it was concluded that three-dose vaccine treatment may be a reliable alternative to surgical castration even when dealing with boar taint from heavy pigs in their full adulthood.

One group of authors, (Zamaratskaia et al., 2008; Zamaratskaia et al., 2012), have stated that Improvac vaccine leads to testis regression and reduction in synthesis and accumulation of boar taint-causing androstenone and skatole. This is in accordance with Dunshea et al. (2001) who have found the highly significant reduction in testosterone concentration and the suppression in testicular growth, for at least 4 weeks of secondary vaccination (approximately 50 % decrease in testes and bulbo-urethral gland weights and length at slaughter). In their study, the testosterone reduction did not have a detrimental effect on growth performance of the Improvac treated pigs. Indeed, they had higher weight gain following the second dose and, over the 4 weeks, they grew more rapidly than control boars, they were leaner and had better feed conversion efficiency.

Aluwe et al. (2016) have shown contrary how testes weights did not differ significantly between pigs vaccinated 6 weeks before slaughter (IM-6) and pigs vaccinated 4 weeks before

slaughter (IM-4). This probably because extending the time for second vaccination by only two weeks was not enough period to observe significant difference in testes weights.

Font-i-Furnols et al. (2016) have come up with the observation that testes to body volume ratio (Figure 9) is a good predictor for androstenone and could be a good tool at slaughter plants to detect non-effective immunocastration (however, it was more related with androstenone than skatole because an important number of pigs with low skatole levels had volume ratio values higher than 0.55%).

Bonneau (1987) stated that androstenone and skatole concentration increases with the age and live weight which is not in-line with recent research conducted by Font-i-Furnols et al. (2016). They have shown that the relationship between body weight and androstenone level is not very strong because androstenone and its variability increase at the same time with the weight of entire male pigs. Also, Bonneaus` research was contrary to Doran et al. (2002) and Zamaratskaia et al. (2008a), where it was stated that increased levels of skatole in adult entire males are related with the reduction of skatole metabolism in the liver (due to inhibition of its metabolizing enzymes by testicular steroids) and not with live weight. Further, Moore et al. (2017) have shown that 37.5% of the light entire male pigs had skatole levels that exceeded the sensory threshold which was a confirmation that the boar taint is still an issue at low carcass weights of entire males.

Generally, it was common that Improvac vaccine as an alternative method to surgical castration is effective in eliminating boar taint but not entirely.

About consumers' acceptance of boar tainted meat, according to Panella-Riera et al. (2016), it was shown that the acceptability of boar tainted pork can depend on the habit of eating meat from castrated or entire male pigs, procedures for preparing the meat and the ability of consumers to perceive androstenone and/or skatole (these findings are in line with results obtained by Borrisser-Pairo et al. (2017), Channon et al. (2016), Lunde et al. (2008), Aaslyng et al. (2015) and Martinez et al. (2016)). For example, in Spain, United Kingdom and Italy over than 90% of consumers ate fresh pork more than 2 times per week while in France just around 35%. The most consumed product was dry cured ham in Spain and Italy, cooked ham in France and sliced bacon in United Kingdom. There was a group of consumers which refuse tainted meat (16.2–20.0%), but there was also a group that prefer boar tainted meat (12.4–21.7%). Therefore, it is important to obtain knowledge about consumer response towards boar tainted meat.

Channon et al. (2016) have shown that aroma, juiciness and flavour of the pork loin steak was influenced by final endpoint temperature, with lower scores for all when cooked to 75 °C compared with 70 °C. Juiciness was better from females aged for 7 days than for 2 days, while from entire males aged for 7 days juiciness was reduced by 5.3 units compared to 2-day aged pork. In their study, pork from entire male pigs had lower scores for juiciness, flavour, overall liking and quality grade than castrates and females, which was in accordance with results obtained from Aaslyng et al. (2016).

Meinert et al. (2017) have confirmed a strong link between skatole in the neck fat and in the meat cuts. The intensity of boar taint in the cooked meat was generally highest in the pigs with the highest concentration of either skatole or androstenone in the neck fat with some exceptions. The pigs numerated with numbers 9., 10. and 12. had a very low amount of skatole and androstenone in the neck fat and an intense boar taint flavour in the cooked meat (Table 7). It was concluded that much lower concentrations of skatole and androstenone can be detected in the cooked meat compared to the established thresholds for skatole and androstenone in fat. This corresponds to results from Martinez et al. (2016) where average values for androstenone in frankfurters were also lower than those reported from the neck fat samples. Meier-Dinkel et al. (2016) have reported how the trained experts` fat score predicts consumer liking (high fat scores imply lower consumer liking), which was confirmation that the fat score was positively correlated with the androstenone and the skatole content (in agreement with Aaslyng et al. (2016) & Meinert et al. (2017)). Therefore, sorting of carcasses by olfactory assessment appears reasonable, but not entirely reliable.

