



UNIVERSITY OF
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Evaluating the Predictors of Reptile Population Assemblages Across a Multifunctional Landscape

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project, Marwell Wildlife & University of Southampton**

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ABSTRACT

Global biodiversity is facing severe threats from anthropogenic landscape modification which results in lost and fragmented habitats. Agricultural intensification has been attributed as the reason for such extreme habitat loss, and is threatening the natural integrity and connectivity of landscapes as well as driving reptile declines in the UK. Reptiles are protected in the UK and are an important group for conservation due to their indication of habitat quality and ability to hunt and disperse along connected habitats, which can provide indication of levels of landscape connectivity. This study examined the key environmental and structural predictors of reptile presence within a multifunctional landscape in Hampshire, UK, comparing reptile accounts across arable land, pasture and conservation grassland. Data were collected through reptile and habitat surveys, and analysed using Generalised Linear Models (GLM). The findings of this study are important for informing local and wider conservation efforts. The results illustrate the importance of connectivity on multiple scales, and attribute lack of connected habitat to low reptile counts. Temperature and its related effects were the greatest environmental predictors of reptile population assemblages and seemed to affect their habitat preference. The results indicate that greater plant diversity and structure did not necessarily yield higher reptile counts, meaning conservation efforts need to achieve a balanced habitat that provides adequate cover and thermal requirements for reptiles.

Key words: Reptiles, slow worm, grass snake, landscape connectivity, habitat conservation

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INTRODUCTION

Habitat Loss

As the UK faces significant changes in landscape heterogeneity, natural habitats are becoming increasingly threatened. Managing and conserving native wildlife is more important than ever due to the expansion of urban areas and subsequent degradation of natural landscapes. Agricultural land use has been attributed as one of the primary causes of habitat loss, and modern agriculture has triggered extensive environmental damage (Foley *et al.*, 2005). Recent agricultural intensification in Europe has been associated with a noticeable decline in farmland biodiversity and dramatic changes within agricultural landscapes (Le Féon *et al.*, 2010).

Global biodiversity is severely threatened by human modification of the landscape, particularly those which result in habitat fragmentation and loss (Fischer and Lindenmayer, 2007). Habitat loss is unanimously attributed as the primary cause of global biodiversity loss, and applies to reptile declines as well (Gardner *et al.*, 2007). Species extinction is being driven by habitat destruction, the leading cause of extinction (Pimm and Raven, 2000). Anthropogenic changes to habitats disrupt ecosystems and changes resource availability (Azor *et al.*, 2015). With habitat loss being the leading threat to biodiversity, it is crucial to understand the interactions of biota with their habitat to determine how to approach species-specific habitat conservation (Mayor *et al.*, 2009). To directly inform conservation, it must be understood how species respond to habitat attributes at the local level (Garden *et al.*, 2007) as well as landscape. It is also crucial to examine landscape structure when approaching conservation efforts due to the relationship between landscape structure and ecological processes (Aauri and de Lucio, 2001). Habitat destruction initially causes smaller-scale extinctions at the population level, but can lead to more drastic extinctions as the entire habitat is eventually wiped out (Pimm and Raven, 2000). This highlights the importance of understanding habitat-species interactions to inform conservation management before biodiversity loss reaches the scale of dramatic extinctions.

Landscape connectivity

Landscape connectivity, the degree to which the landscape facilitates or impedes movement among resource patches, is critically threatened by both habitat loss and fragmentation (Taylor *et al.*, 1993). Habitat fragmentation is described by Fahrig (2003) as a landscape-scale process involving habitat loss and habitats becoming divided or broken apart. Loss of landscape connectivity reduces the size and quality of available habitats, disrupting and preventing movement between habitats, and affecting migration patterns resulting in population declines, loss of genetic variation and decreased carrying capacity, and ultimately extinction (Rudnick *et al.*, 2012). Landscape connectivity operates on many scales: it can be structural (based on physical connectivity) as well as functional (the likelihood of species movement between habitats) (Dodd, 2016). An understanding of the implications of habitat change on biodiversity is imperative to create a framework for biodiversity within anthropogenically-driven landscapes (Gardner *et al.*, 2007).

