

Indicators of Diptera diversity in wet grassland habitats are influenced by environmental variability, scale of observation, and habitat type.

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Highlights

- Fine spatial scale analysis is required to investigate dipteran diversity in agri-ecosystems
- The effectiveness of dipteran surrogate indicators is determined by wet grassland category
- The scale at which invertebrate diversity is assessed on farmland needs further investigation
- Marsh fly and hoverfly communities are highly congruent with nine other dipteran families

1. Abstract

In low intensity agri-ecosystems such as wet grassland habitats, the inclusion of invertebrates in conservation assessments and monitoring is usually limited to charismatic groups such as bees or butterflies. However, wet grasslands support a wide range of inveterate groups, some of which may exhibit limited movement not generally represented by more mobile groups such as those typically examined. The use of surrogate species which exemplify broader invertebrate diversity has been suggested as a possible means of including these overlooked invertebrates (such as Diptera) in conservation planning within these habitats. Based on collections made by Malaise trap, we utilized two families of Diptera (Sciomyzidae and Syrphidae) as indicators of a wider range of dipteran diversity (nine Diptera families identified to parataxonomic unit level [PUs]) in wet grassland habitats. We examined the role of environmental variability, spatial scale and habitat type on patterns of cross-taxon congruence for all three assemblages. Both environmental correlation and community

congruence were significantly stronger among assemblages when examined at low spatial scales; highlighting the need to examine dipteran groups at scales untypical of current agri-environmental assessments; namely field and farm level. Furthermore, when wet grasslands were differentiated into two habitat categories (Sedge and Rush dominated grasslands), the significance of the community congruence increased markedly. This correlation was particularly strong between Sciomyzidae and PUs which demonstrated similar differentiation based on habitat type-implicating that assemblages which exhibit comparable ecological partitioning are more likely to be useful surrogates of one another. Correlations between richness, abundance and Shannon's diversity were highly variable among groups, suggesting compositional analysis as the most appropriate examination of dipteran diversity for surrogacy studies. The results indicate that cross-assemblage congruence of Diptera is influenced by similarity of response to environmental variability, scale of observation, and examination of assemblages differentiated into appropriate habitat categories. The results illustrate the need to investigate invertebrate biodiversity surrogates at scales appropriate to the indicator groups and examine congruence among assemblages within specific habitat categories. Such an approach has the potential to maximise gamma diversity in areas where wet grasslands are under threat of intensification or abandonment.

Keywords: Biodiversity; Diptera; Sciomyzidae; Syrphidae; Parataxonomy; Community Congruence

2. Introduction

European wet grassland habitats are typically low-intensity agricultural systems with semi-natural habitats which support a rich mosaic of plant and animal communities (Bignal, E.M. & McCracken, 1996; Bignal and McCracken, 2000; Billeter et al., 2008). While much of the conservation of lowland wet grassland is driven by botanical or ornithological interests, wet grasslands also sustain a high diversity of invertebrates (Drake, 1998; Hayes et al., 2015; Joyce and Wade, 1998; Maher et al., 2014). However, the difficulties associated with collecting comprehensive invertebrate data from habitats have contributed towards the continued limited inclusion of invertebrate groups in broad scale conservation planning and for monitoring conservation objectives (Cardoso et al., 2011). This is particularly relevant in a European agricultural context, where intensification and abandonment of traditional farming practices in areas such as wet grasslands is threatening biodiversity (Henle et al., 2008). The use of invertebrates in the designation and management of agricultural areas considered as being of a high nature value is usually limited (if included at all) to a few well known, easily identifiable, and often iconic groups such as butterflies or bees (Andersen et al., 2004). Other invertebrate groups such as Diptera are largely overlooked despite contributing significantly to the overall biodiversity of such habitats (Keiper et al., 2002).

While the need to include a wider suite of invertebrates in conservation strategies for wet grasslands makes ecological sense, the sheer abundance and diversity of groups such as Diptera are perceived as barriers to their inclusion in routine habitat assessments. Alternative approaches such as the use of selected invertebrate groups as biodiversity surrogates for a broader range of taxa has been suggested as a possible means of including invertebrates in conservation and monitoring programs (Anderson et al., 2011; Duelli et al., 1999; Duelli and Obrist, 2003; Hayes et al., 2015). Such biodiversity indicators generally include a well-studied taxon or group of taxa which are ubiquitous within the habitat of interest and can be easily collected and identified (Lindenmayer et al., 2000; McGeoch et al., 2002). A predetermined measure of the diversity of the selected indicator is then used to reflect the diversity of similar, or sometimes different, taxa. One such approach has been the examination of species richness congruence between indicator groups and the taxa they are deemed to represent (Duelli and Obrist, 2003; Moreno and Sánchez-rojas, 2007; Prendergast, 1997). However, the species richness approach is considered as having variable outcomes due to its dependency on the pairs of taxa under investigation and it provides little insight into overall species representation and composition (Su et al., 2004).

More recently, patterns of congruence derived from community similarity and/or the examination of similarity of community responses to environmental variability have been utilized as approaches to biodiversity surrogacy (Larsen et al., 2012; Paszkowski and Tonn, 2000; Rooney and Azeria, 2015; Rooney and Bayley, 2012; Su et al., 2004). A potential caveat with this method, however, is the effect of spatial scale of observation and habitat differentiation on congruence patterns. Invertebrate diversity may respond to spatial scales not typically considered in conservation strategies (Haslett and Salzburg, 1997; Weaver, 1995), and community composition can be influenced by microhabitat changes across small scales that can have a marked effect on community structures (Cole et al., 2010). In wet grassland habitats, this may be further exacerbated by temporal changes such as periodic inundation in combination with grazing patterns (Carey et al., 2017; Maher et al., 2014; Ryder et al., 2005). Thus, the examination of invertebrate communities may need to be assessed at spatial scales untypical of those employed in conventional biodiversity assessments which are often linked to agri-environmental scheme evaluations at farm or field-level scales.

