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## ESTIMATION OF SMOLTIFICATION OF HATCHERY-REARED SEA TROUT (*Salmo trutta morpha trutta* L.) BASED ON BODY MORPHOLOGY

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ABSTRACT. Detailed morphometric studies (truss network) were carried out using one and two years old sea trout. Additionally, body silvering and condition factors were evaluated. Correlation of these data (in various combinations) with smoltification index calculated from gill  $\text{Na}^+\text{-K}^+\text{-ATPase}$  activity and silvering (Dębowski et al. 1999b) were calculated as well as regression coefficients. Smoltification involved slight changes in fish body proportions: a decrease of head size, body height, and an increase of caudal part. These changes were, however, not sufficient to evaluate the degree of smoltification. Taking into consideration also body length and condition factor it was possible to develop regression model which explained 66% of smoltification index variation in one year old fish, and 57% - in two year old ones. Using additionally silvering value, increased coefficients of determination of the models to 98 and 99% respectively.

Key words: *Salmo trutta*, SMOLTIFICATION, MODEL, MORPHOMETRY, SILVERING, METHOD

## INTRODUCTION

Possibility of evaluating parr – smolt transformation of salmonid fish juveniles, which involves adaptation to sea water (Hoar 1976), is very important for successful fish stocking. Sea water test (Wedemeyer et al. 1980) and measurement of gill  $\text{Na}^+\text{K}^+\text{-ATPase}$  activity (Wedemeyer et al. 1980, Folmar, Dickhoff 1981, Boeuf et al. 1985, Lysfjord, Staurnes 1998) are considered as the most reliable. These methods are, however, difficult to apply in field conditions, as they require special equipment and costly laboratory analyses. In the fishery practice smoltification is usually assessed visually, taking into consideration body silvering and shape. Irrespective of the accuracy of such assessments, no comparisons of various fish groups may be done, and observations by various persons may differ. Silvering may be measured using special apparatuses (Kazakov, Kozlov 1985, Duston 1995), but no standard device is available. Additionally, light conditions may affect the results (Vanstone, Markert 1968, Kazakov, Kozlov 1985). Thus, the use of several parameters together seems the most sensible method (Dębowski et al. 1999b).

Evaluation of smoltification based on morphological changes in fish is a non-destructive alternative (Fessler, Wagner 1969). Such method was applied by Winans, Nishioka (1987), and Beeman et al. (1994, 1995) for various *Oncorhynchus* species. Detailed multifactorial analysis of morphometric data enabled formulation of reliable criteria of smoltification.

Smoltification process is similar in all anadromous salmonid fish (Folmar, Dickhoff 1980, Wedemeyer et al. 1980), but in sea trout it is often less pronounced and more variable (Oakland et al. 1993, Tanguy et al. 1994, Dębowski et al. 1999b).

The aim of the present study was to develop a method of sea trout smoltification assessment based on various sets of morphometric data: from the simplest, obtained with a ruler, to very detailed, obtained using special equipment.

## MATERIAL AND METHODS

### FISH

One and two years old sea trout were used in the experiment. The eggs were stripped from spawners harvested in the lower Vistula River, and the fish were reared in the Department of Salmonid Fish Breeding of the Inland Fisheries Institute in Rutki. Two year old trout were measured 3 times, and one year old ones – 4 times, from March to June 1996, and once in May 1997. The following measurements were performed: caudal length (l. caud. in mm) (L), body weight (in g) (W), body silvering (S) (Kazakov, Kozlov 1985, Dębowski, Radtke 1994). Condition factor was also calculated ( $CF = 10^5 \cdot W \cdot L^{-3}$ ). Detailed characteristics of the fish is presented in Dębowski et al. (1999b).

Dębowski et al. (1999b) calculated discriminant function values for each fish, assumed an index of smoltification (IS):

$$\text{for one year old fish } IS_1 = -0.5055 \cdot S + 0.2346 \cdot A - 5.6353$$

$$\text{for two years old fish } IS_2 = -0.2710 \cdot S - 0.3705 \cdot A + 4.8857$$

(where A - gill  $Na^+ - K^+$  ATPase activity as  $\mu M$  of inorganic phosphorus per mg of protein per hour).

The lower the IS value, the higher the smoltification level (Dębowski et al. 1999b).

