

# Article

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# Two decades of monitoring earthworms in translocated grasslands at Manchester Airport

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# ABSTRACT

Construction of a second runway at Manchester Airport included a mitigation package of habitat restoration with relocation of earthworms as prey items for protected vertebrates. Translocation of turf in blocks was the standard method used for four of five monitored sites with loose soil moved at the other. To assess earthworm communities at these translocated grassland sites, monitoring was undertaken each October (1998-2019) by digging and hand sorting of soil, followed by vermifuge application. Fourteen earthworm species were recorded, representing all ecological groups, but the majority were endogeic species, dominated by Aporrectodea caliginosa. Total earthworm numbers fluctuated during the monitoring period, with lowest density at 4  $m^{-2}$  and highest more than a hundred times larger. The overall mean from all sites across the monitoring period was 151 ind. m<sup>-2</sup>. The differences between sites such as total earthworm numbers and species richness were clearly influenced by the translocation method and specific site topography. Created Hummocks to 3 m for hibernating amphibians proved successful with grassland soil establishing well. Lumbricus terrestris failed to establish due to translocation technique. Using non-metric multidimensional scaling, integration of environmental data with earthworm records showed effects of soil moisture content, pH and rainfall on abundance of ecological groupings and particular species. In general, earthworm community composition was dynamic over the monitoring period suggesting that this and population size needs to be appraised over realistic timescales, which may be best monitored in decades.

# 1. Introduction

## 1.1. Runway 2 mitigation

In the north-west of England, a second airport runway (R2) was constructed 15 km south of the city of Manchester during the late 1990s, after lengthy consultation, and agreement to a £17 million ecological mitigation programme that spanned 15 years [1–3]. Affected habitats covering more than 200 ha were managed using current ecological techniques, such as grassland translocation (February 1998), which would otherwise have been lost below the 3 km runway. The cut turfs of grass (approx. 3 x 1 × 0.15 m) were moved as blocks on flatbed trucks and reinstated intact at receptor areas from where the topsoil had been stripped away. Translocation of this nature may be seen as a last resort during large scale development of valued grassland areas [4]. Here, one

reason for moving these turfs was to maintain the flora, but associated fauna was also of interest. Great crested newt (*Triturus cristatus*) conservation was considered, as local populations were also affected by R2. In addition to new pond creation [5], raised Hummocks (hibernacula for these amphibians) were constructed from rubble and covered with some of the translocated turfs, a relatively novel intervention [6] to provide protection from frost, prevent any possibility of flooding and contain sufficient accessible crevices [5]. Lower lying, wetter grassland (Hollows) was also created with more turfs. Further translocation of grassland at other sites was in support of badger (*Meles meles*) conservation, whose setts were also disturbed by R2 construction. As with the newts, one purpose of translocation was to ensure that adequate food in the form of earthworms was present. Monitoring of earthworms in the translocated grassland areas was initially commissioned to quantify potential food items for these protected vertebrates, as reported by Butt

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**Fig. 1.** (A) Aerial view of Manchester Airport with location of the translocated grassland sites to the south of Runway 2 (H&H – Hummocks and Hollows; S – Slope sites; F – Field); (B) Detail of Hummocks (Hum) and surrounding Hollows; (C) Detail of Slope (S) soil only, and turf transfer; (D) Detail of Field (F) - adapted from Google Maps, 2022.

et al. [7]. After 5 years, it was determined that long-term earthworm monitoring itself was valuable as there is a paucity of such data collection in restoration ecology [8]. To this end, monitoring was continued until 2019.

# 1.2. Turf transfer for earthworms

Turf transfer is a recognised method for introducing earthworms to areas where they may be absent, or abundance is low. This method was pioneered by Stockdill [9] in New Zealand some 40 years ago and is considered particularly successful for endogeic species (e.g., *Aporrectodea caliginosa (sensu* Bouché [10]) that live close to the soil surface. Cocoons and immature specimens of deeper burrowing (anecic) species may also be included in turf transfer, but adults are usually absent. Laying of turf at the receptor sites on to subsoil ought to form a continuum of soil strata and permit earthworm survival. Environmental conditions such as sufficient but not excessive soil moisture, a food source and temperatures within a range permitting annual survival of at least one life stage need to be present for earthworm populations to persist. Conditions provided by turf transfer at Manchester Airport may not in all cases have been conducive for survival of all earthworm species extracted from the donor sites [11].

# 1.3. Aims and objectives

This work began as a short-term annual assessment as part of the R2 mitigation programme but grew into longer-term monitoring of earthworm communities in newly created grasslands with a range of landforms. Aims were to assess the success of this ecological translocation technique (of grass turfs and associated fauna) and the type of structures (e.g., hibernacula) created; to monitor earthworm community attributes at the given sites and, where possible, relate these directly to environmental factors. It was envisaged that dynamism in community development would occur, a form of stability finally reached, with trajectories influenced by nature of soil translocation, the type of landform created and climatic effects. Objectives were: (i) To record earthworm abundance and biomass per unit area, community composition, species richness and diversity; (ii) To compare effects of topography

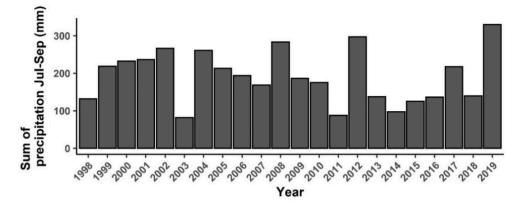


Fig. 2. Precipitation at Sheffield, for three months (July to September) prior to earthworm monitoring (1998–2019).

