

A nitrogen factor for European pike-perch (*Sander lucioperca*), northern pike (*Esox lucius*), and sheatfish (*Silurus glanis*) fillets

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Abstract

Measures for consumer protection against food adulteration and misleading labeling are integrated into EU legislation, including methods for detection of misleading practices. Verification of meat content is available for marine products but not for freshwater fish because of the lack of standard nitrogen factors. The aim of this study was to establish nitrogen factors for European pike-perch *Sander lucioperca* (Linnaeus, 1758), northern pike *Esox lucius* Linnaeus, 1758, and sheatfish *Silurus glanis* Linnaeus, 1758. The study involved analysis of 808 fillet samples obtained in spring (March–April) and autumn (October–November) harvest seasons, 2018–2019, from seven Czech Republic fish rearing facilities. Samples with and without skin were analyzed for nitrogen content, dry matter, protein, ash, and fat according to established ISO methods. The recommended nitrogen factor for European pike-perch with the skin is 3.28 ± 0.09 and without the skin is 3.21 ± 0.09 ; for northern pike with the skin is 3.18 ± 0.09 and without skin is 3.15 ± 0.09 ; and for sheatfish with skin is 2.73 ± 0.13 and without skin is 2.75 ± 0.12 . The established nitrogen factors will enable analysis of meat content to ensure that consumers are purchasing correctly described and labeled fish products.

Keywords

Adulteration, fish-food fraud, fish products, nitrogen factor, fish

Introduction

Freshwater and marine fishes play a significant role in human nutrition as a source of protein and other nutrients (FAO 2016). They provide crucial elements of a healthy diet including digestibility, low fat content, essential vitamins and minerals, and a higher content of unsaturated fatty acids compared to terrestrial animal fat (Adamkova et al. 2011; Lund 2013; Mraz et al. 2017; Linhartova et al. 2018). The nutritional composition of fish muscle is

influenced by many factors such as fish species, age, gender, rearing conditions, breeding technology, and season, etc. (Fajmonova et al. 2003; Buchtova et al. 2008, 2010; Adeniyi et al. 2012).

The annual global per capita consumption of fish products increased from 9.0 kg in 1961 to 20.3 kg in 2017. In this period, fish consumption increased by 3.1 percentage points annually, more than the annual population growth rate of 1.6 percentage points (FAO 2020). The mean consumption of fish and fish products per capita in the EU

was 24.4 kg in 2017 (EC 2020). Variation in consumption among EU countries is significant, which is associated with the eating habits.

Pond fish farming has a long tradition in the Czech Republic, dating to the 12th century with the oldest documented pond in Bohemia established in 1115. In the past ten years, the production of freshwater fish in the Czech Republic reached 20 400–21 800 tons per year, of which approximately 10% is represented by processed fish products for foreign and domestic markets (MA 2019).

Some farmed freshwater fish species popular with consumers are among the more costly foodstuffs, motivating fraud among producers. Meat, including fish meat, carries a high potential for economically motivated adulteration (Čížková et al. 2012; Cavin et al. 2018), most commonly consisting of the addition of undeclared substances or their substitution for genuine ingredients (Everstine et al. 2013; Cavin et al. 2018). These illegal practices can endanger the health of consumers. Examples include the addition of melamine to milk powder used for infant formula, Sudan dyes in chili and paprika, the addition of methanol to spirits, the European horse-meat scandal in 2013 or fipronil contamination of eggs. Another example of misleading of consumers can be undeclared added water (Čížková et al. 2012; Cavin et al. 2018; Morin and Lees 2018).

Consumer protection against adulteration and misleading food labeling is contained in EU legislation. Methods for detection of practices misleading the consumer for pork, chicken, and seafoods have been published in European legislation and in standards of Codex Alimentarius. These are primarily procedures to determine meat content or content of absorbed water (EC 2002; CA 2004; EC 2008). The regulation of meat content in farmed freshwater fish products is not possible because of the absence of established species-specific nitrogen content (nitrogen factor) for freshwater fish, with the exception of tilapia (CA 2004).

The aim of this study was to establish nitrogen factors as determined by the Kjeldahl method (ISO 937 1978) within the context of the Codex standard (CA 2004) for European pike-perch *Sander lucioperca* (Linnaeus, 1758); northern pike *Esox lucius* Linnaeus, 1758; and sheatfish, *Silurus glanis* Linnaeus, 1758) the primary predatory fish species farmed and processed in the Czech Republic for commercial markets. The established nitrogen factors have the potential to provide the basis for verifying content of freshwater fish products.

Methods

Experimental animals and experimental protocol

One-hundred-three market-size European pike-perch (385–2025 g weight) were obtained from five Czech aquaculture facilities: The University of South Bohemia in Ceske Budejovice, Faculty of Fisheries and Protection of Waters (FFPW USB), Vodnany, and the Blatna, Ho-

donin, Klatovy, and Lnare fisheries. Two-hundred-seven market-size northern pike (445–3980 g) were obtained from seven Czech aquaculture facilities: FFPW USB and the Chlumec nad Cidlinou, Blatna, Hodonin, Klatovy, Lnare, and Tabor fisheries. Ninety-four market-size sheatfish (505–9265 g) were obtained from FFPW USB and the Chlumec nad Cidlinou, Blatna, Hodonin, and Klatovy fisheries. The location of the farms is shown in Fig. 1. In order to assess any effect of year, season, and location, fish were collected in the spring (March–April) and autumn (October–November) harvesting seasons in 2018 and 2019. The fish were transported live to the laboratory of the Faculty of Fisheries and Protection of Waters, killed by a blow to the head, weighed, measured, and filleted. Two fillets, one with skin removed, from each fish were individually vacuum packed, immediately frozen, and stored at -32°C until chemical analysis. A total of 808 fillets were chemically analyzed.

The study was conducted according to the principles of the Ethical Committee for the Protection of Animals in Research of the University of South Bohemia, Faculty of Fisheries and Protection of Waters, Vodnany.

Chemical analysis

Samples of fish fillets with and without skin were analyzed for dry matter, protein, fat, and ash. After partial thawing to avoid loss of water and soluble protein fractions, samples were homogenized by grinding on the knife mill PULVERISETTE 11 (FRITSCHE GmbH, Germany).

The determination of percentage of dry matter was based on the standard method ISO 1442 (1997). The homogenized samples were dried with sand up to constant weight at $103 \pm 2^{\circ}\text{C}$ in the laboratory oven Memmert UE 500 (Mettler GmbH + Co. KG, Germany).

The determination of ash content was based on the standard ISO 936 (1998). The homogenized samples were burned in muffle furnace Nabertherm A11/HR (Nabertherm GmbH, Germany) at $550 \pm 25^{\circ}\text{C}$ to a greyish-white color of ash.

Total fat content was assessed based on the standard ISO 1443 (1973). The homogenized samples were hy-



Figure 1. Map of the Czech Republic showing location of aquaculture facilities.

dolyzed by hydrochloric acid, and fat was extracted by light petroleum in the SOXTEC 2050 (FOSS Headquarters, Denmark).

The determination of protein content used the Kjeldahl method based on the standard method ISO 937 (1978). The homogenized samples were digested by sulfuric acid and a catalyzer in the digestion unit KjelROC Digestor 20 (OPSI AB, Sweden) at $420 \pm 10^\circ\text{C}$. Organically bound nitrogen was determined on the KJELTEC 8400 with KJELTEC sampler 8420 (FOSS Headquarters, Denmark). The coefficient 6.25, the conversion factor for meat, was used for calculation of protein content from the nitrogen content.

Analyses of the dry matter, ash, and total fat were conducted in duplicate, and analysis of protein was performed in triplicate for each sample.

Statistical analysis

Kolmogorov–Smirnov and Bartlett’s tests were applied to assess normal distribution data and the homoscedasticity of the variance, respectively. A two-way ANOVA with a subsequent Tukey’s test was performed to test the effects of season, weight of fish, place of rearing, and the difference between fillets with and without skin. The significance level was set at $P < 0.05$. Data were expressed as mean \pm SD (minimum–maximum). The analysis was performed using STATISTICA v.12.0 for Windows (STATSOFT, Inc.).

Principal component analysis (PCA) of the nitrogen and fat content in fillets with and without skin as response values was applied to describe differences in fish samples from different locations. The analysis was complemented with a redundancy analysis (RDA) with whole fish weight as an independent explanatory variable. The ordination plots were produced using Canoco for Windows v.5.10 (Biometris, The Netherlands and P. Šmilauer, Czech Republic).

Results

European pike-perch

The basic composition of fillets with and without skin of European pike-perch is given in Table 1. Dry matter content was significantly lower ($P < 0.01$) in European pike-perch fillets without skin from Lnare (spring 2019, 21.40 ± 0.81), Hodonin (spring 2019, 21.46 ± 0.31 and autumn 2019, 21.49 ± 0.29) and Klatovy (autumn 2018, 21.24 ± 0.73) fisheries compared to those with skin from Blatna in autumn 2018 (23.18 ± 0.42) and autumn 2019 (23.07 ± 0.46). We found no significant differences ($P > 0.05$) in dry matter content between European pike-perch fillets with and without skin within a sampling period.

The ash content was significantly lower ($P < 0.01$) in European pike-perch fillets with skin from Lnare (spring 2018, 1.17 ± 0.04) and Hodonin (spring 2019, 1.17 ± 0.04)

and fillets without skin from Lnare (spring 2018, 1.20 ± 0.03 ; autumn 2018, 1.20 ± 0.12 ; and autumn 2019, 1.18 ± 0.05) and Hodonin (spring 2019, 1.20 ± 0.03), compared to fillets with (1.3 ± 0.08) and without (1.38 ± 0.11) skin from Lnare in spring 2019. We found no significant differences ($P > 0.05$) in ash content of European pike-perch fillets with and without skin within a sampling period.

