

## Article (refereed) - postprint

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Worrall, T.P ; Dunbar, M.J.; Extence, C.A.; Laize, C.L.R.; Monk, W.A.; Wood, P.J. 2014. **The identification of hydrological indices for the characterization of macroinvertebrate community response to flow regime variability**

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**This is an Accepted Manuscript of an article published by Taylor & Francis Group in *Hydrological Sciences Journal* on 17/04/2014, available online:**  
<http://www.tandfonline.com/10.1080/02626667.2013.825722>

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***The identification of hydrological indices for the characterisation of macroinvertebrate community response to flow regime variability***

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**ABSTRACT**

The importance of flow regime variability for maintaining ecological functioning and integrity of river ecosystems has been firmly established in both natural and anthropogenically modified systems. In this paper we examine river flow regimes across lowland catchments in eastern England using 47 variables, including those derived using the Indicators of Hydrologic Alteration (IHA) software. A Principal Components Analysis (PCA) method was used to identify redundant hydrological variables and those that best characterised the hydrological series (1986-2005). A small number of variables (< 6 variables) characterised up to 95% of the statistical variability in the flow series. The hydrological processes and conditions that the variables represent were found to be significant in structuring the instream macroinvertebrate community LIFE scores at both the

family- and species-level. However, hydrological variables only account for a relatively small proportion of the total ecological variability (typically <10%). The research indicates that a range of other factors, including channel morphology and anthropogenic modification of instream habitats, structure riverine macroinvertebrate communities in addition to hydrology. These factors need to be considered in future environmental flow studies to enable the characterisation of baseline/reference conditions for management and restoration purposes.

**Keywords:-** e-flow, inter-annual flow regime, community response, hydromorphology, ecohydrology

## **INTRODUCTION**

It is widely recognised that the river flow regime and its inherent variability is one of the primary factors structuring instream communities (Poff *et al.* 1997, Bunn and Arthington 2002, Lytle and Poff 2004, Durance and Ormerod 2007, Monk *et al.* 2008) and in the absence of other confounding factors, such as pollution, distinct ecological communities have been associated with different flow regimes (e.g. Richter, *et al.* 1996, Wood and Armitage 2004, Monk *et al.* 2006, Durance and Ormerod 2009, Poff *et al.* 2010).

The need to provide water (flow) to protect the environment and instream needs, in addition to anthropogenic requirements, has been increasingly recognised internationally (Acreman and Ferguson 2010, Monk *et al.* 2011). Although most legislation relating to the management of water resources and the protection of the riverine environment does not explicitly use the term 'environmental flows' or 'e-flows', it is widely recognised that the delivery or maintenance of an appropriate flow regime is essential for the sustainable management of riverine ecosystems (Poff *et al.* 2010, Shenton *et al.* 2012). As a result, national guidelines

and legislation are increasingly recognising the need for river flow targets that reflect the competing demands on finite hydrological resources (Acreman and Ferguson 2010, Arthington *et al.* 2006, King and Brown 2010, Peters *et al.* 2012, Shenton *et al.* 2012).

The majority of ‘environmental flows’ research to date has focused on the assessment of different components of the flow regime and the extent to which these components potentially influence aquatic communities in North America (Richter *et al.* 1996, Poff *et al.* 1997, Armanini *et al.* 2012) Europe (Monk *et al.* 2006, 2008, Belmar *et al.* In Press), southern Africa (King and Brown 2010) and Australia (Sheldon and Thoms 2006, Leigh and Sheldon 2009). These studies have subsequently been used to help define sustainable ecological / environmental flow regimes in regulated and anthropogenically modified rivers (Acreman *et al.* 2008, Poff *et al.* 2010, Dunbar *et al.* 2010a, Peters *et al.* 2012).

Internationally, the 32 hydrological indices comprising the Nature Conservancy’s Indicators of Hydrologic Alteration (IHA) variables (Richter *et al.* 1996) have become the basis for characterising the natural flow regime and modifications to it in many locations. These variables have also been widely used for identifying potentially ‘ecologically-relevant’ hydrological drivers of aquatic floral or faunal community structure. The IHA variables quantify five components of the river flow regime, namely (i) magnitude of monthly water conditions; (ii) magnitude and duration of extreme water conditions; (iii) timing of annual extreme weather conditions; (iv) frequency and timing of high and low pulses; and (v) rate and frequency of water condition changes (Richter *et al.* 1996).

In this study we examine the long-term patterns of inter-annual flow regime variability (1986-2005) across 26 lowland river catchments (28 river gauges) from Eastern England, UK

(Fig. 1). A set of 47 hydrological indices (from a pool of over 200 variables) reported to be ecologically relevant in previous research (Richter *et al.* 1996, Poff *et al.* 1997, Extence *et al.* 1999, Wood *et al.*, 2001, Monk *et al.*, 2006) were used to characterise the flow regimes. The potential influence of these hydrological variables freshwater macroinvertebrates were examined in association with family- and species/genus-level macroinvertebrate community data and the Lotic invertebrate Index for Flow Evaluation (LIFE) scores (Extence *et al.* 1999). The LIFE method was developed by Extence *et al.* (1999) to link qualitative and semi-quantitative changes in riverine benthic macroinvertebrate communities to antecedent and current flow characteristics. The LIFE scores for individual families and species are based upon the known associations of British benthic macroinvertebrates to flow velocity. The LIFE method and scores have been widely used in the UK to examine and assess macroinvertebrate community response to flow regime variability in a number of studies (Monk *et al.* 2008, 2012, Dunbar *et al.* 2006, 2010a, 2010b, Dunbar and Mould 2009, Clews and Ormerod 2010, Wilby *et al.* 2011, Vaughan and Ormerod 2012).

This research aims to demonstrate how paired hydroecological datasets can be used to explore the potential hydrological drivers of instream communities and to demonstrate how it is possible to move beyond the identification of ‘ecologically relevant variables’ to testing their expression. We hypothesise that: i) a small subset of hydrological variables (including the IHA variables derived using freely available software) can be identified using a redundancy reduction approach (*sensu* Olden and Poff 2003) to characterise flow regime characteristics at the regional scale; ii) the underlying influence of the flow regime on the instream macroinvertebrate community can be identified in family- and species/genus-level macroinvertebrate community data using multivariate analysis (DCA); and iii) the potential influence of the hydrological variables, identified via the PCA redundancy approach, on the

macroinvertebrate community LIFE scores (Extence *et al.* 1999) at both the family- (LIFE F) and species/genus-level (LIFE S) can be identified and quantified.