(Hummocks and Hollows) and soil translocation methods (turf and loose soil) on contiguous areas; (iii) To evaluate the success of the grassland translocation *per se*.

## 2. Materials & methods

# 2.1. Research sites

Four specific grassland translocation areas were identified by Manchester Airport for monitoring (Fig. 1A), with samples collected annually from 1998 to 2019. "Slope" (53° 20' 39.12" N 2° 16' 45.87" W) (Fig. 1C) is a 0.53 ha site located on a 30° sloping bank between R2 and bounded by roads. Slope consists of a part where turfs were translocated intact (Slope turf) as described, and a second where soil, from the same origin as the turfs, was dug out, transferred, and loose tipped (Slope soil). "Hummocks" and "Hollows" (53° 21' 1.92" N 2° 15' 51.94" W) (Fig. 1B), occupy 0.82 ha and are close to ponds constructed for amphibians. Hummocks consist of raised mounds to 3 m above the surrounding area, contain voids to act as potential hibernacula for great crested newts and were covered with a layer of translocated grass turfs. The surrounding Hollows are lower lying and were initially subject to inundation by water. These were created close to ponds to act as corridors for newts to the hibernacula. The turfs and soil that formed Slope, Hummocks and Hollows all came from the same donor site, a disused brickworks site at Oversleyford. This previously disturbed clay soil had a pH of 7.8 and supported flora including common spotted orchid [1]. Hummocks and Hollows were at times grazed by cattle or horses. A further translocation site of 0.3 ha ("Field"; 53° 20′ 14.16″ N 2° 17' 1.66" W) (Fig. 1D) is adjacent to pasture and a wooded part of the River Bollin Valley. It was constructed using floristically species-rich turfs from an undisturbed semi-natural valley side neutral grassland site and initially had a pH of 7.1 [1,7]. During the monitoring period this grassland (Field) was grazed by sheep.

# 2.2. Monitoring

Sampling for earthworms occurred annually during the second week of October but ceased after 2019 due to restrictions associated with the Covid-19 pandemic. During sampling,  $10 \times 0.1 \text{ m}^2$  quadrats were examined at each grassland translocation site. Soil was dug to 20 cm and hand-sorted in the field on plastic sheeting. Thereafter, a mustard vermifuge (5 g l<sup>-1</sup>) was applied to the pit created [12]. All earthworms collected were preserved in 4% formalin and subsequently identified using Sims and Gerard [13] and the nomenclature of Sherlock [14]. Notes were also recorded of other significant fauna in the soil and a subjective assessment taken of the plants at the site. No quantitative data was recorded for vegetation, but photographic records were made of, for example, vegetation development (Supplementary material 1). Soil samples were collected from sites across the monitoring time frame and subjected to analysis for gravimetric soil moisture content (%) (for 13 out of 22 years) and pH (available years: 1998, 2012, 2014, 2018) using standard techniques [15].

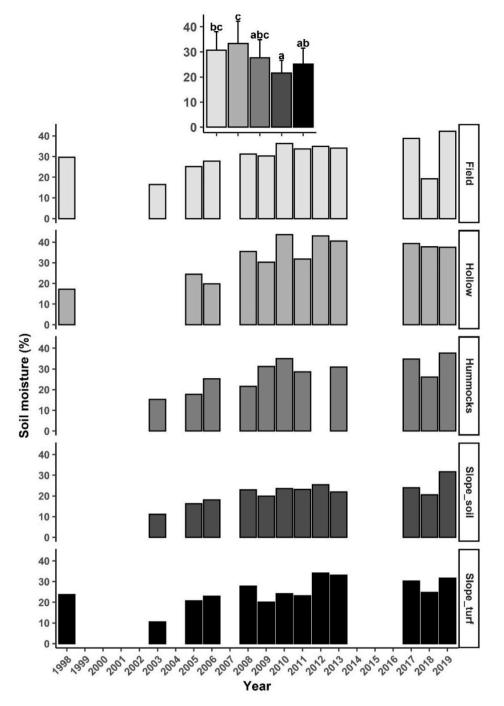
#### 2.3. Climate data

Manchester Airport had its own meteorological recording station, but data collection ceased in 2003. Therefore, a proxy for this was used from Sheffield ( $53^{\circ} 22' 24.6'' \text{ N} 1^{\circ} 29' 24'' \text{ W}$ ; less than 55 km east of the airport), as data from the latest available 15 years (1989–2003) showed a close correlation between recorded rainfall at the two sites (r = 0.913) [16,17]. Fig. 2 shows rainfall data for the three months before monitoring (July–September) over the period 1998–2019. Data were also available for maximum temperature and sunshine hours. Rainfall, maximum daily temperature and sunshine hours were summed for September (rain\_1; tMax\_1; Sun\_1), for August and September (rain\_2; tMax\_2; Sun\_2) and for July to September (rain\_3; tMax\_3; Sun\_3).

