Influence of dietary linseed and sunflower oil on sensory characteristics of rainbow trout (Oncorhynchus mykiss)

Z. Drobná, J. Zelenka, E. Mrkvicová, D. Kladroba

Faculty of Agronomy, Mendel University of Agriculture and Forestry, Brno, Czech Republic

ABSTRACT: This study describes the effects of dietary linseed and sunflower oil on the sensory characteristics of rainbow trout (*Oncorhynchus mykiss*) fillets. Rainbow trout were fed a diet containing either 2.5% or 5% of linseed or sunflower oil or 5% mixture of both oils (2.5% of each). Control group received a commercial feed mixture. A selected and trained panel carried out descriptive sensory tests on fish fillet cross-sections. The addition of vegetable oil into fish feed did not affect firmness, juiciness or intensity of total taste but had a significant impact on colour, intensity of odour and fish oily taste. A higher content of n-3 polyunsaturated fatty acids (PUFA) in linseed oil, in comparison with sunflower oil, is often responsible for the development of an off-flavour. No trout fillet was rejected owing to the off-flavour presence. Both concentrations of linseed oil decreased the intensity of fish oily taste. Therefore, for the partial replacement of fish oil in rainbow trout feed it is recommended the use linseed oil, which is a good source of n-3 PUFA.

Keywords: rainbow trout feeding; vegetable oils; meat quality; organoleptic properties; PUFA

Nowadays, consumers have very high demands on their food. It must be healthy, natural, quality, safe and convenient and, on top of that, it should have pleasant appearance, texture, odour and taste. Fish meat meets the majority of these requirements.

The uniqueness of fish meat in human nutrition lies not only in its high-quality protein, for which there are many competitive alternatives, but also in a high content of long-chain n-3 polyunsaturated fatty acids (PUFA), i.e. all-cis-5,8,11,14,17eicosapentaenoic acid (EPA) and all-cis-4,7,10,13, 16,19-docosahexaenoic acid (DHA) (Sargent and Tacon, 1999). The muscle is often the main part of the fish used for human consumption and salmon has been suggested to be a major human dietary source of n-3 fatty acids (Kiessling et al., 2001). Salmonids are usually considered to be medium-fat fish, with the muscle lipid content in the range of 2–7% (Jobling, 2001).

The health-promoting effects of n-3 PUFA in humans have been reviewed extensively, and they are only briefly summarized here. The evidence of reduced incidence of cardiovascular disease in Eskimos, in the population with high dietary intake of n-3 fatty acids derived from fish, suggests that dietary supplementation of n-3 PUFA may be beneficial in the reduction of cardiovascular risks (Tanakol et al., 1999).

All-cis-5,8,11,14-eicosatetraenoic acid (arachidonic acid) is a precursor of eicosanoids. At dietary intakes of cis, cis-9,12-octadecadienoic acid (linoleic acid – LA) higher than approximately 5% of total energy intake, eicosanoid productions approach their maximum, which is undesirable as illustrated, for example, by the extensive use of steroidal and non-steroidal anti-inflammatory agents blocking eicosanoid production in many human disorders, including cardiovascular and inflammatory disorders and cancers (Sargent and Tacon, 1999). To depress eicosanoid production from arachidonic acid substantially requires a dietary intake of LA lower than 2% of total energy intake. Fortunately, the desired effect can be achieved by even modest dietary intakes of all-cis-9, 12, 15-octadecatrienoic acid (α-linolenic acid – LNA), approximately of 1% of total energy intake. This is caused by the fact that LNA competes very effectively with LA for the common fatty acid desaturases and elongases that convert C_{18} PUFA to their C_{20} and C_{22} homologues (Sargent and Tacon, 1999). For that reason, the current recommendation regarding the consumption of PUFA in human nutrition is 4:1 as the ratio of n-6 to n-3 PUFA (Ralph, 2000).

