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Distribution of juvenile river lamprey (*Lampetra fluviatilis* L.) in different habitats

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The impact of anthropogenic pressure on the distribution of juvenile river lamprey (*Lampetra fluviatilis* L.) in different habitats was investigated. All samples were collected in the midstream of the Minija from June to September 2006. The research covered about 100 meters of the river where, according to the local microenvironment, 20 investigation points were chosen. All the points differed from each other by water depth, the type of substratum, spread of vegetation coverage, and anthropogenic pressure. The highest density of individuals was typical of points with no anthropogenic pressure, in which the substratum contained coarse sand. In such points the density of individuals reached 2 ind m^{-2}. The ANOVA table showed a statistically significant correlation between the density of individuals and anthropogenic pressure ($F = 4.37$, $p < 0.05$). In most points, the biomass was about 200 mg m^{-2}. The study results have shown that lamprey juveniles prefer coarse sand. In such type of substratum, the mean density was about 2 ind m^{-2}. In the substratum that mostly contained pebble, no individuals were found. Statistical analysis showed that the correlation between the length of individuals and water depth was statistically significant ($r = 0.13$, $p < 0.05$). The research showed that lamprey juveniles avoid shallow water. Vegetation coverage in lamprey habitations was a positive factor ($F = 7.65$, $p < 0.05$).

Key words: anthropogenic pressure, river lamprey, *Lampetra fluviatilis* L., water depth, substratum type, spread of vegetation

INTRODUCTION

Anthropogenic impact as an ecological factor has acquired a higher significance during the recent decades. Therefore, research of the areas that experience an anthropogenic influence gives valuable information for environment preservation. Mankind, by intensively exploiting and changing the natural environment for the satisfaction of its needs, broke the balance of the natural factors, which has been formed during a long time. This had a fatal effect on the environment, men and animal communities, on certain species. The greatest threat arose to the species of which a complex reproductive cycle is typical (related with long trips from the sea to fresh waters or in fresh water systems), and which can survive only in habitats of a certain structure or which are very sensitive to changes of water quality (Virbickas et al., 2006).

One of the main problems while trying to preserve the species diversity is protection of the habitats that are best suitable for these species (Primack, 1995; Washitani, Yahara, 1996). In Lithuania, there are 24 species of cyclostomes and fish which our country undertook to protect after joining the Bern Convention and the European Union. One of these species is river lamprey (*Lampetra fluviatilis* L.). Although there is no threat of the disappearance of this species in our waters, their spreading is strongly limited by the size, water body type, obstacles on their migration ways, and also by water quality. As the experience of the Western European countries shows, these fish disappear very rapidly if the river flow is regulated or when the water pollution increases (Virbickas et al., 2006).

Hitherto, in Lithuania there have been no researches into the sensitiveness of juvenile lamprey to the anthropogenic factors and elucidation of the most suitable habitats. Lampreys during the juvenile period are most sensitive and can be easily hurt. The inability of juveniles to survive till maturity affects the population's abundance. As long as Lithuania undertakes to protect the species which has become extinct on the EU scale and lives naturally on different biotopes, this work is important for the evaluation of the significance of the mentioned factors for lamprey larvae and also for the protection of this species.

MATERIALS AND METHODS

The samples were collected in June–September of 2006 in the Middle Minija each second week. In this place the Minija makes a bend, therefore the velocity of its current is not great. The width of the riverbed is 6–7 meters, the average depth being 1.5 m. The bottom consists of coarse sand and gravel, single stones. The river's creek is sandy, the slopes are steep, and on the terrace there is a meadow with bushes and trees.

Lamprey juveniles were gathered in the creek of the Minija River, on its right bank, along a 100-m segment of the river, from 20 places situated each 4–6 meters (Sugiyama, Goto, 2002)

(Fig. 1). Sampling places were chosen taking into account the anthropogenic load in the place. This place is attractive for recreation (the bank and the riverside are sandy, without any macrophytes), a place for the mooring of canoes and a rest place are provided. The study segment of the river distinguishes itself by the biotopical diversity: different macrophytes overgrowing, the size of substratum particles, water depth.

