

Chemical Composition of Fillets of Mirror Crossbreeds Common Carp (*Cyprinus carpio* L.)

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Abstract

This paper presents the results of a study dealing with chemical composition of fillets and fatty acid composition (saturated fatty acid: SFA, monounsaturated fatty acid: MUFA, polyunsaturated fatty acid: PUFA) of lipids. Three groups of the mirror hybrid carp in age of three years: M2 × L15 – the Hungarian Szarvas mirror carp (M2) and the Hungarian hybrid mirror strain (L15), M2 × DOR 70 (the Israeli breed - DOR70), M2 × M72 (Northern mirror carp - M72) were compared with: the pure breed M2 and scaly hybrid ROP × TAT – the Ropsha (ROP) and the Tata (TAT) carp. ROP × TAT hybrid fillets contained (in g·kg⁻¹) more ($P < 0.01$) dry matter (283.1 ± 23.87) and lipids (99.3 ± 30.60). Fat in all of the monitored carp groups was made up of more than 50% of MUFA (from 51 to 64%), 25 - 29% of SFA and 10 - 22% of PUFA. Fillets of mirror hybrids M2 × DOR70, M2 × M72 and breed M2 contained less lipids ($P < 0.01$), less MUFA_{sum} ($P < 0.01$), particularly less oleic acid (C18:1_{n-7}), and more PUFA_{n-3} ($P < 0.01$), more eicosapentaenoic acid (C20:5_{n-3}) and docosahexaenoic acid (C22:6_{n-3}). The differences in fatty acid profile can be related to the different genetic effects of different groups of common carp.

Fish meat, lipid quality, chemical indicator, fatty acid

Fish in general and ocean fish including shellfish in particular are considered an important source of essential n-3 polyunsaturated fatty acids (PUFA_{n-3}), i.e. α -linolenic acid, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) (Ackman 2000). High concentrations of PUFA_{n-3} in foodstuffs are favourable for human health, which has been demonstrated in clinical experiments (Simopoulos 1997). The long-chain PUFA_{n-3} have antiatherosclerotic effects and also beneficial effects on several other diseases (Steffens and Wirth 2007).

N-3 fatty acids are also present in lipids of freshwater fish (Ackman 2002). However, quantity of these acids varies largely in dependence on the fish species (herbivorous, omnivorous or carnivorous), if they are wild fish or farm-raised, on the age of fish and on origin of diets (natural food or cereal supplement) and its composition (rich primarily in PUFA_{n-3} or saccharides) (Steffens 1997). Fresh-water carp may be as nutritionally valuable as ocean fishes (Steffens 1977). Carps reared in ponds on natural food had high content of n-6 as well as n-3 fatty acids in their muscle triacylglycerols, carps fed with a supplementary wheat diet had lower levels of those essential fatty acids (Steffens and Wirth 2007). Donmez (2009) found total EPA and DHA fatty acids concentrations at the level of $14.96 \text{ g} \cdot 100^{-1} \text{ g fat}$ in wild carps. On the other hand, Ackman (2002) reported EPA and DHA concentrations in farmed carp as low as $0.35 \text{ g} \cdot 100^{-1} \text{ fat}$. The fatty acid composition in the muscle of carps can be significantly influenced by the feeding period and the season of the year (Guler et al. 2008). According to Mareš et al. (2009), the presence of cyanobacterial water bloom had a significant effect on the content of individual fatty acids analysed in muscles of the farmed carps. Significant ($P < 0.05$) differences were found in the ratio of PUFA_{n-3/n-6}. In the case of common carp, these differences were based

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on changes in the content of PUFA_{n-3} and _{n-6}, in case of the silver carp only in the content of PUFA_{n-6} that increased during the exposure of fish to the cyanobacterial water bloom. In addition, environmental factors, especially water temperature, influenced the fatty acid composition of plankton (Guo et al. 2008).

The scaly hybrid is commonly farmed for commercial purposes and served as a control group (e.g. Linhart et al. 2002; Kocour et al. 2005a) for comparison of growth and survival of the mirror groups.

The aim of this study was to evaluate the fatty acid profile in the intramuscular fat of different carp groups and to find out whether there is a difference between the sexes.

