**Ponding in intermittent streams: a refuge for lotic taxa and a habitat for newly colonising taxa?**

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**Abstract**

Intermittent rivers are temporally dynamic, shifting between lotic, lentic (ponding) and dry habitat phases, yet almost all research effort has focussed on the lotic phase, with limited research attention on the lentic and dry phases. Information regarding the biological diversity of the lentic phase is vital to quantify the total aquatic biodiversity, their use as flow refugia, and the long-term conservation and management of intermittent rivers. In this study, we compared the diversity and composition of macroinvertebrates from perennial, intermittent and ponded sites in two intermittent rivers in the United Kingdom. We examined whether instream ponding provided refugia for lotic taxa and a habitat for newly colonising taxa. A total of 129 taxa (perennial - 86, intermittent - 82, ponding - 78) were recorded. Instream ponds were found to support heterogeneous communities compared to flowing sites. Twenty-two percent of taxa were recorded only from ponded sites, many of which were lentic specialists, while 38% of taxa persisted in instream ponds after flow had ceased. Results from this study highlight that instream ponds provide an important flow refuge for macroinvertebrates including rheophilic taxa, which move into instream ponds when channels become longitudinally disconnected, and makes a significant contribution to aquatic diversity in intermittent rivers, providing suitable habitat for newly colonising taxa. Aquatic diversity in intermittent rivers may have been underestimated historically, failing to acknowledge the ecological contribution of the lentic phase. Incorporating the ponding phase alongside the lotic phase will ensure the total aquatic biodiversity of intermittent rivers is quantified and effective biodiversity conservation and management strategies are employed.

**Key Words:** beta-diversity; biodiversity conservation; community composition; instream ponds; macroinvertebrates; ponding phase.

## 1. Introduction

International interest in intermittent freshwater ecology has increased significantly in recent years (Leigh and Datry, 2016; White et al., 2018). Intermittent rivers are lotic freshwater ecosystems that experience a predictable or unpredictable loss of surface flow (Stubbington et al., 2017), are common across all continents, and are estimated to comprise more than 50% of river networks globally (Skoulikidis et al., 2017). Their occurrence is likely to increase in the future, in areas where there will be increased climatic drying (due to decreased precipitation and increased temperatures; Doll et al., 2012; Garcia et al., 2017), increases in drought frequency and intensity (Prudhomme et al., 2014), and/or increased anthropogenic water abstraction and impoundment resulting in previously perennial rivers experiencing intermittent flow (Datry et al., 2014). Given the increasing prevalence of intermittent freshwaters, information regarding their ecological diversity and functioning will be vital to the long-term conservation and management of freshwater biodiversity.

Intermittent rivers are temporally dynamic, shifting between lotic, lentic (ponding) and terrestrial habitat phases at a network scale (Gallart et al., 2016), yet almost all research effort has focussed on the lotic phase (Leigh et al., 2016; Soria et al., 2017) with the biological communities in the lentic and dry phases almost entirely ignored (Bonada et al., 2006; Chester and Robson, 2011; Boersma et al., 2014 and; Corti and Datry, 2015). Further, most research on intermittent river ecology has concentrated on arid and semi-arid regions (Leigh et al., 2015; Karaouzas et al., 2018) with limited research attention directed towards intermittent rivers in temperate regions (Stubbington et al., 2017). Flow cessation has been shown to be a key determinant of aquatic macroinvertebrate diversity and composition, with previous studies finding intermittent rivers typically supporting lower macroinvertebrate diversity than perennial rivers (Williams, 2006; Garcia-Roger et al., 2011; Soria et al., 2017). In addition, flow cessation acts as a ramp disturbance increasing in strength and spatial extent through time (Lake, 2003; Datry, 2012), and as a result lotic taxonomic richness decreases as flow intermittency (proportion of year without surface water) increases (Datry et al., 2013). However, aquatic taxonomic richness within intermittent rivers may be significantly underestimated and incomplete, as previous studies have failed to include macroinvertebrate diversity from instream ponds (focussing only on the lotic phase) that form during stream drying (Leigh et al. 2016; Stubbington et al. 2017). The ponding phase may make a contribution to aquatic diversity in intermittent rivers as macroinvertebrate specialists of lentic habitats may colonise the instream ponds from proximal freshwater habitats (Stubbington et al., 2017). In addition, terrestrial macroinvertebrates colonising the dry river bed during the dry phase of intermittent rivers may also provide a significant source of biodiversity for intermittent rivers, and by incorporating the lotic, lentic and dry phase biodiversity, a more accurate representation of the total biodiversity that intermittent rivers support can be obtained (Corti and Datry, 2015).

Aquatic macroinvertebrates present in intermittent rivers during the lotic phase (generalists and specialists) often rely on a combination of traits that promote resistance and/or resilience based strategies to survive streambed drying (Stanley et al., 1994; Fritz and Dodds, 2004). Resilience based strategies refer to an individual's capacity to recolonise after the disturbance and re-establish populations similar to pre-disturbance levels (Bogan et al., 2015). Resistance based strategies refer to the ability of an individual to survive/persist through the dry phase *in situ* (e.g., biological adaptations or refugia). Refuge habitat can play an important role in the persistence of aquatic flora and fauna, reducing the ecological impacts of streambed drying (Chester and Robson, 2011). While some macroinvertebrate species have specific adaptions to survive streambed drying (e.g., desiccation resistant eggs; Williams 2006), many aquatic macroinvertebrate taxa in intermittent rivers in Mediterranean and temperate regions are typically generalist and ubiquitous, and are not well adapted to streambed drying (Datry et al., 2013; Kalogianni et al., 2017), often relying on refugia to persist in intermittent freshwaters. Aquatic macroinvertebrate taxa present in intermittent rivers during the lotic phase may use instream ponds as a refuge to survive and persist through flow cessation (Sheldon et al., 2010; Boersma et al., 2014). When compared to other flow cessation refugia (e.g., damp sediment, leaf litter, underneath stones), perennial and semi-permanent instream ponds were the most important refuge for aquatic macroinvertebrates in intermittent rivers from arid regions (Sheldon et al., 2010;

Chester and Robson, 2011). However, there is a paucity of information on refugia in temperate intermittent river systems, particularly instream ponds. Knowledge of the functioning and capacity of refuges to reduce the effects of streambed drying is increasingly important for ecological conservation in intermittent freshwaters, particularly in regions where freshwater intermittency is likely to increase (Chester and Robson, 2011; Costelloe and Russell, 2014).

While the conceptual and theoretical importance of the ponding phase (as a refuge and/or a site for colonisation) has been recognised (Boulton 2003; Chadd et al., 2017), to our knowledge, this is the first study empirically examining macroinvertebrate diversity and community composition during the lotic and lentic phases in an intermittent river network in a temperate region. This study sought to: (1) quantify the contribution of instream ponding to aquatic macroinvertebrate diversity within intermittent rivers and; (2) examine whether instream ponding provided an area of refuge for lotic taxa during stream desiccation and a habitat for newly colonising taxa. It is hypothesised that (1) ponded sites will be colonised by lentic specialist macroinvertebrate taxa not typically found during the flowing phase, and (2) ponded sites will support macroinvertebrate taxa found during the flowing phase (and thus provide a flow refuge).

## 2. Material and methodology

### *2.1 Study area*

This study was conducted on the Manifold and Hamps Rivers in the English Peak District, United Kingdom (UK; Fig. 1); two meandering rivers characterised by pool-riffle topography. The catchment area of the Manifold and Hamps Rivers is approximately 148.5 km$^2$. The study reaches are situated between 280 m and 150 m above sea level. The mean channel width was larger in the Manifold River (12.2 m, range 10.0–15.4 m) than the Hamps River (7.8 m, range 6.8–10.1 m). Both catchments are dominated by grassland (82.7 %), woodland (5.8 %), and moorland (3.4 %) with substantial riparian vegetation (NRFA, 2017). The Manifold and Hamps catchments are adjacent with comparable meteorological conditions with an average annual precipitation of 964mm, and an average annual minimum and maximum temperature of 5.3 ºC and 11.5 ºC, respectively (1981-2010, UK Meterological Office, 2017). Both rivers flow through gravel drift deposits underlain by carboniferous limestone. Boulder to gravel size clasts dominate both riverbeds.