## **METHODS**

### ***Study sites and data sets***

Following preliminary screening of sites to remove those subject to water quality and other anthropogenic pressures, 28 river flow series (mean daily discharge;  $\text{m}^3 \text{s}^{-1}$ ) from 26 catchments were paired with the long-term (1986-2005) spring and autumn macroinvertebrate community abundance data from 88 sites from eastern England (Fig. 1). Macroinvertebrates were sampled following the standard 3-minute kick method (Murray-Bligh 1999). All sites were centred on riffles and operators sampled the range of aquatic habitats present at a site in proportion to their occurrence. Macroinvertebrates were sorted and identified, with the majority (>90%) of routine identifications being resolved to species- or genus-level over the 20-year study period (excluding Diptera and Oligochaeta). The abundance of macroinvertebrates / taxa was recorded as either total counts or in abundance classes for some older historic samples (prior to 2002); providing a measure of the minimum abundance of each taxa.

Data forming the paired hydrological and ecological datasets comprised (i) spring (March 1st - May 31st) and autumn (1st September - 30th November) (seasonal) macroinvertebrate community data, and (ii) flow series with <10% of daily missing values in any given year. For sites with missing discharge data, values were interpolated using the long-term mean daily discharge series following the procedures outlined in Monk *et al.* (2008). All sites displayed similar hydrological regimes, with winter months experiencing higher flows and the summer months experiencing lower flows.

### ***Data preparation and analysis***

Seasonal paired datasets (spring and autumn) were prepared for analysis to ensure that the influence of antecedent hydrological conditions could be examined in association with the structure of the instream macroinvertebrate communities. Hydrological indices were calculated for hydrological years commencing 1st March - 28/29th February and 1st September - 31st August for the spring and autumn macroinvertebrate biomonitoring periods, respectively. For hydrological indices, that were strongly related to the size of the river / catchment, the series were standardized by deriving z-scores for each site (*sensu* Monk *et al.* 2006) to allow direct comparisons between gauges.

A total of 47 hydrological variables identified in previous research as being ‘ecologically relevant’ (e.g. Richter *et al.* 1996, Poff *et al.*, 1997, Monk *et al.* 2006) were used in the analysis to explore hydroecological associations over the 20-year study period. This included the 32 Indicators of Hydrologic Alteration (IHA) variables, calculated using the Nature Conservancy’s IHA software (version 7.1; Richter *et al.* 1996) and 15 additional variables reported to significantly influence aquatic communities in previous research in the UK (Wood *et al.* 2000, Gibbins *et al.* 2001, Monk *et al.* 2006). Principal Components Analysis (PCA) was used to identify the major sources of statistical variation among the hydrological variables, to identify redundant variables (multicollinearity) and identify a minimum sub-set of variables to characterise the hydrological series (*sensu* Olden and Poff 2003). Three subsets of hydrological indices were used to explore the flow regime variability and flow-ecology relationships (Table 1). The first group (Set 1) comprised all 47 indices recognising that many of the variables may be redundant; the second group of variables (Set 2) comprised the IHA variables, which have been widely used in other environmental flow studies; and the

third group (Set 3) contained the subset of variables identified as a result on the redundancy analysis procedure.

The long-term averaged hydrograph for all 28 gauging stations (1986-2005) and inter-annual LIFE scores (family- and species-level) for all 88 biomonitoring sites (1986-2005) was derived as part of the preliminary analysis. This facilitated the identification of any inter-annual patterns, trends or marked differences between in the series, including known hydrological events (floods or droughts).

Hydrological variables for the spring and autumn periods were analysed using Principal Components Analysis (PCA) within Canoco (ter Braak and Smilauer 1996) to identify redundant variables and the dominant (principal) variables. All hydrological variables were log-e transformed prior to analysis. A series of PCAs were undertaken incorporating a progressively reduced number of variables employing the PCA redundancy reduction approach employed by Olden and Poff (2003) and Monk *et al.* (2007). Given that previous research has indicated that a maximum of 6 hydrological indices (more typically one or two indices) have been incorporated in models characterising flow – ecology relationships (Clausen and Biggs 1997, Monk *et al.* 2006, Belmar *et al.* In Press), the 6 hydrological variables with the highest loadings on the first 2 PC axes were identified from the output of PCAs using Set 1 and Set 2 variables. Following the approach of Olden and Poff (2003), the number of variables used for each axis was proportional to the variance explained by each PC relative to the others (e.g. based on Set 1 variables - PC1 explained 45.3% of the total 69.5% variance explained, therefore four variables were selected from PC1 and two remaining variables were selected from PC2). Highly correlated variables (vectors running along the same axis) identified by significant correlation coefficient values ( $p < 0.05$ ) and those



significantly correlated with multiple variables but with a lower axis loading (shorter vector) were considered redundant and removed from further analyses. However, to enable the use of a single subset of variables for both spring and autumn periods (based on Set 1 and Set 2 variables), the variable(s) which had the greatest loading on both the spring and autumn PCA output models was included in the final selection of indices where a number of similar variables clustered together (e.g., characterising the magnitude of monthly water conditions).

To investigate the temporal variations within the ecological series and if this was related to an underlying environmental gradient associated with the flow regime variability, the spring and autumn macroinvertebrate data were analysed separately using Detrended Correspondence Analysis (DCA) within Canoco. Prior to analysis, the macroinvertebrate data were  $\log_e(x+1)$  transformed to reduce the clustering of common and abundant taxa at the centre of the ordination plot. Following preliminary analysis, rare taxa occurring in only one sample or with an abundance of  $<3$  across all samples were removed from the analysis to reduce their over weighted influence on the output (ter Braak and Smilauer 1996).

Following multivariate analysis of the hydrological series using (PCA) the axes scores for individual sample sites (flow gauges) were extracted and used as independent variables in bivariate correlations and to construct stepwise multiple linear regression models with the macroinvertebrates LIFE scores (family-level, LIFE F, and species/genus-level, LIFE S) as dependent variables. This approach provides a simple means to determine if any gradients represented on the first PC axis can be related to macroinvertebrate community response to flow regime variability. Stepwise multiple linear regressions models were computed using IBM SPSS Statistics (IBM 2012).

## RESULTS

### *Flow Regime Analysis*

The long-term average hydrograph for all sites (1986-2005) clearly indicates periods of reduced river discharge associated with known periods of drought (1989-1992 and 1996-1997) and higher flows (1988, 1998, 2000-2001) within the series (Fig. 2).

