

## QUALITY FEATURES OF FAT TISSUE AS A PLATFORM FOR “IDEAL” BACKFAT VIRTUAL MODEL: A REVIEW

Irina Chernukha<sup>1✉</sup>, Marina Nikitina<sup>1</sup>, Nadezhda Kupaeva<sup>1</sup>, Liliya Fedulova<sup>1</sup>

<sup>1</sup>V. M. Gorbatov Federal Research Center for Food Systems, Talalikhina st., 26, Moscow 109316, Russia  
✉imcher@inbox.ru

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**ABSTRACT**

The intensification of livestock farming and increased selection for “lean meat” breeds has led to the predominance of pigs with altered characteristics of adipose tissue. The meat industry faces the difficult task of providing consumers with high-quality meat and a variety of meat products. While scientists have to develop an optimal strategy to obtain high-quality new meat products with improved nutritional profiles and methods for measuring quality indicators of fat in order to meet both the requirements for a healthy diet of consumers and the technological requirements of the manufacturer.

This review describes the main methods of studying fat and the parameters of the quality of backfat are considered: the thickness of the dorsal fat, the color of the fat, Solid fat content, the determination of the fatty acid composition, and the ratio of fatty acids, iodine number, oxidative degradation. As a result of the systematization and literature analysis on the fatty acid composition of backfat, as well as the recommendations of the WHO, scientists, and nutritionists, a structural-parametric model of the “ideal” fat was formulated. Model summarizes and presents the characteristics of pork adipose tissue, that are optimal for obtaining meat products health-promoting.

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### 1.Introduction

The attitude to fats has changed dramatically over the past 100 years from positive (high nutritional value, energy reservoir) to sharply negative. Since the 1950s, nutritionists have recommended reducing total fat intake in order to decrease the potentially adverse effects of diet on diseases development such as obesity and coronary heart disease resulting from the consumption of excess fat, especially animal origin. Traditionally, animal fat, especially pork fat, is a source of such concerns. However, in recent decades, fat has been considered not only as a source of negative health effects. Taking into account the main potentials of fats, the recommendations of fat intake have moved from quantity of fat toward its quality (Kucha *et al.*, 2018).

### 2. The current state of pig livestock in Russia

Pork is the most consumed meat. Pig farming is intensively developing and occupying more than 30% of the total meat production. From 2016 to 2018 about 3.36-3.74 million tons of pork in carcass weight were produced in Russia, while in 2019 pork production overlapped 4 million tons and accounted for more than 37% of the total meat production. In 2019 about 2505.7 thousand tons of pork in live weight were produced on the territory of the Central Federal District. Enterprises of the Central Black Earth region occupy the largest share of pork production, locate in the Belgorod (17.8%), Kursk (7.9%) and Voronezh (5.9%) regions. Pig processing companies of the Volga region produce 813.7

thousand tons of pork, locate in the Republic of Tatarstan (2.0%), Bashkortostan (1.8%) and Mordovia (1.6%), respectively, in the gross volume of pig production in the country. Siberian producers rank third in the rating of federal districts with a share of 10.0% and a gross volume of 493.7 thousand tons, and enterprises of the Krasnoyarsk Territory, Omsk Region and Altai Territory achieve significant success (Tikhomirov, 2021).

The intensification of pig livestock in Russia and the imposed restrictive measures on the products from a number of foreign countries led to a sharp reduction in pork imports from 619.8 to 79.0 thousand tons and an increase in the cost of a unit of imported products by 80.2%. In 2019, pork occupied the largest share in the structure of the volume of imported supplies (87.2% of the total supply), followed by backfat (8.6%), and offal (4.3%). Brazil (51.8% of all supplies), Chile (23.8%) and Argentina (15.4%) were the main pork suppliers to the Russian market, followed by Paraguay (4.6%), Serbia (2.3%), Belarus (1.6%), Kazakhstan (0.5%); Chile (68.8% of total imports) was the main supplier of backfat (Plugov, 2021).

The volume of exports of pork, offal, backfat and commercial pigs from the Russia also increased from 2017 to 2020. For example, the total volume of exports in 2019 amounted to 108.3 thousand tons, exceeding the volumes for 5 years by 27.5%, for 10 years - by 238.6 times. The structure of total exports was previously dominated by offal, but in 2019 pork accounted for 54.8% in the total volume of export, pork offal - 40.0%, backfat - 4.8%, commercial pigs - 0.4%. Export volumes increased by 947.2% over 5 years, and by 19.9 times over 10 years, while the value of exports amounted to \$ 7.6 million. The total volume of pork backfat exports from Russia amounted to 5.2 thousand tons in 2019, which is 115.7% more than in 2018 (Grigoriev, 2021).

At the same time, there is a demand from domestic and foreign producers for backfat, which is necessary for the production of many types of meat products: the mass fraction of backfat in the recipes of boiled sausages is up to 30%, in semi-smoked, boiled-smoked, raw smoked sausages - up to 60% (Semenova *et al.*, 2015). The annual demand of the Russian meat industry for backfat is about 450 thousand tons. Currently, backfat is produced in Russia in extremely small volumes, because of the predominance of pigs with altered characteristics of adipose tissue in the agro-industrial livestock. Thus, there is a growing market demand for healthier sources of fat. To improve the fat content and its composition, several strategies are used, including feed varying and selection (Martins *et al.*, 2018). The meat industry faces a difficult task to provide consumers with high-quality meat and a variety of meat products, while scientists have to develop an optimal approach to obtaining high-quality products and methods for measuring quality indicators of fat in order to meet both the requirements for a healthy diet of consumers and the technological requirements of the manufacturer. The aim of the article was to review the most recent works on methods of studying adipose tissue and develop the concept of a "ideal" backfat virtual model.

