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A Trial of Natural Habitat Enclosure Traps as a Sampling Tool for Juvenile Crayfish

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Abstract.— Recruitment is a vital factor in the assessment, management and population dynamics of decapods. Since the juvenile stages of crayfish often prefer heterogeneous habitats, sampling with quantitative and reproducible methods have so far been challenging. We evaluate a new quantitative sampling method for juvenile crayfish; the enclosure trap. A field test was carried out during two consecutive years on a population of signal crayfish, *Pacifastacus leniusculus*, in littoral areas of Swedish Lake Erken. The densities of 0+ crayfish varied substantially with year, sampling date and substrate type. As expected, juvenile crayfish densities decreased over time in both study years, indicating a high mortality rate during their first year of life. Juveniles preferred gravel and stone over soft and sand substrates. Mean growth rate varied from 0.15 to 0.22 mm day⁻¹. We evaluate this method and present recommendations for how to design and optimize field studies using enclosure traps. We conclude that enclosure traps can be used to collect valuable data on density, growth and habitat preference of juvenile crayfish, thus providing useful information for studies on population dynamics and increasing the understanding of crayfish recruitment processes. [*Keywords.*— enclosure traps; growth; juvenile signal crayfish; *Pacifastacus leniusculus*; population density; recruitment; sampling method; substrate].

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INTRODUCTION

Recruitment is a vital factor in population dynamics and assessment and management of decapod populations (Caputi et al. 1995; Kirjavainen and Westman 1999; Wahle et al. 2004; Jones and Coulson 2006). When optimal harvesting regimes are designed, aimed at obtaining a high yield but avoiding over-exploitation, reliable assessments of population size and dynamics are required. The early life history of freshwater crayfish has been thoroughly studied (Mason 1978; Taugbøl and Skurdal 1992; Sáez-Royuela et al. 1995; Verhoef et al. 1998; Meade 2002; Savolainen et al. 2003; González et al. 2009; Harlioglu 2009; Kozák et al. 2009; Olsson and Nyström 2009), but usually under controlled conditions or with an experimental approach, with some exceptions (Odelström 1983; Söderbäck 1995; Westman et al. 2002). There is a noticeable lack of data on density and growth of crayfish juveniles in natural habitats, which is needed to assess recruitment success. Documentation on local density and substrate preference in combination with data on coverage of bottom substrate makes it possible to estimate the size of the entire juvenile population. Several studies have suggested the importance of substrate for the habitat preference in freshwater crayfish (Capelli and Magnuson 1983; Lodge and Hill 1994; Streissl and Hödl 2002). Knowledge of bottom substrate can be used in the management of crayfish, for example when identifying essential nursery habitats in threatened populations where recruitment is a bottleneck.

The most common method to sample adult crayfish in lentic systems is using baited traps. Juveniles however, do not often enter traditional baited traps and alternative sampling methods are therefore required. Several such techniques have been described, all with their own characteristics and limitations (Rabeni et al. 1997; Price and Welch 2009; Gladman et al. 2010; Parkyn et al. 2011). We believe that there is a need to further improve methods used to obtain more reproducible and quantitative data, particularly ones that accurately sample lentic habitats. In an effort to create such a quantitative sampling tool for juvenile crayfish, Fjälling (2011) constructed and described the "enclosure trap", a trap developed for the sampling of juvenile crayfish. However, the performance of this new sampling technique has not yet been evaluated under field conditions.

The aim of this study was to (a) assess whether enclosure traps can provide estimates of density, growth and substrate preference of juvenile crayfish in a natural lentic habitat, and (b) to evaluate the method and to provide users with guidelines on its use.

MATERIALS AND METHODS

Study Site

The majority of field work was carried out during July to October 2010 and 2011 in a small bay of Lake Erken (59.854001, 18.651237, WGS84; Figure 1), situated approximately 10 km

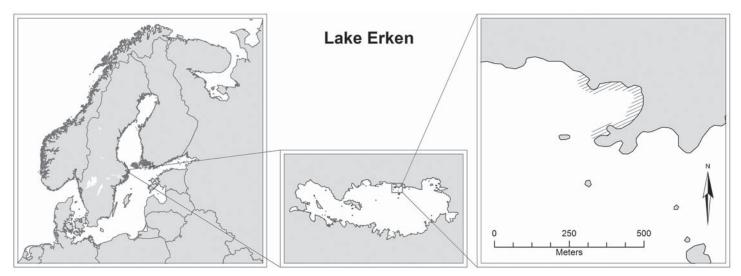


Figure 1. Map showing the location of the study site and Lake Erken. Striated area indicates where enclosure traps were placed.