The fat content was significantly lower ($P < 0.01$) in European pike-perch fillets with (0.14 ± 0.10) and without (0.15 ± 0.14) skin from Lnare (spring 2018), with skin (0.21 ± 0.04) from Blatna (autumn 2019), and without skin (0.21 ± 0.07) from Hodonin (autumn 2019) compared to other groups. We found significant differences ($P < 0.01$) in fat content of fillets with skin and those without skin in European pike-perch from FFPW USB in spring 2018 (1.37 ± 0.50 and 0.69 ± 0.20), Blatna in autumn 2018 (0.75 ± 0.35 and 0.54 ± 0.26) and 2019 (0.2 ± 0.04 and 0.31 ± 0.11), Klatovy in autumn 2018 (0.73 ± 0.31 and 0.56 ± 0.37), and Hodonin in autumn 2019 (0.28 ± 0.15 and 0.21 ± 0.07).

We found no significant differences ($P > 0.05$) in protein content of European pike-perch from with respect to farm, season, year, or presence of skin.

The nitrogen content was significantly lower ($P < 0.01$) in European pike-perch fillets without skin from Hodonin (spring 2019, 3.13 ± 0.05) and Klatovy (autumn 2018, 3.05 ± 0.09) than in those fillets with skin from Lnare in autumn 2019 (3.34 ± 0.06) and Blatna in autumn 2018 (3.32 ± 0.06) and 2019 (3.38 ± 0.05). We found no significant differences ($P > 0.05$) in nitrogen content of European pike-perch fillets with and without skin in a single sampling period.

Principal component analysis extracted the first two axes explaining 74.5% of the total variance. It did not explain the differences between functional traits fillets with and without skin, but a negative correlation of nitrogen and fat content with fish weight was revealed (Fig. 2). In the majority of samples of European pike-perch, PCA discriminated along the PC1 axis explaining 44.0% of the total variance in a cluster along the nitrogen gradient. Samples from FFPW USB (spring 2019) and Blatna (autumn 2019) showed a positive correlation of fat with fish weight. Samples from Klatovy (autumn 2018), Lnare (spring 2019), and Hodonin (spring 2019) were inconsistent and negatively correlated with nitrogen content. Redundancy analysis revealed little correlation of fat content with fish weight as an explanatory factor (first canonical axes explained 56.2% of variability, permutation test on all axes: pseudo- $F = 16.3$, $P < 0.002$). Weight was not considered the main factor in nitrogen content.

Northern pike

The basic chemical composition of northern pike fillets with and without skin are given in Table 2. Dry matter content was significantly lower ($P < 0.01$) in fillets with and without skin from Chlumec nad Cidlinou (spring

Table 1. Live weight and length of European pike-perch (*Sander lucioperca*) and chemical composition of fillets with skin and without skin.

Company	Year	Season	Weight [g] x ± SD (min–max)	Total length [cm] x ± SD (min–max)	Fillet	Dry matter [g · 100 g ⁻¹] x ± SD (min–max)	Ash [g · 100 g ⁻¹] x ± SD (min–max)	Fat [g · 100 g ⁻¹] x ± SD (min–max)	Protein [g · 100 g ⁻¹] x ± SD (min–max)	N [g · 100 g ⁻¹] x ± SD (min–max)
FFPW USB Vodnany	2018	Spring	1359.1 ± 274.9 (1015.0–2025.0)	51.6 ± 3.1 (48.0–57.0)	with skin	22.47 ± 0.33 ^{ab(A)} (21.77–22.94)	1.35 ± 0.62 ^{ab(A)} (1.07–3.21)	1.37 ± 0.50 ^{ab(C)} (0.83–2.59)	20.66 ± 0.35 ^{ab(A)} (20.18–21.47)	3.30 ± 0.06 ^{ab(A)} (3.23–3.43)
					without skin	21.86 ± 0.27 ^{ab(A)} (19.89–20.58)	1.26 ± 0.06 ^{ab(A)} (1.16–1.40)	0.69 ± 0.20 ^{ab(B)*} (0.24–0.97)	20.22 ± 0.27 ^{ab(A)} (21.28–22.27)	3.23 ± 0.03 ^{ab(A)} (3.18–3.29)
	2019	Spring	563.0 ± 119.6 (435.0–805.0)	39.1 ± 2.4 (36.5–44.0)	with skin	22.64 ± 0.26 ^{ab(A)} (22.36–23.17)	1.25 ± 0.04 ^{ab(A)} (1.21–1.32)	0.33 ± 0.08 ^{ab(A)} (0.17–0.42)	20.68 ± 0.27 ^{ab(A)} (20.16–21.07)	3.31 ± 0.04 ^{ab(A)} (3.23–3.37)
					without skin	22.26 ± 0.15 ^{ab(A)} (21.99–22.46)	1.24 ± 0.03 ^{ab(A)} (1.20–1.31)	0.37 ± 0.10 ^{ab(A)} (0.16–0.52)	20.37 ± 0.25 ^{ab(A)} (19.99–20.89)	3.26 ± 0.04 ^{ab(A)} (3.20–3.34)
Fishery Lnare	2018	Spring	437.5 ± 98.9 (330.0–660.0)	35.8 ± 2.1 (36.5–44.0)	with skin	21.72 ± 0.33 ^{ab(A)} (21.33–22.30)	1.17 ± 0.04 ^{ab(A)} (1.11–1.24)	0.14 ± 0.10 ^{ab(A)} (0.02–0.40)	20.38 ± 0.39 ^{ab(A)} (19.78–21.08)	3.26 ± 0.06 ^{ab(A)} (3.16–3.37)
					without skin	21.73 ± 0.25 ^{ab(A)} (21.14–22.00)	1.20 ± 0.03 ^{ab(A)} (1.13–1.24)	0.15 ± 0.14 ^{ab(A)} (0.05–0.57)	20.37 ± 0.31 ^{ab(A)} (19.76–20.81)	3.26 ± 0.05 ^{ab(A)} (3.16–3.33)
		Autumn	722.0 ± 78.0 (605.0–850.0)	42.3 ± 1.0 (40.5–43.5)	with skin	22.46 ± 0.54 ^{ab(A)} (20.95–22.98)	1.27 ± 0.04 ^{ab(AB)} (1.17–1.31)	0.44 ± 0.43 ^{ab(B)} (0.16–1.67)	20.48 ± 0.58 ^{ab(A)} (19.00–21.00)	3.28 ± 0.09 ^{ab(A)} (3.04–3.36)
					without skin	21.79 ± 0.56 ^{ab(A)} (20.49–22.44)	1.20 ± 0.12 ^{ab(A)} (0.95–1.48)	0.29 ± 0.18 ^{ab(B)} (0.12–0.76)	19.77 ± 0.47 ^{ab(A)} (18.67–20.27)	3.17 ± 0.07 ^{ab(A)} (2.99–3.24)
	2019	Spring	562.0 ± 43.6 (500.0–650.0)	38.1 ± 1.4 (36.5–41.0)	with skin	21.91 ± 0.59 ^{ab(A)} (20.62–22.92)	1.37 ± 0.08 ^{ab(B)} (1.23–1.47)	0.47 ± 0.19 ^{ab(B)} (0.32–0.89)	20.18 ± 0.41 ^{ab(A)} (19.49–20.74)	3.23 ± 0.07 ^{ab(A)} (3.12–3.32)
					without skin	21.40 ± 0.81 ^{ab(A)} (19.52–22.82)	1.38 ± 0.11 ^{ab(B)} (1.14–1.50)	0.48 ± 0.25 ^{ab(B)} (0.26–1.13)	19.75 ± 0.57 ^{ab(A)} (18.30–20.33)	3.17 ± 0.09 ^{ab(A)} (2.93–3.25)
		Autumn	1044.5 ± 146.0 (805.0–1305.0)	46.2 ± 2.2 (43.0–48.5)	with skin	22.71 ± 0.71 ^{ab(A)} (21.03–23.63)	1.22 ± 0.04 ^{ab(AB)} (1.13–1.30)	0.41 ± 0.14 ^{ab(B)} (0.20–0.63)	20.90 ± 0.39 ^{ab(A)} (20.19–21.60)	3.34 ± 0.06 ^{ab(A)} (3.23–3.46)
					without skin	22.34 ± 0.68 ^{ab(A)} (20.80–23.16)	1.18 ± 0.05 ^{ab(A)} (1.10–1.29)	0.34 ± 0.14 ^{ab(B)} (0.17–0.56)	20.44 ± 0.31 ^{ab(A)} (19.75–20.96)	3.27 ± 0.05 ^{ab(A)} (3.16–3.35)
Fishery Blatna	2018	Autumn	1230.0 ± 309.2 (765.0–1780.0)	47.3 ± 3.4 (42.0–53.5)	with skin	23.18 ± 0.42 ^{ab(A)} (22.55–23.77)	1.23 ± 0.09 ^{ab(A)} (1.16–1.47)	0.75 ± 0.35 ^{ab(C)} (0.26–1.36)	20.73 ± 0.40 ^{ab(A)} (20.03–21.60)	3.32 ± 0.06 ^{ab(A)} (3.20–3.46)
					without skin	22.54 ± 0.42 ^{ab(A)} (22.04–23.28)	1.28 ± 0.07 ^{ab(A)} (1.20–1.48)	0.54 ± 0.26 ^{ab(B)*} (0.24–1.11)	20.21 ± 0.30 ^{ab(A)} (19.86–20.78)	3.23 ± 0.05 ^{ab(A)} (3.18–3.33)
	2019	Autumn	627.5 ± 97.8 (385.0–815.0)	39.1 ± 2.8 (32.0–48.0)	with skin	23.07 ± 0.46 ^{ab(A)} (22.31–23.28)	1.30 ± 0.04 ^{ab(A)} (1.23–1.35)	0.21 ± 0.04 ^{ab(A)*} (0.04–0.37)	21.12 ± 0.29 ^{ab(A)} (20.69–21.67)	3.38 ± 0.05 ^{ab(A)} (3.31–3.47)
					without skin	22.66 ± 0.43 ^{ab(A)} (21.89–23.28)	1.25 ± 0.06 ^{ab(A)} (1.17–1.35)	0.31 ± 0.11 ^{ab(B)} (0.12–0.48)	20.63 ± 0.22 ^{ab(A)} (20.33–21.06)	3.30 ± 0.04 ^{ab(A)} (3.25–3.37)
Fishery Hodonin	2019	Spring	1210.0 ± 386.9 (660.0–1780.0)	49.3 ± 4.8 (41.0–54.5)	with skin	22.04 ± 0.19 ^{ab(A)} (21.72–22.30)	1.17 ± 0.04 ^{ab(A)} (1.11–1.22)	0.39 ± 0.18 ^{ab(B)} (0.21–0.72)	20.50 ± 0.38 ^{ab(A)} (20.03–20.92)	3.28 ± 0.06 ^{ab(A)} (3.20–3.35)
					without skin	21.46 ± 0.31 ^{ab(A)} (20.91–21.84)	1.20 ± 0.03 ^{ab(A)} (1.15–1.24)	0.26 ± 0.08 ^{ab(B)} (0.14–0.38)	19.59 ± 0.33 ^{ab(A)} (19.29–20.22)	3.13 ± 0.05 ^{ab(A)} (3.09–3.24)
		Autumn	1053.8 ± 112.0 (910.0–1270.0)	46.3 ± 0.9 (45.0–48.0)	with skin	21.97 ± 0.35 ^{ab(A)} (21.45–22.55)	1.24 ± 0.07 ^{ab(A)} (1.11–1.30)	0.28 ± 0.15 ^{ab(B)} (0.05–0.60)	20.44 ± 0.35 ^{ab(A)} (19.86–21.03)	3.27 ± 0.06 ^{ab(A)} (3.18–3.36)
					without skin	21.49 ± 0.29 ^{ab(A)} (21.12–22.0)	1.27 ± 0.06 ^{ab(A)} (1.16–1.36)	0.21 ± 0.07 ^{ab(A)*} (0.10–0.29)	19.83 ± 0.14 ^{ab(A)} (19.57–20.03)	3.17 ± 0.02 ^{ab(A)} (3.13–3.20)
Fishery Klatovy	2018	Autumn	1292.0 ± 128.6 (1150.0–1535.0)	49.3 ± 2.1 (46.5–53.0)	with skin	21.76 ± 0.72 ^{ab(A)} (20.71–22.83)	1.22 ± 0.11 ^{ab(A)} (1.05–1.43)	0.73 ± 0.31 ^{ab(B)} (0.1–1.37)	19.61 ± 0.61 ^{ab(A)} (18.47–20.88)	3.17 ± 0.10 ^{ab(A)} (2.95–3.34)
					without skin	21.24 ± 0.73 ^{ab(A)} (20.20–22.38)	1.29 ± 0.12 ^{ab(A)} (1.07–1.49)	0.56 ± 0.37 ^{ab(A)*} (0.21–1.57)	19.05 ± 0.58 ^{ab(A)} (18.12–19.80)	3.05 ± 0.09 ^{ab(A)} (2.90–3.17)

