Does Rewilding Benefit Dung Beetle Biodiversity?

A case study comparison of rewilding land in West Sussex and nearby organic farmland.

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A case study comparison of rewilding land in West Sussex and nearby organic farmland.

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Abstract

There is growing concern about the conservation of dung beetles in the UK with many species in decline due to various threats, including habitat loss and fragmentation, loss of continual grazing and use of endectocides. Support for rewilding as a solution to agricultural land abandonment, and also as a tool for conservation management to protect biodiversity, is expanding in the UK and throughout Europe. Studies suggest that rewilding as a naturalist grazing model could benefit dung beetle biodiversity but there exists limited data to support this theory. This research investigates the idea further by undertaking a comparative survey study of two rewilding sites at Knepp Castle Estate in West Sussex and two nearby organic farms in order to measure dung beetle biodiversity (using species richness, abundance and evenness) between the two models of land management. Over 12,000 dung beetles were collected at four different sites and identified into three genus groups: Aphodiidae (77%), Geotrupidae (15%) and Onthophagus (8%). Results showed a significant difference between the two models, with higher biodiversity overall at the two rewilding sites than at the organic farms, thus supporting the proposition that rewilding is beneficial for dung beetle biodiversity. The results support the case for small-scale rewilding as a driver for biodiversity and its possible integration into UK agricultural policy for future sustainability.

Key words

Aphodius, Biodiversity, Dung beetles, Knepp Castle Estate, Knepp Wildland Project, Onthophagus similis, Organic Farmland, Rewilding.

Introduction

Dung Beetles

Dung beetles can be found worldwide across a range of geographical landscapes. Their sub-families, Scarabaeinae, Aphodiinae and Geotrupidae, contain three behavioural groups: dwellers (Endocoprids), tunnellers (Paracoprids) and rollers (Telecoprids). Only dwellers and tunnellers can be found in the UK and are dominated by the dweller genus *Aphodius* (Hanski & Camberfort, 1991). Two thirds of these species breed in pastures linked with the traditional farming practices associated with domestic livestock, and the majority of these species are generalist coprophage feeders, although some show preference for certain dung types (Hanski, 1986; Hanski & Camberfort,1991). The evolutionary resource requirements of dung beetles and their preference variations are strongly linked to the diversification of mammals throughout the ages. Dung beetles have continuously adapted alongside the mammals on which they depend (Nichols et al, 2008, Hanski & Camberfort, 1991). Distribution of different dung beetle groups is largely dependent on climate conditions with local and microhabitats determining community assemblage and factors such as vegetation, soil type and seasonality affecting species diversity (Hanski & Camberfort, 1991; Giller,1996; Hortal et al, 2011).

Dung beetles provide important ecological services to agricultural landscapes in a variety of ways. For example, a key service they provide is the breakdown and removal of dung, which helps prevent the build-up of unsuitable pastures and the spread of disease (Gittings et al, 1994). The Australian dung beetle project (1965-1985) which introduced South African and European dung beetles to Australia not only led to improved quality of cattle pastures but reduced the population of pestilent bush flies by around 90% (Doube, 2018; Edwards et al, 2015). Dung beetles also provide a host of nutrient cycling activity including the sequestration of carbon and nitrogen directly into the soil, and the recycling of phosphates

found within animal dung. Furthermore, they enhance soil structures by having a positive influence on the hydrological properties of soil, increasing water infiltration and soil porosity, as well as reducing surface runoff water (Nichols et al, 2008; Manning et al, 2016; Brown et al, 2010). One study has estimated that, without dung beetle activity, the cost of nitrogen loss to the US would be approximately \$58 million a year (Losey & Vaugha, 2006).

Arguably, one of the biggest contributions made by dung beetles is to the cattle industry, by acting as biological control agents for gastrointestinal parasites of livestock. Many cattle parasites require dung to complete their larvae cycles and burying infected dung can considerably reduce the density of these parasites (Fincher, 1981). Studies show a significant reduction of *Ostertaia osteragi* (stiles) larvae as a result of the burying activity of dung beetles (Fincher, 1973). It is estimated that the economic value of dung beetles to the UK cattle industry is £367 million per year, with control of gastrointestinal parasites as a key contributing factor (Beynon et al, 2015). Dung beetles also act as an important food source for hundreds of birds, mammals, reptiles and amphibians (Young, 2015). The genus *Aphodius* are an important food source for the currently threatened Greater Horseshoe Bat (Flanders & Jones, 2009). Evidence suggests that functionally diverse dung beetle assemblages deliver a multitude of ecosystem services, highlighting the potential importance of species rich communities (Manning et al, 2016).

Dung beetles are also seen as a valuable tool by which to measure biodiversity and habitat change (Davis et al 2001; McGeoch et al, 2002) and are increasingly being recognised as key indicator species (Davis et al, 2004). This is because they have the key characteristics of an ideal focal taxon, including adaptability to standardized sampling, taxon accessibility and wide geographical distribution, and are susceptible to environmental changes (Spector, 2006). Their significance within biodiversity monitoring and ecological research worldwide is becoming established, including their possible influence on the effects of greenhouse gases (Verdu et al, 2000; Davis et al, 2004; Spector, 2006; Penttilä et al, 2013 Slade, 2016).

It is clear that dung beetles provide important ecological services within a variety of habitats inter-continently, including a substantial contribution to landscapes in the United Kingdom.

They are also important indicator species that have the potential to further our understanding of changes in the natural world around us.

Despite their importance and ecological contributions, limited research has been conducted on UK dung beetles. Dung beetles are in decline throughout Europe and this is strongly associated with habitat loss, degradation, and fragmentation. The replacement of traditional cattle and sheep farming by either intensive agriculture practices or reforestation is a likely contributor to regional declines in dung beetle communities (Carpaneto et al, 2007; Hutton & Giller, 2003, Buse et al, 2015). Loss of permanent pastures for improved grasslands and a change in agricultural practices have led to a reduction in continuous livestock grazing (Robinson & Sutherland, 2002; Buckingham et al, 2006). Dung beetle communities depend on a continuous supply of good quality dung throughout the year and modern-day farming methods have affected this (Barbero et al, 1998).

A review of Great Britain's Scarabaeoidea by Natural England in 2016 identified 25% of dung beetle species as nationally rare, four as being extinct and more than sixteen that are either endangered, vulnerable or near threatened due to a loss of permanent pastures and cessation of grazing (Natural England, 2016). It is likely that changes in grazing regimes and in livestock husbandry is negatively impacting dung beetle communities (Beyan et al, 2012; Natural England, 2016).

