FISHERIES RESEARCH 151: pp. 12-19. (2014) -- DOI: 10.1016/j.fishres.2013.12.004 1 2 Application of an electrified benthic frame trawl for sampling fish in a very large 3 European river (the Danube River) – Is offshore monitoring necessary? 4 Z. Szalóky<sup>a</sup>, Á.I. György<sup>a</sup>, B. Tóth<sup>b</sup>, A. Sevcsik<sup>b</sup>, A. Specziár<sup>c</sup>, B. Csányi<sup>a</sup>, J. Szekeres<sup>a</sup>, T. 5 Erős<sup>c,\*</sup> 6 7 <sup>a</sup> Danube Research Institute, MTA Centre for Ecological Research, Jávorka Sándor u. 14., H-8 2131 Göd, Hungary 9 <sup>b</sup> Danube-Ipoly National Park Directorate, Költő u. 21, H-1121, Budapest, Hungary 10 <sup>c</sup> Balaton Limnological Institute, MTA Centre for Ecological Research, Klebelsberg Kuno u. 11 3., H-8237 Tihany, Hungary 12 13 \*Corresponding author: 14 Tibor ERŐS 15 Balaton Limnological Institute, MTA Centre for Ecological Research 16 Klebelsberg K. u. 3., H-8237 Tihany, Hungary 17 Tel.: +36 87 448 244 18 Fax.: +36 87 448 006 19 E-mail address: eros.tibor@okologia.mta.hu 20 21

#### 22 Abstract

23

The organization of fish assemblages in offshore, deep channel habitats is poorly known in 24 very large rivers compared with shoreline, littoral areas. We report on the parameters and 25 testing of an electrified benthic frame trawl (EBFT), developed for monitoring the distribution 26 and abundance of benthic fishes in the Danube River, Hungary. We also compare the results 27 of the benthic main channel survey with a shoreline electrofishing (SE) data set. Altogether 28 33 species were collected offshore during the 175 trawling paths (500 m long each). Both 29 sample based and individual based rarefaction showed that night time SE detected 30 significantly more species with increasing sampling effort than day time trawling of offshore 31 areas. However, offshore surveys detected sterlet Acipenser ruthenus, which could not be 32 detected by SE, even using extreme high sampling effort. Offshore trawling also proved the 33 34 common occurrence and high abundance of the strictly protected endemic Danube streber Zingel streber in the river, which proved to be extremely rare in SE catches. The EBFT 35 caught larger/older individuals of many species than SE, and indicated diverse size/age 36 structure for many species offshore. Our survey revealed that offshore areas are intensively 37 used by a variety of species, which occur relatively evenly, but with variable abundance in the 38 Danube River. We suggest that even a relatively small (i.e. 2 m wide 1 m high) EBFT can be 39 a very useful device for monitoring offshore fish assemblages in very large rivers and provide 40 important data for bioassessment and conservation purposes. 41

42

43 Keywords: trawling, large rivers, shoreline electrofishing, sampling, fish assemblages

#### 45 **1. Introduction**

46

Sampling the biota in the main channel of large rivers presents a continuing challenge for 47 freshwater ecologists. While our knowledge of the organization of shoreline fish assemblages 48 and their representative sampling are increasing (e.g. Jurajda et al., 2001; Erős et al., 2008), 49 information about the composition and spatial and temporal distribution of fishes in deep 50 channel habitats is still relatively sporadic (Dettmers et al., 2001). Inferences about how main 51 channel habitats contribute to the bioassessment of large rivers compared with shoreline 52 monitoring data should also be more precisely developed (de Leeuw et al., 2007; Flotemersch 53 et al., 2011). The highway analogy, a postulate of the flood pulse concept (Junk et al., 1989), 54 which states that the main channel of large alluvial rivers is used by fishes mainly as a route 55 for gaining access to floodplain habitats, has been proved to be oversimplified, because main 56 57 channels were shown to maintain diverse fish assemblages with several species spending most of their life-time offshore (Galat and Zweimüller, 2001; Wolter and Bischoff, 2001; Stewart 58 et al., 2002). However, detailed quantitative studies are restricted to only a very few large 59 rivers even in the relatively well studied temperate large river systems of Europe and North-60 America (see e.g. Wolter and Freyhof, 2004; Gutreuter et al., 2009; Ridenour et al., 2009). It 61 would be thus important to develop deep channel fish monitoring methods in a variety of 62 biogeographical and ecoregions for providing data for both basic research and the 63 conservation management of riverine fish species. 64

With its 2872 km length, the Danube River is the second longest river in Europe.
Although the river is the cradle of Europe's most diverse fish fauna (Reyjol et al., 2007), the
large scale organization of its fish assemblages is relatively poorly known, compared with
other central and especially western European large rivers. The monitoring of its fish
assemblages is mainly based on shoreline electrofishing methods (Hirzinger et al., 2003; Erős,