north of the town of Norrtälje, Sweden. This 23.4 km² lake has a maximum depth of 21 m and a mean depth of 9 m. Forests and fields dominate the surrounding area. The lake is mesotrophic, and in 2008 – 2010 the epilimnial pH was in the range of 8.1 - 8.5 and the total phosphorous $22 - 34 \ \mu g \ L^{-1}$ (SLU Water Chemistry Database 2012). The signal crayfish, *Pacifastacus leniusculus* (Dana), was introduced in 1966 and can now be found throughout the lake. Commercial fishing produces an annual yield of 5 - 10 metric tonnes.

The Enclosure Trap

The traps consist of steel rings which support a cylindrical bag made of monofilament nylon (1 mm mesh) with a radius of 17 cm, covering a surface of 0.09 m². A quantitative measure of juvenile crayfish density was obtained by quickly lifting the traps from the

Table 1. Sampling data on juvenile *P. leniusculus* from a littoral site in Lake Erken. On four occasions one of the traps could not be located (No. of traps 9 or 19), the mean density for these dates was calculated using the remaining traps. The relative density is the quota between the abundance of juveniles of a given sample and the first sampling with crayfish in that specific year. S.E. is standard error.

		No.				5.1
Activity	Date of sampling	of traps	Catch of juveniles	Density (juv m ⁻²)	S.E.	Rel. Density
			Juvennes	(Juv III)	D. L.	Density
Set	06/15/2010	10	-	—	-	-
Lift/Reset	08/11/2010	9	45	53.5	9.9	1.00
Lift/Reset	09/06/2010	9	17	20.2	2.1	0.38
Final lift	10/12/2010	9	11	13.1	1.3	0.24
Set	05/26/2011	20	_	-	_	_
Lift/Reset	07/06/2011	20	0	0.00	-	0.00
Lift/Reset	07/26/2011	19	5	2.81	2.4	1.00
Lift/Reset	08/18/2011	20	5	2.67	1.5	0.95
Lift/Reset	09/28/2011	20	4	2.14	1.8	0.76
Final lift	10/27/2011	20	2	1.07	1.0	0.38

bottom. When the trap is retrieved, the fine meshed-sided bags along the edge of the trap unfolds as it rises towards the surface, thus enclosing all occupant crayfish (Figure 2). For a more detailed description of the enclosure trap see Fjälling (2011).

Field Work

The design of the study during the first year (2010) was focused on testing the performance of the enclosure traps and to estimate growth of juvenile crayfish and temporal variation in juvenile densities. We defined juvenile crayfish as individuals belonging to the young-of-the-year (0+) cohorts. In the second year (2011), the study design was expanded to also include habitat preferences. In the first year (2010) ten traps were placed in an area of roughly 30×30 m, on substrate deemed attractive to crayfish juveniles (i.e., small rocks and gravel), and covered with the natural bottom substrate occurring at the specific sites (Figure 2b). Traps were set and emptied three times before terminating the experiment. The interval between sampling occasions was selected to possibly optimize the effort necessary for obtaining density changes over time and growth rate. In the second year the study design allowed for a combined assessment of the importance of bottom substrate. Assuming that the amount of juveniles caught in a trap is correlated to the composition of its substrate surroundings, it should be possible to determine the importance of that substrate. The second year, twenty traps were set and emptied five times. The sampling interval was increased in 2011 based on experiences from 2010. In 2011, the traps were placed in the same bay as the previous year, but traps were distributed randomly to include all different substrate types found in the area. We standardized the substrate by strewing gravel of 16 - 28 mm diameter (shingle) over the bottom of the trap and gluing it together with aquarium silicone, creating a flexible plate with space between pebbles. The number of traps in each habitat type was roughly proportional to the surface coverage of that specific habitat type. Once a trap was set, natural substrate was scattered over its edges to create a smooth transition between the trap and its surroundings. Consequently the effort was higher than the previous year.

