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RESEARCH ARTICLE

Impact of two telemetry transmitter implantation incision suturing methods on the physiological state and condition of perch (*Perca fluviatilis*)

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Abstract. The aim of this work was to determine the impact on European perch, *Perca fluviatilis* L. (mean body weight – 78.33 g) of the intraperitoneal implantation of telemetry transmitters using different suturing methods. In the first experiment silk sutures were used (experiment I – group ST), while in the second tissue adhesive was used (experiment II – group GT). Following the procedure, the fish were kept for 42 days in a recirculating system. Differences in growth and condition parameters were only noted in the first week of the experiment. Specimens from group GT had lower values for DGR (daily growth rate) and SGR (specific growth rate), but a higher value for FCR (feed conversion ratio) values. For the hematological parameters, lower values of MCV (mean corpuscular volume) and PLT (blood platelets) were noted in group GT, while for the biochemical parameters, lowered ALP (alkaline phosphatase) activity and Mg (magnesium) concentrations were noted in group ST. In group ST, 33.3% of the specimens loss their tags, while in group GT 77.8% did so. Differences in incision healing were only noted in the second week, when specimens in group ST were observed to have fully closed incisions, while in group GT 50% of the incisions were open. Despite the high percentage of implantation incision healing in both groups, because of the high values of tag loss rate, neither method can be recommended for perch. It might be more effective to use tag with external antennae in this species. The method use for closing implantation incisions also must be improved to eliminate tag shedding.

Keywords: perch, biochemical profile, hematological profile, tag retention, telemetric, fish tagging

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Introduction

Stress is a adaptive mechanism that permits organisms, including fish, to cope with stressors, react to changing conditions, and maintain systemic homeostasis (Chrousos 1998). Stress itself is not dangerous for organisms, but it can become so when it is too strong or lengthy. Such situations can trigger physiological mechanisms and reactions that lead to weakened physical condition (Barton and Iwama 1991). Organisms under stress exhibit primary, secondary, and tertiary reactions. The primary response is in the

hypothalamus, where stress is recognized. It emits a nerve impulse that is transmitted by the autonomic nervous system. This stimulates the hypothalamic-pituitary-adrenal axis, which secretes, inter alia, cortisol and catecholamines as hormonal responses (Nematollahi et al. 2013). These hormones cause biochemical, metabolic, hematological, and immunological changes that are the body's secondary reaction. Chronic stress can cause the final, critical stage of response, which is exhaustion, disease, and even death (Barton and Iwama 1991, Barton 2002, Smircich and Kelly 2014). There are numerous reports in the literature on various manipulations that fish are subjected to in aquaculture, e.g., high stocking density, transport, sorting, and artificial reproduction, that can be sources of stress and which can also have negative impacts on fish well-being and health (Falahatkar and Barton 2007).

A variety of biotelemetric techniques are used in ecology studies and population management for many fish species. Telemetry transmitter are inserted surgically through an abdominal incision, which can be stressful for fish (Smircich and Kelly 2014). The source of stress that accompanies transmitter implantation could be the presence of the tag in the body cavity. Nevertheless, implanting fish with tags permits conducting unique research and observing fish in their natural environment from a greater distance independent of season or time of day (Smircich and Kelly 2014). Thanks to this, we are able to collect more precise, more reliable data than previously on many aspects of fish life, including migration, spawning activity, feeding, etc. (Righton et al. 2001, Smircich and Kelly 2014). Telemetry is a reliable research method under the condition that it does not impact the body, well-being, or behavior of the fish (or, possibly, if it does so only to a slight degree). This condition must be met fully if the data sample collected with telemetry studies are to be representative of entire fish populations (Mellas and Haynes 1985). Thus, it is important the determine whether, and to what extent, tag implantation induces stress in fish.

Hematological, biological, and growth parameters are excellent, reliable indicators of stress (Barton 2002). Stressors cause the release of stress hormones, particularly cortisol, which induce a number of biochemical reactions, such as changes in enzyme activity and concentration, and in levels of glucose, total protein, and magnesium (Hoseini et al., 2016). They can also affect the functioning of the immune system (Smircich and Kelly 2014), as is manifested in the less efficient feed utilization, slower growth rates, decreased fish resistance, and even increased mortality (Smircich and Kelly 2014).

In an effort to broaden the understanding of the stress caused by the implantation of telemetry tags in juvenile European perch, *Perca fluviatilis* L., a range of biochemical and hematological determinations were performed. The healing of post-implantation incisions sutured with two methods (silk sutures or tissue adhesive) and indexes of growth, feed conversion ratio, survival, and short-term tag retention were also analyzed.

Materials and methods

Fish and rearing conditions

Fertilized perch eggs were collected from spawning grounds during the natural spawning period of this species in Lake Dgał Wielki (northern Poland) in early April. They were transported to an earthen pond (0.2 ha surface area) located at the Department of Sturgeon Fish Breeding, Inland Fisheries Institute in Olsztyn (IFI Olsztyn) in northern Poland. Initial perch rearing was done there on natural feed to a size of approximately 0.2 g body weight (BW). The fish were caught in mid-June and transferred to tanks in a recirculating aquaculture system (RAS) (2 tanks with volumes of 2 m^3). The perch were trained to consume formulated feed. The feed used was manufactured by Nutreco (Nutra, Trouvit, France), and the feeding procedure was similar to that reported by Policar et al. (2015). The fish were reared under these conditions for approximately ten weeks until they had reached approximately 10 g BW. Next, a portion of this material (approximately 500