Further, the literature suggests that the negative impact of boar taint in consumer acceptability would be less in processed pork than in fresh meat because processing may decrease negative perceptions of androstenone and skatole (in accordance with Lunde et al. (2008), Aaslyng et al. (2015), Martinez et al. (2016) and Borrisser-Pairo et al. (2017)). Also, Bonneau et al. (1992) indicated that androstenone and skatole are volatile compounds and therefore can be evaporated during processing (heating). Thus, Lunde et al. (2008) found that strong food flavour additives like oregano extracts and liquid smoke affected the perception of boar taint. Aaslyng et al. (2015) stated that unpleasant odours or flavours can be masked in cooked meat products (bacon and pork belly roll), where smoking or spices are used in the preparation. Martinez et al. (2016) evaluated also the use of smoking and/or spices for their ability to mask boar taint in frankfurters, manufactured from entire pigs. Even though all masking strategies from their study could decrease boar taint perception in frankfurters, only the use of spices

together with a smoking process caused the androstenone to be imperceptible. While, Borrisser-Pairo et al. (2017) have shown that the fried/breaded with garlic and parsley cooking method was more effective in improving the acceptability even in high sensitive consumers.

Very significant is to note that the methodologies used in different studies were diverse and therefore it was difficult to compare the results among them. The following parts are only some examples which differ: type of meat samples (meat from gilts, surgically and immunocastrated pigs and entire male pigs), the sample location of the carcasses, level of boar taint in the assessed meat samples and different procedures for assessing, the timing of the second vaccination in immunocastrated pigs, two or three doses of immunization, the different genetic crossbreeds, the type of attributes and scales used during the consumer test, cooking procedures, type of pork product for consumer evaluation (bacon, dry cured ham, chops, loin slices, minced meat and other).

Further, different laboratories apply different procedures. Some of them use solvent extraction and others use melting/freezing of the fat tissue in combination with solvent extraction to isolate the boar substances from the fat phase. Most laboratories analyse backfat samples, however the sample location of backfat is not well defined, some take the backfat at the 6th rib (T6) and some analyse belly fat (Haugen et al., 2012). Therefore, it is interesting to underline that it has been shown how the levels of skatole and androstenone may vary depending on where on the carcass the sample has been taken (Andresen, 1975).

To date, there is not yet any accepted official reference method available for the analysis of skatole and androstenone. Moreover, there is no harmonized analytical protocols for analysing skatole and androstenone in boar fat tissue, which makes it difficult to compare results from different laboratories (Haugen et al., 2012) and from different studies as well.

## 4 Conclusion

In preliminary research, it was shown that there are different types of quality and different factors can influence the formation and assessment of quality. Significant and promising results regarding boar taint, as a major disadvantage of pork quality nowadays, have been obtained from several studies and summarized in this paper.

Achieving the goals of the Declaration of 2018 is still far from being met.

Regardless of promising results from all described analytical methods (from "electronic noses", ultra-fast GC and other, up to the insects` evaluation), the sorting of boar tainted carcasses is still based on the olfactory assessment of the animals` backfat. Therefore, there is a huge need for establishing a reliable on-line technology which should provide directing selected tainted carcasses to the appropriate market or further processing, capable of rapid, cost-effective and easy to use boar taint detection.

Within each European country, the percentage of carcasses with high boar taint levels and the results from consumers acceptance studies could be useful for the European pig sector, to establish an official reference method for skatole and androstenone analysis.

Further research is needed to establish an efficient, harmonized system that would deal with the boar taint issue. This because none of the suggested alternatives for surgical castration guarantees the entire elimination of boar taint. Currently, a harmonized system for elimination of boar taint might be a combination of immunocastration, "electronic noses" technology and masking strategy for meat processing.

It was encouraging to see that there is a market for tainted meat lovers, which leads to the conclusion that such market could be bigger if consumers were more informed about boar tainted products and their benefits. To remind, the way how animals are raised and produced, higher protein content and consequently a higher nutritional value, a reduced proportion of adipose tissue and an increased content of unsaturated fatty acids are some of the most important benefits from entire males` carcasses.

### 5 References

- Aaslyng, M. D., Broge, E. H. D. L., Brockhoff, P. B., & Christensen, R. H. B. (2015). The effect of skatole and androstenone on consumer response towards streaky bacon and pork belly roll. Meat Science 110, 52–61.
- Aaslyng, M. D., Broge, E. H. D. L., Brockhoff, P. B., Christensen, R. H. B., (2016). The effect of skatole and androstenone on consumer response towards fresh pork from m. longissimus thoracis et lumborum and m. semimembranosus. Meat Science 116, 174–185.
- Acamovic, N., Kljajic, R., 2003. Development of the system of hazard analysis and critical control points (HACCP) in food production. The monograph. Institute of Veterinary Medicine, Novi Sad.
- Adzitey F., 2011. Effect of pre-slaughter animal handling on carcass and meat quality
   Mini Review, International Food Research Journal, 18, 484-490.
- Aldal I., Andresen O., Egeli AK., Haugen JE., Grødum A., Fjetland O., and Eikaas JLH., (2005): Levels of androstenone and skatole and the occurrence of boar taint in fat from young boars, Livestock Production Science 95, 121–129.
- Aluwe, M., Degezelle, I., Depuydt, L., Fremaut, D., Van den Broeke, A., and Millet, S., (2016). Immunocastrated male pigs: effect of 4 v. 6 weeks' time post second injection on performance, carcass quality and meat quality. Animal Science 10:9, pp 1466-1473.
- Ampuero, S., & Bee, G., 2006. The potential to detect boar tainted carcasses by using an electronic nose based on mass spectrometry. Acta Veterinaria Scandinavica, 48 (Suppl1): S15.
- Andersen H. J., Oksbjerg N., Therkildsen M., 2005. Potential control tools in the production of fresh pork, beef and lamb demanded by the European society, Livestock Production Science, 94, 105–124.
- Andresen Ø., (1975): 5α-Androstenone in peripheral plasma of pigs, diurnal variation in boars, effects of intravenous hCG administration and castration, Acta Endocrinologica 78, 385-391.

