

*Letter to Editor*

# **Use of Artificial Substrates for Sampling Benthic Macroinvertebrates in the Assessment of Water Quality of Large Lowland Rivers**

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## **Abstract**

The objective of this investigation was to evaluate the usefulness of the artificial substrate sampler in collecting macroinvertebrates for water quality assessment of Polish lowland rivers. This paper presents the results of a comparative study between two different sampling techniques, i.e. nettings filled with brick as artificial substrates and handnet sampling. The validity of applying the biotic index method is also demonstrated. The Belgian Biotic Index (BBI) method and the lower Nysa Kłodzka river were chosen for study. Macroinvertebrates were collected seasonally at five sampling sites. Although some invertebrate taxa revealed a specific preference for one of the two tested sampling substrates, the artificial substrates and handnet samples had a similar fauna composition. BBI scores were rather insensitive to the different sampling methods used during the study. Taking into account all sampling seasons, 60% of the BBI values for the two sampling techniques were the same and nearly 27% scored one unit lower or higher. This has led to the conclusion that water quality changes of the river ecosystem in Poland can be demonstrated by means of the analyses of macroinvertebrates colonizing artificial substrates.

**Keywords:** artificial substrate sampler, macroinvertebrates, biotic index, river water quality

## **Introduction**

During the 1970s, a series of intercalibration studies and seminars in the field of biological water quality assessment were organized by the European Communities to make comparisons between different assessment methods. From these exercises, which took place in Germany, the UK and Italy, it clearly appeared that the most practical methods for river water quality assessment are those based on benthic macroinvertebrates [1]. The benthic macroinvertebrate fauna may offer good opportunities to study river water quality as this group is confined for the most part to one locality of the river bed, is relatively easily sampled and manipulated and is relatively quick to react to changes. The response of many common invertebrate species to different types of pollution has been

established [2]. In most European countries and also elsewhere this has led to the development of several biotic indices [3].

One of the major problems, however, is locating and sampling ecologically comparable habitats both exposed and not exposed to the variable one wishes to study (e.g. pollutional stress). This entails finding areas with comparable physico-chemical characteristics, sampling ability and proximity of sites to each other to provide valid comparative data of the similarities and differences. One of the most common problems encountered is selective natural substrate colonization by benthic invertebrates [4, 5]. According to the authors the highest densities of macroinvertebrates are usually associated with microhabitats such as large woody debris and macrophytes, both of which are difficult and time-consuming to sample quantitatively. Additional obstacles to adequate sampling may include stream flow, submerged obstacles, large unsamp-

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lable substrates and vertical distribution of invertebrates within the substrate [6, 7, 8].

A solution to these inherent problems has been the use of artificial substrate sampling devices. Moreover, in cases where conventional sampling techniques for collecting macroinvertebrates (handnet samplers, dredging nets, grabs and air-lift samplers, among others) cannot be applied, colonization samplers could provide a valid alternative. A variety of different types of artificial substrate samplers have been developed and employed in collecting macroinvertebrates which inhabit the benthic areas of both lentic and lotic series. Their designs have included such varied devices as simple concrete slabs or blocks, multiple-plate hardboards, and rock or limestone-filled samplers [9, 10]. Although a variety of methods have been used to collect benthic organisms from lowland rivers, it has been shown that water quality changes of a river ecosystem can be demonstrated by means of the analysis of macroinvertebrates colonizing artificial substrates [11]. Each of these samplers has the advantages of being relatively inexpensive, simple in design, easily used and habitat-adaptable (i.e. they can be used in either streams, deep rivers and lakes). Besides, they all have known substrate characteristics. Because of these characteristics, the sampling effort can be better standardized. For these reasons, since the 1980s more attention in Europe has been paid to the development of standardized macroinvertebrate colonization samplers, as different procedures lead to a considerable variability in water quality assessment [12].

The present day method of determining river water quality in Poland is based mainly on the physical and chemical characteristics of water. Since 1999, an attempt has been made at the elaboration of the biological method, in relation with European Union requirements, for assessing the quality of running water. Results from an investigation, which was carried out all over Poland on 49 rivers and streams [13], clearly shows that the application of a uniform methodology was impossible. In some deep rivers, the use of other field samplers rather than the routine Surber and Eckman-Birge grab was necessary, for example a dragnet. Whereas, consistency of methods when collecting macroinvertebrates in lowland rivers is important for assessment of river conditions and ongoing monitoring.