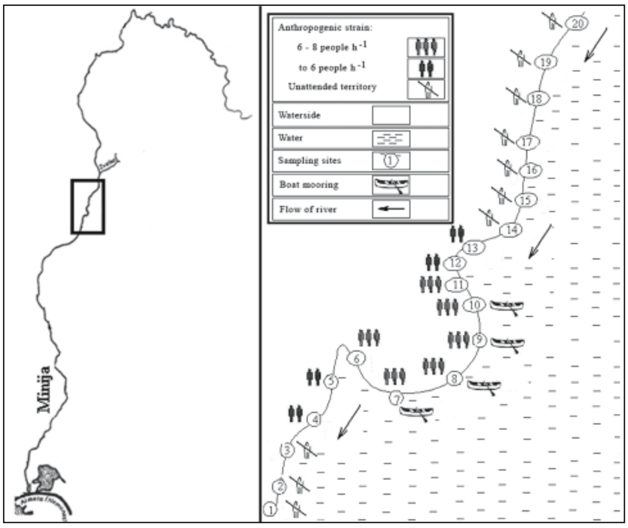

Fig. 1. Study area. Sampling site

In each site, frames 1 m^{-2} size were put and a sand stratum 25 cm deep was sifted. The found individuals were collected and fixed with 70% ethyl alcohol solution. 180 river lamprey samples were analysed. The species were defined at the laboratory (Virbickas, 2001), the dried up individuals were weighed (mg, error 0.01), their length was measured (mm, error 0.1). The individuals were assigned to two length groups, which corresponded to the individuals' age (Sugiyama, Goto, 2002): group I – individuals whose length was up to 5 cm, corresponding to the age less than 2 years and group II – individuals larger than 5 cm aged more than 2 years.

The covering of the places by macrophytes was evaluated visually, and in the data analysis this feature was named as the presence or absence of macrophytes.

According to the granulometric structure of the substratum (Gaigalas, 1995), these sites were divided into four types: 1 – small sand (0.25–0.1 mm); 2 – coarse sand (1–0.5 mm); 3 – gravel (10–1 mm); 4 – pebble (10 cm–10 mm).

The anthropogenic load in these sites was calculated, with regard to the number of people in the study part of the river (those who travelled by canoes and those who walked). The observations were made on sampling days, in the day-time. The number of people during the study period was recalculated into the average rate of visiting people per hour. Anthropogenic load in these places was divided into three types: high – 6–8 people per hour; average – less than 6 people per hour; without any anthropogenic load – unattended territory.

For the evaluation of individuals' density, one-factor dispersion analysis (ANOVA) was used to estimate the average density values, taking into account the limiting values of one independent variable. If the sites differed by two factors (by the type of anthropogenic load and by the type of substratum, two-factor dispersion analysis was used. This analysis permits to evaluate whether the means of dependent interval variables differ in different places, and to ascertain the interaction of the independent factors. All sample pairs were compared by Fisher's LSD (least significant difference) criterion, using the t criterion (when ANOVA shows that the difference of means is statistically significant).

With the help of cluster analysis, similarity by individuals' density in different places was established. The similar places went to the same cluster and the remote ones to the different clusters. The method of centre clusterization was chosen, and according to it the mean values of observation data were calculated in a cluster, and the distance between the clusters was calculated as the distance between these mean values.

For the evaluation of the strength of the correlation among the variables, Pearson's coefficient of linear correlation (r) was used.

RESULTS

The distribution of individuals in the study sites

The distribution of river lampreys in the study sites differing by several biotope's parameters (anthropogenic load intensity, substratum type, water depth, covering with macrophytes) was evaluated (Table 1).

It was established that the average individuals' density in the different sites varied from 0 to 4 ind m^{-2}. In the places where there were no macrophytes, the density fluctuated from 0 to 2 ind m^{-2}, and the type of substratum changed from sand to gravel. In

Table 1. **Characteristics of biotops and river lamprey density ($\overline{x} + s$). Anthropogenic pressure: A – 6–8 people h^{-1}, B – up to 6 people h^{-1}, C – unattended territory. Type of substratum: 1 – fine sand (0.25–0.1 mm), 2 – coarse sand (1 – 0.5 mm), 3 – gravel (10 – 1 mm), 4 – pebble (10 cm – 10 mm); Spread of vegetation cover: – site without vegetation, + site with vegetation**