The differentiation of habitats into categorical groups has also been shown to affect congruence patterns with anthropogenic disturbance and ecoregion having a noticeable influence on congruence measures (Ekroos et al., 2013; Myšák and Horsák, 2014; Rooney and Azeria, 2015; Rooney and Bayley, 2012). The selection of invertebrate biodiversity surrogates, therefore, needs to consider carefully determinants such as the distribution of the indicator taxa relative to the scale of the observation, response of the indicator to ecological variance, and possible ecological relationships between the indicator and the wider community it is chosen to represent (McGeoch, 1998; Paoletti, 1999).

In wet grassland habitats, adult Marshflies (Diptera: Sciomyzidae) are considered as potential bioindicators owing to their ubiquity and ease of capture (Carey and LeRoy et al., 2015; Knutson and Vala, 2011). However, they are known to have highly localised habitat fidelity and exhibit a markedly limited movement (Williams et al., 2010); factors which may restrict their usefulness as surrogates for broader dipteran diversity if the scale of observation utilized is greater than that which accurately reflects their distribution. In contrast to this, adult Hoverflies (Diptera: Syrphidae) are considered suitable bioindicators in agricultural systems (Burgio and Sommaggio, 2007) but are vagile in nature with adults capable of foraging over long distances (Sommaggio, 1999). The use of either of these groups as invertebrate biodiversity surrogates is therefore dependent on the similarity of their response to factors such as spatial scale and habitat differentiation relative to the broader invertebrate diversity for which they are selected to be a proxy

Along with these considerations, the identification of multiple and diverse groups such as Diptera to species level usually requires expertise and time which is not readily available in the context of typical designation or monitoring timeframes. Suggested alternatives to this impediment include attempting to rapidly identify several groups of invertebrates using less traditional taxonomic methods (Cardoso et al., 2011). Rapid biodiversity assessment techniques such as parataxonomy (morphospecies) as described by Oliver et al. (1993) utilizes an approach whereby individuals with similar external morphological traits are grouped together as typological units or Parataxonomic Units (PUs) without the use of taxonomic keys. This work can be carried out by individuals with minimal taxonomic training and possibly even through public participation initiatives such as citizen science (Casanovas et al., 2014). Though the method is subject to debate regarding its effectiveness (Thorsten-Krell, 2004; Ward and Stanley, 2004), when executed with caution, and subject to some level of taxonomic verification, it can be utilized to give ecologically relevant outcomes (Cotes et al., 2009; Obrist and Duelli, 2010; Oliver and Beattie, 1996; Ward and Stanley, 2004). Studies of Diptera in wet grasslands which have utilized the two approaches (taxonomic and parataxonomic) have generally focused on richness correlations of all Diptera and not examined community similarity (Hayes et al., 2015; Ryder et al., 2005). Though useful patterns can be derived from such data, especially in terms of family richness and abundances, a more in-depth investigation of a smaller number of dipteran families using more prolonged sampling and intensive sorting methods might be more insightful (Frouz, 1999).

Given that Sciomyzidae and Syrphidae fulfil the criteria for suitable bioindicators as outlined by McGeogh (1998), we compared measures of their diversity with a broader assemblage of nine Diptera families identified using parataxonomy. By examining agreement among groups in terms of environmental responses we were able to identify the role that environmental factors play in determining community structure of different dipteran assemblages. We also conducted a hierarchical sampling regime from two wet grassland habitat types (Rush dominated and Sedge dominated wet

grasslands) based on samples from individual traps or samples from traps from the same sample patch pooled together. We investigated the role that spatial scale and habitat type played in determining patterns of congruence among the three assemblages (Sciomyzidae, Syrphidae and dipteran PUs) using a range of tests.

Our principal objectives were to:

1. Examine patterns of environmental correlation between the groups at two spatial scales (Trap level and Patch level)
2. Determine whether cross-taxon congruence among groups was affected by scale of observation
3. Investigate the role that habitat type plays in contributing to patterns of cross-assemblage congruence

The results of this investigation are discussed in the context of selecting suitable invertebrate biodiversity indicators within high nature value agri-ecosystems such as wet grasslands.

3. Materials and Methods

2.1 Study area

This investigation was undertaken in the west of Ireland in wet grassland habitats defined according to Fossitt (2000). For inclusion in this classification, grass, rush or small sedge cover needs to exceed 50% and broadleaf herbs, reeds and larger sedges should not dominate i.e. must be <50%. The broadleaf herb component should also be relatively evenly divided between drier grassland and wetland species. We selected five each of two sub-categories of wet grassland based on their dominant vegetation type i.e. wet grasslands dominated by rushes (Family Juncaceae) and wet grasslands dominated by sedges (Family Cyperaceae). All wet grassland sites in this study were actively managed for livestock grazing and were not subject to intensive cutting regimes or application of fertilizers.

2.2 Diptera sampling and determination

Diptera sampling was undertaken between May 1st and September 4th 2014 using black Malaise traps of Townes design (Townes, 1972). Two traps were placed 20m apart in homogeneous patches of vegetation and away from obvious topographical features such as drainage ditches, wet flushes, hedgerows etc. This method was employed to maximise Diptera collections from within the sample patch rather than as a result of movement between habitats or due to the presence of any obvious ecotonal changes (Carey et al., 2017). Collection heads containing a 70% ethanol solution were

positioned in a southerly direction and were collected every 14 days. A portable electric fence was operated to protect the traps from interference by livestock, and vegetation within the enclosure was intermittently shortened and removed to maintain trap efficacy and replicate conditions outside the fenced area. Eleven families of adult Diptera were selected for analysis based on their ease of identification to family level, ubiquity within the habitat, and previous recommendations for use as bioindicators of wetland habitats (Hayes et al., 2015; Speight, 1986). These families were the Dolichopodidae, Empididae, Hybotidae, Limoniidae, Pipunculidae, Scathophagidae, Sciomyzidae, Stratiomyidae, Syrphidae, Tabanidae, and Tipulidae.