Another key factor affecting dung beetles is the use of endectocides, such as Ivermectin on livestock which can influence dung beetle species richness and diversity (Beynon et al 2012; Lumaret *et al.*, 2012; Pérez-Cogollo et al, 2017, Jochmann and Blanckenhorn, 2016). Higher species richness, diversity and functional diversity of dung beetles have been found on

farms with no history of parasitic veterinary treatments (Sand & Wall, 2018). Hutton and Giller (2003) found that intensive farms (those with higher inputs of fertilizer, pesticides, labour and capital) support 38% less dung beetle species than organic farms and concluded a likely contributing factor to be the use of veterinary drugs.

Rewilding

Rewilding is a concept based on the reintroduction of species as key drivers of ecological restoration (Nogués-Bravo et al, 2016). There are a number of perspectives, approaches and definitions which exist in relation to rewilding. This paper broadly defines rewilding as: "The passive management of ecological succession with the goal of restoring natural ecosystem processes and reducing human control of landscapes" (Navarro & Pereira, 2012, p. 904). Rewilding is growing in prominence as a solution to agricultural land abandonment and conservation management throughout Europe (Navarro & Pereira, 2012). There is much debate and controversy surrounding the complexities and benefits of rewilding as an approach, and apprehension remains about its suitability as a model within a modern-day setting. Scientific support for the main ecological assumptions behind rewilding is limited and many believe it is difficult to predict the consequences of the introduction of novel species (Bauer et al 2009; Jørgensen, 2015, Nogués-Bravo et al 2016). That said, the recent expansion of rewilding projects and initiatives within Europe demonstrates its growth and eminence within landscape based conservation (PAN Parks Network, 2002; Rewilding Europe, 2015). There is increasing support for rewilding programmes and their integration in agricultural management schemes (Navarro & Pereira, 2012; Merckx & Pereira, 2015). The recent government 25 year environmental plan makes reference to the Knepp Wildland Project as an interesting case study for landscape scale restoration (HM Government, 2018). It is a small-scale rewilding project based on a less controversial form of rewilding. The Knepp Wildland Project, situated south of Horsham in West Sussex, is the biggest lowland rewilding project in the UK.

The Knepp Estate used to be arable intensively farmed land, but since 2001 its 3,500 acres have been transformed into a *passive rewilding* case study focused on land restoration by natural processes. Principled around a 'process-led' approach, the project relies on a mix of free roaming herbivores that stimulate vegetation and create a montage of habitats. Strongly influenced by Dutch ecologist Frans Vera and his theory of grazing ecology, similar to the Oostvaardersplassen Reserve in the Netherlands, Knepp is a case study for a naturalist grazing model as a stimulant for ecological restoration. Although still in its elementary stages, in less than a twenty year period Knepp has witnessed considerable ecological improvement and is home to a blossoming array of fauna and flora. A 2015 survey revealed the Estate to be home to 567 species of invertebrates (Lyon, 2015) and it is now an established haven for rare species such as turtle doves, nightingales, and purple emperor butterflies. As a result, it has attracted much interest from a number of experts and conservation organisations and is being promoted as a low-cost method of ecological restoration that can replace unsuitable or abandoned farmland in lowland England (Tree, 2017).

Rewilding Versus Organic

Hutton & Giller (2003) found that organic farms had significantly greater dung beetle biodiversity than intensive and rough grazed farms in Ireland. This was largely attributable to the reduced use of chemical fertilisers and veterinary drugs, but also to the possible influence of patchier ecosystem structures and a greater diversity of ungulate species found on organic farms. Therefore, rewilding, as a naturalist grazing model, could be even more beneficial for dung beetle biodiversity. Unlike organic farming, the rewilding model provides dung beetles with a continuous supply of dung throughout the year, as a result of the continual grazing of its free roaming ungulates species. This is essential as it provides dung beetles with the resource they require for colonization and can promote greater numbers of more specialised species (Buse et al, 2015).

A key factor affecting dung beetle diversity is habitat heterogeneity as some specialist species are forest dwellers and prefer shaded areas. Research on the Oostvaarderplassen rewilding reserve suggests that natural grazing refuges benefit invertebrate diversity due to the existence of and differences in vegetative edge effects (Klink, 2016). This supports Barbero et al's (1999) conclusion that patchy ecosystems characterized by open and wooded habitats with a mixture of ungulates are likely to support the highest level of dung beetle diversity. Although many dung beetle species are generalist feeders it is clear that some have particular dung preferences and a mix of wild ungulate species is therefore likely to attract a more diverse array of dung beetle species (Hanski & Camberfort, 1991; Finn & Giller 2002; Whipple & Wyatt Hoback, 2012). Buse et al (2015) highlights that grazing continuity and large pastured areas are important factors for dung beetle diversity and so a rewilding approach to land management could benefit dung beetle communities. The study stresses that these factors are particularly important for species with a low population density which are more vulnerable to local extinction within smaller pastures. It is clear that the conditions created within certain rewilding landscapes have the potential to suit the requirements of dung beetles and could enable communities of species to flourish.

Despite emerging evidence that rewilding could be advantageous for dung beetle conservation, little formal research or monitoring has been undertaken to develop this idea, or comparisons made with an organic farming system. Comparing dung beetle biodiversity in this way is important not only to establish the efficacy of rewilding grazing systems but also to provide empirical evidence for the comparison of these two grazing models. The Knepp Wildland project is a suitable case study by which to investigate biodiversity. Knepp is not only the largest rewilding project in the UK but by virtue of restoration of different areas of land at different times also allows for the comparison of heterogeneous and ecologically diverse sites.

This study addresses the following research question: Is small scale rewilding beneficial for dung beetle biodiversity? Therefore, the aim of this project is to undertake a comparative survey of dung beetle biodiversity between two different grazing models: rewilding (represented by The Knepp Castle Estate case study) and organic farming systems (represented by two organic farms). The key objective of the study is to measure species richness, evenness and abundance between these sites in order to ascertain whether rewilding sites have greater dung beetle biodiversity than the organic farms. *Species richness* can be defined as the total number of different species represented in an ecological community and *species evenness* as the measure of homogeneity of the abundance in a sample of that community (Colwell, 2009).

Study Area

The Knepp Castle Estate is composed of 3,500 acres of heavy weald clay land situated within the Low Weald in West Sussex. The land is sectioned into three blocks; North Block, Middle Block and South Block divided by the A272 road and Shipley/Dial post lane.