- Babol J., Zamaratskaia G., Juneja R. K., Lundström K., (2004): The effect of age on distribution of skatole and indole levels in entire male pigs in four breeds: Yokshire, Landrace, Hampshire and Duroc, Meat Science 67: 351-358.
- 11. Babol, J., Squires, E. J. and Lundström, K. (1998a) Hepatic metabolism of skatole in pigs by cytochrome P4502E1. Journal of Animal Science 76, 822-828.
- Babol, J., Squires, E.J. (1995): Quality of meat from entire male pigs. Food Research International, 28: 201-212.
- Barton Gade, P. A. (1987). Meat and fat quality in boars, castrated and gilts. Livestock Production Science, 16, 187–196.
- Bonneau M., (1987): Effects of age and live weight on fat 5 alpha-androstenone levels in young boars fed two planes of nutrition, Reproduction Nutrition Development 27(2A):413-22.
- Bonneau, M. (1998) Use of entire males for pig meat in the European Union. Meat Science 49: S257-S272
- 16. Bonneau, M., Le Denmat, M., Vaudelet, J. C., Veloso Nunes, J. R., Mortensen, A. B. and Mortensen, H. P. (1992) Contributions of fat androstenone and skatole to boar taint:I. Sensory attributes of fat and pork meat. Livestock Production Science 32: 63-80.
- Bonneau, M., Squires, J. (2004): boar taint: Causes and measurement. In Encyclopedia of Meat Sciences. Eds W.k. Jensen, C. Devine & M. Dikemann, Elsevier, Oxford, ISbN 0-12-464970-X, pp.91-96 (I-S).
- Borrisser-Pairo, F., Panella-Riera, N., Gil, M., Kallas, Z., Linares, M. B., Egea, M., Garrido, M. D., Oliver M. A., (2017). Consumers` sensitivity to androstenone and the evaluation of different cooking methods to mask boar taint. Meat Science 123, 198-204.
- Borrisser-Pairó, F., Panella-Riera, N., Zammerini, D., Olivares, A., Garrido, M. D., Martínez, B., Gil, M., García-Regueiro, J. A., Oliver, M. A., (2016). Prevalence of boar taint in commercial pigs from Spanish farms. Meat Science, 111, 177-182.
- 20. Briyne, N., Berg, C., Blaha, T., and Temple, D., 2016. Pig castration: will the EU manage to ban pig castration by 2018. Porcine Health Management. Available at: <a href="https://porcinehealthmanagement.biomedcentral.com/articles/10.1186/s40813-016-0046-x">https://porcinehealthmanagement.biomedcentral.com/articles/10.1186/s40813-016-0046-x</a>

- 21. Channon, H. A., D'Souza, D. N., & Dunshea, F. R., (2016). Developing a cuts-based system to improve consumer acceptability of pork: Impact of gender, ageing period, endpoint temperature and cooking method. Meat Science 121, 216-227.
- 22. Claus R., Losel D., Lacorn M., Mentschel J., & Schenkel H., (2003): Effects of butyrate on apoptosis in the pig colon and its consequences for skatole formation and tissue accumulation, Journal of Animal Science 81, 239-248.
- 23. Claus R., Schopper D., Wagner HG., (1983): Seasonal effect on seminal plasma of boars, Journal of Steroid Biochemistry 19:725–729.
- Claus R., Weiler U., and Herzog A., (1994): Physiological aspects of androstenone and skatole formation in the boar: A review with experimental data, Meat Science 38: 289– 305.
- 25. Dalmau A., Velarde A., Gispert M., 2009. Standardization of the measure "meat quality "to assess the welfare of pigs at slaughter, in Forkman B. i Keeling L., Assessment of Animal Welfare Measures for Sows, Piglets and Fattening Pigs, Welfare Quality Reports No. 10.
- 26. Danielsen V., Hansen L.L., Møller F., Bejerholm C. Nielsen, S., 2000. Production results and sensory meat quality of pigs fed different amounts of concentrate and ad lib clover grass or clover grass silage, In Ecological Animal Husbandry in the Nordic Countries, Proceedings from NJF-seminar No 303, 16-17 September, DARCOF report, 2, 79-86.
- 27. Dong, X., Bai, Y., Xin, Y., Xu, Q., Chen, G. and Fang, M. (2012) Investigation on the transcription factors of porcine 3-hydroxysteroid dehydrogenase and 17hydroxysteroid dehydrogenase genes. Gene 499, 186-190.
- Doran, E., Whittington, F. W., Wood, J. D., & McGivan, J. D. (2002). Cytochrome P450IIE1 (CYP2E1) is induced by skatole and this induction is blocked by androstenone in isolated pig hepatocytes. Chemico-Biological Interactions, 140, 81– 92.
- 29. D'Souza D. N., Warner R. D., Dunshea F. R. i Leury B. J. (1998b). Effect of on farm and pre-slaughter handling of pigs on meat quality. Australian Journal of Agricultural Research. 49: 1021-1025.
- 30. D'Souza, D. N., Dunshea, F. R., Hewitt, R. J. E., Luxford, B. G., Meaney, D., Schwenke, F., ... van Barneveld, R. J. (2011). High boar taint risk in entire male