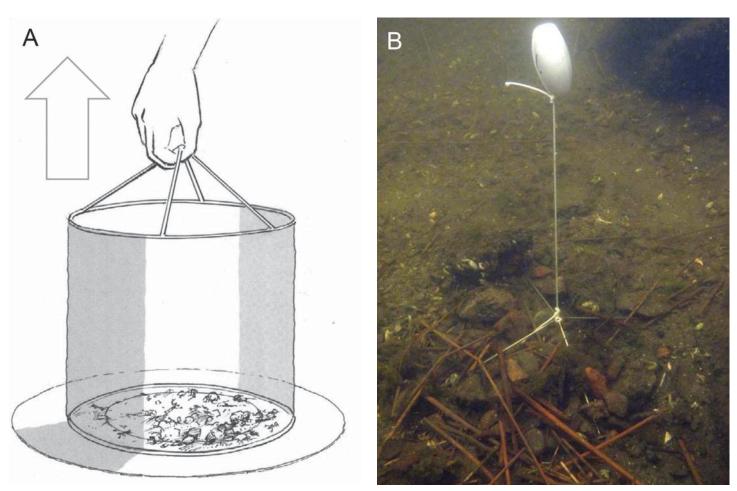


Figure 2. A.) The enclosed trap retrieved by lifting, crayfish enclosed (modified from Fjälling 2011). B.) Photo of enclosure trap set on the bottom with natural substrate.

All traps were placed manually on the bottom by a free diver at a depth of 0.5 - 2.8 m. Each trap was marked with a small white float on a 30 cm line to facilitate finding. The floats had to be submerged due to risk of tangling in sport fishing gear. Trap coordinates were recorded with a handheld GPS. The percentage cover of different bottom substrates within a radius of 3 m from each trap was estimated. Substrate was classified as bedrock, boulder (> 600 mm diameter), large stones (100 – 600 mm), small stones (60 – 100 mm), gravel (2 – 60 mm), sand (0.06 – 2 mm) and soft sediment (silt and clay).

When traps were emptied, all juvenile *P. leniusculus* (juvenile =0+ cohorts) found were counted and the total length was measured from the tip of the rostrum to the solid edge of the middle uropod. Cheliped loss was recorded to assess potential damage induced by the method. The time consumption of all stages in the setting and emptying of traps was estimated in retrospect.

Statistical Analysis

Temporal differences in density and mean total length of juveniles between sampling dates within each year were tested by One-way ANOVA. Water temperature data for the period of hatching and growth of juveniles from Lake Erken was provided by Uppsala University.

Habitat preference was tested by Chi-square tests. Test 1 was used to determine if juveniles generally preferred smaller

substrate to larger substrate, and had two groups; presence or absence of substrate > sand (including all types larger than sand). Test 2 had four groups; soft, sand, \geq gravel (including gravel and all types larger than gravel), and mixed (including all types with both substrate larger than gravel and smaller than gravel). This test would indicate if any of these single substrate types or a combination was preferred over the others.

The number of traps required to adequately estimate density and total length of juveniles for each sampling date was calculated with a bootstrap module in Microsoft Excel. The target level for precision in the estimates of the means was set to a confidence Interval (CI) of maximum 20% of the mean. For total length, the necessary number of juveniles was first calculated, and then the amount of traps needed to catch that number of juveniles was calculated.

RESULTS

Catch and Density

The density in 2010 varied from 52.5 - 13.1 juveniles m⁻². The following year (2011) the density varied from 2.8 - 1.1 m⁻². In 2011, relatively few crayfish juveniles were captured. For both years, there was a decrease in mean density from first to last catch (Table 1). The decrease in mean density over time was significant for all sampling dates in 2010 (P < 0.01), but not for 2011 (P =

Activity	Find suitable location	Find trap	Lift/Empty trap	Sort catch	Prepare trap	Fix trap on location	Setting trap substrate	Substrate survey	Time per trap
Set	4	-	_	_	1	2	4	2	13
Lift/Reset	_	3	1	3	1	2	4	_	14
Final Lift	_	4	1	2	_	_	_	_	7

Table 2. Estimated time consumption for different parts of the sampling process, in minutes.

0.76) (One-way ANOVA). Over the sample interval it decreased with 76% in 2010 and 62% in 2011.

In addition to juvenile crayfish, the traps also caught two adult crayfish, dragonfly nymphs and other insect larvae, zebra mussels, *Dreissena polymorpha* (Pallas), snails and three ruffe 0+, *Gymnocephalus cernuus* (Linnaeus), and one European bullhead, *Cottus gobio* (Linnaeus).