Site	River lamprey density (ind m^{-2})	Anthropogenic pressure (people h^{-1})	Type of substratum	Spread of vegetation coverage
1	0.0 ± 0.00	B	4	–
2	1.3 ± 1.33	B	3	–
3	1.7 ± 1.67	B	3	+
4	4.0 ± 2.65	B	2	+
5	2.0 ± 1.73	B	1	–
6	1.0 ± 1.73	A	2	–
7	0.0 ± 0.00	A	2	–
8	0.0 ± 0.00	A	2	–
9	0.0 ± 0.00	A	2	–
10	0.0 ± 0.00	A	2	–
11	2.0 ± 1.00	A	2	–
12	2.0 ± 1.00	B	2	+
13	2.3 ± 2.52	B	2	+
14	1.3 ± 1.53	C	1	+
15	1.0 ± 1.73	C	1	+
16	0.7 ± 1.15	C	1	+
17	3.3 ± 2.52	C	3	+
18	0.7 ± 1.15	C	2	+
19	1.7 ± 2.89	C	2	+
20	2.7 ± 2.52	C	3	+

the study site with the highest anthropogenic load (6–11 points), the substratum consisted of coarse sand with no macrophytes, and the individuals' density per m^{-2} was 0. The fluctuations of individuals' density in different sites during the different periods of sampling were larger than between sites (0 to 8 ind m^{-2}).

Using the method of group means cluster analysis, we tried to check which sites were most similar by individuals' density. When the Euclidean distance is 1, the study sites make up 7 groups by individuals' density. The most remote site had the highest individuals' density (Fig. 2). This site made a separate group – the 4th one in which the individuals' density was 4 ± 2.65 ind m^{-2}. In this site, the coarse sand substratum prevailed (particle size 0.25–2.00 mm); anthropogenic load was average (less than 6 persons per hour). Few things distinguish the 17th and 20th sites (Euclidean distance 0.8); their densities are respectively 4 ± 2.52 ind m^{-2} and 3.3 ± 2.52 ind m^{-2}. Here, the type of substratum (gravel) and the intensity of anthropogenic load (sites without any anthropogenic load) were similar. The largest group was formed by sites 14, 15, 16, 18 and 19 in which the density varied from 0.67 to 1.67 ind m^{-2}. All these sites differed from the others by the minimal number of visiting people, but the substratum changed from fine sand to coarse sand. The 1st site made a separate group; it differed sharply by the substratum of the largest granulometric structure and by the load of average intensity. The characteristics of sites 7–10 matched absolutely: here the substratum was coarse sand and the anthropogenic load was the highest. There were no individuals in this site.

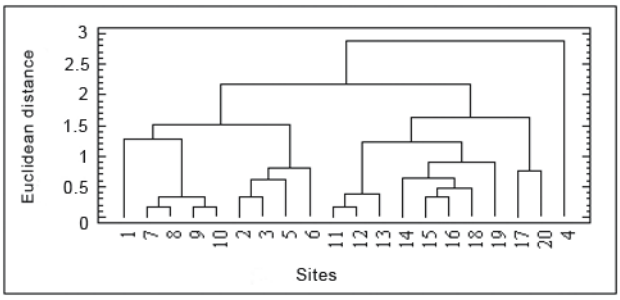

Fig. 2. Dendrogram of sites (density ind m^{-2})

The choice of the habitat by substratum type

The sites in which the substratum was coarse sand differed from others by the highest individuals' density (2.1 ind m^{-2}) (Fig. 3). Meanwhile in the sites in which the substratum consisted mainly of pebble there were no individuals.

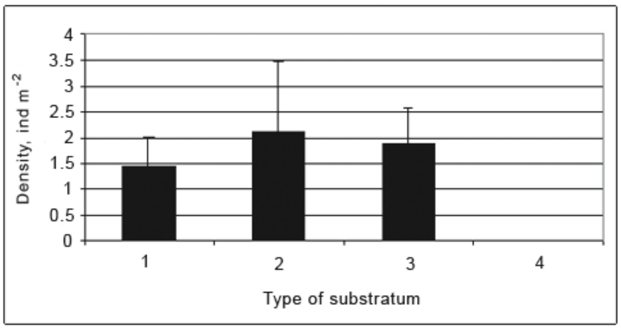

Fig. 3. River lamprey density ($\overline{x} + s$) in different types of substratum: (1 – fine sand (0.25 – 0.1 mm), 2 – coarse sand (1 – 0.5 mm), 3 – gravel (10 – 1 mm), 4 – pebble (10 cm – 10 mm)

The density of individuals and anthropogenic load

More than 50% of all detected individuals were found in the sites in which there were no resorting people. The individuals gathered in the sites of the highest load made approximately 5% of all individuals. The average individuals' density by the load was similar (2 ind m^{-2}) to that of sites with the average load and to of sites without any anthropogenic load (Fig. 4).

