

AN APPRAISAL OF POND-NET SAMPLES FOR BIOLOGICAL MONITORING OF LOTIC MACRO-INVERTEBRATES

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Abstract—The use of a pond-net to sample benthic macro-invertebrates is assessed as a technique for use in a national classification scheme for running water sites. In a field trial, 6 pond-net samples of 3 min duration were taken at each of 4 sites on the River Axe in south-west England. Three different people each took 2 samples at each of the sites. A comparison of the number of families and species caught indicated significant differences with respect to sites, operators and site × operator interactions. Significant inter-operator differences in the number of taxa removed from samples were also shown at the sample processing stage, but only at family level. Despite these differences, clustering and ordination procedures showed strong site faithfulness for each series of 6 samples when data were analysed at species level. Similar analyses at family level produced several misclassifications of samples with those from other sites. Most of these misclassifications were eliminated when categories of abundance were applied to the family level data. A 3 min sample collected approx. 62% of families and 50% of species that could be attained at a site by 18 min netting. The results of this field trial provided justification for the use of a pond-net as the principal sampling technique in the classification exercise.

INTRODUCTION

Biological monitoring, particularly as practised within Water Authorities and River Purification Boards, is highly dependent upon the use of a pond-net to obtain samples. A project* currently being undertaken by the authors utilizes samples taken by Water Authority biologists to provide a classification of lotic sites in Great Britain by means of their benthic macro-invertebrate communities. Consequently, most of the information will be obtained from pond-net collections. This will inevitably involve many different biologists taking samples in a wide variety of rivers throughout England, Wales and Scotland, in which standardization of the pond-net technique cannot be achieved.

Procedures for pond-net sampling have been described by Hynes (1961), Morgan & Egglisshaw (1965), Minshall & Minshall (1966), Frost *et al.* (1971), Armitage *et al.* (1974) and many others. Little attention has been paid however to the precision or reproducibility of such samples as a representation of the community present at a site at the time of sampling. Morgan & Egglisshaw (1965) and Armitage *et al.* (1974) provide justifications for the use of pond-nets in their surveys, whilst Frost *et al.* (1971) evaluate several aspects of this form of sampling for benthic invertebrates. In each of these exercises netting techniques are standardized whereas in the River Communities Project

comparable standardization is not feasible. Therefore before starting the River Communities Project some aspects of the pond-net sample as a descriptor of a benthic macro-invertebrate community were investigated. Studies described in this paper were used to validate the sampling programme adopted for the national classification project.

DESCRIPTION OF SURVEY RIVER AND SITES

The River Axe, in south-west England, was selected for this pilot study since it was conveniently situated near the Freshwater Biological Association (FBA) River Laboratory and was also included in the RCP programme. The river, which is approx. 35 km long, rises near Cheddington (NGR ST 493 503), at an approx. altitude of 190 m before flowing first west and then south to enter the sea at Seaton (SY 256 898).

The 4 study sites selected (Fig. 1) were at Mosterton (ST 457 053); Oathill Farm (ST 402 060); Broom (ST 326 025); and Whitford Bridge (SY 262 953) at 4, 9, 20 and 30 km respectively from the source. Physical features of each site (Table 1) were recorded on standard forms. The most abundant macrophytes at each site were also noted and combined percentage cover estimated (Table 1). Details of water chemistry, as provided by South West Water Authority, are given in Table 2.

Mosterton and Whitford Bridge were the most homogeneous sites with relatively uniform substrata, few macrophytes and little habitat diversity. Broom differed in having extensive growths of several macro-

* The Analysis of Natural River Communities in Great Britain Project—subsequently referred to as River Communities Project or RCP.

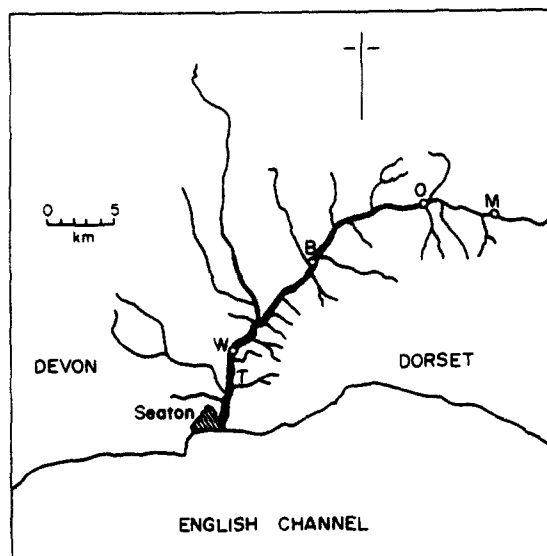


Fig. 1. Map of the River Axe showing sampling sites and tidal limit (T); M = Mosterton; O = Oathill Farm; B = Broom; W = Whitford Bridge.

phytes but the composition of the stream bed appeared to be relatively uniform. The remaining site, Oathill Farm, was the most heterogeneous with diverse substratum and flow conditions ranging from fast riffles to pools, and with macrophytes also present.

METHODS

Three experienced operators (authors MF, JW, PA), each took two pond-net samples at each site. Although pond-net sampling has been frequently described, implementation is very susceptible to individual interpretation. This proved to be the case in this study and the method was applied with considerable variability by operators who had developed their techniques in widely differing streams. A particular source of variation was the extent to which the net was used in a sweeping motion, as opposed to being held static whilst the substratum was disturbed. No attempt was made to standardize the netting techniques of the three operators as this would fail to reflect the variability to be encountered in the full RCP study.