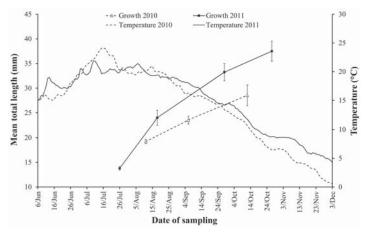


Figure 3. Mean total length of juvenile *P. leniusculus* and water temperature in Lake Erken. Error bars display standard error. The temperature data are mean values for temperatures recorded at 1.5 and 3 m depth.

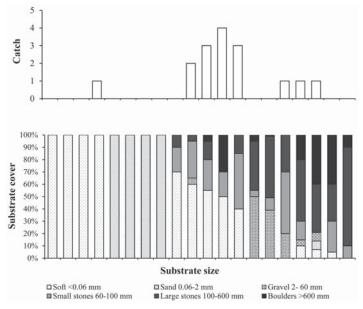


Figure 4. Habitat preference of juvenile crayfish illustrated as total number of juvenile *P. leniusculus* caught per trap (top) and the percentage cover of substrate types within a 3 m radius of each trap (bottom) during 2011 sampling.

Growth

The mean total length of juveniles increased throughout the season for both years (Figure 3), and the One-way ANOVA showed that there were significant differences between all consecutive sampling dates in both 2010 (P < 0.01) and 2011 (P < 0.01). The growth rate in 2010 between first and last sampling (62 days) was 0.15 mm day⁻¹ (initial length was 19.2 and the length at the end of the sampling period was 28.6 mm). In 2011, juveniles were estimated to grow 0.26 mm day⁻¹ (initial length was 13.8 mm and the length at the end of the sampling period was 37.5 mm).

Substrate Preference

Juvenile density was highest in traps that were placed on a substrate larger than sand (mean density = 13.8 crayfish juveniles m⁻¹), whereas the traps on only soft substrate had a lower density (mean density = 0.9 crayfish juveniles m⁻¹), and traps on only sand caught no juvenile crayfish (Figure 4). Catches were significantly higher where substrate material larger than sand was present (P = 0.04, Chi-square test 1), but there was no significant preference for where the substrate was mixed as opposed to composed of a single type or only consisted of material \geq gravel (P = 0.06, Chi-square test 2).

Methodology and Guidelines

The number of traps necessary for estimating total length with CI lower than 20% of the mean ranged between 1 and 40 for our study (Figure 5). Estimates of abundance required more traps, between 30 and 960. Our results indicate that a low abundance of juveniles requires a high number of traps.

The time to find, empty, and reset the enclosure traps varied greatly with depth, trap substrate and timing (Table 2). As a result, the total time of each sampling round was dependent on the actions taken; setting at beginning of experiment, lifting and setting during the season, or just lifting at the end of the experiment. The average time per trap was longest when traps were both emptied and reset in the middle of season, and shortest when traps were only emptied at the last sampling.

No mortality or escape was observed during sampling and cheliped injuries were only found in 2 individuals out of all juveniles captured.

DISCUSSION

Catch, Density and Growth

Results from previous studies have indicated that recruitment success of *P. leniusculus* can vary greatly between years (Abrahamsson 1971; Kirjavainen and Westman 1999) with significant effects on population size. There was an apparent

Study aim	No. of sampling occasions	No. of traps	Sampling timing	Importance of timing	Substrate type
Growth	High	Low	All season	Low	Artificial
Density/Mortality	Medium-High	High	All season	Low	Natural
Index for relative recruitment success	Low	Medium	Late	High	Artificial
Hatching success (Egg to juvenile ratio)	Low	Medium	Early	High	Artificial
Habitat preference	Low	High	Early	Low	Natural
Hatching occurrence	Low	Low	Early	High	Artificial

 Table 3. Recommendations for investigation design given various study objectives when sampling juvenile crayfish.

difference between the two years in our study, possibly demonstrating the annual variation in recruitment success of this species.

