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SHORT COMMUNICATION

In vivo calibration of juvenile crayfish body length and weight with a photographic-computer method

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Abstract. The aim of this work was to develop a photographic-computer method to determine in vivo the body length and weight of juvenile crayfish based on standards that describe the dependence curves of these characters. The measurement procedure that was developed requires one digital group photograph of a sample generally comprised of thirty or so individuals and an image scale, which is visible within the frame. A special program is used to analyze the image according to the scale on the image. Further, only the cephalothorax length (CL) of specimens from the studied sample that appear in the photograph is measured. Total length (TL) and weight (W) are then calculated with correlation formulae that were developed for the characters analyzed. The following dependencies were calculated from the formulae developed for two-month-old narrow-clawed crayfish, *Astacus leptodactylus* (Esch.) (n = 101, W = 225.9 ± 67.2 mg, TL = 18.7 ± 2.1 mm): $TL = 1.739\ CL + 1.1903$, $W = 0.1306\ TL^{2.533}$, $W = 0.7635\ CL^{2.4476}$. All of the dependencies were highly significant statistically ($P < 0.001$).

Keywords: crayfish, body weight, body length, method, measurement

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Measuring individual body length and weight is one of the basic procedures undertaken in scientific study. Various measuring implements are used, such as rulers or slide-calipers, which are adapted to make the task easier (Hepper 1965, Stypińska et al. 1978). However, these measuring methods are not applicable to small individuals as they are very difficult to manipulate. This is why the body length and weight of such individuals is determined after they have been sacrificed. When taking measurements during an experiment there is one other negative issue, apart from humanitarian concerns; it is necessary to take samples from the experimental stock for measurements during the experiment. This can reduce dramatically the stocking density of the investigated population, which can impact the results obtained. To date, the proportions among a range of body parts have been determined and the corresponding formulae have been written to describe the dependencies between them and the length and weight of crayfish of various species (Kossakowski 1966, Huner et al. 1988, Chybowski 2007). Since in vivo determinations of crayfish body weight using electronic scales are not currently difficult, one can use these formulae to calculate basic body length. Individual body weight can also be calculated with specimen length alone using the same dependence formulae. Previous studies have confirmed that it is possible to take in vivo measurements of body length (even of one-month old specimens) by analyzing photographs with the appropriate computer software (Ulikowski and Krzywosz 2006). However, all of the dependence formulae applied to date were developed based on larger individuals (usually above 6 cm total length). Thus, it is justifiably doubtful whether these are appropriate for smaller juveniles whose body proportions might differ from those of fully-formed individuals.

The aim of the study was to develop an in vivo photographic-computer method for determining the weight and length of juvenile crayfish based on formulae that describe the dependencies of the biometric characters analyzed.

The dependence curves and the formulae needed for describing them were determined using individual measurements of two-month old individuals of narrow-clawed crayfish, *Astacus leptodactylus* (Esch.). The crayfish were obtained from a six-week rearing period on commercial feed under controlled conditions (Nutreco Company, Perla Larva 3.0 – composition according to manufacturer: protein 62%, fat 11%, fiber 0.8%, ash 1.0%). Feed was delivered twice per day *ad libitum*. The crayfish were reared in individual compartments (12 × 12 cm) at a stocking density of one per compartment. The water level flowing trough the compartments was 10 cm. The water circulated through a recirculation system equipped with a purification system, and the water temperature was maintained at a constant of 18°C.