Efforts at standardization were confined to the equipment used, the duration of netting and the sampling aims. A standard FBA pond net, (900 μm mesh, 230 \times 255 mm frame, 275 mm bag depth), fitted to a 1.5 m handle, was used throughout the survey. Each sample was of 3 min duration as this was the period most commonly used by Water Authority biologists. Each operator attempted to sample all available habitats at each site in proportion to their occurrence, using whatever technique seemed appropriate to the prevailing conditions. No precise definition of the collecting area was made prior to sampling, thus allowing the constraints of time and habitat diversity to operate freely.

At the end of each sampling period each operator transferred the net-bag contents into heavy duty polythene bags containing formalin. There were considerable differences between operators in the degree of on-site elutriation prior to transfer of the catch to polythene bags.

All samples were taken between 10.00 and 16.00 h on 11 April 1978. Mosterton was the first site to be sampled followed by successive downstream sites. The sampling order of the three operators varied at each site.

Table 1. Features of the study sites at the time of sampling (11-04-78)

Parameter	Mosterton	Oathill Farm	Broom	Whitford Bridge
Altitude (m)	88	82	46	9
Mean channel width (m)	2.5	7.0	8.0	25.0
Midstream water depth (m)	0.10	0.25	0.43	0.65
Surface velocity (cm s^{-1}) as estimated by timed float	51-75	76-100	> 100	76-100
Dominant substratum particle size	Pebbles	Pebbles	Pebbles	Pebbles
Percentage macrophyte cover	16-64 mm 3	16-64 mm 5	16-64 mm 40	16-64 mm 0

Table 2. Mean annual water chemistry (Jan.-Dec. 1978) at the four study sites. All values based on 8-10 samples

Parameter	Mosterton	Oathill Farm	Broom	Whitford Bridge
pH	7.9	7.8	7.9	8.1
Conductivity (micro-siemens, 20°C)	455	434	426	374
Dissolved oxygen (% saturation)	91.0	91.3	98.2	107.3
Dissolved oxygen ($\text{mg O}_2 \text{l}^{-1}$)	10.3	10.2	11.0	11.8
Total BOD ₅ (ATU) ($\text{mg O}_2 \text{l}^{-1}$)	3.25	3.15	2.47	2.09
Nitrogen (total oxidized) (mg l^{-1})	2.84	3.92	4.54	3.23
Phosphate as P (mg l^{-1})	0.14	0.22	0.29	0.21
Chloride (mg l^{-1})	21.1	25.6	25.0	23.7
Calcium (mg l^{-1})	95.7	76.1	70.8	61.6

Five people including the 3 field operators were involved in sample sorting and identification. Cross-checks on identifications were made to ensure accuracy and consistency between these 5 processors. Individual samples were sorted for approx. 2 h, although this varied with the precise nature of the material each contained. Animals were picked by eye from flat-bottomed trays. For greater efficiency samples were divided into manageable aliquots.

Data of two types were derived from the samples. Animals were first identified to family level and estimates of abundance were made for each family. All animals in a fixed proportion of each sample were either removed, or, if readily identifiable, counted. The normal proportion examined was either 25, 50 or 100%, depending on sample size, and an appropriate multiplication factor applied in order to estimate numbers in the whole sample. For this purpose the sorting trays were divided into 4 equal sections and each aliquot spread as evenly as possible over the tray. These methods were considered adequate for the crude level of quantification involved. Five categories of abundance were recognised according to an approximate logarithmic scale.

Estimated numbers	1-9	10-99	100-999	1000-9999	≥10,000
Category	1	2	3	4	5

These data could also be used qualitatively.

The second approach was to obtain as extensive a species list as possible. In this case any unexamined proportion of the sample was scanned for additional species. Where this involved gains at family level these were added to the family data set without applying a multiplication

factor. Species level data were strictly qualitative and all identifications were taken as far as possible with the taxonomic keys available (Armitage *et al.* 1979). Most groups could be identified to species level, but many Diptera could not. In other instances species which could not easily be distinguished from each other were placed in artificial taxa to separate them from other members of the genus [e.g. *Polycelis nigra* group included *P. nigra* (Müller) and *P. tenuis* (Ijima)]. Taxonomic precision can be assessed by reference to Appendix I.

Details of mathematical and statistical techniques applied will be discussed, as encountered, in subsequent sections.

RESULTS

Quantitative differences

The first consideration of this study was whether the variation in sampling techniques adopted by different field operators would provide a series of samples which were adequate to distinguish between the benthic macro-invertebrate communities of the 4 sites.

Numbers of taxa per sample are given in Tables 3 and 4. Examination of the mean number of taxa per sample caught by each person suggested that inter-operator differences could be of major importance. The mean number of taxa caught by each operator varied from 31.9 to 37.5 families per sample and from

Table 3. (a) Number of families per sample; (b) Number of families per sample as a proportion of the total number of families captured at the site. Subscripts given to 3(a) indicate field sampling order and also apply to 3(b)

	Site	Operator						Site means	Number of families caught
		1		2		3			
(a)	Mosterton	28 ₃	32 ₄	31 ₁	24 ₂	27 ₅	33 ₆	29.2	49
	Oathill Farm	33 ₂	25 ₃	46 ₅	50 ₆	41 ₁	39 ₄	39.0	60
	Broom	43 ₁	34 ₃	38 ₂	45 ₄	34 ₅	32 ₆	37.7	61
	Whitford Bridge	32 ₅	28 ₆	33 ₂	33 ₄	33 ₁	30 ₃	31.5	50
	Operator means	31.9		37.5		33.6			
(b)	Mosterton	57.1	65.3	63.3	49.0	55.1	67.3	59.5	
	Oathill Farm	55.0	41.7	76.7	83.3	68.3	65.0	65.0	
	Broom	70.5	55.7	62.3	73.8	55.7	52.4	61.7	
	Whitford Bridge	64.0	56.0	66.0	66.0	66.0	60.0	63.0	