Furthermore, DHA plays a special role in human nutrition for its high concentration in neural tissues (Tanakol et al., 1999). Although humans can undoubtedly convert LNA to EPA and thence to DHA, the rate of production of DHA from LNA need not be sufficiently fast at all times and under all conditions, especially in the case of premature infants (Sargent and Tacon, 1999). Evidence is also emerging concerning the role of EPA and DHA in various mental disorders, including schizophrenia, and in aggressive behaviour (Sargent and Tacon, 1999).

Although fish are not the only source of fatty acids in human nutrition, fish contain by far the highest concentrations of EPA and DHA of any of the foods commonly available to humans. Freshwater fish, including trout, are capable to convert LNA to its higher biologically active homologues – EPA and DHA, and also LA to arachidonic acid (Sargent et al., 1993, 1995). In contrast, no marine fish so far studied can carry out these conversions (Sargent et al., 1993, 1995). Thus, the presence of EPA and DHA in a high concentration in fish oil is an essential dietary constituent of fish feed.

Vegetable oils can partially substitute EPA and DHA in trout feed. LNA-rich oils, such as linseed oil, should be used for the partial substitution of fish oil in fish feed because they are much more acceptable from the human nutritional perspective, especially considering the innate ability of freshwater fish to convert LNA to EPA and DHA.

This study was focused on the effects of linseed and sunflower oils in a trout diet on sensory attributes of rainbow trout fillets.

MATERIAL AND METHODS

Materials and samples preparation

A detailed description of the fish, experimental design, feeding schedules, sampling, sampling times and locations of the samples are given in Zelenka et al. (2003) and they are briefly summarized below.

For this experiment, one-year-old rainbow trout (*Oncorhynchus mykiss*) were used of the aver-

age body weight 257 g. All fish were divided into six groups. One group was fed the control diet – commercial mixture Extruded Trout Grower TroCo SUPREME-16 EX (produced by Coppens International bv, The Netherlands). The other groups of fish received one of the five experimental diets in which either 2.5% or 5% of the commercial mixture was replaced by sunflower oil (S), linseed oil (L) or by a mixture of both oils at the ratio of 2.5% L:2.5% S. The composition of diets, their nutritive values and basic production parameters were shown in Zelenka et al. (2003).

After 75 days of fattening, specimens of each group were selected so that the differences in their live body weight would be as low as possible. Selected fish were individually weighed and killed. After gutting, they were put into freeze bags, chilled on ice until rigor shortening took place (24 h) and frozen until the sensory analyses were carried out.

The frozen fish were allowed to thaw to an internal core temperature of 1°C. The fish were individually wrapped in a double aluminium foil, placed on an oven-plate and baked in an electric household oven at 200°C for 10 min to the final core temperature of 80°C, as recommended by Chambers and Robel (1993). Different baking and core temperatures as well as different baking time are reported by other authors (Ågren and Hänninem, 1993; Johansson et al., 2000; Warm et al., 2000). The temperature was measured by inserting a digital thermometer into fish. The fish were baked in their own juice.

Sensory analysis

Sensory evaluation was performed by a trained internal sensory panel (Mendel University Brno, Czech Republic) consisting of eight persons. All volunteers were selected on the basis of their interest and availability. Their sensorial capacities of memorising stimuli and discriminating intensities were screened following the ISO guidelines (ISO 8586-1, 1993). In agreement with Majou et al. (2001), the panellists attended regular training sessions necessary to develop their sensory performances with respect to fish assessment.

Sensory sessions were conducted in a test room (ISO 8589, 1988) with 10 individual booths. Adapted flavour profile method and texture profile meth-

od (Meilgaard et al., 1991; Hylding and Nielsen, 2001; Majou et al., 2001) were used to evaluate the sensory characteristics of samples. The panellists were familiarized with the sensory methodology and sensory attributes during the training sessions. Sensory attributes were selected from those previously reported in the literature (e.g. Chambers and Robel, 1993; Johansson et al., 2000; Warm et al., 2000) and confirmed during the initial training session.