individuals) were transferred in polyethylene bags (20 l water + 20 l oxygen) to the Department of Aquaculture (IFI Olsztyn, Poland) (transport time 2 h). The fish were placed in tanks with volumes of 0.2 m^3 in a RAS. During rearing in RAS, the following water parameters were maintained: temperature 19.7°C ± 0.1; pH range 7.80–8.01; oxygenation at rearing tank outflows did not decrease below 7.3 mg O_2 l^{-1}; total ammonia nitrogen concentration (TAN = NH_4+-N + NH_3-N) measured at rearing tank outflows did not exceed 0.2 mg TAN l^{-1}, and that of nitrites (NO_2-N) did not exceed 0.1 mg NO_2-N l^{-1}. The fish were fed daily for 16 h d^{-1} with an automated feeder (Fischtechnik GmbH, Nienburg, Germany). The feed used was T-T Nutra MP (Skretting, Holland) with a chemical composition of: protein – 50%, crude fat – 20%, cellulose – 2.4%, ash – 8%.

Experimental groups and tagging

After reaching approximately 70 g BW, all of the fish were anesthetized with etomidate (Propiscin, IFI Olsztyn, Polska, 0.2% etomidate) in an aqueous solution at a concentration of 1.5 ml l^{-1}, tagged intraperitoneally (near the first dorsal fin radius) with passive integrated transponders (PIT; Fish Eagle, Lechlade, Great Britain) (material – bio-glass; length – 12.0 ± 0.4 mm; diameter – 2.12 ± 0.07 mm; weight – 93 mg) (Zakęś and Hopko 2013). Seventy-two fish were tagged. Two experiments were conducted. In experiment I, fish standard length (SL) was 16.28 ± 0.68 cm (mean ± SD), and BW was 78.33 ± 10.01 g (mean ± SD), while in experiment II SL was 15.58 ± 0.51 cm, and BW was 73.07 ± 6.25 g. The two experiments differed in the method used to suture the incision after tags implantation. In experiment I, non-absorbable surgical silk (Jedwab Polski Sp. z o.o., Milanówek, Poland) was used to make two sutures to close the implantation incision, while in experiment II Surgibond tissue adhesive was used (SMI AG, St. Vith, Belgium). The fish in both experiments were surgically implanted with tags (declared weight 0.6 g, length 13 mm, diameter 5 mm; model F1515, ATS Inc., Isanti, MN, USA). The actual weight of the tags was 0.604-0.653 g (mean 0.63 g). The relative weight of the tags in both experiments was < 1.01% perch BW. Before tags implantation, the perch were anesthetized in an aqueous solution of etomidate (Propiscin, IFI Olsztyn, Poland) at a concentration of 1.5 ml l^{-1}. After 3-4 min, the fish were in a state of general anesthesia that was apparent from the lack of balance and no reaction to external stimuli (Rożyński et al. 2016). The transmitter was implanted in the body cavity through a 10-15 mm incision approximately 20 mm anterior to the base of the pectoral fin, and the antenna was directed outside the body cavity between the pectoral and anal fins (Wagner et al. 2011). After the procedure, the incision was disinfected with Betadine (Lavipharm, Peania, Greece). The length of the transmitter implantation procedure using the two methods was timed (± 1 s). After implantation the fish were held in containers (volume 0.08 m^3) with fresh, aerated water until they recovered basic vital functions. In each of the experiments, 18 individuals were tagged (experiment I – silk sutures, group ST; experiment II – tissue adhesive, group GT) and then stocked into three rearing tanks with volumes of 0.2 m^3 (6 individuals per tank). The fish from the control group (18 individuals tagged only with PIT; experiment I – group SC; experiment II – group GC) were subjected to the same procedure as the experimental fish (excluding the incision and transmitter implantation) and were stocked into three rearing tanks (6 individuals per tank). The mean stocking biomass in the tanks was 2.34 kg m^{-3} (experiment I) and 2.27 kg m^{-3} (experiment II). The fish were reared for six weeks in both experiments.

Measurements of water temperature (± 0.1°C) and oxygen concentration (± 0.01 mg O_2 l^{-1}) were taken daily at the rearing tank inflows and outflows. The other water parameters, i.e., TAN (± 0.01 mg TAN l^{-1}), NO_2-N (± 0.01 mg NO_2-N l^{-1}), and pH (± 0.01) were measured a the tank outflows weekly. In both experiments the mean water temperature was 21.0°C ± 0.1. The oxygen concentration at the tank outflows did not decrease below 7.43 O_2 l^{-1} (83.9% saturation) in experiment I or 7.36 O_2 l^{-1} (83.5% saturation) in experiment II. The oxygen concentration at

Table 1
Assessment criteria for silk suture/tissue adhesive retention, macroscopic assessment of incisions, and changes in implantation incisions of perch tagged with telemetry transmitters