### 3. Qualitative characteristics of the backfat

The quality of the backfat is evaluated after slaughter. The thickness of dorsal fat and color characteristics is measured, chemical and spectral analysis is carried out (Hoa *et al.*, 2019), as well as the fatty acid (FA) composition is determined and quality coefficients are calculated. The main traits of pig backfat and methods for their studying are presented in Table 1.

**Table 1.** The main traits for assessing the quality of pig backfat

Trait	The principle of determination or equation	Reference
<b>The thickness of dorsal backfat</b>	Measured with a flexible ruler with an accuracy of 0.5 mm: - at the 6-7 thoracic vertebrae; - at the 10-11 thoracic vertebrae;	Garskaya, Peretyatko, 2021
	- midline, between the 4th and 5th lumbar vertebra level;	Wang <i>et al.</i> , 2021
	- at the 14th rib level.	Ayuso <i>et al.</i> , 2020
<b>Color of backfat</b>	- CIELAB or CIEXYZ: In CIELAB color space L * (the lightness/darkness); a * (the redness/greenness); b * (the yellowness/blueness): b * value is considered the ideal objective measurement of the yellowness of a fat surface; L * value correlates well with the FAs composition.	Brewer <i>et al.</i> , 2001
	- colorimetric, spectrophotometric.	Hoa <i>et al.</i> , 2019
<b>Solid Fat Content</b>	SFC is the percentage of solids in fat at specified temperatures and determined by dilatometry; NMR and differential scanning calorimetry.	Kucha <i>et al.</i> , 2018
<b>SFA :UFA</b>	Ratio of saturated fatty acids to unsaturated fatty acids	Nistor <i>et al.</i> , 2012
<b>Iodine value</b>	- iodometric Wijs/ Hanuš method - calculated according to the equations: IV ( <i>for triglycerides</i> ) = [C16:1] × 0.95 + [C18:1] × 0.86 + [C18:2] × 1.732 + [C18:3] × 2.616 + [C20:1] × 0.785 + [C22:1] × 0.723; brackets indicate the concentration of FA in %; IV ( <i>for free FA</i> ) = [C16:1] × 0.9976 + [C18:1] × 0.8986 + [C18:2] × 1.810 + [C18:3] × 2.735 + [C20:1] × 0.8175 + [C22:1] × 0.7497	Kyriakidis and Katsiloulis, 2000; ISO 3961:2018.
	- coefficient for free FA is calculated according to the equation: $K_{FAi} = \frac{M_{j2} \times k}{M_{FAi}}$ where $M_{j2}$ – molar mass of molecular iodine, g, $M_{FAi}$ – molar mass of the i-th FA, k – the number of double bonds in the molecule of the i-th FA - coefficient for triglycerides is calculated according to the equation: $K_{FAi} = \frac{M_{j2} \times k}{(M_{FAi} + \frac{1}{3} M_{gl} - M_{H2O})}$ where $M_{gl}$ – molecular weight of glycerin, g, $M_{H2O}$ – molecular weight of water, g; IV ( <i>for free FA</i> ) = $\sum K_{FAi} \times m_{FAi}$ , where $K_{FAi}$ – coefficient for i-th FA determined for free FA or for triglycerides, $m_{FAi}$ – mass content of i-th FA in the sample, %.	Spiridonov <i>et al.</i> , 2016
	- regression equations.	Paulk <i>et al.</i> , 2015
<b>Fatty acid composition</b>	- Fatty acid methyl esters analysis using GC/MS; - Near infrared reflectance (appropriate for C16:0, C18:0, C18:1 and C18:2); - Raman spectroscopy	Ayuso <i>et al.</i> , 2020; De Marchi <i>et al.</i> , 2012; Ros-Freixedes <i>et al.</i> , 2014
<b>Oxidative Degradation</b>	- peroxide value (PV); - malondialdehyde (MDA)	Barriuso <i>et al.</i> , 2013; Qiu <i>et al.</i> , 2013.

**3.1. The thickness of subcutaneous backfat** is traditionally determined after slaughter, but modern methods of ultrasound examination in real time allow predicting the thickness of subcutaneous and intramuscular fat *in vivo* (Jung *et al.*, 2015). The thickness of dorsal backfat is traditionally measured in Russia at the 6-7 and 10-11 thoracic vertebrae (Garskaya and Peretyatko, 2021), in China – midline, between the 4th and 5th lumbar vertebra level (Wang *et al.*, 2021), while in Italy – at the 14th rib level (Ayuso *et al.*, 2020).

**3.2. Backfat Color** of mainly depends on the freshness and fatty acids composition. Fresh fat has a white or slightly pinkish color. A high content of unsaturated fatty acids causes a grayish tinge of fat and occurs in young animals. Unusual color, an orange for example, as a rule, is a result of the reaction of unsaturated fatty acids with molecular oxygen by a free radical mechanism (Domínguez *et al.*, 2019; Pereira and Abreu, 2018). Various equipment is used for the studying the color of fat, which evaluate the following CIE L\* a\* b parameters: for lightness from black (0) to white (100) - L\*; from green (-) to red (+) - a\* and from blue (-) to yellow (+) b\* (CIE, 1986; Hoa *et al.*, 2019). The b\* value is considered the ideal objective measurement of the yellowness of a fat surface (Brewer *et al.*, 2001). In subcutaneous fat CIEL\*a\*b\* variables significantly affect color with L\* and chroma being the most affected. L\* value correlates well with the FAs composition (Kucha *et al.*, 2018). In (Maw *et al.*, 2003) a correlation was found between increased yellowness and an elevated percentage of linoleic and  $\alpha$ -linolenic acid, which accompanied with a decrease in the levels of palmitic, palmitoleic and oleic acids. Elevated transparency and softness of fat correlated with a decrease in the percentage of palmitic, stearic and oleic acid while increasing the ratio of linoleic and  $\alpha$ -linolenic acid. At the same time, an increased percentage of myristic acid was associated with a decrease in red color.