The investigation reported in this paper was carried out to :

- evaluate the use of the colonisation sampler in the biological surveillance of water quality in deep lowland Polish rivers;
- evaluate the application of the biotic index method in processing the data.

For study purposes, the Nysa Kłodzka river and the Belgian Biotic Index method (one of the widely used indices) were selected. The investigation focused on the lower course of the river, difficult to sample due to its physical features (large, deep, with a locally steep bank and fast current).

## Materials and Methods

### Description of the Study Area

The Nysa Kłodzka river, which is 182 km long and drains an area of 4565 km<sup>2</sup> in south west Poland, has its source at the Puchacz hillside in the Śnieżne Mountains (975m a.s.l.) and the river – confluence near the town of Lewin Brzeski (140 m a.s.l.).

The study was carried out in the lower Nysa Kłodzka river, within the area of Opole voivodeship. Five monitored stations were localized along the river stretch of 80 km length, from the Nysa retention reservoir to the river-mouth. The river gradient of the lower course is relatively constant, falling at a rate of 77 m in 100 km. The river bed consists mainly of gravel and sand (respectively 2-20 mm and 0.2-2 mm in diameter) with boulders (20 mm) or silt (0.2 mm) additions. Aquatic vegetation is scarce and dominated by carexes *Carex* sp. Steep riverbanks, artificial in part, are mostly overgrown with grass and willow *Salix* sp. The depths at the investigation stations varied between 1 and 2 m. The main form of pollution in the lower Nysa Kłodzka river are nitrogen and phosphorus compounds received from an agricultural catchment area and treated sewage effluents from Nysa, a town of 50,000 inhabitants.

Two sampling methods were used to sample each station on the river on three occasions, in the spring, summer and autumn of 2000.

The artificial substrate sampler consisted of a polyethylene netting (a potato bag of 6 dm<sup>3</sup> volume and the mesh size of ca. 10 mm), filled with pieces of brick (5 – 10 cm diameter). Each season a total of 15 netting samplers was used to sample macroinvertebrate population at sites located near the following towns: Nysa, Piątkowice, Malerzowice Wlk., Głęboko, and Lewin Brzeski. The samplers (three replicates at each site) were placed along the bank of the watercourse and attached with a nylon rope to tree trunks, roots of trees and branches suspended over the water. The whole set of artificial substrates was installed as inconspicuously as possible. All samplers were left in the river water for 30 ± 3 days to allow sufficient time for colonization. Afterwards, the samplers were removed individually with special care being taken to minimize losses of organisms as a result of water leakage. After lifting from the water surface the substrates were immediately transferred into lidded buckets and transported to the laboratory. The nettings and pieces of brick were then sieved through a graded series of sieves (with mesh sizes of 10 mm, 1 mm and 500 µm) to facilitate sorting of live material. From the taxonomic groups which were present in large numbers, a subsample of 100 individuals was taken. Once separated, the macroinvertebrates were preserved in 70% ethanol for further identification and enumeration.

On the same day the artificial substrates were removed, an active sampling was done with a handnet sampler, which consisted of a metal frame (30 x 30 cm) and a conical bag of 50 cm length with the mesh size of 300

µm. The bottom net samples consisted of four one minute kick samples. Once obtained, each sample was processed as previously described for the artificial substrates.

For data processing all artificial substrates (AS) and kick samples (H) were first considered as separate units/subsamples and then as pooled samples of the three (AS) and four (H) replicates together. This was done to avoid the possibility of poor sampling effort impact on biotic index values. The artificial substrates and handnet samples were analyzed qualitatively and quantitatively. Water quality was expressed by means of the Belgian Biotic Index, based on the analysis of macroinvertebrates identified up to a practical level (genus or family). These levels are called "systematic units". The biotic index is a numerical value between 10 (excellent) and 0 (bad water quality), derived from the number of systematic units and the presence of indicator groups [14].

Computations were carried out using the STATISTICA 5.0 PI statistical package.