Rank	Rating criterion
	Assessment criteria for silk suture/tissue adhesive retention (Deters et al. 2010)
0	Lack of silk sutures/tissue adhesive
1	Silk sutures/tissue adhesive partially cover incision
2	Silk sutures/tissue adhesive fully cover incision
	Macroscopic assessment criteria of incision site (Miller et al. 2014)
0	Incision fully closed and healed/no trace of incision
1	Incision fully closed but not healed
2	Incision healing, sides of incision only partially connected with tissue
3	Incision healing, but sides of incision not closed/not connected with tissue
4	Less than 50% of incision open
5	More than 50% of incision open
6	Incision fully open
	Assessment criteria for implantation incision
0	Clean incision
1	Some redness
2	Inflammation
3	Infection/necrosis

the tank inflows remained within the range of 90–98% saturation. The concentration of TAN and NO_2-N at the tank outflows did not exceed 0.08 mg TAN l^{-1} and 0.019 mg NO_2-N l^{-1} (experiment I) and 0.08 mg TAN l^{-1} and 0.013 mg NO_2-N l^{-1} (experiment II). Water pH at the tank outflows in experiment I ranged from 7.74 to 7.93, while in experiment II it was 7.75 to 7.99. The fish in both experiments were fed the same Aller Bronze 3 mm feed with the following chemical composition: protein – 45%, crude fat – 15%, carbohydrates – 24%, cellulose – 3%, ash – 7%, and a digestible energy of 17.6 MJ kg^{-1} (AllerAqua, Denmark). The feed was delivered with an automated feeder (Fischtechnik GmbH, Nienburg, Germany) for 18 h d^{-1} (09:00–03:00). The feed ratio was determined weekly at 1.5% fish biomass.

Study procedures

Immediately before transmitter implantation, all of the fish were measured (SL ± 0.1 cm) and weighed (BW ± 0.01 g). Measurements of individual fish were taken, and the state of the silk sutures or tissue adhesive (Table 1; Deters et al. 2010) and the healing of the implantation incision were assessed (Table 1; Miller et al. 2014) weekly (on day 7 (d7), d14, d21, d28, d35, d42). Throughout the experiment the occurrence of redness, edema, inflammation, infection, and/or tissue necrosis at the incision site was also monitored (Table 1). During measurements, each specimen was identified with a PIT tag reader (Fish Eagle, Lechlade, Great Britain. The data collected permitted calculating the following parameters: daily

growth rate – DGR (g d^{-1} = (BW_2 – BW_1) × t^{-1}; specific growth rate – SGR (% d^{-1}) = 100 × ($lnBW_2$ – $lnBW_1$) × t^{-1}; Fulton's condition coefficient – F = 100 × BW × SL^{-3}; feed conversion ratio – FCR = TFS × $(FB – IB)^{-1}$, where: BW_1 – initial fish body weight (g), BW_2 – final fish body weight (g), t – rearing time (days), SL – fish standard length (cm), FB – final stock biomass (g), IB – initial body mass (g), TFS – total feed supply (g). Additionally, the possible occurrence of PIT and/or transmitter and fish mortality in the rearing tanks was monitored daily.

On the final day of the experiment (d42), approximately 1 mm^2 of blood was drawn directly from the caudal vein of each specimen with a heparinized syringe (Smiths Medical International ASD, Inc., St. Paul, Minnesota, USA). The samples were used to determine the following hematological parameters: white blood cell count (WBC); red blood cell count (RBC); hemoglobin (HGB); hematocrit (HCT); platelets (PLT); and the following erythrocyte indexes: mean corpuscular volume (MCV); mean corpuscular hemoglobin (MCH); and mean corpuscular hemoglobin concentration (MCHC). Portions of the blood samples were centrifuged at a speed of 4000 rpm for 3 min (Fresco 17, Thermo Scientific, Waltham, USA). The material obtained was used to determine the following biochemical indexes: creatinine (CREA); total protein (TP); total bilirubin (BIL-T); alanine aminotransferase (ALT); alkaline phosphatase (ALP); calcium (Ca); albumin (ALB); globulin (GLB); glucose (GLU); magnesium (Mg); ammonia (NH3). Hematological measurements were done with a BC-2800 VET semi-automatic hematology analyzer (Mindray, Shenzhen, China), while biochemical measurements were done with a BS-120 automatic chemistry analyzer (Mindray, Shenzhen, China).

Statistical analysis

Statistical analysis was performed with Statistica 12 (StatSoft, Inc., USA). The data were tested to determine if they met the criteria for normal distribution. Levene's test was used to check the homogeneity of variance. The statistical significance of data on growth and incision status was verified with repeated measures variance analysis (ANOVA). Further analysis was performed with post-hoc Tukey's test. However, the statistical significance of the hematological and biochemical indices was checked with the Mann-Whitney U test.

Results

The implantation and suturing procedure in experiment I was, on average, 2 min 25 s (range 01:59 – 02:59), while that with tissue adhesive in experiment II was 1 min 14 s (range 01:02–01:45), which was about 50% of that in experiment I. In experiment I (silk sutures), no significant differences were noted between the growth, condition, or FCR parameters in group SC vs. group ST. This result refers to the specific weeks of the experiment and to the entire period during which the fish were held in the RAS (Table 2; $P > 0.05$). In experiment II (tissue adhesive), after the first week following tags implantation, the fish from group GT exhibited a significantly lower growth rate (DGR, SGR) and a higher FCR than in control group GC (Table 2; $P < 0.05$). Beginning in the second week, the values of these indexes in groups GT and GC were similar and remained so until the conclusion of the experiment (Table 2). Mortality was only noted in experiment II. One specimen each died in groups GT and GC. The losses occurred at the end of the experiment, i.e., in the fifth week in group GT and in the sixth in group GC (Table 2).