**3.3. Solid fat content (SFC)** could be defined regarding firmness and the percentage of solids in fat at specified temperatures.

Firmness is usually measured by several instrumental techniques for instance by a penetrometer, a texture analyzer and Instron materials testing machine (Dransfield and Jones, 1984; Warnants *et al.*, 1998; Glaser *et al.*, 2000). SFC is an important feature of fat that affects the appearance, flavor, melting rate, shelf life and stability of fat-based foods. It is known that the melting point of FA with an even and odd number of carbon atoms differs. The C', C'' and C forms are the only ones which melt. Namely, A', B', C', C'' and D' forms can be observed for the odds, and A, B, C and E forms for the evens (Gbabode *et al.*, 2010). The length of FA positively correlated with the melting point. Thus, melting point for C14 average 54.0°C, while for C26 – 88.2°C (Francis *et al.*, 1930). According to the melting point, it is possible to empirically proposing the FA composition of adipose tissue and vary the melting point by lifetime or technological manipulation.

SFC is particularly of primary importance when considering fat quality because soft fats could be difficult to dimension and process thus causing the decrease of high-quality cuts that could lead to monetary loss of value. The softness or oiliness of fat depends on the FA composition which also affects the SFC of pork fat at any given temperature. It was found that the melting point of lipids, as well as the hardness of fat, are closely related to the concentration of stearic acid (18: 0) and palmitic acid (C16:0), and has a linear correlation with iodine value at 20°C (Davenel *et al.*, 1999).

Products with an ideal SFC is desirable during processing, which provide the product to remain solid at room temperature, but at the same time give consumers the desired texture in the mouth. In other words, the melting point of backfat is recommended not lower than 35°C for the technological processing in the production of meat products using heat treatment (cooking, smoking, etc.), while the melting point of fat in the finished product may

be lower (32 - 36°C). Thus, the SFC is an important parameter of fat quality control to achieve high quality of the final product.

Traditionally, dilatometry is used to study SFC in routine practice. New methods, such as Nuclear magnetic resonance (NMR) and differential scanning calorimetry (DSC), are also developed. NMR results are calculated based on the relative number of protons present in the triglycerides in the solid and liquid phases, while DSC data are obtained by the melting enthalpies of these triglycerides (Márquez *et al.*, 2013). NMR methods are fast, non-destructive and rarely required recalibration over a long period of measurement. However, sample preparation (tempering) is required before measuring at each required temperature. DSC provides the possibility of tempering fat at various temperatures before measurement, providing a thermogram that includes the entire temperature range, as a result of a single measurement, from which an SFC can be obtained by partially integrating the thermogram. Thus, DSC provides information about the thermal transitions that the fat may undergo during processing since adding fat to a product without the desired melting profile could cause encapsulation of other ingredients. DSC measurement of melting characteristics is an internationally accepted conventional method by the American Oil Chemists Society.

Currently, NMR and DSC methods still require sample preparation and cannot be used for rapid assessment of fat SFC. Therefore, attempts are still being made to find more suitable methods for determining the SFC (Kucha *et al.*, 2018).

**3.4.The ratio of saturated to unsaturated fatty acids** is one of the ways to describe the relative composition of the fatty acid profile (Azain, 2001; Nistor *et al.*, 2012). The total amount of saturated (SFA), monounsaturated (MUFA) and polyunsaturated (PUFA) fatty acid and the ratio between FA, especially SFA:PUFA, are often used to determine the quality of fat with higher levels of saturation indicating a more desirable quality, while

increased unsaturation indicate an undesirable quality of fat. The ratio of all PUFA to SFA, the target value for which is from 0.4 to 0.58, for pork fat may exceed these values and this may be a favorable factor for pigs and other monogastric animals compared to ruminants (Nieto and Ros, 2012).

It is considered that a good quality fat from the standpoint of technological suitability should have a composition of 12% or more of C18: 0, the proportion of C18: 2 should be 12-15% of the total amount of FA. Thus, the ratio of these two key FA should be 1:[1-1.25] (FAO Food and Nutrition Paper, 2010). The total ratio of unsaturated  $\omega 6/\omega 3$  FA is preferable to be more than 3.5-4.5 (Debreceńi *et al.*, 2017). In addition, according to (Pascual *et al.*, 2007) C18: 2w6 can be considered as a marker of the feed, and C16:0 as a marker of de novo synthesis. It is advisable to pay attention to their content in pork.

**3.5.The iodine value (IV)** characterizes the degree of unsaturation of carbon bonds in the structure of fatty acids. This parameter is used for standardization of the characteristics and composition of lipids in fat and is the most commonly used industry standard for quantifying the degree of unsaturation of pork. The advantage of the iodine value is that it corresponds to the amount of iodine that can interact with fatty acids by double bonds (White and Latour, 2008). IV also characterizes the resistance of fat to oxidation (Kyriakidis and Katsiloulis, 2000). A standardized method for calculating the iodine value is used by titration, and the calculated value for free fatty acids is also used ISO 3961:2018. In (Spiridonov *et al.*, 2016) a new method for calculating the IV was proposed, taking into account the content of all unsaturated fatty acids identified in the fat. Acceptable IVs range from less than 70 g/100 g to 75 g/100 g (Corominas *et al.*, 2014). In (Paulk *et al.*, 2015) based on the literature data, equations were proposed to predict the IV in the pork carcass back, belly, and jowl fat, when fattening with various sources of FA.

A high IV and a decrease in the ratio of saturated to unsaturated FA indicate a decrease in the hardness of fat. Many producers use IV for evaluation of carcass quality and IV > 65 for some processes may be unacceptably high, while IV > 75 is the threshold value for many technological processes. Currently, most US enterprises set the IV standard at a level below 74 g/100 g. (Seman *et al.*, 2013). In Russia, fat suitable for long-term storage and for the production of delicatessen meat products should have a IV of no more than 70 g / 100 g (Spiridonov *et al.*, 2016).