Data are mean ± standard deviation (minimum value–maximum value), $n = 10$. Values with different small letters in superscripts are significantly ($P < 0.01$) different among the locality groups. Values with different capital letters in superscripts are significantly ($P < 0.01$) different among the season groups in one locality. *Denotes significant differences among fillets with skin or fillets without skin values over one sampling ($P < 0.01$).

2018, 21.29 ± 0.65 and 21.39 ± 0.96), Klatovy (autumn 2019, 21.14 ± 0.62 and 21.35 ± 0.78) and Lnare (autumn 2018, 21.37 ± 0.92 and 21.21 ± 0.95) compared to those from FFPW USB in spring 2019 (23.47 ± 0.33 and 23.19 ± 0.41) and Tabor in spring 2019 (23.52 ± 0.82 and 23.22 ± 0.74). We found no significant differences ($P > 0.05$) between fillets with skin and those without skin in dry matter at a single sampling time.

The ash content was significantly lower ($P < 0.01$) in northern pike fillets with skin (1.17 ± 0.06) from Chlumec nad Cidlinou (autumn 2018) and in fillets with (1.13 ± 0.12) and without (1.16 ± 0.12) skin from Lnare (autumn 2019) compared to fillets with skin from FFPW USB in spring 2018 (1.52 ± 0.45) and from Tabor in autumn 2018 (1.42 ± 0.06). We found no significant differences ($P > 0.05$) in ash content of fillets with and without skin at a single sampling time.

The fat content was significantly lower ($P < 0.01$) in northern pike fillets with skin from Klatovy (autumn

2019, 0.28 ± 0.19), Lnare (spring 2018, 0.33 ± 0.19 ; autumn 2018, 0.45 ± 0.20 ; and autumn 2019, 0.41 ± 0.16), and Tabor (spring 2018, 0.43 ± 0.27), and in northern pike fillets without skin from Lnare (autumn 2018, 0.38 ± 0.28 and autumn 2019, 0.44 ± 0.21) and Klatovy (spring 2018, 0.39 ± 0.29 and autumn 2019, 0.40 ± 0.26) compared to other groups. We found significant differences ($P < 0.01$) between fillets with skin and fillets without skin in the fat content of northern pike from Klatovy in spring 2018 (0.64 ± 0.28 and 0.39 ± 0.29), Chlumec nad Cidlinou in autumn 2018 (0.68 ± 0.19 and 0.91 ± 0.32) and Fishery Lnare in spring 2018 (0.33 ± 0.19 and 0.63 ± 0.34).

No significant differences ($P > 0.05$) were observed in protein content of northern pike fillets with respect to farm, season, year, and presence/absence of skin.

The nitrogen content was significantly lower ($P < 0.01$) in northern pike fillets with (3.08 ± 0.08) and without (3.06 ± 0.06) skin from Chlumec nad Cidlinou (spring 2018) and fillets without skin (3.06 ± 0.11) from Blatna

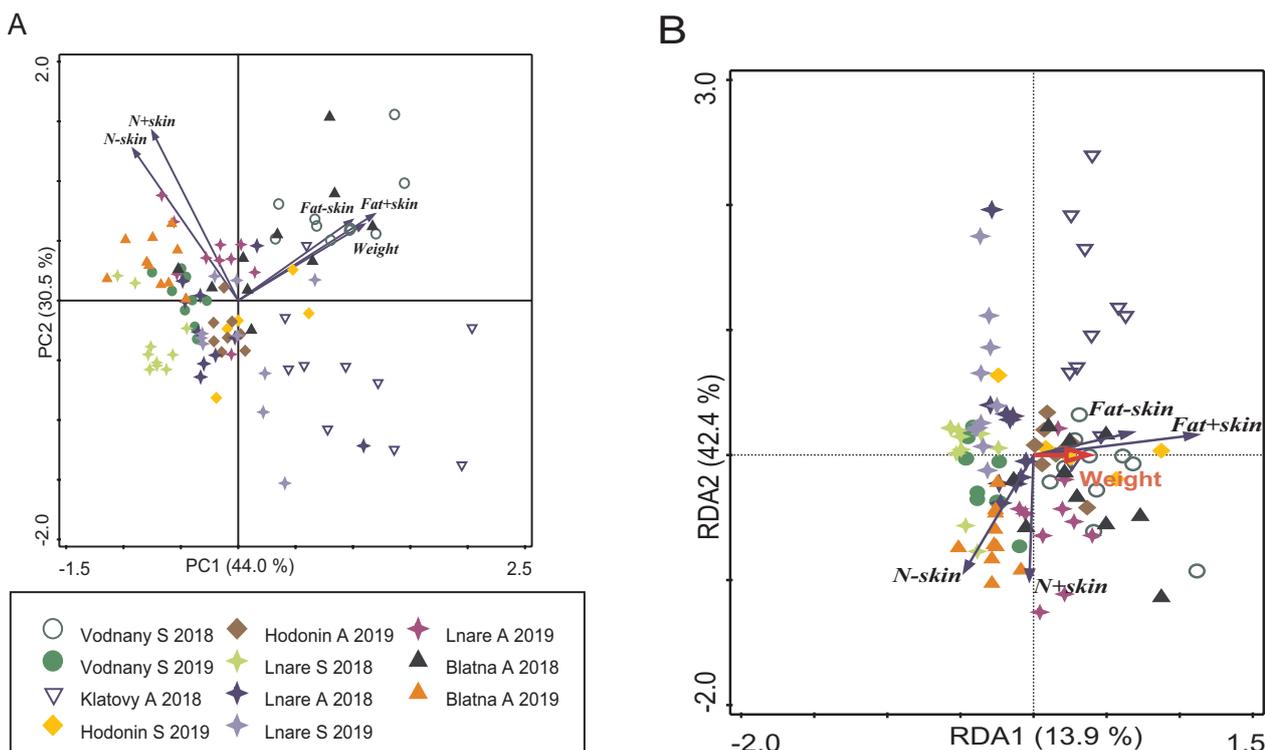


Figure 2. Ordination plots of sample distribution after principal component analysis (PCA) of functional traits as response variables (A) and redundancy analysis (RDA) (B) of functional traits as response variables and weight of European pike-perch, *Sander lucioperca* as explanatory variable. N+skin = nitrogen concentration in fillet with skin, N-skin = nitrogen concentration in fillet without skin, Fat+skin = fat percentage in fillet with skin, Fat-skin = fat percentage in fillet without skin, Weight = body weight of individual fish; abbreviations used in legend: S = spring sampling, A = autumn sampling). The length of the arrow reflects the power of the variable to differentiate the samples.