Restoration of each section has taken place at different times and been managed in different ways, leading to the development of a varied rewilding landscape. The North and South blocks were chosen as the suitable survey sites because they represented the greatest divergence between sites within the Knepp estate. (See figure 1 & 2)

The North Block

The extension of 236 hectares of land north of the A272 led to the creation of the Northern Block in 2006, the most wooded area of the estate. Once a mixed farm focused on the production of dairy, it has since been reseeded with a grass mix of seven different species. The landscape is grazed grassland combined with open woodland pastures and has 108 Longhorn cows that freely graze within its landscape. (See figure 3)

The South Block

The inclusion of the southern block (473.17 hectares in size) has taken place in stages, with fields taken out of production at different times. Natural regeneration over a six year period before free roaming herbivores were introduced has led to a higher density of scrubland than in the North, with huge areas of wetlands dominated by sallow. The South is now home to 165 Fallow Deer, 94 Long-horn cattle, 10 Exmoor Ponies, 7 Tamworth Pigs and a small red deer population. (See Figure 3)

[Figure 1 inserted here]

Organic Farms

Two organic farms, Rudgwick Organic Farm and Lee House Farm, were selected as suitable comparison sites as they met the following criteria: they were within 25 kilometres of Knepp Castle Estate; they have been categorised as the same soil type as Knepp (Type 18 SoilScapes, 30th March 2018) and they have government approved organic certification (*Organic Farmers & Growers* GB-ORG-02 & *Organic Food Federation* - GB-ORG-04). Dung beetle diversity is greatly influenced by soil type and it was therefore important to choose organic farms with the same soil type as Knepp in order to accurately compare diversity. (See figure 2 for location of sites)

[Figure 2 inserted here]

Rudgwick Farm is situated 15 kilometres North West of the Knepp Castle estate and has been a registered Organic beef farm since 1994. A small farm with 44 hectares of land in 9 fields, it is dedicated to the rearing and production of beef with a total of 88 cattle including young livestock. (See figure 3)

Lee House Farm is situated 18 kilometres north west of Knepp Castle estate and 6 kilometres north west of Rudgwick Farm. Lee House Farm has approximately 100 hectares of land totalling 20 fields and is a mixed organic livestock farm. In both farms, the pasteurised fields are surrounded by woodland edges and small hedgerows. Certain fields are rotationally cattle grazed during summer months, June to September and cattle are kept indoors over the winter period. (See figure 3)

[Figure 3 inserted here]

Methodology

Experimental Design

Pitfall trapping was the chosen sampling technique for the dung beetle survey. Although pitfall trapping is not without its biases and can underestimate species richness (Price & Feer, 2012), it is the most widely used and robust technique for a rapid biodiversity assessment of dung beetles (Denver Museum of Nature & Science, 2007). It is an effective method for the collection of a large number of data specimens necessary for comparing biodiversity between sites (Sutherland, 2006). The pitfall trap design constructed was based on the national recording scheme for Scarabaoidea (Mann, D.J 2018, National Recording Scheme for Scarabaoidea) (See appendix 1 for a full description of the pitfall trap design).

A total of 160 pitfall traps were laid out across the four sites. A sample size of 40 traps per site was chosen in order to allow for a statistically meaningful comparison between sites, with a degree of statistical power sufficient to detect significant differences and reduce the likelihood of a Type II error (Fields, 2014). Dead pitfall trapping was undertaken due to the large survey sampling size and problems with identification of live specimens (Larsen &

Forsyth, 2005; Sutherland, 2006). Ethical approval was given and a health and safety risk assessment done before the field survey was undertaken. (See appendix 2)

Four randomly chosen fields were selected at each site in order to collect a representative set of data in a total of 16 fields (See figure 3). Due to the size of the Knepp landscapes, fields for these sites were randomly selected within a 3 kilometre radius of the main parking zones. At Lee House Farm, fields that were not used for cattle grazing were also removed from the selection criteria. A map was used to number fields at each site and entered into a random computerised generator in order to minimise human bias errors.

Pitfall traps were placed along parallel transect lines using a tape measure, starting 50 meters in from the corner edge of each field with a 100 metres between transect lines.

Transects lines were chosen as the most time efficient method, although grid transects may have provided a more complete coverage of the area and addressed detection biases more sufficiently. A total of five pitfall traps were located along each transect line at 10 metre intervals, enough distance to eliminate potential interference between traps that could have affected results. Insufficient trap spacing can affect distribution of species abundance across traps and a minimum of 3 meters between traps is recommended (Denver Museum of Nature & Science, 2007). Larsen & Forsyth (2005) findings suggest 50 metre spacing is the ideal distance to minimize trap interference in dung beetle studies but this was not possible due to limitations in the area size of fields. Distance measures for the study were instead based on previous field work research and also on estimated field sizes to ensure that replication was possible across all fields (Sand & Wall, 2018; Larsen & Forsyth, 2005; Hutton & Giller, 2003).

Survey implementation and identification

All pitfall traps were baited simultaneously on the same day, 31st July 2018, in order to avoid inconsistency of local weather patterns which could have affected results. Baiting and collection was effectively carried out with the help of a group of volunteers that had been

given previous training. Traps were baited with fresh cow dung collected from the same site and frozen for at least 24 hours before the survey. This was necessary to reduce the risk of bio-hazards and to ensure all specimens collected were from the pitfall traps themselves and not from the existing bait. Pitfall traps were left for three days before collection took place, on 4th August 2018, in order to allow enough time for the effective colonisation and capture of the dung beetle community at each site.

All contents from each pitfall trap were collected, but not specimens still residing in the baited dung, so at to reduce possible human error bias. Specimens from each pitfall trap were placed into sample pots, coded with a unique number and filled with ethanol to preserve contents for later identification. At each field an information form was completed at the time of collection in order to obtain important information that could have influenced results, such as damage to pitfall traps and observed fresh dung near pitfall traps.

Identification took place at the Oxford Natural History Museum where specimens were sorted by hand into the dung beetle genus groups: *Onthophagus, Aphodiinae* or *Geotrupidae* and other dung fauna family groups: *Staphylinidae, Hydrophilidae, Histeridae*. All specimens were placed under a binocular microscope with x10 magnification and dung beetles were identified to a species level using Jessops (1986) and Skidmore (1991) key guides. Specimens were checked and validated by Darren Mann (Oxford University Natural History Museum).

Statistical analysis

Differences in mean observed species richness per sample were tested using a General Linear Mixed Model (GLMM) in the software programme SPSS (IBM Corporation, 2017). Within the mixed effect model, nested variables were used to test for significant effects with 'field' randomised as a nested variable within site and a Type III test of fixed effects applied.

A pairwise comparison test using Bonferroni's corrected p-values was then used to compare significant differences in and between sites (Bland & Altman, 1995).

The programme EstimateS (Colwell, 2016 version 9.1.0) was used to formulate rarefaction curves using individual-based abundance data (set at 100 randomization runs with 95% confidence intervals) to further analyse patterns of species richness (Colwell & Coddington, 1994; Gotelli and Colwell, 2001). This additional analysis was undertaken because it addresses possible result biases that can occur due to differences in sampling effort. Abundance variations differ depending on sample size and this can cause problems when accurately measuring species richness. Species-abundance distributions curves were plotted individually and then compared in order to estimate the extent of sampling and significant differences between sites.