**3.6.FA profile of pork fat.** Fatty acid methyl esters (FAMES) analysis using GC/MS is the most common method for determining the FA profile of pork fat. The FA composition is usually expressed as a set of percentages corresponding to the relative content of each FA in the total number of determined FA (Ros-Freixedes and Estany, 2014). FA are classified into saturated and unsaturated (UFA) according to the chemical structure. UFAs are divided into monounsaturated and polyunsaturated. The FA proportions determine the physical properties and distinguish one fat from others: SFA are solid at room temperature, have a higher melting point than MUFA, PUFA, which contain at least one double bond in their structure. Moreover, as the number of double bonds increases, the fat becomes more unsaturated with a lower melting point and a softer consistency at room temperature. Various factors such as feed, gender, breed and age influence on FA composition.

Among the modern methods for assessing the FA profile, NIRS is used and let to develop quantitative models for FAs and the FA classes including SFA, MUFA, and PUFA. This method allows analyzing fat samples both in homogenized or intact forms. Better predictions were obtained for palmitic acid (C16: 0), stearic acid (C18: 0), oleic acid (C18: 1) and linoleic acid (C18: 2), while poor prognosis obtained for the remaining FA was associated with low concentrations of FA in the sample (De Marchi *et al.*, 2012).

Raman Spectroscopy (RS) is a vibrational spectroscopy method based on the shifts in the wavelength or frequency of an exciting incident beam of radiation that result from inelastic scattering on the interaction between the photons and the sample molecules. RS is also used to evaluate fatty acid classes using partial least square regression (PLSR) in combination with various data preprocess. Olsen, Ruke, Egeland and Isaksson (2008) used RS to develop prediction models for the quantification of omega-3 and omega-6 in adipose tissue by analyzing Raman spectra of pork subcutaneous fat. An accuracy of R 20.97 and 0.91 and a prediction error of 0.99 and 0.23 were achieved for omega-3 and omega-6, respectively, based on preprocessed PLSR and first derivative spectra and selected wavelengths. Surprisingly, the study showed poor results in predicting the ratio of omega-6 and omega-3 in non-extracted fat with a determination coefficient of 0.31.

In another study on the determination of FA groups using RS (Berhe *et al.*, 2016) the method of multidimensional PLSR modeling was used on the fingerprint region of the spectra of subcutaneous pork fat samples and obtained reliable results for predicting SFA, MUFA, PUFA and IV, achieving reliable R<sup>2</sup> with a reasonable root mean square error of prediction. In addition, individual fatty acids including C14: 0; C16: 0; C16: 1 cisΔ9; C17: 0; C18: 0; C18: 1 cisΔ9; C18: 1 cisΔ11; C18: 2 cisΔ9, 12; C18: 1 cisΔ9, 12, 15; C120: 0; C20: 1 trans Δ11; C20: 2 cisΔ11, 14; C20: 3 cisΔ8, 11, 14; C20: 4 cisΔ11; C20: 1 cisΔ5, 11, 14 were predicted with R<sup>2</sup> of 0.67, 0.89, 0.56, 0.07, 0.72, 0.82, 0.43, 0.90, 0.87, 0.18, 0.46, 0.78, 0.35, 0.60, 0.87 and prediction errors of 0.06, 0.20, 0.20, 0.09, 0.87, 1.3, 0.18, 1.84, 0.22, 0.05, 0.09, 0.10, 0.05, 0.05, and 0.22, respectively. This study showed RS's ability to determine the quality of pork fat. Moreover, Berhe *et al.* (2016) revealed that the poor prediction of the ratio of omega-6 to omega-3 obtained by Olsen, Rukke, Egeland and Isaksson could be linked to the modelling of FAs based on the same Raman spectra information arising as a consequence of



strong correlation of a less abundant FA to a more abundant FA or their groups (IV, SFA, MUFA, PUFA). In fact, this means that the high coefficient of determination obtained for both less common and more common FAs can be changed or for the entire group of FAs. The study considers that a good prediction model for FA can be obtained by RS when using the PLSR algorithm. It is important to investigate the correlation structure of individual FAs and the degree of unsaturation using other non-destructive spectroscopic methods that demonstrate high collinearity of their spectra (Kucha *et al.*, 2018).

**3.7.Oxidative degradation** of lipids is an important indicator of the quality of fats, meat and meat products, which degree effects on sensory properties (color, aroma, taste, texture) and nutritional value of foods, can negatively affect human health. Lipid oxidation products are harmful to the body due to carcinogenic and atherosclerotic effects, changes in the composition of cell membranes and a decrease in high-density lipoproteins (Reitznerová *et al.*, 2017).

PUFA are more prone to oxidation than MUFA and SFA, and form hydroperoxides with conjugated double bonds (conjugated diene hydroperoxides and conjugated triene hydroperoxides). UFA with two double bonds form conjugated dienes hydroperoxide, while fatty acids with three double bonds form conjugated trienes hydroperoxide (Papuc *et al.*, 2018).

Further, the primary decomposition products (hydroperoxides) decompose to form various low molecular weight secondary compounds, including aldehydes, ketones and alcohols – MDA, pentanal, propanal and hexanal. These compounds are responsible for the development of rancidity, undesirable odor and color changes (Barriuso *et al.*, 2013). 4-hydroxy-2-nonenal is the most permanent aldehyde substance formed during the peroxidation of PUFA  $\omega 6$ , such as C18:2 $\omega 6$  and C20, and accumulates in membranes at concentrations of 5-10  $\mu\text{M}$ . Lipid oxidation of PUFA  $\omega 3$  leads to the formation of 4-hydroxy-

2-hexanal, the concentration of which in meat can reach 120  $\mu\text{M}$  (Kanner, 2007). Dialdehyde, including malonic dialdehyde (MDA), glyoxal and acrolein are other important reactive aldehydes formed as a result of the peroxidation of linolenate lipids and arachidonic acid. In many cases, MDA is the most common individual aldehyde, which is formed in foods undergoing lipid peroxidation. Its concentration in meat products can reach more than 300  $\mu\text{M}$  (Reitznerová *et al.*, 2017).