(autumn 2019) compared to fillets with skin from Chlumec nad Cidlinou (autumn 2019, 3.22 ± 0.07), Lnare (spring 2018, 3.23 ± 0.09), and fillets with and without skin from Tabor (autumn 2018, 3.22 ± 0.03 and 3.22 ± 0.05 and spring 2019, 3.25 ± 0.07 and 3.22 ± 0.07) and Klatovy (spring 2018, 3.24 ± 0.07 and 3.22 ± 0.06). We found no significant differences ($P > 0.05$) in nitrogen content of northern pike fillets with and without skin at a single sampling.

The PCA distribution of samples in northern pike did not show separate clusters, and the samples were discriminated against both axes (Fig. 3). The first two axes explained 75.42% of total variance. No association of nitrogen content with farm source or season or in functional traits of fillets with and without skin was observed. The RDA showed that fish weight slightly positively explained the functional traits (first two canonical axes explained 53.79% of variability, permutation test on all axes, pseudo- $F = 11.5$, $P < 0.002$).

Sheatfish

The basic chemical composition of sheatfish fillets with and without skin is provided in Table 3. The dry matter content was significantly lower ($P < 0.01$) in sheatfish

fillets with skin from FFPW USB (spring 2018, 19.07 ± 0.57 and spring 2019, 19.91 ± 0.91), from Blatna (autumn 2018, 21.97 ± 0.58 and autumn 2019, 20.77 ± 2.06) and those without skin from FFPW USB (spring 2018, 18.84 ± 0.58) and Blatna (autumn 2018, 21.15 ± 0.36 and autumn 2019, 19.86 ± 1.81) compared to the remaining samples. We found significant differences ($P < 0.01$) in dry matter of sheatfish fillets with skin and those without from FFPW USB in spring 2019 (19.91 ± 0.91 and 25.64 ± 2.49) and from Chlumec nad Cidlinou in spring 2019 (30.56 ± 2.01 and 27.25 ± 2.22).

The ash content was significantly lower ($P < 0.01$) in sheatfish fillets without skin from Chlumec nad Cidlinou (spring 2019, 0.98 ± 0.07 and autumn 2019, 0.97 ± 0.09), Klatovy (autumn 2019, 0.98 ± 0.05), and Hodonin (spring 2019, 0.98 ± 0.08) and fillets with skin from Chlumec nad Cidlinou (spring 2019, 0.90 ± 0.05 and autumn 2019, 0.92 ± 0.06), Blatna (autumn 2019, 0.95 ± 0.05), Klatovy (autumn 2019, 0.92 ± 0.05), and Hodonin (spring 2019, 0.92 ± 0.08 and autumn 2019, 0.95 ± 0.05) compared to sheatfish fillets without skin (1.15 ± 0.05) from Blatna in autumn 2018. We found significant differences ($P < 0.01$) in ash content of fillets with skin and those without skin from Chlumec nad Cidlinou in spring 2019 (0.90 ± 0.05 and 0.98 ± 0.07).

The fat content was significantly lower ($P < 0.01$) in sheatfish fillets with skin from FFPW USB (spring 2018,

Table 2. Live weight and length of northern pike (*Esox lucius*) and chemical composition of fillets with skin and without skin.

