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Fish quality

**SOME OF FISH SPECIES AS A SOURCE OF n-3 POLYUNSATURATED
FATTY ACIDS**

**WYBRANE GATUNKI RYB JAKO ŹRÓDŁO n-3 POLIENOWYCH
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Muscle lipids of ten fish species (freshwater: zander, whitefish, roach, burbot and crucian carp; farmed: rainbow trout and carp; and Baltic species: garfish, flounder and sprat) were analysed for fatty acids composition (GC/MS), lipid fractions (HPLC) and susceptibility to oxidation. 100 g of meat of lean freshwater and Baltic fish (roach, flounder, burbot and zander) can provide 50% less n-3 PUFAs than the meat of rainbow trout or whitefish and 4–6 times less than sprat. The susceptibility to oxidation of freshwater fish lipids was higher than in the Baltic species of similar PUFAs content.

INTRODUCTION

The importance of fish lipids for human diet arises from the presence of n-3 family, polyunsaturated fatty acids (n-3 PUFAs). The n-3 PUFAs reduce the level of triacylglycerols, VLDL, and LDL-cholesterol (for patients with hyperlipidaemia) in blood serum, and reduce high blood pressure, thus preventing or curing coronary heart diseases and arteriosclerosis (Kromhout et al. 1985; Kromhout 1988; Sanders 1988). They reduce the risk of breast, pancreas, prostate gland or large intestine cancers; alleviate immunological diseases (arthritis, rheumatism, and kidney disorders) and chronic inflammations. The compounds have a positive effect on development of foetal neural system and its further functioning, improve brain functioning, and eyesight and enhance learning abilities. They

also support treatment of dyslexia, aggression, schizophrenia, psoriasis as well as many other pathological states, though the mechanism of the positive effect of n-3 PUFAs has not been fully explained yet (Lands 1986, 1997; Stansby 1990; Nelson 1997; Ziemiański 1998; Cichoń 1998; Carlson and Neuringer 1999).

Among the long-chain n-3 PUFAs that are present in fish lipids, docosahexaenoic acid (DHA) demonstrates stronger biological activity than eicosapentaenoic acid (EPA) (Suzuki et al. 1995). For human diet, marine fish remain the most important source of the long-chain polyunsaturated fatty acids. Freshwater species contain less n-3 PUFAs, however they have more n-6 PUFAs than marine fish (Sikorski and Kołakowska 1990). The ratio of n-3 PUFAs to n-6 PUFAs in total lipids of freshwater fishes changes mostly between 0.5 and 3.8, whereas for marine fishes it is 4.7–14.4 (Henderson and Tocher 1987). According to the same authors, freshwater eel had the ratio 0.6, whereas the same species caught in seawaters had 2.1.

Fish consumption has recently decreased in Poland, reaching at present about 6 kg per person annually. This places Poland, as far as fish consumption is concerned, behind other European countries as well as behind most of the countries of the world. In Denmark, France, the UK or the USA the level of fish consumption per capita is several times higher than in Poland (Anon 1997; Bykowski 1998). Therefore, besides well-known deep-sea fishes such as mackerel, pilchard or herring, less popular Baltic and freshwater fishes should be considered as a source of healthy fish lipids. Another point is that freshwater fish is easier available fresh, whereas marine, especially deep-sea fish is in most cases stored frozen for a long time before reaching the consumer. This can lead to losses in the content of favourable n-3 PUFAs as well as to accumulation of noxious products of lipid oxidation (Kołakowska et al. 1998). Not only will such lipids lose their beneficial effect, but can cause pathological changes as well (Ziemiański et al. 1991, 1992).

In our previous studies, we examined bream and roach as n-3 PUFAs source (Kołakowska et al. 1991, 1993, 1998). Baltic herring was thoroughly studied (Szczygielski 1998) as well as Atlantic herring, mackerel, and their products: smoked (Kołakowska et al. 1998), salted and canned (Kołakowska et al. 1998, 1999). Antarctic krill was also studied in this respect (Kołakowska et al. 1994).

The aim of this study was to analyse selected fishes coming from Polish inland waters and the Baltic Sea for the content of n-3 PUFAs. Some of these species were also investigated for the composition of lipids and their susceptibility to oxidation.