## Results

The qualitative and quantitative analysis of the bottom fauna showed that the macroinvertebrate community of the Nysa Kłodzka river was characterized by high biodiversity. The total amount of identified systematic units was 57. In spite of the fact that artificial substrates are clearly selective for certain species, the total number of taxa on artificial substrates and natural substrates sampled with the handnet (kick samples) was nearly the same, respectively 49 and 45 taxa collected during the study (Table 1). Exceptions were the following groups of macroinvertebrates which were not present on the artificial sampler: *Heteroptera* and *Lepidoptera*, as well as *Hemiclepsis* (*Hirudinea*), *Gomphus* (*Odonata*), *Lacophilus* (*Coleoptera*), and *Lepidostomatidae* (*Trichoptera*). Certain *Mollusca* (like *Planorbis*, *Ancylus*, *Valvata* and *Dreissena*), *Trichoptera* (*Psychomeidae*) and *Diptera* (*Sciomyzidae* and *Ceratopogonidae*) on the contrary were not present in the handnet samples.

From the qualitative analysis it could be concluded that most taxa have no distinct preference for a particular type of substrate, i.e. natural or artificial. For some collected taxa, most active swimmers, a lower numerical presence on artificial substrates was observed. On the other hand, taxa with a positive thigmotaxis were more numerous on artificial samplers. It is clear that leeches preferred the artificial substrate. *Chironomidae* (*Diptera*) also followed this trend and were found in much larger numbers on artificial substrates than in the handnet samples.

The data also showed that all methods provided sufficient biological information to allow reliable water quality assessment based on the BBI method. This was true for all habitats under study. Irrespective of sampling season, the differences in numbers of taxa identified in particular samples (artificial substrates AS via handnet samplers H) ranged between 0-3 taxa. When all sampling seasons were taken together, the regression analysis clearly dem-

Table 1. Macroinvertebrate taxa presented on artificial substrates in comparison with handnet samples.

Taxon	Handnet	Artificial substrate
<i>Turbellaria</i>	+	+
<i>Planaria</i>		
<i>Oligochaeta</i>	+	+
<i>Lumbriculidae</i>		
<i>Tubificidae</i>	+	+
<i>Hirudinea</i>		
<i>Erpobdella</i>	+	+
<i>Glossiphonia</i>	+	+
<i>Helobdella</i>	+	+
<i>Hemiclepsis</i>	+	-
<i>Mollusca</i>		
<i>Planorbis</i>	-	+
<i>Valvata</i>	-	+
<i>Ancylus</i>	-	+
<i>Bithynia</i>	+	+
<i>Lymnaea</i>	+	+
<i>Anodonta</i>	+	-
<i>Pisidium</i>	+	+
<i>Sphaerium</i>	+	+
<i>Dreissena</i>	-	+
<i>Crustacea</i>		
<i>Asellus</i>	+	+
<i>Oronectes</i>	+	+
<i>Ephemeroptera</i>		
<i>Heptagenia</i>	+	+
<i>Paraleptophlebia</i>	+	+
<i>Caenis</i>	+	+
<i>Baetis</i>	+	+
<i>Seratiella</i>	+	+
<i>Behnigia</i>	-	+
<i>Odonata</i>		
<i>Platycnemis</i>	+	+
<i>Ischnura</i>	+	+
<i>Calopteryx</i>	+	+
<i>Platetrium</i>	+	+
<i>Gomphus</i>	+	-
<i>Ophiogomphus</i>	+	+
<i>Trichoptera</i>		
<i>Polycentropodidae</i>	+	+
<i>Hydropsychidae</i>	+	+
<i>Hydroptilidae</i>	+	+
<i>Limnephilidae</i>	+	+
<i>Leptoceridae</i>	+	+
<i>Lepidostomatidae</i>	+	-
<i>Psychomeidae</i>	-	+

Table 1 continues on next page...