Modern methods for lipid oxidation assessment can measure changes in primary products (changes in FAs and formation of lipid hydroperoxides and conjugated dienes or trienes) and changes in secondary products (formation of carbonyls, aldehydes, volatiles, MDA). The most common approach to assessing the degree of oxidation is to measure both primary and secondary oxidation compounds (Domínguez *et al.*, 2019). A significant decrease in the content of UFAs is expected during lipid oxidation, since they are the main substrate for oxidative reactions. However, the analysis of FAs is an additional indicator of the degree of oxidation, since the effective loss of UFA can be quantified only at the last stage of the oxidation process (Guyon *et al.*, 2016). The measurement of the hydroperoxides generation, also called the peroxide value (PV), has long been used as the main indicator of the formation of primary oxidation compounds in meat and meat products. A low PV may indicate both early and late oxidation (Domínguez *et al.*, 2015). In this regard, the determination of the PV is effective only at the initial stages of oxidative processes. It is recommended to monitor the change in the PV over time in order to get complete information about the state of lipid oxidation and to know whether the lipid is at the site of growth or decay of the hydroperoxide concentration curve (Yang and Boyle, 2016). Methods for determining peroxides are based on their reducing ability (Barriuso *et al.*, 2013). This requires extraction of lipids, which contain hydroperoxides that easily oxidize inorganic ions such as iodine or

iron. Iodometric titration and Ferric-xylenol Orange (FOX) are two main methods used for the determination of peroxides. FOX is a simpler method, does not depend on the availability of oxygen (Gómez and Lorenzo, 2013). The results obtained by the FOX method correlate better with other oxidation parameters than iodometric titration (Nuchi *et al.*, 2009).

Another marker of primary oxidation is the formation of conjugated compounds (dienes and trienes), the measurement of which is used to monitor oxidation in meat and meat products. The method of its determination includes extraction of conjugated compounds with a small amount of organic solvent (hexane/ isopropanol or chloroform / methanol mixture), and then the concentration of conjugated dienes and trienes is measured in the organic phase at 234 and 268 nm, respectively, on a spectrophotometer (Domínguez *et al.*, 2019). This method is simple and does not require expensive reagents; small amount of sample is used. However, this method has a significant number of disadvantages, including low sensitivity, so it is recommended to use other methods together with the measurement of conjugated compounds.

7-ketocholesterol, 20 $\alpha$ -hydroxy-cholesterol, 25-hydroxycholesterol,  $\alpha$ ,  $\beta$ -epoxycholesterol, and 7 $\alpha$ , 7 $\beta$ -hydroxycholesterol are the most commonly presented in meat and meat products. Concentrations between 57 to 71  $\mu\text{g}/100\text{ g}$  can be found in meat and meat products (Domínguez *et al.*, 2019). GC-MS is the most accurate and frequently used method for quantifying cholesterol oxidation products (Chiu *et al.*, 2018). However, the method requires expensive equipment and a number of complex processes, including extraction, saponification, purification and derivatization of lipids in order to increase their volatility and thermal stability, as well as reduce contamination (traces of cholesterol and /or partial glycerides). Measurement of primary oxidation products provides reliable information only in the early stages, since they

are unstable and decompose rapidly, reducing their content as oxidation increases. Therefore, the measurement of secondary compounds is more suitable for determining the degree of oxidation of meat or meat products. Secondary compounds are stable, and they also cause the appearance of rancid taste and smell, which affects the quality of meat.

MDA and hexanal are the most important and widespread aldehydes used as lipid peroxidation indices (Qiu *et al.*, 2013). In fact, different studies established values of 2–2.5 mg MDA/kg as the accepted limit in which there is no rancidity in fat and meat (Campo *et al.*, 2006; Zhang *et al.*, 2019). The main method of quantitative determination of MDA is the method based on MDA, as specific product of lipid peroxidation, reaction with thiobarbituric acid (TBA) lead to formation of a colored complex with maximum absorption at 532 nm (Mousavi *et al.*, 2018).

However, the reaction with TBA is not specific to MDA. There are several aldehydes and other oxidation products that also react with TBA, so the method is called thiobarbituric acid reactive substances (TBARS) (Yang and Boyle, 2016) and is used to evaluate total lipid oxidation, rather than quantifying MDA (Banerjee *et al.*, 2017). There are several variants of the TBARS method with different conditions for extracting MDA from food samples.

HPLC and GC methods provide better specificity and sensitivity when detecting MDA. However, the TBARS spectrophotometric methods are the most widely used to assess the oxidative state of meat and fat in routine analysis due to their simplicity and low cost (Pereira and Abreu, 2018). This method has a good correlation with sensory deterioration of the products quality (Domínguez *et al.*, 2019).

In addition to MDA, several aldehydes, including alkanols, 2-alkenals and 2, 4-alkadienals, are formed during lipid hydroperoxides decomposition. Determination of p-Anisidine value is a general spectroscopic method for measuring the amount of secondary lipid oxidation products formed during the



decomposition of peroxides and hydroperoxides (Majchrzak *et al.*, 2018). This method requires extraction of lipids, however, the p-Anisidine value is considered a good indicator, since it correlates with other indicators of both primary (PV) and secondary oxidation (TBARS), as well as with deterioration of organoleptic qualities (Barriuso *et al.*, 2013). The measurement of carbonyls (aldehydes and ketones) also makes it possible to track secondary lipid oxidation processes. Methods for determining carbonyls are based on interaction with 2,4-dinitrophenylhydrazine in an aqueous solution, as a result of which carbonyls turn into orange-colored hydrazones extracted by hexane. This is a simple and fast method that correlates well with the deterioration of the taste resulting in rancid taste (Domínguez *et al.*, 2019).