Sciomyzidae and Syrphidae were utilized as the principal biodiversity indicators and identified to species level using Rozkošný (1987) and Vala (1989) for sciomyzids and Ball & Morris (2013) and Stubbs & Falk (2002) for syrphids. The remaining nine families were identified using parataxonomy, a rapid biodiversity assessment method. Non-specialist individuals (undergraduate students hereafter referred to as parataxonomists) utilized a simplified character key derived from Oosterbroek (2007) and Unwin (1981) to assist them in the removal of the remaining nine families from bulk samples. Subsequent to initial sorting to family level, the specimens were categorised into parataxonomic units (PUs) based on their external morphological features without the use of keys (Oliver and Beattie, 1996, 1993). Each newly assigned PU was digitally photographed and the image inserted into a shared data base allowing each parataxonomist access to the image for referral. The initial voucher specimen was preserved in 100% ethanol for determination to species level by taxonomists. These voucher specimens were utilized to determine the level of ‘splitting’ whereby a taxonomic species was split into two or more PUs. Oliver and Beattie (1996) recommend that a random subset of ~30 individuals from each PU is also maintained (as vouchers) to determine the degree of ‘lumping’ i.e. when two or more taxonomic species are classed within the same PU. Our study opted to exclude this verification as it would have entailed the identification of over 3000 individual specimens by taxonomists. Instead, only the original voucher specimens were identified by specialists and, therefore, only species splitting and not lumping was accounted for. Omitting the ‘lumping’ protocol due to time constraints is likely to have led to underestimations of species richness, but was countered by the speed of the species determination by specialist taxonomists who needed to verify ~30 times less specimens. This effectively maximised taxonomic input by minimising effort; an important consideration due to the increasingly limited availability of taxonomic specialists (Cardoso et al., 2011) and requirement for rapid assessments of biodiversity in line with typical conservation strategies. The original inventory was then subjected to changes based on the species determinations and the corrected PU data utilized for analysis. The percentage of splitting error for each PU family was reported according to Oliver and Beattie (1996).

2.3 Environmental data

A series of environmental variables was recorded at each site throughout the collection period and mean values calculated. These included vegetation height as well as the percentage cover of grasses, sedges, rushes, moss, moribund material, broadleaf herbs and bare ground. Each of the variables was estimated using five 50 x 50 cm quadrats placed randomly within 10m of each Malaise trap location and within the homogeneous vegetation patch. Soil samples were extracted from the same random sampling areas and mean pH, soil carbon content, and soil moisture were analysed (Anon, 1990). The depth of standing water at each site was recorded in the October following trap removal and prior to any extensive winter inundation which would have rendered water depth measurements unsafe.

2.4 Data analysis

Prior to multivariate analysis invertebrate abundance data was $\log_{10}(x+1)$ transformed to reduce the influence of numerically dominant species and to approximate multivariate normality. Species which were considered outliers (>2.0 standard deviations) using the Sørensen distance measure were also removed (McCune and Mefford, 2011). Environmental data (with the exception of soil pH) was arcsine square root transformed or $\log_{10}(x+1)$ transformed to improve linearity and to approximate normality (McCune and Mefford, 2011).

Non-metric multidimensional scaling (NMS) ordinations (McCune and Grace, 2002) of samples was undertaken using the Sørensen distance in PC-Ord v.6 (McCune and Mefford, 2011). Samples which were identified as extreme outliers with standard deviations >3.0 using the Sørensen distance measure were removed prior to NMS ordination. We utilized 250 runs of real data to 250 runs with randomised data to determine the number of significant axes. An orthogonal principal axis output was selected for each NMS to illustrate maximum community variation along axis 1. Environmental data were utilized as a second explanatory matrix and variables with Pearson r^2 values >0.2 overlain as a bi-plot (McCune and Mefford, 2011). Multi-Response Permutation Procedures (MRPP), which are non-parametric procedures for the testing the hypothesis of no difference between two groups, were utilized to examine for significant differentiation between habitat types based on the species/PU composition of each assemblage (McCune and Mefford, 2011).

A Principal Components Analysis (PCA) with a cross products matrix based on correlation was utilized to evaluate the environmental data. MRPP was also utilized to examine habitat differentiation based on PCA results. The PC1 score obtained from this analysis was used to rotate the NMS ordination of each assemblage to ensure a standard alignment for comparisons between community structure and NMS axes. Such rotation does not alter the relative position of each sample in species space and allows for comparison between assemblages (McCune and Mefford, 2011; Rooney and Bayley, 2012). Subsequent to rotation, we examined the response of the three assemblages to environmental variables by comparing the Pearson correlation coefficients of each significant axis derived from the NMS using Spearman Rank correlations in PAST (Hammer et al., 2001). All

responses were considered at trap scale (each individual Malaise trap [n=20]) and patch scale (where data from pair-wise traps from the same vegetation patch were combined [n=10]).

We compared changes in assemblage structure within fields (i.e. between pair-wise traps) using the Sørensen similarity as a measure of differentiation between pair-wise traps. Similarity scores obtained for each assemblage were compared using linear correlation (Pearson's R) to investigate whether patterns of differentiation between pair-wise traps was congruent between assemblages. For each assemblage, we also tested the relationship between the level of community similarity between pair-wise traps and the differentiation of environmental variables between pair-wise traps using Spearman Rank correlations in PAST (Hammer et al., 2001).

Community congruence between indicator groups was tested using Partial Mantel tests controlling for geographical and environmental autocorrelation using Sørensen distance measures for species/PU data and Euclidean distance measure for control matrices (McCune and Mefford, 2011). Partial Mantel tests were carried out across all samples, at two spatial scales (patch and field), and within habitat types at trap scale using PC-Ord V.6.