All fish were recorded using S-VHS camera, and then measured using computer image analysis system MultiScan (Dębowski et al. 1998). Four body heights (Fig. 1), and 28 distances between certain landmarks in the fish body - truss network (Fig. 2) were measured (Dębowski et al. 1999a).



Fig. 1. Body heights

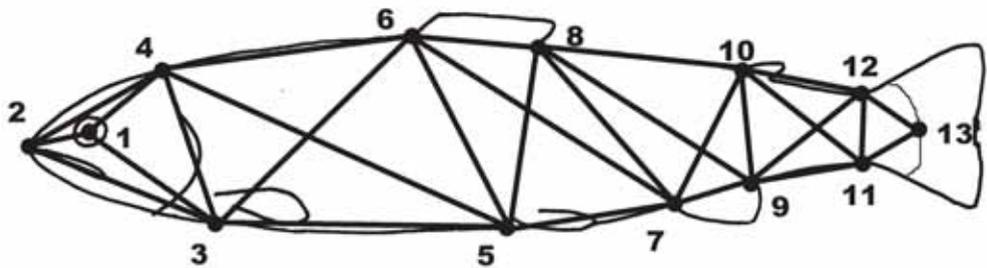


Fig. 2. Truss network characters

## STATISTICAL ANALYSIS

The analysis was carried out for all parameters together, separately for each age group. Morphometric data were standardised to avoid the effect of fish body length variability. In case of easily measured parameters, calculations were simplified for the field use, and expressed as:

$$W_i = \frac{w_i}{L}$$

where:

$W$  – relative height,

$w$  – measured height,

$i = 1, 2, 3, 4$ .

The data obtained using truss network method were standardised according to Karakousis et al. (1993):

$$X = \ln(x) - \beta (\ln(L) - \ln(L_m))$$

where:

$X$  – standardised value,

$x$  – measured value,

$\beta$  – regression coefficient of  $\ln(x)$  versus  $\ln(L)$ ,

$L_m$  – mean fish length.

To reduce the number of truss network variables, factorial analysis was performed, and several principal components were found (PC $_j$ , where  $j$  – number of components) which were used in further analysis.

Multivariate stepwise regression was calculated for smoltification index (IS) versus the obtained variables. Several regression models were tested:

- Model enabling evaluation of morphometric changes during smoltification (variables: principal components PC $_j$ )
- Model providing information on smoltification from truss network (PC $_j$ ), body length, and condition factor (CF).
- As above, additionally including the silvering factor (S).
- Model enabling evaluation of humping changes during smoltification (variables: relative heights W1, W2, W3, W4).
- Model enabling evaluation of smoltification from body height (Wi), length (L), and condition factor (CF).
- As above, additionally including the silvering factor (S).

## RESULTS

Six principal components were obtained from factorial analysis for each age group (Tabs. 1, 2). They explained 77.1% of total variation of measurements for one year old fish, and 79.5% for two year old ones. No correlation was found of none of the factors with the fish length ( $p < 0.001$ ), indicating that they were related to fish shape, and not size (Humphries et al. 1981).

Multivariate stepwise regression analysis resulted in models describing relationship between smoltification index (IS) and various series of the data (Tab. 3). Changes of fish body shape during smoltification are described by the equations 1.1, 1.2, 4.1, and 4.2. The relationship is, however, not very high; the models explain 33-53% of IS variation. Equations 4.1 and 4.2 indicate that the height of front and rear body parts (but not caudal peduncle) decreased during smoltification in both age groups. More detailed information is provided by the equations 1.1 and 1.2 (Fig. 3). Generally, head size, body and caudal peduncle height decreased in smolts, the fish body becoming

TABLE 1

Loadings from principal component analysis of morphometric characters of 68 one-year old trout. Characters are distances between landmarks (Fig.2). Loadings  $>|0.7|$  are distinguished.

