



# Systematic point sampling of fish communities in medium- and large-sized rivers: sampling procedure and effort

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**Abstract** Compared with small rivers and streams, the study of fish communities in large rivers remains challenging as spatial and temporal data variability can be greatly influenced by sampling strategy and operator choice. In an attempt to limit this variability, a new sampling protocol for fish communities in medium- to large-sized rivers was developed, based on point sampling by electric fishing and using standardised procedures and effort. Here, change in data quality (assemblage abundance, richness, structure and biotic index) with increasing sampling effort (from 1 to 100 sampling points) was evaluated. A total of 75 sampling points are proposed as the standard number of samples per site. Broadly, the results show that the application of 75 sampling points provides a reproducible representation of fish community structure in medium and large rivers, with little additional information provided by further sampling except under certain conditions, when 100 points are recommended to maintain data quality.

**KEY WORDS :** electric fishing standardisation, non-wadeable rivers, partial sampling technique, survey.

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## Introduction

Accurate assessment of river ecosystems in space and time requires standardised methods that provide comparable data of equivalent scientific quality. For riverine fish, however, obtaining high-quality field data with an acceptable degree of precision is still a major problem.

Unlike streams and small rivers, where sampling of an entire reach is possible, the study of fish communities in large, non-wadeable rivers remains one of the most difficult problems in freshwater ecology. As suggested by Persat and Copp (1989), rather than maintaining the delusion that estimates of absolute fish density or biomass may be obtained in large rivers, it is better to admit

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that such precise estimates are practically impossible to achieve. Instead, the random character of the samples should be accounted for by choosing an appropriate sampling method and strategy. Regardless of whether relative or absolute parameters are to be estimated, it is essential that fishing procedures be standardised in all possible respects (Bohlin *et al.* 1990).

Prior to 2006, several electric fishing sampling strategies and techniques (*sensu* Copp 2010) had been applied in non-wadeable, medium and large river reaches for the French national fish survey (Kestemont & Goffaux 2002): (1) continuous bank sampling, that is, continuous sampling of a 2-m wide stretch along both river banks; (2) ambience sampling (inspired from the works of Pouilly 1994 and Capra 1995), that is, space-stratified sampling of several meso-habitat units with ambiances of 5–100 m<sup>2</sup>; and (3) point abundance sampling using electric fishing (PASE; Nelva *et al.* 1979). Fishing teams, however, reported several methodological problems when using these methods for large-scale and long-term monitoring. (1) Continuous bank sampling was time consuming and required great effort as a large fish sample was frequently collected. This was particularly the case since ratification of the European standard for fish sampling with electricity (EN14011 2003), which requires that the minimum length of study site be 20× the river's width (10× for large and homogenous rivers). (2) The ambience sampling approach also generated a large fish sample and suffered from low sample reproducibility as the number and surface area of sampling units were both variable and non-standardised. Both continuous and ambience sampling generated large samples (e.g. several thousands of fish), and extended periods of fish handling (e.g. size and weight measures, pathological examination) result in elevated fish mortalities, posing a particular problem for long-term monitoring.

Of the methods used, PASE appeared to be most suitable in terms of sample size produced. PASE originally gained acceptance from the scientific community for sampling of young-of-the-year fish (YOY, for a review see Copp 2010) and has since been used in numerous ecological studies around Europe in rivers of various size (e.g. Copp 1989; Copp *et al.* 1994, 2005a; Pires *et al.* 1999; Wolter & Bischoff 2001; Fladung *et al.* 2003; Valová *et al.* 2006), lakes or wetlands (Perrow *et al.* 1996; Cucherousset *et al.* 2006). In 2006, the French National Agency for Water and Aquatic Environments (ONEMA) implemented a point sampling approach for monitoring medium- and large-sized rivers. Several aspects of the original sampling approach of Nelva *et al.* (1979), however, were adapted to increase sampling efficiency and improve repeatability with respect to monitoring goals and constraints. For example,

fishless points occur relatively frequently in large rivers and are often considered by operators to be unproductive, which results in a gradual shift toward the selection of point samples in areas where fish are more likely to be captured.

To reduce potential operator bias, the random sampling strategy was replaced with a systematic strategy that ensured proportional sampling of all fishable habitats. To reduce variability in the area fished, and for health and safety reasons, the anode was not thrown in front of the boat, as originally described by Nelva *et al.* (1979), but rather immersed in the water by the operator as described by Copp and Garner (1995). Finally, to increase and standardise the size of each sample, and hence the likelihood of capturing large-bodied and/or rare fishes, the anode was moved around each sample point for a minimum of 15 s and maximum of 30 s in a 1-m-diameter circle, a modification of the PASE sampling technique applied in shallow lakes (Perrow *et al.* 1996).

The sampling effort required to provide a reliable picture of the fish assemblage at a given site is of great importance. In attempting to evaluate the number of PASE samples required to estimate YOY density accurately, Garner (1997) suggested that as many samples as possible should be taken, with a minimum of 50. Other studies have compared the efficiency of point sampling with other methods for sampling YOY (e.g. Janáč & Jurajda 2005). Relatively little quantitative evidence exists, however, as regards the reliability of results obtained from point abundance sampling of adult fish – but see Perrow *et al.* (1996) for a comparison with stop-nets in shallow lakes, Brosse *et al.* (2001) for a comparison with scuba sampling in reservoir littoral areas, Lapointe *et al.* (2006) for a test of sampling duration in a large river, and Brousseau *et al.* (2005) for a comparison of transect and point sampling in the littoral zone of large lakes. Previous studies (Persat & Copp 1989; Pretty *et al.* 2003) have applied from 20 to 50 sampling points per site, but without further examination of the reliability of the results. Bady and Pont (2008) proposed that accurate evaluation of a fish assemblage should be based on samples of at least 100 individuals, independent of sampling strategy, which suggests that the minimal sampling effort for point sampling should also approach this figure.

