

## COMPARISON OF DIFFERENT GENOTYPES OF CARP (*CYPRINUS CARPIO*) ON FATTY ACID COMPOSITION

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**Summary.** The aim of the study was to evaluate different genotypes of carp on fatty acid composition, influence of Ukraine and Hungary carps breed on Šilavotas carp fatty acid composition.

Šilavotas pure-bred carp, and their crosses with the Hungarian and Ukrainian carp was 6.5 months grown on identical area, volume and the bottom of the soil in relation to the ponds. Water temperature, oxygen saturation, and the pH was identical.

Studies have shown that for all of the total carp groups of polyunsaturated fatty acids (PUFA) content was 25.951 percent. This is a good indicator, whereas a similar PUFA percent fatty acids, is among the more valuable the marine species. Large and monounsaturated fatty acids (MUFA) the total amount - 47.775 percent. The total amount of saturated fatty acid (SFA) was relatively small, so the PUFA and SFA ratio was close to one. In experimental groups, the n-6/n-3 ratio were positive and it's range from 0.95 to 4.72.

Analysis of different genotypes of carp fatty acid composition, we determined that pure-bred carp had more PUFA, but among the most useful of polyunsaturated fatty acids- eicosapentaenoic (EPA) and docosahexaenoic (DHA) was less than combined with The Ukrainian and Hungarian experimental carps groups. Especially good positive effect had Šilavotas carp on EPA fatty acid in relation to Ukrainian carps. Compared with the pure – bred EPA fatty acid crossbred with Ukrainian carp was 1,171 % higher. The amount of DHA fatty acid, the highest concentration was determine in Hungarian carp, it increased 0,422 %. So, in concluded, it can be said that carps of Šilavotas crossing with Ukrainian and Hungarian carps had a positive effect on fatty acid composition.

**Keywords:** Breeds of carps, crossbred, aquaculture, fatty acid, polyunsaturated fatty acids, monounsaturated fatty acids, saturated fatty acid

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**Introduction.** In Lithuania carp is the main commercial fish growing in ponds. 2010 - 2011 m. carp accounted for about 93 percent of marketable fish production content and the about 87 per cent marketable fish production values (Masiulienė J. 2013; Vaikutis, 2009). In recent years, carp weight in the total fish production in ponds grown is in tend to decrease, but the species remains the most important aquaculture species.

The breeding-selection work in Lithuania is carried out in a single farm, Šilavotas section. Within 30 years, purposeful breeding-selection work, it bred Lithuanian carp breed – Šilavotas carp (Milerienė et al., 2011). These varieties of carp has good exterior - compact, intermittent, their breeders have good fertility characteristics, respond well to the pituitary gland injections, easy to spawn, larvae of a strong and viable. Šilavotas carp grow well and are productive. Šilavotas carp varieties derived from crosses Bubiai carp females with German carp males (B x V) (Milerienė E. 2011).

Since 1972 Šilavotas section for crossing, is used Ukraine (U) carp breed and in the later period, in order to enrich the gene pool, use Paros (P) and Hungary (H) carps.

Carp growing in Ukraine has already started in the XV century. Ukrainian varieties of carp has a high growth rate, good exterior, good compound feed conversion ratio. Economic properties of Ukrainian carp well transmit to their descendants and the crossing them with other varieties of carp (Tomilenko V.G. et al 2012; Privezencev J.A. et al, 2007).

Good results are obtained for crossing, and the use of Hungarian carp (H.M. Nielsen et al, 2010).

Fish are increasingly used in a healthy human diet, not only because of their protein with all 8 essential amino acids, but also because of their valuable human health fat. Their number depends on many factors - genetic characteristics of sex, age, feed. One of the main factors determining the fat content of the fish meat is their type. Some fish fillet fat content is low (less than 2 %) for example, zander (*Stizostedion lucioperca*), European perch (*Perca fluviatilis*) Atlantic cod (*Gadus morhua*). Other fish fillet fat content of more than 10 %, for example, Atlantic salmon (*Salmo salar*), European eel (*Anguilla anguilla*). Meanwhile, the carp for the fat content file occupies an intermediate position (Mráz, 2012). The references demonstrate the fatty acids in the difference between the various carp crossbred not much. It is therefore appropriate to examine whether there is a fatty acids differences between similar environments Šilavotas grown carp and their hybrids with other varieties of carp.