Company	Year	Season	Weight [g] $\bar{x} \pm SD$ (min–max)	Total length [cm] $\bar{x} \pm SD$ (min–max)	Fillet	Dry matter	Ash	Fat	Protein	N	
						[g · 100 g ⁻¹] $\bar{x} \pm SD$ (min–max)	[g · 100 g ⁻¹] $\bar{x} \pm SD$ (min–max)	[g · 100 g ⁻¹] $\bar{x} \pm SD$ (min–max)	[g · 100 g ⁻¹] $\bar{x} \pm SD$ (min–max)	[g · 100 g ⁻¹] $\bar{x} \pm SD$ (min–max)	
FFPW USB Vodnany	2018	Spring	1721.4 ± 469.1 (1260.0–2550)	63.1 ± 4.5 (59.0–74.0)	with skin	21.63 ± 1.03 ^{ab(A)} (19.79–23.36)	1.52 ± 0.45 ^{ab(A)} (1.17–2.60)	1.13 ± 0.63 ^{c(B)} (0.49–2.61)	19.73 ± 0.61 ^{a(A)} (18.59–20.52)	3.16 ± 0.10 ^{ab(A)} (2.97–3.28)	
					without skin	21.78 ± 1.12 ^{ab(A)} (19.84–23.59)	1.30 ± 0.14 ^{ab(A)} (1.20–1.70)	1.18 ± 0.40 ^{c(B)} (0.67–1.78)	19.79 ± 0.60 ^{a(A)} (18.57–20.79)	3.17 ± 0.10 ^{ab(A)} (2.97–3.33)	
		2019	Spring	1609.0 ± 473.6 (945.0–2470.0)	55.5 ± 4.0 (48.0–60.0)	with skin	23.47 ± 0.33 ^{ab(A)} (22.84–24.05)	1.27 ± 0.06 ^{ab(A)} (1.17–1.40)	0.72 ± 0.30 ^{ab(A)} (0.26–1.31)	20.12 ± 0.26 ^{a(A)} (19.66–20.49)	3.21 ± 0.04 ^{ab(A)} (3.15–3.28)
						without skin	23.19 ± 0.41 ^{ab(A)} (22.53–23.90)	1.31 ± 0.07 ^{ab(A)} (1.21–1.47)	0.59 ± 0.22 ^{ab(A)} (0.27–0.99)	20.00 ± 0.30 ^{a(A)} (19.51–20.51)	3.20 ± 0.05 ^{ab(A)} (3.12–3.28)
	Fishery Chlumec nad Cidlinou	2018	Spring	1728.5 ± 464.81 (880.0–2415.0)	64.9 ± 5.5 (53.0–72.0)	with skin	21.29 ± 0.65 ^{ab(A)} (20.30–22.33)	1.24 ± 0.15 ^{ab(A)} (1.06–1.64)	0.60 ± 0.36 ^{ab(A)} (0.18–1.48)	19.24 ± 0.48 ^{a(A)} (18.21–20.03)	3.08 ± 0.08 ^{ab(A)} (2.91–3.20)
						without skin	21.39 ± 0.96 ^{ab(A)} (20.27–23.40)	1.24 ± 0.05 ^{ab(A)} (1.13–1.32)	0.61 ± 0.47 ^{ab(A)} (0.06–1.52)	19.10 ± 0.37 ^{a(A)} (18.37–19.63)	3.06 ± 0.06 ^{ab(A)} (2.94–3.14)
Autumn			1459.0 ± 457.4 (530.0–2405.0)	57.1 ± 5.9 (43.0–67.5)	with skin	22.10 ± 0.55 ^{ab(A)} (20.96–22.99)	1.17 ± 0.06 ^{ab(A)} (1.07–1.29)	0.68 ± 0.19 ^{ab(A)*} (0.39–1.08)	19.62 ± 0.47 ^{a(A)} (18.65–20.34)	3.14 ± 0.07 ^{ab(A)B)} (2.98–3.25)	
					without skin	22.24 ± 0.79 ^{ab(A)} (20.80–23.20)	1.24 ± 0.07 ^{ab(A)} (1.14–1.34)	0.91 ± 0.32 ^{ab(B)} (0.45–1.45)	19.51 ± 0.52 ^{a(A)} (18.33–19.99)	3.12 ± 0.08 ^{ab(A)B)} (2.93–3.20)	
2019		Spring	1125.0 ± 271.1 (615.0–1545.0)	55.0 ± 4.3 (46.0–63.0)	with skin	21.92 ± 0.51 ^{ab(A)} (20.54–23.27)	1.34 ± 0.09 ^{ab(A)} (1.28–1.61)	1.02 ± 0.39 ^{ab(B)} (0.46–1.78)	19.56 ± 0.57 ^{a(A)} (18.49–20.72)	3.13 ± 0.09 ^{ab(A)B)} (2.96–3.31)	
					without skin	21.64 ± 1.02 ^{ab(A)} (20.00–23.24)	1.35 ± 0.07 ^{ab(A)} (1.26–1.51)	0.96 ± 0.38 ^{ab(B)} (0.48–1.77)	19.39 ± 0.58 ^{a(A)} (18.38–20.64)	3.10 ± 0.09 ^{ab(A)B)} (2.94–3.30)	
	Autumn	1653.5 ± 278.5 (1240.0–2075.0)	59.5 ± 3.6 (54.5–66.0)	with skin	22.51 ± 0.51 ^{ab(A)} (21.55–23.00)	1.24 ± 0.02 ^{ab(A)} (1.21–1.28)	0.59 ± 0.20 ^{ab(A)} (0.25–0.95)	20.13 ± 0.42 ^{a(A)} (19.34–20.59)	3.22 ± 0.07 ^{ab(B)} (3.09–3.29)		
				without skin	22.62 ± 0.54 ^{ab(A)} (21.72–23.44)	1.29 ± 0.04 ^{ab(A)} (1.23–1.35)	0.65 ± 0.26 ^{ab(A)} (0.38–1.05)	20.00 ± 0.27 ^{a(A)} (19.49–20.54)	3.20 ± 0.04 ^{ab(A)B)} (3.12–3.29)		
Fishery Blatna	2018	Autumn	1143.3 ± 361.2 (685.0–1820.0)	54.7 ± 4.4 (46.5–60.0)	with skin	22.77 ± 0.41 ^{ab(A)} (22.12–23.45)	1.33 ± 0.10 ^{ab(A)} (1.33–1.52)	0.67 ± 0.20 ^{ab(A)} (0.38–1.01)	20.03 ± 0.26 ^{a(A)} (19.60–20.32)	3.20 ± 0.04 ^{ab(A)B)} (3.14–3.25)	
					without skin	22.68 ± 0.43 ^{ab(A)} (21.99–23.31)	1.37 ± 0.10 ^{ab(A)} (1.20–1.55)	0.71 ± 0.38 ^{ab(A)} (0.14–1.33)	19.87 ± 0.18 ^{a(A)} (19.60–20.12)	3.18 ± 0.03 ^{ab(A)} (3.14–3.22)	
		2019	Autumn	1141.5 ± 322.9 (675.0–1850.0)	56.4 ± 4.31 (48.5–64.0)	with skin	21.50 ± 0.78 ^{ab(A)} (19.47–22.57)	1.21 ± 0.04 ^{ab(A)} (1.14–1.27)	0.65 ± 0.29 ^{ab(A)} (0.17–1.06)	19.35 ± 0.70 ^{a(A)} (18.13–20.67)	3.10 ± 0.11 ^{ab(A)} (2.90–3.31)
						without skin	21.60 ± 0.95 ^{ab(A)} (19.11–22.76)	1.28 ± 0.05 ^{ab(A)} (1.22–1.38)	0.80 ± 0.42 ^{ab(A)} (0.25–1.88)	19.13 ± 0.65 ^{a(A)} (17.66–19.69)	3.06 ± 0.11 ^{a(A)} (2.83–3.15)
	2019	Spring	1378.0 ± 384.2 (865.0–2030.0)	59.3 ± 5.8 (50.5–68.5)	with skin	21.83 ± 0.70 ^{ab(A)} (20.55–22.87)	1.28 ± 0.05 ^{ab(A)} (1.20–1.36)	0.62 ± 0.19 ^{ab(B)} (0.21–0.88)	19.74 ± 0.77 ^{a(A)} (18.16–21.03)	3.16 ± 0.12 ^{ab(A)} (2.91–3.36)	
					without skin	21.62 ± 0.59 ^{ab(A)} (20.30–22.19)	1.28 ± 0.06 ^{ab(A)} (1.19–1.38)	0.53 ± 0.22 ^{ab(B)} (0.24–1.05)	19.43 ± 0.61 ^{a(A)} (18.21–20.33)	3.11 ± 0.10 ^{ab(A)} (2.91–3.26)	
Autumn		1450.6 ± 240.0 (1075.0–1790.0)	59.8 ± 4.4 (52.0–67.5)	with skin	21.79 ± 0.60 ^{ab(A)} (20.62–22.58)	1.31 ± 0.04 ^{ab(A)} (1.27–1.42)	0.34 ± 0.14 ^{ab(A)*} (0.11–0.57)	20.23 ± 0.68 ^{a(A)} (18.65–20.83)	3.24 ± 0.11 ^{ab(A)} (2.98–3.33)		
				without skin	22.05 ± 0.81 ^{ab(A)} (20.74–23.28)	1.39 ± 0.05 ^{ab(A)} (1.27–1.45)	0.56 ± 0.29 ^{ab(B)} (0.17–1.10)	20.00 ± 0.61 ^{a(A)} (19.13–20.94)	3.20 ± 0.10 ^{ab(A)} (3.06–3.35)		
Fishery Klatovy	2018	Spring	1360.0 ± 551.1 (725.0–2795.0)	57.1 ± 5.9 (48.5–70.0)	with skin	22.26 ± 0.87 ^{ab(A)} (20.98–23.92)	1.28 ± 0.05 ^{ab(A)} (1.18–1.35)	0.64 ± 0.28 ^{ab(B)} (0.14–2.28)	20.23 ± 0.44 ^{a(A)} (19.44–20.80)	3.24 ± 0.07 ^{ab(A)} (3.11–3.33)	
					without skin	22.04 ± 0.60 ^{ab(A)} (21.11–23.21)	1.29 ± 0.04 ^{ab(A)} (1.22–1.36)	0.39 ± 0.29 ^{ab(A)*} (0.12–1.19)	20.13 ± 0.40 ^{a(A)} (19.19–20.63)	3.22 ± 0.06 ^{ab(A)} (3.07–3.30)	
		Autumn	1537.5 ± 461.4 (510.0–2200.0)	59.0 ± 7.4 (42.0–70.0)	with skin	22.16 ± 1.19 ^{ab(A)} (19.10–23.93)	1.23 ± 0.31 ^{ab(A)} (0.59–1.59)	0.50 ± 0.17 ^{ab(B)} (0.14–0.68)	19.75 ± 0.89 ^{a(A)} (17.43–21.06)	3.16 ± 0.14 ^{ab(A)} (2.79–3.37)	
					without skin	22.08 ± 1.34 ^{ab(A)} (18.48–23.38)	1.30 ± 0.24 ^{ab(A)} (0.82–1.68)	0.62 ± 0.19 ^{ab(B)} (0.09–0.83)	19.70 ± 0.81 ^{a(A)} (17.53–20.68)	3.15 ± 0.13 ^{ab(A)} (2.80–3.31)	
	2019	Autumn	1375.0 ± 385.9 (755.0–2115.0)	57.4 ± 4.9 (50.5–66.5)	with skin	21.14 ± 0.62 ^{ab(A)} (19.77–21.93)	1.26 ± 0.05 ^{ab(A)} (1.18–1.34)	0.28 ± 0.19 ^{ab(A)} (0.08–0.58)	19.43 ± 0.47 ^{a(A)} (18.70–20.22)	3.11 ± 0.07 ^{ab(A)} (2.99–3.24)	
					without skin	21.35 ± 0.78 ^{ab(A)} (19.56–22.24)	1.36 ± 0.10 ^{ab(A)} (1.21–1.50)	0.40 ± 0.26 ^{ab(A)} (0.08–0.90)	19.44 ± 0.42 ^{a(A)} (18.43–19.81)	3.11 ± 0.07 ^{ab(A)} (2.95–3.17)	
Fishery Lnare	2018	Spring	1322.5 ± 431.1 (615.0–1960.0)	57.2 ± 7.8 (44.5–68.5)	with skin	22.32 ± 0.94 ^{ab(A)} (19.69–23.09)	1.27 ± 0.10 ^{ab(A)} (1.16–1.51)	0.33 ± 0.19 ^{ab(A)*} (0.07–0.67)	20.17 ± 0.54 ^{a(A)} (19.16–21.07)	3.23 ± 0.09 ^{ab(A)} (3.07–3.37)	
					without skin	22.61 ± 0.78 ^{ab(A)} (20.59–23.37)	1.32 ± 0.13 ^{ab(A)} (1.18–1.67)	0.63 ± 0.34 ^{ab(B)} (0.27–1.35)	19.97 ± 0.60 ^{a(A)} (18.48–20.83)	3.20 ± 0.10 ^{ab(A)} (2.96–3.33)	
		Autumn	1327.0 ± 524.1 (805.0–2105.0)	58.9 ± 7.5 (50.0–70.0)	with skin	21.37 ± 0.92 ^{ab(A)} (19.44–22.80)	1.35 ± 0.07 ^{ab(A)} (1.25–1.50)	0.45 ± 0.20 ^{ab(A)} (0.14–0.99)	19.65 ± 0.42 ^{a(A)} (19.14–20.54)	3.13 ± 0.07 ^{ab(A)} (3.06–3.29)	
					without skin	21.21 ± 0.95 ^{ab(A)} (19.15–22.73)	1.35 ± 0.11 ^{ab(A)} (1.09–1.53)	0.38 ± 0.28 ^{ab(A)} (0.12–0.93)	19.42 ± 0.56 ^{a(A)} (18.35–20.52)	3.11 ± 0.09 ^{ab(A)} (2.94–3.28)	
	2019	Spring	981.0 ± 434.4 (445.0–1650.0)	49.7 ± 6.2 (40.0–58.5)	with skin	22.65 ± 0.71 ^{ab(A)} (21.23–23.56)	1.31 ± 0.05 ^{ab(A)} (1.22–1.38)	0.72 ± 0.30 ^{ab(B)} (0.22–1.27)	19.48 ± 0.27 ^{a(A)} (19.47–20.24)	3.17 ± 0.04 ^{ab(A)} (3.12–3.24)	
					without skin	22.72 ± 0.87 ^{ab(A)} (21.00–23.86)	1.37 ± 0.06 ^{ab(A)} (1.29–1.49)	0.71 ± 0.36 ^{ab(B)} (0.26–1.52)	19.85 ± 0.49 ^{a(A)} (19.07–20.66)	3.18 ± 0.08 ^{ab(A)} (3.05–3.31)	
2019	Autumn	1136.0 ± 209.9 (715.0–1435.0)	54.5 ± 3.01 (47.0–58.0)	with skin	22.02 ± 0.68 ^{ab(A)} (20.50–23.03)	1.13 ± 0.12 ^{ab(A)} (0.99–1.28)	0.41 ± 0.16 ^{ab(A)} (0.15–0.70)	19.95 ± 0.34 ^{a(A)} (19.51–20.46)	3.19 ± 0.05 ^{ab(A)} (3.12–3.27)		
				without skin	22.05 ± 0.61 ^{ab(A)} (21.20–23.05)	1.16 ± 0.12 ^{ab(A)} (0.99–1.32)	0.44 ± 0.21 ^{ab(A)} (0.12–0.78)	20.02 ± 0.29 ^{a(A)} (19.44–20.45)	3.20 ± 0.05 ^{ab(A)} (3.11–3.27)		

Table 2 continues on next page.