Statistically significant differences were confirmed when assessing the retention of silk sutures (group ST) or tissue adhesive (group GT) up to the third week of the experiment (Table 1; Fig. 1; $P < 0.05$). In group ST, the silk sutures were lost at an equal rate throughout experiment I, while in most cases in group GT the tissue adhesive peeled off within the first week of implanting the tags (experiment II). The loss of sutures, either silk or tissue adhesive, was directly correlated with transmitter shedding. During the course of the experiment, six specimens from group ST shed their transmitter, mainly in weeks three to five in experiment I (33.3%

Table 2

Rearing parameters of perch (*P. fluviatilis*) tagged with radio transmitter (experiment I – control group (SC) and group with silk sutures (ST) and experiment II – control group (GC) and group with tissue adhesive (GT)) on subsequent days of rearing (d0 – day of tag implantation, d7, d14, d21, d28, d35, d42, respectively, 7, 14, 21, 28, 35, 42 days after tag implantation) (mean values ± SD, n = 18). Details in Materials and methods section. Groups with different letter indexes within one experiment differ significantly statistically ($P \leq 0.05$)

	Experiment I		Experiment II	
Parametr / day of rearing	Group SC	Group ST	Group GC	Group GT
Standard lengh (cm)				
d0	16.02 (± 0.69)	16.28 (± 0.68)	15.88 (± 0.41)	15.58 (± 0.51)
d42	17.19 (± 0.79)	17.21 (± 0.83)	17.15 (± 0.61)	16.61 (± 0.44)
Body weight (g)				
d0	80.92 (± 10.63)	77.70 (± 10.020)	76.79 (± 6.83)	72.44 (± 6.26)
d7	83.49 (± 11.88)	78.68 (± 10.75)	79.56 (± 6.98)	72.81 (± 5.94)
d14	86.86 (± 12.54)	81.77 (± 11.76)	82.72 (± 7.37)	75.92 (± 6.75)
d21	90.26 (± 12.99)	84.98 (± 12.90)	85.13 (± 8.22)	77.72 (± 6.96)
d28	93.76 (± 14.25)	88.52 (± 14.19)	87.20 (± 8.95)	80.33 (± 6.78)
d35	97.35 (± 15.61)	91.68 (± 15.17)	90.46 (± 9.59)	82.96 (± 7.39)
d42	100.97 (± 16.46)	95.22 (± 16.59)	93.54 (± 11.50)	85.37 (± 7.63)
Daily growth rate (g d^{-1})				
d0-d7	0.37 (± 0.37)	0.14 (± 0.24)	0.39 (± 0.20)^{b}	0.05 (± 0.27)^{a}
d7-d14	0.48 (± 0.28)	0.44 (± 0.30)	0.45 (± 0.18)	0.44 (± 0.27)
d14-d21	0.49 (± 0.25)	0.46 (± 0.24)	0.34 (± 0.18)	0.26 (± 0.21)
d21-d28	0.50 (± 0.26)	0.51 (± 0.26)	0.30 (± 0.19)	0.37 (± 0.20)
d28-d35	0.51 (± 0.29)	0.45 (± 0.27)	0.47 (± 0.23)	0.39 (± 0.19)
d35-d42	0.52 (± 0.43)	0.51 (± 0.27)	0.38 (± 0.38)	0.34 (± 0.32)
d0-d42	0.48 (± 0.25)	0.42 (± 0.23)	0.39 (± 0.19)	0.30 (± 0.14)
Specific growth rate (% d^{-1})				
d0-d7	0.43 (± 0.44)	0.16 (± 0.29)	0.51 (± 0.24)^{b}	0.08 (± 0.37)^{a}
d7-d14	0.56 (± 0.33)	0.54 (± 0.36)	0.56 (± 0.20)	0.59 (± 0.35)
d14-d21	0.55 (± 0.32)	0.53 (± 0.26)	0.40 (± 0.19)	0.33 (± 0.27)
d21-d28	0.53 (± 0.26)	0.56 (± 0.26)	0.34 (± 0.20)	0.48 (± 0.25)
d28-d35	0.52 (± 0.28)	0.49 (± 0.29)	0.52 (± 0.24)	0.48 (± 0.22)
d35-d42	0.51 (± 0.45)	0.52 (± 0.25)	0.38 (± 0.45)	0.41 (± 0.38)
d0-d42	0.51 (± 0.27)	0.47 (± 0.24)	0.45 (± 0.19)	0.39 (± 0.17)
Fulton's condition coefficient				
d0	1.96 (± 0.12)	1.80 (± 0.11)	1.91 (± 0.10)	1.92 (± 0.14)
d7	1.92 (± 0.12)	1.80 (± 0.12)	1.91 (± 0.07)	1.91 (± 0.15)
d14	1.95 (± 0.12)	1.86 (± 0.11)	1.92 (± 0.07)	1.89 (± 0.16)
d21	1.91 (± 0.14)	1.84 (± 0.11)	1.77 (± 0.08)	1.76 (± 0.12)
d28	1.93 (± 0.13)	1.84 (± 0.12)	1.86 (± 0.08)	1.89 (± 0.14)
d35	1.94 (± 0.15)	1.88 (± 0.13)	1.87 (± 0.09)	1.88 (± 0.12)
d42	1.97 (± 0.14)	1.85 (± 0.13)	1.85 (± 0.09)	1.86 (± 0.13)