Table 4. (a) Number of species per sample; (b) Number of species per sample as a proportion of the total number of species captured at the site. Field sampling order is the same as in Table 3(a)

	Site	Operator						Site means	Number of species caught
		1		2		3			
(a)	Mosterton	43	47	45	37	34	52	43.0	93
	Oathill Farm	45	35	76	85	65	62	61.3	119
	Broom	69	59	55	70	51	44	58.0	114
	Whitford Bridge	48	40	52	46	50	47	47.2	94
	Operator means	48.3		58.3		50.6			
(b)	Mosterton	46.2	50.5	48.4	39.8	36.6	55.9	46.2	
	Oathill Farm	37.8	29.4	63.9	71.4	54.6	52.1	51.5	
	Broom	60.5	51.8	48.2	61.4	44.7	38.6	50.9	
	Whitford Bridge	51.1	42.6	55.3	48.9	53.2	50.0	50.2	

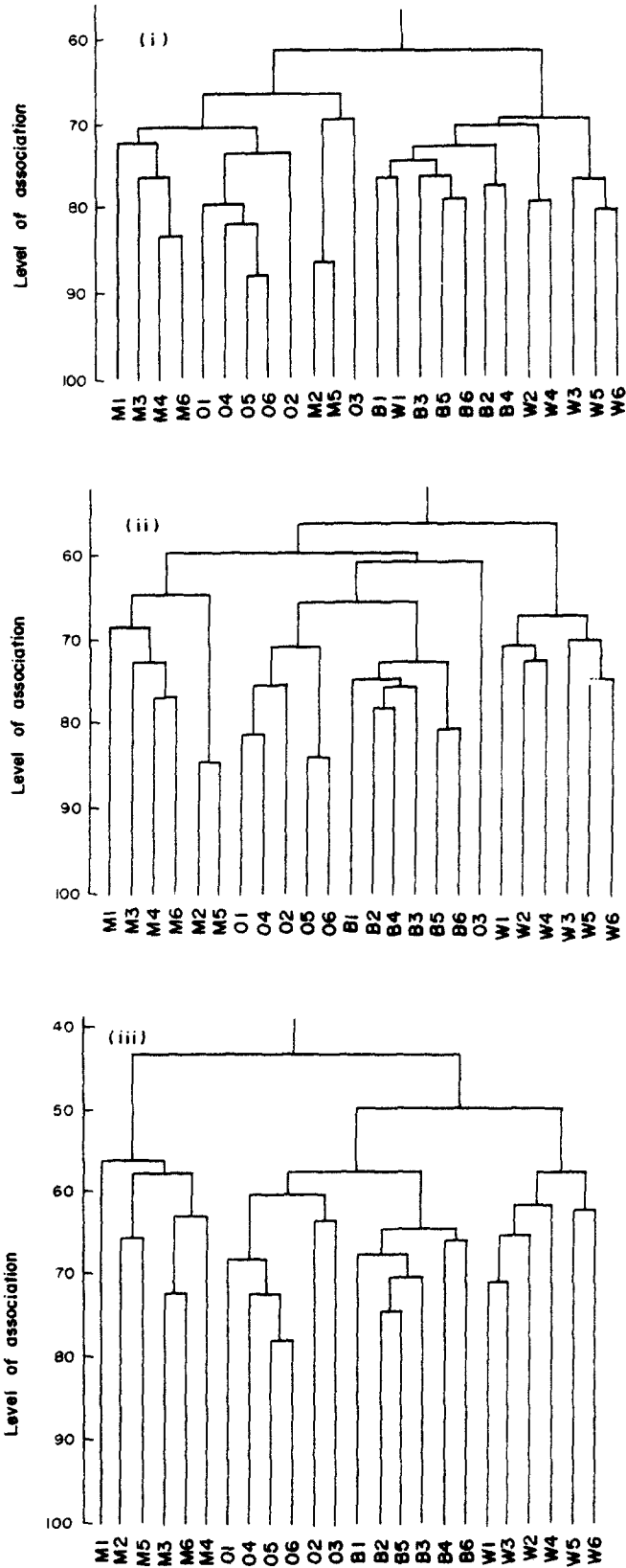


Fig. 2. Hierarchical classification of samples by average linkage clustering, using the Czekanowski similarity index. (i) Family presence/absence data; (ii) Family log categories data; (iii) Species presence/absence data; M = Mosterton; O = Oathill Farm; B = Broom; W = Whitford Bridge.