# 2.4. Statistical analyses

Earthworm species richness was derived from the mean of counted earthworm species sample<sup>-1</sup> site<sup>-1</sup> and Shannon index was assessed by package 'vegan' [18] with function 'diversity'. All parameters, except Shannon index and soil moisture were square root transformed (sqrt). All parameters were analysed using linear mixed models (LMM) with package 'nlme' [19] and function 'lme' using the residual maximum likelihood (REML) method in R [20]. One-way LMM (1-way LMM) included sites (5 levels; Field, Hollow, Hummock, Slope turf, Slope soil) as fixed factor, except for Allolobophora chlorotica (3 levels; Field, Hummock, Hollow), and random effects were fitted per year (22 levels; 1998 to 2019) and samples (N = 5-10) with compound symmetry variance-covariance structure for repeated measurements [21,22]. For specific changes between years, a two-way LMM (2-way LMM) was applied with fixed factors site, year, and random factor sample. Function 'anova.lme (type = marginal)' was employed for analyses of variance (ANOVA) using Wald-type F-tests and type III hypotheses and Tukey post-hoc test for pairwise comparisons (P < 0.05) with package 'emmeans' and function 'emmeans' [23]. Residual distribution was revised visually by using QQ-plots and for homogeneity of the variance, residuals were plotted against fitted values. All data provided are mean values and standard deviation (mean  $\pm$  SD).

Due to low numbers of earthworms collected for some species, only *A. caliginosa, Aporrectodea rosea, Aporrectodea longa* and *Lumbricus rubellus* were selected for detailed analyses. *A. chlorotica* was found in sufficient numbers but could only be analysed at three sites due to a failed check of assumptions with or without transformation (log or sqrt). Data for *Lumbricus terrestris* was not analysed, because of low



**Fig. 3.** Gravimetric soil moisture content (%) at 0–15 cm in October of 1998–2019 at five sampling sites. Inserted plot shows related overall mean and 1-way LMM with fixed factor site (P < 0.01) and degrees of freedom = 4, F = 5.21.

abundance, but is presented to offer a more complete species spectrum.

# To integrate environmental data (soil moisture, rainfall, temperature and sunshine) plus soil pH for given years, ordination for rank orders were based on the abundance of species or ecological groups and was obtained by non-metric multidimensional scaling (NMDS) [24,25] with function 'metaMDS' (package 'vegan') and Bray-Curtis distances. Ordination was solved with two or six dimensions for ecological groups or species respectively with a stress score of <0.1 after an interaction of 20 tries [26,27]. Vector fitting was performed by function 'envfit' (package 'vegan') with scaling 'species' for earthworm species or ecological groups and 'site' for environmental parameters per site and years.

# 3. Results

# 3.1. Environmental data

After 2003, a dry year, a general trend of increasing soil moisture was recorded, with another fall at most sites in 2018 (Fig. 3). Across the monitoring period, Slope\_soil ( $21.6 \pm 5.11\%$ ) had the lowest level of soil moisture, with Hollows ( $33.4 \pm 8.86$ ; P < 0.05) the highest. Soil pH at Slope\_soil reduced during monitoring from above neutral to 6.2 by 2018. At Field, the pH of turf from a different origin reduced from around neutral to 5.5. The other three sites retained a soil pH of 7.5–7.8 throughout monitoring (data not shown).

#### Table 1

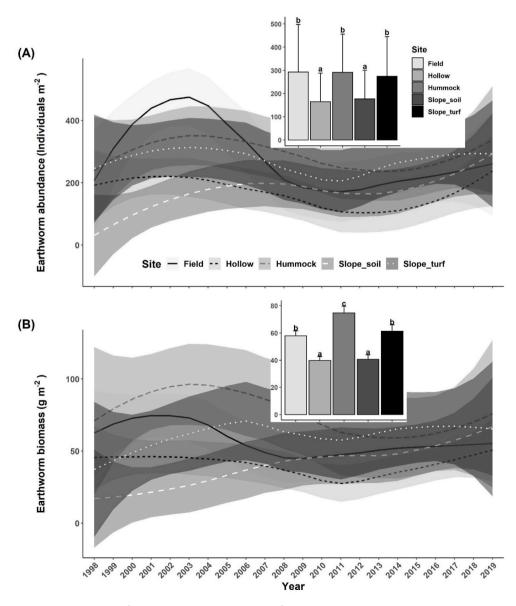
Proportion of earthworm species at Manchester Airport from 1998 to 2019 by abundance and biomass across all sampled sites (Ecological categories: Endogeic (En); Anecic (An); Epigeic (Ep)).