As regards the systematic point sampling protocol recently implemented by ONEMA, it remains unclear how many points are needed to obtain a reliable picture of the entire fish assemblage. The main objective, therefore, was to evaluate how estimates of fish assemblage structure in medium and large rivers change when PASE sampling effort is increased. To this end, a systematic

sampling approach was applied, using a standard of 100 point samples (e.g. Copp 1997), in 12 rivers with different habitat characteristics and recorded fish data at each point. The levels of sampling effort applied in previous studies using PASE, that is, 25 points (Persat & Copp 1989), 50 points (Pretty *et al.* 2003), 75 points (as proposed by ONEMA for their national fish survey) and 100 points (Copp 1997), were then used to evaluate the influence of sampling effort on fish assemblage results. This addressed four specific questions: (1) Are 25, 50, 75 or 100 points enough to catch a minimum of 100 individuals (as recommended by Bady & Pont 2008); (2) how many species are not captured when using 25, 50 or 75 points compared with 100 points? (3) is an equivalent picture of fish assemblage structure found when applying 25, 50, 75 and 100 sampling points? and (4) can river characteristics affect data quality in relation to sampling effort, that is, should the sampling effort be adapted according to river characteristics?

## Materials and methods

### Study sites and sampling method

Sampling took place during autumn 2004 at 12 river sites of differing size and habitat heterogeneity (Table 1). Following the European standard (EN14011 2003), the minimum length ( $L$ ) of river reach depended on river width ( $w$ ) as follows:  $L = 20 \times w$  if  $w < 30$  m;  $L = 600$  m if  $30 \text{ m} < w < 60$  m; and  $L = 10 \times w$  if  $w > 60$  m.

For the purposes of this study, electric fishing was undertaken only in those zones where it is most efficient

(i.e. depth  $< 1$  m and water velocity  $< 1 \text{ m s}^{-1}$ ) within each river reach (deeper zones from a boat and shallower by wading) using a Heron-type electric generator with a 35-cm ring anode (electric output ranging from 400 to 600 V and 2 to 4 A).

Unlike the original point abundance sampling strategy (Nelva *et al.* 1979; Persat & Copp 1989), systematic point sampling consists of collecting numerous point samples evenly distributed over the entire study site or reach (where electric fishing is both efficient and secure). The fishing team moves upstream, by wading or by boat, from one bank to the other in a zigzag manner, taking equidistant point samples in fishable zones (see Fig. 1 for several possible sampling procedures). Upon arrival at each sampling point, the anode is immersed and moved around in a 1-m-diameter, horizontal circle for 15–30 s. To define the influence range of the anode, several preliminary tests were performed under different conductivity conditions ( $230\text{--}550 \mu\text{S cm}^{-1}$ ) consisting of repeated measurements of voltage gradient between two points 10 cm apart at different distances from the centre of the anode (i.e. 0.5, 1, 1.5, 2, 3 and 4 m). Galvanotaxis can be achieved at voltage gradients as low as  $0.1 \text{ V cm}^{-1}$  when using direct current. This value was, therefore, taken into account when computing the actual influence range. These tests showed that voltage gradients of  $0.1 \text{ V cm}^{-1}$  were always measured within 1.5 m from the centre of the anode (Table S1), and therefore, moving the anode around a horizontal circle of 1 m diameter would result in a sampling unit surface area of  $\approx 12.5 \text{ m}^2 (= 3.14 \times (0.5 + 1.5)^2)$ . To limit anode influence between each sample site, a 5 m minimum distance was set between sampling points when fishing by

**Table 1.** Characteristics of sites where systematic point sampling tests took place. Note that habitat heterogeneity was visually estimated by members of the fishing teams

| River            | Latitude     | Longitude   | Drainage area<br>(km <sup>2</sup> ) | Distance from source<br>(km) | Mean width<br>(m) | Mean depth<br>(m) | Habitat<br>heterogeneity | Fishing |
|------------------|--------------|-------------|-------------------------------------|------------------------------|-------------------|-------------------|--------------------------|---------|
| Aisne            | 49.398267° N | 3.474425° E | 5589                                | 278                          | 60                | 3                 | Low                      | Boat    |
| Besbre           | 46.201575° N | 3.667964° E | 360                                 | 39                           | 18                | 0.6               | High                     | Wading  |
| Bouzanne         | 46.635576° N | 1.592040° E | 480                                 | 69                           | 12                | 0.6               | Low                      | Wading  |
| Charente         | 45.623221° N | 0.035769° W | 4404                                | 267                          | 50                | 3                 | Medium                   | Boat    |
| Ill              | 48.655005° N | 7.842170° E | 4736                                | 213                          | 50                | 1.2               | Low                      | Boat    |
| Loire            | 45.307270° N | 4.118988° E | 3254                                | 100                          | 50                | 0.9               | Medium                   | Mixed   |
| Meuse –<br>Chooz | 50.111496° N | 4.802046° E | 10387                               | 476                          | 70                | 1                 | Medium                   | Mixed   |
| Meuse –<br>Han   | 48.870220° N | 5.542255° E | 2590                                | 173                          | 25                | 0.7               | High                     | Mixed   |
| Rhône            | 45.609840° N | 4.800908° E | 51 080                              | 530                          | 90                | 2.9               | Low                      | Boat    |
| Tarn             | 44.114737° N | 1.159162° E | 15 132                              | 369                          | 155               | 3.8               | Low                      | Boat    |
| Taurion          | 45.987946° N | 1.866993° E | 276                                 | 24                           | 18                | 0.5               | High                     | Wading  |
| Vienne           | 47.150880° N | 0.305559° E | 20 358                              | 346                          | 120               | 0.8               | High                     | Boat    |

wading and 10 m when sampling from a boat. At each sampling site, 100 sampling points were performed, covering the entire river reach. At each point sampled, captured fishes were identified to species level and counted.