So, the aim of the study was to evaluate Šilavotas carp and their crossbred with Ukrainian and Hungarian carp on fatty acid composition (quantitative and qualitative).

### **Materials and methods**

The experimental was carried out in 2013, the Fisheries Service under Lithuanian Ministry of Agriculture and Aquaculture Inland Department Šilavotas section, LUHS VA in Animal productivity laboratory, KTU Food Institute.

Research carried out in accordance with the new edition of 01.01.2013 11.06.1997 Lithuanian Republic animal care, storage and use (Law of the Republic of Lithuania Animal Welfare and Protection Act) (Valstybės žinios 2012, no. 122-6126) and an executive act - the State Food and veterinary service Order on animals used for experimental and other scientific research, storage, maintenance and operation of approval "Valstybės žinios", 2009.01.22, no. 8-278). It is also in line with 2010 September 22nd European Parliament and Council Directive 2010/63 / EU and EC recommendations 2007/526 EC "Animal Handling and storage of experimental and other purposes." It was formed by three different genotype groups carp: Šilavotas carp, Šilavotas x Ukraine crossbred and Šilavotas X Hungarian carp hybrids. Hybrids were obtained using artificial fertilization. Caviar of Šilavotas female with Ukrainian and Hungarian carp male sperm. Reproducer has bred Šilavotas fish farm ponds. Fertilization was carried out farm incubator. Larvae and yearling carp bred in farm ponds. Annuals carp (avg. weight 104 g) were placed into three identical ground floor and benthic animals with respect to ponds. Carp were grown from 7 th of April until 10 th of October.

All three ponds water temperature varied depending on the climatic conditions (12 - 23 C), the average of May, June, July, August temperature was 18 C °.

The dissolved oxygen content depends on the water temperature, time of day, water plants and algae growing phase. On average to 6-8 mg / l. The water temperature and dissolved oxygen levels were measured oximeters Marvet Junior 2000.

The carps were fed with compound feed. Nutrition value of compound feed is presented in Table 1.

Table 1. Nutrition value of compound feed, %

Crude protein %	20,18
Crude fat %	4,78
Crude fibre %	4,59
Crude ash %	5,40
Calcium ( Ca ) %	0,75
Phosphorus ( P ) %	0,76
Available phosphorus	0,36
Natrium ( Na ) %	0,09
Magnesium ( Mg ) %	0,17
Chlorine ( Cl ) %	0,14
Salt %	0,19
Natrium chloride	0,69
Lysine %	1,05
Methionine %	0,33
Lysine methionine	0,3119
Methionine+cysteine %	0,66
Lysine : methionine + cystine	0,6637
Threonine %	0,77
Lysine threonine	0,7334
Tryptophan %	0,26
Lysine tryptophan	0,2436
Isoleucine %	0,71
Leucine %	1,37
Valine %	0,92

Carp were overfishing in 2013 October 9-10. Were calculated each of the total weight of carp, amount of carp. For further research, randomly selected 10 carp from each group.

The investigation of anatomical parts of the body samples were taken for further experimentals in LUHS VA Animal productivity laboratory. Before the study carps were euthanized, brain contusion fish hitting the skull, and the death was confirmed by knife, physically destroying the brain. Each carp further investigation was cut fillet.

Extraction of lipids for fatty acid analysis was performed with chloroform/methanol (2:1 v/v) as described by Folch et al. (1957). Fatty acid methyl esters (FAME) were prepared using the procedure of Christopherson and Glass (1969). The samples, according to current standards - ISO 5555: 1997 Animal and vegetable fats and oils. The sampling, prepared in accordance with ISO 661: 1997 Animal and vegetable fats and oils. Sample preparation was selected for the qualitative and quantitative analysis of fatty acids. The sample (100 ± 0,001g homogenized product) were covered with 25 ml of hexane and shaken for 5 min., And then left for 30 minutes, again vigorously. Fat was separated hexane blow away to the tube (4 ml). Fatty acids methylation to methyl esters was used in 2 mol / l KOH anhydrous methanol solution of 200 ml was placed in tubes containing extracts. Samples were shaken for 1 min., With (Vortex) mixer for 30 minutes. The top layer of the prepared sample were taken of 2 ml, of which the chromatography was used 2 ml.

Chromatographic analysis by gas chromatograph Shimadzu GC -2010 Plus.