All of the individuals (n = 101) measured had the full complement of walking legs and no visible body deformations. The measurement of individual crayfish body weight (W, to the nearest 1 mg) was done on an electronic scale (Axis AM-50, Poland). The specimens were dried gently with paper towels before they were placed on the scale. After weighing, the individual was placed in a Petri dish filled with water. This dish was then placed under a camera mounted on a tripod (Canon EOS 5D + Canon EF 24-105 f/4 L IS USM lens, distance from the dish – 0.5 m), and a digital photograph was taken. A photograph was taken of the Petri dish and the scale (range of 5 cm, to the nearest 0.1 mm), which was then used to calibrate the measurement module of the computer program used to analyze the picture. All of the photographs were taken with the same camera and tripod settings. Total body length (TL – from the rostrum to the end of the telson) was then measured, as was the cephalothorax length (CL – from the anterior edge of the rostrum to the posterior edge of the carapace) using a computer program for image analysis (MultiScan 8.1, Computer Scanning Systems LTD, Poland; to the nearest 0.1 mm). This program was calibrated previously using a photograph of the scale. The data obtained was subjected to statistical analyses using Statistica 7.1 (Statsoft, USA). The normality of the range of the data obtained was verified with the Chi^2 test. The mean value of the TL/CL proportion and its coefficient of variation (V) were calculated. Linear and non-linear regression models were used to fit the dependence curves between TL and CL, W and TL, and W and CL along with the corresponding descriptive formulae. The statistical significance among the relationships was determined with the determination coefficient (r^2) and the level of significance (P).

The measurements and weights of the crayfish analyzed were (mean ± standard deviation (range)): W = 225.9 ± 67.2 mg (61-406 mg); TL = 18.7 ± 2.1 mm (12.2-23.1 mm); CL = 10.1 ± 1.2 mm (6.1-12.4 mm). The mean value for the TL/CL proportion of the two-month-old specimens was 1.86 (V = 3.36%). This is lower than values reported by other authors for adult specimens of this same species (Kossakowski 1962, Śmietana 1998, Andrzejewski et al. 2001, Chybowski 2007). Simultaneously, it is higher than the figure obtained in earlier studies of one-month-old individuals (Ulikowski and Krzywosz 2006). These results indicate that the values of these proportions change as individuals age (Table 1).

The dependence between TL and CL (mm-mm) in the two-month-old specimens from the current study is illustrated by simple regression (Fig. 1), described by the following formula:

$$TL = 1.739\ CL + 1.1903;\ r^2 = 92.2\%,\ P < 0.001 \qquad (1)$$

This dependence is similar to that obtained for one-month individuals (Ulikowski and Krzywosz 2006), while it differs from that described for adult specimens of this species (Stypińska et al. 1978, Chybowski 2007). This is likely the result of differences confirmed in the

Table 1

Comparison of proportions between total length (TL) and cephalothorax length (CL) and the coefficient of variation (V) of adult individuals from different Polish lakes and ponds and juvenile *A. leptodactylus* reared under controlled conditions

Origin of research material	Sex or age and rearing time	TL/CL	V (%)	Authors
	♂	1.90	1.10	
Mazurian Lakes	♀	2.00	2.50	Kossakowski (1962)
	♂	1.86	3.67	
Mazurian Lakes	♀	2.00	2.50	Śmietana (1998)
	♂	1.95	3.31	
Lake Bylice Małe	♀	2.01	2.48	Śmietana (1998)
	♂	1.90	2.63	
Lake Rzeplino	♀	2.02	1.99	Śmietana (1998)
	♂	1.91	1.52	
Lake Sumin	♀	2.04	6.68	Śmietana (1998)
	♂	1.91	2.41	
Lake Cerkiew Nowa	♀	1.92	2.83	Śmietana (1998)
	♂	1.90	4.60	
Lak Gaj	♀	2.05	2.57	Andrzejewski et al. (2001)
Dgał Hatchery Ponds and Lake	♂	1.96	5.40	
Samin	♀	2.01	5.20	Chybowski (2007)
Recirculation system	one-month indiv. 4 weeks	1.82	4.56	Ulikowski and Krzywosz (2006)
Recirculation system	two-month indiv. 6 weeks	1.86	3.36	Current study

mean values of TL/CL proportions of adult and juvenile individuals. There are also differences in the TL/CL dependence among adult males and females (Stypińska et al. 1978, Podsiadło and Olech 1994).