EstimateS (Colwell, 2016 version 9.1.0) was also used to calculate and plot two species diversity indices: the Shannon exponential mean (EH⁰), and Simpson (inverse) index (1/D) (Jost, 2006; Magurran 2004). The diversity indices were used in order to apply an alternative measure of biodiversity, one that considers relative abundance of species and evenness. The exponential and inverse formulae were chosen as they transform indices into effective numbers of species (the true diversity of the community in question) for a more accurate interpretation.

Results

A total of 12,178 adult dung beetles belonging to 13 different species were collected. The total number of species found at each site and the total number of individuals for each species are shown in Table 1. When considering species richness, *Aphodius* was the dominant genus with 10 species (77%) followed by *Geotrupes* (2 species, 15%) and *Onthophagus* (1 species, 8%). However, when species abundance was considered,

Scarabaeinae was the overwhelmingly dominant taxon, 93% of all individuals, due to the high volume of *Onthophagus similis* identified. The most abundant species, *O. similis*, comprised a total of 11,399 individuals, followed by *Acrossus rufipes* (399 individuals, 3.2%) and *Coloboterus erraticus* (209 individuals, 1.7%).

90% of individuals collected across all sites came from South Knepp, 96% (10,617) of which were identified as *O. similis*. At South Knepp a total of 11 different species were identified. At North Knepp, 624 individuals were collected, comprising 10 different species. At Rudgwick Farm, there were 233 individuals comprising 6 different species and at Lee House Farm there were 279 individuals comprising 8 different species.

The total number of dung beetles collected and identified at the rewilding sites was 11,066 compared to 512 individuals at the organic sites. This demonstrates a large difference in abundance between the two grazing models. (See table 1 for raw data summary)

[Table 1 to be inserted here]

General Linear Mixed Model

The results from the Type III test of fixed effects analysis showed that site (*independent variable*) had a significant effect on species richness (F3,12 = 8.535, P < 0.01). A further comparative analysis between sites using Bonferroni's corrected p-values showed that South Knepp had significantly higher observed species richness per pitfall trap than either organic farms but not in comparison to North Knepp. There were no significant differences in observed species richness between the other three sites: North Knepp, Rudgwick Farm and Lee house farm. (See figure 4)

[Figure 4 inserted here]

Rarefaction analysis

Species accumulation curves demonstrate stabilisation patterns for each of the sites, indicating a relatively complete sampling of the community. Inspection of the width of the 95% confidence intervals suggests that at North Knepp and Rudgwick Farm all species were estimated as likely to have been sampled. However, at South Knepp and Lee House Farm it is more likely that there was greater variation in the number of species sampled. (See figure 5 a.b.c.d). A comparison of species accumulation curves between sites was difficult to measure due to the large abundance of *O.similis* at South Knepp (see figure 6a). A revised analysis with *O. similis* excluded from the data set was therefore completed in order to analyse results without such a skew in the data set. The results of this suggest that there is significant difference between North Knepp and Rudgwick Organic farm. This may be seen from the non-overlapping confidence intervals between the two sites (see figure 6b). A relatively low species count across each of the sites may begin to explain the limited differentiation between curves.

[Figure 5. a.b.c.d & 6.a.b. inserted here]

Species diversity indices

The inverse Simpson index (1/D) and exponential Shannon index (EH⁰) graphs showed similar results to the rarefaction analysis and suggested Rudgwick Farm (1/D = 2.15 & EH⁰ = 2.68) and North Knepp (1/D = 1.56 & EH⁰ = 2.32) to be the most bio-diverse sites and South Knepp (1/D = 1.8 & EH⁰ = 1.23) to be one of the least diverse sites (see figure 7a and 8b). This is perhaps not surprising given the index calculations focus on relative abundance and the large population of *O. similis* found at South Knepp, which comprised approximately 96% of all individuals. Species diversity analysis was also conducted excluding the species *O. similis* to ascertain biodiversity without the influential dominance of this species. This was done in order to acquire a more holistic analysis without such a possible skew in the data set. This resulted in North Knepp becoming the most bio-diverse followed by South Knepp,

and Rudgwick Farm the least (See figure 7b and 8b). When *O.Similis* is removed both index graphs show the biggest difference between North Knepp (1/*D*=3.72, i = 90 & °*H*0 =5 i= 131 &) and Rudgwick Farm (1/D=1.68, i = 90 & °*H*0 =2.29 i = 131 &) supporting findings from the rarefaction analysis. A comparison of the graphs with and without *O.similis* show a mix of results but suggest North Knepp to be the most consistently diverse site when considering all diversity index analyses.

[Figure 7a.b & 8a.b inserted here]

Discussion

Rewilding versus an organic farming grazing model

The results suggest rewilding sites have significantly higher dung beetle abundance and species richness than the organic farms. This is particularly apparent when looking at individual numbers for both Knepp sites in comparison to the two organic farms (see table 1). South and North Knepp sites combined have more than twenty times the number of individual dung beetle specimens than the organic sites. Although this is largely attributable to the abundance of one species, *O. similis* (approx. 40 times difference between the models) there were also noticeable abundance differences for the other three most common species: *A. rufipe, C. erraticus,* and *V. stic*ticus. The species *C. erraticus,* in particular had approximately 28 times more individuals at the Knepp sites compared to the organic farms (see table 1). Overall, total abundance was greatest at South Knepp by a considerable margin when comparing all sites.

Statistical analysis using the GLMM demonstrated that South Knepp had significantly higher observed species richness than the two organic farms, with a greater number of species on

average per pitfall trap (see figure 1). As species richness is one of the most valid measures of biodiversity (Colwell, 2009), these results strongly indicate that the rewilding landscape at South Knepp supports greater dung beetle biodiversity than at the other sites. There are a number of different factors that can account for this, which will now be discussed in detail.

Resource and habitat specialisation

South Knepp can be defined as the most rewilded part of the estate with a mixture of ungulate species that are not present at the other sites. It is this mix of ungulate species that could explain the higher diversity and abundance of dung beetle species at South Knepp. Evidence suggests dung beetles have preferences to certain dung types (Estrada et al, 1993; Finn & Giller, 2002; Whippple & Hoback, 2012). Moreover, generalist species that are less particular in their resource selection can utilise a more varied range of dung types, leading to reduced resource competition (Hanski, 1991, Davis and Sutton, 1997). A recent review by Buse et al (2018) provides evidence of different dung specialisation among over 100 dung beetle species found in central Europe. Data from this study shows that two dung beetle species A. fimerarius and E. Pusillus, found at the rewilding sites but not organic sites, have a varied range of dung type preference and utilisation history. E. Pusillus, found only at South Knepp, is a known pasture specialist that shows preference to all dung types and is known to occupy wild boar dung (Buse et al, 2018). The residence of Tamworth pigs at the South Knepp could therefore be a contributing factor to its presence. Similarly, A. fimerarius, known to prefer large herbivore dung, was found at South and North Knepp but not at the organic farms. Evidence suggests this species has a preference for Fallow & Roe deer dung (Buse et al, 2018) and both these deer species reside at South Knepp. Roe deer does naturally occur at North Knepp and it could be that the presence of deer species at South Knepp and Middle Knepp, situated in close proximity to North Knepp, is a reason for A. fimerarius at both sites. There is however limited data relating to dispersals rates of dung beetles and therefore caution must be exercised in taking this as an assumption. The

presence of these species only at the rewilding sites supports the proposition that South Knepp may attract a large number of generalist species because of a more varied array of dung resource availability.

