Fatty acid composition and cholesterol content of the fat of pigs of various genotypes

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Abstract. The authors determined the fatty acid composition and the fat cholesterol content of the fat of Mangalica, Hungarian Large White \times Hungarian Landrace and Mangalica \times Duroc pigs. It was established that no significant difference among the three genotypes could be detected with respect to saturated, unsaturated or the essential fatty acids – not in regard to cholesterol content. The findings of these investigations indicate that in the three pig genotypes studied fat cholesterol content varies between 71 and 109 mg/100 g. Attention is also drawn to the high oleic acid content (relative percentage 43.57–44.81) and linoleic acid content (relative percentage 10.63–11.47) of pig fat.

1 Introduction

The fatty acid composition of the fat content of foodstuffs is of extremely great importance with respect to healthy human nutrition. A number of studies have

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reported on the substantial effect which different ratios of saturated and unsaturated fatty acids may exert on the health of those consuming them. While saturated fatty acids are considered a risk factor for cardiovascular diseases (*Burr et al.*, 1989; *Hrboticky & Weber*, 1993), polyunsaturated fatty acids are regarded as assisting in the prevention of disease (*Simopoulos*, 1991; *Weber et al.*, 1993; *Willett*, 1994). Since it was revealed that the fat contained by foodstuffs of animal origin is very rich in saturated fatty acids, the popularity of pig and cattle meat products for human consumption has recently suffered a decline, while that of poultry, fish and various sea foods – which contain high levels of unsaturated fatty acids – has increased.

The task of improving the fatty acid composition of foods of animal origin constitutes a great challenge both for livestock producers and for those involved in the production of foodstuffs. In the case of monogastric animals, such as the pig, there is a reasonably good possibility for the breeder to influence by varying the composition of the diet, the body composition of the pigs produced and the composition of foodstuffs derived from them (*Bee & Wenk*, 1994; *Klingenberg* et al., 1995; *Overland et al.*, 1996). Despite the fact that the fatty acid composition of the fat of the various regions of the body is relatively constant, when different diets are fed, significant differences are observed between the individual tissues in relation to the fatty acid composition of the diet. In addition, genotype-dependent differences have also been detected in pigs (*Nurnberg et al.*, 1994), in cattle (*May et al.*, 1994) and in poultry (*Reidy et al.*, 1994). According to the findings of *Sather et al.* (1996), there exists a relation of inverse proportionality between the degree of leanness and the hardness of the fat of Lacombe, Landrace and Yorkshire pigs.

In recent years, zoologists and livestock breeders worldwide have joined forces in the interest of saving indigenous and introduced domestic livestock breeds from extermination. The best strategy for preventing the disappearance of such breeds is to strive to maintain genetic diversity, for which it is precisely the indigenous breeds which can prove useful. In 1972, the recommendation made by the Stockholm Conference of the United Nations relating to the safeguarding of gene stocks and the establishment of a gene bank was accepted in Hungary. In 1993, the Rio Conference reaffirmed the importance of the role of gene banks, and drew attention to the fact that the risk of genetic drift is making its effect felt increasingly strongly in the course of genetic breed improvement.

The future of the Mangalica breed, indigenous to Hungary, is largely dependent on how its products can be utilized and how long-term market opportunities for these can be ensured. The Mangalica pig is now enjoying a renaissance in Hungary: this is due, on the one hand, to endeavours to return to the traditional breeds, and to the new market opportunities presented by the production of Serrano type ham processed by means of specialized Spanish technology, on the other hand. The ham of the Mangalica pig is extremely suitable for the processing of products of this kind, as due to its meat:fat ratio and the distribution of the fat between its muscle fibres the ham does not dry out even during the long-term maturing process. The meat of this breed is of outstanding quality; it has high dry matter content and its red colour corresponds to current requirements. Its palatable flavour is derived from the fat surrounding the muscle tissue.

It is a generally known fact that not only the quantity of fat consumed is of significance from the aspect of nutrition but also the ratio with respect to each other of the various fatty acids within the quantity of the fat consumed. The lipid theory attributes the now widespread incidence of arteriosclerosis, together with high blood pressure or cardiac infarction developing as a consequence of this, to the cholesterol content, on the one hand, and to the lower unsaturated fatty acid content of fats of animal origin, on the other hand. According to this theory, lower cholesterol intake and the consumption of higher quantities of unsaturated fatty acids can lead to a reduction of approximately 10% in the cholesterol level of the blood plasma. A positive relation has been ascertained between the cholesterol level of the blood and arteriosclerosis in the majority of the individuals examined. The cholesterol level of the blood plasma is now regarded as an indicator of arteriosclerosis.