Principal Components Analysis of both spring and autumn hydrological datasets using both Set 1 (47 variables) and Set 2 (32 IHA variables) accounted for a similar amount of the variance across the first four PC axes (ranging between 69.8-72.5%) and facilitated the identification of 6 variables that facilitated the identification of the major sources of statistical variability in the hydrological series and minimised redundancy across all of the analyses (Table 2). Given the strong similarity in the PCA output for both spring and autumn periods, only output / figures for the latter are presented herein (Fig. 3). Examination of the PCA ordination biplots for both seasons indicated the vectors of the hydrological variables formed similar clusters on PC axis 1 and PC axis 2 (Fig. 3) although the order of the hydrological variables on PC axis 2 were reversed in some output (see Fig. 3a and 3b as an illustration). Five indices plotted on the negative end of PC axis 1 using both Set 1 and Set 2 of hydrological variables (Base Flow Index - BF, Reversals - Rev, Fall Rate - FR, Low Pulse Occurrences - LP# and Low Pulse Duration - LPD). The remaining hydrological variables plotted positively on PC axis 1 for Set 2. The 15 additional variables used in Set 1 (total of 47 indices) found to be potentially important in previous research on UK rivers also plotted positively on PC axis 1. However, none of these variables were more heavily loaded on PC axis 1 or axis 2 than any of the IHA variables (Set 2).

The subset of six variables identified to minimise redundancy among hydrological variables (Set 3) were consistently highly loaded on PC axis 1 and axis 2 of Set 1 (47 variables) and Set 2 (32 IHA variables) for both the spring and autumn datasets. The first four axes of the PCA using these variables (Set 3) accounted for 95.2% of the total variance in the hydrological dataset recorded for the autumn period (Fig. 3c) and 94.9% for spring within the hydrological dataset (Table 2).

When the sample scores for individual years from Set 3 were averaged and plotted, a clear inter-annual pattern of flow variability was observed on PC axis 1 (Fig. 4). Hydrological years associated with periods of high magnitude drought that spanned more than one year (1989-1992 and 1996-1997) plotted at the negative end of PC axis 1. Years characterised by higher flows and flooding (most markedly 2001) plotted at the positive end of PC axis 1 (Fig. 4). The second axis reflected inter-annual differences associated with periods of prolonged low flow (e.g., 1989-1992 and 1996-1997) or sustained higher river discharge (e.g., 1994-1995 and 2001).

### ***Macroinvertebrate Analysis***

Four DCAs were undertaken using spring and autumn macroinvertebrate community datasets for the family- and species/genus-level data. The cumulative percentage of variance of the macroinvertebrate community explained on the first four DCA axes were similar for both seasons (Table 3). However, analyses using the family-level macroinvertebrate data were able to account for a greater proportion of the variance (18.5% and 19.1%, respectively) than the species-level macroinvertebrate data (9.4% and 10.8%). When the mean sample scores for each year (1986-2005) were plotted (Fig. 5a and 5b), a similar gradient to that recorded for the PCA was observed on DCA axis 1. The macroinvertebrate community for the

majority of years coinciding with low-flow and drought conditions (1989-1991; 1996-1997) and sustained higher flow (1987-1988; 2000-2002) plotted at opposite ends of DCA axis 1 for both the family- and species/genus-level data (Fig. 5a and 5b). When the macroinvertebrate biplots were examined taxa from LIFE Flow Groups I and II (preferring faster flow velocities) were located at one end of axis one while those in LIFE Flow Group VI, V and VI (preferring slower flow velocities) were located at the opposite end (see supporting information). Years marking the transition between low-flow/drought and higher flows were more variable depending on the taxonomic level considered. The majority of transition/intermediate flow years plotted in the middle of DCA axis 1 (1994, 1995, 1998, 1999, 2003 and 2005 for family-level - Fig. 5a; 1989, 1992, 1994, 2003 and 2005 for species-level - Fig. 5b). Axis 2 of the DCA reflected inter-annual changes of the community associated with periods of prolonged low flow and high flow; although axis 2 accounted for a relatively low proportion of the variance recorded (<5%).

### ***Hydroecological relationships***

Plotting the seasonal (spring and autumn) family- (LIFE F) and species-level (LIFE S) LIFE scores alongside the long-term average hydrograph (Fig. 2) indicated that during periods of reduced discharge LIFE scores were typically depressed, with the lowest LIFE scores (LIFE F and LIFE S) being recorded between autumn 1991 and spring 1993 (Fig. 2). In contrast, the highest average LIFE scores were associated with periods of sustained elevated discharge (1986-1988 and 2000-2002 for both LIFE F and LIFE S; and moderate elevation during 1994-1995 for LIFE F) (Fig. 2).

To examine the gradients identified in the PCA and DCA further, LIFE F and LIFE S were correlated with samples scores from PCA axis 1 and DCA axis 1, respectively. There was a

relatively weak positive correlation between PCA axis 1 scores for all three sets of hydrological variables and the LIFE F and LIFE S scores for both the autumn and spring survey periods (Table 4a). The correlation coefficients recorded were higher for LIFE F in all instances, although all were significant (Table 4a). Stepwise multiple linear regression models generated for the LIFE scores using the PCA axis 1 scores and the reduced set of 6 hydrological variables (Set 3) were able to explain between 8-9% of the variance in the LIFE F score for both the autumn and spring periods, whilst only 4-6.3 % of the variance in the LIFE S score was able to be explained (Table 4b). For two of the models, PCA axis 1 scores were the most influential variable (LIFE S - autumn and LIFE F - spring). For autumn LIFE F, the annual 7-day minimum flow (7-day min) was the most influential variables and for spring LIFE S the fall rate was the most influential variable. Strong negative correlations were recorded between DCA axis 1 sample scores and LIFE F for both autumn ( $r = -0.830$ ,  $P < 0.01$ ) and spring ( $r = -0.805$ ,  $P < 0.01$ ) and LIFE S scores (autumn:  $r = -0.855$ ,  $P < 0.01$ ; and spring:  $r = -0.772$ ,  $P < 0.01$ ) indicating the presence of an environmental gradient reflecting flow regime variability (Table 4c). However, given the interest in the response of macroinvertebrate LIFE scores to flow variability, the DCA samples scores were not used in the development of stepwise multiple linear regression models at this stage.

## **DISCUSSION**

Until relatively recently the majority of environmental flow studies were primarily centred on components of the long-term flow duration curve considered to be important and relevant to instream ecology and habitats (Gao *et al.* 2012, Peters *et al.* 2012). The results of this research indicate that the influence of both long and short term antecedent flow regime characteristics can be clearly identified using benthic community data and the LIFE score. In addition, the results demonstrate the power and value of integrating long-term hydrological

and ecological datasets within instream flow assessment studies (Monk *et al.* 2006, Belmar *et al.* In Press).