Approximately 10 km of the mid reaches of the Manifold and Hamps Rivers are intermittent (Fig. 1) and drying occurs seasonally (typically from early June to September) due to reduced precipitation and water flowing through underground karst caves/channels and resurfacing downstream (Fig. 1). In 2016, when the study was undertaken, drying occurred irregularly between June and October as there were sporadic periods of rainfall which caused the intermittent section to flow on a number of occasions, typically for < 48 hours. Upstream and downstream of the intermittent section, the flow regime is perennial. The downstream perennial section of the Manifold River has a mean discharge of 3.58 $m^3/s$ (1968-2016; NRFA 2017). During stream desiccation, small ponds form on the dry bed (in the deeper areas of the river channel where river water persisted) of the Manifold River along the downstream end of the intermittent river channel (Fig. 1). The instream ponds were discrete (disconnected from ponds upstream or downstream), typically < 50 $m^2$ (although one pond had an area of 216 $m^2$), shallow (depth < 40 cm), dominated by bedrock or gravel size clasts and had limited aquatic macrophyte coverage. The instream ponds typically held water for ~1.5 months (developed in late June 2016 and dried in mid-August 2016) before drying completely (although 4 ponds contained water for the entire period of flow desiccation). Macroinvertebrates were sampled in the perennial section and during the flowing phase of the intermittent section from the 23rd - 25th May 2016. A total of 17 ponds were sampled for aquatic macroinvertebrates during the ponding phase in the intermittent section of the Manifold River on the 19$^{th}$ and 20$^{th}$ July 2016, after lotic conditions had ceased. All ponded sites were sampled downstream of the confluence of the two sub catchments (the Hamps River flows into the Manifold River approx. 5 km upstream of the ponded sites; Fig. 1).

### *2.2 Macroinvertebrate sampling*

A total of 3 reaches in the perennial section and 4 reaches in the intermittent section within the Manifold and Hamps Rivers were sampled for aquatic macroinvertebrates during the flowing phase (Fig. 1). Reach length was calculated as 10× the mean width of the active channel to ensure distinct aquatic mesohabitats (e.g. submerged macrophytes, overhanging vegetation, different substrate sizes and hydrological flows) were included. The active channel was defined as the area of inundated bed sediments between edges of terrestrial vegetation and abrupt changes in slope (Corti and Datry, 2015). At each reach, sampling took place along 3 transects, separated by 2 × mean wetted width from each reach (Corti and Datry, 2015). At each transect, 5 surber samples (size: 0.33 × 0.3m, sampling area: $0.1m^2$, mesh size: 1 mm) were taken, equidistantly spaced across the transect. Each surber sample was undertaken for 60 seconds.

All instream ponds were sampled using a standard sweep technique in the different mesohabitats present in the ponds (Hill et al., 2017). The length of time allocated to sample aquatic macroinvertebrates in each pond was proportional to its surface area, up to a maximum of 3 min (see Hill et al., 2015 for full methodology). The allocated sampling time was divided equally between the different mesohabitats present (i.e. open water, submerged macrophytes, and emergent macrophytes), although if one mesohabitat dominated sampling time was further divided to reflect this (Biggs et al., 1998). Immediately after sampling, macroinvertebrates were placed into plastic bags, preserved in 70% ethanol and taken to a laboratory for sorting and identification. Macroinvertebrate taxa were identified to species level wherever possible, although Pisidium and Sphaerium were identified to genus level, Physidae, Diptera larvae and Zonitidae were identified to family level and Hydrachnidia, Oligochaeta and Collembola were identified as such.

*2.3 Statistical analyses*

Macroinvertebrate data from the 5 surber samples along each transect were pooled to provide a measure of alpha diversity and community assemblage for each transect. The macroinvertebrate dataset was converted into a presence-absence matrix prior to analyses to reduce sampling bias. Gamma diversity is defined here as the total diversity within each sample group; perennial, intermittent and ponded. Alpha diversity is defined as the taxonomic richness within an individual sample site (transect). The Chao 2 estimator (in the vegan package; Oksanen et al., 2017) was used to estimate gamma diversity for perennial, intermittent and ponded sites. Differences between estimated gamma diversity from perennial, intermittent and ponded sites were considered significant if the 95% Confidence Intervals (95% CI) did not overlap. Rarefaction was undertaken to estimate taxonomic richness within perennial, intermittent and ponded sites for a given number of individuals, as sample size and sampling procedures can significantly affect the ecological diversity recorded (McCabe and Gotelli, 2000). The least abundant sample had 63 individuals; as a result, 63 individuals were randomly sampled from each perennial, intermittent and ponded site, and the rarefied species richness was recorded. Differences in taxonomic and rarefied alpha diversity between perennial, intermittent and ponded sites were examined using a one-way analysis of variance (one-way ANOVA in the stats package) test (preliminary examination of the diagnostic plots of a linear model of the macroinvertebrate dataset, indicated that the underlying assumptions of parametric tests were not violated). Pairwise comparisons using Tukey post-hoc tests (in the stats package) were undertaken to determine where significant differences among the perennial, intermittent and ponded sites occurred.

Beta-diversity is defined here as the variation in community composition between samples (perennial, intermittent and ponded sites) in a predefined geographic area (Koleff et al., 2003), in this case the Manifold and Hamps catchments. Non-Metric Multidimensional Scaling (NMDS: using the Sorensen dissimilarity) was undertaken (using the *metaMDS* function from the vegan package) to examine and visualise the variability in macroinvertebrate community assemblage between the perennial, intermittent and ponded sites. Differences in community assemblage among the perennial, intermittent and ponded sites was statistically examined using a permutational multivariate analysis of variance (PerMANOVA). Pairwise comparisons (with Bonferroni correction) were calculated to quantify where differences among perennial, intermittent and ponded sites occurred. To examine the heterogeneity of macroinvertebrate assemblages within perennial, intermittent and ponded sites, homogeneity of multivariate dispersions were calculated (using the *betadisper* function in the vegan package). Differences in the homogeneity of multivariate dispersions between the three groups were tested statistically using an ANOVA and pairwise Tukey HSD tests. To examine the underlying processes driving macroinvertebrate community heterogeneity in perennial, intermittent and ponded communities, total beta-diversity (Sorenson dissimilarity) of the macroinvertebrate communities from perennial, intermittent and ponded sites were partitioned into nestedness and turnover components using the function *beta.multi* from the package betapart (Baselga et al. 2010; Baselga et al., 2017). Macroinvertebrate assemblages are considered to be nested when sites with low taxonomic richness comprise subsets of sites with greater taxonomic richness, while species turnover reflects the replacement of taxa from one site to another (Hill et al., 2017). In addition, indicator value analysis was undertaken using the function *multipatt* in the indicspecies package (De Caceres and Jansen, 2016) to identify which taxa were characteristic of perennial, intermittent and ponded sites. All statistical analyses were performed in the R environment (R Development Core Team, 2016).

## 3. Results

### *3.1 Macroinvertebrate diversity (alpha and gamma)*

In total, 129 macroinvertebrate taxa from 58 families and 18 orders were recorded from the perennial (86 taxa), intermittent (82 taxa) and ponded (78 taxa) sites in the Manifold and Hamps Rivers. A total of 30 taxa were collected as single occurrences. Estimated gamma diversity was not significantly different between perennial (estimated gamma: 110.1 95%, Confidence Interval (CI): 85.7-134.6), intermittent (estimated gamma: 114.7 95%, CI: 82.2-147.1) and ponded sites (estimated gamma: 102.5, CI: 78.7-126.3) as the 95% confidence intervals overlapped (Table 1; Fig. 2a). Alpha diversity was significantly different (ANOVA df = 2, $F_{2, 38} = 7.64$, $p < 0.01$) between the perennial, intermittent and ponded sites (Fig. 2b). Post hoc Tukey tests found perennial sites had higher alpha diversity ($p < 0.01$) than ponded sites (Table 1; Fig. 2b). Similarities in alpha diversity were found between intermittent and ponded sites, and between perennial and intermittent sites ($p > 0.05$). No significant difference in rarefied taxon richness was recorded among perennial, intermittent and ponded sites ($p > 0.05$).