2007). For example, the second Joint Danube Survey (JDS2), organized by the International 70 Comission for the Protection of the Danube River (ICPDR) in 2007, provided the first data 71 about the longitudinal distribution of fish assemblages along the river with a standardized 72 methodology (http://www.icpdr.org/jds/). However, the electrofishing based surveys were 73 restricted to inshore areas, and consequently did not provide information on main channel fish 74 assemblages. Further, although national monitoring programs intend to address the deepwater 75 main channel species in some countries (e.g. long line sampling in Austria) a really effective 76 77 and routinely used methodology for sampling main channel species, to our knowledge, has not been developed yet. Such an easily applicable monitoring methodology would be essential 78 for example to provide complementary information about the occurrence and abundance of 79 conservationally important species. 80

Many small, benthic species are important for conservation purpose (Labonne et al., 81 82 2003; Ridenour et al., 2009), yet they are especially difficult to collect with conventional fishing gears (e.g. trammel nets, gillnets) used to sample deep habitats in slow flowing large 83 rivers (Murphy and Willis, 1996; Herzog et al., 2005; Freedman et al., 2009). Additionally, 84 entangling nets and hook and line (i.e. long line) sampling can injure these small fish 85 seriously. Note that we by no means refer to boatable (raftable), but rather shallow (i.e. less 86 than 2 m deep) rivers which are usually sampled with electrofishing from boats or with boom 87 mounted electrofishing ships (see e.g. Hughes et al., 2002). Monitoring the populations of 88 benthic species with the more intensively used hydroacustic methods is still problematic, 89 since their exact identification still present difficulties for researchers, especially in case of 90 91 species from the same genus. Naturally, the combination of hydroacustic surveys with a suitable fishing gear can be fruitful, because the latter can help to collect fish for 92 identification. For this purpose, trawling is the most preferred fish sampling method of the 93 main channel trough (Dettmers et al., 2001; Wolter and Bischoff, 2001; De Leeuw, 2007; 94

Doyle et al., 2008). Recently, different trawling gears has been developed and their efficiency
tested for the more effective sampling of benthic species in very large rivers (Herzog et al.,
2005; Freedman et al., 2009; Ridenour et al., 2009; 2011).

In this study, we report on the application of an electrified benthic frame trawl (EBFT), 98 developed for monitoring the distribution and abundance of benthic fishes in the Danube 99 River. Specifically, we show the parameters of the EBFT device and provide the first detailed 100 data on sampling effort species richness relationships, abundance, and size structure of the 101 102 most common benthic species in the offshore, deep channel habitats of this very large river. We also compare the results of our benthic main channel survey with an extensive shoreline 103 electrofishing (SE) data set. Based on these comparisons we evaluate the applicability of main 104 channel benthic surveys for the study of fish assemblages in a very large river for 105 bioassessment and conservation purposes. 106

107

## 108 2. Material and methods

109

#### 110 *2.1. Study area*

111

The Danube has a drainage area of approximately 796,250 km<sup>2</sup>. River regulation, namely 112 the construction of hydroelectric schemes, especially in the Upper Danube (i.e. in Germany 113 and Austria), and channelization have profoundly modified the physical structure of the 114 Danube throughout its course. The Hungary section, referred to as the 'Middle Danube', runs 115 for 417 km and has a mean annual discharge of 2000 m<sup>3</sup> s<sup>-1</sup>. The main channel has a 116 substratum dominated by gravel and sand, a mean depth of 4 m and a mean velocity of 0.6 m 117  $s^{-1}$ . The banks are relatively natural (except the section lying within Budapest), interrupted 118 with embanked rip-rap shorelines of  $\sim 100-1000$  m long sections. 119

# 121 2.2. Data collection

122

To construct the sampling device (EBFT) to be effective in catching small sized benthic 123 species we combined the design of conventional trawl nets and framed sledge nets, the latter 124 used to sample fish fry in deep habitats (Fig. 1). This consisted of a 3.4 cm diameter stainless 125 126 steel frame (2 m long  $\times$  1 m high) to which a drift net was attached. The drift net was 5 m 127 long and consisted of a 5 mm-stretch inner mesh bag and an 8 mm-stretch outer mesh bag. A buoy was attached to the codend with a rope to indicate the position of the net while fishing. 128 129 We used weighted metal wheels to help keeping the device close to the bottom and also keeping the frame 6 cm above the bottom to prevent the filling of the net with the substrate 130 material. We electrified the frame with a 40 m long electrode cable which was connected to a 131 Hans-Grassl EL65 IIGI electrofishing device operated with a VANGUARD HP21 14.9 KW 132 generator. A 6 m long copper cathode cable was hanged freely and pulled approx. 2 m before 133 the electrified frame (Fig. 1). Preliminary catching experiments, (specifically with and 134 without electricity, different positions of cathode cables, different net mesh sizes and frame 135 sizes) showed that this construction yielded the most acceptable compromise between 136 catching rates and sampling from boat with a four person crew (see Szalóky et al., 2011). In 137 this crew, 2 people handled the framed net, one handled the electrofishing device and one 138 operated the boat. Fishing (hereafter trawling) was conducted with a 6.3 m long boat powered 139 by a 50 horsepower outboard Mercury four stroke engine. 140

When starting trawling, the EBFT operators lowered the frame to the bottom while the boat was slowly moving downstream with the flow. Trawling route was started to be measured by a GPS only after EBFT reached the bottom, which could be easily felt while holding the central rope (Fig. 1) and right after electroshocking started. The direct current

(approx. 350 V, 33 A) was given for 5-8 s with 3-5 s breaks between the operations to
minimize fright bias and injury of fish. The applied trawling speed was slightly higher than
the current velocity of the river (approx. 60 cm s<sup>-1</sup>). Each haul had a length of 500 m.
Trawling was carried out during daytime. This study contains data of the samples collected in
April 2011 - September 2011 period.