Cont. Table 2

	Experiment I		Experiment II	
Parametr / day of rearing	Group SC	Group ST	Group GC	Group GT
Feed conversion ratio*				
d0-d7	4.59 (± 3.84)	12.64 (± 8.65)	2.84 (± 0.69)[a]	16.56 (± 13.57)[b]
d7-d14	2.70 (± 0.65)	3.01 (± 1.34)	2.68 (± 0.29)	2.50 (± 0.30)
d14-d21	2.73 (± 0.44)	2.88 (± 0.90)	4.15 (± 1.88)	5.02 (± 2.22)
d21-d28	2.72 (± 0.23)	2.73 (± 0.67)	5.11 (± 2.70)	4.08 (± 2.76)
d28-d35	2.75 (± 0.22)	2.98 (± 0.34)	2.80 (± 0.19)	3.11 (± 0.36)
d35-d42	3.12 (± 1.21)	2.84 (± 0.62)	3.65 (± 1.37)	3.57 (± 1.13)
d0-d42	2.82 (± 0.51)	3.13 (± 0.70)	3.28 (± 0.80)	3.73 (± 0.58)
d28-d35	-	1 (5.6)	-	0
d35-d42	-	2 (11.1)	-	0
d0-d42	-	6 (33.3)	-	14 (77.8)
Tag loss rate (ind., %)				
d0-d7	-	0	-	0
d7-d14	-	0	-	4 (22.2)
d14-d21	-	1 (5.6)	-	7 (38.9)
d21-d28	-	2 (11.1)	-	3 (16.7)
d28-d35	-	1 (5.6)	-	0
d35-d42	-	2 (11.1)	-	0
d0-d42	-	6 (33.3)	-	14 (77.8)
Survival (%)				
d0-d42	0	0	5.6	5.6

*FCR was calculated for rearing tanks (n = 3)

tagged fish), while in group GT, 14 specimens (77.8%) loss their tag, mainly in weeks two to four in experiment II (Table 2).

Observations of incision healing indicated that only in week two were there statistically significant differences in the speed of incision healing (Table 1; Fig. 2; $P < 0.05$). Significant progress in incision healing from rank 5 to 2 was noted in group ST. However, after the second week of the experiment little progress was observed in the fish from group GT with incision healing progressing from rank 5 to between ranks 5 and 4. After the third week following tag implantation, the incision healing assessments of the two groups were very similar, and the healing rate in both groups oscillated around rank 2 (Table 1; Fig. 2; $P > 0.05$). No further statistically significant differences were noted through to the end of the experiment, and observations of both groups indicated that incision healing after six weeks was rank 1 (Table 1; Fig. 2; $P > 0.05$).

The percentage of specimens in each of the experiments exhibiting some redness at the tag implantation incision site was similar (Table 3; Fig. 3; $P > 0.05$). The most specimens exhibiting this symptom were noted after the first week of the experiment (experiment I – 66.7%, experiment II – 61.1%). In subsequent weeks, the number of fish with this symptom decreased. After six weeks, only one specimen from both experiments exhibited this symptom (5.56%). However, clear inflammation was observed only in experiment II (tissue adhesive) in three specimens. In the first week after implantation, two specimens from

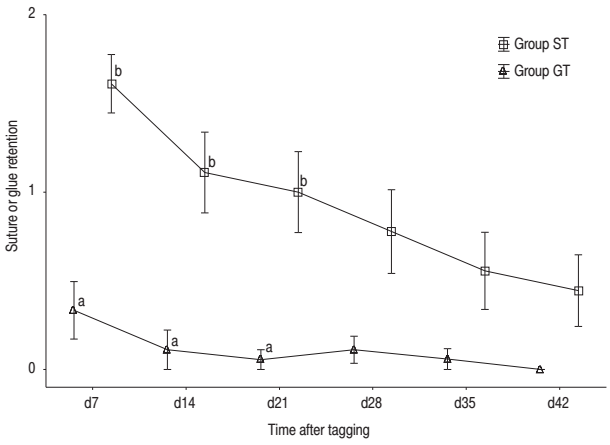

Figure 1. Retention of silk sutures or tissue adhesive in two groups of perch (*P. fluviatilis*) in which two suturing methods were used after transmitter implantation (silk suture group (ST) and tissue adhesive group (GT)) (see Table 1). Groups with different letter indexes within one week differ significantly statistically ($P \leq 0.05$) (mean values ± SE).

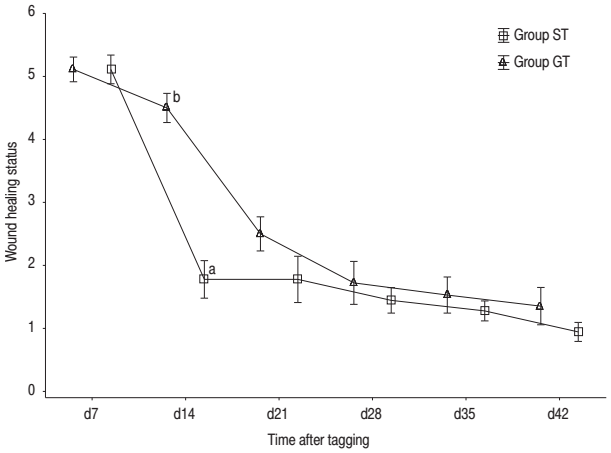

Figure 2. Incision healing in two groups of perch (*P. fluviatilis*) in which two suturing methods were used after transmitte implantation (silk suture group (ST) and tissue adhesive group (GT)) (see Table 1). Groups with different letter indexes within one week differ significantly statistically ($P \leq 0.05$) (mean values ± SE).

group GT exhibited this symptom (which subsided after a week), while in the second week of the experiment this symptom appeared in another specimen and persisted until the end of the experiment. Difference between groups ST and GT throughout the experiment were not statistically significant (Fig. 3; $P > 0.05$). Throughout the experiment, no specimens in either group were noted to have had infection or necrosis at the transmitter implantation incision site.