carcasses. In R. J. Van Barneveld (Ed.), Manipulating Pig Production XIII (pp. 259). Werribee, Australia: Australasian Pig Science Association.

- 31. Du, M., Yin, J., & Zhu, M. J. (2010). Cellular signaling pathways regulating the initial stage of adipogenesis and marbling of skeletal muscle. Meat Science, 86, 103–109.
- 32. Dunshea F. R., Allison J. R. D., Bertram M., Boler D. D., Brossard L., Campbell R., Crane J. P., Hennessy D. P., Huber L., de Lange C., Ferguson N., Matzat P., McKeith F., Moraes P. J. U., Mullan B. P., Noblet J., Quiniou N., and Tokach M. (2013). The effect of immunization against GnRF on nutrient requirements of male pigs: a review. Animal 7, 1769-1778.
- 33. Dunshea, F. R., Colantoni, C., Howard, K., McCauley, I., Jackson, P., Long, K. A., Lopaticki, S., Naugent, E. A., Simons, J. A., Walker, J., and Hennessy, D. P., (2001). Vaccination of boars with a GnRH vaccine (Improvac) eliminates boar taint and increases growth performance. Journal of Animal Science, 79:2524-2535.
- 34. Earley B., and Crowe M. A., (2002): Effects of ketoprofen alone or in combination with local anesthesia during the castration of bull calves on plasma cortisol, immunological, and inflammatory responses, Journal of Animal Science 80: 1044-1052.
- 35. Ellis, M., F. K. McKeith and K. D. Miller. 1999. The effect of genetic and nutritional factors of pork quality- Review. Asian-Aust. J. Anim. Sci. 12:261-270.
- 36. Elsbernd, A. J., Patience, J. F., & Prusa, K. J., (2016). A comparison of the quality of fresh and frozen pork from immunologically castrated males versus gilts, physical castrates, and entire males. Meat Science 111, 110-115.
- 37. Enser, M. (1999). Nutritional effects on meat flavor and stability. In R. I. Richardson, & G. C. Mead (Eds.), Poultry Meat Science. Poultry Science. Symposium Series (Vol. 25) (pp. 197–215). Wallingford, UK: CABI Publishing.
- 38. Enser, M., Richardson, R. I., Wood, J. D., Gill, B. P., & Sheard, P. R. (2000). Feeding linseed to increase the n-3 PUFA of pork: fatty acid composition of muscle, adipose tissue, liver and sausages. Meat Science, 55, 201–212.
- Faustman, C., Sun, Q., Mancini, R., & Suman, S. P. (2010). Myoglobin and lipid oxidation interactions: Mechanistic bases and control. Meat Science, 86, 86–94.
- 40. Fitzgerald M., (1994): Neurobiology of Fetal and Neonatal Pain. In: Textbook of Pain. Eds. Font i Furnols M., Gispert M., Diestre A., & Oliver M. A., (2003). Acceptability of boar meat by consumers depending on their age, gender, culinary

habits, and sensitivity and appreciation of androstenone odor. Meat Science, 64(4), 433–440.

- 41. Font i Furnols, M., Gispert, M., Diestre, A., & Oliver, M. A. (2003). Acceptability of boar meat by consumers depending on their age, gender, culinary habits, and sensitivity and appreciation of androstenone odour. Meat Science, 64(4), 433–440.
- 42. Font-i-Furnols, M., Gispert, M., Soler, J., Diaz, M., Garcia-Regueiro, J. A., Diaz, I., & Pearce, M. C. (2012). Effect of vaccination against gonadotropin-releasing factor on growth performance, carcass, meat and fat quality of male Duroc pigs for dry-cured ham production. Meat Science, 91, 148–154.
- 43. Font-i-Furnols, M., Carabús, A., Muñoz, I., Čandek-Potokar, M., Gispert, M., (2016). Evolution of testes characteristics in entire and immunocastrated male pigs from 30 to 120 kg live weight as assessed by computed tomography with perspective on boar taint. Meat Science 116, 8-15.
- 44. Food and Agriculture Organizations of the United Nations (FAO). Animal Production and Health. 2017. Available at: http://www.fao.org/ag/againfo/themes/en/pigs/home.html
- 45. Friis C., (1993): Distribution, metabolic fate and elimination of skatole in the pig. In Bonneau M (ed), Measurement and Prevention of Boar Taint in Entire Male Pigs. Paris, France; Institute National de la Recherche Agronornique (INRA), p113–115.
- 46. Gardner G. A., Cooper T. J. H., (1979). An evaluation of feeding liquid sugar to pigs' lairaged overnight before slaughter, In Proceedings of the 25th European Meeting of Meat Research Workers, Budapest, Hungary, 5-8.
- 47. Gibson G. R., & Roberfroid M. B., (1995): Dietary modulation of the human colonic microbiota—introducing the concept of prebiotics, Journal of Nutrition 125, 1401–1412.
- 48. Grandin T., (1994). Methods to reduce PSE and Blood splash. Proc. Allen D. Leman Swine Confr. University of MN. 21:206-209
- 49. Hansen L. L, Larsen A. E, Jensen B. B, Hansen-Møller J., Barton-Gade P., (1993). Influence of stocking rate and temperature on feces deposition in the pen and its consequences on skatole concentration (boar taint) in subcutaneous fat. In Bonneau M (ed), Measurement and Prevention of Boar Taint in Entire Male Pigs. Paris, France; Institut National de la Recherche Agronornique (INRA), p151–157.