Earthworm species (Ecol cat.)	Abbreviation	Proportion of earthworm		
		abundance	biomass	
Aporrectodea caliginosa (En)	Acal	0.442	0.317	
Allolobophora chlorotica (En)	Achlo	0.063	0.030	
Aporrectodea icterica (En)	Aicter	0.020	0.032	
Aporrectodea longa (An)	Along	0.097	0.224	
Aporrectodea rosea (En)	Aros	0.213	0.075	
Eiseniella tetraedra (Ep)	Etetr	0.002	0.000	
Lumbricus castaneus (Ep)	Lcast	0.006	0.003	
Lumbricus rubellus (Ep)	Lrub	0.096	0.168	
Lumbricus terrestris (An)	Lter	0.019	0.099	
Murchieona muldali (Ep)	Mmuld	0.008	0.001	
Octolasion cyaneum (En)	Ocyan	0.020	0.041	
Octolasion lacteum (En)	Olact	0.014	0.012	

#### 3.2. Earthworms

Sampling over 22 years produced 14 species of earthworm, twelve of which are shown in Table 1. In addition, specimens of *Bimastos rubidus* and *Dendrobaena octaedra* were found once (2017 in Slope\_soil) and twice (2019 in Slope\_turf and in Field), respectively, but not considered in the statistical analyses. Most earthworms found were endogeic species (Table 1). Total earthworm abundance fluctuated during the monitoring period, with lowest density at 4 m<sup>-2</sup> and highest more than a hundred times larger. Similarly, biomass ranged from 1 g to 110 g m<sup>-2</sup>. Statistically, Hollows and Slope\_soil produced less earthworms in total (170  $\pm$  5.5 indiv. m<sup>-2</sup>) than the other three sites (286  $\pm$  7.79 indiv. m<sup>-2</sup>; Fig. 4A). For total biomass, Hummocks (9.1  $\pm$  6.07 g m<sup>-2</sup>) significantly exceeded all site results (5.96  $\pm$  0.57 g m<sup>-2</sup>; Fig. 4B), with Hollows (3.93  $\pm$  3.38 g m<sup>-2</sup>) the lowest of all sites. Interactions of year and site were significant for both earthworm biomass and abundance (*P* < 0.0001) (Table 2A).

Throughout monitoring, the species richness at the sites ranged from a minimum of 2 (Slope\_soil in 2003) to a maximum of 9 (Field in 2012).



**Fig. 4.** (A) Earthworm abundance (Individuals m<sup>-2</sup>) and (B) earthworm biomass (g m<sup>-2</sup>) from 1998 to 2019 at five sampling sites. Inserted plots show related means over 22 years of 1-way LMM with fixed factor site (P < 0.001) and degrees of freedom = 4.  $F_{\text{biomass}} = 36.7$ ;  $F_{\text{abundance}} = 35.5$ . Sites having no letter in common are significantly different by pairwise comparison (Tukey; P < 0.05). Mean  $\pm$  SD, N = 5–10.

#### Table 2

ANOVA results of 2-way LMM of (A) total earthworm abundance (Individuals m<sup>-2</sup>), total earthworm biomass (g m<sup>-2</sup>), species richness, diversity and (B) species abundance (Individuals m<sup>-2</sup>) with fixed factors site (S; Field, Hollow, Hummock, Slope\_turf, Slope\_soil) and year (Y; 1998 to 2019). Degrees of freedom: S = 4; Y = 21; S × Y = 84; N = 5–10.

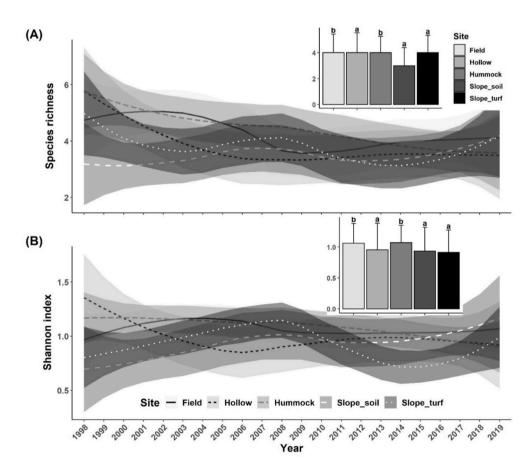
(A)			(B)					
Parameter	<i>F- P-</i> value value		Species Abundance	F- value	P-value			
Earthworm abundance			Acal					
S	0.821	0.512	S	0.268	0.899			
Y	11.3	< 0.0001	Y	7.97	< 0.0001			
S  imes Y	4.38	< 0.0001	S   imes  Y	3.62	< 0.0001			
Earthworm			Aros					
biomass								
S	0.667	0.615	S	2.85	0.023			
Y	4.48	< 0.0001	Y	6.34	< 0.0001			
S  imes Y	3.11	< 0.0001	$S \times Y$	3.05	< 0.0001			
Species richness			Along					
S	0.379	0.824	S	0.579	0.678			
Y	4.19	< 0.0001	Y	3.84	< 0.0001			
S  imes Y	3.36	< 0.0001	$S \times Y$	3.04	< 0.0001			
Shannon index			Lrub					
S	0.54	0.707	S	0.351	0.844			
Y	2.24	0.001	Y	1.76	0.019			
$S  \times  Y$	2.85	< 0.0001	$S\timesY$	4.56	< 0.0001			

Field and Hummocks  $(4.2 \pm 0.06 \text{ species site}^{-1})$  contained more species than the other 3 sites  $(3.36 \pm 0.11 \text{ species site}^{-1})$  considered over the monitoring period (Fig. 5A). Table 2A shows that although site was not significantly different for species richness, it was for year and for

interaction between site and year.