MATERIAL AND METHODS

The following species were studied: crucian carp (*Carassius carassius* L.), length 19–28 cm, weight 0.3–0.7 kg, from Przeclaw-Staw fish farm; carp (*Cyprinus carpio* L.), 1.2–1.4 kg, Gryfino fish farm; rainbow trout (*Oncorhynchus mykiss* Walb.), 250–270 g, Łoźnica fish farm; zander (*Stizostedion lucioperca* L.), 1.5–1.8 kg, from the Odra and Regalica rivers; roach (*Rutilus rutilus* L.), 0.2–0.3 kg; burbot (*Lota lota* L.), 1.3–1.5 kg; whitefish (*Coregonus lavaretus* L.), 0.4–0.6 kg; garfish (*Belone belone* L.), 0.3–0.5 kg; European flounder (*Platichthys flesus* L.), 0.2–0.3 kg; sprat (*Sprattus sprattus* L.). The carp, trout, and garfish were caught in May, while the other fish in the autumn (September–November).

Approximately 10 kg of whole fish represented a unit sample. The fish were collected within 10–20 hours after catch, thereafter being washed, headed and gutted, washed again, left to drain off, filleted (except for sprat), and finally ground with skin on by means of an electric meat grinder with a 2-mm cutting plate. So prepared, the sample was directly taken to analyses.

Analytical methods

Lipid extraction was done according to Bligh-Dyer method (Bligh and Dyer 1959). The content of lipids was determined gravimetrically through evaporation of a fixed amount of the extract.

The composition of fatty acids was obtained by gas chromatography with a mass spectrometer (Hewlett Packard II, FID, capillary column SP 2560), under conditions as described in the previous paper (Kołakowska and Szczygielski 1994).

The lipid composition was determined with the HPLC method (Waters, PDA 996 detector, Si 60 column, n-hexane: propanol-2:0,1 M acetate buffer) according to Holger et al. (1982).

The susceptibility to oxidation was analysed by irradiating the lipids with UV of 344 nm for 2 hours in Petri dishes. Detailed procedure of the analysis has been previously described (Kołakowska 1991). The oxidation level was determined at 0, 30, 60, 120, and 180 min. of irradiation by obtaining the peroxide value with the thiocyanate technique (PTS, 1994), absorbance at 285 nm, absorbance at 470 nm (Pye Unicam), fluorescence spectrum (spectrofluorimeter Hitachi) Ex 365 nm.

The results that are presented in tables and figures are the means of three parallel analyses carried out on the same lipid extract.

RESULTS

The muscle tissue (together with skin) of the studied fish, coming mainly from November and May catches, contained 1.61–19% of fat (Tab. 1). The meat of carp, sprat,

Table 1

Content of lipids in muscle (with skin)
of the studied fishes

Species	Date of catch	Lipids % w.w.
Crucian carp	6 Nov 99	4.10
Zander	9 Nov 99	1.88
Whitefish	29 Nov 99	8.75
Burbot	29 Nov 99	1.61
Flounder	8 Nov 99	4.67
Sprat	6 Nov 99	12.24
Carp	19 May 99	19.05
Rainbow trout	4 May 99	7.86
Roach	12 May 99	2.17
Garfish	12 May 99	2.44

whitefish, and rainbow trout was the most fatty. The muscle tissue of crucian carp and flounder had above 4%, and zander, garfish and burbot had less than 2%.

The lipids differed in fatty acids composition (Tab. 2), both in respect to the proportion of the groups of acids (saturated, monoenoic, and polyenoic) and to the proportion of particular fatty acids.

Two most important cultured freshwater species—carp and rainbow trout—differed in the content of lipids and the composition of fatty acids in muscle tissue. The studied carp had a large amount of muscle lipids, however with relatively poor composition of fatty acids. Monoenoic fatty acids—mainly 18 : 1 (oleic acid)—comprised more than a half of the profile, whereas unsaturated acids made up 25% of the total. The content of polyunsaturated acids in carp's muscle lipids was, besides roach, the lowest of all the studied fishes, and it was practically only linoleic acid. Carp's muscle lipids contained very little n-3 PUFAs—linolenic acid only. Long-chain n-3 PUFAs, EPA or DHA were not detected in the examined carp.