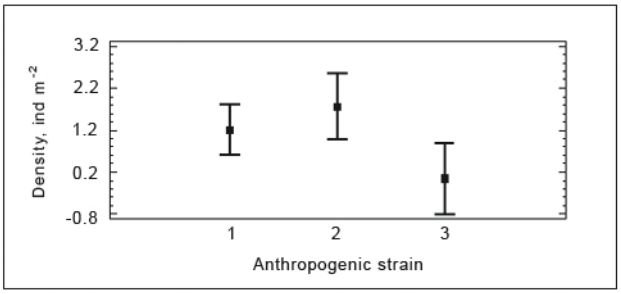

Fig. 4. River lamprey density ($\overline{x} + s$) in sites with different anthropogenic pressure: 1 – unattended territory; 2 – up to 6 people h^{-1}; 3 – 6–8 people h^{-1}

However, although the mean values of these fields were similar, in the sites not affected by the anthropogenic load the values of the density were more remote from the mean. The sites with the highest density were those in which the anthropogenic load was evaluated as the average or as zero. In the sites with a high load there were no individuals or their density was 0.2 ind m^{-2}.

When two factors – substratum type and the load – were accounted for the density was the highest (3 ind m^{-2}) in the substratum of coarse sand in the absence of anthropogenic load (Table 2).

Table 2. **River lamprey density ($\overline{x} + s$) in the sites with different anthropogenic pressure (A – 6–8 people h^{-1}, B – up to 6 people h^{-1}, C – unattended territory) and different type of substratum: 1 – fine sand (0.25 – 0.1 mm), 2 – coarse sand (1 – 0.5 mm), 3 – gravel (10 – 1 mm), 4 – pebble (10 cm – 10 mm)**

Anthropogenic pressure / type of substratum	A	B	C
1	–	2.0 ± 1.73	1.3 ± 1.47
2	0.4 ± 1.47	2.8 ± 1.96	1.2 ± 2.02
3	–	1.5 ± 1.46	3.0 ± 2.52
4	–	0	–

In the sites of average anthropogenic load, the density was 2.8 ind m^{-2}. Here, the individuals preferred fine sand. Approximately the same numbers of them were found in the finest substratum (particle size 0.25–0.1 mm) – about 2 ind m^{-2}. The least lamprey density (about 0.4 ind m^{-2}) was when the load was high. In this case, the most appropriate substratum was found to be coarse sand (particle size 1–0.5 mm). Two-factor dispersion analysis (ANOVA) revealed a statistically significant dependence between individuals' density and anthropogenic load ($F = 4.37$, $p < 0.05$). Multiple class tests showed a different density under the different load: this difference was significant between the sites with a high load and a zero load (1.16) and also between the sites with the average and high load (1.72). When the anthropogenic load was average and when there was no load, the individuals' density did not differ.

The choice of habitat by water depth

Lamprey juveniles for the ripening choose places where the water depth is up to 30 cm. In the sampling sites, the depth was different and varied from 0 to 30 cm. The smallest individuals (length 5 cm) prefer shallower places (depth up to 5 cm) (Fig. 5).