The dependence between W and TL (mg-mm) and between W and CL (mg-mm) of the two-month individuals tested was best described by the regression curve (Figs. 2 and 3), described with the power equations:

$$W = 0.1306\,TL^{2.533};\ r^2 = 79.9\%,\ P < 0.001 \qquad (2)$$

$$W = 0.7635\,CL^{2.4476};\ r^2 = 78.7\%,\ P < 0.001 \qquad (3)$$

The values of the determination coefficients (r^2) and level of significance (P) indicate that all the

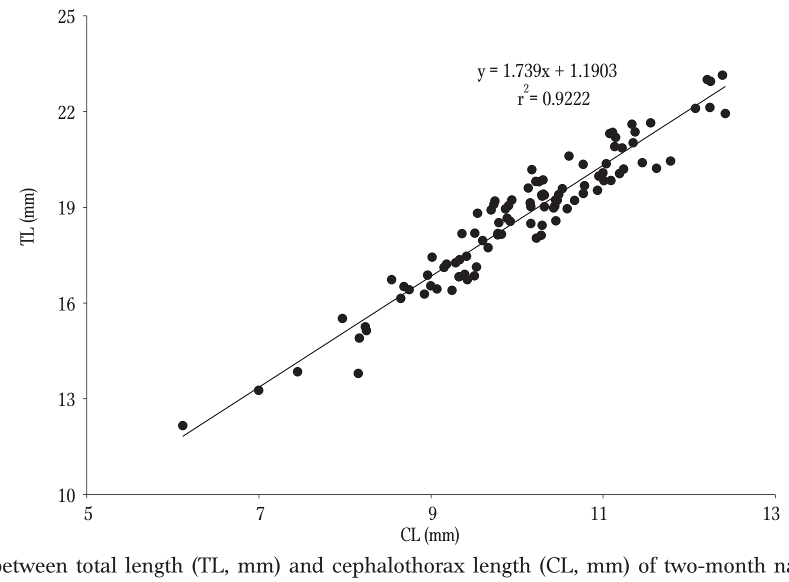

Figure 1. Dependence between total length (TL, mm) and cephalothorax length (CL, mm) of two-month narrow-clawed crayfish, *A. leptodactylus.*

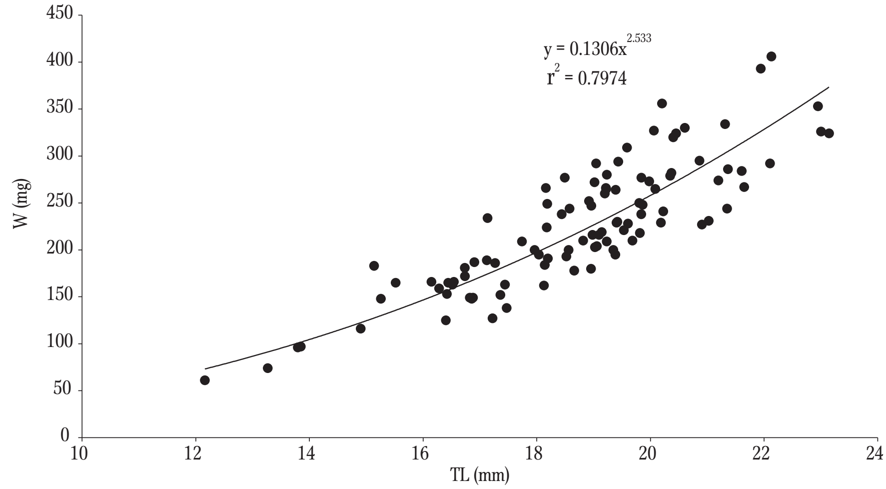

Figure 2. Dependence between body weight (W, mg) and total length (TL, mm) of two-month narrow-clawed crayfish, *A. leptodactylus.*