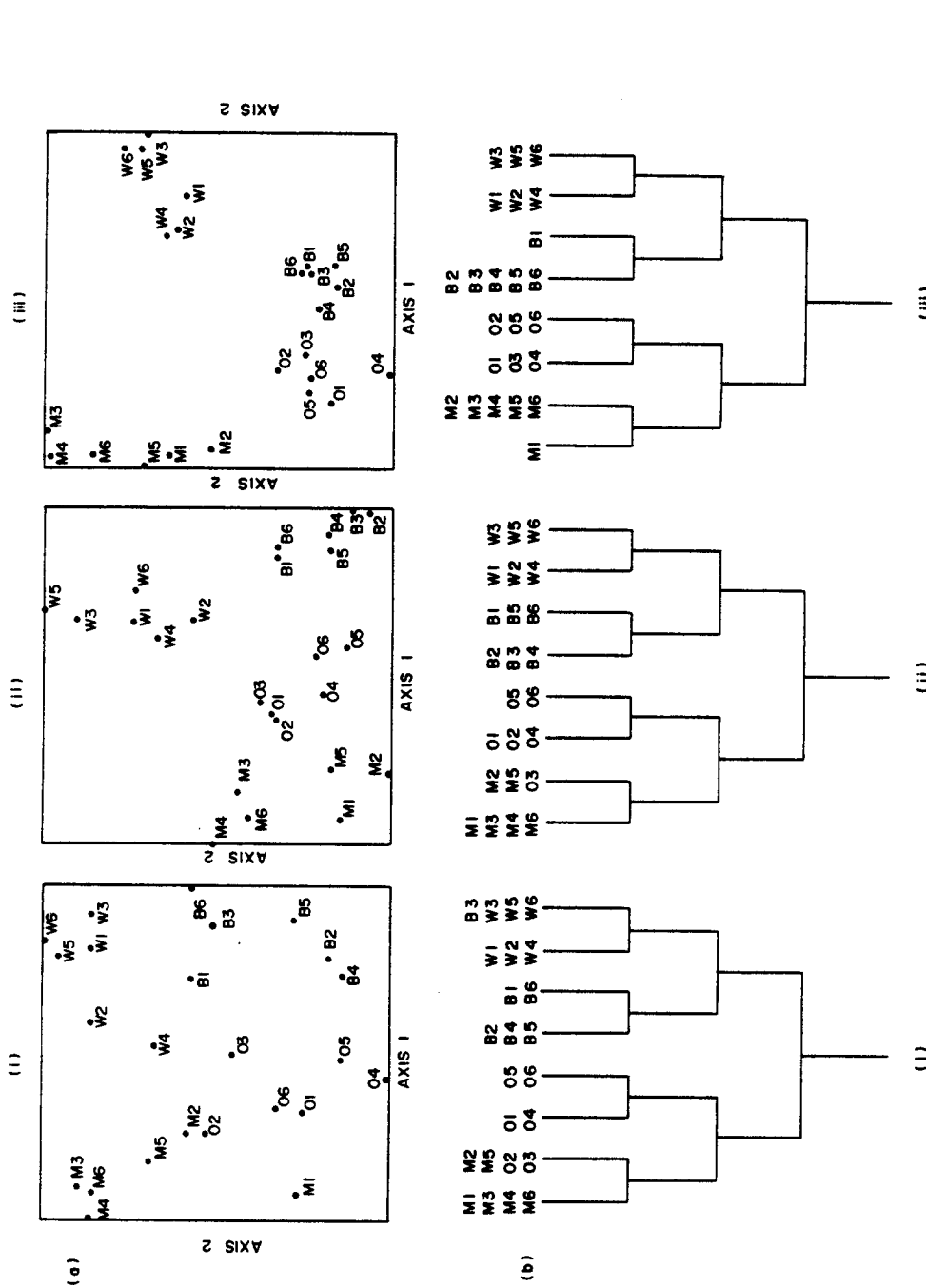


Fig. 3. (a) Reciprocal averaging ordination plots of samples. (b) Hierarchical classification of samples by indicator species analysis. (i) Family presence/absence data; (ii) Species presence/absence data; (iii) Species presence/absence data; M = Mosterton; O = Oathill Farm; B = Broom; W = Whitford Bridge. 1-6 = Sample numbers.

48.3 to 58.3 species per sample. Furthermore the poor performance of operator one at Oathill Farm suggested that such variability might be site dependent. These hypotheses were tested by analysis of variance.

For the two data sets examined (family and species level), differences in the number of taxa per sample were significant with respect to sites, operators and site \times operator interactions. In each case the greatest source of variability arose from site effects. At family level this was expressed by a higher significance level for inter-site variation ($F_{3,12} = 9.43$; $P < 0.01$) than between operators ($F_{2,12} = 4.62$; $P < 0.05$) or for site \times operator interactions ($F_{6,12} = 3.76$; $P < 0.05$). At species level both site ($F_{3,12} = 10.11$; $P < 0.01$) and site \times operator interactions ($F_{6,12} = 5.93$; $P < 0.01$) were significant at the higher level with operator effects the least significant ($F_{2,12} = 4.87$; $P < 0.05$).

Differences in the number of taxa the 5 processors removed from samples were also assessed after first removing site and field operator effects from the data. At family level processor effects were significant ($F_{4,14} = 3.17$; $P < 0.05$) but at species level just failed to be so ($F_{4,14} = 2.51$; $P > 0.05$). It should be noted that different numbers of samples were processed by each of the 5 people involved. The greatest differences detected were between those people who had processed the greatest number of samples.

Qualitative differences

Analysis of variance techniques are purely concerned with number of taxa taken per sample and ignore differences in the type of taxa caught. Clustering and ordination techniques, which take note of the taxa in common between samples, were both used to test whether samples taken from different sites can be sufficiently distinct, on the basis of species content, to withstand operator and identifier effects.

Average (= unweighted mean pair group) linkage was used to generate sample clusters after similarity values were derived from the Czekanowski Index (Czekanowski, 1913). This index was favourably reviewed by Hellawell (1978) for use in the analysis of benthic surveys.

When applied to the 3 Axe data sets, families (presence/absence), families (log categories) and species (presence/absence) successively, the sites segregated with increasing faithfulness (Fig. 2). At family (presence/absence) level the tendency for samples from the same site to cluster together was quite clear but there were also several misplacements. When log categories were added to the family data set a considerable improvement was effected, with only one sample lying outside a perfect inter-site segregation (Operator 1, sample 3, Oathill Farm) but total separation was only achieved by using qualitative species data.

Reciprocal Averaging (RA) (Hill, 1973) was selected as a representative ordination technique. The dependence of sample scores upon the species scores, and vice versa, which this method involves, suggests that it may have a greater ecological validity than many

comparable techniques. It has been favourably compared with Principal Component Analysis (PCA) and polar or Bray-Curtis Ordination (PO) (Gauch *et al.*, 1977).