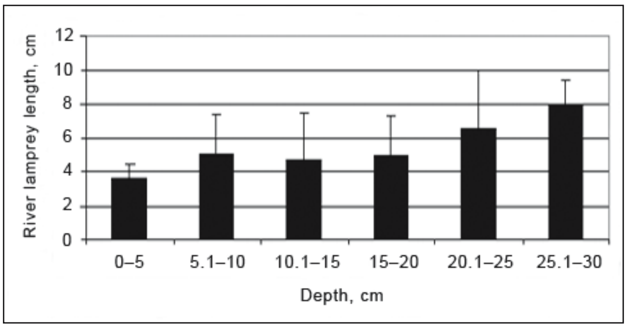

Fig. 5. River lamprey length ($\overline{x} + s$) in different water depth

With the growth of water depth, the mean value of individuals' length increased insignificantly. At the maximum depth this index was 7.8 cm, but the correlation analysis showed a weak significant correlation between individual's length and water depth ($r = 0.13$, $p < 0.05$). The majority of the individuals (40%) were found at a depth of 5–10 cm. With an increase of the depth, this quantity decreased to the moment when at a depth of 25–30 cm the found individuals made up ~1% of all individuals (Fig. 6). Individuals aged two years and more avoid shallow waters – there were no such individuals at a depth of 5 cm. Meanwhile, at a depth up to 5 cm, 4% of smaller individuals were found. At a depth from 5 to 10 cm the situation changes – here more than 20% of individuals from two length groups were found. When the depth increases, a tendency of fewer individuals was observed.

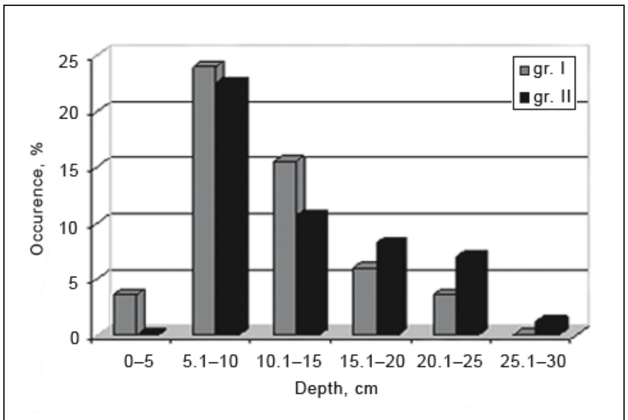

Fig. 6. Various age groups in different water depth (gr. I – individuals up to 5 cm long, gr. II – individuals longer than 5 cm)

The choice of habitat by macrophytes covering

The values of individuals' densities when the covering was of two different types were compared using the one-factor dispersion analysis. The F-test showed a statistically significant difference between the mean values in the sites covered by macrophytes and in those with no macrophytes covering ($F = 7.65$, $p < 0.05$) (Fig. 7). In the sites with macrophytes covering, the density of larvae was 2 ind m^{-2}. Meanwhile in the sites where there were no macrophytes, their density was 0.7 ind m^{-2}.

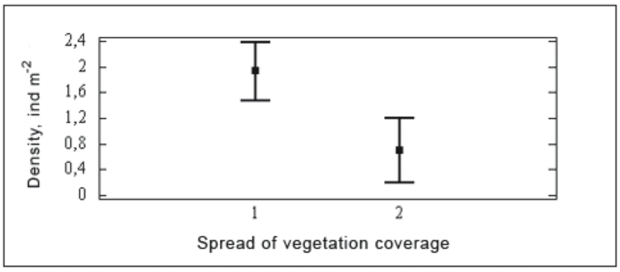

Fig. 7. River lamprey density ($\overline{x} + s$) in the sites with different vegetation density: 1 – sites with vegetation, 2 – sites without vegetation

DISCUSSION

When the research was accomplished and the average individuals' density in the sites had been calculated, it was noted that this index was the highest in the site in which the river makes a bend or where a little bay is, or where it is covered with macrophytes and is poorly resorted by people. In the site in which the bank was bent towards water, there were no individuals at all. Hardisty and Potter (1971) have noticed that larvae burrows are frequently in the cove or in the middle of the river bend, also beyond the obstacles, because in the places where the obstacles occur, the sediments and organic substances accumulate. According to these authors, in such places the sedimentation of organics is more rapid. Therefore, there is a common opinion that places for the burrowing, chosen by lampreys, are on the riversides, which are rather far away from the main current. Hardisty (1986) has ascertained that lampreys accumulate where the flow is slow and a rapid sedimentation of sand and deposits takes place (7 cm s^{-1}).

The density of larvae was studied by several scientists (Maitland, Campbell, 1992). They established that this index can vary strongly depending on place on separate years.