*3.2 Community heterogeneity (beta-diversity)*

The NMDS biplot demonstrates a clear separation between macroinvertebrate assemblages recorded from flowing (perennial and intermittent sites overlap) and ponded sites (Fig. 3a). The pairwise PerMANOVA tests found macroinvertebrate assemblages in the ponded sites were significantly different to macroinvertebrate assemblages from the perennial (PerMANOVA $R^2 = 0.41$, $p = 0.03$) and intermittent sites (PerMANOVA $R^2 = 0.38$, $p = 0.03$). No significant difference in macroinvertebrate community assemblage was recorded between perennial and intermittent sites (PerMANOVA $R^2 = 0.08$, $p = 0.2$). Multivariate dispersion (ANOVA df = 2, $F = 0.06$, $p = 0.93$) was similar among perennial (average median distance to group centroid: 0.31), intermittent (average median distance to group centroid: 0.31) and ponded (average median distance to group centroid: 0.32) sites (Fig. 3b).

A total of 28 taxa were only recorded from ponded sites (22% of total aquatic diversity), 12 in perennial sites (9% of total aquatic diversity) and 8 in intermittent sites (6% of total aquatic diversity) (see Table S1 for the full list of macroinvertebrate taxa recorded only from perennial, intermittent or ponded sites). When considering only intermittent and ponded sites, 33% of taxa were lost (38 taxa were recorded in intermittent sites in May 2016, but not from ponded sites in July 2016), as the intermittent section of the Manifold River transitioned from the flowing to the ponding phase (Fig. 4). This included taxa from the Ephemeroptera (particularly species of Heptageniidae e.g., *Rhithrogenia semicolorata*, *Heptagenia sulphurea*), Plecoptera (e.g., *Isoperla grammatica*) and Trichoptera (e.g., *Hydropsyche siltalai*, *Rhyacophila dorsalis*) orders (Fig. 4). However, 38% of taxa from the intermittent section persisted in the instream ponds after flow ceased (44 taxa were recorded in the intermittent sites in May 2016 and the ponded sites in July 2016, predominantly from the Gastropoda, Coleoptera, Diptera, Ephemeroptera and Trichoptera orders; Fig. 4), and 29% of taxa (34 taxa) were recorded only from the ponded sites (compared to intermittent sites), predominantly from the Hemiptera (all 5 species of Corixidae were recorded only from ponds), Coleoptera (particularly species of Haliplidae, Helophorus and Dytiscidae) and Gastropoda macroinvertebrate groups (Fig. 4). The top five macroinvertebrate taxa identified as indicator species for perennial, intermittent and ponded sites are presented in Table 2 (see Table S2 for the full list of statistically significant indicator taxa).

Macroinvertebrate communities within perennial (0.61), intermittent (0.67) and ponded (0.77) sites demonstrated high beta diversity based on the Sorensen dissimilarity index. Differences in macroinvertebrate community assemblages within pond sites and transects within perennial and intermittent reaches could almost entirely be explained by species turnover rather that spatial patterns of nestedness (*perennial* - species turnover: 85%, nestedness: 15%, *intermittent* - species turnover: 87%, nestedness: 13%, and ponded - species turnover: 94%, nestedness: 6%; Fig. 5). At a reach scale, intermittent reaches on average supported less diversity than perennial reaches, and 82% of taxa recorded in the intermittent section were also recorded in the perennial sites, indicating that intermittent sites were nested subsets of perennial sites.

## 4. Discussion

### *4.1 Contribution of instream ponding to total aquatic diversity and as a site for newly colonising taxa*

This study highlighted that gamma diversity was similar among perennial, intermittent and ponded sites. Similarly, comparable gamma diversity among intermittent and perennial sites was also recorded from the Lathkill River in the same study region (Wood et al. 2005). However, broad-scale studies and meta-analyses of intermittent rivers have found that intermittent reaches typically support a lower macroinvertebrate diversity than perennial reaches (Datry et al., 2013; Leigh and Datry, 2016; Soria et al., 2017). Similar alpha and gamma diversity recorded between perennial and intermittent reaches in this study may reflect the timing of sampling and the spatial arrangement of temporary and perennial reaches. Sampling was undertaken in May (just prior to the initiation of stream drying), when the river had been flowing for a significant length of time (typically flows from September to June). Therefore, there was substantial opportunity for macroinvertebrate taxa to colonise intermittent reaches, and support a comparable diversity to perennial reaches. The Manifold and Hamps Rivers are characterised by mid-reach drying, which provides upstream and downstream sources of colonisation for the intermittent section and may explain the similar diversity recorded among perennial and intermittent reaches in this study (Storey and Quinn, 2008). Alpha diversity was lower in the ponded sites compared to the intermittent and perennial sites. Rosset et al., (2016) also found alpha-diversity to be lower in intermittent ponds than lotic ecosystems. This may reflect (1) the significantly shorter water residence times (reducing opportunities for colonisation and the development of more complex communities) in the ponded sites than in the intermittent and perennial sites, and (2) the hydrological isolation of ponded sites and the hydrological connectivity among intermittent and perennial reaches (increasing opportunities for colonisation of intermittent reaches from upstream perennial reaches; Rosset et al., 2016).

Macroinvertebrate communities from the ponding phase were significantly different (based on NMDS and PerMANOVA analysis) to communities recorded from the perennial and intermittent sites (this was also recorded within intermittent streams in Northern California; Bonada et al., 2006), demonstrating that instream ponds make a significant contribution to total aquatic diversity in temperate intermittent rivers and provide a habitat for newly colonising taxa (accept hypothesis 1). During the transition of the intermittent river from the lotic to lentic phase in this study, many rheophilic taxa were replaced by taxa more commonly found in lentic habitats, particularly actively dispersing taxa such as Hemiptera (Corixidae) and Coleoptera (Dytiscidae/Helophoridae). Similar results were reported from the Fuirosos catchment in Spain where rheophilic taxa were replaced by lentic taxa such as veliids and dytiscids (Acuna et al., 2005). This may be due to: (i) nearby temporary lentic waterbodies drying causing the active dispersal of lentic macroinvertebrate taxa to instream ponded sites, that may hold water for longer than the nearby temporary waterbodies (the instream ponds act as a refuge for a wider area than just the river channel); (ii) conditions may be favourable for lentic macroinvertebrate taxa in instream ponds and competition for resources may be lower as they are newly developed habitats in the summer (although Acuna et al., 2005 found predation pressure to be high in instream ponds); (iii) stochastic processes influencing colonisation and dispersal and; (iv) stream desiccation causing a concentration of macroinvertebrate taxa in the ponds (Acuna et al., 2005). A reduced wetted area may potentially increase the likelihood of sampling the less commonly occurring taxa, compared to a fully flowing river where there is greater habitat area and potentially a reduced likelihood of sampling the less commonly occurring taxa. However, the significant differences in community composition demonstrated in this study between the lotic and lentic phases is unlikely to be the result of sampling issues. Many of the taxa recorded in the ponded sites, but not the intermittent flowing sites were lentic specialists (e.g., Hemiptera - *Sigara dorsalis, Sigara nigrolineata, Calicorixa praeusta, Corixa punctata* and Coleoptera - *Hesperocorixa sahlberghi, Helophorus grandis, Helophorus dorsalis, Haliplus ruficollis*), and are rarely recorded within lotic systems (Friday, 1988; Savage, 1989).

We speculate that several Gastropoda taxa (e.g., *Armiger crista, Valvata cristata, Lymnaea palustris* and *Lymnaea stagnalis)* were recorded only from the ponded sites. This may reflect the transport of eggs of lentic specialist gastropods from nearby lentic waterbodies to the instream ponds by vector species (such as wildfowl), which has been demonstrated in other studies to be a key dispersal strategy for Gastropoda (van Leeuwen et al., 2013; van Leeuwen and van de Velde, 2012). A number of Gastropoda demonstrate specific adaptations to survive in temporary habitats, including desiccation resistant eggs, a diapause state during the dry period, and a protective epigram over the opening during the dry phase (Nicolet et al., 2004; Williams, 2006). Given that the instream ponds examined in this study occur predictably every year, in a similar location on the streambed, some Gastropoda species that have colonized previously may have the specific adaptations required to survive the ponding phase in intermittent rivers *in situ,* and may have further adapted their life cycle to fit the hydrological cycle in instream ponds.