Character	PC1	PC2	PC3	PC4	PC5	PC6
1-2	-.153317	.587117	-.394688	-.313875	.007234	.166867
1-3	<b>.755608</b>	.047325	-.080507	.217163	.100192	-.219969
1-4	.402177	.616201	.052889	.384219	-.233110	.080907
2-3	.413284	.272367	-.307169	-.033513	.211402	-.356711
2-4	.324053	<b>.783005</b>	-.114185	.223712	-.218034	.174967
3-4	<b>.897986</b>	.175512	.022659	.182127	-.047493	-.028603
3-5	.023907	-.073048	.188635	.082113	<b>.816041</b>	.245128
3-6	.544984	-.338171	.052370	.200486	-.199701	.479992
4-5	.213685	-.418054	.083823	.039494	<b>.805541</b>	.069114
4-6	-.083323	<b>-.881432</b>	-.066029	-.078455	.091061	.139307
5-6	<b>.784705</b>	.324463	.096685	.300708	.164029	.252411
5-7	.159853	-.032053	.261371	-.011135	<b>-.751852</b>	.122551
5-8	<b>.819618</b>	.249717	.068719	.206167	-.109932	.309810
6-7	.541503	.465841	.380382	.273151	.053341	.080803
6-8	.193790	.258118	-.637958	-.206361	.185896	-.053773
7-8	.406699	.268576	<b>.744481</b>	.399058	-.038575	.110673
7-9	-.157866	-.233534	-.182585	<b>-.739063</b>	.174962	.058171
7-10	.628001	-.346078	-.055747	-.365313	.077896	.009520
8-9	.213736	.136008	<b>.914245</b>	-.001353	.092653	.087186
8-10	-.320938	-.207796	<b>.728961</b>	-.321771	.118538	-.126350
9-10	<b>.853092</b>	.134978	.065429	.355669	-.037870	.093770
9-11	.020594	-.174223	.076553	<b>.757792</b>	.196288	.142613
9-12	.321725	-.045277	-.017710	<b>.771587</b>	.005961	.336321
10-11	.530776	.214531	.035065	.670801	.140697	.196849
10-12	.256056	.301111	-.066377	<b>.747077</b>	.059315	.299068
11-12	<b>.742698</b>	-.021688	.002855	.043357	-.043467	.233718
11-13	-.140819	-.053869	-.041189	-.156878	-.168723	<b>-.828466</b>
12-13	-.107343	-.087076	-.008953	-.246954	-.040587	<b>-.833933</b>
% of total variance explained	33.5	12.5	10.2	8.7	6.7	5.5

TABLE 2

Loadings from principal component analysis of morphometric characters of 60 two-years old trout. Characters are distances between landmarks (Fig.2). Loadings  $>|0.7|$  are distinguished.

Character	PC1	PC2	PC3	PC4	PC5	PC6
1-2	-.055635	-.483075	-.221925	.223119	-.561213	-.009695
1-3	.358820	.032085	-.060462	.083896	<b>-.718696</b>	.303168
1-4	.264243	-.021445	<b>-.764942</b>	.190200	.095300	-.028775
2-3	.112734	-.231300	-.080033	.116126	<b>-.897723</b>	.206664
2-4	.148977	-.270066	<b>-.779564</b>	.299185	-.215118	.009003
3-4	<b>.733267</b>	.020632	.030945	.108037	-.588554	.033953
3-5	.227081	.112526	.664880	.367966	.435613	-.025085
3-6	<b>.854864</b>	-.007536	.190872	.127184	.240264	-.078572
4-5	.225237	.111592	<b>.914349</b>	.154249	.021187	-.017664
4-6	.297560	.002891	<b>.871568</b>	-.126622	.064254	-.101741
5-6	<b>.860818</b>	.049478	.104250	.357198	-.115325	.003015
5-7	.079468	.298883	-.448130	-.526366	.334956	.147938
5-8	<b>.915522</b>	-.006494	-.146387	.081083	-.132010	.069771
6-7	.325029	.607021	-.136628	.104315	.264992	.173072
6-8	.141839	<b>-.705546</b>	-.352960	.258286	-.000102	.259869
7-8	.244695	<b>.904023</b>	.149763	-.098202	.070898	-.133713
7-9	.231699	.188511	.013952	<b>.802161</b>	-.036947	.221837
7-10	<b>.779678</b>	.039814	.059758	.277427	-.139450	-.136411
8-9	-.032558	<b>.937809</b>	.068385	.232445	.068707	.003747
8-10	-.452282	<b>.768397</b>	.005488	.068374	.052397	-.183721
9-10	<b>.926717</b>	-.087764	.016763	-.082435	-.055274	-.029191
9-11	-.302167	.080191	.223677	<b>-.748480</b>	.125065	.107943
9-12	.008635	-.039802	.052295	<b>-.885062</b>	.035406	-.003874
10-11	.518649	.159737	.471977	-.331941	.094750	.357689
10-12	-.110368	.084038	.354289	-.500638	.100038	.467291
11-12	<b>.802235</b>	.053334	.024283	-.003865	-.053896	-.135054
11-13	.073829	.112828	-.042206	-.063225	.157852	<b>-.798703</b>
12-13	.147124	.192209	.258482	.025981	.280789	<b>-.755893</b>
% of total variance explained	24.5	20.7	11.9	9.6	7.7	5.1