- 50. Hansen L. L., Mejer H., Thamsborg S. M., Byrne D. V., Roepstorff A., Karlsson A. H., Hansen-Møller J., Jensen M. T., & Tuomola M., (2006): Influence of chicory roots (Cichorium intybus L) on boar taint in entire male and female pigs, Animal Science 82, 359-368.
- 51. Haugen, J. E. (2006). The use of chemical sensor array technology, the electronic nose, for detection of boar taint. Acta Veterinaria Scandinavica, 48(Suppl 1), S15.
- 52. Haugen, J. E., Brunius, C., & Zamaratskaia, G. (2012). Review of analytical methods to measure boar taint compounds in porcine adipose tissue: The need for harmonized methods. Meat Science, 90 (1), 9-19.
- 53. Haugen, J. E., Lundby, F., Wäckers F. L., Olson, D., Kauppinen, I, Ferber, A., de Wiel, D., Briens, M., & Tuyttens, F., (2008). Rapid detection methods for boar taint; Fast GC, FTIR-PAS and Biosensing. EAAP meeting, 26-27. March, Girona.
- 54. Hay M., Vulin A., Genin S., Sales P., and Prunier A., (2003): Assessment of pain induced by castration in piglets: behavioral and physiological responses over the subsequent 5 days, Applied Animal Behavior Science 82:201-218.
- 55. Henckel P., Karlsson A., Oksbjerg N., Petersen J.S., 2000. Control of post mortem pH decrease in pig muscles: experimental design and testing of animal models, Meat Science, 55, 131–138.
- 56. Hofmann K. (1994): What is quality? Definition, measurement and evaluation of meat quality. Meat Focus International, 3(2).
- 57. Hwang, Y. H., Kim, G. D., Jeong, J. Y., Hur, S. J., & Joo, S. T. (2010). The relationship between muscle fiber characteristics and meat quality traits of highly marbled Hanwoo (Korean native cattle) steers. Meat Science, 86, 456–461.
- 58. Jensen B. B. & Jensen M. T., (1998): Microbial production of skatole in the digestive tract of entire male pigs. In: Jensen, W.K. (Ed.), Skatole and boar taint. Roskilde, Denmark: Danish Meat Research Institute, 41-75.
- 59. Jensen M. T., Cox R. P., Jensen B. B., (1995): Microbial production of skatole in the hindgut of pigs given different diets and its relation to skatole deposition in back fat, Animal Science 6:293-304.
- 60. Katsamuta M., (2011). Promotion of intramuscular fat accumulation in porcine muscle by nutritional regulation. 82 (1): 17-25.
- 61. Kenji, H., & Mitsuo, T., (2002). Ciencia y tecnologia de las especias. Zaragoza: Acribia.

- 62. Kjeldsen N., (1993): Practical experience with production and slaughter of entire male pigs. In Bonneau M (ed), Measurement and Prevention of Boar Taint in Entire Male Pigs Paris, France; Institut National de la Recherche Agronornique (INRA), p137–144.
- Kocwin-Podsiadła M., 2002. Methods for improvements of pork quality, in: Proceedings V Intern. Tradeshows. Pork and Poultry Farm. 17–19 May, Poznań, Poland, 93–95 (in Polish).
- 64. Lealiifano A. K., Pluske J. R., Nicholls R. R., Dunshea F. R. and Mullan B. P., (2009): Altering the timing of an immunocastration vaccine to optimize pig performance. In "Manipulating Pig Production XII", ed R.J. van Barneveld. (Australasian Pig Science Association: Werribee), 184.
- 65. Lebret B. (2008). Effects of feeding and rearing systems on growth, carcass composition and meat quality in pigs. Animal. 2 (10): 1548–1558.
- 66. Leskanich, C. O., Matthews, K. R., Warkup, C. C., Noble, R. C., & Hazzledine, M., (1997). The effect of dietary oil containing n-3 fatty acids on the fatty acids, physiochemical and organoleptic characteristics of pig meat and fat. Journal of Animal Science, 75, 673–683.
- 67. Lunde, K., Egelandsdal, B., Choinski, J., Mielnik, M., Flåtten, A., & Kubberød, E., (2008). Marinating as a technology to shift sensory thresholds in ready-to-eat entire male pork meat. Meat Science 80, 1264-1272.
- Lundström K., Malmfors B., Malmfors G., Stern S., Petersson H., Mortensen A. B., & Srensen S. E., (1988): Skatole, androstenone and taint in boars fed two different diets, Livestock Production Science 18, 55-67.
- 69. Lundström, K., Matthews, K. R., & Haugen, J. E., (2009). Pig meat quality from entire males. Animal, 3 (11), 1497–1507.
- Martinez, B., Rubio, B., Viera, C., Linares, M. B., Egea, M., Panella-Riera, N., Garrido, M. D., (2016). Evaluation of different strategies to mask boar taint in cooked sausage. Meat Science 116, 26-33.
- 71. Mathur, P. K., ten Napel, J., Bloemhof, S., Heres, L., Knol, E. F., & Mulder, H. A. (2012). A human nose scoring system for boar taint and its relationship with androstenone and skatole. Meat Science, 91 (4), 414-422.
- 72. Meier-Dinkel, L., Strack, M., Höinghaus, K., Mörlein, D., (2016). Consumers dislike boar taint related off-flavors in pork chops regardless of a meal context. Meat Science 122, 119-124.