Community composition changed throughout monitoring and some species present in early years, such as Eiseniella tetraedra (Hollows site only; Table 3) were not recorded after 2005. By contrast, certain species were not recorded during the first years of monitoring but appeared thereafter, e.g., Murchieona muldali and Aporrectodea icterica, which were first noted in 2004 (Field) and 2006 (both Slope sites), respectively. Shannon indices (Fig. 5B) showed that the Field and Hummocks (1.07  $\pm$  0.01H) were more diverse than the other 3 areas (0.93  $\pm$ 0.02H), but overall, there was little change in community diversity over the monitoring period. At the start of monitoring (1998) the sites did not differ in species richness or diversity but in 1999, Slope\_turf and Slope\_soil had lower species richness compared to Hollows (P < 0.05; Tukey; Fig. 5A). For diversity, Slope turf was lowest compared to Hollows and Hummocks (P < 0.05; Tukey; Fig. 5B). Overall, Shannon diversity was not significant for site but was for year and the interaction between site and year (Table 2A).

Table 1 shows that A. caliginosa (0.44), A. rosea (0.21), A. longa (0.10) and L. rubellus (0.10) accounted for 85% of all earthworms collected across all sites during monitoring. The abundance of each of these four species was therefore examined individually (Fig. 6). For all four of these earthworm species (Table 2B), interaction between site and year was important. Year was significant for all three *Aporrectodea* species and for L. rubellus. Only for A. rosea was site alone also important. Over the whole of the monitoring period, both A. caliginosa and A. rosea occurred least frequently at the wetter Hollows and Slope\_soil sites (Fig. 6A; B), with numbers of A. rosea generally decreasing. Abundance of A. longa fell at all sites after the first decade, but overall, more were present at the Hollows and Hummocks sites (Fig. 6C). By



**Fig. 5.** (A) Species richness and (B) Shannon index from 1998 to 2019 at five sampling sites. Inserted plots show related means over 22 years of 1-way LMM with fixed factor site (P < 0.001) and degrees of freedom = 4.  $F_S = 14.3$ ;  $F_H = 7.4$ . Sites having no letter in common are significantly different by pairwise comparison (Tukey; P < 0.05). Mean  $\pm$  SD, N = 5–10.

Table 3

Earthworm	Field	Field		Hollow		Hummock		Slope_soil		Slope_turf		Overall	
species	Mean	$\pm$ SD	Mean	$\pm SD$	Mean	±SD	Mean	$\pm$ SD	Mean	$\pm SD$	Mean	$\pm SD$	
Acal	71.8	63.8	40.0	39.4	81.6	57.6	43.4	50.6	86.6	71.7	64.7	56.6	
Achlo	17.8	25.1	13.8	22.4	2.86	9.39	2.64	6.37	1.70	6.24	7.76	13.9	
Aicter	4.93	21.12	0.05	0.72	0.05	0.71	6.60	14.00	5.94	16.0	3.52	10.5	
Along	10.8	15.7	18.5	18.5	22.3	24.1	4.83	8.85	8.15	13.7	12.9	16.2	
Aros	53.7	51.5	17.3	21.2	25.01	33.11	19.1	22.9	36.5	52.1	30.3	36.2	
Aspp	2.89	19.09	0.412	4.18	0.510	5.80	0.47	4.00	1.23	8.36	1.10	8.28	
Etetr	0.00	0.00	1.13	8.50	0.00	0.00	0.00	0.00	0.00	0.00	0.227	1.70	
Lcast	1.37	5.39	0.57	3.97	1.17	4.07	0.19	1.94	0.66	3.46	0.793	3.77	
Lrub	3.89	9.36	4.95	10.6	26.5	31.1	21.2	26.0	20.5	24.0	15.41	20.2	
Lspp	7.44	13.8	3.66	7.79	8.62	16.6	5.24	15.1	5.38	19.9	6.07	14.7	
Lter	3.42	6.46	1.86	5.54	4.05	7.64	0.75	3.29	2.49	7.01	2.51	5.99	
Mmuld	1.52	6.87	0.258	2.14	0.61	4.00	2.17	7.56	2.45	13.2	1.40	6.75	
Ocyan	6.22	11.0	0.670	3.68	2.14	8.20	2.64	6.66	1.89	5.87	2.71	7.08	
Olact	1.00	5.56	2.06	6.82	4.34	12.6	0.19	1.37	1.70	6.24	1.86	6.51	
Overall site mean $\pm$ SD	292	205	165	124	291	165	176	123	275	171			

Species abundance (Individuals  $m^{-2}$ ) by site over 22 years and overall mean per species and per site. N = 5-10 (earthworm species abbreviations as for Table 1).

contrast to the *Aporrectodea* species examined, *L. rubellus* had very few individuals present in the Hollows or the Field, with significantly more throughout at the Hummocks and the two Slope sites (Fig. 6D).

Trends in the abundance of *L. terrestris* at the monitored sites differed for this species but generally, after a few increases, most decreased to a very low level (Fig. 7A). For *A. chlorotica* numbers mainly remained steady, except at Hollows where they decreased over the first decade before climbing towards the end of monitoring, and at Field where numbers grew initially before dropping and then increasing once again (Fig. 7B). Overall, Field and Hollows had higher abundance than Hummocks.