TABLE 3

Regression models for some sets of variables on indices of smoltification for one-year ( $IS_1$ ) and two-years ( $IS_2$ ) old trout. Variables: principal components (PCj), body heights (Wi), length (L), condition factor (CF) and body silvering (S). „X” denotes variable taken into account. For all models  $p < 0.001$ .

Variables					R <sup>2</sup>	Model
PCj	Wi	L	CF	S		
X					0.33 0.49	$IS_1 = 0.689 * PC1 + 0.620 * PC2 - 0.691 * PC3 + 0.149$ (1.1) $IS_2 = 1.127 * PC1 - 0.486 * PC3 + 0.717 * PC4 - 0.616 * PC5 - 0.538$ (1.2)
X		X	X		0.66 0.57	$IS_1 = 13.357 * CF - 0.092 * L - 0.556$ (2.1) $IS_2 = 12.846 * CF - 0.717 * PC3 - 0.430 * PC5 - 14.082$ (2.2)
X		X	X	X	0.98 0.99	$IS_1 = -0.470 * S + 5.950$ (3.1) $IS_2 = -0.333 * S + 0.006 * L + 0.149 * PC1 - 0.131 * PC5 + 3.168$ (3.2)
	X				0.51 0.53	$IS_1 = 222.19 * W1 - 128.34 * W3 - 18.807$ (4.1) $IS_2 = 96.44 * W1 + 102.50 * W2 - 90.76 * W3 - 24.543$ (4.2)
	X	X	X		0.66 0.53	$IS_1 = 13.357 * CF - 0.092 * L - 0.556$ (5.1) $IS_2 = 96.44 * W1 + 102.50 * W2 - 90.76 * W3 - 24.543$ (5.2)
	X	X	X	X	0.98 0.98	$IS_1 = -0.470 * S + 5.950$ (6.1) $IS_2 = -0.334 * S + 0.005 * L + 16.316 * W1 + 0.734$ (6.2)

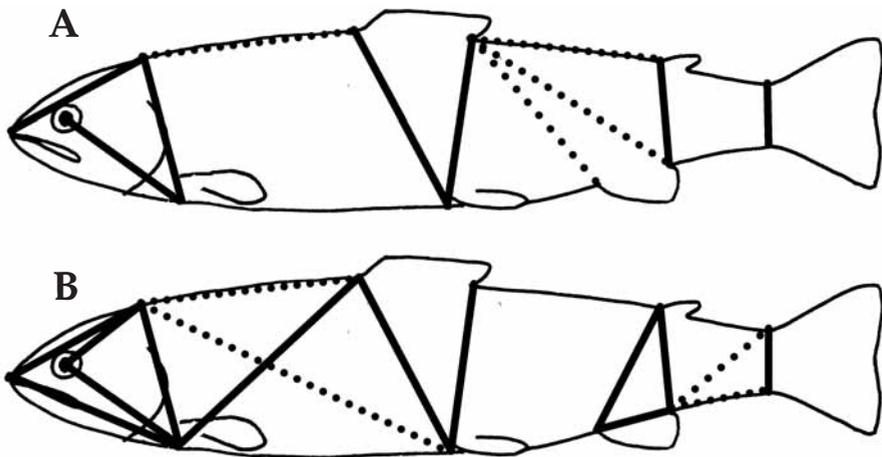


Fig. 3. Morphometric characters positively (solid lines) and negatively (broken lines) correlated with index of smoltification (IS) for one-year odd (A) and two years old (B) trout