Researchers report that a total of 20% of the daily cholesterol requirement is derived from food, the remaining 80% being produced by the organism. The reason for the proportion of cholesterol ingested with the food being so low is that only about half of the cholesterol consumed with food is absorbed, the remainder being excreted undigested from the body of the organism. A number of factors have been found to possibly influence changes in the cholesterol level of the blood plasma, apart from the cholesterol content of the foodstuffs consumed. High blood plasma cholesterol level may also develop by the effect of the intake of large quantities of saccharose. Other aspects which may be significant in changes in cholesterol level are various hereditary factors, lifestyle, level of food consumption and the state of health of the individual.

A number of authors maintain that the fatty acid composition of dietary fats is ideal where saturated (SAFA), monounsaturated (MUFA) and polyunsaturated fatty acids (PUFA) occur in equal proportions. Other authors consider such proportions of PUFA too high due to the risk of lipid peroxidation. They suggest that the proportion of PUFA in dietary fats should not exceed 10-15%, where MUFA content is between 30% and 35% and SAFA content between 50% and 60%. A composition of similar ratio has also been established for hypothetically ideal fatty acid composition (HIF).

A quantity of information has been published recently in connection with the fatty acid composition and cholesterol content of the back fat and other fat of the Mangalica pig. It has been claimed that the fat of the Mangalica pig is softer and more easily digestible than that of the modern pigs. Its softer, granular consistency is attributable to its different and also healthier fatty acid composition. Another view expressed is that the cholesterol content of the fat of the Mangalica pig is substantially lower than that of the fat of the new, intensive genotypes. At present, the validity of this view can be neither corroborated nor refuted, since, as far as the authors are aware, there are no precise relevant experimental data available. The investigations outlined in this paper were performed for the purpose of providing scientific substantiation or disproval of the above assertions; this study included the determination of the fatty acid composition and cholesterol content of the fat of Mangalica, Mangalica \times Duroc F₁ and Hungarian Large White \times Hungarian Landrace F₁ $(MNF \times ML)$ pigs. The MNF \times ML genotype is one of the most extensively used crosses in Hungary, and was therefore quite suitable to act as the control.

2 Materials and method

Experimental circumstances

These investigations were performed with the collaboration of the Hungapig Co. Ltd. and the Animal Breeding and Nutrition Research Institute in Herceghalom, at the new performance testing station. The experimental livestock were all housed in the same indoor area, with 6 pigs to a cage and 2.5 m² ground area per animal. Throughout the study, both the Mangalica pigs and those of the other genotype constructions were fed ad libitum diets of identical composition, provided from self-feeders. The composition of the diets used and their content are shown in *Table 1*. Diet I was fed in the live weight range of 30–70 kg and diet II when the weight of the pigs exceeded 70 kg.

At live weight between 120 and 130 kg, the pigs were slaughtered and their meat classified at the slaughterhouse of the Animal Breeding and Nutrition Research Institute in Herceghalom. After narcosis and slaughter, hanging to drain off the blood, boiling at 60-64 °C and manual singeing away of the hair, the carcasses were divided into parts. During the routine splitting and cutting into pieces of the carcasses, 100 g back fat samples were taken from the region

of the withers. These samples were stored in a freezer prior to laboratory analysis.

Component	Fattening diet I (%)	Fattening diet II (%)
Barley	15.00	15.00
Maize	59.72	57.00
Soybean meal, CP 46%	13.83	14.10
Full-fat soya (heat-treated)	5.00	-
Sunflower meal, CP 40%	-	3.53
Wheat bran	4.00	8.00
MCP (monocalcium phosphate)	0.29	0.20
Lime meal	0.05	0.06
Salt	0.11	0.11
Complete premix I for fattening pigs 2%	2.00	2.00

Table 1: Composition of fattening diets I and II

Examination of fatty acid composition and cholesterol content

Determination of fatty acid composition

A 1 g quantity of adequately homogenized back fat was measured into a 100 cm^3 Erlenmever flask, to which 8 cm^3 concentrated hydrochloric acid was added; the flask was then covered and heated on a steam bath for 60-90 minutes. After cooling, 7 cm^3 of ethanol and 25 cm^3 of ether were added and the flask was shaken for 1 minute. The ether phase was then poured off into a flask, and 25 cm^3 of petrol ether at the boiling point of 40-60 °C was added to the remainder of the sample; this was shaken for 1 minute, and, after separation, the petrol ether phase was poured into the ether phase, followed by homogenization. A quantity of the resultant extract known to contain 150-200 mg fat was then transferred to a round-bottomed flask with a ground glass neck. Subsequent to evaporation, the extract was boiled for 3 minutes with 4 cm^3 of a solution of boron trifluoride in methanol, and, after cooling, mixed with saturated aqueous saline solution. The organic phase was dried on sodium sulphate and then injected into the gas chromatograph. The following simplified procedure was used for the fat: 0.1-0.2 g fat was dissolved in 2 cm^3 of n-heptane, a small quantity of sodium sulphate was added, and 500 μ l was measured from the dehydrated organic phase into a vial; 500 μ l sodium methylate reagent was added to this, after which the liquid was heated at 60 °C for 1 hour, mixing by shaking it in every 10 minutes. Following cooling, 1 cm^3 of n-heptane and 1 cm^3 of water were added, and after shaking for

1-2 minutes, the fluid was injected from the upper organic phase into the gas chromatograph.