Materials and Methods

Starting material and processing

The performance test of mirror common carp groups started in 2003 at the fishfarming company Rybníkářství Tábor, a. s., when fish in the K₀ stage were stocked in ponds. The test was finished at the end of the 2005 in vegetation period, in which the fish reached the harvest size. The fish were tested in ponds and, to guarantee the objectivity of results of performance, growth and survival, an internal control group of carp scaly hybrid was used (Linhart et al. 2002; Kocour et al. 2003; 2005ab). For the experiment, 15 females of Hungarian Szarvas mirror carp (M2) were used in the maternal position. The females were crossed with 25 males from each out of 4 various breeds (top-crossing) using artificial reproduction method described by Kocour et al. (2005a). Paternal breeds were the Hungarian Szarvas mirror carp (M2) for pure breed production, Hungarian Szarvas two-line hybrid strain (L15), the Israeli breed (DOR70) and the Northern mirror carp (M72). The scaly hybrid of the Ropsha (ROP) and the Tata (TAT) carp was used as a control. The fish were reared under standard pond management conditions in the Czech Republic. For their growth, fish had their natural diet available in the ponds (plankton, benthos) over 3 vegetation seasons. Fish usually do not feed in the winter (November - February) period. In the first vegetation season, the fish were fed supplementary feed mix KP1 3 × a week starting when the fish was 2 months old. In the second vegetation period, the supplementary feed mix was replaced with uncrushed wheat, which was also fed to the fish in the third vegetation period. The supplementary feed was fed 3 × weekly throughout the experiment. During the experiment (before and after each vegetation period), data on growth (weight) and survival of fish were recorded.

The final evaluation of the experiment was made at the end of the 2005 vegetation period in three-year-old fish (K₃). From the pond with the highest mean fish weight, 40 carp from each of the 5 groups (i.e. the ROP × TAT control, M2 pure breed, experimental hybrid M2 × L15, M2 × DOR70, M2 × M72) were randomly chosen for chemical examination (a total of 200 fish). Fish carcasses were evaluated according to Gela and Linhart (2000) at University of South Bohemia in České Budějovice and Research Institute of Fish Culture and Hydrobiology in Vodňany, Czech Republic.

Chemical analysis

Four indicators of basic chemical composition (in g·kg⁻¹) were determined in fish meat: content of dry matter (DM), crude protein (CP), fat (F) and ash (A). The DM content was determined gravimetrically following the reference method (International Standard 1997) for determination of moisture content in flesh by drying the sample with sand up to constant weight at +103 ± 2 °C. The CP content was determined as the amount of organically bound nitrogen (recalculating coefficient f₁ = 6.25) using analyzer Kjeltec 2300 (FOSS Analytical AB, Sweden, Höganäs) following the procedure recommended by the producer (Application note 2003). The content of F was determined quantitatively by extraction in diluents using Soxtec (Tecator, Sweden) using the method following recommendation of the producer (Application note 1990). The content of A was determined gravimetrically by burning the weighed sample in muffle oven (Elektro LM 212.11, Germany) at 550 °C until black carbon particles disappeared (International Standard 1998).

To study fatty acid composition in fish meat, 5 male carp and 5 female carp fillets were randomly chosen from each of the groups (a total of 50 fillets). An aliquot (50 g) of each sample (middle part of the fillet) was used for fat determinations. The determination of fatty acid composition was performed by gas chromatography using HP 4890 (Hewlett-Packard, USA) apparatus with flame ionization detector (FID) and capillary column Omega Wax TM250 (30 m × 0.25 mm × 0.25 μm) following extraction with a mixture of methanol and chloroform (Folsch et al. 1957). The optimum temperature gradient was 140 °C to 240 °C (5 °C/min). The injector temperature was 280 °C, the FID temperature was 300 °C. Nitrogen was used as the carrier gas.

Basic statistical values (means, S.D.) of the variables measured were processed in Excel 97. Statistical significance was evaluated using the multifactorial analysis of variance ANOVA Statistica 7.0 (StatSoft CR, s.r.o., Praha, Czech Republic).