Taxon	Handnet	Artificial substrate
<i>Megaloptera</i> <i>Sialis</i>	+	+
<i>Coleoptera</i> <i>Agabus</i>	+	+
<i>Platambus</i>	+	+
<i>Laccophilus</i>	+	-
<i>Orectophilus</i>	+	+
<i>Haliplus</i>	+	+
<i>Heteroptera</i> <i>Micronecta</i>	+	-
<i>Nepa</i>	+	-
<i>Sigara</i>	+	-
<i>Mesovelia</i>	+	-
<i>Gerris</i>	+	-
<i>Lepidoptera</i> <i>Nymphula</i>	+	-
<i>Parpomyx</i>	+	-
<i>Diptera</i> <i>Chironomidea</i>	+	+
<i>Tipulidae</i>	+	+
<i>Simuliidae</i>	+	+
<i>Limoniidae</i>	+	+
<i>Ceratopogonidae</i>	-	+
<i>Sciomyzidae</i>	-	+
<i>Hydrachnella</i>	+	+

onstrated that the number of taxa collected by means of the artificial substrate sampler was closely approximating that of the handnet, with a correlation of 0.7791 ( $p=0.001$ ) (Fig. 1).

Of all the sampling seasons, 60% of the BBI values for the two sampling techniques were the same, nearly 27% scored one unit lower or higher and about 13% differed + or - two units. This similarity was confirmed by the regression lines and the high correlation coefficient of 0.8981 ( $p=0.001$ ), when comparing index values obtained from the analysis of macroinvertebrates collected by means of the artificial substrate samplers and the handnet (Fig. 2). At the same time, a good agreement between these sampling methods has been demonstrated by the variable coefficient calculated for biotic index values. BBI index values did not appear to be significantly influenced by the sampling technique, which gave almost the same mean scores of 7.13 and 7.26 for the handnet and the artificial substrates, respectively. However, the artificial substrates yielded a slightly lower value of the variable coefficient (16.8 for a standard deviation of 1.22) than the handnet (19.0 for a standard deviation of 1.35).

The results of the final classification of water quality assessment were convergent. River water quality derived from BBI values were mainly the same (80%) or differed

with only one value/class from each other. For the biotic index and water quality classification, the sampling procedure by means of the artificial substrate usually provided a higher score.

## Discussion and Conclusions

According to numerous publications [15, 16, 17] artificial substrates have proven their usefulness in river ecosystem assessment. They could be used under different climatic conditions and in different types of watercourses, including streams, slow lowland brooks and rivers as well as canals. In a small space, the samplers provide a great variation of microhabitats allowing colonization by numerous taxa representative for the natural fauna of the watercourse. It has been demonstrated, by comparison of diversity indices between various artificial samplers and conventional sampling methods, that bottom samplers for the collection of benthic macroinvertebrates are more reliable than floating ones. The floating substrates tend to be selectively colonized by beetles, mayflies, and caddisflies. Humphries et al. [18] found that snags and basket-type artificial substrate samplers seemed to collect a greater diversity of aquatic invertebrates than dredging and air-lift sampling. Basket-type samplers collect a selective community (mostly oligochaetes and chironomids), but as shown by Watton and Hawkes [19], the groups collected are the ones which are generally considered important as water quality indicators. The results of the Nysa Kłodzka river investigations reveal similar differences, as described by the authors, in colonization preferences and structural diversity of the benthic community collected by brick-bags and handnet sampling. Some of them were no doubt due to real differences in the macroinvertebrate distribution connected with microhabitat preferences. Larvae of dragonflies *Gomphus*, living in sandy and muddy substrata, were absent in the artificial substrate samples, for instance. However, some variations were probably caused by different sampling efficiency of the methods used during the study. It was possible to sample a wider range of habitats by means of the handnet

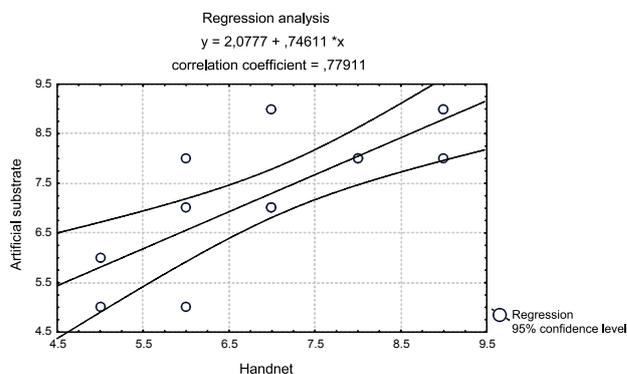


Fig. 1. Regression analysis of number of taxa for artificial substrates versus handnet samples.