# 3.3. Earthworms and the environment

For all environmental data, there were only very weak correlations of  $R^2$ , smaller than 0.05, but nonetheless, NMDS of ecological grouping (Fig. 8A; B) showed a clear separation between sunshine hours, maximum temperature and rainfall along NMDS1, except tMax\_2 +\_3; Sun\_3. The only significant parameters were rain\_1; rain\_2; tMax\_1; soil moisture (P < 0.001; 0.025; 0.031; 0.001 respectively) and soil moisture was closely aligned with rain\_1 + \_2 and weakly negatively related to Sun 1 + 2.

Fig. 8A shows some separation for the sites with anecic species more prevalent in the Hollows site (as represented by the dominance of *A. longa* within this ecological grouping and at this site (Fig. 8C)). Endogeics cluster across all sites, as recorded by the almost ubiquitous nature of this group across all sampling years and sites. *O. cyaneum, A. rosea* and *A. chlorotica* were more associated with Field. Epigeics were drawn more to the Slope\_soil site and typify the large number of *L. rubellus* found there.

From Fig. 8D, species abundance was independent of environmental vectors, similar to ecological grouping (Fig. 8B) with  $R^2 < 0.05$ . However, significant vectors were tMax\_1, tMax\_2, Sun\_1, Sun\_2 and soil moisture (P < 0.007). With consideration of years with soil pH values (4 years only, Fig. 8F), the environmental vectors of soil moisture and pH were more important ( $R^2 = 0.15$  and 0.2, respectively). Here, a clearer separation of Hollows was then seen from Field and both Slope sites with respect to soil moisture content. Earthworm species assignment to sites was little changed, except for *A. chlorotica* which showed a greater association to Hollows.

# 3.4. Vegetation

At the Hummocks, woody plants began to appear on the grassland relatively rapidly and by 2007, small hawthorn (*Crataegus*) and willow (*Salix*) were present with a thick covering of bramble (*Rubus*), which had to be beaten back to allow sampling. On request, these were cut back by contractors in spring 2011 and the grassy areas restored. In 2013, two Great Crested Newts were collected unharmed from soil pits when sampling for earthworms on Hummocks. After 2011, this site had no further direct intervention, but remained mainly tree-free due to sporadic grazing by cattle and horses. The most dramatic vegetation change was recorded at Slope sites where, after 2011, the grassland began to succeed to woodland, dominated by birch (*Betula*), with gorse (*Ulex*) also present on the slope\_soil part. Nevertheless, by 2019 it was still possible to locate open grassland areas for sampling. The nature of the Hollows also changed over the 22 years, with a reduction in the quantity of rushes (*Juncus*) present, particularly after 2007. No major changes in vegetation were recorded at Field.

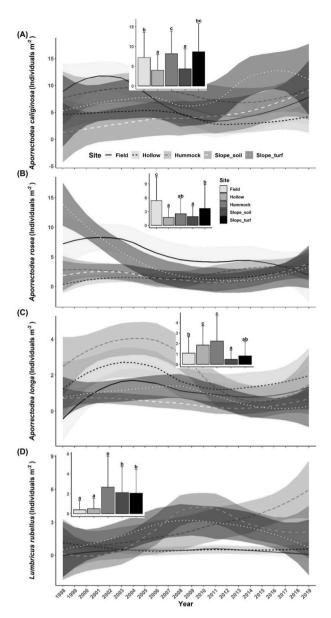
#### 4. Discussion

# 4.1. Food for vertebrates

For food provision of protected amphibians, the transfer of grass turfs at R2 of Manchester Airport could be considered successful. Newts in their terrestrial phase would certainly have gained from the smaller, shallow working (endogeic) earthworm species present in large numbers. Great created newts, discovered in soil pits when sampling for earthworms on the Hummocks, showed that the constructed hibernacula had been successfully colonised for overwintering, as designed [5,6]. However, total earthworm numbers across all sites, 105-187 ind. m<sup>-2</sup>. were much lower than those reported from old English pasture [28]; 390-470 ind.  $m^{-2}$  with a biomass of 52-110 g  $m^{-2}$ . This is likely a function of the immature nature of these soils following the translocation process and the failure of larger (anecic) species to become established. Provision of earthworm food for badgers was therefore not so successful as they primarily prey on L. terrestris and an individual badger can consume from 130 to 200 mature worms from pasture in a single evening [29]. Populations of this deep burrowing species tended to decline in all the translocated grasslands.