Results and Discussion

The traditional approach to the rearing of common carp (*Cyprinus carpio* L.) in the Czech Republic is based on foods naturally occurring in ponds (zooplankton, benthos). The energy-producing component of their diet is supplemented with untreated cereals

Table 1. Chemical composition of fillets of control sealy group (ROP × TAT) and experimental mirror groups (M2, M2 × DOR70, M2 × L15, M2 × M72) of the common carp (*Cyprinus carpio* L.). Values with the common superscript within the row do not differ significantly at the given probability level

Indicators in g·kg ⁻¹	Control group	Experimental groups				P
	ROP × TAT n = 41 Mean ± S.D.	M2 n = 40 Mean ± S.D.	M2 × DOR70 n = 40 Mean ± S.D.	M2 × L15 n = 40 Mean ± S.D.	M2 × M72 n = 39 Mean ± S.D.	
FLW*	2030 ± 176.0 ^a	1699 ± 167.4 ^b	1790 ± 150.9 ^b	1709 ± 197.2 ^b	1772 ± 221.3 ^b	< 0.05
Dry matter	283.1 ± 23.87 ^a	220.6 ± 9.79 ^b	227.1 ± 13.51 ^b	226.1 ± 13.74 ^b	236.7 ± 12.43 ^{ab}	< 0.01
Crude protein	174.1 ± 8.53 ^a	175.7 ± 4.19 ^a	178.2 ± 8.63 ^a	177.9 ± 8.09 ^a	176.2 ± 5.88 ^a	< 0.05
Fat	99.3 ± 30.60 ^a	24.3 ± 12.01 ^b	35.7 ± 19.98 ^b	30.6 ± 14.95 ^b	35.3 ± 15.07 ^b	< 0.01
Ash	10.7 ± 0.75 ^b	11.3 ± 0.59 ^a	11.1 ± 0.56 ^{ab}	11.0 ± 0.61 ^{ab}	11.2 ± 0.71 ^a	< 0.05

* FLW - fish live weight in g (Buchtová et al. 2009)

(wheat). The preference for a feed rich in saccharides leads to an increase in the percentage of the oleic acid (C18:1_{n-9}) in body lipids of the fish, which is produced in the organism by desaturation and elongation of saturated fatty acids (SFA). At the same time, there is a decrease in the percentage of PUFA_{n-3} (Fajmonová et al. 2003; Buchtová et al. 2007).

Regarding basic chemical composition of muscle tissue, we did not expect to find any major differences in percentages of the nutrients monitored between groups of carp tested in our experiment because the fish came from the same environment, were of the same species, age, and had the same access to the same food and supplements. Chemical composition of sexually mature healthy fish is more markedly affected by the ongoing reproductive cycle (Lachowicz and Kołakowski 2001). Quality of edible parts is also affected in fast-growing fish because the change in growth capability is associated with a greater food intake and increased nutrient conversion (Steffens 1974).

We found differences in fillet chemical composition between the ROP × TAT control hybrid and the mirror experimental carp groups. ROP × TAT hybrid fillets (the highest final weight 2030 ± 176.00 g, $P < 0.05$) contained more ($P < 0.01$) dry matter (283.1 ± 23.87 g·kg⁻¹) and lipids (99.3 ± 30.60 g·kg⁻¹) and non-significantly less nitrogenous substances (Table 1). Similar results were reported for the rainbow trout by Corraze et al. (1993). Higher deposition of lipids in meat of ROP × TAT hybrids and better growing are probably consequential to their preference for high-energy cereals in feed and/or to a genetically-based difference in metabolism. Chemical compositions of fillets from mirror carp hybrids (M2 × DOR70, M2 × L15, M2 × M72) and from the pure line carp (M2) were practically identical. All mirror groups showed comparable weight at harvest.

Lipids in all groups of carp examined consisted of more than 50% of MUFA_{sum} (from 51 to 64%). SFA_{sum} and PUFA_{sum} made up 25 to 29% and 10 to 22%, respectively. The oleic acid (C18:1_{n-9}) followed (in descending order) by palmitic acid (C16:0), palmitoleic acid (C16:1), stearic acid (C18:0), linoleic acid (C18:2_{n-6}), eicosenic acid (C20:1_{n-9}), α-linolenic acid (C18:3_{n-3}) and myristic acid (C14:0) were the most abundant among lipids. Concentrations of the remaining fatty acids were below 1% (Table 2). The reason for unfavourable composition of fatty acid spectrum in lipids of all carp groups can be accounted to the type of food dominating in the diet. Steffens and Wirth (2007) reported that different methods of rearing and feeding cause substantial variations in the fatty acid composition of pond fish.