in comparison with the artificial substrates. Thus, hand-net samples included macroinvertebrates collected from different types of the river bed (from stony to muddy), overgrown with aquatic vegetation in various degrees. It is also important to know that qualitative and quantitative fluctuations with time can be caused by such factors as the colonization of the artificial substrates by periphyton (food for many invertebrates), the accumulation of silt and organic matter on the substrate surface, spatial competition, predations, and the natural life history of the organisms [20, 21, 22]. De Pauw et al. [23] also found that the nature of substrates was not a crucial factor for macroinvertebrate colonization. According to these authors, of all materials tested (like plastic tubes, brushes or mats) brick is the most convenient and available worldwide for the purpose of routine biomonitoring.

It appears from this study that the Belgian Biotic Index (BBI) could be applied without any problems in biological water quality assessment. No practical difficulties were encountered with the calculation of the index since all taxa found and indicator groups could be taken into consideration. Other studies have also documented a positive relation between BBI index values and values of the common chemical variables [24]. This confirms the observations by De Pauw and Hawkes [25] on the BBI method application. Regarding the effect of the sampling method on biotic index scores, the BBI proved to be insensitive to the use of two different sampling techniques. In spite of the specific preference of some invertebrate taxa for one of the two tested sampling substrates, the value of water quality derived from BBI were mainly the same or differed with only one unit lower or higher from each other. Data from the present study indicates that water quality of the lower Nysa Kłodzka river can be ranked to the III class at the Nysa sampling site and the II class at the others. However, the final water quality classification at the Malerzowice Wlk. site, according to the BBI values obtained for the artificial substrates, was higher (I class) in comparison with the results for the handnet sampling. A good correlation between BBI values obtained for the artificial substrates and the handnet sampling has

been corroborated by the results of statistical analysis. Four other biotic indices (BMWP, ASPT, EBI, IBE) and artificial sampling methods have been successfully used in assessing river water quality in Italy [26]. The authors reported that there were only small differences among the biotic index scores stemming from the different artificial substrates. Although a common criticism is that these indices show seasonal dependence, Zamora-Munoz et al. [27] found that the variability of BMWP and ASPT was caused more by pollution than by seasonality. This suggests the need to further investigate the biotic indices and artificial substrate usage in biomonitoring studies in Poland.

In summary, the results of the study indicate that water quality changes of the river ecosystem can be demonstrated by means of the analysis of macroinvertebrates colonizing artificial substrates. Artificial substrates have many advantages over conventional sampling techniques for collecting macroinvertebrates because they are inexpensive to construct, simple to use, and seem to collect predominantly macroinvertebrates which are associated with the bottom substrate of the river. Certainly, the main advantage of this technique application in routine monitoring of river water quality is the fact that sampling efforts can be better standardized, as artificial substrates offer the same habitats for colonization by organisms at all sampling sites. Of course, artificial substrate samplers have limitations (the unforeseen losses which often occur, the necessity to visit the site twice, and the long period needed to obtain representative samples, among others) but may be very good tools for surveying water pollution if the drawbacks of using them are understood.

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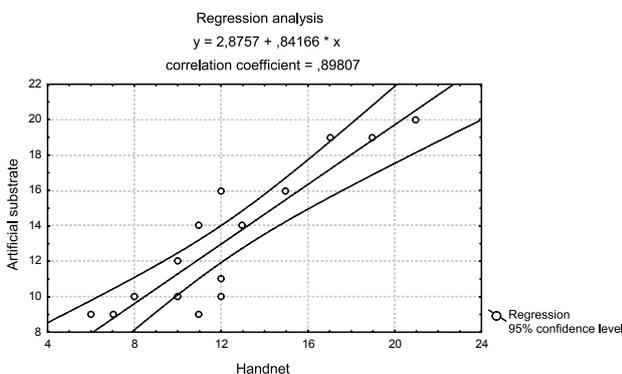


Fig. 2. Regression analysis of BBI values for artificial substrates versus handnet samples.

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