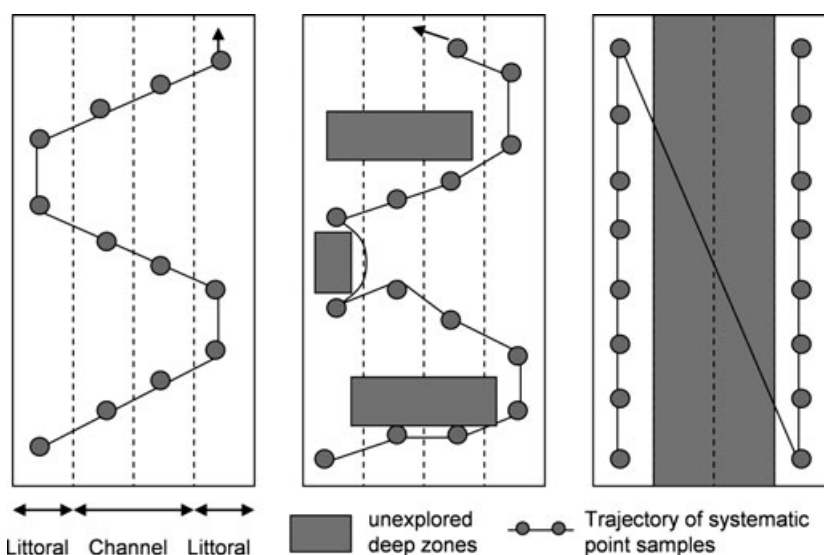
### Data analysis

To assess how fish assemblage data (total number of captures, species richness and assemblage structure) varied with increasing sampling effort (25, 50, 75 and 100 sampling points), the minimum sampling effort required to catch 100 individuals (as per Bady & Pont 2008) was assessed using rarefaction curves (Colwell & Coddington 1994) modified by replacing species richness evaluation by total number of individuals. The total number of captured individuals (TNI) for each river and sampling effort (1–100 points) was computed 1000 times for each river site by permuting the sequence of sampled points. The resulting 1000 simulated TNI values provided a mean and standard deviation for each sampling effort.

During the second step, changes in species richness with increasing sampling effort were assessed using species accumulation curves (SACs) – a commonly used method to determine whether sample size applied is large enough to represent an assemblage accurately (e.g. Angermeier & Smogor 1995; Cao *et al.* 2001; Lapointe *et al.* 2006; Copp 2010). A SAC not achieving an asymptote indicates that not all species have been detected. In this study, SACs were constructed for each sampling site using the vegan library's *specaccum* function (Oksanen *et al.* 2008) in the R software package (R Development Core Team 2011). The exact option from this function was used to obtain the expected SAC (and

its standard deviation) following the method independently developed by Ugland *et al.* (2003) and Colwell *et al.* (2004). The exact accumulation curve is independent of the underlying species abundance distributions, but is strongly influenced by the distribution of species among the samples and the spatial configuration of the samples that are randomised (Ugland *et al.* 2003). To evaluate species richness with increasing sampling effort at each sampling site, the number of species remaining to be captured was estimated for each sampling effort as the difference between the species richness achieved with 100 sampling points and the expected SAC values for each sampling effort. Finally, the mean and standard deviation of non-captured species was computed for the 12 rivers to evaluate the general loss of information.

Species abundance was estimated (log-transformed,  $\ln m^{-2}$ ) for each sampling effort (i.e. from 1 to 100 points) to evaluate the variation in fish assemblage structure at each sampling site with increasing sampling effort. Fish communities assessed under differing sampling efforts were evaluated separately for each site using principal components analysis (PCA). For grouping different sampling efforts providing similar results, cluster analysis was applied to the first four PCA axes (always describing more than 96% of total variability), using Euclidean distances as a measure of similarity and Ward's grouping method. This method uses minimal intra-cluster variance to evaluate distances between clusters. The data from each sampling effort (from  $E_{S1}$  to  $E_{S100}$ ) were then classified into five clusters. This procedure was repeated 1000 times for each site after permuting the sequence of sampled points at the beginning of the analysis. From the resulting 1000 PCA analyses of



**Figure 1.** Schematic examples of systematic point sampling protocols under different habitat conditions (unexplored zones: depth >1 m or depth <1 m and water velocity >1 m s<sup>-1</sup>).

permuted sampling point sequences, the probability of each sampling effort being in the same cluster was computed as  $E_{S100}$  for each site.

As ONEMA proposed a sampling effort of 75 points for their yearly surveys (mainly focused on evaluating river ecosystem status under the European Water Framework Directive), potential differences between  $E_{S75}$  and  $E_{S100}$  over a commonly used water management tool were evaluated further, that is, quality estimates based on the fish assemblage. For each river, the French Fish Biotic Index (FBI; see Oberdorff *et al.* 2002) was computed 100 times using  $E_{S75}$ , that is,  $FBI_{75}$  was computed with the first 75 points resulting from randomly permuted sequences of 100 sampled points, as above.  $FBI_{100}$  was then compared with the distribution of 100 simulated  $FBI_{75}$  results.

Finally, environmental and biological parameters (i.e. habitat heterogeneity, depth, total species richness and frequency of fishless points) and other features related to the fishing method (i.e. fishing by wading, by boat, or mixed) were analysed to evaluate their influence on the probability of  $E_{S75}$  showing an equivalent fish assemblage to  $E_{S100}$ . As the study was conducted on a small number of river sites, selected parameters were expressed in two or three categories and their influence evaluated graphically (box-whisker plot).

## Results

At 11 of the 12 study sites (River Bouzanne excluded), the minimum of 100 individuals was always exceeded with 25 sampling points, TNI increasing rapidly with increasing sampling effort (Table 2). Large standard errors for simulated TNI values were logically associated with high heterogeneity in capture rates between points.

In the majority of cases, SACs (see Appendix 1) closely approached the asymptote, suggesting that 100 sampling points could yield a reliable estimate of total species richness. For some sites, however, and particularly for the River Bouzanne, SACs indicated that, even after 100 sampling points, several species were not detected. When mean species richness estimated from SACs for each sampling effort was compared against species richness at sites sampled with 100 points (Fig. 2), the number of non-captured species decreased rapidly with increasing sampling effort until around 40 sampling points, whereupon it gradually levelled off. This also indicates that several species may still not be detected when applying a sampling effort of 25 or 50 points. Compared with the number of species captured with 100 point samples, the mean number of species ( $\pm$ SD) not captured with 25 or 50 points was 4.6 ( $\pm$ 1.7) and 2.2 ( $\pm$ 1), respectively. With 75 point samples (Fig. 2), the mean number of missed species was 0.9 ( $\pm$ 0.5). Clearly, these numbers would be greater if 100 points were not sufficient to capture all species.