Comparison between the long term flow series and LIFE scores (family- and species-level) indicated that the macroinvertebrate community appeared to reflect changes in the flow regime (Fig. 2). Gradients reflecting inter-annual changes in river flow from higher to lower discharge (years) were identified when analysing the hydrological (PCA) and ecological series (DCA) independently (Fig. 4 and 5). However, while the influence of the antecedent flow regime on the instream community could be detected using multivariate analysis when this was quantified via the development of regression models it only accounted for a relatively small proportion of the statistical variance (<10% for both seasons and taxonomic resolutions). This clearly highlights that there are a range of other important factors structuring the instream community that may include channel morphology and habitat characteristics (Dunbar *et al.* 2010a), biotic interactions (Shenton *et al.* 2012) and water quality (Durance and Ormerod 2009) and may need to be incorporated into future analyses. However, screening of sites and data in the preliminary stages of this research ensured that sites with known water quality pressures were removed.

The IHA methodology was developed to enable the hydrological regime of a river to be quantified via a set of 'ecologically-relevant indicators' (Richter *et al.* 1996). Whilst the selection of these indices was based upon extensive research (Gustard 1984, Kozlowski, 1984, Hughes and James, 1989, Poff and Ward 1989), only a limited number of these studies have been able to integrate hydrological and ecological data over medium- to long-term time periods due to the absence of appropriate ecological datasets in many areas (Shenton *et al.* 2012, Monk *et al.* 2012, Vaughan and Ormerod 2012). Despite major advances in our

understanding for temporal and spatial variability in river flow regime characteristics, the availability of paired hydrological and ecological datasets for specific sites or reaches remains a major obstacle to quantifying the nature of any relationships which could be used to underpin the development of environmental flow criteria in many regions (Arthington *et al.* 2006, Poff *et al.* 2010).

This result of this study demonstrate that the IHA variables can effectively characterise the major sources of statistical variability in the flow regime and form the basis of exploring their potential influence on instream macroinvertebrate communities using a paired long-term data set. The PCA redundancy approach (*sensu* Olden and Poff 2003, Monk *et al.* 2007) demonstrated a high level of multicollinearity between the majority of the 47 hydrological indices. However, it is worth noting that none of the additional 15 hydrological indices, identified in previous hydroecological studies in the UK as being ecologically important (Gibbins *et al.* 2001, Monk *et al.* 2006, 2008), were more heavily loaded on any of the PCA axes and none were included in the final set of 6 variables (Set 3).

The six indices identified by the PCA redundancy approach were able to explain ~95% of the statistical variance within the hydrological series and represent a relatively quick and robust method to screen data that could be easily employed in future hydroecological research. The removal of ‘redundant’ indices followed a simple set of criteria based on identifying the indices most heavily loaded on PCA axes and comparison of the loadings for the two seasons (autumn and spring) for which ecological data were available. The final set of 6 indices (Set 3) included three quantifying low-flow / discharge characteristics (Base Flow Index – BF; Low Pulse Duration – LPD; Annual 7-day minimum flow – 7min). This probably reflects the relatively high contribution of groundwater to many of the rivers in the region studied (Monk

*et al.* 2012) and the occurrence of two high magnitude national scale droughts within the study period (Marsh *et al.* 2007). Two of the variables identified also quantified high flow characteristics (Annual 7-day maximum flow – 7max; Rise Rate – RR) demonstrating that flow regime characteristics across the entire hydrograph are important. However, when these variables and the PCA axis-scores were used to develop stepwise multiple linear regression models only one variable was incorporated into any of the models.

The output derived from the stepwise multiple linear regression models developed indicated that family-level macroinvertebrate data (LIFE F) provided marginally better predictive power than those derived using species-level data (LIFE S); although all models yielded significant output and the total amount of variance explained was relatively low (<10%). This is in contrast to other recent studies in the UK (Monk *et al.* 2012) and Spain (Belmar *et al.* In Press) where models developed using species/genus-level data were able to account for a greater proportion of the variance in relation to flow variability. The reduced predictive capacity associated with the species/genus level data probably reflects the greater complexity of the community data across the 88 macroinvertebrate biomonitoring sampling sites (192 taxa at species/genus-level compared to 73 taxa at the family-level) and natural biogeographical differences in community composition across the sites studied.

## **CONCLUSION**

The temporal variability of river flow regimes is a primary structuring factor of instream communities. The use of IHA indices and the application of the redundancy minimisation approach (*sensu* Olden and Poff 2003, Monk *et al.* 2007) in this study enabled the identification of a small number of variables to characterise the flow regime of 26 catchments in lowland England. The influence of these variables on the macroinvertebrate community



family- and species-level LIFE scores could be identified. However, only a relatively small proportion of the statistical variance within the ecological data could be accounted for by hydrological indices alone. This demonstrates the high level of redundancy associated with hydrological indices and also reinforces the fact that a range of other biotic and abiotic factors (in addition to the flow regime) structure instream communities (Dunbar *et al.* 2010a, Durance and Ormerod 2009, Shenton *et al.* 2012, Vaughan and Ormerod 2012). These factors, including riverine habitat, channel structure characteristics and water quality, need to be incorporated in future analysis and environmental flow studies so that they can help inform future management strategies. The approach used in this study could be easily adapted for use in other locations to characterise flow regime characteristics or reference conditions for natural and semi-natural rivers and, with appropriate recognition of other factors structuring instream communities, for the development of environmental flow criteria for rivers subject to flow regulation and anthropogenic modifications.

## **ACKNOWLEDGEMENTS**

TW acknowledges the support of a Loughborough University Department of Geography scholarship with a CASE award from the Centre for Ecology and Hydrology (CEH) and the Environment Agency of England and Wales (EA). CEH and EA are thanked for providing river discharge series and the EA for providing the macroinvertebrate data. The views expressed in this paper are those of the authors and not necessarily those of the EA.