Conditions applied for gas chromatography:

Equipment:	Chrompack P 9000 gas chromatograph
Column:	$50 \text{ m} \times 0.25 \text{ mm}$ quartz capillary, humidifying phase
	CP Sil-88 (FAME)
Detector:	FID
Injector:	splitter
Gases:	carrier gas helium, 150 kPa, rate of flow $30 \text{ cm}^3/\text{min}$.
	at the detector: air 250 $\text{cm}^3/\text{min.}$, hydrogen 30 $\text{cm}^3/\text{min.}$
Temperatures:	injector 220 °C, detector 220 °C, column initially 100 °C,
	then increasing by $6 ^{\circ}C/min$. to $210 ^{\circ}C$, and subsequently
	isothermal until the process was completed.
Volume injected:	0.5-2 µl

Determination of the cholesterol

The pure fat contained by 5 g back fat was extracted in Soxhlet extraction equipment with n-hexane; the fatty extract was evaporated, and 10 cm³ of 60% potassium hydroxide and 40 cm³ of methanol were added to the residue. The flask was heated in a water bath with a reflux condenser for 30 minutes. After saponification had been completed, the flask was cooled, its contents were washed into a separating funnel with 3×40 cm³ of water, and the cholesterol was extracted with 3×40 cm³ of ether. The unified ether phase was evaporated, after which the residue was dissolved in 4 cm³ of hexane and 0.5 cm³ of methanol, and then injected into the gas chromatograph.

The conditions applied for the gas chromatography procedure were the following:

Equipment:	Chrompack CP 9000 gas chromatograph
Column:	$10 \text{ m} \times 0.25 \text{ mm}$ quartz capillary, humidifying phase
	CP Sil-5 CB
Carrier gas:	helium, pressure 30 kPa
Flow ratio:	50:1
Temperatures:	injector 275 °C, detector 300 °C, column 270 °C
Detector:	flame ionisation detector; hydrogen $30 \text{ cm}^3/\text{min.}$,
	air $300 \text{ cm}^3/\text{min.}$, nitrogen $20 \text{ cm}^3/\text{min.}$
Volume injected:	0.5-2 µl

Statistical evaluation of the results

The Student t-test was applied for the statistical evaluation of the experimental data. The analysis of the basic statistics and the correlation analyses were performed by means of the SPSS for Windows (1996) software package, version 7.5.

3 Results and discussion

Table 2 contains the fatty acid composition of the fat of the pigs of different genotypes in terms of relative mass percentages of the fatty acid methyl esters, while Table 3 shows the cholesterol content of the fat of the pigs of the various breeds.

	Genotype		
Fatty acid	Mangalica, $n = 5$	$MNF \times ML, n = 5$	$\begin{array}{l} \mathrm{Mangalica} \times \mathrm{Duroc}, \\ \mathfrak{n} = 5 \end{array}$
	Mean \pm SD	Mean \pm SD	Mean \pm SD
Capric acid	0.071 ± 0.0087	0.080 ± 0.011	0.082 ± 0.0103
Lauric acid	0.090 ± 0.0081	0.084 ± 0.010	0.086 ± 0.0068
Myristic acid	1.640 ± 0.12	1.458 ± 0.116	1.530 ± 0.083
Pentadecanoic acid	0.040 ± 0.0081	0.058 ± 0.012	0.038 ± 0.0062
Palmitic acid	25.97 ± 0.81	25.04 ± 1.01	26.15 ± 0.978
Palmitoleic acid	2.650 ± 0.47	2.270 ± 0.32	2.490 ± 0.424
Margaric acid	0.280 ± 0.034	0.450 ± 0.098	0.262 ± 0.034
Stearic acid	11.56 ± 1.01	13.63 ± 0.698	12.71 ± 1.633
Oleic acid	44.81 ± 1.71	44.34 ± 1.282	43.57 ± 2.155
Nonadecanoic acid	0.059 ± 0.012	0.074 ± 0.019	0.054 ± 0.0049
Linoleic acid	11.47 ± 1.92	10.63 ± 1.609	11.15 ± 0.724
Arachidic acid	0.170 ± 0.017	0.230 ± 0.022	0.200 ± 0.034
Eicosenoic acid	1.020 ± 0.208	0.750 ± 0.095	0.840 ± 0.139
Linolenic acid	0.570 ± 0.042	0.620 ± 0.081	0.630 ± 0.046
Eicosatrienoic acid	0.074 ± 0.0106	0.084 ± 0.022	0.068 ± 0.0091
Arachidonic acid	0.156 ± 0.027	0.196 ± 0.045	0.150 ± 0.021