We collected altogether 175 samples, 500 m long each, along a  $\sim$  350 km stretch of the 150 417 km Hungarian Danube River section (Fig 2). We used a stratified design to select 151 segments in order to get a representative coverage of the whole main channel area (excluding 152 the section where the capital, Budapest can be found). In each of these segments several, but a 153 minimum of 3 transects were selected randomly, perpendicular to the bank. Along each 154 transect, across the width of the main channel, we generally distributed 5-6 trawl paths, 155 excluding the littoral, less than 2 m deep shoreline zone. These paths were approximately 156 157 equispaced and centred over the approximate place of the main channel centreline (Gutreuter et al., 2009). Note that the number of trawl paths along the transects varied depending on the 158 159 river width. Sometimes the trawl was stopped due to interruption by large rocks or logs. The trawl paths were then grouped into five classes depending on their offshore position, starting 160 conventionally from the right side of the river. As such, the offshore classes 1 and 2 situated 161 on the right side of the centreline, and had a mean ( $\pm$ SD) distance of 74 ( $\pm$ 35) and 123 ( $\pm$ 52) 162 m from the right side of the river, respectively. Class 3 situated approximately at the 163 centreline with a mean distance of 255 ( $\pm$ 65) and 229 ( $\pm$ 66) m from the right and left side of 164 the river, respectively. Classes 4 and 5 situated on the left from the centreline and had a mean 165 distance of 112 ( $\pm 27$ ) and 57 ( $\pm 19$ ) m from the left side of the river, respectively. The mean 166 depths ( $\pm$ SD) for Classes 1, 2, 3, 4, and 5 were 3.7 ( $\pm$ 2.4), 4.6 ( $\pm$ 1.9), 4.4 ( $\pm$ 1.3), 4.4 ( $\pm$ 1.6) 167 168 and 3.9 ( $\pm$ 2.1) m, respectively. Fish were identified and measured to the nearest mm standard lengths (L) and then released back to the river. 169

Besides evaluating main channel fish data, we compared EBFT data with SE monitoring 170 data collected between 2005 and 2011Although the SE and EBFT data were only partly 171 overlapping they are comparable at such a large spatial scale since yearly or seasonal changes 172 were relatively small compared with the effect of the mesohabitat type and spatial position of 173 the sampling sites in the SE data (Erős et al., 2008; Erős et al unpublished data). Briefly, the 174 SE data set contains electrofishing data of altogether 207, 500 m long stretches in the frame of 175 which approximately 48,000 individuals were caught (Table 1). The stretches were fished 176 177 from a boat using either a 7.5 KW or a 13 KW generator powered machine (Hans-Grassl Gmbh EL64 II GI, DC, 7.5 KW, and EL65 II GI, DC, 13KW). Note that preliminary 178 evaluations did not show significant differences in species richness and fish relative 179 abundance distributions between the two machines (Erős et al., unpublished data). Fish were 180 caught with one hand held anode (2.5 m long pole with a ring anode of 40 cm diameter and a 181 182 net mesh size of 6 mm) while slowly moving downstream with the boat as per Wolter and Freyhof (2004). The cathode, a 5 m long copper cable, was floated at the rear of the boat. We 183 used night time sampling because former surveys of the Danube (Erős et al., 2008) and other 184 systems (Wolter and Freyhof, 2004) justified that it is more efficient than daytime sampling 185 of shoreline fish assemblages. 186

187

### 188 2.3. Statistical analysis

189

We used both sample based and individual based rarefaction analyses to examine
changes in the number of species with increasing sampling effort (Gotelli and Colwell, 2001;
Erős et al., 2008; Flotemersch et al., 2011). The analyses were conducted to compare the
differences in the number of species between 1) the SE and the EBFT collections, and 2) the
different offshore distance classes of the EBFT collections (1-5 classes).

We used the nonparametric Kruskall-Wallis ANOVA to test whether total median CPUE 195 data (i.e. median of the total number of individuals captured per a 500 m long sampling 196 transect) differ among the five offshore classes. To test for significant differences in 197 assemblage structure among the offshore classes, we used ADONIS in Vegan package of R 198 (R Development Core Team, 2013), which is the more robust version of nonparametric 199 permutational analysis of variance (PerMANOVA) method developed by Anderson (2001). 200 The analysis was run using 999 permutations of the raw catch per unit effort (CPUE) data of 201 202 fishes (i.e. number of individuals captured per a 500 m long sampling transect) and the Bray-Curtis measure. 203

Between gear (i.e. SE vs. EBFT) differences in the  $log_{10}(x+1)$  transformed CPUE data of the benthic species were tested with two-sample t-test. Standard length distributions of fish in cumulated samples were compared between the SE and the EBFT with the nonparametric Kolmogorov-Smirnov test, and median fish sizes were tested for significant differences with the Mann-Whitney U-test.