No statistically significant differences were noted among the hematological parameters tested in experiment I (Table 3; $P > 0.05$), while in experiment II significant differences were noted in the values of MCV and PLT (Table 5; $P < 0.05$). The MCV in group GT was approximately 6% lower than in control group GC, while the PLT value in group GT compared to that in group GC was approximately 26% lower.

The biochemical tests indicated significantly lower activity (by approximately 40%) of the enzyme ALP and lowered magnesium concentrations (by approximately 10%) in group ST in experiment I (Table 4; $P < 0.05$). No statistically significant intergroup differences were noted in any of the other parameters tested in experiment I or in any of those tested in experiment II (Table 4; $P > 0.05$).

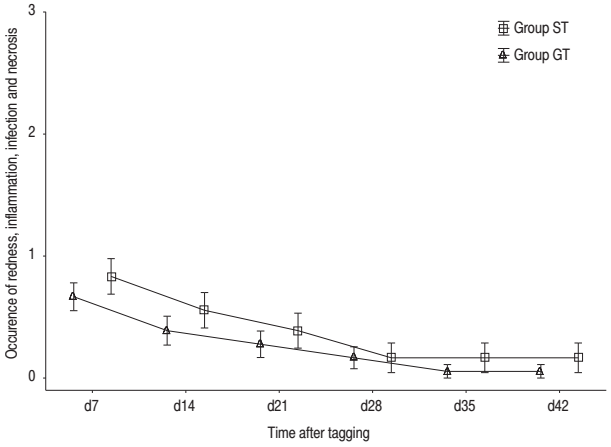

Figure 3. Occurrence of redness, inflammation, and infection/necrosis in two groups of perch (*P. fluviatilis*) in which two suturing methods were used after transmitter implantation (silk suture group (ST) and tissue adhesive group (GT)) (see Table 1). Groups with different letter indexes within one week differ significantly statistically ($P \leq 0.05$) (mean values ± SE).

Discussion

The advantage of using surgical tissue adhesive to suture transmitter implantation incisions is undoubtedly the brevity of the procedure. In the current study, the average tag implantation time when using tissue adhesive was half as long as that when using

Table 3

Hematological indexes in two groups of perch (*P. fluviatilis*) in which different transmitter implantation suturing methods were applied (experiment I – control group (SC) and silk suture group (ST) and experiment II – control group (GC) and tissue adhesive group (GT)) (mean values ± SD, n = 18). Details in Materials and methods section. Groups with different letter indexes within a given experiment differ significantly statistically ($P \leq 0.05$)

		Experiment I		Experiment II	
Parameters	Units	Group SC	Group ST	Group GC	Group GT
WBC	$10^3\ \mu l^{-1}$	127.66 (± 15.32)	133.74 (± 19.42)	125.96 (± 16.36)	120.83 (± 25.72)
RBC	$10^6\ \mu l^{-1}$	1.67 (± 0.19)	1.68 (± 0.14)	1.58 (± 0.14)	1.46 (± 0.17)
HGB	$g\ l^{-1}$	38.09 (± 5.68)	38.56 (± 3.79)	39.38 (± 6.21)	33.91 (± 4.22)
HCT	%	30.76 (± 3.95)	31.18 (± 2.69)	30.56 (± 4.09)	26.41 (± 3.67)
MCV	Fl	142.21 (± 7.35)	143.41 (± 4.74)	148.82 (± 10.50)^b	139.90 (± 8.76)^a
MCH	Pg	27.76 (± 2.36)	27.99 (± 1.59)	30.21 (± 3.05)	28.42 (± 2.66)
MCHC	$g\ l^{-1}$	195.17 (± 9.48)	195.46 (± 8.33)	203.00 (± 9.39)	203.30 (± 10.65)
PLT	$10^3\ \mu l^{-1}$	24.00 (± 6.38)	25.08 (± 9.00)	21.38 (± 6.06)^b	15.90 (± 3.67)^a

Table 4

Biochemical indexes in two groups of perch (*P. fluviatilis*) in which different transmitter implantation suturing methods were applied (experiment I – control group (SC) and silk suture group (ST) and experiment II – control group (GC) and tissue adhesive group (GT) (mean values ± SD, n = 18). Details in Materials and methods section. Groups with different letter indexes within a given experiment differ significantly statistically ($P \leq 0.05$)

		Experiment I		Experiment II	
Parameters	Units	Group SC	Group ST	Group GC	Group GT
CREA	$mg\ dl^{-1}$	0.15 (± 0.09)	0.17 (± 0.16)	0.43 (± 0.99)	0.11 (± 0.08)
TP	$g\ dl^{-1}$	4.34 (± 0.62)	4.08 (± 0.45)	3.72 (± 1.27)	4.01 (± 0.20)
BIL-T	$mg\ dl^{-1}$	0.17 (± 0.10)	0.22 (± 0.16)	0.23 (± 0.16)	0.18 (± 0.07)
ALT	$U\ l^{-1}$	33.29 (± 39.13)	21.77 (± 46.07)	28.86 (± 32.45)	23.25 (± 31.24)
ALP	$U\ l^{-1}$	42.79 (± 13.83)^b	25.54 (± 9.51)^a	35.86 (± 16.20)	37.33 (± 23.97)
Ca	$mg\ dl^{-1}$	12.56 (± 1.09)	11.98 (± 0.77)	11.94 (± 1.54)	11.58 (± 0.59)
ALB	$g\ dl^{-1}$	1.60 (± 0.19)	1.54 (± 0.14)	1.41 (± 0.25)	1.49 (± 0.08)
GLOB	$g\ dl^{-1}$	2.74 (± 0.46)	2.54 (± 0.35)	2.58 (± 0.49)	2.52 (± 0.16)
GLU	$mg\ dl^{-1}$	146.07 (± 63.64)	116.38 (± 49.06)	123.93 (± 53.03)	102.58 (± 53.62)
Mg	$mg\ dl^{-1}$	2.49 (± 0.33)^b	2.24 (± 0.32)^a	2.54 (± 0.27)	2.54 (± 0.31)
NH_3	$\mu g\ dl^{-1}$	487.04 (± 93.30)	420.42 (± 132.56)	470.32 (± 138.03)	388.38 (± 86.77)