Reptile conservation

Reptile species are facing global declines with six identified threats, acting independently and collectively, contributing significantly to this decline: habitat loss and degradation, invasive species, pollution, disease, unsustainable trade and exploitation and global climate change (Gibbon *et al.*, 2000). These threats have been categorised into direct factors including habitat change, invasive species and over-exploitation, whilst global climate change, pollution, and disease are classed as indirect factors (Collins and Storer, 2003). Gardner *et al.* (2007) simplify these categorisations to changes to structural (e.g. changes to habitat/landscape vegetation structure) and non-structural threats. Amphibians and reptiles are the most threatened of all terrestrial vertebrates according to the IUCN (Gardner *et al.*, 2007).

Reptile population declines are worrying on multiple scales. Their declines reflect a more general trend in decline of environmental quality, potentially predicting a 'crisis situation' on a more intense, global scale (Gibbon *et al.*, 2000). However, at a more localised scale, they also hold ecological relevance within many habitats (Todd *et al.*,

2010). Reptiles are an important component of biodiversity and have a supporting role in the biological diversity of vertebrates (Greenberg and Waldrop, 2008).

Reptiles are one of the least studied vertebrate groups, lacking in scientific studies compared with other groups (Gardner *et al.*, 2007; Todd *et al.*, 2010). Knowledge of threats to reptile species is insufficient, and it is estimated that the number of reptile species on red lists is highly underrepresented (Brooks *et al.*, 2002). Despite this stigma, they are important for a multitude of reasons and play significant roles in natural systems as predators, prey, seed dispersers and commensal species (Böhm *et al.*, 2013). Reptiles serve as bioindicators for environmental health, and their specific microhabitat associations provide ideal study systems to illustrate the biological and evolutionary processes underlying speciation (Böhm *et al.*, 2013). Reptiles are an important indicator of habitat health due to their sensitivity to landscape changes and habitat disturbance/fragmentation (Keinath *et al.*, 2017). Furthermore, in the instance of finding grass snakes, it can be generally assumed that amphibians are also present in the ecosystem as grass snakes prey nearly exclusively on anurans (Keinath *et al.*, 2017). These factors, as well as evidence of their global-scale decline, make reptiles of conservation interest and importance for study. There is a lack in scientific literature focusing on the threat of structural habitat change on reptiles (Gardner *et al.*, 2007), which is what this study aims to research in the context of a multifunctional landscape.

UK reptiles

There are seven species of native reptiles in the UK: adder (*Vipera berus*), barred grass snake (*Natrix helvetica*), common grass snake (*Natrix natrix*), smooth snake (*Coronella austriaca*), common lizard (*Zootoca vivipara*), slow worm (*Anguis fragilis*) and sand lizard (*Lacerta agilis*) (Figure 1). As *N. helvetica* was only very recently classified as a different species to *N. natrix* (Kindler *et al.*, 2017), grass snake species were not differentiated in this study. Due to their vulnerability, all UK-native reptiles are protected under Schedule 5 of the Wildlife and Countryside Act (1981) (Platenberg and Griffiths, 1999), under which *L. agilis* and *C. austriaca* receive full protection.



Figure 1: Adult grass snake (A) and adult male slow worm (B) (Images: authors own).

All UK species prefer well-structured habitat that provides them with warmth, shelter and foraging opportunities (Jofré *et al.*, 2016). As reptiles are ectotherms and vulnerable to predators, particularly avian, they depend greatly on habitat structure (mainly vegetation structure) at and just above ground level (JNCC, 2004). The decline of British reptiles is a result of anthropogenic activities (Dunford and Berry, 2012), with habitat change widely accepted as the primary cause (Jofré and Reading, 2012).

Importance of landscape and habitat structure for reptiles

Landscape structure is an essential aspect of conservation research due to the relationship of the structure of the land and ecological processes (Atauri and de Lucio, 2001). Atauri and de Lucio (2001) found that species richness responds in varying degrees depending on landscape heterogeneity. The most important factor in herpetofauna species richness was found to be abundance of specific land-use types. Focusing on landscape features is necessary to inform appropriate management, as land planning and management is a wide scale endeavour (Atauri and de Lucio, 2001). Landscape heterogeneity is changing significantly across Europe and in the UK due to agricultural practice, policy changes, urban development and so on. These changes influence biological diversity (Atauri and de Lucio, 2001),

highlighting a need for research into species distribution across sites of varying land use is particularly important.