Another explanation for greater biodiversity at South Knepp could lie in differences in scrub and vegetation densities that attract a number of habitat specialist species. Natural regeneration at South Knepp over a six year period before grazers were introduced has led to large scrub and wetland areas (Tree, 2017; Tree 2018). Evidence suggests that differences in vegetative edge effects can often influence species diversity (Robert et al 2008; Klink et al 2006; Benton et al 2003). This is particularly the case for dung beetles as it is known that many species have specialist habitat requirements; some prefer shaded areas often choosing to inhabit forest floors (Audino et al 2014; Roslin, & Koivunen, 2001; Hanksi, 1991. The presence of *B.ictericus* at South Knepp but not at the other sites supports this theory as it is a species known to prefer well drained soils (DUMP, 2016). Its presence also indicates an improvement of soil conditions at South Knepp which could be a result of the natural regeneration of water courses (Tree, 2018). The shaded specialist species *P.borealis* was also found at both North and South Knepp but not at the other farms. This further supports the notion that a rewilding grazing system may support a larger variety of specialist species.

Patch size, grazing continuity and dung quality

Other analysis focused on alternative measures of biodiversity showed mixed results. Although species richness is a strong indicator of biodiversity, it is the evenness of species distribution that is also seen as a key measure of diversity (Colwell, 2009). When species evenness was considered using diversity indices, Rudgwick Farm and North Knepp were revealed as the most bio-diverse sites, although this changed with the removal of *O. similis*, indicating its strong impact on the biodiversity metrics. Conceptual and statistical problems

associated with the use of diversity indices, such as sensitivity to sample size and lack a probabilistic basis (Sandoval& Barrantes, 2009) means caution should be exercised when considering these results in isolation. Overall however, analysis from the diversity indices suggests North Knepp to be the most diverse when species evenness is considered. This echoes findings from the rarefaction analysis which suggests a significant difference between North Knepp and Rudgwick Farm.

Patch size could be one explanation for the more even spread of species at North Knepp compared to other sites. North Knepp has larger open and semi open pastures which could affect population densities, especially in relation to vulnerable species (Buse et al, 2015; Roslin, 2000; Burke & Goulet, 1998). Different pasture size areas can affect resource availability influencing competition between populations within a community (Hanksi, 1991). It has been suggested that patch size areas larger than 130 ha (hectare) the most effective for species richness and help aid vulnerable populations (Buse et al, 2015). South and North Knepp sites both meet this size criterion but the organic farms do not, illustrating the possible importance of patch size area on diversity.

Grazing continuity and history are also key factors that could explain greater dung beetle biodiversity at both rewilding sites. Species need available resource all year round to feed and breed and both sites provide this with their continuous free roaming grazing regimes. Land with a longer history of grazing has shown to be higher in dung beetle diversity (Buse et al. 2015). A longer grazing history allows for a longer period of colonisation of dung beetle communities. North Knepp has a longer grazing history than South Knepp, which may be another factor explaining a more evenly spread variance of dung beetle species at Knepp North. The greater number of generalist and specialist species identified at the rewilding sites support Buse et al (2015) findings that grazing continuity could play an important role in dung beetle diversity.

Another factor affecting dung beetle diversity is quality of dung, as moisture content and consistency influence resource requirements and attractiveness for dung beetles (Hanski, 1991). Dung produced by native breeds is better than intensely managed dairy and beef as it produces less watery content (Greenham, 1972). Knepp has introduced long-horn cattle, a native breed originating in the northern counties of England, and this may also have contributing positive effects on dung beetle diversity. It should be noted that at the organic farms a runnier dung consistency was observed than at South and North Knepp. The use of ivermectins can also impact on dung consistency effecting dung beetle breeding patterns, (Roncalli, 1989; Wall & Strong, 1987) and the absence of the use of invermectins at Knepp may be a further contributing factor. However, it should be noted that the use of invermectins on organic farms is usually restricted, explaining higher dung beetle biodiversity on organic farms than on intensive farms (Barbero et al, 1998; Sand & Wall, 2018).

Overall findings suggest that both rewilding sites, North and South Knepp, have varying degrees of biodiversity within them, a likely result of different restoration management. Despite variations, overall they both show similar biodiversity patterns and as a rewilding model demonstrate a stronger representation of biodiversity than the organic farms. As explored above, there are a number of factors that account for this and explain why rewilding sites provide suitable conditions for dung beetle biodiversity. However, limited research on this subject matter means the extent of these influencing factors are hard to quantify.

Onthophagus similis

An interesting finding from the study was the acute colonisation of one species, *O. similis*, which accounted for approximately 86% of all individuals and was particularly dominant at South Knepp. As this study is not longitudinal and there is no previous data for annual comparisons, it is not known if this mirrors a seasonal trend. Nor can the reason for the large

presence of *O.simils* be established with any degree of certainty. That said, weather conditions may explain such a large abundance of this species captured across sites. The genus *Onthophagus* is known to favour summer climates and warm conditions, a reason why the genus dominates in Mediterranean regions (Robinson, 2013). The study was undertaken during a particularly hot summer with temperatures reaching 31 degrees over the survey period. Resource is scarcer during droughts as dung dries up faster, affecting resource availability (Halffter & Edmonds, 1983). The possible effects of this on the dung beetle communities surveyed are unknown. Nor is it known if the colonisation of *O.similis* influenced the presence or absence of other species. Warm weather conditions do however provide a valid explanation for the high volume numbers of *O.similis* across all four sites.

The results also pose interesting questions about the possible future effects of climate change on UK dung beetle assemblages; will there be a growth in *Onthophagus* species as UK summer temperatures increase? Studies have shown the importance of dung beetles as key indicator species and how they can be used to monitor climate change (Penttilä et al, 2013; Slade et al, 2016). A study by Robinson (2013) investigated the relative abundance of *Onthophagus* species in British assemblages of dung beetles as evidence of Holocene climate change. More research in this area and long-term data monitoring may help to answer some of these questions.