On the other hand, muscle lipids of rainbow trout consisted of almost 30% PUFAs, and there were five times more n-3 acids than the n-6 ones. The dominant polyenoic acid was DHA. Having nearly 8% of lipids, rainbow trout can provide approximately 2 g of n-3 PUFAs from 100 g of meat (Tab. 3).

The freshwater burbot, zander and roach, as well as marine garfish, were lean fishes, containing about 2% of muscle lipids.

Zander and burbot, the leanest of the freshwater fishes, did not show high differences in fatty acids composition. Both species had above 30% of polyenoic acids with dominant n-3 PUFAs and DHA. However, in the lipids of zander, DHA was present in significantly lower amount, but shorter polyenoic acids and n-6 acids were more abundant comparing to burbot. As a result, the ratio of n-3 to n-6 PUFAs was considerably higher in burbot, being nearly 14, whereas in zander—about 9 (Tab. 2).

Table 2

Fatty acid composition of lipids in muscle studied fishes species

Fatty acid	Rainbow trout	Garfish	Roach	Carp	Sprat	Crucian carp	Zander	Flounder	Whitefish	Burbot
12 : 0	Tr	Tr	Tr	0.29	Tr	Tr	Tr	Tr	Tr	Tr
14 : 0	6.17	3.04	2.86	1.66	5.89	2.47	2.83	4.70	1.74	2.49
15 : 0	0.49	0.41	0.86	Tr	1.49	2.67	1.49	1.00	0.79	0.44
16 : 0	21.53	26.14	24.13	17.73	23.62	18.14	21.96	22.70	21.40	24.29
17 : 0	Tr	Tr	1.28	Tr	Tr	2.69	1.40	1.70	0.72	0.86
18 : 0	2.87	5.23	5.85	5.05	2.12	3.60	3.61	2.63	4.73	4.22
Sum SFA	31.06	34.82	34.98	24.73	33.12	29.55	31.28	32.73	29.38	32.30
15 : 1	0.00	0.81	Tr	Tr	Tr	Tr	Tr	Tr	Tr	Tr
16 : 1	6.50	8.82	12.59	5.29	5.94	10.94	12.85	17.33	12.71	6.27
17 : 1	Tr	0.54	1.00	Tr	1.33	1.10	0.70	2.00	0.62	0.87
18 : 1	22.36	24.24	30.34	47.95	24.17	24.40	19.25	22.24	34.79	24.86
20 : 1	5.22	2.60	2.90	2.10	0.38	1.15	0.15	3.34	1.97	0.60
22 : 1	5.90	2.00	Tr	Tr	0.18	Tr	0.19	0.09	Tr	Tr
24 : 1	Tr	Tr	Tr	Tr	0.64	Tr	Tr	Tr	Tr	Tr
Sum MUFA	39.98	39.01	46.83	55.34	32.64	37.58	33.13	44.98	50.09	32.60
16 : 2 n-6	Tr	Tr	Tr	Tr	Tr	Tr	0.58	0.39	Tr	Tr
18 : 2 n-6	4.94	1.77	1.41	17.78	2.80	5.38	2.69	1.20	0.92	2.17
18 : 3 n-3	1.64	Tr	0.55	1.49	3.12	4.76	3.72	1.58	0.70	1.40
18 : 4 n-3	1.12	0.32	Tr	Tr	2.92	0.91	1.07	1.11	0.40	0.82
20 : 2 n-6	Tr	Tr	Tr	0.19	0.24	Tr	Tr	Tr	0.27	Tr
20 : 3 n-3	Tr	Tr	Tr	Tr	Tr	0.27	Tr	Tr	Tr	Tr
20 : 4 n-3	0.56	Tr	2.70	0.47	0.53	3.16	2.93	1.12	0.95	3.50
20 : 5 n-3	4.40	1.42	4.03	Tr	9.37	4.98	8.78	9.02	4.75	6.38
22 : 2 n-6	Tr	Tr	Tr	Tr	0.41	0.64	0.26	Tr	Tr	Tr
22 : 3 n-3	Tr	Tr	Tr	Tr	Tr	0.29	0.16	Tr	Tr	Tr
22 : 4 n-3	Tr	Tr	Tr	Tr	Tr	0.64	0.21	Tr	Tr	Tr
22 : 5 n-3	0.76	0.85	0.74	Tr	0.40	1.95	1.76	0.98	0.98	0.88
22 : 6 n-3	15.54	21.81	8.76	Tr	14.45	9.93	13.46	6.90	11.56	17.21
Sum PUFA	28.96	26.17	18.19	19.93	34.24	32.88	35.60	22.29	20.53	32.36
Sum (n-3) PUFA	24.02	24.40	16.78	1.96	30.79	26.87	32.07	20.70	19.34	30.19
(n-3)/(n-6)	4.9	13.8	11.9	0.1	8.9	4.5	9.1	13.0	16.3	13.9