The decrease in density of crayfish juveniles found during the first months after hatching was expected as a consequence of predation from fish, invertebrate larvae, adult crayfish and mortality during molting events. Brewis and Bowler (1983) estimated crayfish juvenile mortality at 15% each week during the molting period. Our results indicate a weekly mortality of 7 - 17% for 2010 and 2 - 12%for 2011. Our density estimates are comparable with several other studies. Odelström (1983) who used a diver operated dredge-sieve to sample juvenile crayfish in Lake Erken found the density to be in the range of 4.5 to 2.4 individuals m⁻² during the summer season, which is consistent with our data from 2011. However, it should be noted that the population was still expanding in the early 1980's. Other studies of species of crayfish in streams and lakes using a variety of methods have reported densities ranging from 0.35 - 27.9juveniles m⁻² (Rabeni et al. 1997; DiStefano et al. 2003; Larson et al. 2008, Parkyn et al. 2011).

Both adult crayfish and dragonfly nymphs were caught in the traps, they are both considered predators on juvenile crayfish (Jonsson 1992, *A. astacus*; Herberholz et al. 2004, *P. clarkii*). Thus, it is interesting to note that the traps can also be used to assess food availability for older crayfish, as well as the presence of some important juvenile predators.

The growth rate of 0.15 - 0.22 mm day⁻¹ found in this study is comparable to that of other studies in the wild (Söderbäck 1995; 0.16 mm day⁻¹; Kirjavainen and Westman 1999; 0.15 mm day⁻¹).

Substrate Preference

Our results indicate that substrate size is important for habitat preference in juvenile crayfish. However, it must be emphasized that the number of crayfish juveniles in our samples were relatively low. Thus, the results must be interpreted with caution. Nevertheless, our results are in accord with several earlier studies. Blake and Hart (1993) found that *P. leniusculus* juveniles were almost completely absent on soft substratum, and that material with a size of 12-29 mm was preferred to that of 8-16 mm. A study on the crayfish *Orconectes rusticus* (Girard) conducted by Stein and Magnuson (1976) found that juveniles prefer pebbles and gravel to sand in the presence of a fish predator. Kershner and Lodge (1995) illustrated that the density of *O. rusticus* juveniles was higher on

cobble substrate than on a substrate consisting primarily of sand or macrophytes growing on sand or soft sediment.

Methodology and Guidelines

Our assessment shows that enclosure traps can be a valuable method for acquiring quantitative measures of juvenile crayfish density and growth. Data can be used for several purposes; measuring variation in recruitment, identifying essential nursery habitats, identifying important processes in crayfish population dynamics and several other significant areas of research and management. However, the presence, density and activity of juvenile crayfish varies considerably depending on substrate, depth and predation intensity (Hamrin 1987, Blake and Hart 1993, Kershner and Lodge 1995, Englund and Krupa 2000) and this will influence the optimal sampling design. Based on our experience we stress the importance of a careful design of sampling intensity and timing, the choice of substrate and the spatial design of sampling sites like habitat coverage and trap spacing (Table 3).

Number of traps

Our results suggested that in cases where the objective is to follow juvenile crayfish growth during the season, or to register if hatching has occurred, fewer traps are necessary, but in order to make a good estimate of juvenile density or mortality, the required number of traps would need to be greater (Table 3). Since time is a limiting factor, the use of many traps would be facilitated by an improved methodology. The original idea behind the enclosure traps was to use a design that allowed for deployment from a boat with traps on a line, thus eliminating the need for diving and allowing for a quicker and more frequent sampling.

Number of sampling rounds

For continuous data on growth and density throughout the growth season, given that precise estimates can be made in each round, we suggest a minimum of five sampling occasions. If data are to be used only for comparing relative recruitment success between years, for habitat preference or for hatching occurrence or success, one sampling occasion may suffice, depending on the number of traps used, juvenile density and timing.

Sampling timing

The importance of timing is greater with fewer sampling rounds. If there is only one round, a favorable time should be at

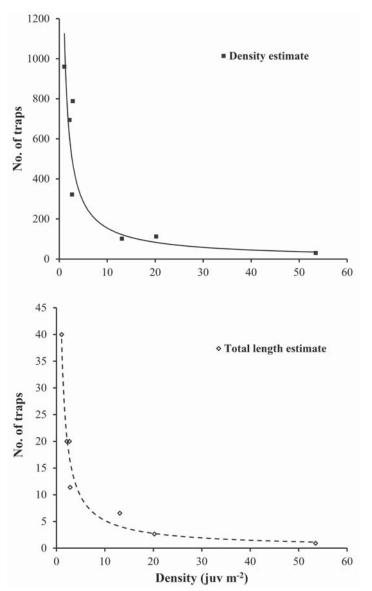


Figure 5. Number of traps necessary to estimate juvenile population density (top) and mean juvenile total length (bottom) with CI < 20 % of mean values for different densities of juveniles. Trend line equations: Density estimate $y = 1194.6x^{-0.9}$, $R^2 = 0.94$; Total length estimate $y = 41.5x^{-0.9}$, $R^2 = 0.96$.