Conclusion

The results of this study provide strong evidence of the benefits of rewilding for dung beetle biodiversity. Previous studies have asserted that rewilding could help increase dung beetle diversity (Buse et al 2015; Hutton & Giller, 2003; Barbero et al, 1998) but few studies have been undertaken to further investigate this. Overall results from this study indicate that dung beetle biodiversity is significantly higher at the rewilding sites than at the organic sites, and thus provide empirical support for this proposition. That said, it should be stressed that this study only sampled one rewilding site and two organic farms. Studies using a larger number

of representative sites and long term research are needed to provide further evidence of the benefits of rewilding on dung beetle biodiversity. Rewilding is growing in popularity as a conservation management solution for increasing biodiversity (Navarro & Pereira, 2012). Case studies such as Knepp are examples of the potential ecological benefits of rewilding within a short time period. This study provides further evidence of this biodiversity with the identification of 12 different dung beetle species collected from one survey across both Knepp sites.

Given the ecological services that dung beetles provide, particularly within a UK agricultural setting, it is surprising that a greater attention has not been given to dung beetles by conservation organisations and government bodies. The case for the integration of rewilding land management within agri-environmental policy and provision of subsidies has already been proposed by Merckx & Pereira (2015). They strongly promote the suggestion that less-productive agricultural land should be ecologically restored through rewilding and the management of natural succession. This study promotes the idea that rewilding is beneficial for dung beetle biodiversity, and as such, could provide enhanced ecosystem services in these areas. Further research into this would be valuable and may provide insight and possible support for the integration of rewilding into a UK agricultural policy.

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I would like to thank all the landowners who allowed me to perform pitfall trapping on their pastures (Knepp Castle Estate, Rudgwick Farm, Lee House Farm), all the volunteers who helped with the pitfall trap baiting and collecting, Darren Mann for his invaluable support and taxonomic assistance and Sam Cotton and Mark Steer for their valuable contributions.

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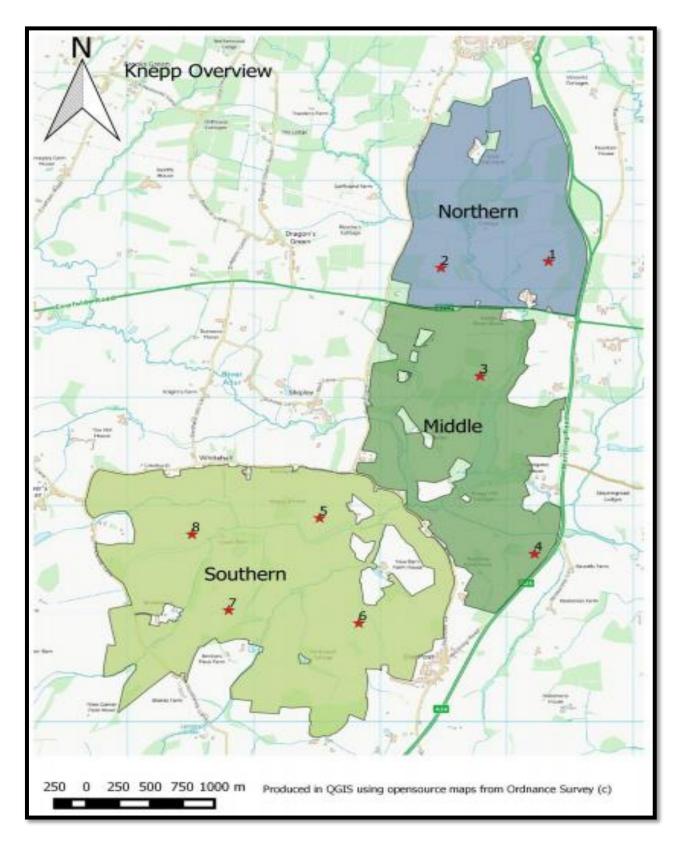


Figure 1: Map of Knepp Castle Estate by Graeme Lyons 2015 permissions for use granted.



Figure 2: Map showing all four site locations, South Knepp, North Knepp, Rudgwick Farm & Lee House Farm in West Sussex - created in ArcGIS.



Figure 3: Map of each site location with each of the four survey fields outlined in red - created in ArcGIS.

Table 1: Breakdown of species and number of individuals across all four sites

Species	South Knepp	North Knepp	Rudgwick Farm	Lee House Farm			
		Number of individuals					
Onthophagus Similis	10617	503	143	136			
Acrossus rufipes	179	59	68	93			
Coloboterus erraticus	181	21	5	2			
Violinus sticticus	44	14	0	27			
Bodilopsis rufa	6	2	12	8			
Aphodius fimerarius	2	2	0	0			
Esymus Pusillus	2	0	0	0			
Teuchestes fossor	3	19	1	1			
Bodiloides ictericus	1	0	0	0			
Otophorus haemorrhoida	alis 6	2	4	3			
Planolinus borealis	1	1	0	0			
Geotrupes stercorarius	0	1	0	0			
Geotrupes spiniger	0	0	0	1			
Total no: of species	11	10	6	8			
Total no: of individuals	11042	624	233	279			

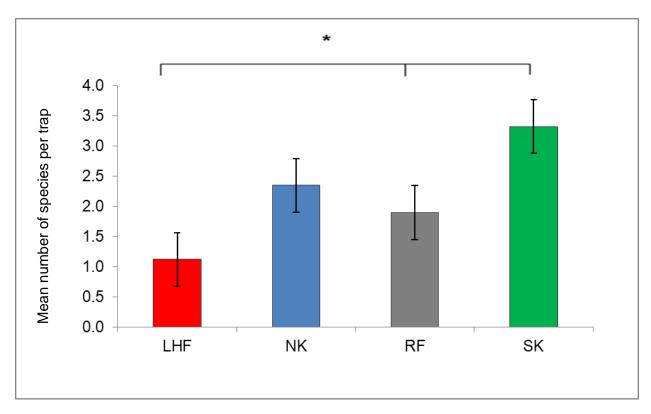


Figure 4. Mean (+SE) species richness (S) per pitfall trap (per site) with standard error. Asterick denotes significant difference at the .05 level of South Knepp (SK) following Bonferroni's comparison test.

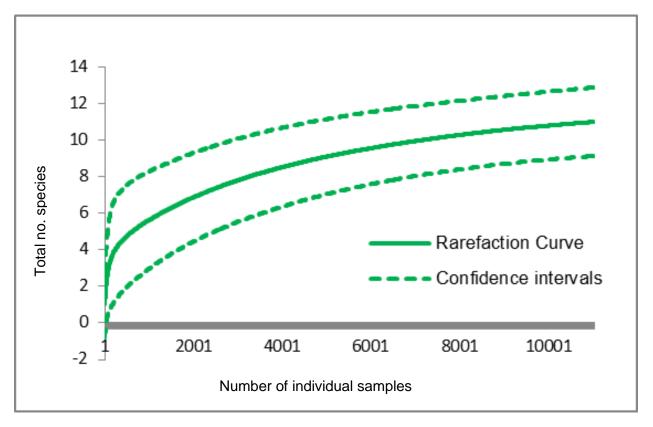


Figure 5a: individual-based rarefaction curve for South Knepp with 95% confidence intervals