Applying RA (with rare species down-weighted) to the 3 Axe data sets presents the same sequence of improvement in site separation (Fig. 3a) as shown by clustering. First axis ordination scores from RA can be used as a basis for dichotomous classification of sites by Indicator Species Analysis (ISA) (Hill *et al.*, 1975). When applied to the 3 data sets (Fig. 3b) 3 samples were apparently misclassified at family level (presence/absence), only one when log categories were used and perfect site segregation was attained at species level.

Taxon accretion

The sampling exercise also produced information on taxon accretion with repeated collections. In order to eliminate operator effects the proportion of taxa collected after each 3 min period was expressed as the mean of all possible sequences for the 6 samples at each site. This also enabled the highest and lowest possible catches after each 3 min interval to be presented (Fig. 4). The calculated values are based on the total 18 min catch representing 100% as a convenient way of comparing sites.

At family level, considering all sites, a mean of 62.3% of the full catch was captured after 3 min. The extreme values ranged from 41.7 to 83.3%. After 6 min this rose to 78.3% (range 59.2–90.2%) and then to 87.3% (73.5–95.0%) after 9 min. At species level the mean proportions taken after each time interval were lower. The respective values for 3, 6 and 9 min, with ranges, were 49.6% (29.4–71.4%), 68.1% (46.2–86.7%) and 79.7% (63.9–94.1%). This was because although the chances of a family occurring in an early sample were high this did not necessarily mean that all species in that family were represented.

DISCUSSION

Inherent in biological monitoring programmes is the underlying assumption that a sample taken from a site is adequate to define that site and to allow comparisons to be made with any other site. The River Communities Project involves an estimated 40–60 biologists collecting field samples from a total of 267 sites spread over 41 rivers throughout Great Britain. Most of these samples will be taken by pond-net. The exact technique of netting used by each person will depend, not only upon individual interpretations of the method, but also upon river conditions. Factors such as current speed, water depth, substratum stability and macrophyte cover can all be expected to affect technique. In the face of this variation it is important to establish that such inter-operator differences are not so extreme as to outweigh inter-sample differences.

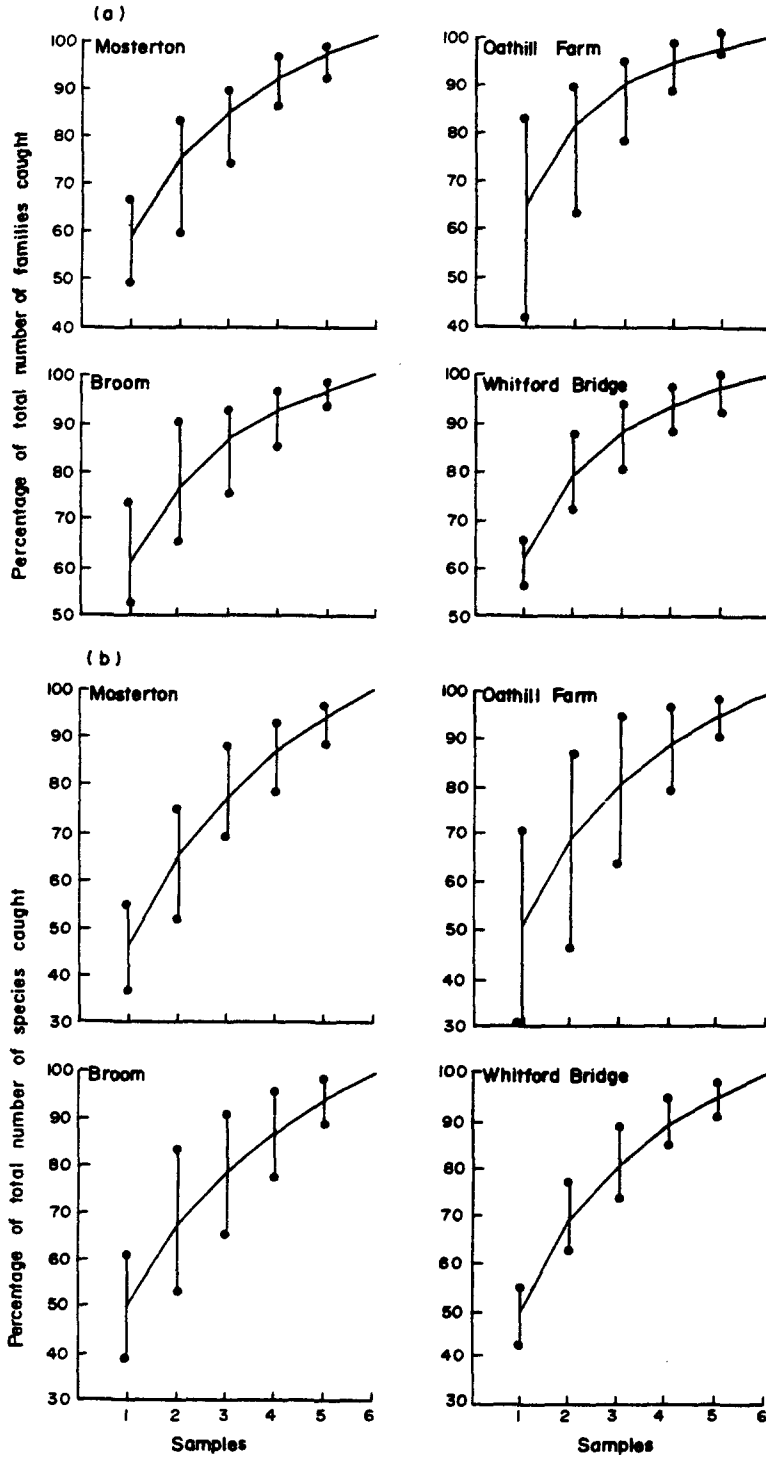


Fig. 4. Taxon accretion for each site. Mean, maximum and minimum values derived from all possible combinations of sample order. (a) Family accretion. (b) Species accretion.