Based on mean probability ( $P$ ), there was a negligible chance of obtaining a similar fish assemblage from 25 point samples as from 100 samples at the 12 river sites ( $P = 0.006 \pm 0.004$ ; Fig. 3). As sample number increased from 50 to 75 samples,  $P$  ( $\pm$ SD) increased rapidly from 0.16 ( $\pm$ 0.06) to 0.75 ( $\pm$ 0.07), but less rapidly between 75 and 100 points. Thus, 50 point samples are insufficient for assessing fish assemblage structure, as there is a low probability that the outcome will not change with increasing sampling effort. Instead, 75 point samples appear to be a good compromise as the probability remains relatively high ( $P = 0.75$ ) that similar a fish assemblage structure will be observed as that from 100 sample points. When tested with FBI,  $FBI_{100}$  fell

**Table 2.** Mean total number of individuals (TNI  $\pm$  SD) captured with sampling efforts ( $E_S$ ) of 25, 50, 75 and 100 points resulting from the rarefaction procedure

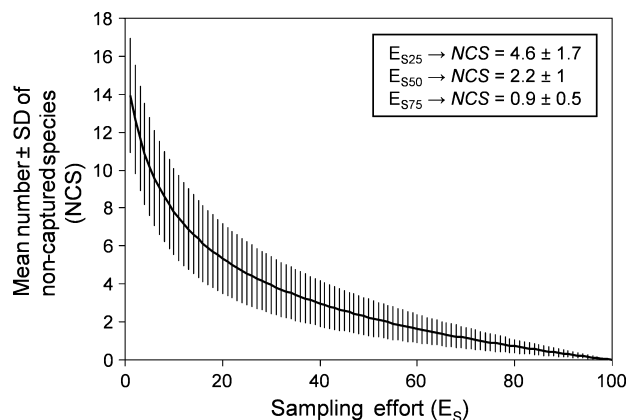
| River         | TNI ( $E_{S25}$ ) $\pm$ SD | TNI ( $E_{S50}$ ) $\pm$ SD | TNI ( $E_{S75}$ ) $\pm$ SD | TNI ( $E_{S100}$ ) |
|---------------|----------------------------|----------------------------|----------------------------|--------------------|
| Aisne         | 133 $\pm$ 41               | 264 $\pm$ 48               | 396 $\pm$ 41               | 528                |
| Besbre        | 182 $\pm$ 114              | 362 $\pm$ 129              | 537 $\pm$ 114              | 718                |
| Bouzanne      | 26 $\pm$ 19                | 51 $\pm$ 22                | 77 $\pm$ 19                | 104                |
| Charente      | 134 $\pm$ 43               | 267 $\pm$ 49               | 402 $\pm$ 42               | 537                |
| Ill           | 122 $\pm$ 25               | 243 $\pm$ 30               | 365 $\pm$ 25               | 487                |
| Loire         | 233 $\pm$ 116              | 467 $\pm$ 142              | 702 $\pm$ 122              | 945                |
| Meuse – Chooz | 807 $\pm$ 330              | 1595 $\pm$ 371             | 2410 $\pm$ 343             | 3250               |
| Meuse – Han   | 208 $\pm$ 63               | 416 $\pm$ 74               | 626 $\pm$ 62               | 832                |
| Rhône         | 169 $\pm$ 43               | 335 $\pm$ 49               | 504 $\pm$ 43               | 674                |
| Tarn          | 273 $\pm$ 296              | 554 $\pm$ 342              | 829 $\pm$ 301              | 1108               |
| Taurion       | 134 $\pm$ 42               | 266 $\pm$ 47               | 398 $\pm$ 40               | 528                |
| Vienne        | 230 $\pm$ 139              | 474 $\pm$ 165              | 702 $\pm$ 137              | 939                |

TNI, total number of captured individual.

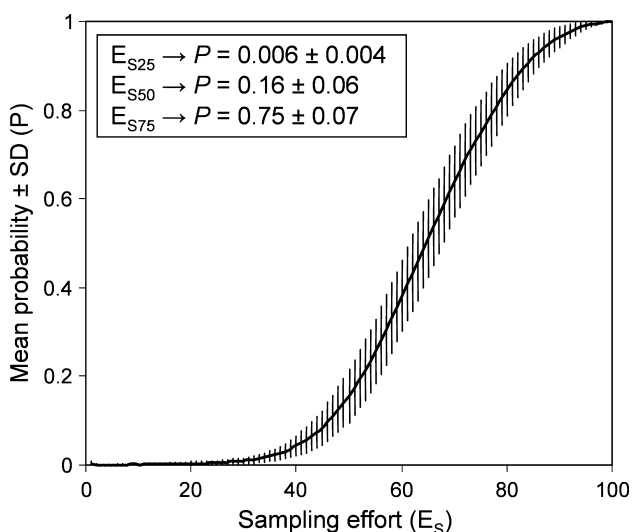


within the simulated distribution of FBI<sub>75</sub> for all 12 rivers, and in most cases, no change was observed in the resulting quality level (Fig. 4).

Finally, habitat variables and fishing method appeared to have a slight influence on the probability of observing an equivalent fish assemblage as E<sub>S100</sub> at E<sub>S75</sub> (Fig. 5). The probability at E<sub>S75</sub> was lower in deep river reaches (>1.5 m) with low habitat variability, the number of species being lower and the frequency of fishless samples >30%. There was no difference in *P* when fishing was undertaken by wading or by using a boat, although *P* was higher when both fishing strategies were employed together.



**Figure 2.** Mean number of species ( $\pm$ SD) remaining to be captured at each sampling effort level (compared with number of species captured with 100 points; see also Appendix 1).