209 Note, that the effectiveness and efficiency of the two sampling gears (i.e. boat electrofishing vs trawling) cannot be directly compared, since the two gears sampled two 210 different habitats during different time of the day. In fact the two gears cannot be used in the 211 same habitat, because it is clear that boat electrofishing is ineffective in deep offshore areas, 212 whereas the use of the EBFT is very laborious and can be even dangerous in shallow 213 shoreline areas, especially during the night. Therefore, it is important to emphasize that the 214 purpose of "between gear comparisons" was to evaluate the complementary information 215 EBFT can provide to SE about the fish assemblages of a very large river. 216

217

218

219 **3. Results** 

221	We collected 33 species and 8112 specimens with the EBFT during the 175 trawling
222	paths. In 4 trawlings we did not catch any fish due to sampling error (i.e. the gear was
223	snagged on logs). These tows were excluded from further analyses. The mean number of
224	species per 500 m long sampling reach was $5.1 \pm 2.1$ (mean $\pm$ SD), which is significantly
225	lower (P<0.05) than the number of species estimated for shoreline electrofishing (SE), where
226	$14.7 \pm 2.1$ species was caught for the same sample unit length (Fig. 3a). Both sample based
227	and individual based rarefaction showed higher increase in the estimated number of species
228	with increasing sampling effort in case of SE compared with offshore sampling with the
229	EBFT (Fig. 3a,b). However, offshore sampling detected sterlet Acipenser ruthenus L., which
230	could not be detected by SE, even using extreme high sampling effort.
231	Sample based rarefaction curves indicated relatively large differences between the EBFT

based samples differing in their offshore position (Fig 4a). Samples which situated in the
centreline of the river (i.e. class 3 samples) tended to have the lowest number of species at
any sample size. However, the differences between the different offshore sample classes were
not really supported by the individual based rarefaction (Fig. 4b). The number of species
varied only between 17 and 19 among the five offshore classes at a standardized number of
individuals collected (i.e. 678 individuals, the total number of individuals collected in
offshore class 4).

Both relative abundance and frequency of occurrence data of fishes differed between the EBFT and the SE samples (Table 1). As expected, benthic species dominated in the catches of the EBFT, while surface oriented, water column and benthic species were all important assemblage constituting species in the SE catches. Of these, the silver bream *Blicca bjoerkna* (L.), the bleak *Alburnus alburnus* (L.), the white-finned gudgeon *Romanogobio albipinnatus* (Lukasch), the schraetzer *Gymnocephalus schraetser* (L.), the bighead goby *Ponticola* 

kessleri (Günther) and the round goby Neogobius melanostomus (Pallas) were the most 245 abundant species using both monitoring methods. However, the EBFT indicated the 246 commonness of some benthic species in the river, which information would have remained 247 hidden using only shoreline surveys (SE). The most striking difference was the common 248 occurrence and relatively high abundance of Zingel species, and especially Danube streber 249 Zingel streber (Siebold) offshore. Mean CPUE of the benthic species showed that Danube 250 streber was the only species which had significantly higher abundance in the main channel 251 252 than in the shoreline catches (Fig. 5). Total CPUE data of EBFT catches varied between 2 and 1761 ind 500 m<sup>-1</sup>, and showed weakly significant differences between the five offshore 253 254 classes (Kruskall-Wallis ANOVA, H=10.07, p=0.039). Similarly, the ADONIS analysis also indicated significant differences in assemblage structure between the offshore classes 255 (pseudo-F=1.62, p=0.015). However, the variance explained was extremely low ( $R^2=0.038$ ), 256 257 which showed that distance from shore cannot explain differences in fish assemblage structure (i.e. raw CPUE data) and that probably the high sample size influenced the result of 258 259 the significance test. This latter result was further confirmed by two-dimensional solution of non-metric multidimensional scaling analysis (NMDS), which yielded almost completely 260 overlapping assemblages among the five offshore classes in the ordination plane (results are 261 not shown for brevity). 262

Median values of standard length data of the abundant species (i.e. >1% relative abundance in the total catch of any method) showed significant differences for most fishes between the EBFT and SE samples (Table 2, Mann-Whitney *U*-tests). In general larger specimens of many cyprinids (e.g. barbel *Barbus barbus* (L.), common bream *Abramis brama* (L.), common nase *Chondrostoma nasus* (L.) and vimba *Vimba vimba* (L.)) were relatively more abundant in the EBFT than in the SE samples (Fig. 6., see Table 2 for Kolmogorov-Smirnov tests).