Table 2. Fatty acid composition (in % of total determined fatty acids), contents of saturated fatty acids (SFA_{sum}), monounsaturated fatty acids (MUFA_{sum}) and polyunsaturated fatty acids (PUFA_{sum}) and PUFA_{n-6/n-3} ratio in intramuscular lipids of control sealy group (ROP × TAT) and experimental mirror groups (M2, M2 × DOR70, M2 × L15, M2 × M72) of the common carp (*Cyprinus carpio* L.). Values with the common superscript within the row do not differ significantly at the given probability level

Fatty acid	Control group		Experimental groups				P
	ROP × TAT n = 10 Mean ± S.D.	M2 n = 10 Mean ± S.D.	M2 × DOR70 n = 10 Mean ± S.D.	M2 × L15 n = 10 Mean ± S.D.	M2 × M72 n = 39 Mean ± S.D.		
C14:0	1.04 ± 0.05 ^a	0.83 ± 0.12 ^b	0.83 ± 0.14 ^b	1.02 ± 0.10 ^a	0.90 ± 0.13 ^{ab}	< 0.01	
C16:0	18.14 ± 0.94 ^b	20.53 ± 1.01 ^a	20.37 ± 1.89 ^a	20.55 ± 2.66 ^a	20.24 ± 1.49 ^{ab}	< 0.01	
C18:0	6.77 ± 0.50 ^a	7.22 ± 0.66 ^a	7.08 ± 0.77 ^a	7.30 ± 0.64 ^a	7.14 ± 0.45 ^a	> 0.05	
SFA _{sum}	25.95 ± 0.50^b	28.58 ± 0.60^{ab}	28.28 ± 0.93^{ab}	28.87 ± 1.13^a	28.27 ± 0.69^{ab}	< 0.05	
C16:1	9.43 ± 0.58 ^a	9.03 ± 0.92 ^a	8.47 ± 1.33 ^a	9.54 ± 0.63 ^a	8.70 ± 0.73 ^a	> 0.05	
C18:1	51.70 ± 1.52 ^a	40.11 ± 4.79 ^b	39.59 ± 5.86 ^b	48.70 ± 3.68 ^a	42.48 ± 4.10 ^{ab}	< 0.01	
C20:1	2.48 ± 0.26 ^a	1.93 ± 0.21 ^b	1.81 ± 0.15 ^b	2.27 ± 0.44 ^{ab}	2.22 ± 0.36 ^{ab}	< 0.01	
MUFA _{sum}	63.61 ± 0.79^a	51.08 ± 1.97^b	49.87 ± 2.45^b	60.50 ± 1.58^a	53.41 ± 1.73^b	< 0.01	
C18:3n3	1.79 ± 0.34 ^a	1.61 ± 0.32 ^a	1.77 ± 0.29 ^a	1.45 ± 0.33 ^a	1.55 ± 0.20 ^a	> 0.05	
C18:4n3	0.23 ± 0.07 ^a	0.17 ± 0.06 ^a	0.21 ± 0.05 ^a	0.18 ± 0.06 ^a	0.19 ± 0.08 ^a	> 0.05	
C20:5n3	0.90 ± 0.19 ^b	3.13 ± 0.95 ^a	3.57 ± 1.30 ^a	1.00 ± 0.34 ^b	2.62 ± 1.15 ^a	< 0.01	
C22:5n3	0.27 ± 0.09 ^b	1.37 ± 0.48 ^a	1.35 ± 0.45 ^a	0.44 ± 0.19 ^b	1.07 ± 0.49 ^a	< 0.01	
C22:6n3	0.66 ± 0.31 ^b	3.99 ± 1.67 ^a	4.84 ± 1.90 ^a	1.05 ± 0.50 ^b	3.43 ± 1.66 ^a	< 0.01	
PUFA _{n3} _{sum}	3.85 ± 0.20^b	10.26 ± 0.70^a	11.74 ± 0.80^b	4.11 ± 0.28^b	8.86 ± 0.71^a	< 0.01	
C18:2n6	5.49 ± 0.36 ^{ab}	5.96 ± 0.49 ^a	5.55 ± 0.34 ^{ab}	5.15 ± 0.69 ^b	5.98 ± 0.79 ^a	< 0.05	
C18:3n6	0.13 ± 0.02 ^a	0.13 ± 0.06 ^a	0.15 ± 0.03 ^a	0.11 ± 0.04 ^a	0.16 ± 0.04 ^a	> 0.05	
C20:4n6	0.83 ± 0.30 ^b	3.33 ± 1.31 ^a	3.70 ± 1.16 ^a	1.06 ± 0.44 ^b	2.82 ± 1.23 ^a	< 0.01	
C22:4n6	0.07 ± 0.03 ^b	0.36 ± 0.16 ^a	0.36 ± 0.12 ^a	0.09 ± 0.05 ^b	0.25 ± 0.15 ^a	< 0.01	
C22:5n6	0.07 ± 0.04 ^b	0.31 ± 0.14 ^a	0.37 ± 0.11 ^a	0.10 ± 0.06 ^b	0.26 ± 0.11 ^a	< 0.01	
PUFA _{n6} _{sum}	6.60 ± 0.15^b	10.09 ± 0.43^a	10.12 ± 0.35^a	6.52 ± 0.26^b	9.46 ± 0.46^a	< 0.01	
PUFA _{sum}	10.45 ± 1.65^b	20.35 ± 1.99^a	21.86 ± 2.06^a	10.63 ± 1.52^b	18.32 ± 1.90^a	< 0.01	
Ratio n-6/n-3	1.76 ± 0.26^a	1.02 ± 0.17^b	0.90 ± 0.15^b	1.62 ± 0.21^a	1.13 ± 0.25^b	< 0.01	