UK reptiles favour a high degree of habitat heterogeneity, south-facing aspects, and the boundaries between two habitat types. These habitat characteristics are the most important determinants of population size and viability for UK reptiles (JNCC, 2004). The availability of breeding sites is also of importance for grass snakes (JNCC, 2004). Currently, the relationship between habitat structure and reptile population status has not been quantified (JNCC, 2004), however this research aims to examine the effect of habitat features on reptile population assemblages.

Important predictors of reptile populations

Generally, the greatest predictors of reptile richness are temperature-related (Qian, 2010; Dodd, 2016). Reptile habitat preference is influenced by their thermal requirements. Therefore, vegetation structure within the habitat is a key predictor of presence as it can alter the microhabitat conditions (Singh *et al.*, 2002). According to JNCC (2004) UK reptiles are dependent upon vegetation structure, topography and refuge availability. They also use the interfaces between certain habitats, as these link habitat types and can determine population viability and size of certain reptiles.

Guisan and Hofer (2003) showed that climatic variables were better at predicting reptile distribution than topographic variables. Distribution was predominately determined by factors related to temperature, although in an Alpine zone where temperature ranges are likely to be more dramatic (Guisan and Hofer, 2003). Guisan and Hofer (2003) also concluded that *A. fragilis* and *N. natrix* had strong responses to their resources (e.g. availability of anuran prey and reproductive sites for *N. natrix*), rather than climatic or topographic predictors, and suggest that conservation approaches must consider resource availability, as well as habitat and landscape provisions.

Study aims and hypotheses

The core aim of this research was to determine the key predictors of reptile population assemblages at multiple levels to determine how habitat structure, landscape connectivity and environmental variables effect population assemblages within a multifunctional landscape. The study particularly examined land use and compared agricultural sites with actively managed conservation sites, using reptiles as a focal group to help to build an understanding of the effect of land use practice and land management on reptiles in the UK.

Aims:

1. To quantify and asses the importance of environmental predictors for reptile population assemblages across the landscape.
2. To examine the suitability of habitat for reptiles and the connectivity within the wider landscape, by evaluating key predictors at microhabitat and landscape levels.
3. To inform conservation management within the study area and wider field of reptile conservation.

Hypotheses:

1. Temperature and related effects will be the greatest environmental predictors of reptile assemblages.
2. Habitat heterogeneity and structural variability will be the most significant physical predictors of reptile assemblages.
3. Land use type will be the greatest driver of reptile assemblages.

MATERIALS AND METHODS

Study area

The study site in southern England encompasses a variety of land use practices which are part of the multifunctional landscape surrounding Marwell Zoo: wildlife protected areas, arable farmland and orchard/pasture. During the study, five grassland sites were selected due to having favourable reptile conditions of south-facing edge habitat of scrub/woodland bordering on grassland (Sewell *et al.*, 2013). All five sites were within and around the Marwell Estate near Colden Common in Owlesbury, Hampshire (Figure 2). The Marwell Estate consists of woodland, grassland and marginal habitats, which total about 45 acres of land (Wilkie *et al.*, 2014) and lie within the South Downs National Park (SDNP), a mosaic of diverse landscape with varying habitat and land use types including grassland, heathland, ancient woodland, open farmland, arable and pastoral fields, villages and recreational areas (South Downs National Park Authority, 2017a). Agriculture is of social, cultural and economic importance within the region, with 85% of the land within the SDNP used for farming (South Downs National Park Authority, 2017b). There were approximately five key habitat types within the study area: arable land, grazed pasture, grassland, mixed woodland, and some small ponds.



Legend

Site 1 Site 2 Site 3 Site 4 Site 5

0 0.5 1 Kilometers

Figure 2: The Marwell Estate and surrounding area, with the study sites highlighted in colour.

Two of the five sites surveyed, Site 1 and Site 5 were agricultural land (orchard/pasture and arable farmland) whilst the other three, Sites 2, 3 and 4, were areas of conservation grassland managed by the conservation organisation Marwell Wildlife.