### 271 **4. Discussion**

272

273 The EBFT proved to be very effective in detecting both benthic and water column species in offshore areas of the Danube River in habitats which are unavailable for 274 conventional boat electrofishing methods. All known benthic species from the last ten years 275 of fish faunistic surveys (Harka and Sallai, 2004; Erős et al., 2008) were collected with the 276 277 one year study using EBFT, with the exception of some rare benthic species, which prefer shallow, slow flowing habitats (e.g. the spined loach Cobitis elongatoides Bacescu and 278 279 Mayer) or species which appear extremely rarely in the Hungarian section of the river (e.g. Danube sturgeon Acipenser gueldenstaedtii Brandt and Ratzeburg). Although sample based 280 rarefaction analyses indicated differences between the EBFT samples differing in their 281 282 shoreline distance position (i.e. offshore classes 1-5), these differences were not supported by individual based rarefaction. These results thus indicate that simple passive sampling effects 283 284 (i.e. the number of individuals caught) can explain the differences found in the number of species between the samples of the offshore classes (Gotelli and Colwell, 2001). Our large 285 scale spatial survey thus revealed that offshore areas are intensively used by a variety of 286 species which are distributed relatively homogenously in the river, at least regarding their 287 occurrence, because their abundance can vary largely at the mesoscale (i.e. between 500 m 288 long sampling stretches). These results on the River Danube, therefore, complement studies 289 from other large rivers (e.g. Dettmers et al., 2001; Wolter and Bischoff, 2001), and support 290 291 the view that main channel offshore areas provide important habitats for riverine fish assemblages which should be more intensively considered by habitat managers. 292 Although it is difficult to make direct comparisons, because of methodological 293

differences, density data of fishes were comparable with or even higher than the values found

for offshore fish assemblages in other large European rivers. For example, Wolter and 295 Freyhof (2004) estimated density values (i.e. mean CPUE per 1200  $\text{m}^{-2}$ , see Table IV daytime 296 data) ranging between 0.01 and 3.7 individuals for the 6 most common species using a 12 m 297 wide and 1.8 m high bottom otter trawl from a ship in the River Oder in Germany. For 298 comparison, our mean CPUE values ranged between 0.3 and 24.2 individuals obtained by 299 fishing 500 m long stretches with the 2 m wide and only 1 m high net (i.e. roughly 1000  $\text{m}^{-2}$ , 300 but lower water column depth) for the 12 most common benthic species. Note that although 301 302 the frame was electrified, we do not believe that these data would largely overestimate actual density values, because the electrofishing rather helped to catch dormant or hidden fish. 303 However, this methodological question remains to be tested in the future by fishing with 304 sonar combined devices (see Juza et al., 2013). Consequently, our study proves the broad 305 applicability of the EBFT in monitoring benthic fishes in the Danube, but how density (i.e. 306 307 CPUE) data would change using larger ships or bigger devices remains the topic of further research (see Juza et al., 2010). In fact it is likely that larger trawls could be more effective in 308 309 catching large specimens of many benthic and water column species in very large rivers, but care should be taken because increasing mesh size of the net may yield the underestimation 310 of the abundance of small benthic species. Nevertheless, due to its relatively easy handling we 311 propose that a two metre wide EBFT can be a reasonable compromise in the monitoring of 312 offshore areas in large rivers, when logistic constraints or other reasons (e.g. manoeuvring, 313 width and depth of the river, stopping of the net) may hinder the routine and wide scale 314 application of large trawls and fishing ships. 315

Night-time sampling of the shoreline using boat electrofishing was highly more effective in detecting species than offshore bottom trawling. Further, SE proved the occurrence of all fishes detected by the EBFT, with the exception of sterlet, which is a strictly deep channel trough species. However, the EBFT provided essential information on the occurrence and

abundance data of some benthic species in the river, which would have been highly 320 321 underestimated using only SE. Most importantly, we revealed the commonness and relatively high abundance of strictly protected Zingel species with the EBFT. Therefore, the EBFT 322 should be an essential device for monitoring spatial and temporal changes in the abundance of 323 these species of high conservation concern. In this respect, monitoring the stock of small 324 bodied benthic species is critically important. Consequently, we believe that night time SE 325 can be a very efficient cost effective method for monitoring fish assemblages for assessing 326 327 environmental health, if only a single device can be applied for logistical difficulties reasons. However, the sampling of offshore habitats is required for monitoring the status of some 328 species of high conservation concern (i.e. NATURA 2000 species, like zingel Zingel zingel 329 (L.) and Danube streber). To emphasize the importance of the previous finding note that the 330 JDS2 survey could not even prove the occurrence of Danube streber in the Hungarian Danube 331 332 river section (Wiesner et al., 2007).

An important methodological question is whether time of the day could influence our 333 comparisons between SE and EBFT catches. Several studies proved that night time 334 electrofishing of shoreline areas is more effective than day time sampling, because most 335 fishes are usually more active at night and many species move from offshore to inshore areas 336 at night (Wolter and Freyhof, 2004; Erős et al., 2008). Therefore, it is likely that for example 337 the shape of the species accumulation curves (Fig. 3) would differ less between the SE and 338 EBFT data set if we used day time SE data. Similarly, some differences in the catches of day 339 time and night time trawlings also exist due to movement of fish from offshore to inshore 340 areas at night (Wolter and Freyhof, 2004), although our preliminary studies could not prove 341 this finding due to large variability in CPUE data between hauls (unpublished data). These 342 differences however, did not influence the main findings of this study about the practical use 343 of offshore trawling in the monitoring of large river fishes for complementing shoreline data 344

sets. Additionally, nigh time trawling can be especially dangerous in the navigation channel
of ships, and therefore only day time sampling is recommended for security reasons and for
not disturbing the traffic of ships in very large rivers.