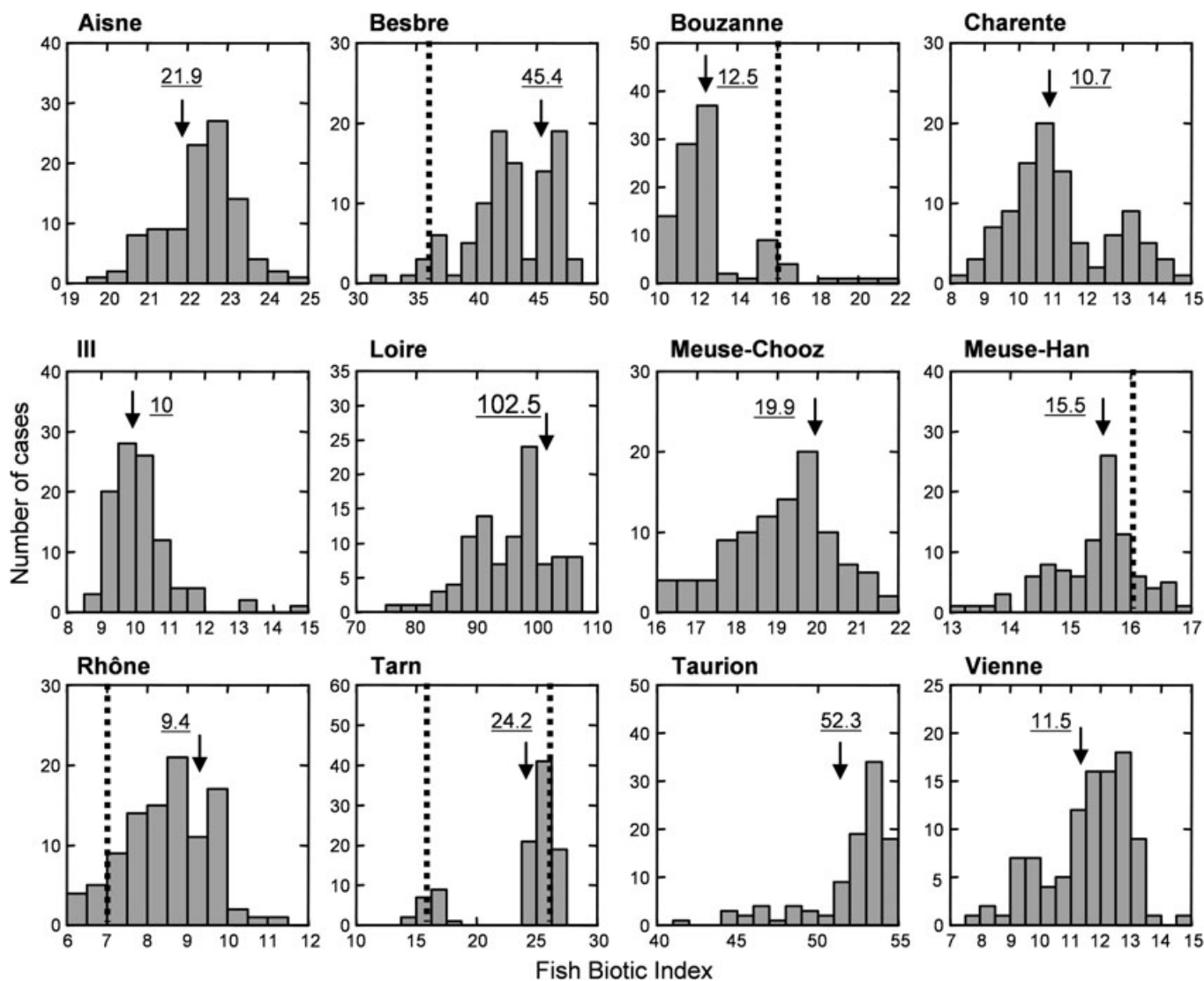
**Figure 3.** Mean probability ( $P \pm$ SD) of estimating a similar fish assemblage as observed with maximum sampling effort (E<sub>S100</sub>) at each sampling effort level.

## Discussion

Many sampling approaches exist for studying fish communities in large rivers (see Casselman *et al.* 1990), each one having advantages and disadvantages. Irrespective, the sampling strategy and technique employed must be appropriate for the purpose of the investigation (Copp 2010). To ensure maximum data quality, and to allow for comparisons of spatial and temporal variability, all possible biases in fishing efficiency must be avoided, in particular, the variability generated by subjective choices of operators (e.g. habitats sampled and fishing effort). The fishing methodology used in this study, that is, an adaptation of the PASE approach for annual routine fish surveys at a national scale, aims to minimise such potential biases and to detect eventual ecological changes.

The results show that for 11 of 12 rivers studied, the recommended minimum number of individuals in a sample required for reliable fish assemblage studies (Bady & Pont 2008) might be achieved with 25 sampling points only, but was more likely to be achieved with  $\geq 50$  points (Table 2). When sampling effort was increased from 75 to 100 point samples (Figs 2 & 3), few new species were captured and the resulting changes in assemblage structure were slight. Therefore, 75 points are, in most cases, adequate for investigations of fish assemblage and species richness. This was supported by the lack of major differences between the FBI<sub>75</sub>-simulated biotic quality classes and the final scores for FBI<sub>100</sub> (Fig. 4). The distributions for simulated FBI<sub>75</sub> were almost normal, with the majority of values close to the final observed value computed with 100 points. In some cases (e.g. the rivers Tarn, Besbre and Charente), however, bimodal-simulated distributions of FBI<sub>75</sub> were observed, although the quality classification was rarely altered. These bimodal distributions were produced as a result of heterogeneity in fish captures between points, that is, the occurrence of one or two points with high fish abundance and random selection by the permutation procedure. On the River Tarn, for example, over 600 juveniles were caught at one sampling point, contrasting the mean of four juveniles per sample for all other point samples. The inclusion, or absence, of this extreme sample logically impacts on the final fish assemblage obtained (as discussed in Persat & Copp 1989) and is reflective of the shoaling behaviour of juveniles of some species (e.g. cyprinids).