Table 2. cont.

Company	Year	Season	Weight [g] x ± SD (min–max)	Total length [cm] x ± SD (min–max)	Fillet	Dry matter [g · 100 g ⁻¹] x ± SD (min–max)	Ash [g · 100 g ⁻¹] x ± SD (min–max)	Fat [g · 100 g ⁻¹] x ± SD (min–max)	Protein [g · 100 g ⁻¹] x ± SD (min–max)	N [g · 100 g ⁻¹] x ± SD (min–max)
Fishery Tabor	2018	Spring	937.0 ± 202.2 (685.0–1315.0)	52.3 ± 4.0 (46.0–60.0)	with skin	21.69 ± 0.58 ^{ab(A)} (20.94–22.75)	1.28 ± 0.06 ^{ab(A)} (1.18–1.38)	0.43 ± 0.27 ^{a(A)} (0.06–1.00)	19.82 ± 0.29 ^{a(A)} (19.38–20.28)	3.17 ± 0.05 ^{ab(A)} (3.10–3.24)
					without skin	21.80 ± 0.65 ^{ab(A)} (20.69–22.71)	1.34 ± 0.07 ^{ab(A)} (1.22–1.48)	0.51 ± 0.24 ^{ab(B)} (0.08–0.95)	19.53 ± 0.36 ^{a(A)} (18.74–20.04)	3.12 ± 0.06 ^{ab(A)} (3.00–3.21)
		Autumn	677.0 ± 80.7 (560.0–800.0)	46.0 ± 1.6 (43.0–48.5)	with skin	22.42 ± 0.45 ^{ab(A)} (21.38–22.97)	1.42 ± 0.06 ^{ab(A)} (1.35–1.57)	0.47 ± 0.13 ^{ab(B)} (0.30–0.74)	20.10 ± 0.18 ^{a(A)} (19.83–20.44)	3.22 ± 0.03 ^{ab(A)} (3.17–3.27)
					without skin	22.57 ± 0.45 ^{ab(A)} (21.46–23.03)	1.38 ± 0.07 ^{ab(A)} (1.30–1.50)	0.55 ± 0.20 ^{ab(B)} (0.20–0.92)	20.07 ± 0.31 ^{a(A)} (19.48–20.58)	3.22 ± 0.05 ^{ab(A)} (3.12–3.29)
	2019	Spring	1565.0 ± 889.1 (760.0–3980.0)	57.7 ± 9.7 (48.0–82.0)	with skin	23.52 ± 0.82 ^{ab(A)} (22.50–25.00)	1.26 ± 0.05 ^{ab(A)} (1.18–1.34)	0.91 ± 0.48 ^{ab(B)} (0.34–2.02)	20.30 ± 0.43 ^{a(A)} (19.36–20.87)	3.25 ± 0.07 ^{ab(A)} (3.10–3.34)
					without skin	23.22 ± 0.74 ^{ab(A)} (21.92–24.39)	1.26 ± 0.06 ^{ab(A)} (1.16–1.36)	0.74 ± 0.42 ^{ab(B)} (0.20–1.62)	20.05 ± 0.41 ^{a(A)} (19.45–20.81)	3.22 ± 0.07 ^{ab(A)} (3.11–3.33)
		Autumn	981.0 ± 269.9 (540.0–1650.0)	51.0 ± 4.2 (43.5–60.0)	with skin	21.84 ± 0.38 ^{ab(A)} (21.07–22.50)	1.35 ± 0.13 ^{ab(A)} (1.20–1.57)	0.68 ± 0.22 ^{ab(B)} (0.27–1.13)	19.94 ± 0.39 ^{a(A)} (19.37–20.37)	3.19 ± 0.06 ^{ab(A)} (3.10–3.26)
					without skin	21.86 ± 0.45 ^{ab(A)} (20.94–22.57)	1.37 ± 0.14 ^{ab(A)} (1.22–1.63)	0.79 ± 0.19 ^{ab(B)} (0.60–1.18)	19.71 ± 0.28 ^{a(A)} (19.38–20.16)	3.15 ± 0.05 ^{ab(A)} (3.10–3.23)

Data are mean ± standard deviation (minimum value–maximum value), $n = 10$. Values with different small letters in superscripts are significantly ($P < 0.05$) different among the locality groups. Values with different capital letters in superscripts are significantly ($P < 0.05$) different among the season groups in one locality. *Denotes significant differences among fillets with skin or fillets without skin values over one sampling ($P < 0.01$).

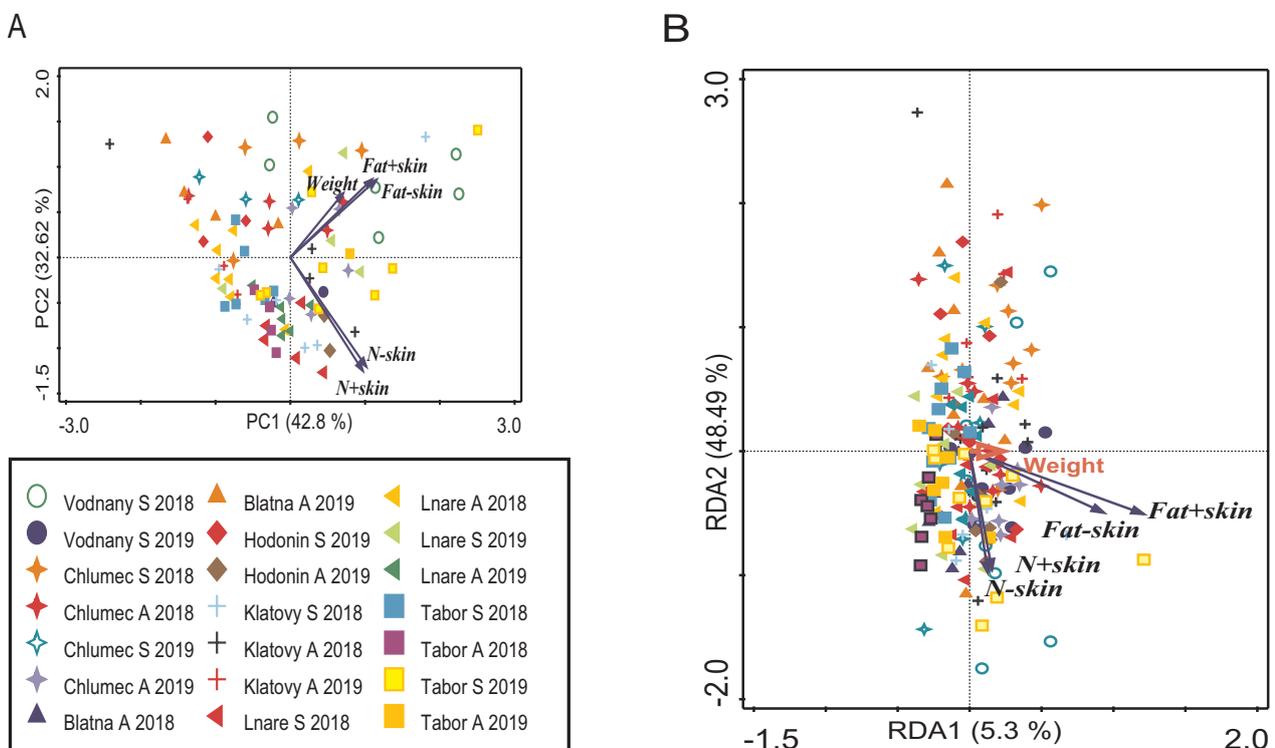


Figure 3. Ordination plots of sample distribution after principal component analysis (PCA) of functional traits as response variables (A) and redundancy analysis (RDA) (B) of functional traits as response variables and weight of northern pike, *Esox lucius* as explanatory variable. N+skin = nitrogen concentration in fillet with skin, N-skin = nitrogen concentration in fillet without skin, Fat+skin = fat percentage in fillet with skin, Fat-skin = fat percentage in fillet without skin, Weight = body weight of individual fish; abbreviations used in legend: S = spring sampling, A = autumn sampling). The length of the arrow reflects the power of the variable to differentiate the samples. Sample scores were limited to 90 points for better fit.