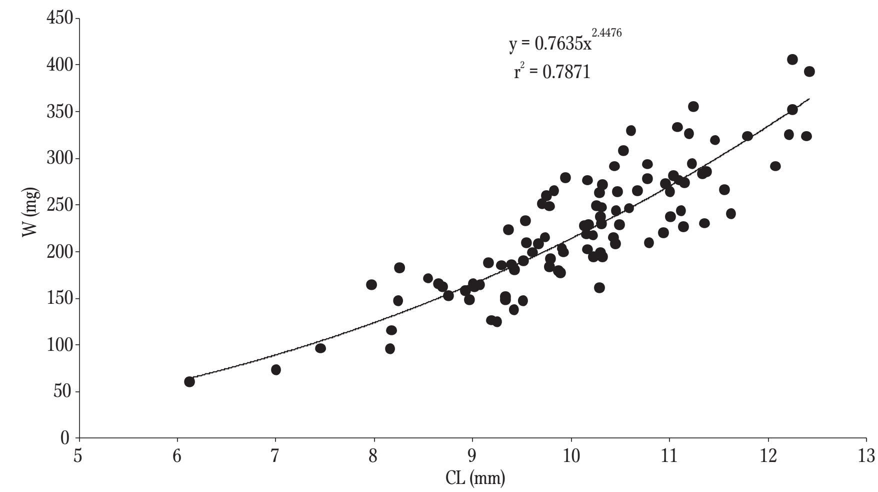

Figure 3. Dependence between body weight (W, mg) and cephalothorax length (CL, mm) of two-month narrow-clawed crayfish, *A. leptodactylus.*

dependences described are highly significant. According to Chybowski (2007), the regression coefficient described by the power equations is best at determining the dependence between W and TL in adult *A. leptodactylus*, but also in *Astacus astacus* L., *Pacifastacus leniusculus* Dana, and *Orconectes limosus* (Raf.). However, the parameters of the regression curve equations reported by Chybowski differ from the results of the current study on two-month-old individuals. This indicates that it is appropriate to apply different dependence formulae for TL and CL, W and TL, and W and CL for juvenile and adult individuals. This suggests that the aim of subsequent studies should be to determine the formulae for the dependencies mentioned above for individuals older than those used in the present study, for example one-year olds.

The equations for TL and CL, W and TL, and W and CL dependencies in two-month-old individuals presented in this paper permit using the photographic-computer method to determine in vivo the weight and length of juvenile crayfish. The entire measurement procedure is based on taking one digital group photograph that includes the measurement scale and the studied sample that is usually comprised of about thirty individuals. Then the photograph files are read by an image analysis program (e.g., MultiScan), and the measurement module is calibrated according to the scale in the photograph. Further, measurements are taken just of the cephalothorax of the individuals visible in the photograph, while total length and weight are calculated according to the formulae developed. One more advantage of the method presented is the possibility of taking monitoring measurements during experiments without having to remove individuals from the rearing tanks. It is enough to take a digital photograph of the tank bottom with the scale, while further analysis and results are performed by the image analysis program.

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Streszczenie

Przyżyciowa kalibracja długości i masy ciała młodocianych raków przy zastosowaniu metody fotograficzno-komputerowej

Celem było opracowanie fotograficzno-komputerowej metody przyżyciowego określania masy i długości ciała młodocianych raków. Na podstawie pomiarów masy i długości ciała dwumiesięcznych osobników raka błotnego, *Astacus leptodactylus* (Esch.) (n = 101, W = 225,9 ± 67,2 mg, TL = 18,7 ± 2,1 mm) wyznaczono wzory opisujące zależności pomiędzy masą (W), długością głowotułowia (CL) i długością ciała (TL). Wyliczona średnia wartość proporcji TL/CL badanych dwumiesięcznych osobników wyniosła 1.86 (V = 3,36%). Natomiast opracowane wzory zależności przedstawiały się następująco: TL = 1,739 CL + 1,1903, $W = 0{,}1306\ TL^{2{,}533}$, $W = 0{,}7635\ CL^{2{,}4476}$. Wszystkie wymienione zależności miały statystycznie wysoką istotność (P < 0,001). Po opracowaniu krzywych zależności i opisujących je wzorów proponowana procedura pomiarowa sprowadza się do wykonania jednej zbiorowej cyfrowej fotografii badanej próby liczącej przeważnie kilkadziesiąt osobników razem z umieszczoną w kadrze wzorcową skalą. Następnie plik fotografii wczytuje się do komputerowego programu do analizy obrazu (np. MultiScan) i kalibruje jego moduł pomiarowy według skali widocznej na wczytanej fotografii. Dalej wykonuje się pomiary tylko długości głowotułowia osobników badanej próby widocznych na fotografii, a długość całkowitą i masę ciała oblicza się z opracowanych wzorów zależności między TL a CL, W a TL oraz W a CL.