*4.2 The ponding phase as a refuge for lotic taxa*

In total, the instream ponds supported 38% of taxa that were recorded from the intermittent flowing sites, indicating that instream ponds can provide suitable refuge habitat from stream drying (accept hypothesis 2). Similarly, Chester and Robson (2011) and Fritz and Dodds (2004) found instream perennial ponds to be the most important refuge habitat in intermittent rivers. As well as macroinvertebrates, instream ponds have been shown to be an important refuge for fish during stream cessation (Magoulick and Kobza, 2003; Arthington, 2005; Marshall et al., 2016). This study was undertaken in a temperate region where the ponds maintained water for at least 1.5 months (four ponds held water for the entire stream desiccation period and occur predictably every year), so there was sufficient water residence time to support some taxa that were unable to disperse elsewhere in the stream channel between wet phases. However, in Mediterranean regions, where residence times of water in instream ponds is potentially much shorter (drying before the onset of the wet phase) and less predictable, the capacity for ponds to provide a refuge habitat may be significantly reduced.

A significant number of macroinvertebrate taxa in intermittent rivers in temperate and Mediterranean regions are often generalist (Datry et al., 2013) and not well adapted to flow cessation (although some macroinvertebrate taxa have adapted to streambed drying; Williams 2006). As a result, there is often a reliance on refuge habitat to persist in intermittent rivers, particularly by taxa that employ resistance-based strategies to survive streambed drying *in situ*, or those who have not reached their adult life stages before the onset of stream cessation (Sheldon et al., 2010; Beorsma et al., 2014). While river bed drying can prevent macroinvertebrate fauna from completing their aquatic life stage, instream ponds may provide an extended wetted habitat period in intermittent rivers, enabling some intermittent stream taxa (e.g., Diptera) to complete their juvenile aquatic life stage (although pond hydroperiod length must exceed remaining invertebrate development time) and emerge as adults (Drummond et al., 2015). However, instream ponds may not provide a suitable refuge habitat for all macroinvertebrate taxa. In this study, a number of Plecoptera (5 species, particularly from Perlidae and Perlodidae families), Ephemeroptera (9 species, particularly from the Heptageniidae family) and Trichoptera (15 species, noticeably from the Hydropsyche family) taxa that were recorded from the intermittent sites were not recorded from the ponded sites. Many of these taxa are rheophilic, requiring stream flow to complete their life histories (e.g., to obtain oxygen and food; Mackay and Wiggins, 1979; Elliot et al., 1988; Edington and Hildrew, 1995; Wallace et al., 2003; Walters and Post, 2011; Lancaster and Downes, 2013), which instream ponds do not provide, potentially resulting in the loss of some taxa during the transition from flowing to lentic conditions. However, the loss of some Ephemeroptera, Plecoptera and Trichoptera taxa during the ponding phase may also reflect their life cycle, typically emerging as flying adults during the summer months (Wise, 1980; Petersen et al., 1999; Poepperl, 2000; Greenwood et al., 2001; Dobrin and Giberson, 2003; Feeley et al., 2009), and therefore not requiring flow refugia. When flow resumes these taxa are likely to lay eggs in the intermittent reaches, and recolonise intermittent reaches via drift from upstream perennial reaches; Vander Vorste et al., 2016).

### *4.3 Conservation implications*

Given that almost all intermittent studies have focused only on the lotic phase of intermittent rivers, the total aquatic diversity in intermittent rivers may have been significantly underestimated historically. Intermittent rivers are often viewed as species-poor compared to perennial rivers (e.g. Storey and Quinn, 2008; Garcia-Roger et al., 2011; Soria et al., 2017), but this is likely to be inaccurate as previous studies have failed to consider the contribution of the ponding phase to intermittent river diversity. In addition, there is significant taxonomic richness supported during the dry phase, which has also been overlooked in intermittent river research (although see Corti and Datry, 2015). The likely underestimation of aquatic diversity in some intermittent systems may be a key factor driving the limited conservation attention received by intermittent rivers, with lotic conservation efforts almost exclusively focused on perennial rivers (Leigh et al., 2016; Skoulikidis et al., 2017). The limited conservation efforts directed towards intermittent rivers focus on protecting/maintaining diversity from the lotic phase (Skoulikidis et al., 2017). An acknowledgement of the faunal diversity from all phases of intermittent rivers and streams is required to ensure that the total biodiversity and conservation value is identified, and the development of more accurate and effective conservation strategies, particularly regarding the persistence of particular target species (Vardakas et al., 2017) or the conservation of the maximum possible diversity within intermittent systems.

There is a need to preserve and maintain instream ponds in an intermittent river network as they are often the only suitable refugium for fully aquatic taxa and fish during streambed drying (Sheldon et al., 2010; Marshall et al., 2016). The development of instream ponds during the dry phase (that will recurrently appear every year during drying) may significantly increase the resilience and persistence of biological communities to drying and drought episodes. However, given the wide refuge requirements of freshwater biota in intermittent rivers, no single type of refugia would provide adequate conditions for all taxa. Instream ponds need to be considered alongside a spatial and temporal mosaic of refugium (e.g., damp sediment, leaf litter, hyporheic) to maximise the resilience and persistence of aquatic floral and faunal communities in intermittent streams (Sheldon et al., 2010). In addition, instream ponding during the summer months may play an important role in sustaining macroinvertebrate taxa at a landscape-scale. Instream ponds may act as an aquatic refuge for a wider area than the stream channel (Chester and Robson, 2011), providing refuge habitat for taxa within a landscape mosaic of waterbodies with intermittent regimes (e.g., temporary ponds and wetlands).

Lotic, lentic and terrestrial systems have traditionally been studied in isolation (Soininen et al., 2015). Given that intermittent rivers transition between integrated lotic, lentic and dry phases there is a need to transcend traditional research boundaries (Larsen et al., 2016) to fully understand the ecological and hydrological functioning of all phases of intermittent rivers. Integrative research examining the ecological functioning and interaction/linkages among lotic, lentic and dry phases in intermittent rivers across different climatic regions and temporal scales is required to (1) understand and predict ecological responses in rivers with increasing intermittency and surface water loss (Leigh et al., 2016; Datry et al., 2014), and (2) provide significantly more detailed and accurate information for the development of bioassessment tools (incorporating lotic, lentic and terrestrial biota to monitor the overall health of the intermittent river) and monitoring and conservation strategies (Stubbington et al., 2018).

## 5. Conclusion

This study has demonstrated that instream ponds can provide an important site of refuge for macroinvertebrate taxa and make a significant contribution to aquatic diversity, providing suitable habitat for newly colonising taxa in temperate regions. Some rheophilic taxa were replaced by taxa more commonly found in lentic habitats as the river transitioned from the flowing phase to the ponding phase. Aquatic diversity in intermittent rivers is likely to have been significantly underestimated historically, as most previous biodiversity studies focussed on the lotic phase. Given that intermittent rivers are coupled lotic-lentic-terrestrial systems, there is a need for intermittent river research to transcend traditional, isolated scientific boundaries to better understand the integrated ecological functioning of biotic communities in intermittent systems. It is clear that current conservation strategies are failing to preserve a major aquatic biodiversity resource, whereby intermittent rivers can provide a site for the conservation of lotic, lentic and terrestrial taxa. Quantifying the biological diversity from all phases of intermittent rivers (lotic, lentic and terrestrial) will ensure the total biodiversity, and the most effective conservation, monitoring and management approaches are realised by scientists, policy makes and society.

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**References**

Acuña, V., Muñoz, I., Giorgi, A., Omella, M., Sabater, F., Sabater, S., 2005. Drought and postdrought recovery cycles in an intermittent Mediterranean stream: structural and functional aspects. J. Nam. Benthol. Soc 24, 919-933.

Arthington, A. H., Balcombe, S. R., Wilson, G. A., Thoms, M. C., Marshall, J. C., 2005. Spatial and temporal variation in fish assemblage structure in isolated waterholes during the 2001 dry season of an aridzone floodplain river, Cooper Creek, Australia. Mar. Freshw. Res. 56, 25-35.

Baselga, A. 2010. Partitioning the turnover and nestedness components of beta diversity. Global Ecol. Biogeogr. 19, 134-143.