Larger specimens of many species were generally caught offshore, which suggests that 348 older age classes prefer these areas to shoreline areas as habitats. Therefore, to provide more 349 detailed information on the size structure of riverine fish assemblages offshore monitoring 350 351 would be essential. It is also clear, that shoreline areas present only a small fraction of large river habitats. For example, in the River Danube shoreline areas which can be effectively 352 monitored with electrofishing comprise only a maximum of 10 or 20 m wide zone of the 300-353 354 500 m wide channel. Therefore, although doubtlessly highly important for the diversity of fish assemblages, sampling the shoreline exclusively provide a biased picture of the composition 355 and structure of the fish assemblages in this very large river. Consequently, we recommend 356 357 the monitoring of offshore areas for a better understanding of fish assemblage composition and dynamics in the Danube and in other large rivers. 358

359

#### 360 Acknowledgements

361

This study is part of the doctoral (PhD) thesis of Zoltán Szalóky under the supervision of
Tibor Erős. The work of Tibor Erős was supported by the Bolyai János Scholarship of the
Hungarian Academy of Sciences and the OTKA K104279 research grant. We thank Clayton
Ridenour for pre-reviewing the ms and Péter Sály for drawing Fig. 1.

366

#### 367 **References**

368

Anderson, M.J., 2001. A new method for non-parametric multivariate analysis of variance.

Austr. Ecol. 26, 32–46.

- 371 De Leeuw, J.J., Buijse, A.D., Haidvogl, G., Lapinska, M., Noble, R., Repecka, R., Virbickas,
- T., Wisniewolski, W., Wolter, C., 2007. Challenges in developing fish-based ecological
  assessment methods for large floodplain rivers. Fish. Manage. Ecol. 14, 483-494.
- 374 Dettmers, J.M., Gutreuter, S., Wahl, D.H., Soluk, D.A., 2001. Patterns in abundance of fishes
- of the main channel of the upper Mississippi river system. Can. J. Fish. Aquat. Sci. 58,
  933-942.
- Doyle, W., Paukert, C., Starostka, A., Hill, T., 2008. A comparison of four types of sampling
  gear used to collect shovelnose sturgeon in the Lower Missouri River. J. Appl. Ichthyol.
  24, 637-642.
- Erős, T., 2007. Partitioning the diversity of riverine fish: the roles of habitat types and nonnative species. Freshw. Biol. 52, 1400-1415.
- Erős, T., Tóth, B., Sevcsik, A., Schmera, D., 2008. Comparison of fish assemblage diversity
  in natural and artificial rip-rap habitats in the littoral zone of a large river (River Danube,
  Hungary). Int. Rev. Hydrobiol. 93, 88-105.
- Flotemersch, J.E., Stribling, J.B., Hughes, R.M., Reynolds, L., Paul, M.J., Wolter, C, 2011.
- 386 Site length for biological assessment of boatable rivers. River Res. Appl. 27, 520-535.
- 387 Freedman, J.A., Stecko, T.D., Lorson, B.D., Stauffer, J.R., 2009. Development and efficacy
- of an electrified benthic trawl for sampling large-river fish assemblages. N. Am. J. Fish.
  Manage. 29, 1001-1005.
- Galat, D.L., Zweimüller, I. 2001. Conserving large-river fishes: is the highway analogy an
  appropriate paradigm? J. N. Am. Benthol. Soc. 20, 266-279.
- 392 Gotelli, N.J., Colwell, R.K. 2001. Quantifying biodiversity: procedures and pitfalls in the
- measurement and comparison of species richness. Ecol. Lett. 4, 379-391.

- Gutreuter, S., Vallazza, J.M., Knights, B.C., 2009. Lateral distribution of fishes in the mainchannel trough of a large floodplain river: Implications for restoration. River Res. Appl.
  26, 619-635.
- Harka, Á., Sallai, Z., 2004. The fish fauna of Hungary. NIMFEA, Szarvas. (in Hungarian)
- Herzog, D.P., Barko, V.A., Scheibe, J.S., Hrabik, R.A., Ostendorf, D.E. 2005. Efficacy of a
  benthic trawl for sampling small-bodied fishes in large-river systems. N. Am. J. Fish.
  Manage. 25, 594-603.
- 401 Hirzinger, V., Keckeis, H., Nemeschkal, H.L., Schiemer, F., 2003. The importance of inshore
- 402 areas for adult fish distribution along a free-flowing section of the Danube, Austria. River
  403 Res. Appl. 20, 137-149.
- Hughes, R.M., Kaufman, P.R., Herlihy, A.T., Intelman, S.S., Corbett, S.C., Arbogast, M.C.,
  Hjort, R.C., 2002. Electrofishing distance needed to estimate fish species richness in
  raftable Oregon rivers. N. Am. J. Fish. Manage. 22, 1229-1240.
- 407 Junk, W.J., Bayley, P.B., Sparks, R.E., 1989. The flood pulse concept in river-floodplain
- 408 systems, in: Dodge, D.P. (Ed.), Proceedings of the International Large River
- 409 Sysmposium (LARS). Can. Spec. Publ. Fish. Aquat. Sci.106, pp. 110-127.
- 410 Jurajda, P., Reichard, M., Hohausová, E. Černý J. 2001. Comparison of 0+ fish communities
- 411 between regulated-channelized and flooplain stretches of the River Morava. Arch.
- 412 Hydrobiol., Suppl. Large Rivers 12, 187-202.
- Jůza, T., Čech, M., Kubečka, J., Vašek, M., Peterka, J., Matěna, J., 2010. The influence of the
- trawl mouth opening size and net colour on catch efficiency during sampling of early fish
- 415 stages. Fish. Res. 105, 125-133.