Site 1

Site 1 was privately-owned farmland with a mixture of beef pasture and orchard. There is a woodland corridor which borders the grassland, providing shelter and adding complexity to the vegetation structure. Two of the three transects for this site ran along the north of the orchard, whilst the third ran along the north of a field used for beef pasture.

Site 2 – Hurst Farm Field (4.6 Ha)

Hurst Farm Field has been relatively intensively managed for its hay production to maximise hay yields. To control noxious weeds the site received both chemical fertiliser and herbicide in 2011. The field was fertilised with organic matter (products from pond de-silting) in 2015, but has not been fertilised since. The hay is cut annually in July to minimise disturbance to wildlife and to allow seeds to set after re-seeding (Wilkie, 2017).

Site 3 – West Copse Field (10.3 Ha)

West Copse Field has received targeted conservation monitoring, resulting in the restoration of the northern half to chalk meadow. It has wide field margins surrounding the entire field, which are cut on rotation every five years. These margins provide rough grassland edges to encourage biodiversity. No fertiliser is added to this field. The hay is also cut annually in July, allowing the seed to set with minimal habitat degradation (Wilkie *et al.*, 2014; Wilkie, 2017).

Site 4 – Valley Top Field (4.5 Ha)

Valley Top Field sits within the zoo boundary and is bordered by hedgerows on the north, east and south sides and fencing on the west side. It receives no nutrient input and is also annually cut in July, with some spot-treatment carried out for ragwort as needed (Wilkie, 2017).

Site 5

Site 5 consists of arable farmland bordered by woodland and woodland corridors. The land is cultivated with rapeseed and is harvested annually.

Reptile Surveys

Artificial cover objects

To survey the reptile species present within the study area, artificial cover objects (ACO) were placed along transects within each site. Each site had three transects, totalling 15 transects and 195 ACO across all sites. ACO attract reptiles which shelter

and bask as they provide the surface cover needed for both refugia and thermoregulation (Figure 3). Using ACO is a straightforward method of reptile sampling as the ACO is simply lifted and the reptiles underneath counted and recorded (Hill, 2005; Dodd, 2016). The method has been proven to considerably increase detection rate of UK-native reptiles, and reduces instances of misidentification (Sewell *et al.*, 2012). ACO provide a standardised level of survey effort, meaning it is an easily replicated method of surveying. Additionally, it incurs less bias from the observer than surveying reptiles using only visual encounter methods (Dodd, 2016).

Roofing felt is an effective material for reptile sampling with low cost and labour implications, and was considered the best material for this research based on recommendations by JNCC (2004). ACO are recommended to be larger than 0.5m by 0.5m (Dodd, 2016), and for this study 1m by 0.5m pieces of roofing felt were used. The ACO was deployed along transects with a standard number per transect.



Figure 3: A) Roofing felt used as ACO. B) A grass snake and two slow worms using ACO found during a reptile survey (Images: authors own).

Transects and refugia placement

Each site had a total of three transects of equal length, approximately 120m with 13 ACO per transect. ACO were spaced roughly 10m apart, following the proposed standard method for surveying reptiles by Reading (1997). Transects were generally situated along south-facing edge habitat between grassland and scrub, an important linear feature for reptiles (JNCC, 2004; NARRS, 2014; Jofré *et al.*, 2016). ACO were placed in inconspicuous areas so the reptiles sheltering underneath them were not left vulnerable to disturbance by the public (Hill, 2005). Whilst Hill (2005) and Reading (1997) recommend placing the ACO in an array pattern of 37 refugia, the National Amphibian and Reptile Recording Scheme (NARRS) and JNCC Guidelines for reptile surveys were followed instead, which recommend placing ACO in areas of deep cover or edge of dense vegetation and away from sparse cover (JNCC, 2004; NARRS, 2014). This was more in line with the study aims, which examines connectivity of edge habitat in conjunction with reptile counts.

Survey technique

Reptile surveys took place for 10 weeks during April, May and June, a time period recommended in the NARRS (2014) guidelines. Using ACO to survey reptiles is particularly effective during this period, especially May and June, as reptiles actively seek out warm surface cover for thermoregulation during these months (Reading, 1997; Dodd, 2016). Additionally, the breeding season falls within April and May when reptiles are more active and less cautious, and therefore easier to sample (Hill, 2005).