In the present work, the majority of the collected individuals (~60%) were found in the substratum of coarse sand formed by particles 1 – 0.5 mm in size. Here, the highest individuals' density in comparison with the other types of substratum was also defined. Although the statistical analysis did not show the significance of substratum type for the individuals' density, the results are partly confirmed by other researches related to choosing the place for burrowing. The scientists that studied the environmental conditions in sites of lamprey burrowing estimated that the optimal particle size in the sediments in which larvae are found is 0.18–1.38 mm (clay, deposits and sand fraction) (Maitland, Campbell, 1992).

The importance of substratum particle size (including sand, deposits and clay), slow current and water depth for the lamprey larvae was also marked by many other authors (Kainua, Valtonen, 1980; Maitland, 1980; Potter et al., 1986; Almeida, Quintella, 2000). All these authors also put an accent on the importance of organics in the sites of burrowing. Here, the current velocity is an important factor: it can predetermine the substratum type in a certain site (Hardisty, Potter, 1971).

Some authors (Hardisty, 1986; Sugiyama, Goto, 2002) state that the choice of habitats in lamprey larvae depends on the individual's length. Sugiyama and Goto (2002) compared the choice of the habitats of two length groups (group I – individuals up to 5 cm length, group II – individuals whose length was more than 5 cm), taking into account the environmental factors. The studies showed a statistically significant difference between these groups in respect of choosing the substratum type. The most appropriate size of substratum particles for the group of little individuals was 0–17 mm, meanwhile the individuals, whose length was more than 5 cm chose a substratum in which the particles 0–2 mm in size prevailed (Sugiyama, Goto, 2002).

However, Manion and McLain (1971) established that a high density of larvae is found in the sites where 90% of substratum consists of sand particles up to 0.5 mm. There were no lampreys in a finer substratum (clay).

In Finland, Kainua and Valtonen (1980) reported that the majority of lamprey larvae were found in a substratum in which the size of particles varied from 0.05 to 0.20 mm. The research revealed that the size of particles of the most suitable substratum may vary, and this is confirmed by the results of the present work.

Usually, sites appropriate for juvenile lampreys are found not in all parts of river systems (Maitland, Campbell, 1992). An absolute absence of such places is possible in some rivers. Changes of the stream regimen can affect strongly the density of lampreys, because these changes can modify the characteristics of the natural bottom or they can remove the area of slow, calm water and to wash away the sites abundant in silt.

Lamprey larvae avoid too shallow waters. But too deep water is also a negative factor for these individuals. Besides, the importance of environmental variables varied depending on size: no individuals was up to 5 cm were found deeper than 25 cm, meanwhile individual, more than 5 cm, were not found at a depth less than 5 cm. These data confirm the results reported by other researchers (Sugiyama, Goto, 2002). A statistically significant difference in choosing the water depth was defined: larger larvae chose deeper waters (0 to 38 cm), meanwhile the depth chosen by smaller individuals does not excesed 24 cm. It was also noted by Morman et al. (1980) that larger larvae are found in deeper waters.

Thus, disturbance of the natural water level, water diversion may be harmful to larvae (Maitland et al., 1988), destroying their habitats and abolishing rapids and canals in the places where the main sandy habitats are situated. It may make habitats unstable in respect of water level, disturbing the living places of larvae and keeping them above the water; in this way lampreys may be totally removed from the river (Maitland et al., 1988).

The results of our work also confirm that destroying the substratum is significant for lamprey larvae: there were no individuals found in the sites frequently visited by people.

Places covered with macrophytes are attractive for lamprey larvae. The largest density of these individuals (~2 ind m^{-2}) was established exactly in places overgrown with macrophytes. The relation between larva detection frequency and the bank's covering with macrophytes was studied by Potter et al. (1986). They reported that bank plant cover had a positive effect on larva density. Moreover, places covered only partly are suitable for the growth of diatom algae. This group of algae is one of the feeding components found in lamprey guts in most cases (Jurgaityte, 2005). Ryapolova (1972), while studying juvenile lampreys, agreed with the statement that 0+ larvae are mostly found in sand biotopes. However, she asserted that, although larvae can be found between algae and macrophytes, preference is given to the substratum without any vegetative cover. In this paper, it is impossible to say unambiguously how important macrophytes are for juvenile lampreys. But Kelly and King (2001) have found the largest larvae concentration found in the places where water plant islets were frequent.