The use of a pond-net as a monitoring technique has been used by many workers including Morgan & Egglisshaw (1965) and Armitage *et al.* (1974). In the quoted examples more than 1 collector was used to obtain data which were then to be compared. In both instances the authors suggest or imply that inter-

operator differences had no marked impact upon the results. Where co-workers are able to standardize methodologies this may seem to be a reasonable assumption. However work by Mackey *et al.* (in preparation) on Berkshire chalk-streams has demonstrated that inter-operator differences in the number of taxa

caught by pond-net can often be highly significant when samples are standardized principally by time.

Information collected on the Axe has shown that demonstrable inter-operator differences that exist, when techniques are poorly standardized, vary in importance according to the taxonomic and quantitative status of the data. When the qualitative family data were used misclassifications were commonplace, but when species level data were used misclassifications were eliminated. This improvement is not unexpected as longitudinal replacement of closely related species is a common feature of river systems (Illies & Botosaneanu, 1963; Hynes, 1970; Hawkes, 1975). Changes of this type would not be detected at family level, as demonstrated by the greater similarity coefficients between sites when family rather than species data are used (Fig. 2).

Of equal interest is the improvement in inter-site segregation if the family level data is crudely quantified. Attempts to derive quantitative data from pond-net samples have usually involved establishing a relationship between the number of individuals collected in a fixed area or period of netting and numbers captured by quantitative samplers used at the same site and time (Hynes, 1961; Morgan & Egglshaw, 1965; Armitage *et al.*, 1974). By use of a multiplication factor, taxa caught by pond-net could then, in theory, be expressed as numbers per unit area of stream-bed. Armitage *et al.* (1974) cautiously argue that, because of differences in river conditions, data derived from pond net samples can still only be considered precise enough to allow gross comparisons between population densities of different streams, sites or seasons. Such caution is wise in view of the difficulties encountered in using, so called, quantitative sampling equipment to obtain population densities (Needham & Usinger, 1956; Chutter & Noble, 1966; Dickson *et al.*, 1971; Chutter, 1972; Resh, 1979; Downing, 1979). Nevertheless, Morgan and Egglshaw (1965) were able to establish reproducible results from replicate samples at a given site. They were also able to demonstrate that, in comparing pond-net, shovel and cylinder samples at a fixed site, the same species were recorded as being the most abundant by each method. This held even though the pond-net samples were taken exactly one year after the others. Morgan & Egglshaw (1965) concluded that it was not necessary to obtain absolute values of the quantity of the bottom fauna if the differences between catches were directly proportional to the differences between streams.

Application of crude logarithmic categories to the Axe family data appeared to be justified since it improved site recognition. What cannot be established on the basis of the data available is whether differences in abundances reflected actual variation in community structure at the sites or merely differing degrees of difficulty in catching each taxon under variable stream conditions. The degree to which the composition of a pond-net sample is a measure of the

community it purports to represent is in need of more detailed investigation.

The greatest tendency for sample misclassification occurred at Oathill Farm, which exhibited the greatest habitat diversity of the 4 sites. Failure to exploit all sources of faunal richness, as was established to be the case with operator one, resulted in an inability to demonstrate the discrete identity of this site at family level even if the data were quantified. Even here however identification to species provided perfect segregation.

The excellent segregation of Axe sites, by multivariate techniques, when data were examined at species level, has provided justification for using a large number of people to collect pond-net samples in the RCP exercise. The test sites were closely spaced geographically and were relatively similar in the chemical characteristics of their waters (Table 2). Much greater differences in environmental conditions and benthic communities will be encountered over the range of sites surveyed in the main programme.

As a consequence of the justification this trial provided, biologists co-operating with the RCP study were asked to sample for 3 min with a standard FBA, or equivalent pond-net, such that all available habitats at the site were sampled in proportion to their occurrence.

Macan (1957), Hynes (1961) and Morgan & Egglshaw (1965) have all indicated that the minimum number of times in a year that British streams need to be sampled in order to collect most of the major species, is two. It was decided that to be reasonably sure of attaining this end each RCP site should be sampled 3 times, roughly equivalent to spring, summer and autumn samples, within a 12 month period. Constraints of time made it impossible to evaluate by field collections, the proportion of taxa present at a site that might be captured by such a sampling regime. The Axe data did, however, provide secondary information on the nature of species accretion with successive samples of the type to be used in the RCP study.

The result that approx. 62, 78 and 87% of families and 50, 68 and 80% of species in a series of 6 samples are caught after the first, second and third sample is in general accord with other published information. Frost *et al.* (1971) used kick-sampling techniques to sample the Downing Stream, Flintshire. In a series of 10 successive samples taken within the space of a few minutes 86% of the total number of taxa recorded were captured in the first 2 samples. They give no indication of the taxonomic precision upon which this result is based and the value is slightly higher than the equivalent two catch value for Axe family level data. This is explicable in terms of the greater standardization of field methodology used. Their value however does fall clearly within the equivalent range for Axe family level data and just within the species range. Morgan & Egglshaw (1965) collected 6 single kick-samples from each of several sites in a Highland

stream and paired each 2 successive kicks. This gave 3 paired kicks per site. The proportion of the total site fauna occurring in the first pair of kicks varied between 51–87% at the various stations. Their level of identification was nearer to the species level of the Axe study and the range of results in the two works are very similar. Comparison of the 3 studies in geographically well spaced streams supports the extrapolation of the Axe results to a wider national context.