silk sutures. Fish tagging procedures that are too long can result in various undesirable side effects including increased exposure to stress and prolonged recovery from anesthesia that can even result in death (Neely et al. 2009). Noteworthily, even the time required to perform the transmitter implantation procedure and suturing with surgical silk (2 min 25 s) in the current study was within the time period recommended in the literature for this type of procedure (Sandstrom et al. 2013).

Low stock biomass growth was noted in both groups, but particularly in group GT, of fish tagged with radio transmitter after the first week of the experiment in comparison to the control groups. A slight decrease in the value of this parameter was even observed in one of the tanks stocked with fish from group GT; however, 14 days after tagging the perch with radio transmitter and until the conclusion of the study, differences in growth rates were not significant. Discernibly lower fish body weight increases shortly following tagging are reported in other studies of percid fishes, which were tagged with, for example, PIT (Baras et al. 2000, Zakęś and Hopko 2013). Similar observations are reported in juvenile Chinook salmon, *Oncorhynchus tshawytscha* (Wal.) (BW 50.5 g) tagged with acoustic transmitter (3.6% of the fish BW), when lower body weight increases in the tagged fish group occurred until day 35 of the experiment. However, despite the lower values of this parameter in the first phase of the experiment, similarly to the case of the current study, the final body weight values were comparable to those observed in the control group of untagged fish (Ammann et al. 2013). Tagging juvenile European seabass, *Dicentrarchus labrax* (L.) (BW 173 g) intraperitoneally with dummy tags also did not impact final body weight values. However, lower growth was observed in a group of fish tagged with external tags (Anras et al. 2003). Similarly, juvenile Green sturgeon, *Acipenser medirostris* Ayres (BW 347 g) were studied to assess the impact surgical implantation transmitter (1.31% of the fish BW) had on growth after 140 days of rearing; no statistically significant differences were noted in either total length or body weight in any of the groups (Miller et al. 2014). The results of the current study could indicate that the reaction of a given species to tag implantation could also depend on the method used to suture the incision after implantation. It was precisely in group GT in the first week following radio transmitter implantation that significantly lower values of growth indexes DGR and SGR were noted. The FCR coefficient that was nearly sixfold higher than that in the control group should be noted as it undoubtedly indicates that the fish in this group consumed only a slight amount of food in the first week of the experiment. Similar results are reported for juvenile European seabass (BW 173 g) tagged with intraperitoneal transmitter (2.4% of the fish BW). In the first weeks after tags implantation higher values were noted in FCR, and these did not decrease until three weeks following tagging (Anras et al. 2003). The phenomenon of a significant decrease in growth and FCR indexes were not, however, noted in juvenile pikeperch, *Sander lucioperca* (L.) (BW 60 g) that were subjected to the same radio transmitter tagging procedure as the perch in the present study (M. Rożyński et al. unpublished materials). This could indicate that the reaction to the tag implantation procedure could be species specific.

It is noteworthy that high FCR values were confirmed in all of the groups, including the controls, in the current study, and they were higher than those routinely noted when feeding perch formulated feed (Policar et al. 2015). It is highly likely that this can be explained by the frequent manipulations the fish were subjected to during the weekly individual examinations.

High levels of tag rate were noted in juvenile perch. During the six weeks of the experiment, 33.3% of the fish in group ST loss their tags, while in group GT 77.8% of them did. In experiment I, the radio tags were lost between weeks three and six, while in experiment II this occurred between weeks two and four. Tag loss rate over such a long period of time during the experiment was undoubtedly linked with the use of silk sutures and tissue adhesive at the transmitter implantation site. The silk sutures

remained in the incision site longer and held the two edges of the incision longer than did the tissue adhesive, which, in most instances, peeled off during the first week of the experiment.