Table 3

Content of n-3 PUFAs in tissue (with skin) in g/100g

Species	n-3 PUFA	DHA
Crucian carp	1.10	0.41
Zander	0.60	0.25
Whitefish	1.69	1.01
Burbot	0.49	0.28
Garfish	0.64	0.56
Roach	0.36	0.19
Carp	0.37	Tr
Rainbow trout	1.89	1.22
Flounder	0.97	0.32
Sprat	3.77	1.77

Roach had the least polyenoic acids from among the freshwater fishes (except for carp). Those acids did not exceed 20% of total fatty acids, and although they were n-3 PUFAs, the roach caught in May was a poor source of those acids (as calculated per 100 g of meat—Tab. 3).

Garfish, which was also treated as a lean fish, had about 20% less polyenoic acids than zander and burbot, but the amount of EPA and DHA was similar in all three species and was about 23% of total fatty acids. In garfish, however, almost only DHA accounted for n-3 PUFAs. EPA was present in less than 2%, which was the lowest level of EPA for all the species, except for carp. The content of DHA was 22% and was the highest among all the studied fishes. Therefore, garfish is the richest source of DHA, except for fatty fishes, as it is demonstrated in Tab. 4.

Garfish, which was also treated as a lean fish, had about 20% less polyenoic acids than

Table 4

Lipids composition in some of the studied fishes (% of fraction)

Species	Triacyl-glycerols	Cholesterol	Free Fatty Acids	Phosphacyl-glycerols	Lysophosphacyl-glycerols
Crucian carp	50.26	6.54	22.82	20.38	—
Zander	48.93	4.36	19.44	27.27	—
Whitefish	52.07	6.95	18.07	22.91	—
Burbot	40.37	5.81	28.33	25.31	0.18
Flounder	57.58	24.97	8.73	8.09	0.63
Sprat	50.96	17.31	17.68	0.92	13.13

Crucian carp and flounder, the fishes of similar lipid content but entirely biologically and environmentally different, significantly differed in the composition of lipids in muscle tissue as well.

Flounder had extremely high cholesterol fraction, little phosphoacylglycerols, especially phosphatidylcholine, and a large amount of monoenoic acids, with the 16 : 1 acid in higher quantity than the other fishes. There were little polyenoic acids, only 22% and the ratio of EPA to DHA was nearly as 1 : 1, with EPA slightly prevailing, which was characteristic only for flounder.

The differences in fatty acids composition for flounder and crucian carp were well characterised by the n-3/n-6 ratio, which was over 4 in crucian carp and about 13 in flounder.

Crucian carp's lipids were the most similar to trout ones in fatty acids composition. The n-3/n-6 ratio in both species was about 4, however the lipids of crucian carp were clearly poorer in DHA, though they contained more alpha-linolenic acid than the lipids of trout.

Whitefish muscle tissue contained over 8% of lipids (Tab. 1), a half of which were triacylglycerols (Tab. 4). The characteristic feature of whitefish was a high, nearly 35%, content of 18 : 1 acid with 20% of polyenoic acids. However, the ratio n-3/n-6 was the highest among the studied fishes.

Sprat, caught in November, had over 12% of fat in the headless (Tab. 1), more than a half of which consisted of triacylglycerols and relatively high amount of cholesterol (Tab. 4). The contents of saturated, monoenoic and polyenoic acids were similar. Comparing to the other studied fishes, sprat lipids contained the most polyenoic acids, despite a little amount of phosphoacylglycerols fraction (Tab. 4).