- Meinert, L., Claudi-Magnussen, C., & Støier, S. (2013). Limits for the detection of boar taint. Fleischwirtschaft International, 2,101–102.
- 74. Meinert, L., Lund, B., Bejerholm, C., Aaslyng, M. D., (2017). Distribution of skatole and androstenone in the pig carcass correlated to sensory characteristics. Meat Science 127, 51-56.
- 75. Meisinger D., 2002. A system for assuring pork quality, National Pork Board, USA.
- Migdal W., Ţivković B., Migdal L., (2009): Piglet castration, Biotechnology in Animal Husbandry 25 (5-6), p 839-847.
- 77. Millet S, Gielkens K, De Brabander D and Janssens GPJ (2011). Considerations on the performance of immunocastrated male pigs. Animal 5, 1119-1123.
- Miyahara M., Matsuda S., Komaki H., Sakari H., and Tsukise A., (2004): Effects of sexual distinction on growth rate and meat production in three-way cross pigs, Japanese Journal of Swine Science 41, 228–236.
- 79. Moore, K. L., Mullan, B. P., Dunshea, F. R., (2017). Boar taint, meat quality and fail rate in entire male pigs and male pigs immunized against gonadotrophin releasing factor as related to body weight and feeding regime. Meat Science, vol. 125, pp. 95-101.
- Morel, P. C. H., McIntosh, J. C., and Janz, J. A. M., 2006. Alteration of the Fatty Acid Profile of Pork by Dietary Manipulation- Review. Asian-Aust. J. Anim. Sci. Vol 19, No. 3: 431-437.
- 81. Morgan, M. T. and J. C. Forrest. 1997. Color vision systems and tetrapolar electrodes for assessing pork quality. pp 93-109 in Proc. of Pork Quality Summit. National Pork Producers Council. Des Moines, IA.
- Morgan, M. T., Klaus, J. R., and Berg, E. P., 1997. Objective color standards for pork. pp 138-141 in Proc. 50th Ann. Recipr. Meat Confr.
- 83. Mortensen A. B., Sørensen S. E., (1984): Relationship between boar taint and skatole determined with a new analysis method. In Proceedings of the 30th Eurpoean meeting of meat research workers Bristol, 394-396.
- 84. Myer, R. O., Johnson, D. D., Knauft, D. A., Gorbet, D. W., Brendemuhl, J. H., & Walker, W. R. (1992). Effect of feeding high-oleic acid peanuts to growing-finishing swine on resulting carcass fatty acid profile and on carcass and meat quality characteristics. Journal of Animal Science, 70, 3734–3741.