More recently, other analytical methods have been developed for the determination of lipid oxidation products in meat and meat products (chemiluminescence, fluorescence

emission, RS, infrared spectroscopy or NMR). It is worth noting that the oxidative susceptibility is more influenced by the unsaturation of fat than the amount of fat.

#### 4. Fatty acid composition of adipose tissue vs consumer health: “ideal” backfat virtual model

Consumer concern about the composition of the food product, the desire for healthy food has become synonymous with low-fat, or even free-fat food. Moreover, this approach primarily concerns animal fats. Anxiety about the harmful effects of cholesterol and saturated fatty acids on the cardiovascular system has led to changes in pig livestock technologies everywhere. Pig breeds with the thickness of the backfat lower 1.0 cm have been bred. Combined feeds contribute to a change in the FA composition of fat towards a decrease in the proportion of saturated fats. In this regard, fat was also evaluated by its atherogenicity (AI) and thrombogenicity (TI) indices.

$$AI = \frac{C12:0 + C14:0 + C16:0}{n-3PUFA + n-6PUFA + MUFA}, \quad (1)$$

$$TI = \frac{C14:0 + C16:0 + C18:0}{0.5 \times MUFA + 0.5 \times n-6PUFA + 3 \times n-3PUFA + n-3PUFA/n-6PUFA} \quad (2)$$

Currently, these indicators are calculated according to the equations proposed by (Ulbricht and Southgate, 1991) in various modifications, for example, according to the equation described by Campo *et al.* (2013).

The values of both indicators are linked to certain FA composition. The positive role of omega 3 FA in reducing the risk of atherosclerosis is known, as well as C 12:0, C 14:0 и C 16:0 FA contributes to an increase of low-density cholesterol level in the blood, whereas C 18:0 does not demonstrate such an effect.

Systematization and analysis of literary (Fallon S., 1999; Pascual *et al.*, 2007; Domínguez *et al.*, 2015; Semenova *et al.*,

2019; Huang *et al.*, 2020) and own data on the FA composition assessment of pork fat, as well as WHO, scientists and nutritionists recommendations (FAO Food and Nutrition Paper, 2010) resulted in Table 2 in the form of a structural-parametric model, which are summarized and presented characteristics of pork adipose tissue optimal for obtaining products with health-promoting properties.

The structural-parametric model is a square matrix and is formed in the blocks (Ivashkin, 2004). They are placed along the main diagonal and combine operators of functional relationships within the selected groups of parameters reflecting the variety of existing known and theoretically defined relationships

between the content of individual FAs and the characteristics of fat. If the parameters belonging to the same group are independent, the corresponding block on the main diagonal is a unit matrix. If there are relationships between parameters, the extra-diagonal elements of the selected blocks describe

interaction operators both within groups and between parameters of other groups. Then the extra-diagonal square of the original block matrix correspond to the operators of direct and indirect intergroup influence of individual parameters belonging to different functional groups.

**Table 2.** “Ideal” backfat structural-parametric model

k	Traits of “ideal” backfat								SFA														MUFA						PUFA								
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	
1	x																					*	!!	*	*	*	!!	*	!!	*	*	*	*	*	*	*	
2		x							*	*	*	*		*		*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*
3			x											!!		!!		!!		*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	
4				x										*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*
5					x														!!				*	*	*	*	*	*	*	*	*	*	*	*	*	*	
6						x			*	*	*	*		*		!!		!!		!!		*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*
7							x							*			!!	!!	!!	!!	!!		*	*	*	*	*	*	*	*	*	*	*	*	*	*	
8								x														*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	
9		*				*			x																												
10		*				*				x																											
11		*				*					x																										
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25	*	*	*	!!	*	*																		x													
26	!!	!!	!!	*	*	!!	*	*																	x												
27	*	*	*	*	*	*		*	*																	x											
28	*	!!	*	*	*	*		*																			x										
29	!!	*	*	*	!!	!!	*	*		!!																		x									
30	!!	*	*	*	*	*		*	*		!!																		x								
31	*		*	*	*	*		*	*																						x						
32	*	*	!!	!!	*	*		*	*																							x					
33	*	*	!!	*	*	*		!!	!!																								x				
34																																		x			
35																																			x		
36	*	*	!!	*	*	*		!!	!!																											x	

Note:

1 – IV; 2 – Trans fat content; 3 – AI; 4 – IA/IT ratio; 5 – Saturated index (SI); 6 – Melting point; 7 – Index  $\omega$  3; 8 –  $\omega$  6;  $\omega$  3; 9 – C4:0; 10 – C6:0; 11 – C8:0; 12 – C10:0; 13 – C11:0; 14 – C12:0; 15 – C13:0; 16 – C14:0; 17 – C15:0; 18 – C16:0; 19 – C17:0; 20 – C18:0; 21 – C19:0; 22 – C20:0; 23 – C14:1; 24 – C 15:1; 25 – C16:1; 26 – C18:1; 27 – C20:1; 28 – C 22:1; 29 – C18:2  $\omega$  6; 30 – C18:3  $\omega$  3; 31 – C 20:2  $\omega$  6; 32 – C20:4  $\omega$  6; 33 – C 20:5  $\omega$  3; 34 – C 22:2  $\omega$  6; 35 – C 22:5  $\omega$  3; 36 – C 22:6  $\omega$  3

\* – low correlation; \*\* – medium correlation; !! – significant correlation.

$$\Phi(x) = 1 - \sqrt{\frac{1}{n} \sum_{i=1}^n b_i \left( \frac{x_i - x_i^0}{\Delta x_i^l} \right)^2} \rightarrow \max \quad (3)$$

where n – the number of estimated indicators, according to the structural-parametric model, their number is equal to 8;  $x_i$ ,  $x_i^0$  – actual and desired value;  $\Delta x_i^l$  – the maximum deviation from the desired value for the  $l$ -th quality level;  $b_i$  – the weighting coefficient of the  $i$ -th parameter.