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**Table 1 Hydrological variables used in Principal Components Analysis (PCA) with descriptions and abbreviations used in Figures and text. Set 1= 47 hydrological variables, Set 2= 32 Indicators of Hydrologic Alteration (IHA) variables and Set 3 = PCA redundancy reduction variables.**

Hydrological variable	Description	Abbreviation	Set 1	Set 2	Set 3
September median	Monthly median flow	Sep	✓	✓	
October median	Monthly median flow	Oct	✓	✓	
November median	Monthly median flow	Nov	✓	✓	
December median	Monthly median flow	Dec	✓	✓	
January median	Monthly median flow	Jan	✓	✓	
February median	Monthly median flow	Feb	✓	✓	
March median	Monthly median flow	Mar	✓	✓	
April median	Monthly median flow	Apr	✓	✓	
May median	Monthly median flow	May	✓	✓	
June median	Monthly median flow	Jun	✓	✓	
July median	Monthly median flow	Jul	✓	✓	
August median	Monthly median flow	Aug	✓	✓	
1-day min	Annual 1-day minimum flow	1min	✓	✓	
3-day min	Annual 3-day minimum flow	3min	✓	✓	
7-day min	Annual 7-day minimum flow	7min	✓	✓	✓
30-day min	Annual 30-day minimum flow	30min	✓	✓	
90-day min	Annual 90-day minimum flow	90min	✓	✓	
1-day max	Annual 1-day maximum flow	1max	✓	✓	
3-day max	Annual 3-day maximum flow	3max	✓	✓	
7-day max	Annual 7-day maximum flow	7max	✓	✓	✓
30-day max	Annual 30-day maximum flow	30max	✓	✓	
90-day max	Annual 90-day maximum flow	90max	✓	✓	
Base flow	7-day minimum discharge divided by the mean annual daily discharge	BF	✓	✓	✓
Date min flow	Julian date of annual minimum flow	Dmin	✓	✓	
Date max flow	Julian date of annual maximum flow	Dmax	✓	✓	
Low pulse occurrences	Number of low pulses	LP#	✓	✓	
Low pulse duration	Duration of low pulses	LPD	✓	✓	✓
High pulse occurrences	Number of high pulses	HP#	✓	✓	
High pulse duration	Duration of high pulses	HPD	✓	✓	
Rise rate		RR	✓	✓	✓
Fall rate		FR	✓	✓	✓
Reversals		Rev	✓	✓	
Mean 7-day prior season		Mean7	✓		
Mean 30-day prior season		Mean30	✓		
Mean 90-day prior season		Mean90	✓		
Mean 180-day prior season		Mean 180	✓		
Max 7-day prior season		Max7	✓		
Max 30-day prior season		Max30	✓		
Max 90-day prior season		Max90	✓		
Max 180-day prior season		Max180	✓		
Min 7-day prior season		Min7	✓		
Min 30-day prior season		Min30	✓		
Min 90-day prior season		Min90	✓		
Min 180-day prior season		Min180	✓		
DWF		DWF	✓		
6-month Q10		Q10	✓		
6-month Q95		Q95	✓		

**Table 2 Summary of Principal Components Analysis output a) Cumulative percentage of variance explained by the first four PC axes and their eigenvalues for the spring and autumn sampling periods and for each of the three sets of hydrological variables used (b) PCA axes 1 and 2 scores for the six hydrological variables identified by the redundancy minimisation approach (See Table 1 for abbreviations and definitions of hydrological variables)**

(a)

Season/Variable set (no. of indices)	Autumn set1 (47)	Autumn set2 (32)	Autumn set3 (6)	Spring set1 (47)	Spring set2 (32)	Spring set3 (6)
Axis 1	45.30%	48.90%	52.70%	48.40%	47.30%	54.80%
<i>EIG</i>	0.45	0.49	0.53	0.48	0.47	0.55
Axis 2	57.30%	59.40%	76.30%	62.20%	62.00%	77.70%
<i>EIG</i>	0.12	0.11	0.24	0.14	0.15	0.23
Axis 3	65.50%	66.00%	88.00%	68.00%	68.40%	88.60%
<i>EIG</i>	0.81	0.07	0.12	0.06	0.06	0.11
Axis 4	69.80%	70.40%	95.20%	72.20%	72.50%	94.90%
<i>EIG</i>	0.04	0.04	0.07	0.04	0.04	0.06

(b)

Index	Autumn set1		Autumn set2		Autumn set3		Spring set1		Spring set2		Spring set3	
	Axis1	Axis2	Axis1	Axis2	Axis1	Axis2	Axis1	Axis2	Axis1	Axis2	Axis1	Axis2
BF	-0.27	0.49	-0.37	-0.80	-0.44	-0.80	-0.25	0.75	-0.13	-0.82	-0.29	0.92
FR	-0.85	0.15	-0.87	-0.05	-0.94	-0.02	-0.87	0.02	-0.85	-0.20	-0.90	0.08
LPD	-0.36	-0.21	-0.32	0.41	-0.40	0.61	-0.40	-0.37	-0.46	0.29	-0.63	-0.27
RR	0.79	-0.17	0.81	0.06	0.89	0.04	0.84	-0.01	0.83	0.18	0.88	-0.04
7max	0.77	-0.35	0.83	0.37	0.81	0.29	0.81	-0.26	0.78	0.47	0.81	-0.29
7min	0.83	0.11	0.79	-0.55	0.69	-0.56	0.76	0.60	0.86	-0.45	0.74	0.60

**Table 3 Cumulative percentage of variance cumulatively explained by each of the first four DCA axes and their eigenvalues for autumn and spring sampling periods and for family- and species-level macroinvertebrate data**

Season/Taxonomic resolution	Axis 1	<i>EIG</i>	Axis 2	<i>EIG</i>	Axis 3	<i>EIG</i>	Axis 4	<i>EIG</i>
<i>Autumn Family</i>	8.4%	0.2004	12.4%	0.0971	15.7%	0.0791	18.5%	0.0670
<i>Autumn Species</i>	4.2%	0.3262	6.2%	0.1553	7.8%	0.1263	9.4%	0.1191
<i>Spring Family</i>	8.3%	0.1958	12.6%	0.1052	15.9%	0.0789	19.1%	0.0757
<i>Spring Species</i>	4.3%	0.3170	6.7%	0.1787	8.8%	0.1578	10.8%	0.1438



**Table 4 (a) Pearson's correlation coefficients between autumn and spring PCA axis 1 sample scores for set 1-3 hydrological variables and family level (LIFE F) and species level (LIFE S) LIFE scores, (b) Stepwise multiple linear regressions model output for autumn and spring family level (LIFE F) and species level (LIFE S) LIFE scores using the 6 hydrological variables identified to minimise redundancy and including PCA axis 1 sample scores (c) Pearson's correlation coefficient between DCA axis 1 sample scores and autumn and spring LIFE S and LIFE F scores**

(a)

Season/Variable set	No. of variables	LIFE S	LIFE F
<i>Autumn set1</i>	47	0.135**	0.277**
<i>Autumn set2</i>	32	0.149**	0.296**
<i>Autumn set3</i>	6	0.127**	0.262**
<i>Spring set1</i>	47	0.235**	0.318**
<i>Spring set2</i>	32	0.220**	0.317**
<i>Spring set3</i>	6	0.222**	0.288**