- Nanni Costa, L., Tassone, F., Comellini, M., Ielo, M. C., Lo Fiego, D. P., & Russo, V. (2008). Effect of the slaughterhouse on the incidence of defects in raw pig ham destined to the dry-curing process. Veterinary Research Communications, 32 (1), S351–S353.
- 86. National Pork Producers Council. 2000. Pork Composition and Quality Assessment Procedures. NPPC. Des Moines, IA.
- Ngapo, T. M., Martin, J. F., & Dransfield, E. (2007a). International preferences for pork appearance: I. Consumer choices. Food Quality and Preference, 18 (1), 26–36.
- Ngapo, T. M., Martin, J. F., & Dransfield, E. (2007b). International preferences for pork appearance: II. Factors influencing consumer choice. Food Quality and Preference, 18 (1), 139–151.
- 89. Nicolau-Solano, S. I., Whittington, F. M., Wood, J. D. and Doran, O., (2007) Relationship between carcass weight, adipose tissue androstenone level and expression of the hepatic 3-hydroxysteroid dehydrogenase in entire commercial pigs. Animal 1, 1053-1059.
- 90. OECD-FAO Agricultural Outlook, Meat consumption. Edition 2016. Available at: <a href="https://data.oecd.org/agroutput/meat-consumption.htm">https://data.oecd.org/agroutput/meat-consumption.htm</a>
- 91. Olsson, V., and Østergaard Brandt, S., (2015). Chapter 5: The quality of local and regional food. Interdisciplinary perspectives on local and regional food in the South Baltic Region, Kristianstad University Press, 74.
- 92. Øverland M., Berg J., Matre T., (1995): The effect of feed and feeding regime on skatole and androstenone levels and on sensory attributes of entire male and female pigs. In from the Proceedings of the EAAP Working Group Production and Utilization of Meat from Entire Male Pigs Milton Keynes, UK. 27–29 September.
- 93. Panella-Riera, N., Blanch, M., Kallas, Z., Chevillon, P., Garavaldi, A., Gil, M., Gil, J. M., Font-i-Furnols, M., Oliver, M. A., (2016). Consumers' segmentation based on the acceptability of meat from entire male pigs with different boar taint levels in four European countries: France, Italy, Spain and United Kingdom. Meat Science 114, 137-145.
- 94. Park, G. B., Moon, S. S., Ko, Y. D., Ha, J. K., Lee, J. G., Chang, H. H., & Joo, S. T. (2002). Influence of slaughter weight and sex on yield and quality grades of Hanwoo (Korean native cattle) carcasses. Journal of Animal Science, 80, 129-136.
- 95. Parkinson, A. (2001) Biotransformation of xenobiotics. In: Toxicology: The basic science of poisons, sixth edition, edited by Klaassen C. C., New York, McGraw-Hill.

- 96. Pinna, A., Schivazappa, C., Virgili, R., Parolari, G. (2015). Effect of vaccination against gonadotropin-releasing hormone (GnRH) in heavy male pigs for Italian typical drycured ham production. Meat Science, vol.110, pp. 153-159.
- 97. Pisula A. and Florowski T. (2009). Critical Points in the Development of Pork Quality
   A Review. Polish Journal of Food and Nutrition Sciences. 15/56 (3): 249–256.
- 98. Prunier A., Bonneau M., von Borell E. H., Cinotti S., Gunn M., Fredriksen B., Giersing M., Morton D. B., Tuyttens F. A. M., and Velarde A., (2006): A review of the welfare consequences of surgical castration in piglets and the evaluation of non-surgical methods, Animal Welfare 15(3): 277-289.
- 99. Rideout T. C., Fan M. Z., Cant J. P., Wagner-Riddle C., & Stonehouse P., (2004): Excretion of major odor-causing and acidifying compounds in response to dietary supplementation of chicory inulin in growing pigs, Journal of Animal Science 82, 678-684.
- 100.Rosenvold K., Andersen H. J. (2003a). Factors of significance for pork quality-a review, Meat Science. 64: 219–237.
- 101.Savell J. W., Mueller S. L., Baird, B. E., 2005. The chilling of carcasses, Meat Science, 70, 3, 449–459.
- 102.Shackelford, S. D., Reagan, J. O., Haydon, K. D., & Miller, M. F. (1990). Effects of feeding elevated levels of monounsaturated fats to growing-finishing swine on acceptability of boneless hams. Journal of Food Science, 55, 1485–1517.
- 103.Sheard, P. R., Enser, M., Wood, J. D., Nute, G. R., Gill, B. P., & Richardson, R. I. (2000). Shelf life and quality of pork and pork products with a raised n-3 PUFA. Meat Science, 55, 213–221.
- 104.Sinclair, P. A., Gilmore, W. J., Lin, Z., Lou, Y., and Squires, E. J. (2006) Molecular cloning and regulation of porcine SULT2A1: relationship between SULT2A1 expression and sulfoconjugation of androstenone. Journal of Molecular Endocrinology 36, 301-311.
- 105.Sinclair, P. A., Hancock, S., Gilmore, W. J. and Squires, E. J. (2005) Metabolism of the 16-androstene steroids in primary cultured porcine hepatocytes. The Journal of Steroid Biochemistry and Molecular Biology 96, 79-87.
- 106.Smith, S. B., Kawachi, H., Choi, C. B., Choi, C. W., Wu, G., & Sawyer, J. E. (2009). Cellular regulation of bovine intramuscular adipose tissue development and composition. Journal of Animal Science, 87, E72–E82.