As demonstrated in Tab. 3, the richest source of n-3 PUFAs were (respectively): sprat, rainbow trout, whitefish, and crucian carp. However, the level of n-3 PUFAs in sprat was 3 times higher than in crucian carp. The analysed muscle tissue of lean fishes, both freshwater and marine, can provide less than half of the n-3 PUFAs comparing to sprat, trout, whitefish and crucian carp. Considering DHA—the most important acid—only sprat, trout and whitefish can provide over 1 g of DHA from 100 g of muscle tissue. Garfish is also worth considering, as it contains more DHA than the muscles of flounder or crucian carp, which contain twice more total lipids.

Table 5

Relationship between peroxide value changes (y)
and lipid UV exposure time (x)

Species	Linear function parameters $y = bx - a$		
	b	a	R
Crucian carp	12.83	170.44	0.94
Zander	19.37	36.87	0.95
Flounder	3.80	43.11	0.96
Sprat	6.44	109.76	0.97

Among the fishes whose lipids were analysed for susceptibility to oxidation (Fig. 1, Tab. 5) the lipids of flounder appeared to be resistant to photooxidation, and they specifically contained the least polyenoic acids. The lipids of zander, on the other hand, with high level of polyenoic acids, displayed the most rapid oxid-

ation. A similar effect was noted in the case of crucian carp but not sprat, whose lipids—rich in polyenoic acids—were relatively resistant to oxidation.

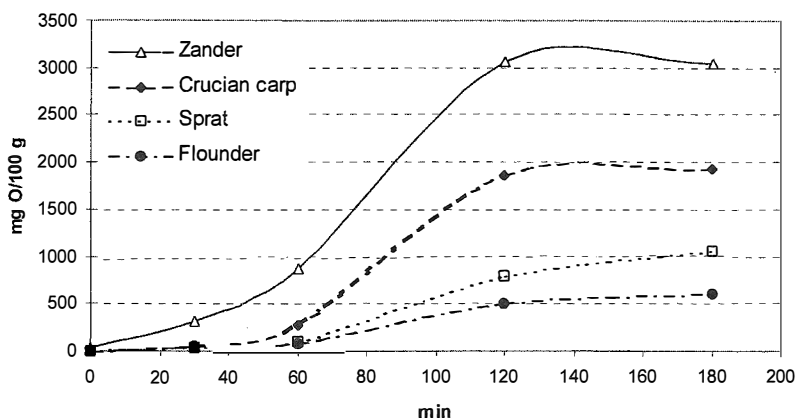


Fig. 1. Changes in peroxide value of fish lipids during UV exposure

DISCUSSION

The studied species are harvested in Polish waters and are available on the market. Rainbow trout and carp are the most popular farming species, however, as it was demonstrated in this study, both fishes differ greatly as far as nutritional value of their lipids is concerned. 100 g of trout fillets provides nearly 2 g of n-3 PUFAs along with about 8 g of lipids, which makes the fish a valuable source of the acids in concern. These results do not vary from the data collected by Hepburn et al. (1986), Westre et al. (1993), and Hyvonen and Koivistoinen (1994). The carp of the studied lot, although very fatty (nearly 20%), had almost no n-3 PUFAs, and as such, the tasty fish presents little nutritional value from the point of view of its lipids. According to the monograph by Henderson and Tocher (1987), carp lipids contained as much as 18% of n-3 PUFAs. Ackman (1974) observed 10% of n-3 PUFAs in carp, in another paper (Ackman 1988) recorded 0.67 g of DHA per 100 g of the fish. None of these papers however quotes the total content of lipids, which makes further comparisons impossible. On the other hand, the data by Hepburn et al. (1986), concerning the content of n-3 PUFAs in 100 g of carp with 5% of fat, are similar to the results of this study. The above divergences are understandable, as the composition of fatty acids heavily depends on the composition of the feed. Therefore, the lipids of both carp and trout, although not to the same degree, can be modified in the process of rearing in respect to fatty acids composition, especially for triacylglycerols (Farkas et al. 1980).

The muscle tissue of the lean freshwater and Baltic fishes, having about 2% of fat, i.e. zander, burbot, roach, and garfish, as well as those with about 4% of fat, can hardly be treated as a considerable source of n-3 PUFAs. What is more, zander and crucian carp

demonstrate high susceptibility to lipid oxidation, which even at low content of lipids can lead to rapid quality deterioration, e.g. during cold storage.