Baselga, A., Orme, D., Villeger, S., De Bortoli, J., Leprieur, F. 2017. Partitioning beta diversity into turnover and nestedness components. R package version 1.3. Available online at: https://mran.microsoft.com/web/packages/betapart/betapart.pdf (accessed 20/11/2017).

Biggs, J., Fox, G., Whitfield, M., Williams, P., 1998. A guide to the methods of the National Pond Survey, Pond Action.

Boersma, K. S., Bogan, M. T., Henrichs, B. A., Lytle, D. A., 2014. Invertebrate assemblages of pools in arid-land stream have high functional redundancy and are resistant to severe drying. Freshw. Biol. 59: 491-501.

Bogan, M. T., Boersma, K. S., Lytle, D. A., 2015. Resistance and resilience of invertebrate communities to seasonal and supraseasonal drought in arid-land headwater streams. Freshw. Biol. 60, 2547-2558.

Bonada, N., Rieradevall, M., Prat, N., Resh, V. H., 2006. Benthic macroinvertebrate assemblages and microhabitat connectivity in Mediterranean-climate streams of northern California. J. Nam Benthol, Soc. 25, 32-43.

Boulton, A. J. 2003. Parallels and contrasts in the effects of drought on stream macroinvertbrate assemblages. Freshw. Biol. 48, 1173-1185.

Chadd, R. P., England, J. A., Constable, D., Dunbar, M. J., Extence, C. A., Leeming, D. J., Murray-Bligh, J. A., Wood, P. J., 2017. An index to track the ecological effects of drought development and recovery on riverine invertebrate communities. Ecol. Indic. 82, 344-356.

Chester, E. T., Robson, B. J., 2011. Drought refuges, spatial scale and recolonization by invertebrates in non-perennial streams. Freshw. Biol. 56, 2094-2104.

Corti, R., Datry, T., 2015. Terrestrial and aquatic invertebrates in the riverbed of an intermittent river: parallels and contrasts in community organisation. Freshw. Biol. 61, 1308-1320.

Costelloe, J. F., Russell, K. L., 2014. Identifying conservation priorities for aquatic refugia in an arid zone, ephemeral catchment: a hydrological approach. Ecohydrology. 7, 1534-1544.

Datry, T., 2012. Benthic and hyporheic invertebrate assemblages along a flow intermittence gradient: effects of duration of dry events. Freshw. Biol. 57, 563-574.

Datry, T., Larned, S. T., Fritz, K. M., Bogan, M. T., Wood, P. J., Meyer, E. I., Santos, A. N., 2013. Broad-scales patterns of invertebrate richness and community composition in temporary rivers: effects of flow intermittence. Ecography 36, 1-11.

Datry, T., Larned, S. T., Tockner, K. 2014. Intermittent rivers: a challenge for freshwater ecology. BioScience 64, 229-235.

De Caceres, M., Jansen, F., 2016. Relationship between species and groups of sites. R package version 1.7.6. Available online at: https://cran.r-project.org/web/packages/indicspecies/indicspecies.pdf (accessed 20/11/2017).

Dobrin, M., Giberson, D.J., 2003. Life history and production of mayflies, stoneflies, and caddisflies (Ephemeroptera, Plecoptera, and Trichoptera) in a spring-fed stream in Prince Edward Island, Canada: evidence for population asynchrony in spring habitats? Can. J. Zool. 81, 1083-1095.

Döll, P., Schmied, H. M., 2012. How is the impact of climate change on river flow regimes related to the impact on mean annual runoff? A global scale analysis. Environ. Res. Lett. 7, 14–37.

Drummond, L. R., McIntosh, A. R., Larned, S. T., 2015. Invertebrate community dynamics and insect emergence in response to pool drying in a temporary river. Freshw. Biol. 60, 1596-1612.

Edington, J. M., Hildrew, A. G., 1995. A revised key to the caseless caddis larvae of the British Isles. Freshwater Biological Association Scientific Publication No. 53. Freshwater Biological Association, Cumbria.

Elliot, J. M., Humpesch, U. H., Macan, T. T., 1988. Larvae if the British Ephemeroptera. Freshwater Biological Association Scientific Publication No. 49. Freshwater Biological Association, Cumbria.

Feeley, H., Baars, J.R., Kelly-Quinn, M., 2009. The life history of Perla bipunctata Pictet, 1833 (Plecoptera: Perlidae) in the upper river Liffey, Ireland. Aquat. Insects. 31: 261-270.

Friday, L. E. 1988a. A key to the adults of British Water Beetles. Field Studies. 7, 1-151.

Fritz, K. M., Dodds, W. K., 2004. Resistance and resilience of macroinvertebrate assemblages to drying and flood in tallgrass prairie stream. Hydrobiologia 527, 99-112.

Gallart, F., Llorens, P., Latron, J., Cid, N., Rieradevall, M., Prat, N. 2016. Validating alternative methodologies to estimate the regime of temporary rivers when flow data are unavailable. Sci. Total Environ. 565, 1001-1010.

Garcia, C., Gibbins, C. N., Pardo, I., Batalla, R. J., 2017. Long term flow change threatens invertebrate diversity in temporary streams: evidence from an island. Sci. Total Environ. 580, 1453-1459.

García-Roger, E., del Mar Sanchez-Montoya. M., Gomez, R., Suarez, M. L., Vidal-Abarca, R., Latron, J., Rieradevall, M., Prat, N., 2011. Do seasonal changes in habitat features influence aquatic macroinvertebrate assemblages in perennial versus temporary Mediterranean streams? Aquat. Sci. 73, 567–579.

Greenwood, M. T., Bickerton, M. A., Petts, G. E., 2001. Assessing adult Trichoptera communities of small streams: a case study from Charnwood Forest, Leicestershire, UK. Aquat. Conserv. Mar. Freshwat. Ecosyst. 11, 93-107.

Hill, M. J., Mathers, K. L., Wood, P. J., 2015. The aquatic macroinvertebrate biodiversity of urban ponds in a medium sized European town (Loughborough, UK). Hydrobiologia. 760, 225-238.

Hill, M. J., Heino, J., Thornhill, I., Ryves, D. B., Wood, P. J., 2017. Effects of dispersal mode on the environmental and spatial correlates of nestedness and species turnover in pond communities. Oikos. doi: 10.1111/oik.04266.

Kalogianni, E., Vourka, A., Karaouzas, I., Vardakas, L., Laschou, S., Skoulikidis, N.T., 2017. Combined effects of water stress and pollution on macroinvertebrate and fish assemblages in a Mediterranean intermittent river. Sci. Total Environ. 603, 639-650.

Karaouzas, I., Smeti, E., Vourka, A., Vardakas, L., Mentzafou, A., Tournes, E., Sabater, E., Munoz, I., Skoulikidis, N., Kalogianni, E. 2018. Assessing the ecological effects of water stress and pollution in a temporary river -implications for water management. Sci. Total Environ. 618, 1591-1604.

Koleff, P., Gaston, K. J., Lennon, J. J., 2003. Measuring beta diversity for presence-absence data. J. Anim. Ecol. 72, 367-382.

Lake, P. S. 2003. Ecological effects of perturbation by drought in flowing waters. Freshw. Biol. 48, 1161-1172.

Lancaster, J., Downes, B. J., 2013. Aquatic Entomology. Oxford University Press: Oxford.

Larsen, S., Muehlbauer, J.D., Marti, E., 2016. Resource subsidies between stream and terrestrial ecosystems under global change. Global Change Biol. 22, 2489-2504.

Leigh, C., Datry, T., 2016. Drying as a primary hydrological determinant of biodiversity in river systems: a broad scale analysis. Ecography. DOI: 10.1111/ecog.02230.

Leigh, C., Boulton, A. J., Courtwright, J. L., Fritz, K., May, C. L., Walker, R. H.. Datry, T., 2016. Ecological research and management of intermittent rivers: an historical review and future directions. Freshw. Biol. 61, 1181-1199.

Leigh, C., Bush, A., Harrison, E. T., Ho, S. S., Luke, L., Rolls, R. J.. Ledger, M. E., 2015. Ecological effects of extreme climatic events on riverine ecosystems: insights from Australia. Freshw. Biol. 60, 2620-2638.

Mackay, R. J., Wiggins, G. B., 1979. Ecological diversity in Trichoptera. Annu. Rev. Entomol. 24, 185-208.