Sciomyzidae, Syrphidae and PU richness from each sample was calculated as a proportion of the total richness of each assemblage across all sample sites (Finch and Löffler, 2009). Shannon's entropy (previously known as Shannon's diversity) was utilized as a measure of the diversity of each assemblage (Ellison, 2010; Jost, 2007), along with raw abundance values. We utilized Spearman rank correlations to investigate patterns of cross-taxon congruence of each of these values using PAST (Hammer et al., 2001). Correlations were investigated at two spatial scales (Trap scale [n=20], Patch scale [n=10]) and within habitat types (rush or sedge dominated sites) at trap scale (n=10).

5. Results

5.1. General results

A total of 105,666 individuals from eleven families of Diptera were collected from the sampling sites and subjected to taxonomic or parataxonomic identification. Sciomyzidae (1,975 individuals) and Syrphidae (9,568 individuals) were determined to species level with 34 and 72 species identified, respectively. This represents 53% of Sciomyzidae and 40% of Syrphidae from the Irish fauna (Chandler et al., 2008).

A total of 105 parataxonomic units were identified from the nine remaining Diptera families. Once splitting had been accounted for, this was reduced to 85 with an overall percentage splitting error of 24% (Table 1). Dolichopodidae (Long-legged flies) represented the most abundant family (45,337) with Stratiomyidae (Soldierflies) contributing the least number of individuals (685). Scathophagidae (Dung flies) showed the highest percentage splitting error of PU allocation owing to the markedly different body size and colouration attributed to the yellow dung fly (*Scathophaga stercoraria* L.

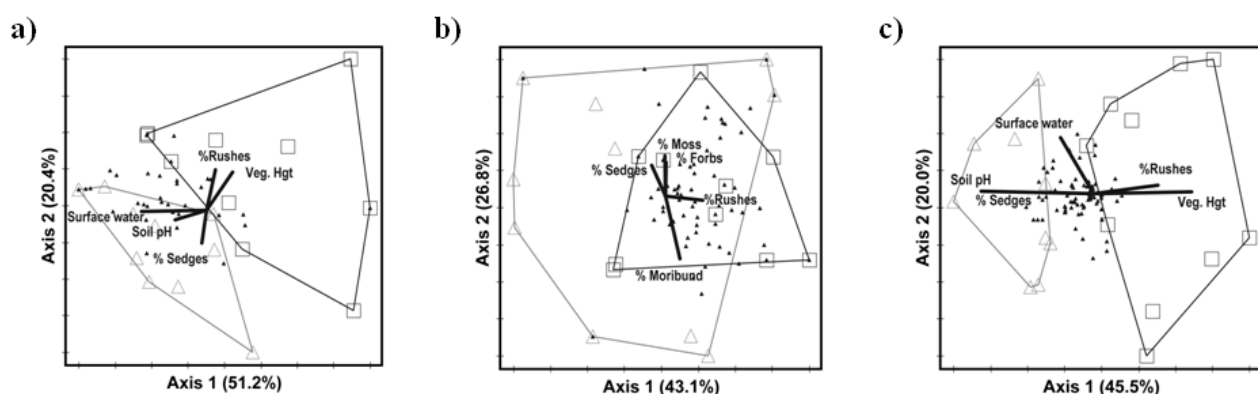
1758). Pipunculidae (Big-headed flies) showed the lowest level of splitting error, though it is likely that ‘lumping’ of species occurred in the allocation of PUs to this family owing to marked similarity among sibling species.

Table 1: Number of individuals, species, and Parataxonomic units (PUs) per Diptera family. The percentage splitting error refers to the proportion of splitting within each family whereby a species was classified as more than one PU by parataxonomists.

Family (Individuals)	Number of species	Number of PUs	% splitting error
Dolichopodidae (45337)	15	17	13
Limoniidae (13796)	14	15	7
Empididae (11987)	15	18	20
Scathophagidae (8933)	7	14	100
Hybotidae (6098)	12	13	8
Pipunculidae (3129)	4	4	0
Tabanidae (2820)	3	4	33
Tipulidae (1338)	5	8	60
Stratiomyidae (685)	10	12	20
Overall	85	105	24

5.2. Response to environmental variability

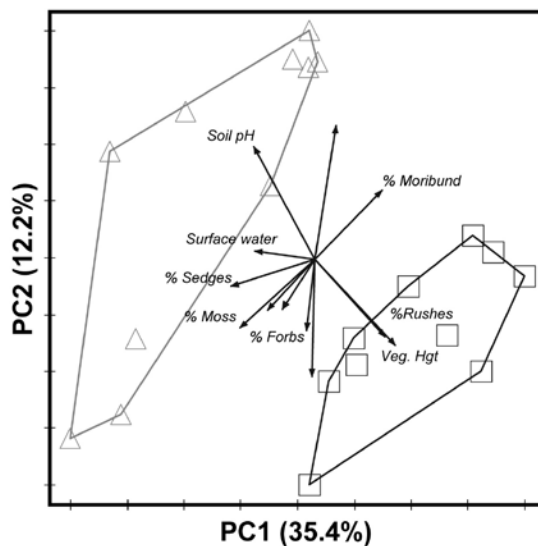
NMS ordinations of samples in species/PU-space produced three dimensional solutions which explained >80% of the variation for each assemblage (Sciomyzidae 86.0%, Syrphidae 84.3%, PUs 80.4%). Environmental variables with a Pearson r^2 score of >0.2 are shown as bi-plots (Figure 1). Stress levels for each ordination were all <11.5 with values of ~10 are considered suitable for reasonable interpretation (McCune and Mefford, 2011). Multi-response permutation-procedure (MRPP) analysis showed significant differences between the two habitat types for all three assemblages using the Sørensen distance measure. PUs showed the most significant differentiation between habitat types ($A=0.080$, $P=3 \times 10^{-5}$), followed by Sciomyzidae ($A=0.062$, $P=6 \times 10^{-4}$) and then Syrphidae ($A=0.030$, $P=0.027$).