(b)

AUTUMN	Adjusted R sq.	F	No. of samples	Most influential predictor variables
<i>LIFE F</i>	0.084	101.927***	1103	(+)7 day min
<i>LIFE S</i>	0.063	75.603***	1103	(+)Axis1
SPRING	Adjusted R sq.	F	No. of samples	Most influential predictor variables
<i>LIFE F</i>	0.088	102.949***	1051	(+)Axis1
<i>LIFE S</i>	0.041	45.718***	1051	(+)Fall rate

\*\* $p < 0.01$   
 \*\*\* $p < 0.001$

(c)

Season/Taxonomic resolution	LIFE S	LIFE F
<i>Autumn Family</i>	-0.776**	-0.830**
<i>Autumn Species</i>	-0.855**	-0.733**
<i>Spring Family</i>	-0.746**	-0.805**
<i>Spring Species</i>	-0.772**	-0.665**

## List of Figures

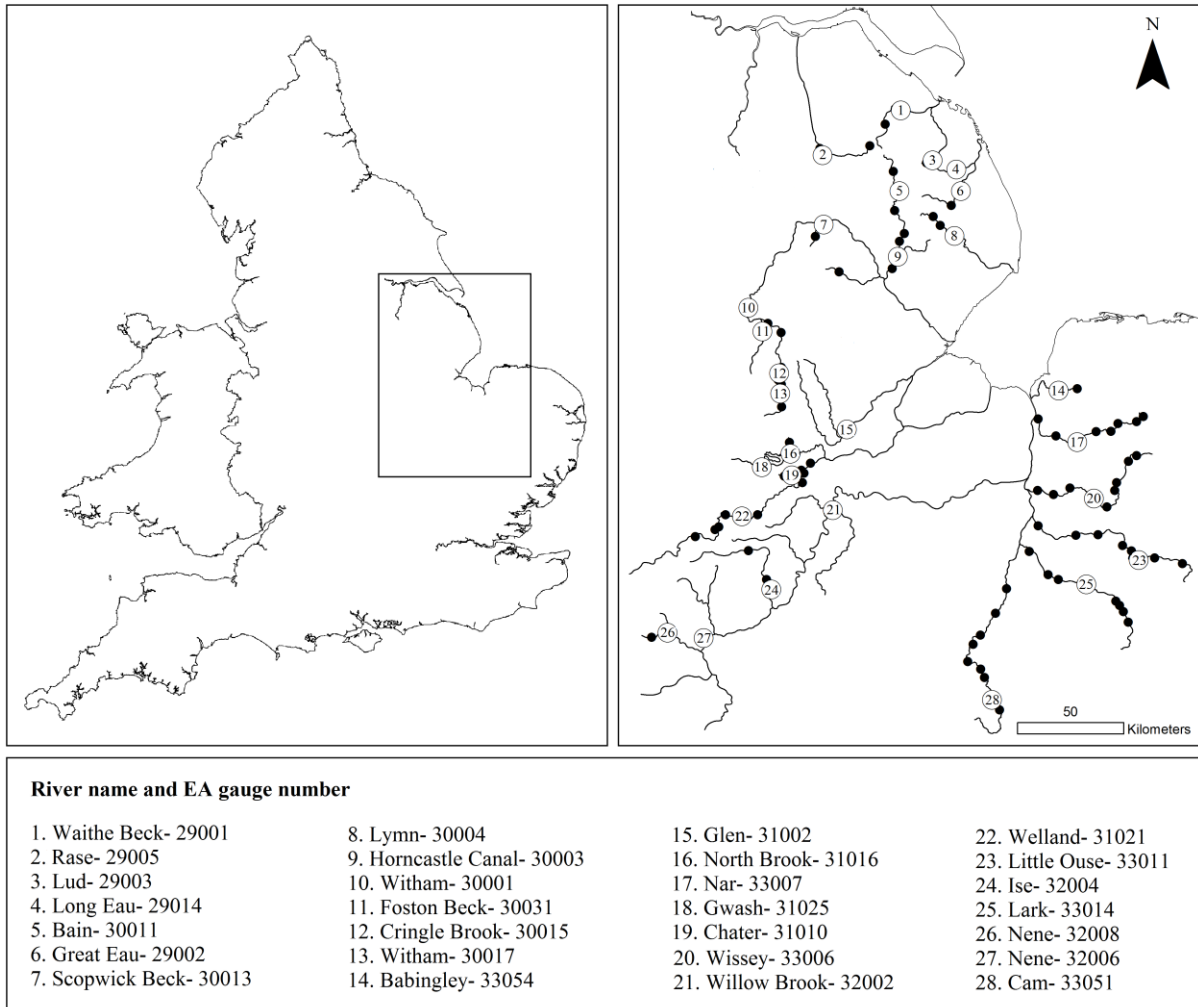


Fig. 1 Map of England and Wales showing locations of the 28 Environment Agency gauging stations used in the macro-scale investigation (numbered) and the 88 Environment Agency biomonitoring sites (indicated by ●)