Magoulick, D. D., Kobza, R. M., 2003. The role of refugia for fishes during drought: a review and synthesis. Freshw. Biol. 48, 1186-1198.

Marshall, J.C., Menke, N., Crook, D.A., Lobegeiger, J.S., Balcombe, S.R., Huey, J.A., Fawcett, J.H., Bond, N.R., Starkey, A.H., Sternberg, D., Linke, S., 2016. Go with the flow: the movement behaviour of fish from isolated waterhole refugia during connecting flow events in an intermittent dryland river. Freshw. Biol. 61, 1242-1258.

McCabe, D. J., N. J. Gotelli, 2000. Effects of disturbance frequency, intensity and area on assemblages of stream macroinvertebrates. Oecologia 124: 270–279.

UK Meteorological Office. 2017 Available online at: http://www.metoffice.gov.uk/public/weather/climate/gcqtvzgq0 (accessed 28/11/2017).

Nicolet, P., Biggs, J., Fox, G., Hodson, M.J., Reynolds, C., Whitfield, M., Williams, P., 2004. The wetland plant and macroinvertebrate assemblages of temporary ponds in England and Wales. Biol. Conserv. 120, 261-278.

NRFA 2017. National River Flow Archive: 28031 – Manifold at Ilam. Available online at: http://nrfa.ceh.ac.uk/data/station/meanflow/28031 (accessed 02/10/2017).

Oksanen, J., Blanchet, F. G., Friendly, M., Kindt, R., Legendre, P., McGlinn, D., Minchin, P. R., O'Hara, R. B., Simpson, G. L., Solymos, P., Henry, M., Stevens, H., Szoecs, E., Wagner, H., 2017. Vegan: Community Ecology Package. R package version 2.4-0. Available online at: http://CRAN.R-project.org/package=vegan (accessed 11/11/2017).

Petersen, I., Winterbottom, J.H., Orton, S., Friberg, N., Hildrew, A.G., Spiers, D.C., 1999. Emergence and lateral dispersal of adult Plecoptera and Trichoptera from Broadstone Stream, UK. Freshw. Biol. 42, 401-416.

Poepperl, R., 2000. The filter feeders *Hydropsyche angustipennis* and *H. pellucidula* (Trichoptera: Hydropsychidae) in a Northern German lowland stream: microdistribution, larval development, emergence pattern, and secondary production. Limnologica 30, 65-72.

Prudhomme, C., Guintoli, I., Robinson, E. L., Clark, D. B., Arnell, N. W., Dankers, R., Fekete, B. M., Franssen, W., Gerten, D., Gosling, S. N., Hagemann, S., Hannah, D. M., Kim, H., Masaki, Y., Satoh, Y., Stacke, T., Wada, Y., Wisser, D., 2014. Hydrological droughts in the 21st Century, hotspots and uncertainties from a global multimodel ensemble experiment. Proc. Natl. Acad. Sci. U.S.A. 111, 3262-3267.

R Development Core Team. 2016. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing. Vienna, Austria.

Rosset, V., Ruhi, A., Bogan, M.T. and Datry, T., 2017. Do lentic and lotic communities respond similarly to drying?. Ecosphere, 8, Article e01809.

Savage, A. A. 1989. Adults of the British aquatic Hemiptera Heteroptera: a key with ecological notes. Freshwater Biological Association Scientific Publication No. 50. Freshwater Biological Association, Cumbria.

Sheldon, F., Bunn, S. E., Hughes, J. M., Arthington, A. H., Balcombe, S. R.a Fellows, C. S. 2010. Ecological roles and threats to aquatic refugia in arid landscapes: dryland river waterholes. Mar. Freshw. Res. 61, 885-895.

Skoulikidis, N.T., Sabater, S., Datry, T., Morais, M.M., Buffagni, A., Dörflinger, G., Zogaris, S., del Mar Sánchez-Montoya, M., Bonada, N., Kalogianni, E., Rosado, J., 2017. Non-perennial Mediterranean rivers in Europe: status, pressures, and challenges for research and management. Sci. Total Environ. 577, 1-18.

Soininen, J., Bartels, P., Heino, J., Luoto, M.. Hillebrand, H., 2015. Toward more integrated ecosystem research in aquatic and terrestrial environments. BioScience 65, 174-182.

Soria, M., Leigh, C., Datry, T., Bini, L. M., Bonada, M., 2017. Biodiversity in perennial and intermittent rivers: a meta-analysis. Oikos. doi: 10.1111/oik.04118.

Stanley, E. H., Buschman, D. L., Boulton, A. J., Grimm, N. B., Fisher, S. G. 1994. Invertebrate Resistance and Resilience to Intermittency in a Desert Stream. Am. Midl. Nat. 131, 288-300.

Storey, R. G., Quinn, J. M., 2008. Composition and temporal changes in macroinvertebrate communities of intermittent streams in Hawke's Bay, New Zealand. – N. Z. J. Mar. Freshw. Res. 42, 109–125.

Stubbington, R., Chadd, R., Cid, N., Csabai, Z., Miliša, M., Morais, M., Munné, A., Pařil, P., Pešić, V., Tziortzis, I., Verdonschot, R.C., 2018. Biomonitoring of intermittent rivers and ephemeral streams in Europe: Current practice and priorities to enhance ecological status assessments. Sci. Total Environ. 618, 1096-113.

Stubbington, R., England, J., Wood, P. J., Sefton, C. E. M., 2017. Temporary streams in temperate zones: recognising, monitoring and restoring transitional aquatic-terrestrial ecosystems. Wiley Interdiscip. Rev. Water. doi: 10.1002/wat2.1223.

Van Leeuwen, C.H., Huig, N., Van der Velde, G., Van Alen, T.A., Wagemaker, C.A., Sherman, C.D., Klaassen, M., Figuerola, J., 2013. How did this snail get here? Several dispersal vectors inferred for an aquatic invasive species. Freshw. Biol. 58, 88-99.

Van Leeuwen, C.H., van der Velde, G., 2012. Prerequisites for flying snails: external transport potential of aquatic snails by waterbirds. Freshw. Sci. 31, 963-972.

Vander Vorste, R., Malard, F., Datry, T., 2016. Is drift the primary process promoting the resilience of river invertebrate communities? A manipulative field experiment in an intermittent alluvial river Freshw. Biol. 61, 1276-1292.

Vardakas, L., Kalogianni, E., Papadaki, C., Vavalidis, T., Mentzafou, A., Koutsoubas, D., 2017. Defining critical habitat conditions for the conservation of three endemic and endangered cyprinids in a Mediterranean intermittent river before the onset of drought. Aquat. Conserv. Mar. Freshwat. Ecosyst. doi: 10.1002/aqc.2735.

Wallace, I. D., Wallace, B., Philipson, G. N., 2003. Case-bearing caddis larvae of Britain and Ireland. Freshwater Biological Association Scientific Publication No. 61. Freshwater Biological Association, Cumbria.

Walters, A.W., Post, D.M., 2011. How low can you go? Impacts of a low-flow disturbance on aquatic insect communities. Ecol. Appl. 21, 163-174.

White, J. C., House, A., Punchard, N., Hannah, D. M., Wilding, M. A., Wood, P. J., 2018. Macroinvertebrate community responses to hydrological controls and groundwater abstraction effects across intermittent and perennial headwater streams. Sci. Total Environ. 610-611, 1514-1526.

Williams, D. D. 2006. The Biology of Temporary Waters. Oxford University Press, Oxford.

Wise, E. J., 1980. Seasonal distribution and life histories of Ephemeroptera in a Northumbrian river. Freshw. Biol. 10, 101-111.

Wood, P. J., Gunn, J., Smith, H., Abas-Kutty, A. 2005. Flow permanence and macroinvertebrate community diversity within groundwater dominated headwater streams and springs. Hydrobiologia. 545, 55-64.