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292 Figure 1: NMS ordinations of samples in species/PU space. Environmental variables with r^2 values
 293 >0.2 are shown as bi-plots. The principal orthogonal axes are shown with the percentage of variation
 294 associated with each axis. Samples are shown as open squares (rush dominated wet grassland) and
 295 open triangles (sedge dominated wet grassland), species/PUs as black triangles. **a)** Sciomyzidae, **b)**
 296 Syrphidae and **c)** and PUs. Only axes 1 & 2 are illustrated which explain a cumulative variation of:
 297 Sciomyzidae (71.6%), Syrphidae (69.9%); PUs (65.5%). Habitat types were significantly different
 298 using MRPP analysis for all three groups **a)** $A=0.062$, $P=6 \times 10^{-4}$ **b)** $A=0.030$, $P=0.02700$ **c)**
 299 $A=0.080$, $P=3 \times 10^{-5}$.

300 Principal components analysis of environmental variables showed that PC1 explained 35.4% of the
 301 variance between samples (Figure 2). MRPP analysis confirmed that habitats were significantly
 302 different from one another based on environmental data ($A=0.249$, $P=2 \times 10^{-5}$). The scores from PC1
 303 were utilized to rotate the NMS ordinations for examination of cross-assemblage agreement based on
 304 environmental correlations. All three assemblages showed significant correlations based on NMS axis
 305 one irrespective of spatial scale of observation (Table 2). At trap scale ($n=20$), Sciomyzidae and
 306 Syrphidae showed significant correlations across all axes. This was reduced to the first two axes at
 307 patch scale. PUs showed significant correlations with the other groups only on axis one. It is worth
 308 noting that the negative correlation between Syrphidae and PUs is likely an artefact of the rotation of
 309 the NMS using only PC1 scores (Rooney and Bayley, 2012).



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311 Figure 2: Principal Components Analysis of sampling patches showing the environmental variables
 312 most strongly associated with PC1. Samples are differentiated into habitat type (Rush wet grassland:
 313 open squares; sedge wet grassland: open triangles). Habitat types were significantly different based on

MRPP analysis (A=0.249, P=0.00002). Arrows with no associated environmental variable were associated with PC2.

Table 2: Environmental correlations (Spearman's r) between dipteran assemblages based on the axes

	Axis 1			Axis 2			Axis 3		
	Sciomyzidae	Syrphidae	PUs	Sciomyzidae	Syrphidae	PUs	Sciomyzidae	Syrphidae	PUs
Trap scale (n=20)									
Sciomyzidae	1			1			1		
Syrphidae	0.73**	1		0.65*	1		0.73**	1	
PUs	0.98***	0.70**	1	-0.01	-0.53	1	-0.38	-0.07	1
Patch scale (n=10)									
Sciomyzidae	1			1			1		
Syrphidae	0.85***	1		0.79**	1		0.07	1	
PUs	0.76**	0.73**	1	-0.49	-0.80**	1	-0.13	-0.05	1

of the NMS ordinations rotated by PCA score. (P<0.05*, P<0.01**, P<0.001***)

5.3. Community similarity between pair-wise traps

Sciomyzidae and PUs showed a significant relationship in terms of how their assemblages responded to differentiation between pair-wise traps i.e. increased dissimilarity between pair-wise traps was congruent among both assemblages ($r^2 = 0.84$, $P=0.002$) (Figure 3). None of the measured environmental variables were significantly correlated with changes in community similarity of Sciomyzids or PUs between pair-wise traps i.e. none of the measured variables appeared to significantly affect differences in community structure between traps from the same patch. Syrphidae were not significantly correlated with Sciomyzidae ($r^2 = -0.23$, $P=0.470$) or PUs ($r^2 = -0.17$, $P=0.063$) in terms of similarity of community differentiation between pair-wise traps. However, differentiation of Syrphidae between pair-wise traps was significantly correlated with differences in the percentage cover of broadleaf herbs between pair-wise traps (Spearman's $r = 0.82$, $P=0.004$); a relationship that was not significant for Sciomyzidae or PUs.

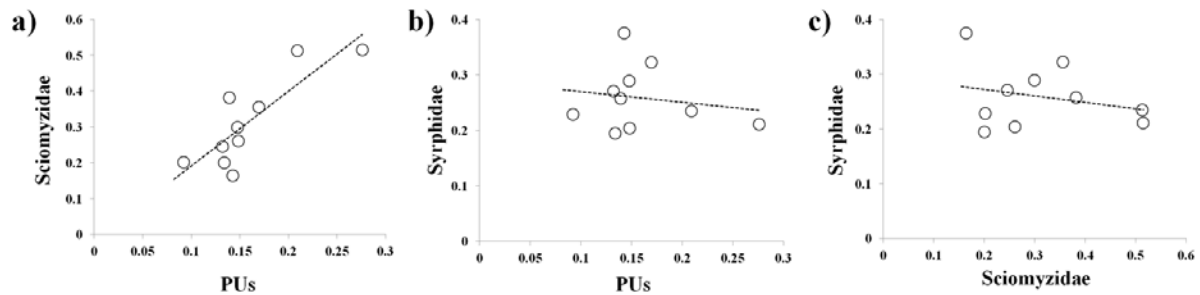


Figure 3: Linear regression illustrating the congruence of changes in assemblage structure between pair-wise traps using the Sorensen distance measure. Increasing values are indicative of greater dissimilarity. **a)** Sciomyzidae and PUs were significantly correlated ($r^2 = 0.84$, $P=0.002$), **b)** Syrphidae and PUs ($r^2 = -0.17$, $P=0.0630$) ($r^2 = -0.23$, $P=0.470$) and **c)** Syrphidae and Sciomyzidae ($r^2 = -0.23$, $P=0.470$) were not significantly correlated.