Clear differences in fatty acids composition were observed among particular species. From the nutritional point of view, the composition of garfish's lipids is favourable, whereas n-3 PUFAs in flounder are scarce and its lipids contain much cholesterol. No data concerning the composition of Baltic flounder fatty acids have been found in the available literature. There is also very little information on the lipid composition of the other studied species. Sykora and Valenta (1979) studied the composition of fatty acids in muscles and internal organs (white muscle, dark muscle, liver, and brain), and Hyvonen and Koivistoinen (1994) in skinless fillets of roach. The results of the present study correspond to the data by these authors as well as to the previous studies (Kołakowska 1998), and confirm little value of roach lipids as to the content of n-3 PUFAs. Hyvonen and Koivistoinen (1994) also analysed the muscle of zander, whose DHA content was 0.301 g per 100 g of tissue, thus being slightly higher than observed in this study—probably because skinless fillets were taken for the analysis.

For all the studied species, n-3 PUFAs dominated in the polyenoic acids and, with the exception for flounder, DHA prevailed over EPA.

The n-3/n-6 PUFAs ratio changed between 4.5 (except for carp) and 16.35. The low values referred to the lipids of crucian carp and rainbow trout, though the freshwater fishes had this index generally much higher than observed by Henderson and Tocher (1987). The sprat caught in November was the richest source of n-3 PUFAs, also because its high content of fat. Moreover, the lipids of sprat were at least twice as resistant to oxidation than the same unsaturation degree lipids of freshwater fishes. Sprat's lipids oxidation rate was similar to Baltic herring, though depending on the period of catch, as was demonstrated previously (Kołakowska et al. 1992). The fatty acids composition of Baltic herring also shows seasonal changes (Szczygielski 1998). Such changes in the life cycle of freshwater fishes are probably smaller, which was indicated by the studies on bream (Kołakowska et al. 1994), however, this should be confirmed by studies on other species as well. A range of other environmental and biological factors can influence the content and composition of lipids. Serrini et al. (1996) found differences in n-3 PUFAs content for a coregonid species living in different parts of Italy.

CONCLUSIONS

1. Among the studied fishes, the muscle tissue of sprat, rainbow trout, and whitefish were a good source of n-3 PUFAs. The cultured carp, which was analysed in this study, cannot be considered as a source of n-3 acids.

2. The n-3/n-6 PUFAs ratio in the muscle lipids of Polish freshwater fishes is higher than described in reference literature.
3. In comparison with Baltic fishes of similar polyunsaturated fatty acids content, the susceptibility to oxidation of the muscle lipids of freshwater fishes was higher.

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WYBRANE GATUNKI RYB JAKO ŹRÓDŁO n-3 POLIENOWYCH
KWASÓW TŁUSZCZOWYCH

STRESZCZENIE

Analizowano tkankę mięśniową (ze skórą) 10 gatunków ryb, poławianych w polskich zbiornikach wodnych i M. Bałtyckim. Z ryb słodkowodnych analizowany był: sandacz, sieja, płoć, miętus, karaś, hodowlanych: pstrąg i karp oraz ryb bałtyckich: belona, stornia i szprot. Określono skład kwasów tłuszczowych lipidów mięśniowych metodą GC/MS (we wszystkich próbach), frakcji lipidów metodą HPLC (w 6 gatunkach), podatność lipidów na utlenianie za pomocą testu fotooksydacji (w 4 gatunkach). Stwierdzono istotne różnice w składzie lipidów tkanki mięśniowej badanych ryb i ich podatności na utlenianie. Stosunek n-3/n-6 PUFA mieścił się w przedziale od około 4 do 16. Lipidy karpia zawierały jedynie śladowe ilości n-3 PUFA, zaś badany pstrąg był dobrym ich źródłem. Spośród badanych ryb najbogatszym źródłem n-3 PUFA były w kolejności malejącej: szprot, pstrąg, sieja, karaś, z tym, że w mięsie szprota było ich 3-krotnie więcej niż w mięsie karasia. 100 g tkanki mięśniowej ryb chudych słodkowodnych i morskich: płoć, stornia, miętus, sandacz może dostarczyć tylko 50% tej ilości n-3 PUFA co pstrąg i sieja i 4–6-krotnie mniej niż szprot.

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