Most searches took place in the morning when reptiles were likely to be basking (Reading, 1997). Visits were weather-dependent, with heavy rain and extreme temperatures (under 9°C and over 20°C) being unsuitable to survey (NARRS, 2014). Each transect was surveyed twice a week where possible so that each ACO was surveyed approximately 20 times.

Age class for slow worms and grass snakes was classified as either juveniles or adults. This is simpler with slow worms due to their bright gold colour as juveniles

(Beebee and Griffiths, 2000), and proved to be less feasible with grass snakes. However, juvenile grass snakes were classified as being less than approximately 55cm, as adults are roughly 50-60cm in length, dependent on sex (Beebee and Griffiths, 2000). Snakes estimated to be longer than 55cm were classed as adult. Sex was also recorded for slow worms as it is easily determined in the field by the presence of dark stripes along the females flanks and back.

Environmental variables

To understand environmental predictors of reptile presence and richness, several environmental variables were measured at the start of each transect. The variables measured were temperature, humidity, cloud cover, wind speed, ultraviolet radiation (UV) and overall weather condition. Temperature, humidity and wind speed were measured using a portable Kestral 3000 weather station. Cloud cover, estimated to the nearest 5%, and weather condition were based on visual assessment. Maximum UV data were obtained from the Reading University monitoring site (Defra, 2017).

Habitat structure

To determine species-habitat relationships in forest ecosystems (or in this case the edge habitat between woodland and grassland) it is necessary to understand the three-dimensional vegetation structure of the habitat (Froidevaux *et al.*, 2016). Data on the structural complexity, vegetation diversity, and microhabitat features for each transect were collected to understand the habitat structure and features corresponding with reptile presence. Habitat measurements were taken at 10m intervals along transects, corresponding with the placement of ACO.

Structural complexity and diversity of vegetation were measured using indices of vertical vegetation diversity for three height classes: percent cover 0-1m, 1-2m and 2-3m above ground, similar to a study by Sitters (2016). From this, a modified Foliage Height Diversity Index (FHD) was determined for each vertical point from 0m to 4m by estimating percentage cover of vegetation composition for each height class. The

formula for the Shannon Weiner index was used to calculate a value for the modified FHD index, following methods used by Berger and Puettmann (2000).

At each point the average vegetation height between 0m to 1m was also recorded (Berger and Puettmann, 2000). Proportions of vegetation composition for each vertical section was estimated and dominant vegetation type recorded for each point. Vegetation composition was recorded based on the following vegetation groups:

- Grasses
- Herbaceous plants
- Woody plants
- Open space

In total, a vertical column of 0-4m³ was assessed at each point to understand habitat heterogeneity of transects. The total percentage of vertical vegetation cover for each vertical column was calculated to determine habitat complexity and structure, as this influences thermoregulation and prey availability for reptiles (Willson, 1974; Dodd, 2016). These methods were based on those in a study by Willson (1974) which used both FHD and vegetation cover to determine the correlation between bird species diversity and habitat structure, although these methods were modified to accommodate for reptile species. The methods also mirror those used in a study on sand lizards which measured the structural complexity of sand lizard habitat by examining the horizontal vegetation layers of different height groups (House and Spellerberg, 1983).

To further characterise structural habitat features used by the reptiles in this study, additional measures of microhabitat features included presence of a canopy and presence of a dense canopy. Canopy cover and presence of connected dense vegetation were assessed similarly to a study by Singh *et al.*, (2002), which examined habitat use of lizards for thermoregulation in Australia. The following was recorded at each point:

- Presence/absence of a canopy (layer of foliage greater than 3m high)

- Presence/absence of a dense canopy (thick layer of foliage greater than 3m high allowing very little light through)
- Presence/absence of a dense ground vegetation patch greater than 0.5m³, further categorised into connected and unconnected to other patches.

Statistical analysis

Separate analyses were carried out using R statistical software (R Core Team, 2017) for environmental and habitat measurements due to differences in scale of analysis. Environmental measurements were analysed based on measurements taken per transect, whereas habitat measurements were based on microhabitat features within the transects and were measured at points along each transect in line with ACO.