# 4.2. Earthworms and the environment

Results showed that annual conditions had a profound effect on measured and derived earthworm attributes. Comparisons of earthworm abundance showed strong differences across the monitoring period with reduced numbers particularly in 2003, 2008, 2011 and 2018. Of these years, all but 2008 occurred when rainfall data and soil moisture readings were appreciably lower. Across much of Europe [30,31] and specifically in the UK, 2003 registered the driest February to October period since 1921 [32] and was therefore a drought. It has been demonstrated that the activities of earthworms are drastically affected by rainfall, with drought as the main factor when viewed across a selection of European



**Fig. 6.** Species abundance (Individuals m<sup>-2</sup>) of (A) *A. caliginosa*, (B) *A. rosea*, (C) *A. longa* and (D) *L. rubellus* from 1998 to 2019 at five sampling sites. Inserted plots show related species abundance over 22 years of 1-way LMM with fixed factor site (P < 0.001) and degrees of freedom = 4.  $F_{Acal} = 28.2$ ;  $F_{Aros} = 31.1$ ;  $F_{Along} = 32.7$ ;  $F_{Lrub} = 69.7$ . Sites having no letter in common are significantly different by pairwise comparison (Tukey; P < 0.05). Mean  $\pm$  SD, N = 5–10.

agroecosystems [33]. Global modelling [34], has shown that earthworm abundance, biomass and species richness, are attributes that are all considerably impacted negatively by low level of precipitation, more than soil properties, as might be anticipated. Long-term investigations, as here, may assist in determination of factors causing such annual fluctuations in earthworm data, but more frequent sampling throughout the year might also be advantageous.

A combination of earthworm data from two decades with available environmental data allowed for some separation of effects at the given grassland translocation sites. Nevertheless, results showed few major relationships between earthworm numbers and measured environmental factors. One exception was with *A. chlorotica*, a species renowned for a tolerance to wet soils [35], with high numbers recorded in Field and Hollows aligning with a large environmental soil moisture vector. For overall earthworm abundance, specific years had an important effect. Even though e.g., precipitation and soil moisture records were markedly different over time, these resulted in small discernible environmental effects. Site specific differences were more apparent with better conditions in Field and less so on Slopes sites.

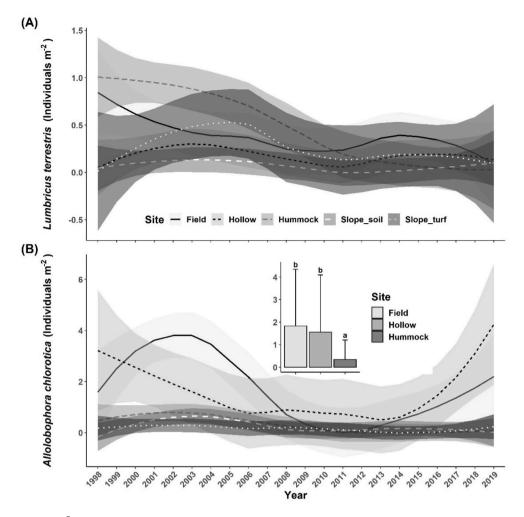
# 4.3. Topography and translocation technique

Type of soil translocation was directly compared at the two Slope sites and showed that initial numbers of earthworms were dramatically lower when soil alone was loosely tipped compared with adjacent turfs, both deposited on to exposed subsoil. Earthworm abundance at Slope\_soil required the whole monitoring period to approach the numbers found at Slope\_turf. A similar situation was recorded for earthworm biomass, but here the two Slope sites were comparable after a decade. This was despite a decrease in soil pH at the Slope\_soil site from above neutral to 6.2 at final sampling, which may have partially resulted from a growing dominance of gorse, an invasive species, known to severely reduce soil pH [36]. The high proportion of *L. rubellus* found on the Slope sites is testament to a tolerance of lower soil pH [13] and colonisation ability. This pioneering species has been shown to rapidly invade restored soils, by comparison to other earthworms [37], can disperse at 7–14 m y<sup>-1</sup> and likely arrived from surrounding tree-covered areas.

Results strongly advocate the use of turf stripping and transfer over the translocation of loose soil to promote earthworm abundance [8], although the cost of the latter on a large scale may prohibit its use [4]. Nevertheless, this is a technique that may not have been investigated as fully as it may warrant [38]. Any translocation as part of a mitigation scheme needs to be fully justified and must consider aspects that include species ecology, logistics and cost of the action.

Site topography of the Hummocks and Hollows, deliberately created for amphibian usage, also influenced earthworm-related results. The Hummocks held a greater abundance and biomass of earthworms throughout the monitoring period. This may seem counter intuitive, given drought years, but here the high (>40%) soil moisture content of the Hollows may have been detrimental to the existence of many common endogeic earthworm species, such as A. caliginosa and A. rosea, and certainly to some epigeics such as L. rubellus. The latter is sensitive towards flooding, with escape and avoidance behaviour recorded as main mechanism of survival in laboratory experiments [35]. Hollows also suffered from the 2003 drought conditions, when numbers of moisture-loving A. chlorotica fell dramatically and semi-aquatic E. tetraedra went unrecorded and were not seen at all after 2005. Data from comparative, if smaller, raised and lower adjacent systems (abandoned ridge and furrow agroecosystems in NW Scotland [39], also pointed to similar numbers and biomasses of earthworms in the two soil components. These findings were also ascribed to changes in soil moisture levels with annual extremes (wet and dry) proving negative to earthworm abundance.