Statistical characteristics of the sample (arithmetic mean - M, standard error – SE, minimum – mi, maximum - max) were calculated using statistical software SPSS (version 15, SPSS Inc., Chicago, IL).

### Results and discussions

The differences between the fish fat content is very high. It depends on the species and other factors. For example, some species of fish fillet is only less than 2 % of fat. For example zander (*Stizostedion liucoperrca*), European perch (*Perca fluviatilis*, Atlantic cod (*Gadus morhua*). Meanwhile, other fish, such as Atlantic salmon (*Salmo salar*), European eel (*Anguilla anguilla*) has more than 10 % of fat. But saltwater fish contain higher levels of polyunsaturated fatty acids than freshwater fish. (Henderson et al., 1987; Connan M. et al., 2010).

Influence of fat content and composition is depend on many factors, but the most important is - the type of the fish, diet (feed composition), bioactive compounds, genetics (genetic background), sex, age, fish body tissue type (white muscles, red muscle, adipose and skin) (Mraz, 2011; Fauconneau 1995).

Between carp species, fat content is also distributed unevenly. In Serbia polyculture ponds, carp meat fat content of 6.85 %, White hypophthalmichthys - 4.07 %., Amur - 6.39 % (Cirkovic M. et al., 2011).

Fatty acid composition of fat carp depends on a variety of carp, pond type. Naturally feeding carp fat have more linolenic acid and eicosapentaenoic (EPA) and dokozaeksaen (DHA). Meanwhile, the compound feed fed carp fatty acids contains more arachidic and oleic acids (Faimonova, 2003).

For the human body is very important unsaturated fatty acids, biologically they are more valuable than the saturated fatty acids. Polyunsaturated fatty acids accumulate in the brain and reproductive cells. Omega - 3 (alpha-linolenic) and omega - 6 (linolenic) acids is necessary for many metabolic processes, is a source of energy component to all cell membranes, they inhibit inflammation, regulates coagulation and blood pressure, strengthen the immune system (Dagilytė, 2006).

Polyunsaturated fatty acids (PUFA) content of the species is variety. Yellowfin bream (*Acanthopagrus hasta*) fat of these acids is 29.1 % sea bass (*Dicentrarchus labrax*) - 36,1%. Rainbow trout (*Onchorhynchus mykiss*) - 28.4 %, Salmon (*Salmo salar*) - 20 %, Halibut (*Scaophthalmus maximus*) - 47.3 %, Perch (*Perca fluviatilis*) - 35, 0 %, Sturgeon (*Acipenser oxyrinchus desotoi*) - 25,0 % pike (*Esox lucius*) - even 58.3 % (Hossain, 2011). Polyunsaturated fatty acids are quantitatively different and carp fish species belonging to the family of fat - Asp are 24,60 % , Bream-17.07 %, Common barbel - 26,31 %. Carp - 19.7 %. (Ljubojevic et al 2013). It is also very different concentration of PUFA, when carps are growing by natural conditions and feeding farmed. Aquaculture reared rainbow trout fat is 28.4 % of PUFA and the wild - 40.5 %, Freshwater perch among these data is 35.0 and 43.0 %, Pike 58, 3 and 31.3 % (Hossain, 2011).

When analysed data of mono-unsaturated fatty acids, these acids enhance cell membrane permeability, and easily penetrates into cells hormones and other biologically active compounds. The use of MUFA reduces the risk of developing cardiovascular disease (Schwingshackl et al, 2011). Among freshwater fish have the most MUFA is in bream (56.9 %) and carp (52,94 %), Asp, Common barbel and sturgeon has about 45-47 %. MUFA, pike - only 31,66 % (Ljubojevic et al. 2013).

Our research showed that fatty acid composition (Table 2), separately for each group (Table 3) and compared to Šilavotas and their crossbred with the Hungarian and Ukrainian carp fat composition (Table 4).

Table 2. Average of fatty acid concentration in carp meat

Variable	n	M	SE	Min	Max
SFA	30	24.842	0.237	22.54	27.43
MUFA	30	47.775	0.598	42.27	55.75
PUFA	30	25.951	0.485	19.84	31.51
Neident.	30	1.4327	0.0573	1	2.26
PUFA/SFA	30	1.0463	0.0204	0.83	1.29
n3suma	30	7.843	0.482	3.69	16.18
N6suma	30	18.109	0.558	12.07	23.91
n6/n3	30	2.604	0.186	0.95	4.72

From data in Table 2, we can see that the most useful for human of polyunsaturated fatty acids in carp meat was high - 25.951 % (varried from 19,84 to 31.51 %). This is a very good indicator, since it is the polyunsaturated fatty acids are useful for the human body. Some scientifics shown that various PUFA percentage of carp meat is from 12.05 % (Faimonova E. et al, 2003) to 32.58 % (Stanchev M. et al 2014).