High tag loss was observed in juvenile rainbow trout, *Oncorhynchus mykiss* (Wal.) tagged with acoustic tags of various sizes (Sandstrom et al. 2013). Throughout the 143-day study, in the group of trout (BW 112 g) tagged with larger tags (2.3% of the fish BW) 25% of the specimens shed tags (10 of 40 specimens), while in the group of smaller fish (BW 108 g) tagged with smaller tags (1.7% of the fish BW) 15% of the fish shed tags (6 of 40 specimens). No differences in body weight among the specimens that retained or lost tags were confirmed in either group (Sandstrom et al. 2013). In the current study, most of the tags were shed through the implantation incision in both experiments, and only in one case was the tag lost through the antenna exit site. Some of the tags in the rainbow trout were shed through the implantation incision, while the other tags were lost from other sites (Sandstrom et al. 2013). Chisholm and Hubert (1985) report that tags can also be shed through the digestive tract. However, in the present study such a case was not observed. No tag shedding was noted in Chinook salmon (BW 50.5 g) throughout the 221-day study (Ammann et al. 2013). These authors attribute the high tags retention to low water temperature and fish activity. In another study conducted on the same species in a similar developmental stage, but in which the fish were held at higher temperatures, numerous tags were shed. The high tags shedding in the current study can be explained by perch behavior. This species is a shoaling fish, and interactions among individuals are strong (Magnhagen 2015). Observations of the tanks in which the fish were held indicated that the perch attacked the radio tags antennae of other specimens. This resulted in the tag moving inside the body cavity, slower healing or separation of the implantation incision, and, in extreme cases, tag shedding. It should be noted that in pikeperch of a similar size that were subjected to the same radio transmitters implantation procedure and held under the same conditions, tag retention (after 35 days) in specimens in which incisions were sutured with either silk sutures or surgical tissue adhesive was > 90% (M. Rożyński et al., unpublished materials). This fact confirms the significant impact that the behavior of a given species has on the outcome of using telemetry tags.

In both groups of tagged fish both the silk sutures and the tissue adhesive remained in place at the incision site for a short time. Most of the specimens group ST lost the silk sutures in the second or third week after implantation, while the surgical tissue adhesive (group GT) peeled off in the first week after the procedure. From the fourth week, the rank of the assessment of silk sutures and tissue adhesive was 0 (lack of sutures) or 1 (silk sutures/tissue adhesive partially cover incision) (Deters et al. 2010). While tissue adhesive usually remains in place on an incision for about one week, closing an incision with non-absorbable sutures should be more durable (Thorstad et al. 2013). In rainbow trout (BW 100 g) the majority of non-absorbable sutures were lost during a 64-day study, and only in two specimens (5%) did the sutures remain in place until the conclusion of the experiment (d64) (Sandstrom et al. 2013). In a study of largemouth black bass, *Micropterus salmoides* (Lacepède) implanted with acoustic telemetry devices, Caputo et al. (2009) observed sutures as long as a year after the implantation procedure (17.6%). The fast suture loss in the current study of perch was probably the result of interactions among individuals, which included specimens grabbing and tugging the ends of the sutures that they reacted to as if they were potentially food.

In experiment I, the incisions of most of the specimens were already healing (rank 2) in the second week of the study, and slight progress was noted in subsequent weeks; however, during this time six fish shed their tags through the implantation incision. In experiment II, numerous tags were lost through the implantation incision, because the tissue adhesive peeled off in the first week of the experiment before the edges of the incision were closed (rank 4-5), and numerous tags were shed through the implantation incision. In group GT, incision healing (rank 2) was

observed in the third and fourth weeks, but most of the specimens had, by this time, shed their tags from the body cavity. On the last day of the experiment, 22% of the specimens in group ST had fully healed incisions (the incision site was practically imperceptible), while 61% of them were healed and the incision was visible. In comparison, with group GT, fully healed incisions were noted in a smaller number of specimens (6%), while healed incisions in which the incision remained visible was noted in a greater number of specimens (82%). In juvenile European seabass (BW 173 g), 30% of specimens had fully healed implantation incisions after 47 days (end of experiment) (Anras et al. 2003), while in Chinook salmon (BW 50.5 g) tagged intraperitoneally with acoustic tags, all incisions (100%) were fully closed after 34 days of the experiment and at 104 days the incisions were barely perceptible (100%) (Ammann et al. 2013). In the present study, the healing of implantation incisions was successful, without complications caused by infection, and the incisions were in similar states in the various weeks of the experiment in both groups of fish (ST, GT) tagged with radio tags. As early as two weeks after tag implantation, the incisions of most of the specimens (61-51%) were clean and partially healed. Incision redness was noted in approximately 39% of the fish in group ST and 44% of the fish from group GT. Additionally, three specimens from group GT were observed to have inflamed incisions (in two fish until the second week of the study, and in one perch from weeks two to six). These symptoms were evidently linked with the relatively easy opening of the implantation incisions that had been sutured with tissue adhesive and tag shedding. It can be concluded that both of the implantation suturing methods were safe for perch, but better results (e.g., higher tag retention) can be achieved when silk surgical sutures are used.

Changes in the hematological profile of perch tagged with radio tags in comparison to the control group were only observed in group GT. Lower MCV and PLT values could be evidence of lowered iron levels and disruptions in water-electrolyte balance. The causes of these changes were doubtless the longer incision healing time in group GT that resulted in excessive blood loss (Neves et al. 2016). ALP activity and systemic magnesium concentration are closely correlated in the body, and contribute to the creation of bone tissue and the maintenance of homeostasis. The lowered levels of these two indexes in group ST could indicate impaired liver function associated with the synthesis of factors responsible for systemic balance, increased protein catabolism, and wound healing. Low ALP activity could stem directly from a decelerated rate of glycogen synthesis, malnutrition, or the wasting/exhaustion of the body (Shaffi 1979).

The radio tags implantation method used in juvenile perch with two methods of implantation suturing did not negatively affect growth parameters or physiological processes. Despite the high percentage of fish with healed incisions in both experiments, the high number of tags shed means that neither of the two methods studied can be recommended for radio tags implantation in perch as was done in the study. For perch it might be more effective, for example, to use transmitters with internal antennae. The implantation suturing method also needs to be improved to increase the tags retention index.

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