Parameters of FA composition are presented in three blocks: SFA (k=9 to 22),

MUFA (k=23 to 28), PUFA (k=29 to 34). Knowledge and analysis of patterns,

relationships, including structural ones, allow creating a model of "ideal" backfat. Block "Traits of "ideal" backfat" includes indicators forming the quality functionality (Nikitina *et al.*, 2019). Qualitative indicators can make an equivalent contribution to the formation of the quality of an "ideal" backfat, or they can have their own weight value (contribution) different from other indicators. The determination of weight coefficients can be carried out by methods of expert assessments, factor experiment, etc.

The value of the quality functional changes from 1 when the obtained values completely coincide with the optimal ones (the best quality) to 0 when the quality level limit is reached (the limit value).

The FA composition of adipose tissue has a significant effect on the quality traits of the "ideal" backfat: blocks of SRA, MUFA, PUFA in the structural-parametric model. The presence of a correlation in the matrix table is marked with signs (\*, \*\*, !!), which corresponds to a low, medium and significant correlation between fatty acids and indicators of the quality of "ideal" backfat. Numerical values can be obtained using correlation and regression analysis.

The equations, ratios, criteria used in the assessment of FA composition are presented in Table 3, and the optimal values are also given, which must be achieved in order to have the

most satisfying (sufficient, significant) effect on the qualitative traits the "ideal" backfat.

## 5. Discussions

Various factors should be taken into account in order to meet industry requirements and consumers demands for a quality of product during the developing an optimal pig livestock system. On the one hand, saturated animal fats contribute to the cardiovascular diseases development, on the other hand, fat are an important component of the human diet, being the main source of energy, and contains a range of essential fatty acids that have a positive effect on consumer health. At the same time, adipose tissue is important and determines the quality of pork. However, less attention has been paid to the qualitative characteristics of fat in recent decades compared to other characteristics of meat quality, including pH, moisture loss, moisture content, tenderness and color. Traditionally, approaches for assessing the quality of fat, such as gas chromatography, iodometric titration and spectrophotometric methods, are widely popular for assessing fatty acids, iodine value (IV) and fat resistance to oxidation. The IV not exceeding 0.74 g/100g can be considered as the optimal value (Seman *et al.*, 2013). The creation of an effective method for assessing the traits of fat quality and the development of criteria and indicators of the "ideal" backfat are very relevant today.

**Table 3.** Optimal backfat traits for a healthy diet

Reference \ Trait	Value	Equations/ criteria of optimality*
<b>MUFA:PUFA (ω3, ω6):SFA ratio</b>	1:1:1	$\sum_{k=9}^{22} q_k lx + \sum_{k=23}^{28} q_k lx = 2 \sum_{k=29}^{36} q_k lx$
FAO Food and Nutrition Paper, 2010		
<b>MUFA:PUFA:SFA ratio</b>	0.48:0,12:0.4	$0.3 \sum_{k=9}^{22} q_k lx = 1.2 \sum_{k=23}^{28} q_k lx$ $\sum_{k=9}^{22} q_k lx + \sum_{k=23}^{28} q_k lx = 1.5 \sum_{k=29}^{36} q_k lx$
Fallon S., 1999		
<b>PUFA:SFA ratio</b>	0.4–0.6	$0.4 \leq \frac{\sum_{k=29}^{34} q_k lx}{\sum_{k=9}^{22} q_k lx} \leq 0.6$
Nieto&Ros, 2012; Ulbricht & Southgate, 1991		
<b>MUFA:PUFA ratio</b>	2,5–3,5	$2.5 \leq \frac{\sum_{k=23}^{28} q_k lx}{\sum_{k=29}^{34} q_k lx} \leq 3.5$
Lisitsyn <i>et al.</i> , 2013		

<b>SI</b>	$\leq 1$	$SI = \frac{q_{16} + q_{18} + q_{20}}{\sum_{k=23}^{28} q_k + \sum_{k=29}^{36} q_k}$
Martins et al., 2015		
<b><math>\Sigma</math> C14 and C 16</b>	25%	$(q_{16} + q_{18}) = 0.25 \sum_{k=9}^{36} q_k$
<b>C18:0 content</b>	$\geq 12\%$	$q_{20} \geq 0.12 \sum_{k=9}^{36} q_k$
<b>C18:2 content</b>	12–15 %	$q_{20} \geq 0.12 \sum_{k=9}^{36} q_k$
<b>18:3n–3:18:2n–6 ratio</b>	1–(3–5)	$1 \leq \frac{q_{30}}{q_{29}} \leq 5$
Lisitsyn et al., 2013		
<b>C18:0:C18:2 ratio</b>	1.1–1.25	$1.1 \leq \frac{q_{20}}{q_{29}} \leq 1.25$
FAO Food and Nutrition Paper, 2010		
<b><math>\Sigma</math> C 11, C 13, C 15, and C 17 (Mainly due to the pentadecanoic FA)</b>	0.4–0.6%	$0.04 \sum_{k=9}^{36} q_k \leq (q_{13} + q_{15} + q_{17} + q_{19}) \leq 0.06 \sum_{k=9}^{36} q_k$
<b><math>\Sigma</math> C22:5 and C22:6n3</b>	$\geq 0.25\%$	$(q_{35} + q_{36}) = 0.025 \sum_{k=9}^{36} q_k$
<b>Arachidonic cis–5, 8, 11, 14– C20:4<math>\omega</math>6 content</b>	0.8–1.1	$0.8 \leq q_{32} \leq 1.1$
<b>FA medium chain (C11 – C16): FA long chain (&gt;C17) ratio</b>	1.6–2.0	$1.6 \leq \frac{\sum_{k=13}^{18} q_k}{\sum_{k=19}^{22} q_k + \sum_{k=26}^{36} q_k} \leq 2$
<b><math>\omega</math>6:<math>\omega</math>3 ratio</b>	3,5–4,5	$3.5 \leq \frac{q_{29} + q_{31} + q_{32} + q_{34}}{q_{30} + q_{33} + q_{35} + q_{36}} \leq 4.5$
Debrecéni et al., 2017		

Note: \* according to the structural–parametric model (in corresponding units): q – FA content of fatty acid in the “ideal” backfat; k – FA identifier, for example  $q_{32}$  – corresponds to C20:4, a  $q_{22}$  – C20:0, etc.; l – the amount of fat in the “ideal” backfat; x – the amount of the “ideal” backfat.