1.17 ± 0.36 and spring 2019, 0.74 ± 0.54) and Blatna (autumn 2018, 1.83 ± 0.72 and autumn 2019, 2.42 ± 2.34) and without skin from FFPW USB (spring 2018, 0.79 ± 0.31 and spring 2019, 0.54 ± 0.48) and Blatna (autumn 2018, 1.20 ± 0.59 and autumn 2019, 1.53 ± 2.16) compared to other groups. We found significant differences

($P < 0.01$) in fat content of fillets with and without skin in sheatfish from FFPW USB in spring 2018 (1.17 ± 0.36 and 0.79 ± 0.31), Chlumec nad Cidlinou in spring 2019 (13.41 ± 2.67 and 9.33 ± 2.89), Blatna in autumn 2019 (2.42 ± 2.34 and 1.53 ± 2.16), and Klatovy in autumn 2019 (8.63 ± 2.80 and 6.92 ± 2.41).

Table 3. Live weight and length of sheatfish (*Silurus glanis*) and chemical composition of fillets with skin and without skin.

Company	Year	Season	Weight [g] x ± SD (min–max)	Total length [cm] x ± SD (min–max)	Fillet	Dry matter [g · 100 g ⁻¹] x ± SD (min–max)	Ash [g · 100 g ⁻¹] x ± SD (min–max)	Fat [g · 100 g ⁻¹] x ± SD (min–max)	Protein [g · 100 g ⁻¹] x ± SD (min–max)	N [g · 100 g ⁻¹] x ± SD (min–max)
FFPW USB Vodnany	2018	Spring	900.0 ± 76.9 (750.0–1000.0)	50.5 ± 1.2 (48.0–53.0)	with skin	19.07 ± 0.57 ^(A) (18.22–20.27)	0.99 ± 0.01 ^(A) (0.97–1.00)	1.17 ± 0.36 ^(B) (16.42–17.61)	17.03 ± 0.36 ^(A) (16.42–17.61)	2.73 ± 0.06 ^(A) (2.63–2.82)
					without skin	18.84 ± 0.58 ^(A) (17.72–20.02)	1.03 ± 0.04 ^(A) (0.98–1.13)	0.79 ± 0.31 ^{(A)*} (0.45–1.40)	16.88 ± 0.51 ^(A) (15.69–17.60)	2.70 ± 0.08 ^(A) (2.51–2.82)
	2019	Spring	744.0 ± 161.0 (505.0–985.0)	48.8 ± 3.1 (44.0–53.0)	with skin	19.91 ± 0.91 ^(A) (18.83–21.38)	1.04 ± 0.74 ^(A) (0.98–1.09)	0.74 ± 0.54 ^(A) (0.16–1.79)	17.52 ± 0.69 ^(A) (16.58–18.62)	2.80 ± 0.11 ^(A) (2.65–2.98)
					without skin	25.64 ± 2.49 ^{(B)*} (21.64–31.63)	1.07 ± 0.04 ^(A) (0.95–1.12)	0.54 ± 0.48 ^(A) (0.16–1.48)	17.52 ± 0.70 ^(A) (18.40–20.79)	2.80 ± 0.11 ^(A) (2.68–3.01)
Fishery Chlumec nad Cidlinou	2018	Autumn	2027.0 ± 1170.9 (885.0–4115.0)	59.95 ± 9.31 (48.0–75.5)	with skin	24.82 ± 2.42 ^(A) (22.52–30.48)	0.99 ± 0.07 ^(A) (0.89–1.07)	6.61 ± 2.98 ^(A) (3.06–13.33)	16.59 ± 0.58 ^(A) (15.71–17.67)	2.66 ± 0.09 ^(A) (2.51–2.83)
					without skin	24.82 ± 3.45 ^(A) (20.67–30.38)	1.05 ± 0.08 ^(A) (0.84–1.12)	7.21 ± 4.00 ^(A) (3.08–15.02)	16.35 ± 0.67 ^(A) (14.67–17.18)	2.62 ± 0.11 ^(A) (2.35–2.75)
	2019	Spring	3220.0 ± 426.3 (2435.0–3725.0)	76.9 ± 3.6 (70.0–82.0)	with skin	30.56 ± 2.01 ^(C) (26.27–32.55)	0.90 ± 0.05 ^{(A)*} (0.81–0.98)	13.41 ± 2.67 ^(B) (7.71–16.50)	16.31 ± 0.61 ^(A) (15.73–17.66)	2.62 ± 0.10 ^(A) (2.52–2.83)
					without skin	27.25 ± 2.22 ^{(B)*} (23.25–30.68)	0.98 ± 0.07 ^(A) (0.83–1.06)	9.33 ± 2.89 ^{(B)*} (4.76–13.93)	17.33 ± 0.65 ^(A) (16.12–18.00)	2.77 ± 0.01 ^(A) (2.58–2.88)
		Autumn	3892.5 ± 732.0 (2515.0–4815.0)	77.8 ± 6.8 (55.5–91.0)	with skin	25.48 ± 1.32 ^(A) (23.46–27.19)	0.92 ± 0.06 ^(A) (0.82–1.06)	7.60 ± 1.62 ^(A) (5.16–10.03)	16.33 ± 0.62 ^(A) (14.95–17.22)	2.61 ± 0.10 ^(A) (2.39–2.76)
					without skin	24.24 ± 1.60 ^(A) (22.31–27.34)	0.97 ± 0.09 ^(A) (0.85–1.15)	5.93 ± 1.65 ^(A) (2.91–8.68)	16.77 ± 0.44 ^(A) (16.05–17.66)	2.68 ± 0.07 ^(A) (2.57–2.81)
Fishery Blatna	2018	Autumn	2552.5 ± 1290.3 (1005.0–4870.0)	72.0 ± 12.3 (55.5–91.0)	with skin	21.97 ± 0.58 ^(A) (21.16–23.09)	1.08 ± 0.13 ^(A) (0.76–1.29)	1.83 ± 0.72 ^(A) (1.04–3.03)	18.23 ± 0.32 ^(A) (17.60–18.67)	2.92 ± 0.05 ^(A) (2.82–2.99)
					without skin	21.15 ± 0.36 ^(A) (20.42–21.77)	1.15 ± 0.05 ^(B) (1.08–1.26)	1.20 ± 0.59 ^(A) (0.39–2.52)	18.12 ± 0.36 ^(A) (17.60–18.62)	2.90 ± 0.06 ^(A) (2.82–2.98)
	2019	Autumn	2527.5 ± 1481.6 (515.0–6115.0)	72.4 ± 11.01 (60.0–100.0)	with skin	20.77 ± 2.06 ^(A) (19.11–26.50)	0.95 ± 0.05 ^(A) (0.87–1.03)	2.42 ± 2.34 ^(A) (0.81–9.05)	17.26 ± 0.64 ^(A) (16.17–18.15)	2.76 ± 0.10 ^(A) (2.59–2.90)
					without skin	19.86 ± 1.81 ^(A) (18.57–24.94)	1.01 ± 0.07 ^(A) (0.93–1.12)	1.53 ± 2.16 ^{(A)*} (0.22–7.71)	17.21 ± 0.33 ^(A) (16.65–17.59)	2.75 ± 0.05 ^(A) (2.66–2.81)
Fishery Hodonin	2019	Spring	5892.9 ± 1055.4 (4805.0–7780.0)	93.3 ± 6.8 (87.0–105.0)	with skin	26.09 ± 3.05 ^(A) (21.57–30.37)	0.92 ± 0.08 ^(A) (0.83–1.04)	7.94 ± 3.79 ^(A) (2.51–13.69)	17.35 ± 0.81 ^(A) (16.00–18.42)	2.78 ± 0.13 ^(A) (2.56–2.95)
					without skin	25.17 ± 3.19 ^(A) (21.14–29.78)	0.98 ± 0.08 ^(A) (0.88–1.11)	6.84 ± 3.78 ^(A) (2.39–12.97)	17.52 ± 0.74 ^(A) (16.17–0.74)	2.80 ± 0.12 ^(A) (2.59–2.92)
	Autumn	4979.0 ± 1798.8 (3120.0–9265.0)	83.5 ± 9.5 (72.0–101.0)	with skin	27.07 ± 2.47 ^(A) (23.18–32.86)	0.95 ± 0.05 ^(A) (0.83–1.01)	8.95 ± 3.01 ^(A) (4.60–16.57)	17.06 ± 0.62 ^(A) (15.80–18.04)	2.73 ± 0.10 ^(A) (2.53–2.89)	
				without skin	25.64 ± 2.49 ^(A) (21.64–31.63)	1.03 ± 0.05 ^(A) (0.95–1.13)	7.11 ± 2.72 ^(A) (3.16–13.82)	17.29 ± 0.54 ^(A) (16.21–18.08)	2.77 ± 0.09 ^(A) (2.59–2.89)	
Fishery Klatovy	2019	Autumn	5220.0 ± 2247.8 (1955.0–8615.0)	86.7 ± 13.9 (64.0–105.0)	with skin	26.66 ± 2.08 ^(A) (22.77–30.97)	0.92 ± 0.05 ^(A) (0.84–1.00)	8.63 ± 2.80 ^(A) (4.16–13.10)	16.99 ± 0.72 ^(A) (15.61–18.51)	2.72 ± 0.12 ^(A) (2.50–2.96)
					without skin	26.04 ± 2.44 ^(A) (22.14–31.28)	0.98 ± 0.05 ^(A) (0.90–1.05)	6.92 ± 2.41 ^{(A)*} (3.23–11.10)	17.17 ± 0.46 ^(A) (16.17–17.74)	2.75 ± 0.07 ^(A) (2.59–2.84)

Data are mean ± standard deviation (minimum value–maximum value), $n = 10$. Values with different small letters in superscripts are significantly ($P < 0.05$) different among the locality groups. Values with different capital letters in superscripts are significantly ($P < 0.05$) different among the season groups in one locality. *Denotes significant differences among fillets with skin or fillets without skin values over one sampling ($P < 0.01$).