## Tables

**Table 1 -** Summary table of macroinvertebrate diversity recorded from the perennial and intermittent transects during lotic conditions (May), the instream ponds during the ponding phase (July) of the intermittent section, and for all sampling sites combined form the Manifold and Hamps Rivers, UK.

| | Perennial | Intermittent | Ponding | Combined |
|---|---|---|---|---|
| **Total richness** | 86 | 82 | 78 | 129 |
| **Estimated total richness (95% CI)** | 110.1 (85.7 - 134.6) | 114.7 (82.2 - 147.1) | 102.5 (78.7 - 126.3) | 155.5 (130.6 - 180.4) |
| **Mean (±SE)** | 32.7 (± 3.2) | 26.3 (± 2.4) | 20.9 (± 1.2) | 25.3 (± 1.4) |
| **Median** | 30 | 23 | 21 | 23 |
| **Min** | 20 | 16 | 10 | 10 |
| **Max** | 46 | 42 | 29 | 46 |

**Table 2 -** Top 5 macroinvertebrate taxa identified as indicator species for the perennial and intermittent reaches during lotic conditions (May) and from the instream ponds during the ponding phase (July) of the intermittent section in the Manifold and Hamps Rivers, UK (see supplementary material Table S1 for the full list of statistically significant species indicator values). * = p <0.05, ** = p <0.01.

| **Perennial** | Stat | **Intermittent** | Stat | **Ponding** | Stat |
|---|---|---|---|---|---|
| *Ephemera danica* | 0.73** | Ecdyonurus spp. | 0.5* | *Helophorus brevipalpis* | 0.94** |
| *Esolus parallelepipedus* | 0.70** | Heptagenia spp. | 0.5* | *Centroptilum pennulatum* | 0.87** |
| *Psychomyia pusilla* | 0.70** | | | Dytiscidae Larvae | 0.78** |
| Trichoptera Pupae | 0.58* | | | *Potamopyrgus antipodarum* | 0.78** |
| *Baetis niger* | 0.52* | | | Haliplidae Larvae | 0.77** |

**Figure Captions**

**Fig. 1** – Location of the 7 study reaches during lotic conditions inMay (blue circles=perennial sites, green circles=intermittent sites) and the 17 instreamponds (grey circles) surveyed in July during the ponding phase of the intermittent section of the Manifold and Hamps Rivers in the Peak District, UK.

**Fig. 2 -** Species accumulation curves of macroinvertebrate richness (a) and median richness of macroinvertebrates (b) recorded from the perennial and intermittent reaches during lotic conditions (May) and from the instream ponds during the ponding phase (July) of the intermittent section. Boxes show 25th, 50th, and 75th percentiles and whiskers show 5th and 95th percentiles.

**Fig. 3 -** Non-metric multidimensional scaling plots of dissimilarity in (a) aquatic macroinvertebrate communities (Sørenson dissimilarity) and boxplots of multivariate dispersion distances (b) for macroinvertebrate communities recorded from the perennial and intermittent transects during lotic conditions (May) and from the instream ponds during the ponding phase (July) of the intermittent section. Perennial sites = black circles, intermittent sites= grey diamonds and ponded sites = open circles.

**Fig. 4 -** Number of taxa recorded from the intermittent (surveyed in May 2016 during lotic conditions) and ponded sites (surveyed in July 2016 during ponding phase) from the Manifold and Hamps Rivers that were recoded only from the ponded sites (unique), recorded from both the intermittent and ponded sites (persisted) and recorded only from the intermittent sites (lost).

**Fig. 5 -** Relative contribution of species turnover and nestedness to total community dissimilarity (in bold) within the perennial and intermittent sites (transects) during lotic conditions (May) and from the instream ponds during the ponding phase (July) of the intermittent section.

**Fig. 1**

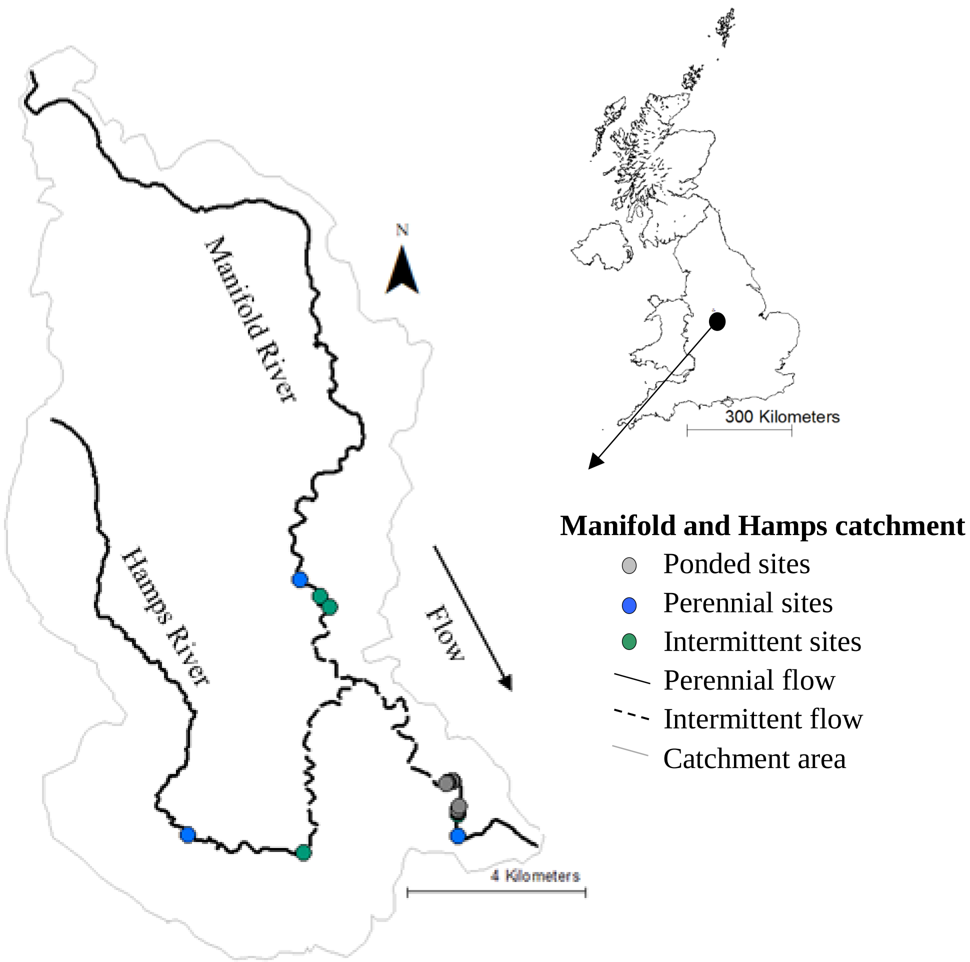

N
Manifold River
Hamps River
Flow
300 Kilometers
4 Kilometers
Manifold and Hamps catchment
Ponded sites
Perennial sites
Intermittent sites
Perennial flow
Intermittent flow
Catchment area

**Fig. 2**

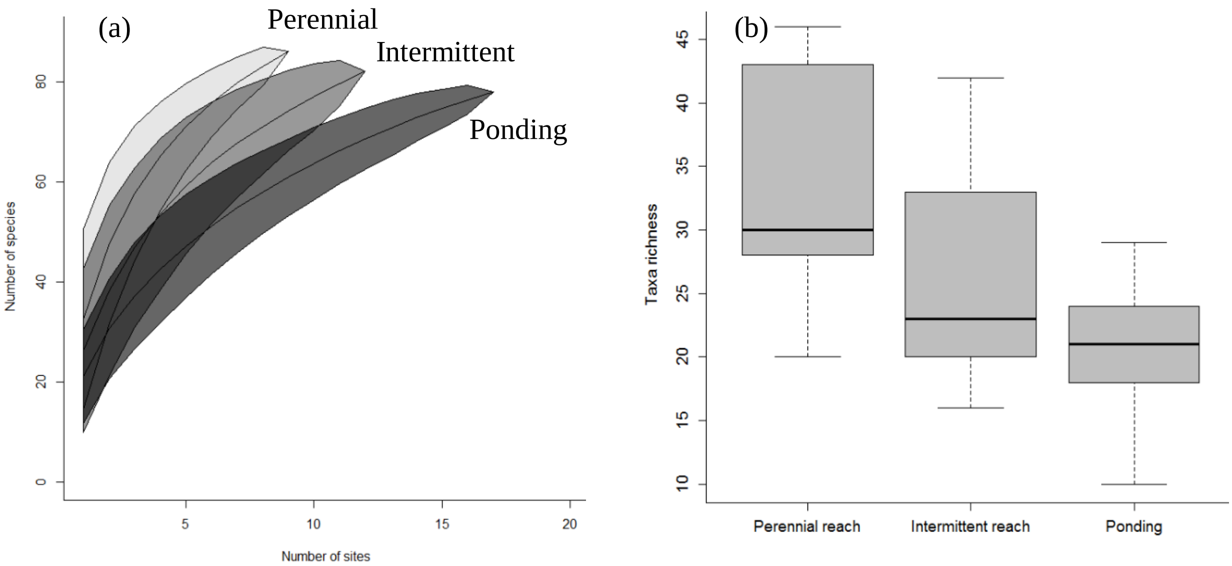

(a)
Perennial
Intermittent
Ponding
Number of species
0
20
40
60
80
5
10
15
20
Number of sites
(b)
Taxa richness
10
15
20
25
30
35
40
45
Perennial reach
Intermittent reach
Ponding