This article describes the criteria for the optimality of pig fat tissue – the first stage on the way to creating “ideal” backfat virtual model.

A comprehensive assessment of the fatty acid (FA) composition and the application of accumulated knowledge to the identification of quality–determining FA have great potential. So, for example, it is known that myristic and myristoleic FA have an effect on the melting of fat. Some monounsaturated FA may protect against the risk of cardiovascular diseases development (Briggs et al., 2017). Odd-numbered FAs are rarely detected in pork fat, or are found in trace amounts, for example, C:19 and C:21 FA (Lisitsyn et al., 2013). The total increase in odd-numbered FAs content begins with C:15. The recommended PUFA:SFA ratio is higher than 0.4. Such values can be achieved by the in vivo influence

on pig adipose tissue through the use of certain ingredients in feed. J.V. Pascal et al obtained pork with a PUFA:SFA ratio of 0.6–0.74, using linolenic acid–rich feed when raising pigs of the Landrace and Duroc breed (Pascual et al., 2007). The  $\omega$ –6: $\omega$ –3 ratio is an important trait. Experts point out that the  $\omega$ –6 –  $\omega$ –3 FA should be at the level of 4–5% of the fat component of the diet. At the same time, scientists differ in determining the optimal  $\omega$ –6: $\omega$ –3 ratio, which ranges from 3:1 to 10:1, while nutritionists claim: the  $\omega$ –6: $\omega$ –3 ratio of a healthy adult diet should be in the range 4:1–6:1 (FAO Food and Nutrition Paper, 2010).

The negative attitude towards trans fats is well known. WHO recommends the use of technologies that do not cause the formation of trans fats. This means, that the product should be free of trans–fatty acids of non–natural origin. However, trans isomers of FA

are found in nature. For example, vaccenic FA, trans-11-octadecenoic acid (C18:1  $\omega$ 7) is synthesized in ruminants in significant quantities. At the same time, some authors note its presence in pork (Domínguez *et al.*, 2019; Aboagye *et al.*, 2020).

Vaccenic acid is metabolized to form rumenic, or conjugated linoleic acid – polyunsaturated FA ( $\omega$  –6), in organism. The "omega 3 index" indicator proposed by Harris and von Schacky is included in the number of fat characteristics and calculated as the sum of eicosapentaenoic and docosahexaenoic fatty acids (Harris and von Schacky, 2004). These FA are essential and contribute to the elimination of low-density lipoproteins, reduction of inflammatory processes, decrease the risk of cardiovascular diseases. Docosahexaenoic acid (DHA) is found in large quantities in brain tissues and the retina of the eye, being their structural component (Arterburn *et al.*, 2006).

Eicosapentaenoic acid is a precursor of prostaglandins, promotes the formation of anti-inflammatory protective function of the organism. In adipose tissue, these FA are contained in very small amounts, which indicates the need to replenish these FA. Although these acids can be synthesized from alpha-linolenic FA, this process is very complex and inefficient with coefficient of efficiency only of 0.1. At the same time, it has been shown that the content of these two FA in human blood in an amount of 8% dramatically reduces the risk of mortality from cardiovascular diseases (Huang *et al.*, 2021), and in concentrations above 4% of the total fatty acid content they could contribute to the normal metabolism of brain cells, retina, heart tissue. This indicator should tend to 1 in the adipose tissue of pigs. It is advisable to pay attention to such a long-chain FA as eicosatriene (C20:3  $\omega$ –3), which is usually either not detected during analysis, or is detected in trace amounts. The presence of this FA in detectable amounts indicates the priority activity of synthesis of  $\omega$  – 3 PUFA due to  $\omega$  – 6 PUFA (Pascual *et al.*, 2007).

As mentioned above, many doctors consider fat as a potential source of cholesterol and a promoter of cardiovascular diseases. It is worth noting that currently not all cholesterol is considered harmful to health, but its individual fractions, especially cholesterol of low density lipoproteins. The equation proposed by Ulbricht TLV and Southgate DAT in 1991 for calculating the atherogenicity of a food product by the ratio of individual fatty acids divided fatty acids into those that have a significant effect on the risk of cardiovascular diseases development and others that, on the contrary, contribute to the removal of excess fats from the body. Previously, we determined the atherogenicity and thrombogenicity indices of various pork samples obtained from animals of different breeds and location on carcasses. There was no correlation between the total fat content and the atherogenicity of the studied samples. However, the correlation between the indices and the location on carcasses has been established. The minimum value of AI = 0.53 was found in the outer part of the hip of carcass, and the maximum value was 0.84 in the spinal-lumbar part of the middle carcass. At the same time, the atherogenicity of pork is lower than that of beef. Own and literary data allowed determining the optimal ratio of AI:TI ranges from 0.4 to 0.6. The focus on the indicators of thrombogenicity and atherogenicity of pork made it possible to formulate more precisely the requirements for pigs and the obtained raw materials.

## 6. Conclusions

Summarizing, pork backfat meeting the arisen in the review traits as much as possible, will contribute, in addition to its main purpose – energy nutrition of the body, also to maintaining its normal functioning, reduction the risk of cardiovascular diseases development and other alimentary-dependent diseases.

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