When analysed data of MUFA concentration of 30 carps, we determined that it ranged from 42.27 % to 55.75 % (Average - 47.775 %).The concentration of MUFA, PUFA depend not only on the above written factors but also on the season. The highest amount of MUFA and PUFA carp meat were in the end of feeding period (October). While the SFA amount of in that period were the minimum (Trbović 2013). Our study was also carried out in a middle of October, at the end of a period of carp nutrition.

When analysed saturated fatty acids in carp fat were relatively small, so the PUFA and SFA ratio was close to one. A similar (Mraz, 2012), or extent lower PUFA / SFA (0.71) (Stanchev 2014).

It is an important indicator of n-6/n-3 polyunsaturated fatty acids, since of the human body both acid metabolism must be activated by the same enzymes. This creates competition: omega-6 acids in excess of hinder the omega-3 fatty acids metabolic of exchange and prevents their appropriation. The World Health Organization recommended omega 6 to omega 3 ratio is 5: 1 (Dagilytė, 2006). Our findings show a very good n-6/n-3 ratio (from 0.95 to 4.72), as even and dietary kept in a turkey this ratio ranges from 5.10 to 6.68 (Juodka, 2010).

Table 3. Fatty acid concentration depends from genotype

Variable	Breed	n	M	SE	Min	Max
SFA	SxU	10	24.092	0.396	22.54	26.08
	SxV	10	24.449	0.219	23.18	25.21
	Silavotas	10	25.985	0.335	23.92	27.43
MUFA	SxU	10	48.95	1.22	42.27	55.75
	SxV	10	47.773	0.849	42.59	51.79
	Silavotas	10	46.603	0.976	43.12	52.16
PUFA	SxU	10	25.402	0.95	19.84	31.51
	SxV	10	26.389	0.706	22.55	30.59
	Silavotas	10	26.062	0.905	21.1	30.67
Neident.	SxU	10	1.561	0.124	1.07	2.26
	SxV	10	1.387	0.094	1	1.85
	Silavotas	10	1.35	0.0691	1.01	1.72
Sumatran	SxU	10	1.234	0.119	0.876	1.893
	SxV	10	0.9674	0.0343	0.806	1.101
	Silavotas	10	0.8852	0.0283	0.754	1.098
PUFA/SFA	SxU	10	1.054	0.0337	0.88	1.29
	SxV	10	1.081	0.0295	0.92	1.23
	Silavotas	10	1.004	0.0408	0.83	1.28
n3suma	SxU	10	10.727	0.745	7.76	16.18
	SxV	10	6.54	0.331	5.39	8.5
	Silavotas	10	6.263	0.438	3.69	8.3
n6suma	SxU	10	14.676	0.392	12.07	16.67
	SxV	10	19.852	0.457	17.17	22.09
	Silavotas	10	19.798	0.824	16.01	23.91
n6/n3	SxU	10	1.413	0.795	0.95	1.78
	SxV	10	3.082	0.12	2.55	3.64
	Silavotas	10	3.316	0.27	1.93	4.72

Analysis of results by genotypes shows that Šilavotas breeds carp had more PUFA than Šilavotas Ukraine and hybrids, but the most useful fatty acids eicosapentaenoic (EPA) and docosahexaenoic (DHA) it contains the least. EPA (20: 5 (n-3) Šilavotas carp fat were on average 1.385 % (from 0.67 to 2.08 %) SxV - by 1.413 % (from 1.06 to 1.89) and this has been SxU best – 2.556 % (from 1.63 to 4.36). DHA (22: 6 n-3) % among the fatty acids comprised: Šilavotas 1.733 % (from 1.07 to 2.47) SXV - 2.155 % (1.53 to 3.26) and SxU - 1.939 % (from 1.26 to 2.73 %). From the data we can see that Šilavotas carp improved of DHA and EPA and the direction of increasing the Ukrainian and Hungarian breeds carp use appropriate as omega - 3 fatty acids reduce heart disease, the risk of thrombus formation, acts anti-inflammatory. Omega - 3 fatty acids are also used in case of high blood pressure and rheumatoid arthritis. On average, 1 g of EPA and DHA per day are recommended for the prevention of heart disease. 2 - 4 for g per day, helps to reduce triglyceride levels (Covington, 2004, Griffin et al., 2006).