Although it is clear that 3 samplings, each of 3 min duration, will not give a comprehensive family/species list, there is good reason to believe that the samples to be obtained for the RCP study will contain sufficient information for a preliminary classification of benthic macro-invertebrate communities of streams and rivers. The initial classification may then be extended and refined by more detailed studies to examine the relationships between physical and chemical features of rivers and the communities they support.

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APPENDIX I

A list of the taxa recorded in the River Axe during this study, indicating the level of taxonomic precision used. Upper case taxa are those used at "family" level; lower case taxa are those used at "species" level.

PLANARIIDAE

Polycelis nigra group¹

NERITIDAE

Theodoxus fluviatilis (L.)

HYDROBIIDAE

Potamopyrgus jenkinsi (Smith)

Bithynia tentaculata (L.)

Bithynia leachi (Sheppard)

LYMNAEIDAE

Lymnaea peregra (Müller)

PHYSIDAE

Physa fontinalis (L.)

PLANORBIDAE

Gyraulus albus (Müller)

Gyraulus laevis (Alder)

Bathymphalus contortus (L.)

Hippeutis complanatus (L.)

SPHAERIIDAE

Sphaerium corneum (L.)

Pisidium amnicum (Müller)

Pisidium casertanum (Poli)

Pisidium personatum Malm

Pisidium milium Held

Pisidium subtruncatum Malm

Pisidium henslowianum (Sheppard)

Pisidium hibernicum Westerlund

Pisidium nitidum Jenyns

NAIDIDAE

Nais elinguis (Müller)

Nais bretscheri Michaelsen

- TUBIFICIDAE**
Tubifex tubifex (Müller)
Tubifex ignotus (Stolc)
Psammoryctides barbatus (Grube)
Limnodrilus claparedeianus Ratzel
Limnodrilus hoffmeisteri Claparède
Limnodrilus udekemianus Claparède
Peloscoclex ferox (Eisen)
Rhyacodrilus coccineus (Vejdovsky)
Aulodrilus plurisetus Piguet
- ENCHYTRAEIDAE**
 Enchytraeidae
- LUMBRICULIDAE**
Lumbriculus variegatus (Müller)
Stylogrilus heringianus Claparède
- LUMBRICIDAE**
Eiseniella tetraedra (Savigny)
- PISCICOLIDAE**
Piscicola geometra (L.)
- GLOSSIPHONIIDAE**
Theromyzon tessulatum (Müller)
Glossiphonia complanata (L.)
Helobdella stagnalis (L.)
- ERPOBDELLIDAE**
Erpobdella octoculata (L.)
- SPERCHONIDAE**
Sperchon hibernicus Halbert
Sperchon setiger Thor
- LEBERTIIDAE**
Lebertia (Pilolebertia) inaequalis (Koch)
Lebertia (Pilolebertia) insignis insignis Neuman
Lebertia (Pilolebertia) porosa porosa Thor
- HYGROBATIDAE**
Hygrobates (Hygrobates) fluviatilis (Ström)
Hygrobates (Hygrobates) nigromaculatus Lebert
Atractides (Atractides) nodipalpis nodipalpis (Thor)
- ASELLIDAE**
Asellus meridianus Racovitza
Asellus aquaticus (L.)
- GAMMARIDAE**
Crangonyx pseudogracilis Bousfield
Gammarus pulex (L.)
- BAETIDAE**
Baëtis vernus Curtis
Baëtis buceratus Eaton
Baëtis rhodani (Pictet)
Baëtis muticus (L.)
Centroptilum luteolum (Müller)
- HEPTAGENIIDAE**
Rhithrogena semicolorata (Curtis)
Heptagenia sulphurea (Müller)
Ecdyonurus sp.
- LEPTOPHLEBIIDAE**
Paraleptophlebia submarginata (Stephens)
Habrophlebia fusca (Curtis)
- EPHEMERIDAE**
Ephemera danica Müller
- CAENIDAE**
Caenis rivulorum Eaton
Caenis moesta group²
- NEMOURIDAE**
Protonemura montana Kimmins?
Amphinemura sp.
Nemurella picteti Klapálek
Nemoura cambrica (Stephens)
Nemoura erratica Classen
- LEUCTRIDAE**
Leuctra hippopus (Kempny)
Leuctra nigra (Olivier)
- PERLODIDAE**
Isoperla grammatica (Poda)
- AGRIIDAE**
Agrion splendens (Harris)
- APHELOCHEIRIDAE**
Aphelocheirus aestivalis (Fabricius)
- CORIXIDAE**
Sigara dorsalis (Leach)
Micronecta sp.
- HALIPLIDAE**
Brychius elevatus (Panzer)
Haliplus lineatocollis (Marshall)
- DYTISCIDAE**
Potamonectes depressus complex
Oreodytes sanmarki (Sahlberg)
- GYRINIDAE**
Orectochilus villosus (Müller)
- HYDROPHILIDAE**
Hydraena gracilis Germar
Helophorus brevipalpis Bedel
- DRYOPIDAE**
Helichus substriatus (Müller)
- ELMINTHIDAE**
Elmis aenea (Müller)
Limnius volckmari (Panzer)
Oulimnius tuberculatus (Müller)
- SIALIDAE**
Sialis lutaria (L.)
Sialis nigripes Pictet
- RHYACOPHILIDAE**
Rhyacophila dorsalis (Curtis)
Agapetus sp.
Glossosoma sp.
- POLYCENTROPODIDAE**
Polycentropus flavomaculatus (Pictet)
Polycentropus irroratus (Curtis)
- PSYCHOMYIIDAE**
Tinodes waeneri (L.)
Tinodes unicolor (Pictet)?
Psychomyia pusilla (Fabricius)
- HYDROPSYCHIDAE**
Hydropsyche pellucidula (Curtis)
Hydropsyche sitalai (Döhler)
Hydropsyche instabilis (Curtis)
Hydropsyche contubernalis McLachlan
Cheumatopsyche lepida (Pictet)
- HYDROPTILIDAE**
Hydroptila sp.
- LIMNEPHILIDAE**
Drusus annulatus Stephens
Limnephilus lunatus Curtis
Anabolia nervosa Curtis
Potamophylax group³
Halesus group⁴
- MOLANNIDAE**
Molanna angustata Curtis
- BERAEIDAE**
Beraea pullata (Curtis)
Beraeodes minutus (L.)
- GOERIDAE**
Goera pilosa (Fabricius)
Silo sp.
- SERICOSTOMATIDAE**
Sericostoma personatum (Spence)
- BRACHYCENTRIDAE**
Brachycentrus subnubilus Curtis
- LEPIDOSTOMATIDAE**
Lepidostoma hirtum (Fabricius)
Lasiocephala basalis (Kolenati)
- LEPTOCERIDAE**
Athripsodes cinereus (Curtis)
Athripsodes bilineatus (L.)
Mystacidus azurea (L.)
- TIPULIDAE**
Dicranota sp.
Eloeoephila sp.
Antocha vitripennis Meigen