More intensive sampling is likely to produce only limited additional information. Under certain conditions, such as deep river sites with low habitat heterogeneity, rivers with low species richness or rivers with frequent fishless points (see Fig. 5), 100 points are recommended to increase data quality. Further, if the TNI from 75



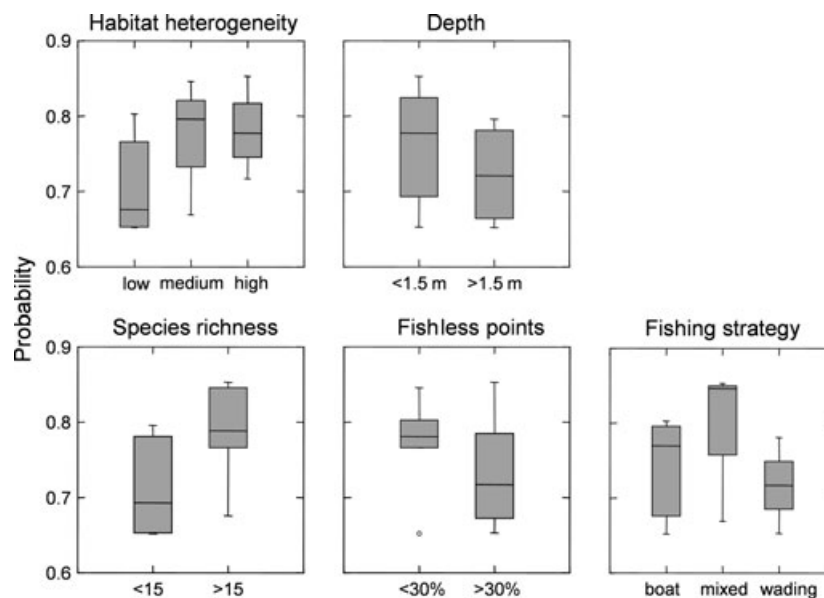
**Figure 4.** Distribution of 100 simulated values using the Fish Biotic Index (FBI) of Oberdorff *et al.* (2002), computed with  $E_{S75}$  using a randomisation procedure (i.e. 100 permutations of the sampling points and selection of the first 75 points to compute  $FBI_{75}$ ) for each study site. The numbers and arrows indicate the FBI computed with  $E_{S100}$ , and the dashed lines indicate the limits of different FBI quality classes.

points does not exceed 100 individuals, then sampling effort should also be increased to 100 point samples. These results confirm previous studies suggesting that greater sampling effort is needed in homogeneous (regulated) rivers with low-density fish communities (Angermeier & Smoger 1995).

The SAC asymptote was still not completely stabilised after 100 sampling points at several sites (see Appendix 1), suggesting that all species present were still not captured. Accumulation curves are strongly influenced by the distribution of species among randomised points (Ugland *et al.* 2003), so when more (rare) species are captured at single sampling points (from 100 performed), the asymptote will not be achieved. This means that,

even when all species from a site had been sampled and several rare species were present at single sampling points, the resulting SAC would incorrectly indicate non-stabilised species richness.

The opposite situation could also occur, where all species are not captured but those captured occur in at least two samples, then the SAC will incorrectly indicate stabilised species richness. As a consequence of these limits, SAC estimates of missing species should be treated with caution. Following a similar sampling principle, Smith and Jones (2005) proposed completion of the sampling protocol with targeted sampling of rare species, which are generally difficult to catch. In the case of systematic point sampling with pre-defined point placement



**Figure 5.** Influence of habitat parameters and fishing method on the probability ( $P$ ) that  $E_{S75}$  will estimate an equivalent fish assemblage to  $E_{S100}$ .

(Fig. 1), complementary targeted sampling would certainly be helpful in detecting rare species. Consequently, in addition to the standard number of sampling points, it is recommended that 10 additional sampling points be collected from rare habitats (i.e. those potentially inhabited by rare species) to improve estimates of species occurrence and to gain valuable local knowledge on the habitat preferences of rare species for defining aquatic conservation priorities. The data from such additional sampling points, however, should be excluded from standard analyses of spatial and temporal variability if the additional points are not always performed.

Species abundance is a key variable in ecology and is used in most fish biotic indices (e.g. see Oberdorff *et al.* 2002; Pont *et al.* 2006; and Roset *et al.* 2007 for a review). Measurement of species abundance, however, is complicated in large rivers. While the area sampled per point was evaluated under a range of river conditions, several uncontrollable variables influenced the attraction zone around the anode, including species fished, fish size, temperature and fish orientation with respect to the anode (Regis *et al.* 1981; Zalewski & Cowx 1990; Scholten 2003). Also, the systematic point sampling is only applied in zones where electric fishing is most efficient (i.e. depth <1 m and water velocity <1 m s<sup>-1</sup>), and deep or fast-flowing zones, if present, are not sampled. Uncertainties still remain, therefore, when determining abundance or biomass per m<sup>2</sup> using this sampling technique. Abundance data, in particular, should be treated with caution because of the large variation in catch that can occur between sampling points.

For small- and medium-sized wadeable rivers, previous comparisons between classical depletion sampling methods and point sampling methods have shown good agreement at the assemblage (Pretty *et al.* 2003) or population (Laffaille *et al.* 2005) scale. In larger and deeper rivers, however, higher fish densities and species richness are generally observed near the shoreline (Wolter & Bischoff 2001), with fewer and larger individuals of some species found exclusively in mid-channel. In such cases, the overall impression of fish assemblage given by systematic point sampling is certainly biased, as only fishable (i.e. where electric fishing is efficient) and accessible zones are sampled (Persat & Olivier 1991; see also Fig. 1). Other sampling methods such as seine netting (see Cowx *et al.* 2001) or electrified mini-trawls in deeper waters (e.g. see Gerdeaux & Jestin 1979) can be applied in these omitted zones, but in fisheries management and environmental monitoring programmes (e.g. application of the European Water Framework Directive), the aim is to assess the ecological status of rivers based on biological indicators, such as fish, and to study changes in spatial and/or temporal trends. Precise knowledge of the entire fish assemblage may not be necessary when selecting a suitable sampling method, which should: (1) detect significant assemblage changes related to variations in environmental conditions and human disturbances and (2) ensure constant bias over space and time (Bohlin *et al.* 1990). Several studies have already employed PASE for spatial and temporal analysis of fish communities (e.g. Persat & Copp 1989; Copp &



Jurajda 1993, 1999; Copp *et al.* 2005a,b; Santoul *et al.* 2005; Daufresne & Boët 2007), and it was, therefore, assumed that systematic point sampling will also prove suitable for such studies.