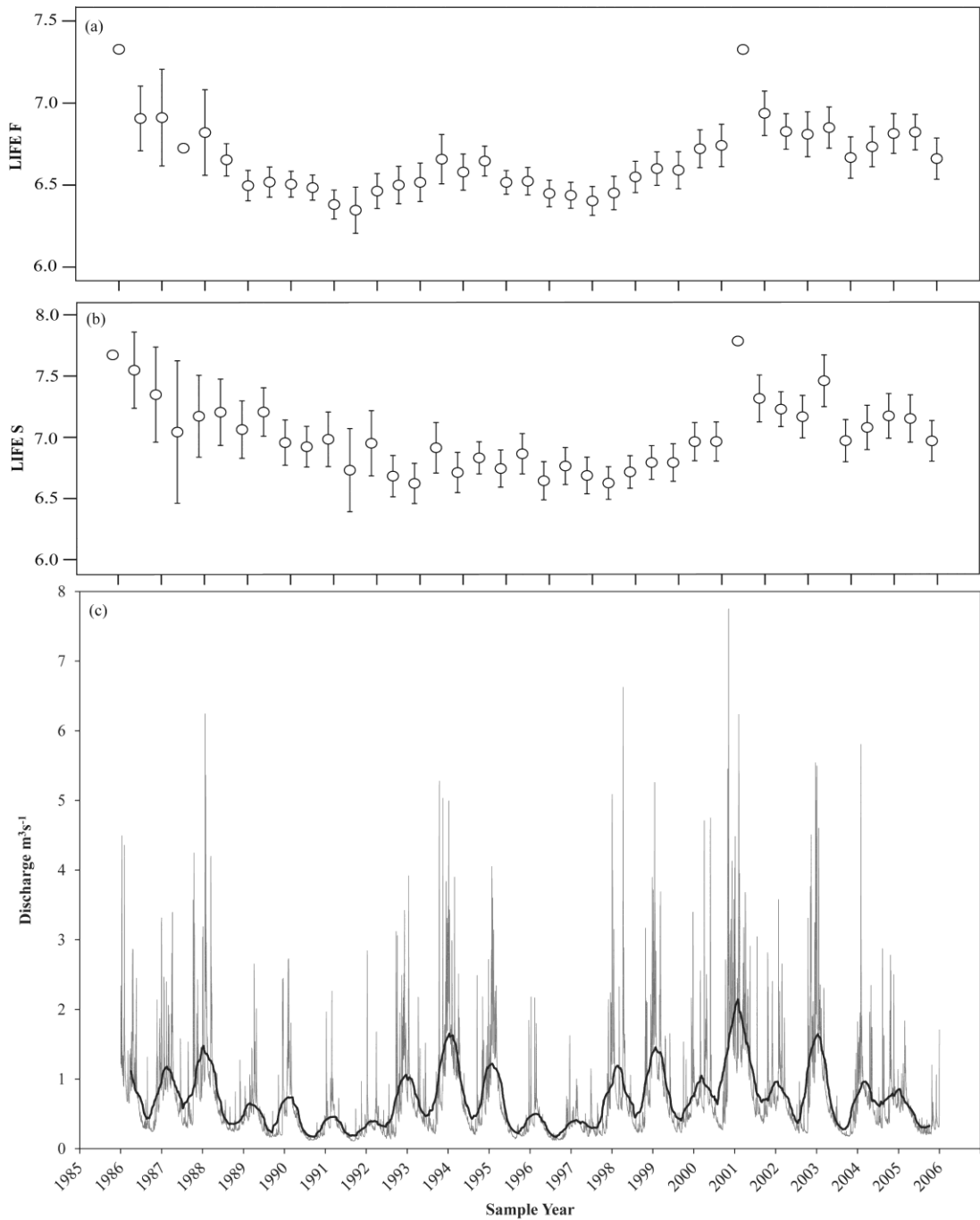


Fig. 2 Time series plots (1986-2005) for: (a) mean (+/- 95% confidence intervals) Family level LIFE scores (LIFE F), (b) mean (+/- 95% confidence intervals) Species level LIFE scores (LIFE S), (c) mean long term hydrograph for the 28 Environment Agency gauges and the six month rolling mean discharge plot (red line). Note - 95% confidence intervals have been removed from 1986 and 2001 on LIFE F and LIFE S plots due to the high level of variance recorded due to reduced sample number during these years.

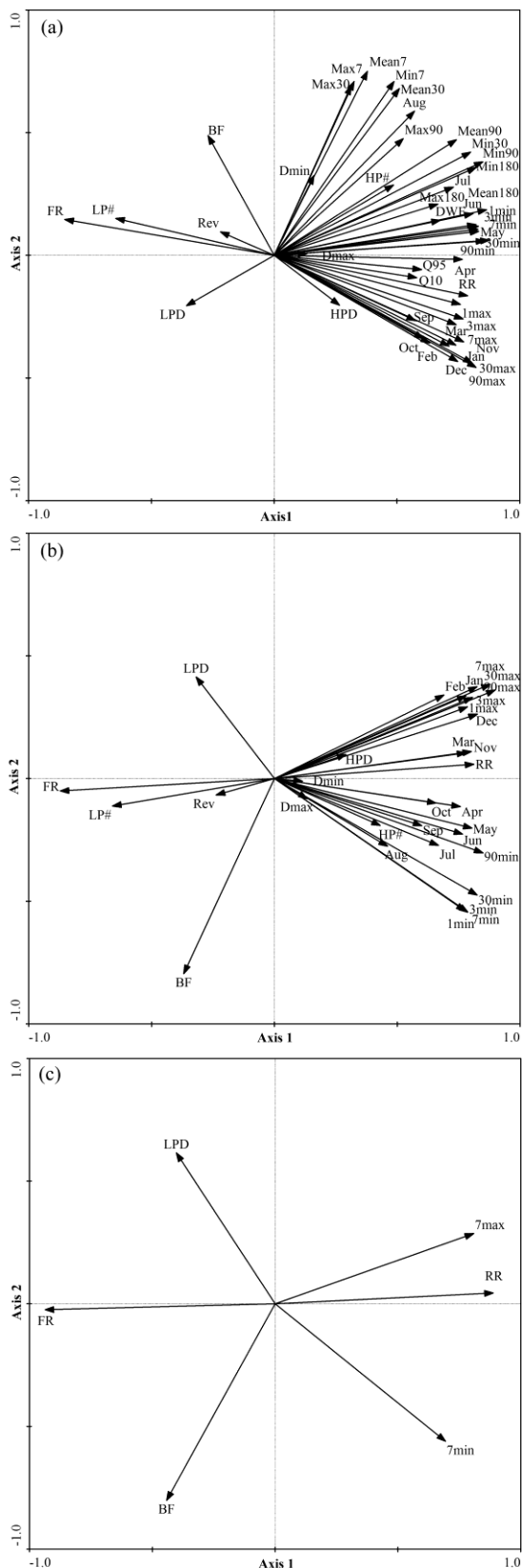


Fig. 3 Principal Components Analysis plots of hydrological variables for the autumn sampling period for (a) Set 1 - 47 hydrological variables, (b) Set 2 = 32 Indicators of Hydrologic Alteration (IHA) variables, and (c) Set 3 - PCA redundancy reduction variables (see Table 1 for abbreviations)