The 100-mm unstructured line scales with two anchor points were used. The anchor points were placed 15 mm and 85 mm from zero on the scale and were labelled as follows. Odour: slightly intensive – very intensive; colour: greyish – white (middle) – yellowish; firmness: very soft – very firm; juiciness: very dry – very juicy; total taste: slightly intensive – very intensive; oily taste: slightly intensive – very intensive. In the case of any offodour and/or off-flavour presence, panellists had to specify it.

After heat treatment, each fish was cut into eight pieces. This "sub-samples" without skin and red muscle were presented to the panel in a sequence ensuring that each panellist received the same part of the fish every time. All samples were labelled with 3-digit numbers, randomised and served warm $(40-50^{\circ}C)$. The odour was evaluated immediately while flavour and texture attributes were evaluated after cutting the sample. Colour assessments were conducted against a white background.

Between sample evaluations the panellists were required to eat unflavoured bread, rinse their mouth thoroughly with drinking water and, if necessary, to use vodka to minimize the carry-over effect. Six fish samples, one fish from each diet group, were assessed in one session. The sensory evaluation was carried out in eight sessions during four separate days.

Statistical analysis

The three-way analysis of variance (ANOVA) with interactions was used. Apart from the diet influence, the effect of panellists and sessions was considered as well. Interactions between all main effects were calculated. Student-Newman-Keuls multiple comparison test was carried out to determine the significance of differences between the groups. The principal component analysis (PCA) was conducted for all judgements across all treatments on the mean judgement scores for each attribute and each treatment using the correlation matrix. The Unistat Package, version 4.53 (Unistat Ltd., London, England) was used for all calculations.

RESULTS

The mean scores for all evaluated attributes are shown in Table 1. Graphic presentation of these values is given in Figure 1.

The presence of linseed or sunflower oil in a diet did not influence firmness, juiciness and total taste of rainbow trout fillets (Table 2). As expected, statistically significant differences between panellists (P < 0.001) as well as significant panellist × session (P < 0.001) interactions were found for each attribute. The treatment × panellist interactions for all attributes, except oily taste, were statistically significant (P < 0.05).

As the ANOVA results showed significant *F*-values, the Student-Newman-Keuls multiple comparison test was used for each attribute to find out which means of treatment were significantly different (Figure 1). The odour of fillets obtained from trout fed the diet containing 2.5% sunflower oil, 2.5% linseed oil or mixture of both oils (2.5% sun-

Diet	Odour	Colour	Firmness	Juiciness	Total taste	Oily taste
Control	55.9 ± 1.39	56.2 ± 1.16	53.3 ± 1.54	44.2 ± 1.34	47.2 ± 1.30	46.2 ± 2.03
2.5% sunflower oil	59.7 ± 1.70	54.5 ± 1.78	51.6 ± 1.87	47.8 ± 1.26	48.7 ± 1.44	50.7 ± 2.10
5% sunflower oil	55.0 ± 1.81	51.3 ± 0.98	52.6 ± 1.76	47.8 ± 1.52	48.0 ± 1.54	46.0 ± 2.08
2.5% linseed oil	58.2 ± 1.46	52.6 ± 1.08	48.9 ± 1.71	47.3 ± 1.33	48.2 ± 1.41	42.9 ± 2.17
5% linseed oil	53.2 ± 1.47	54.8 ± 1.41	48.0 ± 1.58	48.6 ± 1.54	47.8 ± 1.47	42.4 ± 2.07
2.5% linseed oil + 2.5% sunflower oil	59.5 ± 1.69	52.0 ± 1.05	51.7 ± 1.86	45.3 ± 1.28	46.3 ± 1.50	50.8 ± 2.15

*mean (mm of the unstructured line scale) ± standard error of the mean



Figure 1. The effect of trout diet on the sensory characteristics of fillets – means within an attribute followed by the same letter are not significantly different from one to another at P < 0.05

flower oil + 2.5% linseed oil) was significantly more intensive (P < 0.05) than from the others. Panellists perceived no off-odour in evaluated samples. Fillets from the control group of trout had a stronger shade of yellow compared to the colour of fillets from the other groups. This colour differences were statistically confirmed at P < 0.05.