- 416 Jůza, T., Rakowitz, G., Draštík, V., Blabolil, P., Herzig, A., Kratochvíl, M., Muška, M., Říha,
- M., Sajdlová, Z., Kubečka, J. 2013. Avoidance reactions of fish in the trawl mouth
  opening in a shallow and turbid lake at night. Fish. Res. 147, 154-160.
- 419 Labonne, J., Allouche, S., Gaudin, P., 2003. Use of a generalized linear model to test habitat
- 420 preferences: the example of *Zingel asper*, an endemic percid of the River Rhone. Freshw.
- 421 Biol. 48, 687-697.
- Murphy, B.R., Willis, D.W. (Eds.), 1996. Fisheries techniques, 2nd ed.. American Fisheries
  Society, Bethesda, Maryland.
- 424 R Development Core Team, 2013. R: A language and environment for statistical computing.
- 425 R Foundation for Statistical Computing, Vienna, Austria. http://www.R-project.org/
- 426 Reyjol, Y., Hugueny, B., Pont, D., Bianco, P.G., Beier, U., Caiola, N., Casals, F., Cowx, I.,
- 427 Economou, A., Ferreira, T., Haidvogl, G., Noble, R., De Sostoa, A., Vigneron, T.,
- 428 Virbickas, T., 2007. Patterns in species richness and endemism of European freshwater
- fish. Global Ecol. and Biogeog., 16, 65-75.
- 430 Ridenour, C.J., Doyle, W.J., Hill, T.D., 2011. Habitats of age-0 sturgeon in the Lower
- 431 Missouri River. Trans. Am. Fish. Soc. 140, 1351-1358.
- 432 Ridenour, C.J., Starostka, A.B., Doyle, W.J., Hill, T.D., 2009. Habitat used by Macrohybsis
- 433 chubs associated with channel modifying structures in a large regulated river:
- 434 implications for river modifications. River Res. Appl. 25, 472-485.
- Stewart, D.J., Ibarra, M., Barriga-Salazar, R., 2002. Comparison of deep-river and adjacent
  sandy-beach fish assemblages in the Napo River Basin, eastern Ecuador. Copeia 2002,
  333-343.
- 438 Szalóky, Z., György, Á.I., Csányi, B., Tóth, B., Sevcsik, A., Szekeres, J., Erős, T., 2011.
- 439 Development and testing of an electrified benthic trawl for monitoring benthic fish

- assemblages in the River Danube. Pisces Hungarici 5: 37-42. In Hungarian with asummary in English.
- 442 Wiesner, C., Schotzko, N., Cerny, J., Guti, G., Davideanu, G., Jepsen, N., 2007. Technical
- report with results from the fish sampling and analyses from the Joint Danube Survey
- 444 2007. International Commission for the Protection of the Danube River, Vienna. 73pp.
- 445 Wolter, C., Bischoff, A., 2001. Seasonal changes of fish diversity in the main channel of the
- large lowland river Oder. Reg. Rivers. Res. Manage. 17, 595-608.
- 447 Wolter, C., Freyhof, J., 2004. Diel distribution patterns of fishes in a temperate large lowland
- 448 river. J. Fish Biol. 64, 632-642.

# **Table 1**

The species composition, relative abundance (A%) and frequency of occurrence (FrO%) of
fishes collected by shoreline electrofishing (SE) and by trawling with the electrified benthic
framed net (EBFT) in the River Danube, Hungary.

	SE		EBFT		
Species name	A%	FrO%	A%	FrO%	
Abramis brama (L.)	0.84	42.51	0.97	16.00	
Acipenser ruthenus L.	-	-	0.02	1.14	
Alburnus alburnus (L.)	29.80	98.55	4.25	36.00	
Anguilla anguilla (L.)	0.01	0.48	-	-	
Aspius aspius (L.)	1.94	78.26	0.04	1.14	
Babka gymnotrachelus (Kessler)	0.69	41.06	0.53	8.57	
Ballerus ballerus (L.)	0.04	4.35	0.02	1.14	
Ballerus sapa (Pallas)	0.52	34.30	0.57	13.71	
Barbus barbus (L.)	0.59	35.75	1.64	37.14	
Blicca bjoerkna (L.)	14.36	91.79	9.27	49.71	
Carassius gibelio (Bloch)	0.28	28.50	0.01	0.57	
Chondrostoma nasus (L.)	2.46	55.56	0.12	4.57	
Cobitis elongatoides (Bacescu and Mayer	0.00	0.48	-	-	
Ctenopharyngodon idella (Valenciennes)	0.00	0.48	-	-	
<i>Cyprinus carpio</i> L.	0.26	16.91	0.01	0.57	
<i>Esox lucius</i> L.	0.17	17.39	-	-	
Eudontomyzon mariae (Berg)	0.01	2.90	-	-	