<i>Molophilus</i> sp.	CHIRONOMINI ⁷
<i>Tipula montium</i> group ⁵	<i>Chironomus</i> sp.
PSYCHODIDAE	<i>Microtendipes</i> sp.
<i>Pericoma</i> sp.	<i>Demicryptochironomus vulneratus</i> (Zetterstedt)
<i>Psychoda brevicornis</i> Tonnoir	<i>Pentapeditum</i> group ⁸
PTYCHOPTERIDAE	TANYTARSINI
<i>Ptychoptera</i> sp.	<i>Tanytarsus</i> group ⁹
CERATOPOGONIDAE	<i>Rheotanytarsus</i> group ¹⁰
Ceratopogonidae	<i>Cladotanytarsus</i> sp.
TANYPODINAE	SIMULIIDAE
<i>Thienemannimyia</i> group ⁶	<i>Simulium</i> (<i>Wilhelmia</i>) <i>equinum</i> L.
<i>Apsectrotanypus trifascipennis</i> (Zetterstedt)	<i>Simulium</i> (<i>Wilhelmia</i>) <i>lineatum</i> Meigen
DIAMESINAE	<i>Simulium</i> (<i>Simulium</i>) <i>reptans</i> L. var. <i>galeratum</i> Edwards
<i>Pothastia longimana</i> Kieffer	<i>Simulium</i> (<i>Simulium</i>) <i>argyreatum</i> Meigen
<i>Pothastia gaedii</i> (Meigen)	<i>Simulium</i> (<i>Simulium</i>) <i>ornatum</i> Meigen
<i>Diamesa</i> sp.	<i>Simulium</i> (<i>Simulium</i>) <i>rheophilum</i> Knoz
ORTHOCLADIINAE	RHAGIONIDAE
<i>Eukiefferiella</i> sp.	<i>Atherix ibis</i> (Fabricius)
<i>Brillia modesta</i> (Meigen)	TABANIDAE
<i>Rheocricotopus</i> sp.	Tabanidae
<i>Parametricnemus stylatus</i> (Kieffer)	MUSCIDAE
<i>Corynoneura</i> sp.	<i>Limnophora</i> sp.
<i>Thienemanniella</i> sp.	Indet. sp.
<i>Epoicocladus flavens</i> (Malloch)	STRATIOMYIDAE
<i>Nanocladus rectinervis</i> (Kieffer)	<i>Oxycera</i> sp.
<i>Orthocladus</i> group ⁷	EMPIDIDAE ¹¹
PRODIAMESINAE	<i>Clinocera</i> type
<i>Prodiamesa olivacea</i> (Meigen)	<i>Chelifera</i> type
	<i>Wiedemannia</i> type
	<i>Hemerodromia</i> type

¹ *Polycelis nigra* group includes *Polycelis nigra* (Müller) and *P. tenuis* (Ijima).

² *Caenis moesta* group includes *Caenis moesta* Bengtsson and *C. macrura* Stephens.

³ *Potamophylax* group includes *Potamophylax cingulatus* (Stephens), *P. latipennis* (Curtis), *P. rotundipennis* (Brauer), *Allogamus auricollis* (Pictet) and *Chaetopteryx villosa* (Fabricius).

⁴ *Halesus* group includes *Halesus radiatus* (Curtis), *H. digitatus* (Schrank) and *Hydatophylax infumatus* (McLachlan).

⁵ *Tipula montium* group includes *Tipula montium* Egger, *T. lateralis* Meigen and *T. couckeii* Tonnoir.

⁶ *Thienemannimyia* group includes the genera *Thienemannimyia*, *Arctopelopia*, *Rheopelopia* and *Conchapelopia*.

⁷ *Orthocladus* group includes the genera *Orthocladus* and *Cricotopus*.

⁸ *Pentapeditum* group includes the genera *Pentapeditum* and *Polypeditum*.

⁹ *Tanytarsus* group includes the genera *Tanytarsus* and *Micropsectra*.

¹⁰ *Rheotanytarsus* group includes the genera *Rheotanytarsus* and *Paratanytarsus*.

¹¹ The division of Empididae follows Brindle A. (1964) *Entomologist* 97, 162-165.

It is not implied that each member of each of these groups was recorded in the River Axe.