- 107.Statistiks sentralbyrå Norway. Meat production. 2015. Available at: <u>https://www.ssb.no/en/jord-skog-jakt-og-fiskeri/statistikker/slakt/aar/2016-04-05</u>, accessed on 16 March 2017.
- 108.Sundrum, A., Bütfering, L., Henning, M., Hoppenbrock, K. H., 2000. Effects of onfarm diets for organic pig production on performance and carcass quality. J. Anim. Sci. 78, 1199–1205.
- 109.Taylor A. A., Weary D. M., Lessard M., & Braithwaite L., (2001): Behavioral responses of piglets to castration: the effect of piglet age, Applied Animal Behavior Science 73, 35-43.
- 110.Terlouw E. M. C., Astruc T. and Monin G. (2004). Effect of genetic background, rearing and slaughter conditions on behaviour, physiology and meat quality of pigs. In Proceedings of the EU Workshop on Sustainable Pork Production: Welfare, Quality, Nutrition and Consumer Attitudes (ed. AH Karlsson and HJ Andersen), pp 113–125. Copenhagen, Denmark.
- 111.Trautmann, J., Meier-Dinkel, L., Gertheiss, J., & Mörlein, D. (2016). Boar taint detection: A comparison of three sensory protocols. Meat Science, 111,92–100.
- 112.USDA-FAS (United States Department of Agriculture). 2016. Available at: <u>https://apps.fas.usda.gov/psdonline/circulars/livestock\_poultry.pdf</u>, accessed on 15 March 2017.
- 113.Walker N., (1978): Boars for meat production the effect of single-sex or mixed-sex groups on growth performance and carcass characteristics, Agricultural Research Institute of Northern Ireland, 26:7–10.
- 114.Walstra P., Claudi-Magnussen C., Chevillon P., von Seth G., Diestre A., Matthews K. R., Homer D. B., Bonneau M., (1999): An international study on the importance of androstenone and skatole for boar taint: levels of androstenone and skatole by country and season, Livestock Production Science 62: 15-28.
- 115.Warriss, P. D., 2010. An Introductory text. Meat Science, 2nd edition, ch 6, ch 1, ch 3 available at:

https://books.google.no/books?hl=no&lr=&id=NbzTWCl8LkC&oi=fnd&pg=PR5&d q=meat+quality+chapter+6+meat+science+P+D+Warriss&ots=la\_IHBZiIB&sig=Z2s q0qtibmFavoy2Lg8r\_2Z6bVQ&redir\_esc=y#v=onepage&q&f=true

- 116.Weiler, U., Gotz, M., Schmidt, A., Otto, M., and Muller, S., (2013). Influence of sex and immunocastration on feed intake behavior, skatole and indole concentrations in adipose tissue of pigs. Animal 7, 300-308.
- 117.Wesoly, R., Stefanski, V., & Weiler, U. (2016). Influence of sampling procedure, sampling location and skin contamination on skatole and indole concentrations in adipose tissue of pigs. Meat Science, 111, 85–91.
- 118.Wood J. D. (1990). Consequences for meat quality of reducing carcass fatness. In J. D.Wood and M. Enser (Eds.), Reducing fat in meat animals (pp. 344–397). London: Elsevier Applied Science.
- 119.Wood J. D., Buxton P. J., Whittington F. M and Enser M., (1986): The chemical composition of fat tissues in the pig: effects of castration and feeding treatment, Livestock Production Science 15, 73–82.
- 120.Wood, J. D., Richardson, R. I., Nute, G. R., Fisher, A. V., Camp, M. M., Kasapidou, E., Sheard, P. R., and Enser, M., 2003. Effects of fatty acids on meat quality: a review. Meat Sci. 66:21-32.
- 121.Xargay, J., Fàbrega, E., Gispert, M., Francas, C., Soler, J., Tibau, J., ... Font i Furnols, M. (2010). Effect of pig breed on androstenone and skatole concentrations. Paper presented at the EAAP working group meeting "Production and Utilisation of meat from entire male pigs". Bristol: UK.
- 122.Zamaratskaia G., (2004). Factors involved in the development of boar taint influence of breed, age, diet and raising conditions, Doctoral thesis, Swedish University of Agricultural Sciences, Uppsala.
- 123.Zamaratskaia G., Babol J., Andersson H. K., Rydhmer L., and Lundström K., (2003). Relationships between testicular hormones, androstenone and skatole in entire male pigs fed raw potato starch. EAAP Working Group on Production and Utilization of Meat from Entire Male Pigs, Dublin, Ireland.
- 124.Zamaratskaia, G., and Squires, E.J., (2009). Biochemical, nutritional and genetic effects on boar taint in entire male pigs. Animal 3, 1508-1521.
- 125.Zamaratskaia, G., Chen, G. and Lundstrom, K., (2006). Effects of sex, weight, diet and hCG administration on levels of skatole and indole in the liver and hepatic activities of cytochromes P4502E1 and P4502A6 in pigs. Meat Science 72, 331-338.

- 126.Zamaratskaia, G., Oskam, I., Ropstad, E., Tajet, H., & Andresen, Ø., (2008a). Effect of Hcg stimulation on hepatic activities of cytochrome P4502E1 and P4502A in entire male pigs. Reproduction in Domestic Animals, 43, 147–152.
- 127.Zamaratskaia, G., Rydhmer, L., Andersson, H. K., Chen, G., Lowagie, S., & Andersson, K., (2008). Long term effects of vaccination against gonadotropin-releasing hormone, using Improvac<sup>™</sup>, on hormonal profile and behavior in pigs. Animal Reproduction Science, 108, 37–48.
- 128.Zamaratskaia, G., Zlabek, V., Chen, G. and Madej, A., (2009). Modulation of porcine cytochrome P450 enzyme activities by surgical castration and immunocastration. Animal 3, 1124-1132.
- 129.Zamaratskaia, G., Zlabek, V., Ropstad, E., & Andresen, Ø., (2012). In vitro and in vivo association of porcine hepatic cytochrome P450 3A and 2C activities with testicular steroids. Reproduction Domestic Animals, 47, 891–898.



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