* FLW - fish live weight in g (Buchtová et al. 2009)

Marketable carp reared on the basis of natural food in ponds exhibit high contents of $n-6$ as well as $n-3$ fatty acids, on the other hand carp fed supplementary wheat, which is characterized by a low content of PUFA $_{n-3}$ (Steffens et al. 1998), resulted in somewhat lower concentrations of these acids and higher oleic acid content.

The cereal-based supplement offered was probably sought after more readily ($P < 0.05$) by the faster growing scaly hybrid ROP \times TAT. Hence, lipids of this hybrid contained the highest concentrations ($51.70 \pm 1.52\%$) of the oleic acid (C18:1 $_{n-9}$). With respect to lipid quality, experimental mirror carp M2 \times DOR70, pure line M2 carp and mirror carp M2 \times M72 demonstrated a more desirable fatty acid composition than carp from the control ROP \times TAT group. Although their fillets contained less ($P < 0.01$) lipids, they also contained less ($P < 0.01$) MUFA $_{\text{sum}}$, less oleic acid (C18:1 $_{n-9}$), and more ($P < 0.01$) PUFA $_{n-3}$, more eicosapentaenoic acid (C20:5 $_{n-3}$) and docosahexaenoic acid (C22:6 $_{n-3}$), while percentages of the essential α -linolenic acid (C18:3 $_{n-3}$) in lipids of all carp groups were identical. Derivates of these PUFA $_{n-3}$, so-called eicosanoids (especially prostacyclins), play a positive role in the prevention of cardiovascular diseases (Steffens 1997). In contrast to ROP \times TAT hybrids, the slowly growing mirror carp groups probably ingested in higher rate natural pond foods containing more EPA and DHA. Concentrations of these fatty acids are high especially in zooplankton and chironomid larvae (Steffens et al. 1998). According to Guo et al. (2008), fatty acid composition of plankton varies with seasonal changes. The content of PUFA $_{n-3}$ (especially DHA) is high when cryptophytes and DHA-rich copepods become an important group of plankton. However, the fatty acid profiles of the fish larvae did not always agree with those of the plankton.