5.4. Cross-assemblage congruence

Mantel R values were weak but significant between Sciomyzidae and PUs across all samples but only at trap level (Table 3). This relationship was maintained even when geographic and environmental autocorrelation was controlled for. There was also a significant relationship between Syrphidae and PUs at trap scale but this was not maintained when either geographic or environmental autocorrelation was controlled for. There were no significant correlations between any of the three assemblages at patch level. When correlations among assemblages were examined within habitat types, Sciomyzidae and PUs were significantly congruent within both habitats (Table 4) even when geographic and environmental autocorrelation were controlled for. Sciomyzidae were also significantly correlated with Syrphidae in both habitats, however, in the sedge habitats this was a result of geographic and/or environmental co-response i.e. trap proximity or similarity of environmental variables at the trap

locations appeared to influence congruence. Syrphidae were congruent with PUs in Rush dominated wet grasslands but not in Sedge dominated habitats.

Table 3: Mantel R statistics of assemblage concordance at two spatial scales using the Sorensen distance measure for species/PU data and the Euclidean distance measure to control for location (geographic) and environmental variability. (P<0.05*, P<0.01**, P<0.001***)

Controlled effect	None			Geographical			Environmental		
	Sciomyzidae	Syrphidae	PUs	Sciomyzidae	Syrphidae	PUs	Sciomyzidae	Syrphidae	PUs
Trap scale (n=20)									
Sciomyzidae	1			1			1		
Syrphidae	0.08	1		0.03	1		0.06	1	
PUs	0.44**	0.20*	1	0.41**	0.13	1	0.44**	0.15	1
Patch scale (n=10)									
Sciomyzidae	1			1			1		
Syrphidae	-0.16	1		-0.17	1		-0.16	1	
PUs	-0.03	0.12	1	-0.03	0.09	1	-0.01	0.18	1

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Table 4: Mantel R statistics of assemblage concordance within habitats using the Sørensen distance measure for species/PU data and the Euclidean distance measure to control for location and environmental variability. (P<0.05*, P<0.01**, P<0.001***)

Controlled effect	None			Geographical			Environmental		
	Sciomyzidae	Syrphidae	PUs	Sciomyzidae	Syrphidae	PUs	Sciomyzidae	Syrphidae	PUs
Sedge grasslands (n=10)									
Sciomyzidae	1			1			1		
Syrphidae	0.27*	1		0.15	1		0.20	1	
PUs	0.71**	0.04	1	0.68**	0.13	1	0.71**	0.02	1
Rush grasslands (n=10)									
Sciomyzidae	1			1			1		
Syrphidae	0.35*	1		0.24*	1		0.36*	1	
PUs	0.72**	0.51**	1	0.67**	0.42**	1	0.75**	0.47*	1

5.5. Cross-taxon congruence of richness, abundance and Shannon's entropy

Patterns of cross-taxon congruence were not significant for richness among any of the groups (Table 5). Sciomyzidae and PU abundance was significantly positively correlated across all samples at trap scale (n=20) but not at patch scale (n=10) or within habitat types. Syrphidae and PU abundance was significantly positively correlated, but only within rush dominated wet grasslands. There was also a significantly positive relationship between Shannon's entropy of Syrphidae and PUs across all samples but only at trap scale (n=20).

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424 Table 5: Spearman Rank correlations of richness, abundance and Shannon’s diversity (entropy) of each group. (P<0.05*, P<0.01**, P<0.001***)

	Overall (Trap scale n=20)			Overall (Patch scale n=10)			Sedge Habitats (n=10)			Rush Habitats (n=10)		
Richness	Sciomyzidae	Syrphidae	PUs	Sciomyzidae	Syrphidae	PUs	Sciomyzidae	Syrphidae	PUs	Sciomyzidae	Syrphidae	PUs
Sciomyzidae	/	-0.003	0.261	/	-0.055	0.212	/	-0.6	0.044	/	0.482	0
Syrphidae		/	0.055		/	0.024		/	0.314		/	0.360
PUs			/			/			/			/
Abundance	Sciomyzidae	Syrphidae	PUs	Sciomyzidae	Syrphidae	PUs	Sciomyzidae	Syrphidae	PUs	Sciomyzidae	Syrphidae	PUs
Sciomyzidae	/	0.002	0.543*	/	0.018	0.045	/	-0.067	0.382	/	0.249	0.079
Syrphidae		/	0.310		/	0.503		/	0.552		/	0.746*
PUs			/			/			/			/
Shannon’s H’ (Entropy)	Sciomyzidae	Syrphidae	PUs	Sciomyzidae	Syrphidae	PUs	Sciomyzidae	Syrphidae	PUs	Sciomyzidae	Syrphidae	PUs
Sciomyzidae	/	-0.381	-0.307	/	-0.467	-0.285	/	-0.164	-0.309	/	-0.321	-0.539
Syrphidae		/	0.448*		/	0.587		/	0.224		/	-0.018
PUs			/			/			/			/

426

427 **6. Discussion**

428 Community congruence between dipteran groups examined at low spatial scales and within district
429 habitat types provided the most useful predictors of selected dipteran diversity. Assemblages which
430 showed similar responses to environmental variables and exhibited shared community differentiation
431 based on habitat type were most significantly correlated. Diptera which display limited movement
432 (Sciomyzidae) were more indicative of changes in wider dipteran community structure (represented
433 by PUs) than those which are generally more mobile in nature (Syrphidae). The results highlight the
434 variation that occurs between groups when a range of statistical expressions and scales of observation
435 are utilized in biodiversity indication. The results also suggest compositional analysis of Diptera as
436 the most appropriate approach to the investigation of this overlooked group as previously suggested
437 (Hughes et al., 2000).