**Fig. 3**

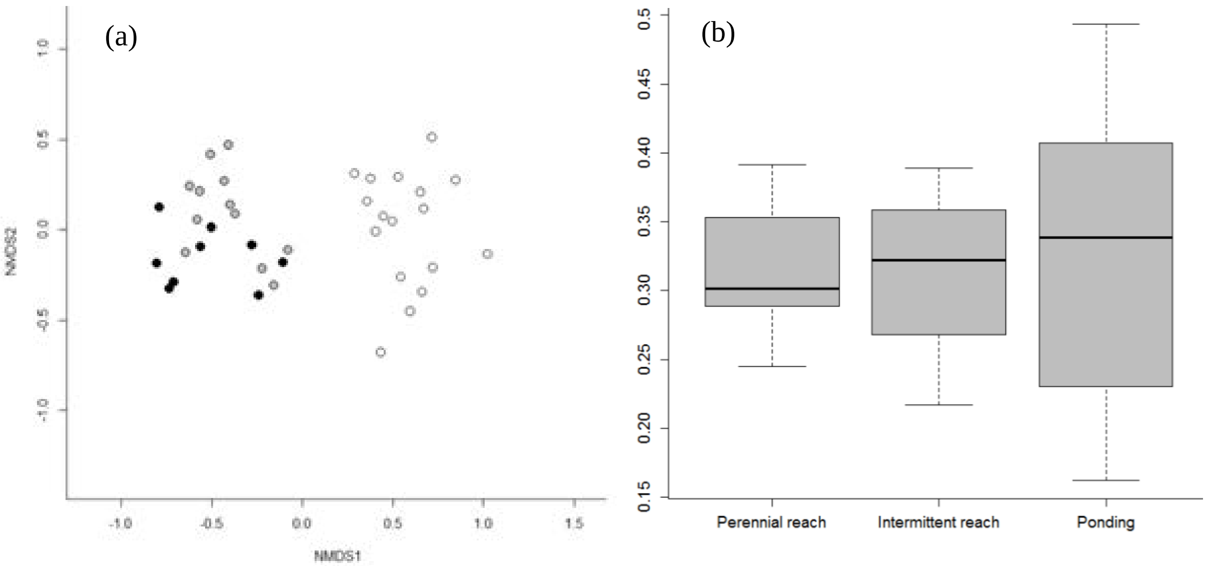

(a)
NMDS1
NMDS2
-1.0
-0.5
0.0
0.5
1.0
1.5
(b)
0.15
0.20
0.25
0.30
0.35
0.40
0.45
0.5
Perennial reach
Intermittent reach
Ponding

**Fig. 4**

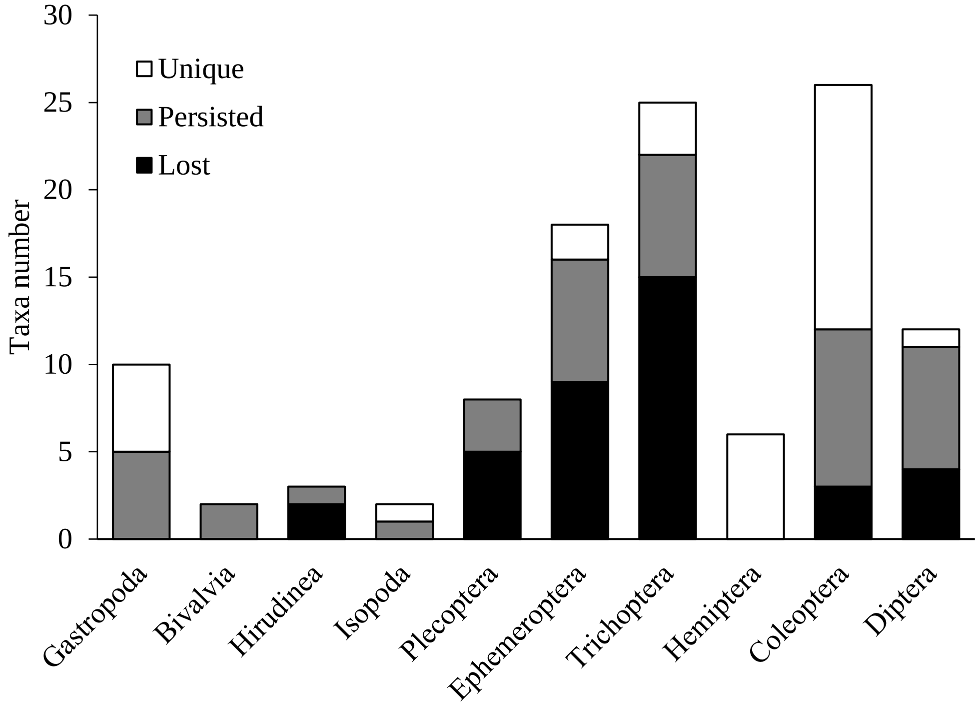
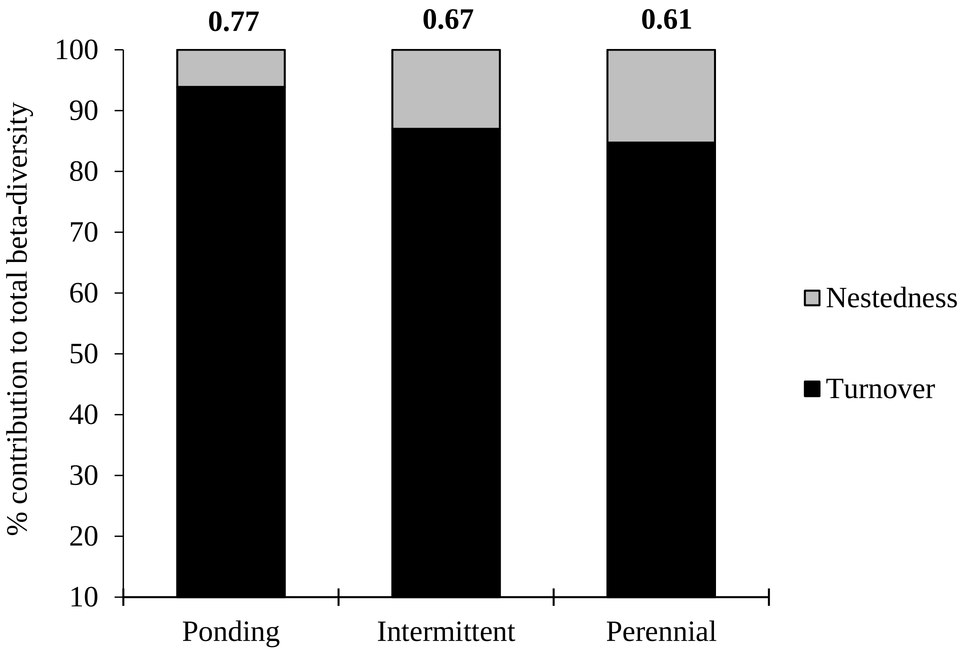

Unique
Persisted
Lost
Taxa number
0
5
10
15
20
25
30
Gastropoda
Bivalvia
Hirudinea
Isopoda
Plecoptera
Ephemeroptera
Trichoptera
Hemiptera
Coleoptera
Diptera

**Fig. 5**


0.77
0.67
0.61
100
90
80
70
60
50
40
30
20
10
% contribution to total beta-diversity
Ponding
Intermittent
Perennial
Nestedness
Turnover