Regarding the assessments of texture attributes, no statistically significant differences were observed. Generally, the white muscle of trout fillets from all diet groups was evaluated as firmer and drier. No statistically significant influence of dietary sunflower or linseed oil content on the total taste of fillets was found. According to the panellists' assessments, the taste of all sensory evaluated fillets corresponded to the typical taste of trout, without any occurrence of off-flavour. For oily taste, highly significant differences (P < 0.001) were observed between treatments (Table 2). The multiple comparison test (Figure 1) divided all diet groups of trout into three "sub-groups" that differed significantly (P < 0.05). The most intensive oily taste was perceived in fillets from the trout fed 2.5% sunflower oil and/or mixture of both oils (2.5% sunflower oil + 2.5% linseed oil). On the other hand, the lowest intensity of oily taste was perceived in fillets of the trout fed both 2.5% and 5% linseed oil. The last "sub-group" of fillets consisted of trout from the control group and from the diet group fed 5% sunflower oil.

PCA of the panellists' mean scores across all treatments for each attribute and across all attributes for each treatment was carried out. The way of using the attributes as a whole to describe

Source of variance	df	Odour	Colour	Firmness	Juiciness	Total taste	Oily taste
Treatments (T)	5	*	**	NS	NS	NS	***
Panellists (P)	7	10 AF 10	***	***	***	***	***
Sessions (S)	7	NS	*	*	NS	NS	**
$T \times P$	35	46	**	16-16-	*	*	NS
$T \times S$	35	NS	NS	NS	NS	NS	NS
$P \times S$	49	***	***	***	***	***	***

Table 2. Analysis of variance - the significance of F-tests for all sensory attributes

*P < 0.05, **P < 0.01, ***P < 0.001

NS = not significant; df = degrees of freedom



Figure 2. Results of principal components analysis of the descriptive analysis of sensory data for sensory attributes and diet groups, prefix C = control, 2.5S = 2.5% sunflower oil, 5S = 5% sunflower oil, 2.5L = 2.5% linseed oil, 5L = 5% linseed oil, SL = 2.5% linseed oil + 2.5% sunflower oil; suffix – numbers correspond to panellists 1 through 8

the treatments by the panel is shown in Figure 2. PCA accounted for 76.61% of the variance in the data with the first three components, whereas two components accounted for 65.54%. The first principal component (PC1) accounted for 52.86% of the variance (Table 3) and was highly loaded in the positive direction with firmness and highly loaded in the negative direction with juiciness (Figure 2). The second principal component (PC2) accounted for an additional 12.68% of the variance in the data set and was highly loaded in the positive direction with oily taste.

The location of the panellists on the PCA plot (Figure 2) showed similarities and differences in

Table 3. The percentage of total variance attributable to the first two principal components for all diet groups

Diet	PC1 (%)	PC2 (%)
Control	59.95	11.09
2.5% sunflower oil	62.60	23.55
5% sunflower oil	40.76	33.16
2.5% linseed oil	57.50	24.45
5% linseed oil	54.07	23.06
2.5% linseed oil + 2.5% sunflower oil	64.94	22.33

their assessment of each diet group of trout. The percentage of the total variance attributable to the first two principal components for all diet groups is shown in Table 3. In general, the panellists who were grouped together were most alike in their judgement, while the panellists who were located further from the others were more dissimilar.