more elongated. In models comprising also other variables: length (L) and condition factor (CF), coefficients of determination increased (Tab. 3, equations 2.1, 2.2, 5.1, 5.2). In one year old fish, 66% determination may be obtained using length, and condition factor only (equations 2.1 and 5.1). In two years old fish, body length did not increase the coefficient, and condition factor did it only slightly (equations 2.2 and 5.2). Inclu-

sion of the silvering factor (S) into the models considerably changed the coefficient, which reached from 98 to 99% (Tab. 3, equations 3.1, 3.2, 6.1 and 6.2). In case of one year old fish, silvering factor alone was sufficient (3.1, 6.1), and in two years old trout – also body length and two principal components were necessary (3.2), or length and height of front part of the body (6.2).

## DISCUSSION

Wedemeyer et al. (1980), and Virtanen and Soivio (1985) observed that almost each smoltification indicator clearly visible in wild fish did not apply to reared fish, and that environmental stressors under artificial conditions could considerably affect smoltification (Wedemeyer 1982). At the same time, level of smoltification of the released fish and stocking site determined the success of stocking (Farmer et al. 1978, Isaksson et al. 1978, McCleave 1978, Bartel, Dębowski 1996). Thus, reliable assessment of smoltification is of key importance. Dębowski et al. (1999b) undertook an attempt to increase accuracy of such assessments by taking into consideration several parameters together. For each fish, smoltification index was calculated (IS) from silvering as well as gill  $\text{Na}^+\text{-K}^+\text{ATPase}$  activity, silvering being the most important factor (Dębowski et al. 1999b). Thus, it is obvious that among the developed regression models, those containing variable S are the most accurate. The apparatus for silvering measurements is, however, a unique experimental device, available only for research purposes. In the fishery practice, only simple measurements may be done (body length and height), sometimes also body weight. Such measurements produce inaccurate smoltification estimates: explaining 66% of variation for one year old fish, and 53% for two year old ones.

Body shape changes involved in smoltification i.e. relative decrease of body height, and lengthening of its rear part, were similar to those observed in Arctic char, *Salvelinus alpinus* (L.), (Damsgard 1991), and *Oncorhynchus* sp. (Beeman et al. 1994, 1995). Decrease of head size occurred also in Arctic char but not in *Oncorhynchus* sp. Body shape differences observed by Beeman et al. (1994, 1995) were sufficient to develop discriminant function, which made it possible to distinguish smolt from parr. In case of sea trout, considerable changes were also observed, but their values explained only about 50% of IS variation.

The results of the present study indicate that body silvering measurements are necessary for accurate evaluation of smoltification. For one year old fish, this parame-

ter alone is sufficient, and for two year old ones – body length, and some other morphometric data increase model accuracy. If silvering values are not available, smoltification assessment is much less accurate, especially in case of two-year-old fish. In such cases the assessment is based on body length and condition factor (one year old fish), and some morphometric data (two-year-old fish).

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## STRESZCZENIE

### OCENA STOPNIA SMOLTYFIKACJI POCHODZĄCYCH Z WYLĘGARNI TROCI (*Salmo trutta morpha trutta* L.) NA PODSTAWIE DANYCH MORFOLOGICZNYCH

Przeprowadzono wiosną szczegółową morfometrię (truss network) jedno- i dwuletnich troci. Dokonano także pomiarów wysrebrzenia ciała i obliczono współczynniki kondycji. Zbadano regresje różnych zestawów tych danych na obliczony na podstawie aktywności skrzelowej  $\text{Na}^+$ - $\text{K}^+$  ATPazy i wysrebrzenia indeksu smoltyfikacji (Dębowski et al. 1999b). W związku ze smoltyfikacją ryby zmieniają kształt: zmniejsza się relatywnie głowa i wysokość ciała, wydłuża się część ogonowa. Zmiany te jednak nie są duże i nie wystarczające aby na ich podstawie oceniać stopień smoltyfikacji. Uwzględnienie dodatkowo długości i współczynnika kondycji pozwala na sformułowanie modelu regresji wyjaśniającego 66% zmienności indeksu smoltyfikacji u ryb jednorocznych i 57% - dla ryb dwuletnich. Dodanie zmiennej opisującej wysrebrzenie zwiększa współczynnik determinacji modeli do, odpowiednio, 98 i 99%.

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