The highest concentration of MUFA were in SxU carp meat (48.95 % , min. 42.27 max. 55.75), the lowest - in breed Šilavotas carp meat (46.603 % . min. 43.12 max. 52.16).

The highest concentration of SFA was in Šilavotas carp (25.985 % , min. 23.92., max. 27.43), the lowest SxU carp (24.092 % , min. 22.54, max. 26.08) meat.

All of these indicators suggest that carp hybridization with Ukraine and with the Hungarian carp fatty acids in terms of quality produce positive results.

The same conclusion can be drawn, and the examination of PUFA / SFA ratio of n-3 and n-6 fatty acids and the amount of n6 / n3 ratio, which is particularly good SxU – 1.413 (min. 0.95 max. 1.78), the while the SxV and bred carp among Šilavotas it exceeded 3.

Table 4. In comparisons purebred Šilavotas carp and their hybrids with Ukrainian and Hungarian carp fatty acid composition

Variable	Genotype	n	M	SE	Min	Max
SFA	Crossbreed	20	24.271	0.224	22.54	26.08
	Silavotas	10	25.985	0.335	23.92	27.43
MUFA	Crossbreed	20	48.361	0.735	42.27	55.75
	Silavotas	10	46.603	0.976	43.13	52.61
PUFA	Crossbreed	20	25.896	0.587	19.84	31.51
	Silavotas	10	26.062	0.905	21.1	30.67
Neident.	Crossbreed	20	1.474	0.0782	1	2.26
	Silavotas	10	1.35	0.0691	1.01	1.72
Sumatran	Crossbreed	20	1.1008	0.0676	0.806	1.893
	Silavotas	10	0.8852	0.0283	0.754	1.098
PUFA/SFA	Crossbreed	20	1.0675	0.022	0.88	1.29
	Silavotas	10	1.4	0.0408	0.83	1.28
n3suma	Crossbreed	20	8.634	0.623	5.39	16.18
	Silavotas	10	6.263	0.438	3.69	8.3
n6suma	Crossbreed	20	17.264	0.662	12.07	22.09
	Silavotas	10	19.798	0.824	16.01	23.91
n6/n3	Crossbreed	20	2.248	0.204	0.95	3.64
	Silavotas	10	3.316	0.27	1.93	4.72

Table 4 shows that although, purebred Šilavotas carp meat (n = 10) total quantity of PUFA was different very low (0.166 %) higher than ŠxU and SxV crossbred group (n = 20), but PUFA qualitative composition was higher in crossbred group, also demonstrated higher EPA and DHA values were higher and fatty acid of linolenic (C18: 3 n-3) content. Crossbred group, the median of the fatty acid value was 3.685 % while bred carp 2.512 % higher alpha-linolenic acid content is very important, because this acid helps maintain normal cholesterol levels in the blood (Nacionalinis maisto ir veterinarijos rizikos vertinimo institutas, 2013). Šilavotas bred carp content of PUFA was slightly higher for others, not so valuable, polyunsaturated fatty acids. Linoleic acid (C18: 2n-6) in Šilavotas bred carp fat composition consisted of 15,47 %. (min. 12.94 max. 18,97) and crossbred of carp meat was 12.51 %. (min. 9.16 max 15.72). Total of n-3 fatty acids as well was higher in the crossbred - 8.634 %, when pure Šilavotas carp meat fat were 2.371 % less. Therefore, n6/n3 ratio was better among the crossbred – 2.248 (pure - 3.316).

### Conclusions

Concentration of fatty acid in carp meat were: polyunsaturated fatty acid - 25.95 %, monounsaturated fatty acid – 47.77 %, omega 6/omega 3 varied from 0.95 to 4.72 point. The most PUFA and SFA was determined purebred Šilavotas carp meat, but the optimal n-6 /n-3 ratio - 1.41: 1.00 was obtained from the crossing Šilavotas and Ukraine carp varieties. So in conclusions, pure-bred carp hybridization with the Ukrainian and Hungarian carp on fatty acids concentration had a produce positive results.

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