For the control group of trout, the assessments of all panellists were very similar and highly correlated with the positive direction on PC1. In contrast, the assessments of fillets of trout fed 2.5% sunflower oil made by nearly all panellists were loaded in the negative direction of PC1. Panellist number 8, whose assessment highly negatively correlated with PC2, was identified as an outlier for this diet group. According to the evaluation of 5% sunflower oil diet group, the panel was divided into two small groups and some free panellists. In the negative direction of PC1, there were located two groups of panellists with similar judgements within the group but panellist number 2 with a dissimilar judgement was found in the opposite direction.

The ratings of panellists for the trout fed 2.5% linseed oil were very similar although the outlier of group was identified (panellist number 6). Similarly like the panellists' assessments of fillets of trout fed

the 5% sunflower oil diet, the judgements of trout fed the higher concentration of linseed oil were highly dissimilar. The majority of the panellists' assessments were loaded in the positive direction on PC1 but two free panellists (number 3 and 7) were found in the positive direction on PC2. The panel evaluations of the last diet group (2.5% sunflower + 2.5% linseed oil) were highly correlated with the positive direction on PC1 even though two outlier panellists (number 2 and 6) with high positive correlation with PC2 were identified.

DISCUSSION

The quality assessment of rainbow trout must be considered from two aspects. One important attribute is its nutritional value as a source of n-3 PUFA but sensory attributes like colour, texture, odour and flavour need to be taken into account as well.

The influence of dietary linseed and sunflower oil on the fatty acid composition of rainbow trout fillets was already reported by Zelenka et al. (2003) and it was shortly summarized there. The content of saturated fatty acids in the meat of fish of all experimental groups was lower (P < 0.01) than in controls. The percentage of monounsaturated fatty acids in the meat of control fish was higher in comparison with groups receiving 5% of oil in the diet (P < 0.01) and with the group fed the mixture with 2.5% linseed oil (P < 0.05) but did not differ from the 2.5% sunflower oil group (P > 0.05). The meat of control group contained significantly less PUFA than the meat of fish fed 5% sunflower oil (P < 0.05), 5% linseed oil (P < 0.05) and mixture of 2.5% sunflower oil and 2.5% linseed oil (P < 0.01) in their diet. In the fillets from trout on the diet containing sunflower oil significantly higher contents of n-6 PUFA (P < 0.01) were found compared to the other diet groups. In contrast, its partial replacement by linseed oil increased the n-3 PUFA value (P < 0.01) in fish meat. No statistically significant differences (P > 0.05) in EPA and DHA content were found between the fillets of trout on different diets. The highest ratio of n-3/n-6 PUFA was statistically confirmed (P < 0.01) in the diet group containing 5% linseed oil.

Due to this fact, the partial replacement of fish oil by linseed oil in fish feed is much more acceptable from the human nutrition aspect than its replacement by sunflower oil. However, the abundance of fatty acids with a high degree of unsaturation in fillets could affect the sensory characteristics of trout and, especially, might be responsible for the development of an off-flavour resulting from lipid oxidation.

In addition to this, sensory characteristics of fish meat often depend on the proportion of red and white muscle. Most of the fish muscle is white. The red muscle is located just under the skin along the body size. The typical pelagic fish such as herring may contain up to 50% of dark muscle (Warm et al., 2000), whereas rainbow trout contains only 5% of red muscle. There are many differences in the chemical composition of these two muscle types (Ingemansson et al., 1993; Amerio et al., 1996). The high lipid content of red muscle can give high intensities of desirable sensory notes but a shorter shelf-life due to lipid oxidation resulting in off-flavour. According to Kiessling et al. (2001), the white muscle is a tissue showing the largest changes in fatty acid composition in relation to the feed ration. They also supported an earlier funding that the white muscle of rainbow trout functions as a short-term rather than long-term fat depot.