438 Assessments of invertebrate cross-taxon taxon congruence in agricultural systems may be limited in
439 spatial resolution so that patterns can be examined at scales such as field or farm level which
440 coordinate with administrative requirements e.g. agri-environmental payments are allocated at such
441 scales (Anderson et al., 2011; McMahon et al., 2012). While this is a pragmatic approach to aligning
442 assessment and monitoring of biodiversity with policy, the distribution of certain organisms such as
443 Diptera may occur at more restricted scales. Sciomyzidae, for example, have been shown to exhibit
444 habitat fidelity across scales as low as 23m (Williams et al., 2010), and Hughes et al. (2008)
445 determined high levels of dipteran community dissimilarity (0.52 Bray-Curtis) between two Malaise
446 traps sited 25m apart in a meadow.

447 Therefore, the evaluation of patterns of congruence between surrogates and wider diversity of such
448 organisms should occur at scales appropriate to the community distribution of the target taxa
449 (McGeoch, 1998). Subsequent monitoring of such groups which take place at fixed sampling sites can
450 then provide accurate insights into the success of typical conservation strategies such as habitat
451 maintenance or restoration; but inclusive of organisms that inhabit areas at lower spatial scales.

452 In the present study, environmental correlations between groups based on axis 1 of the NMS
453 ordination were of most interest as they explained the largest amount of variation owing to the use of
454 orthogonal principal outputs in the analysis. While all three dipteran assemblages showed some
455 congruence in terms of environmental correlates, the strongest relationship was seen between
456 Sciomyzidae and PUs at trap level. Based on the NMS output, Sciomyzidae and PU assemblages were
457 largely influenced by the same environmental variables (% Rush, % Sedge, Soil pH, vegetation height
458 and surface water depth) whereas Syrphidae were influenced by a different combination of
459 environmental factors (% Rush, % Sedge, % Moss, % Forbs, % Moribund). It is unsurprising that

Syrphidae communities responded primarily to variables associated with vegetation type and structure as adults are largely dependent on pollen and nectar resources for food (Ricarte et al., 2011). Sciomyzidae on the other hand are known to respond to factors such as soil moisture, hydroperiod and vegetation type (Maher et al., 2014; Williams et al., 2009b) and exhibit very limited movement within wet grassland habitats (Williams et al., 2010). Their response to factors such as soil pH and water depth suggests that environmental influences that may affect their malacophagous larvae could also influence adult distributions.

The strong correlation between PU and Sciomyzidae assemblages in terms of environmental correlates at trap level not only illustrates that PUs are influenced by comparable environmental factors as Sciomyzidae, but that they also respond at similar scales. Fine scale habitat features are known to affect arthropods in grassland systems, especially more sedentary groups (Cole et al., 2010). Within wet grassland habitats, variations in hydrological regime and grazing patterns are known to have significant influences on dipteran diversity – in particular Sciomyzidae (Maher et al., 2014; Ryder et al., 2005; Williams et al., 2009a). Changes in environmental factors across small spatial scales e.g. between pair-wise traps 20m apart, could therefore have a more marked effect on groups such as Sciomyzidae which are characterised as having limited movement. This response to small scale features could be a result of their reduced ability to respond to disturbances or stressors, or alternatively, it may be an indication of the availability of a specific resource located at finer scales. Factors that influence dispersal of Diptera adults include larval development, sexual behaviour and food requirements (Delettre et al., 1998). Analysis of Syrphidae community differentiation between pair-wise traps within each site was not congruent with Sciomyzidae or PUs but was significantly influenced by changes in the percentage cover of broadleaf herbs. As Syrphidae adults are largely dependent of pollen and nectar as food sources (Sutherland et al., 2001) and patches of increased broadleaf herb cover within sites most likely attracted Syrphidae adults, this is not surprising. Sciomyzidae adults, on the other hand, can be restricted to very small areas characterised by dominant vegetation type (Williams et al., 2010) and their distribution may potentially be influenced by the availability of larval food sources i.e. aquatic and semi-aquatic molluscs which often have very clustered distributions (Knutson and Vala, 2011). Sciomyzidae community differentiation between pair-wise traps was anticipated in our results but interestingly was not correlated with any of the measured environmental variables examined in this study. However, the significant correlation between Sciomyzidae and PU community structure between pair-wise traps strongly suggests that the factors contributing towards community differentiation across small spatial scales for these two groups are similar.

This result may explain why highly mobile adult Syrphidae were not strongly correlated with the other dipteran assemblages in terms of environmental correlates or community congruence between pair-wise traps as they likely respond to environmental heterogeneity at much broader scales

(Hendrickx et al., 2007). In contrast, groups which share a decreased ability to disperse and/or have high habitat fidelity have previously been shown to have increased levels of congruence (Grenouillet et al., 2008). However, such patterns of congruence might only be recognisable at spatial scales where these taxa respond similarly to environmental stressors (McGeoch, 1998).

The partitioning of data into biogeographical units, discrete habitat types or subsets of taxa based on community structure is also known to affect congruence patterns between assemblages showing both increased and decreased correlations (Myřák and Horsák, 2014; Pawar et al., 2007; Rooney and Bayley, 2012). In the present study, all three assemblages showed significant community differentiation between the two habitat types, and community congruence between groups within these two wet grassland habitats were markedly more significant than congruence from groups examined at overall wet grassland level. Wet grassland in Ireland is broadly defined, and generally classified as having >50% cover of grasses, small sedges or rushes, with the remaining vegetation comprised of a mixture of wetland and drier grassland herbs (Fossitt, 2000). However, dominant vegetation type is known to significantly contribute towards dipteran community structure (Hughes et al., 2008) and our results imply that assessments of dipteran diversity in wet grassland habitats should consider sub-categories based on dominant vegetation type. Selecting a biodiversity surrogate requires knowledge of the scale of distribution of the proposed surrogate within the habitat of interest and an examination of its relationship with the predicted taxa below this scale (McGeoch, 1998). In the present study, examining the wet grassland habitats as distinct habitat-types increased community congruence among the groups, in particular between Sciomyzidae and PUs which displayed similar differentiation between the two wet grassland habitats. Strong community congruence between groups that share similar responses to environmental variables and gradients such as habitat change has been previously found among arthropods in grassland habitats (Oertli et al., 2005).