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## References

- Angermeier P.L. & Smogor R.A. (1995) Estimating number of species and relative abundances in stream-fish communities: effects of sampling effort and discontinuous spatial distributions. *Canadian Journal of Fisheries and Aquatic Sciences* **52**, 936–949.
- Bady P. & Pont D. (2008) *Workpackage 3.2: Evaluation of Existing European Fish Index (M9–M12). Version 3.1*. Aix en Provence: Cemagref, available on [http://efi-plus.boku.ac.at/downloads/EFI+-%200044096%20Deliverable%20D3\\_3.pdf](http://efi-plus.boku.ac.at/downloads/EFI+-%200044096%20Deliverable%20D3_3.pdf) 39 pp.
- Bohlin T., Heggberget T.G. & Strange C. (1990) Electric fishing for sampling and stock assessment. In: I.G. Cowx & P. Lamarque (eds) *Fishing with Electricity*. Oxford: Blackwell Scientific Publications, pp. 112–139.
- Brosse S., Laffaille P., Gabas S. & Lek S. (2001) Is scuba sampling a relevant method to study fish microhabitat in lakes? Examples and comparisons for three European species. *Ecology of Freshwater Fish* **10**, 138–146.
- Brousseau C.M., Randall R.G. & Clark M.G. (2005) *Protocol for Boat Electrofishing in Nearshore Areas of the Lower Great Lakes: Transect and Point Survey Methods for Collecting Fish and Habitat Data, 1988 to 2002*. Canadian Manuscript Report of Fisheries and Aquatic Sciences No. 2702, Fisheries and Oceans Canada, 106 pp.
- Cao Y., Larsen D.P. & Hughes R.M. (2001) Evaluating sampling sufficiency in fish assemblage surveys: a similarity-based approach. *Canadian Journal of Fisheries and Aquatic Sciences* **58**, 1782–1793.
- Capra H. (1995) *Amélioration des modèles prédictifs d'habitat de la truite fario: échelles d'échantillonnage, intégration des chroniques hydrologiques*. PhD Thesis, Villeurbanne: University of Lyon I, 268 pp.
- Casselman J.M., Penczak T., Carl L., Mann R.H.K., Holčák J. & Woitowich W.A. (1990) An evaluation of fish sampling methodologies for large river systems. *Polskie Archiwum Hydrobiologii* **37**, 521–551.
- Colwell R.K. & Coddington J.A. (1994) Estimating terrestrial biodiversity through extrapolation. *Philosophical Transactions of the Royal Society of London. Series B, Biological Sciences* **345**, 101–118.
- Colwell R.K., Mao C.X. & Chang J. (2004) Interpolating, extrapolating, and comparing incidence-based species accumulation curves. *Ecology* **85**, 2717–2727.
- Copp G.H. (1989) Electrofishing for fish larvae and 0+ juveniles: equipment modifications for increased efficiency with short fishes. *Aquaculture and Fisheries Management* **20**, 453–462.
- Copp G.H. (1997) Microhabitat use of fish larvae and juveniles in a highly regulated section of the River Great Ouse. *Regulated Rivers: Research & Management* **13**, 267–276.
- Copp G.H. (2010) Patterns of diel activity and species richness in young and small fishes of European streams: a review of 20 years of point abundance sampling by electrofishing. *Fish and Fisheries* **11**, 439–460.
- Copp G.H. & Garner P. (1995) Evaluating the microhabitat use of freshwater fish larvae and juveniles with point abundance sampling by electrofishing. *Folia Zoologica* **44**, 145–158.
- Copp G.H. & Jurajda P. (1993) Do small riverine fish move inshore at night? *Journal of Fish Biology* **43** (Suppl. A), 229–241.
- Copp G.H. & Jurajda P. (1999) Size-structured diel use of river banks by fish. *Aquatic Sciences* **61**, 75–91.
- Copp G.H., Gutí G., Rovný B. & Černý J. (1994) Hierarchical analysis of habitat use by 0+ juvenile fish in Hungarian/Slovak flood plain of the Danube River. *Environmental Biology of Fishes* **40**, 329–348.
- Copp G.H., Spathari S. & Turmel M. (2005a) Consistency of diel behaviour and interactions of stream fishes and invertebrates during summer. *River Research and Applications* **21**, 75–90.
- Copp G.H., Vranovský M., Černý J. & Kováč V. (2005b) Dynamics of young fishes and zooplankton in a lentic side-channel of the River Danube over a 24-hour cycle in late summer. *Biologia* **60**, 179–188.
- Cowx I.G., Nunn A.D. & Harvey J.P. (2001) Quantitative sampling of 0-group fish populations in large lowland rivers: point abundance sampling by electric fishing versus micromesh seine netting. *Archiv für Hydrobiologie* **151**, 369–382.
- Cucherousset J., Paillisson J.-M., Carpentier A., Eybert M.-C. & Olden J.D. (2006) Habitat use of an artificial wetland by the invasive catfish *Ameiurus melas*. *Ecology of Freshwater Fish* **15**, 589–596.
- Daufresne M. & Boët P. (2007) Climate change impacts on structure and diversity of fish communities in rivers. *Global Change Biology* **13**, 2467–2478.
- EN14011 (2003) Water Quality – Fish Sampling with Electricity. European Standard EN 14011.
- Fladung E., Scholten M. & Thiel R. (2003) Modelling the habitat preferences of preadult and adult fishes on the shoreline of the large, lowland Elbe River. *Journal of Applied Ichthyology* **19**, 303–314.