Gasterosteus aculeatus L.	0.00	0.48	-	-
Gymnocephalus baloni Holcík and Hensel	0.47	26.57	0.63	2.86
Gymnocephalus cernua (L.)	0.21	10.14	0.05	1.71
Gymnocephalus schraetser (L.)	4.08	72.46	4.83	36.00
Hypophthalmichthys molitrix (Valenciennes)	0.00	0.48	-	-
Lepomis gibbosus (L.)	0.02	2.90	-	-
Leuciscus idus (L.)	2.46	75.36	0.64	16.00
Leuciscus leuciscus (L.)	0.03	4.35	-	-
Lota lota (L.)	2.77	43.96	0.01	0.57
Neogobius fluviatilis (Pallas)	0.91	46.86	0.23	6.86
Neogobius melanostomus (Pallas)	18.32	84.54	52.18	52.00
Pelecus cultratus (L.)	0.07	14.49	0.02	1.14
Perca fluviatilis L.	0.69	21.74	0.02	1.14
Ponticola kessleri (Günther)	3.01	82.13	1.06	14.86
Proterorhinus marmoratus (Pallas)	0.11	10.63	0.02	1.14
Pseudorasbora parva (Temminck and Schlegel)	0.00	0.97	-	-
Rhodeus sericeus (Pallas)	0.03	0.97	-	-
Romanogobio albipinnatus (Lukasch)	6.63	73.43	8.12	75.43
Rutilus rutilus (L.)	1.46	56.04	0.02	1.14
Rutilus virgo (Heckel)	0.35	29.47	0.21	8.00
Sabanejewia aurata (De Filippi)	0.11	5.80	0.11	2.29
Sander lucioperca (L.)	3.27	79.23	0.43	14.86
Sander volgensis (Gmelin)	0.43	40.58	0.04	1.14
Scardinius erythrophthalmus (L.)	0.01	2.42	-	-

Silurus glanis L.	0.08	11.11	0.05	1.71
Squalius cephalus (L.)	1.11	42.51	-	-
Vimba vimba (L.)	1.10	42.03	0.30	6.86
Zingel streber (Siebold)	0.06	6.76	11.81	74.29
Zingel zingel (L.)	0.27	28.50	1.73	25.14
Number of species	45		33	
Number of individuals	47731		8112	
Number of samples	207		171	

456 **Table 2** 

457 Standard length (mm) data of the most common benthic species collected by shoreline electrofishing (SE) and trawling with the electrified

458 benthic framed net (EBFT) in the River Danube, Hungary. Differences in the distribution and median values of data between the sampling gears

459 were examined with non-parametric Kolmogorov-Smirnov test and Mann-Whitney U test, respectively.

	SE	EBFT				Kolmogorov-Smirnov test	Mann-Whitney U test			
	Median	Min.	Max.	N	Median	Min.	Max.	N	р	р
Abramis brama	78	21	440	242	325	100	509	78	<0.001	<0.001
Barbus barbus	71	35	600	105	360	40	610	127	<0.001	<0.001
Blicca bjoerkna	85	33	280	2005	91	35	330	642	<0.001	<0.001
Chondrostoma nasus	110	45	450	416	273	107	420	10	<0.05	<0.001
Gymnocephalus schraetser	70	11	220	1162	75	40	170	387	<0.001	< 0.001
Lota lota	230	28	480	605	240	240	240	1	-	-
Neogobius melanostomus	62	18	165	1470	40	15	125	1957	<0.001	< 0.001
Ponticola kessleri	70	30	165	385	72	45	115	85	< 0.001	ns

Zingel zingel	81	60	350	68	84	45	275	133	ns	ns
Zingel streber	54	45	95	15	66	29	175	932	<0.01	< 0.05
Vimba vimba	85	32	185	323	181	80	380	23	< 0.001	< 0.001
Romanogobio albipinnatus	71	18	150	1388	64	18	125	617	< 0.001	< 0.001

462 Legends to figures

463

464 Fig. 1. Schematic picture and parameters of the electrified benthic framed trawl (EBFT)
465 developed to sample fish in the offshore areas of the Danube River, Hungary.

466

Fig. 2. The spatial distribution of samples along the 350 km long section of the Danube River
Distances (m, mean±SD) of five classes of electrified benthic framed trawl (EBFT) samples
from the right and left (in parentheses) river banks are indicated. Shoreline electrofishing (SE)
samples were taken at mainly 2-5 m distances off the shore. Solid and open circles represent
SE and EBFT samples, respectively.

472

473 Fig. 3. Estimated number of species (±95% CI.) as a function of (a) number of samples and
474 (b) number of individuals collected with shoreline electrofishing (SE) and the electrified
475 benthic framed trawl (EBFT) in the Danube River, Hungary.

476

Fig. 4. Estimated number of species (±95% CI) as a function of (a) number of samples and (b)
number of individuals collected with the electrified benthic framed trawl (EBFT) in five
classes of samples differing in their offshore position in the Danube River, Hungary (Fig. 2).

Fig. 5. Mean (±SD) catch per unit effort data (CPUE, ind 500 m<sup>-1</sup>) of the benthic species in
the EBFT collections in the Danube River, Hungary. The symbol \* indicates significant
differences at p<0.05 level.</li>

484

- Fig. 6. Length frequency distribution of the most common benthic species collected with
  shoreline electrofishing (SE) and the electrified benthic framed trawl (EBFT). See Table 2 for
  sample numbers.