There were no significant differences ($P > 0.05$) in protein content of sheatfish fillets of different farms, seasons, years, or with/without skin.

The nitrogen content was significantly lower ($P < 0.01$) in sheatfish fillets with skin from Chlumec nad Cidlinou (spring 2019, 2.62 ± 0.10 and autumn 2019, 2.61 ± 0.10), and fillets without skin from Chlumec nad Cidlinou (autumn 2018, 2.62 ± 0.11 and autumn 2019, 2.68 ± 0.07) compared to those with (2.92 ± 0.05) and without (2.90 ± 0.06) skin from Blatna in autumn 2018. Nitrogen content of fillets with skin (2.62 ± 0.10) significantly ($P < 0.01$) differed from those without skin (2.77 ± 0.01) in sheatfish from Chlumec nad Cidlinou in spring 2019.

The first two axes of PCA in sheatfish explained 92.80% and the PC1 axis explained 62.9% of the total variance (Fig. 4). The samples were not clearly separated into specific clusters, but a gradient of samples negatively correlated with fat content. RDA distribution showed that fish weight was a stronger explanatory factor for functional traits data (first two canonical axes explained 77.80% of variability, permutation test on all axes: pseu-

do- $F = 25.7$, $P < 0.002$) in sheatfish than in European pike-perch and northern pike.

Nitrogen factors

Nitrogen factor indicates the content of nitrogen determined by Kjeldahl (ISO 937 1978). The term was chosen in the context of the Codex standard (CA 2004). The recommended nitrogen factor was determined from all samples of each fish species and expressed as mean and standard deviation. The recommended nitrogen factors for European pike-perch, northern pike, and sheatfish fillets with skin and without skin determined by Kjeldahl methods are given in Table 4. The recommended nitrogen factor for European pike-perch with skin is 3.28 ± 0.09 and 3.21 ± 0.09 without skin, for northern pike with skin is 3.18 ± 0.09 and without skin is 3.15 ± 0.09 , for sheatfish with skin 2.73 ± 0.13 and without skin 2.75 ± 0.12 . Codex Alimentarius recommended value ± 10% of variation (CA 2004).

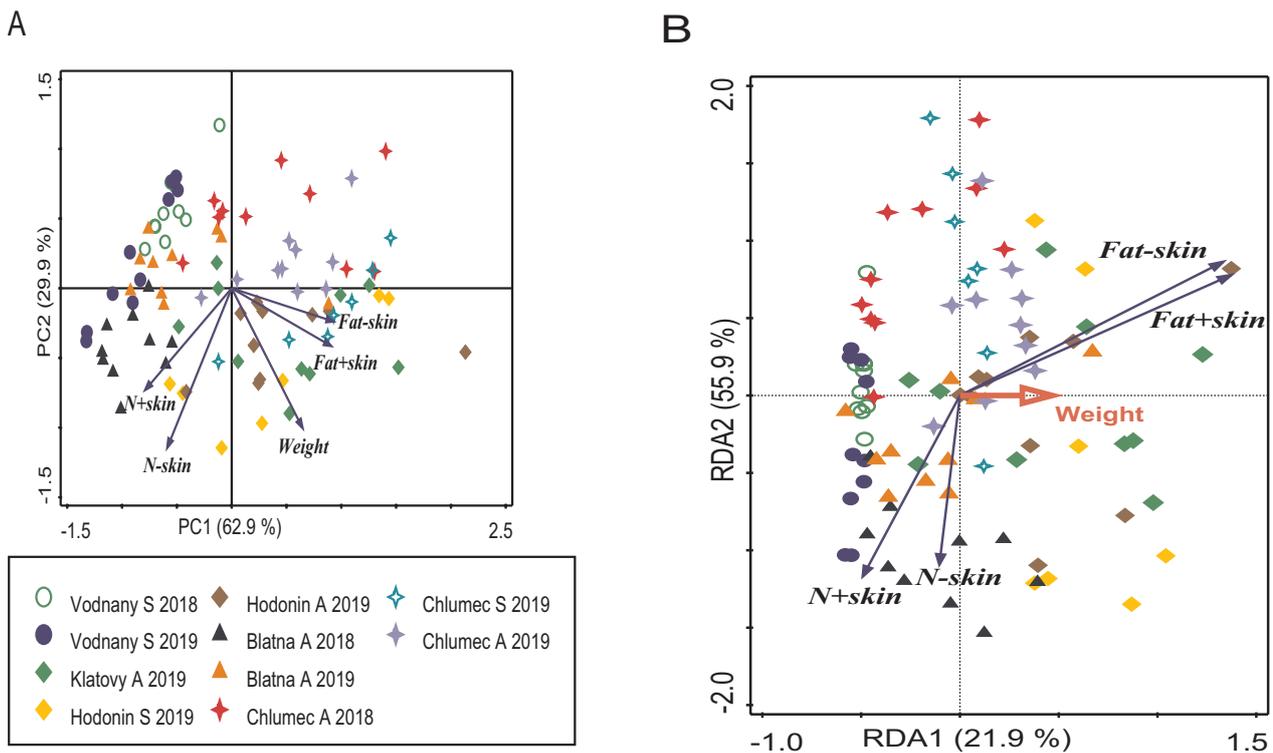


Figure 4. Ordination plots of sample distribution after principal component analysis (PCA) of functional traits as response variables (A) and redundancy analysis (RDA) (B) of functional traits as response variables and weight of sheatfish, *Silurus glanis* as explanatory variable. N+skin = nitrogen concentration in fillet with skin, N-skin = nitrogen concentration in fillet without skin, Fat+skin = fat percentage in fillet with skin, Fat-skin = fat percentage in fillet without skin, Weight = body weight of individual fish; abbreviations used in legend: S = spring sampling, A = autumn sampling). The length of the arrow reflects the power of the variable to differentiate the samples.

Discussion

Fillet samples with and without skin from European pike-perch, *Sander lucioperca*; northern pike, *Esox lucius*; sheatfish, *Silurus glanis* from several rearing locations and different harvest seasons and years were analyzed for dry matter, protein, fat, and ash content. To date, there are no established nitrogen factors determined by the Kjeldahl method for European pike-perch, northern pike, and sheatfish with and without skin.

The basic nutrient values obtained in this study are similar to those reported by Policar et al. (2016) in European pike-perch, Salama and Davies (1994) and Modzelewska-Kapituła et al. (2017) in northern pike, and Jankowska et al. (2007) in sheatfish. The water percentage decreased, and the fat and protein percentages increased, with increasing body weight and length, whereas the ash content remained fairly constant. Our findings are in agreement with those reported for other fish species (McComish et al. 1974; Elliott 1976; Costopoulos and Fond 1989; Brown and Murphy 1991; Clawson et al. 1991; Salama and Davies 1994).

Obtained nitrogen factors, taking into account the fat content of fish with skin and without skin, for European pike-perch, northern pike, and sheatfish determined by

Kjeldahl method are given in Table 4. All these values take into account the fat contents of the fish fillets with skin and without skin. The fat content of European pike-perch and northern pike fillets was low as shown in Table 1 and Table 2. The sheatfish fillets contained higher fat percentage with larger inter-sample differences as shown in Table 3 nevertheless fat content showed no significant influence ($P > 0.05$) on protein (nitrogen) content. We found no association of nitrogen content with rearing location, sampling season, or presence of skin in European pike-perch and northern pike. Nitrogen and fat content in sheatfish were negatively correlated. It is not necessary to establish nitrogen factors on a fat-free basis as is the case for some fish species (Colwell et al. 2011).

Table 4. Recommended nitrogen factors.

Species	Fillet	Nitrogen factor (Kjeldahl)
European pike-perch (<i>Sander lucioperca</i>)	with skin	3.28 ± 0.09
	without skin	3.21 ± 0.09
Northern pike (<i>Esox Lucius</i>)	with skin	3.18 ± 0.09
	without skin	3.15 ± 0.09
Sheatfish (<i>Silurus glanis</i>)	with skin	2.73 ± 0.13
	without skin	2.75 ± 0.12

Note: The recommended nitrogen factor was determined from all samples each fish species and mentioned as mean and standard deviation. Codex Alimentarius apply the recommended value ± 10% of variation (CA 2004).

The established nitrogen factors allow analysis of products from European pike-perch, northern pike, and sheatfish, in accordance with the EU legislation. Regulation (EU) No 1169/2011 of the European Parliament and of the Council on the provision of food information to consumers (EC 2011), including the requirement for a Quantitative Ingredients Declaration (QUID) label specifying the quantity of fish content. On the basis of recommended nitrogen factors is possible to calculate the value of QUID (CA 2004) for analyzed samples which are verified for labeled QUID information.

There are limitations in the use of nitrogen factors. They are calculated as mean values with standard deviations, and it is important to bear in mind the effects of season, weight, fishery location, and nutritional status on natural values and the analytical variability of their determination and to apply the recommended value of $\pm 10\%$ to the factor (CA 2004).

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Conclusions

Determined nitrogen factors for European pike-perch, northern pike, and sheatfish would help to ensure that consumers are buying correctly labeled or described fish products. The recommended nitrogen factor for European pike-perch with skin is 3.28 ± 0.09 and without the skin is 3.21 ± 0.09 , for northern pike with skin is 3.18 ± 0.09 and without the skin is 3.15 ± 0.09 , for sheatfish with skin 2.73 ± 0.13 and without the skin 2.75 ± 0.12 . Codex Alimentarius recommend allowing $\pm 10\%$ variation (CA 2004).

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