A further factor that may have influenced earthworms on the grassy Hummocks site was management of woody growth and brambles in 2011. After this, earthworm abundance and biomass both increased. Such aboveground management of plants may seem unlikely to affect earthworms, but Butt and Briones [40] found that scrub management had a significant positive effect on earthworms by promoting numbers, biomass, and species richness. This was at a post-industrial site in NW England where the earthworm community was dominated by endogeic species, as here. Succession of grassland to woodland was most dramatic at the Slope sites which saw the open area slowly covered by birch and gorse over the final decade of monitoring. This also indicates that mitigation measures put in place need to be managed long-term. Nevertheless, this provided an opportunity to record changes to earthworm community composition, e.g., with A. rosea largely replaced by A. icterica. Over the longer-term, further soil and earthworm sampling, in what was translocated grassland, may reveal a community change towards woodland-associated species, as latterly seen with the initial presence of B. rubidus and D. octaedra. Earthworm community change



**Fig. 7.** Abundance (Individuals m<sup>-2</sup>) of (A) *L. terrestris* and (B) *A. chlorotica* from 1998 to 2019 at five sampling sites. Inserted plot shows related species abundance of *A. chlorotica* over 22 years of 1-way LMM with fixed factor site (3 levels; Field, Hummock, Hollow; P < 0.001) and degrees of freedom = 2. F = 14.1. Sites having no letter in common are significantly different by pairwise comparison (Tukey; P < 0.05). Mean  $\pm$  SD, N = 5–10.

has previously been recorded associated with primary vegetation succession, across sand dune systems [41] or within secondary succession of abandoned fields [42] and although relatively stable communities may ultimately be found, future climate changes may reveal more dynamic situations.

The failure of *L. terrestris* to become firmly established may have been a result of low initial inoculum, as adults would have retreated deeper into burrows during turf extraction. Transfer of cocoons and hatchlings would have occurred, but the shallow nature of the turf and a failure to fully integrate with the sub-soil would not have assisted establishment. When sampling, a clear division of turf and sub-soil was still apparent in places after more than a decade. *L. terrestris* burrows ought to have assisted in a unification of the soil strata, but seemingly did not in many areas. *L. terrestris* numbers may grow and the influence of this K-selected earthworm species [43] on soil properties and as a food source for badgers may increase with time, but current trends suggest otherwise.

# 4.4. Lessons learned

Grassland turf translocation, with creation of appropriate receptor landforms (raised hibernacula and wetlands), can promote amphibian conservation, with the presence of earthworms seen as vital due to the ecosystem services that they provide in functioning soils [44]. Nevertheless, these grasslands need appropriate management, through grazing or cutting to prevent vegetation succession to woodland. In addition, water levels need to be monitored and potentially adjusted if amphibians are the target species.

The current work showed that earthworms present in translocated turfs could promote sustainable communities over time, even if at relatively low abundance. The dynamic nature of the earthworm communities was determined mainly by soil moisture content, itself a function of precipitation. Earthworm species have markedly different survival strategies to changing soil conditions and those, such as *A. caliginosa*, which show little response to flooding [35], are likely to fare well.

The long-term nature of this study has shown benefits and, if resources allow, could be replicated in other works. Should future investigations of grassland translocation be undertaken, they might also usefully monitor nearby grasslands, not subjected to mitigation measures, to provide genuine control data and allow effects of climate and translocation to be more easily separated. Nevertheless, it is suggested that further monitoring of these R2 sites e.g., every 5 years, could still provide valuable data of earthworm community dynamics in translocated grasslands.

# Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence

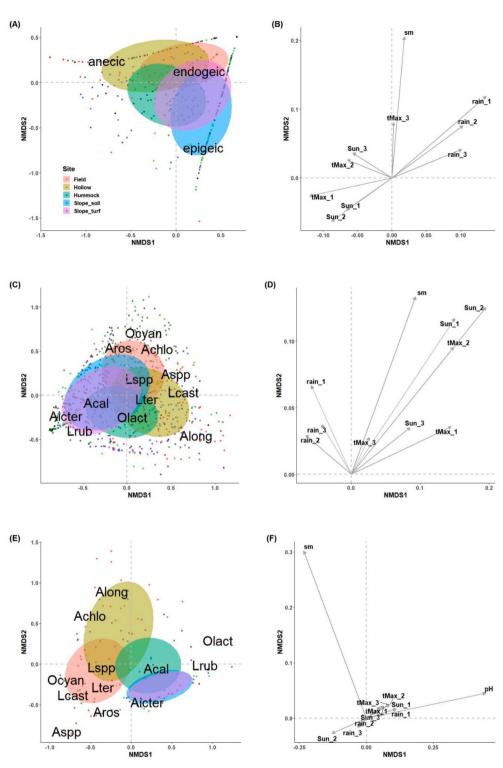


Fig. 8. Nonmetric multidimensional scaling (NMDS) of (A) ecological groups of earthworms and (C) earthworm species found at five sampling sites at Manchester Airport from 1998 to 2019 and (B, D) site parameters, respectively. Site parameters include sum of precipitation of September (rain\_1), August and September (rain\_2), July to September (rain\_3), maximum temperature °C (tMax\_), sunshine hours (Sun\_) accordingly and soil moisture at sampling date (sm). Additionally (E, F), soil pH is included from available years (1998, 2012, 2014, 2018).

the work reported in this paper.

# Data availability

Data will be made available on request.

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# Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ejsobi.2022.103443.

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