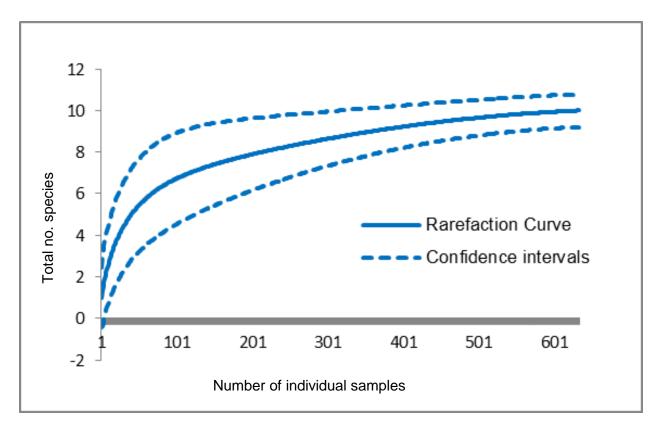


Figure 5b: individual-based rarefaction curve for North Knepp with 95% confidence intervals

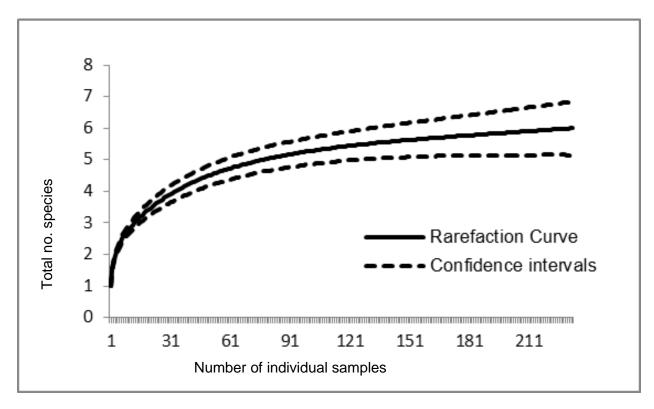


Figure 5c: individual-based rarefaction curve for Rudgwick Farm with 95% confidence intervals

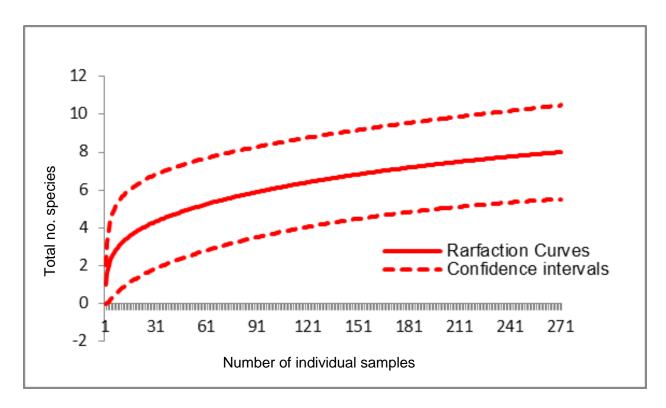


Figure 5d: individual-based rarefaction curve for Lee House Farm with 95% confidence intervals

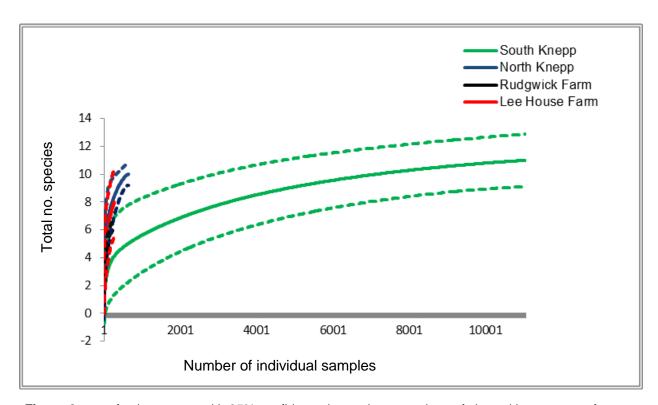


Figure 6a: rarefaction curves with 95% confidence intervals comparison of sites with presence of *O. similis*.

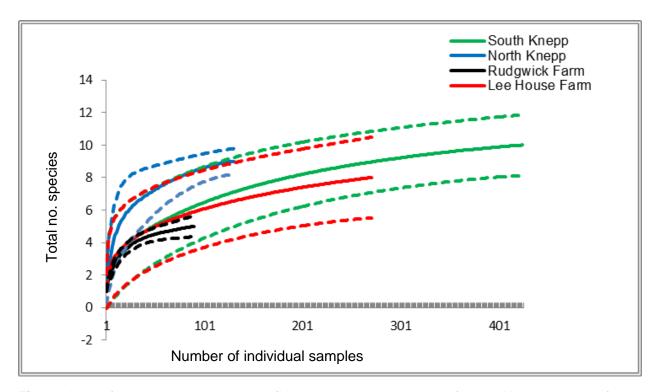


Figure 6b: rarefaction curves with 95% confidence intervals comparison of sites without presence of *O.similis*.

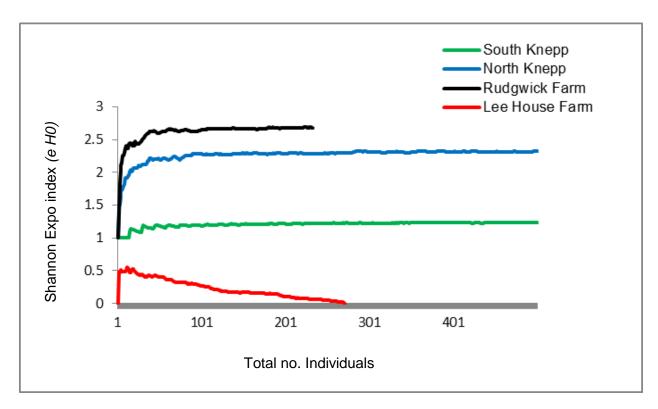


Figure 7a: mean value accumulation of Shannon exponential (eH0) comparison of sites with *O.similis*. (Higher values indicate greater bio-diversity)

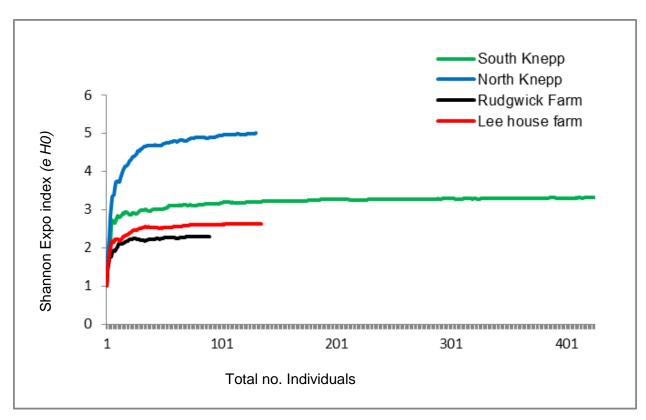


Figure 7b: mean value accumulation of Shannon exponential (e H0) comparison of sites without *O.similis*. (Higher values indicate greater biodiversity)