the end of the growing season, since the recruitment of a certain year is often not possible to estimate until the end of season when a cohort has experienced the substantial mortality that occurs during the first summer. Also, DiStefano et al. (2003) found that estimates of crayfish density could be less variable in autumn than spring. One drawback of a late sampling may be that there can be greater bottom coverage of macrophyte algae, making it difficult to set and find the traps. The water temperature may also be useful when deciding sampling timing since the activity of signal crayfish is reduced at low temperatures (Bubb et al. 2002, 2004; Usio et al. 2006).

Regardless of when the first sampling takes place, it is important that the traps are not retrieved before they are colonized by the food items of crayfish and algal growth, to match the surrounding habitat. Through visual inspections, we concluded that traps should be deployed at least two weeks before that time.

Substrate type

Our results demonstrate the importance of substrate for juvenile crayfish habitat preference. In our study, we tried both covering the enclosure traps with natural substrate (2010) and with a standardized substrate that was covered/integrated with natural substrates on site (2011). A third option is to only use a single standardized artificial substrate. Using a natural substrate probably renders a measure that is as close to natural densities as possible. In many habitats, however, covering the enclosure trap with natural substrate without affecting its function is hard and often time consuming. When the natural substrate consisted of loose material from the bottom, visibility became very poor, soft clay and debris was disturbed when substrate was moved and in areas with boulders and large stones it was hard to find natural substrates that could be used in traps. Using artificial substrates could be a way to reduce time per trap and consequently make it possible to increase the number of traps. One potential bias with artificial substrates, however, is that they might attract juveniles when deployed in less preferable habitats, thus resulting in higher densities than natural substrates. However, no such patterns were found in our results.

If the intention is to catch many juveniles, for example, to record growth during the season, to estimate relative recruitment success, hatching success or hatching occurrence, the traps should be set with an artificial substrate and concentrated on gravel and/ or rocks. Areas with soft sediment or sand should be avoided. For collecting data on density or substrate preference, traps should be set with a natural substrate. In the case of substrate preference and/or spatial distribution, a stratified approach is advocated, calculating the number of traps necessary for each substrate type. This requires an estimate of the percentage of the bottom covered by different substrates.

Time consumption

The major limiting factor during sampling was visibility and the traps were hard to locate even with GPS coordinates known and traps marked with (submerged) white floats. This was especially the case late in the season when substantial algal growth on traps and floats made them difficult to distinguish. There were also severe algae blooms at different times which blocked out sunlight and increased the turbidity, making visibility very low. Moreover, the precision of a standard GPS was too low to pinpoint the exact location. The amount of time for sampling would decrease substantially if traps were only to be deployed and emptied once, at the end of the season.

Crayfish welfare

The low frequency of injured juveniles indicates that the traps do not harm crayfish during emptying and handling. Data on cheliped loss from juvenile crayfish sampling is scarce, but it has been commonly observed when using electrofishing (Westman et al. 1979; Price and Welch 2009; Gladman et al. 2010). Alonso (2001) found cheliped loss on 26.8% of all caught crayfish when using

electro-fishing, with smaller crayfish suffering more injuries. The use of enclosure traps is thus a less destructive method compared to electro-fishing. There are several reasons for minimizing cheliped loss as a result of sampling. If part of the objective of catching juveniles is to determine the frequency of cheliped loss in the population, a destructive methodology will create errors. It has also been suggested that cheliped loss can increase mortality and limit growth in juveniles (Mason 1978; Hirvonen 1992; Taugbøl and Skurdal 1992). This would have a negative effect if juveniles are released after sampling or if they are intended for further study.

Concluding remarks

Useful data on density, growth and substrate preference of juvenile crayfish was collected using the enclosure trap, and it is our impression that it is a useful tool in studies of recruitment and early development of freshwater crayfish. However, additional comparisons with other methods as well as extended tests in other lakes and/or streams needs to be done before the enclosure trap can be accepted as a standardized method for juvenile crayfish monitoring surveys.

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