The cover as a significant factor was noted when the juvenile's habitats in Sweden were analysed (Hardisty, Potter, 1971). This factor works in the habitats as the cover for juvenile. The loss of underwater flora can induce a more intensive washout of deposits from the upper reaches and an increase of bank instability, as well as erosion (Lamprey Baseline Report, 2005). Riverside flora can be important for the formation of slow waters on the coast where the bed of the deposits can be a suitable habitat for the juveniles (Lamprey Baseline Report, 2005).

Summarizing the results of the study, we may agree with Kirchhofen (1995) who has defined the indicators important for lamprey preservation, paying attention to the influence of habitat abolishment, removal of migration barriers and stopping sudden pollution in order to improve water quality, and also to the riversides' and flora's improvement and maintenance and to returning the engineering canals to a more natural state.

Thus, the most important factors that affect the density of lamprey juveniles are the loss of bank's diversity and destroying the river bottom. The loss of the flora as the source of food and cover and bank abolishment influence the life of larvae. However, further ecological researches are necessary to define the environmental factors that favour a high density of *Lampetra fluviatilis*.

CONCLUSIONS

1. During the period of the research, the individuals' density was highest in the places that were not affected by anthropogenic load. ANOVA analysis showed a statistically significant correlation between the individuals' density and anthropogenic load ($F = 4.37, p < 0.05$).

2. The density of juvenile lampreys is highest in the substratum of coarse sand (2 ind m^{-2}). No individuals were found in the substratum in which pebble prevailed.

3. Juvenile lampreys avoid shallow waters. No individuals more than 5 cm in length were found at a depth less than 5 cm.

4. Macrophyte covering is a positive factor for lamprey habitats: juvenile lamprey prefer places with macrophyte covering ($F = 7.65, p < 0.05$).

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UPINĖS NĖGĖS (*LAMPETRA FLUVIATILIS* L.) JAUNIKLIŲ PASISKIRSTYMAS SKIRTINGO TIPO BUVEINĖSE

Santrauka

Mėginiai rinkti 2006 m. birželį–rugsėjį Minijos vidurupyje. Darbe nagrinėjamas antropogeninės apkrovos poveikis upinės nėgės (*Lampetra fluviatilis* L.) jauniklių pasiskirstymui skirtingo tipo buveinėse. Mėginiai imti kas dvi savaites 100 m upės atkarpoje (20 stebėjimo taškų), atsižvelgiant į vietos mikroaplinką – substrato dalelių dydį, vandens gylį, makrofitų dangos paplitimą bei antropogeninę apkrovą. Nustatyta, kad didžiausiu individų tankiu pasižymėjo taškai, kurių neveikė antropogeninė apkrova, o substratą sudarė stambus smėlis. Čia individų tankis siekė 2 ind. m^{-2}. ANOVA analizė parodė statistinį reikšmingumą tarp individų tankio ir antropogeninės apkrovos ($F = 4{,}37$, $p < 0{,}05$). Daugelyje taškų biomasė kito apie 200 mg m^{-2}. Tyrimo rezultatai parodė, kad nėgių jaunikliai pirmenybę teikia stambaus smėlio substratui (substrato dalelių dydis 0,25–2,0 mm). Šiame substrate vidutinis individų tankis – 2 ind. m^{-2}. Substrate, kurio pagrindinę dalį sudarė gargždas, individų neaptikta. Statistinė analizė parodė silpną reikšmingą individo ilgio ir vandens gylio ryšį (koreliacijos koeficientas $r = 0{,}13$ ($p < 0{,}05$)). Atlikus tyrimą nustatyta, kad nėgių jaunikliai vengia seklių vandenų. Tuo tarpu makrofitų danga nėgių buveinėse veikė kaip teigiamas veiksnys ($F = 7{,}65$, $p < 0{,}05$). Skirtingo apaugimo taškuose nustatytas statistiškai reikšmingas vidutinių tankio reikšmių skirtumas: nėgių jaunikliai rinkosi vietas, kurios apaugusios makrofitais (tankis 2 ind. m^{-2}). Taškuose be makrofitų ši reikšmė buvo apie 0,7 ind. m^{-2}.

Raktažodžiai: antropogeninis poveikis, upinė nėgė, *Lampetra fluviatilis* L., vandens gylis, substrato tipas, augmenijos paplitimas