The examination of Diptera community congruence within these habitat categories at the lowest spatial scale provided the most significant insight into relationships between potential biodiversity surrogates (predictor taxa) and the wider assemblage as represented by PUs (predicted taxa). However, while Sciomyzidae and Syrphidae were both useful surrogates of the PU assemblages in rush dominated habitats, only sciomyzids were significantly congruent with PU assemblages in sedge dominated wet grasslands. The result further emphasises the need to include multiple biodiversity surrogates in evaluations and monitoring rather than reliance on one group (Rooney and Bayley, 2012). The results also illustrate the importance of examining community congruence between taxa based on distinct partitioning of communities.

Species richness is largely utilized as a measure of the success of conservation actions (Su et al., 2004) and is a relatively straightforward value to communicate to policy makers. However, changes in the species richness of a target group may not be indicative of the response of a wider suite of

organisms, especially in agricultural systems (Billeter et al., 2008). Correlations between richness, abundance, and Shannon's entropy in this study varied between groups as is generally reported in studies investigating cross taxon congruence (Báldi, 2003; Hayes et al., 2015; Legakis et al., 2004). Only three significant correlations were recorded using these measures, all of which were found when the lowest spatial scale (trap level) was examined. Hayes et al. (2015) reported a significant relationship between Sciomyzidae richness and overall Diptera PU richness in a comparative study of wet grassland conducted at sampling scales similar to the trap level utilized in this investigation; albeit using a sweep net. Though our study was limited to the use of nine families of Diptera to PU level, it is interesting to note that Sciomyzidae abundance and PU abundance was significantly correlated overall at trap level. In general, the richness and abundance of a group tends to be significantly correlated so it could have been expected that if abundance between two groups was significant, then the richness measures would also have been co-correlated. However, an underestimation of PU richness as a result of the conservative allocation of PUs in this study i.e. lumping, may have contributed towards a lack of significance between the richness of PUs and Sciomyzidae. Maher et al. (2014) illustrated patterns that suggested increased richness and abundance of Sciomyzidae was indicative of decreased abundance of Syrphidae in wet grassland habitats. However, these findings were reflective of reduced flowering plant diversity due to increased hydroperiod which positively affected Sciomyzidae but negatively affected Syrphidae.

Syrphidae diversity (Shannon's entropy) and PU diversity (Shannon's entropy) were also significantly correlated; but only at trap scale. It could be considered that the most robust PU measure utilized within these correlations was abundance which was a raw value not subject to interpretation. It is therefore interesting that the most significant correlation among these abundance measures was seen between Syrphidae abundance and PU abundance in rush dominated habitats. This result suggests that within such habitats, areas of rush dominated wet grasslands with high abundances of Hoverflies could be important for maintaining high abundances of other Diptera; a pattern also reflected in the community congruence of these two groups with this habitat type. However, the results also highlight the variability of congruence when using richness, abundance and Shannon's entropy as measures of each group and suggest compositional analysis as a measure of Diptera for surrogacy evaluation.

Agricultural landscapes such as those frequent in the west of Ireland are a mosaic of improved grasslands interspersed with pockets of wet grassland habitat. Realistically, land managers may be required to maintain a suite of wet grasslands whilst others are sacrificed to drainage and/or change of use. Dipteran diversity provides one practical way of maximising landscape-level complementarity in the assemblage conserved. The suite of sites to be preserved, which would maximise gamma diversity of the eleven families studied here, could reasonably be predicted by reference to changes in Sciomyzidae composition dissimilarities (if the sites were *Carex* or *Juncus*-dominated) or with reference to Syrphidae composition (if the sites were *Juncus*-dominated).

7. Conclusion

The inclusion of overlooked groups such as Diptera in conservation strategies is unlikely to reach the thresholds of more iconic invertebrate groups so the use of surrogate taxa is increasingly probable. If such action is required, the surrogates selected for representation of wider dipteran diversity need to reflect the distribution of Diptera at both habitat level and at smaller scales within those habitats. Our results demonstrate that the use of community similarity at low spatial scales and within specific habitat types appears to be the most useful surrogate of dipteran diversity in wet grasslands. However, it may well be most beneficial for conservationists to adopt several invertebrate groups should a surrogate approach be utilized to account for the range of dispersal and distribution patterns of wet grassland invertebrates. While a more extensive study of invertebrates in high nature value farming systems is required before a practical solution to invertebrate diversity surrogacy can be selected, in the case of Diptera within wet grassland habitats, our results suggest that both Sciomyzidae and Syrphidae meet these requirements; though the former appear to represent the predicted taxa (PUs) better in this study. In light of the increasing intensification and abandonment of wet grassland habitats, the inclusion of easily captured, readily identified, and ubiquitous groups as seen in this study may provide essential information on the status of dipteran assemblages within these areas and inform for the selection of areas for conservation. We recommend that considering samples from low spatial scales rather than administrative units such as field or farm level in cross-taxon congruence investigations may highlight patterns of correlation among invertebrate groups which are generally overlooked.

8. Acknowledgements

This research was funded by the Irish Research Council and the Thomas Crawford Hayes Research Grant. We would like to thank the landowners and who accommodated our sampling regime. A special note of thanks to Enora Aubry, Celine Talarczyk and Maxime LeRoy and several other students for their assistance with sampling, sorting and PU allocation.

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