In this study the panellists evaluated only the white muscle of rainbow trout. No trout fillet was rejected due to the off-flavour presence. Generally, the use of vegetable oil in fish feed significantly affected (P < 0.05) odour, colour and oily taste intensity of trout fillets. On the other hand, it had no statistically significant influence (P > 0.05) on firmness, juiciness and total taste intensity.

Although the odour intensity of fillets of trout fed 5% linseed oil was comparable to the intensity of control group, the lower concentration of linseed oil supported the intensity of this attribute. The presence of dietary linseed oil led to a reduction of the yellowish shade of white muscle and to a decrease in oily taste intensity compared to the intensity of control group.

The presence of sunflower oil in the diet had the same influence on the intensity of odour as linseed oil. Once again, dietary sunflower oil led to a reduction of the yellowish shade of white muscle. In the fillets of trout fed 2.5% sunflower oil stronger oily taste was perceived than in the fillets from control group or from diet group fed 5% sunflower oil.

In the case of diet group on 2.5% sunflower oil and 2.5% linseed oil mixture, the panellists perceived stronger odour compared to the control group. The intensity of this attribute was comparable to the intensity of both 2.5% sunflower oil and 2.5% linseed oil diet groups. The yellowish shade of white muscle was reduced again. The use of the mixture of vegetable oils increased the intensity of oily taste identically like in the case of 2.5% sunflower oil diet group.

There are many variations in fish handling and cooking which can alter the fate of lipid stores outside the flesh and thereby affect the amount of lipids lost or absorbed during cooking. In the study of Ågren and Hänninem (1993), the influence of cooking on the fatty acid concentrations in fish flesh was observed. The total concentration of fatty acids highly increased (70%) in boiled trout samples, slightly decreased (5%) in baked trout samples and increased (32%) in microwave samples. It is possible that the shorter cooking time in the microwave oven decreased the loss of fluidified lipids outside the flesh. In practice, all usual cooking methods can be used without significant loss of n-3 PUFA. Similarly, the ratio of n-3/n-6 PUFA in the carp meat was not changed by the heat treatment in the experiment of Fajmonova et al. (2003).

The use of a trained panel to do the descriptive analysis of a product is a standard practice in sensory evaluation. However, even with training, panellists can vary in their perception of the attributes. Significant differences (P < 0.001) between the panellists might be caused by differences in the panellists' use of the scale for both the range of scores and the location of scoring (Wilkinson and Yuksel, 1997). The significant panellists × treatments interactions can be caused by the panellists' attachment of different meaning to the same attribute (Dijksterhuis, 1995) or by the ability to detect small differences in attribute intensities (Schiffman and Lockhead, 1983).

Up to now, several statistical methods have been used to examine judgements or panel performance. According to King et al. (2001), the principal components analysis is recommended because of a simple visual display of judgement relationships and identification of attributes having the greatest effect on panellists' scores. In this study, panellists No. 2 and 6 were identified by PCA as possible outliers who could be responsible for significant treatments × panellists interactions.

CONCLUSION

Although the farmed rainbow trout (*Oncorhynchus mykiss*) are not so a good source of n-3 fatty acids

as the marine fish, the use of linseed oil in fish feed positively affects the content of n-3 PUFA in fish fillets. This is, of course, very important for human diet in inland countries.

Taste and texture traits are the most important consequential factors in determining consumer preference for trout flesh. In this study, the presence of sunflower or linseed oil in fish feed had no pronounced influence on sensory characteristics, except the intensity of fish oily taste.

For the partial replacement of fish oil, the use of 2.5% linseed oil in rainbow trout feed is recommended. The lower concentration of linseed oil increased odour intensity and decreased the intensity of unpleasant oily taste in trout fillets.

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Corresponding Author

Prof. Ing. Jiří Zelenka, CSc., Faculty of Agronomy, Mendel University of Agriculture and Forestry, Zemědělská 1, 613 00 Brno, Czech Republic

Tel. +420 545 133 159, fax +420 545 133 199, e-mail: zelenka@mendelu.cz