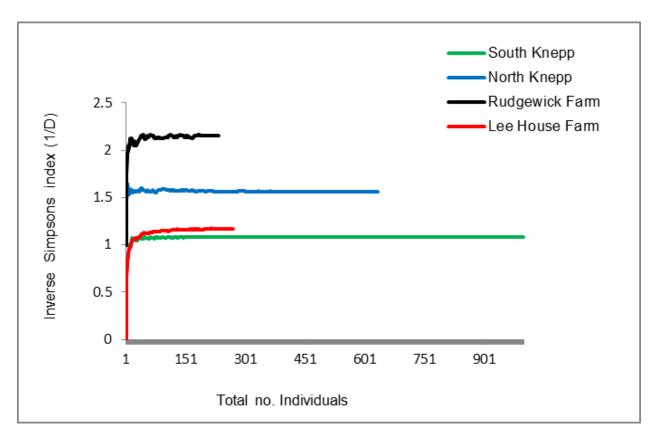


Figure 8a: mean value accumulation of Inverse Simpson (1/D) comparison of sites with *O.similis*. (Higher values indicate greater bio-diversity)

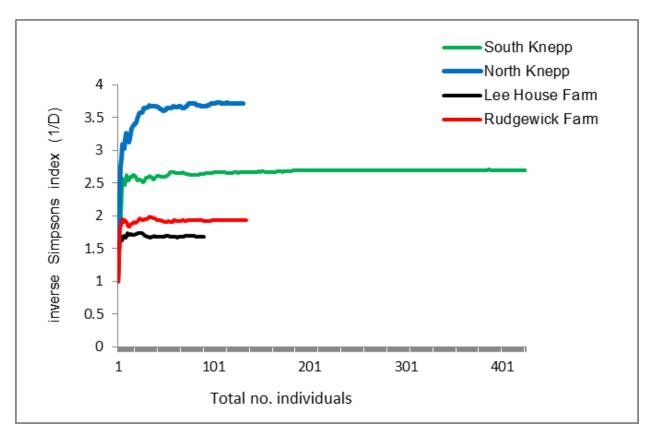


Figure 8b: mean value accumulation of Inverse Simpson (1/D) comparison of sites without O.similis. (Higher values indicate greater bio-diversity)

Appendices

Appendix 1: Pitfall Trap Description with photos of pitfall traps at Lee House Farm and South Knepp.

A hole for each pitfall trap was dug using a pick axe and spade. A 1 litre plastic bucket was then inserted inside the hole with the lip of the bucket placed level with the ground surface. A piece of square mesh 18 cm X 18cm in size was pegged down over the bucket using two metal tent pegs. A protection cover was made and placed over each pitfall trap using two paper plates and four wooden sticks. A unique number for each pitfall trap was written on flag tape and tied to a bamboo stick next to each trap for identification purposes.





Appendix 2: Copy of Risk Assessment Form

Hazards Identified	Existing Control Measures	S	L	Risk	Additional Control Measures	S	L	Risk	By whom and	Date
(state the potential harm)				Level				Level	by when	completed
Injury from thorny	Wear suitable footwear and proceed with caution	2	5	10	Buddy system will be in place, volunteers	1	5	5	Student and	Day of
vegetation	especially when ground wet.				working in pairs. There will also be a first aid				volunteers	work
					kit onsite.					
Tripping or falling on uneven	Wear suitable footwear and proceed with caution	2	5	10	Buddy system in place and first aid kit	1	5	5	Student and	Day of
ground or into water	especially when ground wet.								volunteers	work
Insect stings or bites	Use appropriate insect repellents	2	2	4	First aid kit	1	2	2	Student and	Day of
_									volunteers	work
Skin irritation from plants.	Avoid coming into contact with sap from species	2	2	4	First aid kit	1	2	2	Student and	Day of
	such as hogweed.								volunteers	work
	_									
Sunburn or heatstroke	Wearing suitable clothing and using sunscreen	2	2	4	First Aid Kit and water taken onsite	1	2	2	Student and	

Page 1 of 3

	creams					Т			volunteers	
Lightning strike, flooding	Listen to weather forecast & avoiding days when storms or heavy rain predicted.	2	1	2		2	1	2	Student and volunteers	Day of work
Attack by livestock	Avoid approaching animals especially with young.	4	2	8	Buddy system in place and First aid kit	4	1	4	Student and volunteers	Day of work
Propylene Glycol (non-toxic preservative)	Wear gloves at all time when handling solution	2	2	4	First aid kit	2	2	4	Student and volunteers	Day of work
Lyme disease	Wear suitable clothing; check for ticks & remove any asap; visit doctor's if site of any bite gives cause for concern.	3	2	6	Buddy system to check for ticks. First aid kit to include appropriate equipment for removing ticks.	3	1	3	Student and volunteers	Day of work
Disease from handling cattle dung.	Wear disposal gloves when handling dung at all times. Wash hands thoroughly with anti-septic after gloves are thrown away.	3	1	3	Full briefing given to volunteers and safety procedures including appropriate storage and transportation processes.	3	1	3	Student and volunteers	Day of work
Lone person working	Ensure Estate Office or landowner knows who onsite is; take fully charged mobile phone; ensure spouse/partner/other knows location of site & expected time of return.	2	5	10	Buddy system and first aid kit	1	5	5	Student	Day of work
Travel to site: risk of delays/harm to researcher(accident)/harm to unknown third parties(accident)	Researcher holds valid insurance and license to drive appropriate vehicle.	5	2	10	Plan of travel and contact details of volunteer and next of kin. Safety procedures followed.	5	1	5	Student and volunteers	Day of work

RISK MATRIX: (To generate the risk level).

Very likely 5	5	10	15	20	25	
Likely 4	4	8	12	16	20	

Page 2 of 3

Possible	3	6	9	12	15
3					
Unlikely	2	4	6	8	10
2					
Extremely unlikely	1	2	3	4	5
1					
Likelihood (L)	Minor injury – No first aid	Minor injury – Requires First	Injury - requires GP treatment	Major Injury	Fatality
 ↑	treatment required	Aid Treatment	or Hospital attendance		
Severity (S)	1	2	3	4	5

ACTION LEVEL: (To identify what action needs to be taken).

POINTS:	RISK LEVEL:	ACTION:			
1-2	NEGLIGIBLE	No further action is necessary.			
3-5	TOLERABLE	Where possible, reduce the risk further			
6 - 12	MODERATE	Additional control measures are required			
15 – 16	HIGH	Immediate action is necessary			
20 - 25	INTOLERABLE	Stop the activity/ do not start the activity			

S