- Garner P. (1997) Sample sizes for length and density estimation of 0+ fish when using point sampling by electrofishing. *Journal of Fish Biology* **50**, 95–106.
- Gerdeaux D. & Jestin J.-M. (1979) Exemple d'application du chalut électrifié dans un milieu tempéré très minéralisé. *Annales de Limnologie* **14**, 281–287.
- Janáč M. & Jurajda P. (2005) Inter-calibration of three electric fishing techniques to estimate 0+ juvenile fish densities on sandy river beaches. *Fisheries Management and Ecology* **12**, 161–167.
- Kestemont P. & Goffaux D. (2002) *Metric Selection and Sampling Procedures for FAME (D 4 – 6)*. Final Report. Development, Evaluation & Implementation of a Standardised Fish-based Assessment Method for the Ecological Status of European Rivers – A Contribution to the Water Framework Directive (FAME), 88 p.
- Laffaille P., Briand C., Fatin D., Lafage D. & Lasne E. (2005) Point sampling the abundance of European eel (*Anguilla anguilla*) in freshwater areas. *Archiv für Hydrobiologie* **162**, 91–98.
- Lapointe N.W.R., Corkum L.D. & Mandrak N.E. (2006) Point sampling by boat electrofishing: a test of the effort required to assess fish communities. *North American Journal of Fisheries Management* **26**, 793–799.
- Nelva A., Persat H. & Chessel D. (1979) Une nouvelle méthode d'étude des peuplements ichthyologiques dans les grands cours d'eau par échantillonnage ponctuelle d'abondance. *Comptes Rendu de l'Académie des Sciences* **289**, 1295–1298 (In French).
- Oberdorff T., Pont D., Hugueny B. & Porcher J.P. (2002) Development and validation of a fish-based index for the assessment of 'river health' in France. *Freshwater Biology* **47**, 1720–1734.
- Oksanen J., Kindt R., Legendre P., O'Hara B., Simpson G.L., Solymos P. et al. (2008) *Vegan: Community Ecology Package*. R package Version 1. 15–1, 190 pp. available on <http://cran.r-project.org/>, <http://vegan.r-forge.r-project.org/>
- Perrow M.R., Jowitt A.J.D. & Zambrano González L. (1996) Sampling fish communities in shallow lowland lakes: point-sample electric fishing vs electric fishing within stop-nets. *Fisheries Management and Ecology* **3**, 303–313.
- Persat H. & Copp G.H. (1989) Electric fishing and point abundance sampling for the ichthyology of large rivers. In: I.G. Cowx (ed.) *Developments in Electrofishing*. Oxford: Blackwell Scientific Publications, pp. 197–209.
- Persat H. & Olivier J.-M. (1991) The point abundance sampling, a fishing strategy for large rivers: short presentation of the concept, its appliance and some results. In: M. Peňáz (ed.) *Biological Monitoring of Large Rivers*. Brno and Bratislava: Czechoslovak Academy of Sciences, and Slovak Academy of Sciences, pp. 104–113.
- Pires A.M., Cowx I.G. & Coelho M.M. (1999) Seasonal changes in fish community structure of intermittent streams in the middle reaches of the Guadiana basin, Portugal. *Journal of Fish Biology* **54**, 235–249.
- Pont D., Hugueny B., Beier U., Goffaux D., Melcher A., Noble R. et al. (2006) Assessing river biotic condition at a continental scale: a European approach using functional metrics and fish assemblages. *Journal of Applied Ecology* **43**, 70–80.
- Pouilly M. (1994) *Relations entre l'habitat physique et les poissons des zones à cyprinidés rhéophiles dans trois cours d'eau du bassin rhodanien: vers une simulation de la capacité d'accueil pour les peuplements*. PhD Thesis, Villeurbanne: University of Lyon I, 256 pp.
- Pretty J.L., Harrison S.S.C., Shepherd D.J., Smith C., Hildrew A.G. & Hey R.D. (2003) River rehabilitation and fish populations: assessing the benefit of instream structures. *Journal of Applied Ecology* **40**, 251–265.
- R Development Core Team (2011) *R: A Language and Environment for Statistical Computing*. Vienna: R Foundation for Statistical Computing, ISBN 3-900051-07-0, URL <http://www.R-project.org/>
- Regis J., Pattee E. & Lebreton J.D. (1981) A new method for evaluating the efficiency of electric fishing. *Archiv für Hydrobiologie* **93**, 68–82.
- Roset N., Grenouillet G., Goffaux D., Pont D. & Kestemont P. (2007) A review of existing fish assemblage indicators and methodologies. *Fisheries Management and Ecology* **14**, 393–405.
- Santoul F., Cayrou J., Mastrorillo S. & Céréghino R. (2005) Spatial patterns of the biological traits of freshwater fish communities in south-west France. *Journal of Fish Biology* **66**, 301–314.
- Scholten M. (2003) Efficiency of point abundance sampling by electro-fishing modified for short fishes. *Journal of Applied Ichthyology* **19**, 265–277.
- Smith K.L. & Jones M.L. (2005) Watershed-level sampling effort requirements for determining riverine fish species composition. *Canadian Journal of Fisheries and Aquatic Sciences* **62**, 1580–1588.
- Ugland K.I., Gray J.S. & Ellingsen K.E. (2003) The species–accumulation curve and estimation of species richness. *Journal of Animal Ecology* **72**, 888–897.
- Valová Z., Jurajda P. & Janáč M. (2006) Spatial distribution of 0+ juvenile fish in differently modified lowland rivers. *Folia Zoologica* **55**, 293–308.
- Wolter C. & Bischoff A. (2001) Seasonal changes of fish diversity in the main channel of the large lowland River Oder. *Regulated Rivers: Research & Management* **17**, 595–608.
- Zalewski M. & Cowx I.G. (1990) Factor affecting the efficiency of electric fishing. In: I.G. Cowx & P. Lamarque (eds) *Fishing with Electricity*. Oxford: Blackwell Scientific Publications, pp. 89–111.

**Supporting Information**

Additional Supporting Information may be found in the online version of this article:

**Appendix 1.** Species accumulation curves for each study site following Ugland *et al.* (2003).

**Table S1.** Voltage gradient measurements between two points 10 cm apart at different distances from the centre of the anode (35 cm diameter) at different sampling sites.