Fatty acid composition of M2 \times L15 mirror carp was demonstrably different from FA composition of other mirror carp groups (comparable growth intensity, content of lipids and other nutrients in muscle tissues) particularly in lower ($P < 0.01$) percentage of PUFA $_{\text{sum}}$ ($n-3$ and $n-6$ fatty acids), and was almost identical with lipid composition of the control ROP \times TAT hybrids. The exception was the palmitic acid (C16:0) concentration, which was higher ($P < 0.01$) in M2 \times L15 mirror carp, with consequently higher SFA $_{\text{sum}}$ ($P < 0.05$). This finding may be connected with a change in nutrient habits of M2 \times L15 carp with higher cereal supplement ratio in the diet. Another hypothetical reason for differences in fatty acid composition may lie in differences in genetic specifications between groups tested, which could have resulted in differences in activity levels of the appropriate desaturation and elongation enzymes. Significant difference in PUFA content between crossbreds was found in a previous study in common carp (Buchtová et al. 2007). Hoffman and Prinsloo (1995) investigated fatty acid profile in 4 different genetic strains of *Clarias gariepinus* grown under identical environmental conditions. Authors found significant differences in the content of C16:0, C18:1 $_{n-9}$, C22:5 $_{n-3}$, C22:6 $_{n-3}$ and $n-3/n-6$ ratio. In another experiment, the effect of nutrition on the fatty acid profile in one selected strain was also verified. Similarly, Erickson (1992) found differences in content of PUFA, peroxidizability index for susceptibility of UFA to oxidize, and ratio of PUFA/ α -tocopherol equivalents when comparing three strains of channel catfish (*Ictalurus punctatus*).

Significant differences in the fat between female and male carp were found in one case. Male M2 \times M72 mirror carp fat contained less eicosenoic acid (C20:1) compared to their female counterparts ($1.92 \pm 0.22\%$ vs. $2.53 \pm 0.10\%$, $P < 0.01$). No other sex-related differences in FA concentrations were found.

The results show that differences in the fatty acid profile in various genetic groups of common carp may exist. The lower quality of fatty acid profile in carp reared under pond management may be ascribed to the negative effect of cereal supplementation and/or to the genetic effects. The basic prerequisite for sustainable carp production with desirable muscle lipid compositions should be seen in the development of new feeding and breeding procedures. Supplemental feeding should contain more vegetable oils that

will help to increase PUFA_{sum} concentrations in muscle lipids of the cyprinids, similarly to the approach used in ocean and fresh-water salmoniform fish (Zelenka et al. 2003; Steffens and Wirth 2007). Salmoniform fish feeds commonly contain e.g., rape seed or flax seed oil as substitutes for fish oil, which is very expensive and in short supply. At the same time, experiments are under way to study sesamin, a biologically active substance present in sesame oil, which significantly effects enzyme activity and enhances fatty acid elongation and desaturation capability (Trattner et al. 2008). With regard to breeding, strains and/or crossbreeds with more optimal fat metabolism should be looked for, together with possibilities for an effective selection program.

Chemické složení filetů lysých hybridů kapra obecného (*Cyprinus carpio* L.)

Studie prezentuje výsledky chemického složení filetů včetně složení mastných kyselin tuku (nasycené mastné kyseliny: SFA, mononenasyčené mastné kyseliny: MUFA, polyne-nasyčené mastné kyseliny: PUFA). K vyšetření byly použity filety tří lysých hybridů kapra ve věku tři let: M2 × L15 - maďarský "Szarvas" lysec (M2) a maďarská lysá linie (L15), M2 × DOR70 izraelské plemeno (DOR70) a M2 × M72 severský lysý kapr (M72), které byly srovnávány s filety čisté linie M2 a šupinatým hybridem ropšinského (ROP) a tatajského plemene (TAT). Filet hybrida ROP × TAT obsahoval více ($P < 0.01$) sušiny ($283.1 \pm 23.87 \text{ g} \cdot \text{kg}^{-1}$) a lipidů ($99.3 \pm 30.60 \text{ g} \cdot \text{kg}^{-1}$). Tuk všech sledovaných skupin kapra byl složen z více jak 50% MUFA (od 51 do 64%), 25 - 29% SFA a 10 - 22% PUFA. Filety lysce M2 × DOR70, lysce M2 × M72 a čisté linie M2 obsahovaly méně ($P < 0.01$) MUFA, zejména kyseliny olejové (C18:1_{n-9}) a více ($P < 0.01$) PUFA_{n-3}, zejména kyseliny eikosapentaenové (C20:5_{n-3}) a dokosaheptaenové (C22:6_{n-3}). Rozdílné zastoupení mastných kyselin u jednotlivých skupin kaprů může souviset s jejich rozdílným genetickým vybavením.

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