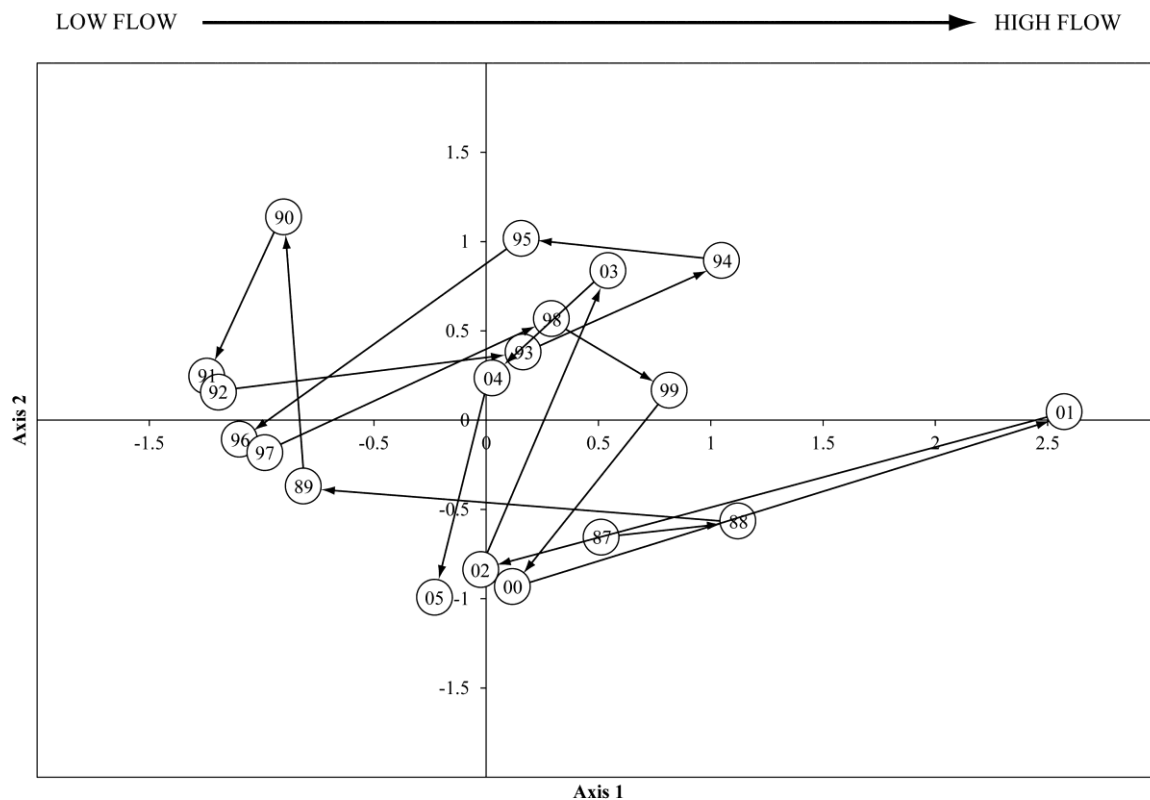


Fig. 4 Principal Components Analysis plot of the centroid for individual years (axes 1 and 2), generated using Set 3 hydrological variables (redundancy reduction), for all 28 Environment Agency flow gauges used in the study. Arrows show change between individual years. The first axis reflects variation in the magnitude of flow from low-flow (drought) to high flow along axis 1.

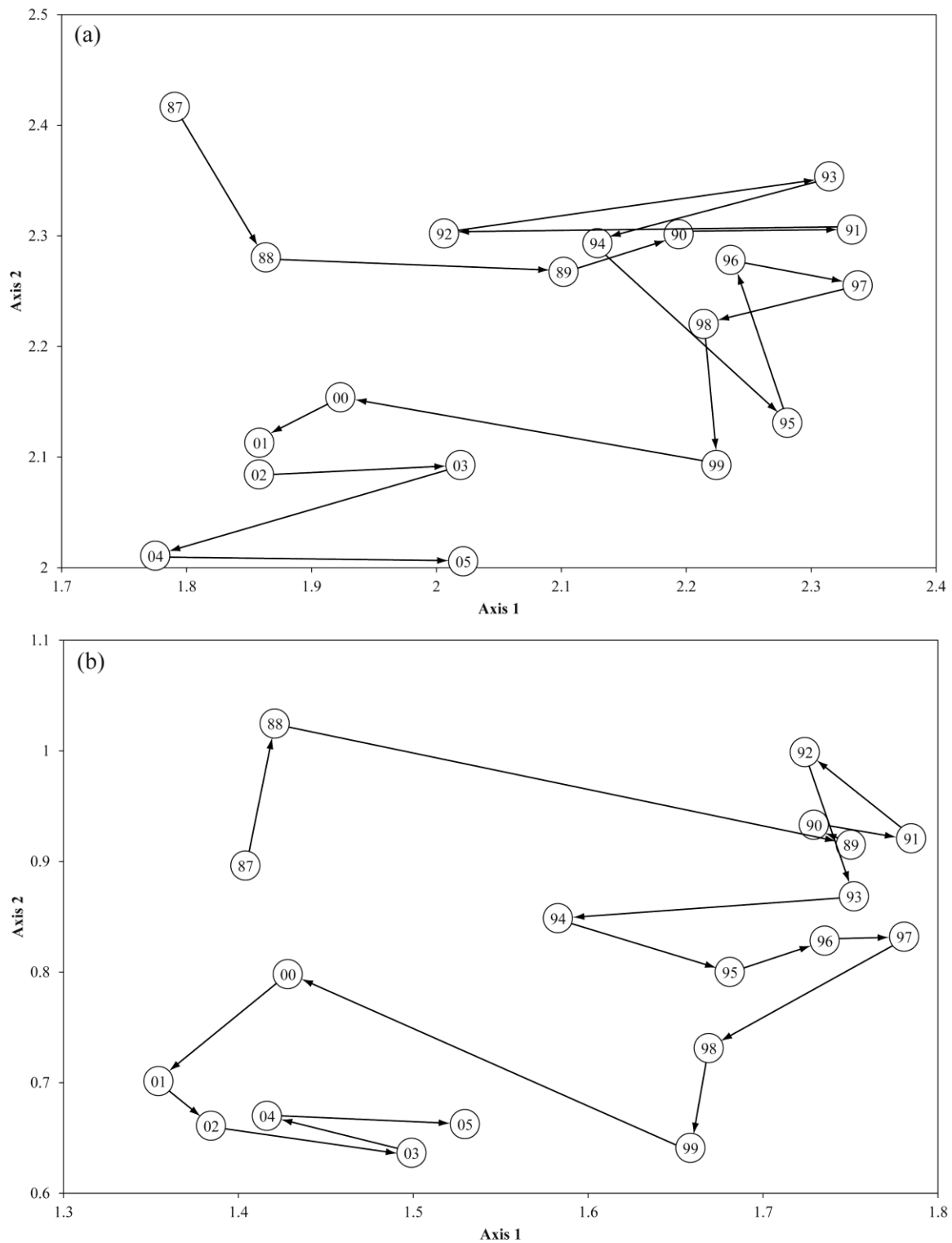


Fig. 5 Detrended Correspondence Analysis plot of the centroid for individual years for the autumn macroinvertebrate sampling period for: (a) macroinvertebrate community data at the family level, and (b) macroinvertebrate community data recorded at the genus/species level. Arrows show change between individual years.