Table 2: Fatty acid composition of the fat of the pigs of variousgenotypes (relative percentage of fatty acid methyl esters)

No significant difference (at P = 0.05 level) between the individual genotypes was detected either for unsaturated essential fatty acids or for unsaturated non-essential fatty acids, with the exception of eicosanoic acid. With respect to saturated fatty acids, with the exception of capric, lauric and palmitic acid, the difference between the genotypes proved significant at P = 0.05 level. Of these saturated fatty acids, in the case of stearic, margaric, pentadecanoic and nonadecanoic acid, the MNF × ML genotype contained the higher proportion, only myristic acid being determined in higher quantities in the Mangalica pig. This signifies that the ratio of saturated fatty acids in comparison with the unsaturated fatty acids was the highest in the MNF × ML pigs (41.12:58.88), although the difference was not significant (this ratio proving to be 39.87:60.13 for the Mangalica). The value for the Mangalica × Duroc genotype was found to be closer to that obtained for the MNF × ML group.

 Table 3: Cholesterol content of the fat of the pigs of various genotypes

Genotype	Cholesterol content $(mg/100 g)$
	mean \pm SD
Mangalica, $n = 5$	88.40 ± 10.08
Hungarian Large White \times Hungarian Landrace, $n = 5$	83.60 ± 11.77
$\mathrm{Mangalica}\times\mathrm{Duroc},n=5$	92.00 ± 8.72

The data was obtained from the Student-Newman-Keuls range test, designed for the purpose of distinguishing significantly different genotypes. It can be ascertained from the data that the control group differed non-significantly from the Mangalica pigs for every fatty acid under examination.

All of the three genotypes included in this study were found to greatly deviate from the hypothetically ideal ratio with respect to fatty acid composition (HIF). Ratios for saturated fatty acids were calculated at only approximately 40% instead of 53-62%, while those for unsaturated fatty acids proved to be around 60% rather than 38-47%. The values for oleic acid (43-44%) were substantially higher than those reported in the literature, while those for linoleic acid (10-11%) and those for linolenic acid (0.5-0.7%) were found to correspond to the literature data.

On the basis of these investigations, it may be established that no substantial difference was ascertained with respect to either the monounsaturated, or the polyunsaturated, or the saturated fatty acids (stearic acid being the exception among the fatty acids present in concentrations above 10%) on the examination of the fatty acid composition of the fat of these three pig genotypes. In the case of palmitic acid, oleic acid and linoleic acid, which together amount to more than 80% of fatty acid content, the mean values obtained practically concur. Thus, it is possible to draw the conclusion from these investigations that the fatty acid composition of the fat of the Mangalica pig is, practically speaking, totally identical in value to that of the fat of the Hungarian Large White × Hungarian Landrace and the Mangalica × Duroc genotype constructions. There are therefore no grounds for any assumption that the fat of the Mangalica breed has a more favourable fatty acid composition, which would render it more easily digestible and healthier for humans than that of the intensive breeds.

A similar conclusion can be drawn with regard to the cholesterol content of the fat of these genotypes. On the basis of the average for nine animals, the cholesterol content of the fat of the Mangalica was measured at 88.44 mg/100 g, that of the Hungarian Large White × Hungarian Landrace at 83.60 mg/100 g and that of the Mangalica × Duroc F_1 genotype at 92.00 mg/100 g. No significant difference at P < 0.05 level was detected between the three genotypes with respect to fat cholesterol content; variance within the genotypes proved greater than between genotypes. Thus, there is no truth in the reports indicating that the fat of the Mangalica pig contains less cholesterol than that of the more generally produced types of fattening pig.

However, on the basis of the findings of these investigations, the authors wish to draw attention to the observation that the fat of all three genotypes examined proved to contain 43-45% oleic acid and 10-12% linoleic acid, and it is thus extremely rich in unsaturated fatty acids and the essential linoleic acid if pigs are kept on a fattening diet based on one of the feed mixes currently in widespread use. The linolenic acid content (0.57-0.63%) and arachidonic acid content (0.15-0.20%) of the fat of the pigs examined proved low, while in comparison with the other fats studied, the stearic acid content was observed to be extremely low (11.56-13.63%).

The measurements made in this study indicate that the cholesterol content of pig fat varies between 71 and 109 mg/100g. This cholesterol content is substantially lower than that of the kidney, liver, egg yolk, bone marrow or cod liver oil.

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