Model selection

Generalised linear models (GLM) were employed to analyse the effect of environmental (Appendix 1) and habitat variables (Appendix 2), as well as their interactions, on predicting reptile presence and abundance. GLMs were used as they cope well with analysis of non-normal ecological data (Bolker *et al.*, 2009).

Models were selected by comparing the Akaike Information Criterion (AIC) values of each, with the aim of using a model with the lowest possible AIC result. Prior to running the models, data were explored by assessing distribution central values and the variance to mean ratio was calculated. Most of the data was highly skewed towards zero due to an excess of zeros in the frequency plots, and had a high variance-mean ratio. These indicated that the data were zero-inflated and highly over-dispersed (Zeileis *et al.*, 2008; Rodriguez, 2013; Crawley, 2015). To account for this, a zero-inflated Poisson model was initially employed for each response group. High deviance compared to the degrees of freedom indicated over-dispersion, which was resolved by specifying a quasipoisson distribution for the model. If inspection of diagnostic plots still indicated a poor fit, the data were inspected further. Calculations of probability of zero counts showed whether the model was underfitting the number of zero counts in the data. When this was the case, a hurdle model was used, which

works in two parts to analyse both zero count processes and positive count processes. The hurdle model was fitted through the 'pscl' package in R (Zeileis *et al.*, 2008). The data were then tested through calculations and plots to see if the hurdle model had addressed the issue of zero-inflated data. If plotting the data showed it was still over-dispersed, the model was fit with a negative binomial distribution rather than a Poisson distribution, using the 'MASS' package in R (Venables and Ripley, 2002). This was then plotted to see if there was a reduced difference between observed and predicted counts, and therefore if the model fit the data better.

Two series of models were fitted using this procedure, the models for environmental predictors and for habitat predictors. The same procedure was carried out further for the various response groups within each dataset: total counts, species counts, and age and sex class. These models were run with the aim of showing the amount of variation in reptile counts can be explained by the predictor variables (Guisan and Hofer, 2003).

Land classification maps

All maps were created through ArcMap 10.2.2 software (ESRI, 2014). In addition to context maps showing sites and transects, two maps were created in ArcMap for analysis purposes: a map of land use types and a map of connectivity of edge habitat. Edge habitat was chosen as a measure of connectivity as it is commonly used by both grass snakes and slow worms (Platenberg and Griffiths, 1999; Sewell *et al.*, 2013).

A supervised classification of land use type was carried out using a maximum likelihood classifier to categorise land types using an Ordnance Survey base map. The map was assigned six categories based on visual analysis of map features: woodland, grassland/natural cover, water, roads, paths and buildings. Training sites for a supervised classification model were digitised by drawing polygons around the features which most represented the colour palette of that category. A land cover map with digitisations of the six categories of land was produced. Land classification was checked against an aerial image to reduce risk of error in classification.

Landscape connectivity index

Connectivity with the wider landscape was also evaluated numerically through an index based on likelihood of links with edge habitat. Connectivity of edge habitat was selected over measuring connectivity of woodland or grassland, as both grass snakes and slow worms use the edge of woodland and grassland for both basking and shelter (Jofré *et al.*, 2016). Grass snakes especially show a preference for habitat boundaries, particularly for dispersal, using them as corridors of access for different areas within the landscape (Reading and Jofré, 2009).

Edge habitat was digitised into polygons for an area spanning a 1500m radius around a central point in the Marwell estate. These polygons were drawn and analysed using ArcMap extension software, Conefor (Saura and Torné, 2009). This software provided outputs based on likelihood of links to other habitats. These values were determined for each transect and were fed into the GLM as part of the habitat data.

RESULTS

The following analysis on reptiles applies only to grass snakes and slow worms, which were the only reptile species found in this study.

Reptile counts

Throughout the duration of the survey period, a total of 235 grass snakes and 548 slow worms were counted. This does not take recapture into account, which means many of these observations may be repeated. West Copse Field consistently yielded the most reptile observations. The total counts for each transect over the survey period is shown in Figure 4.

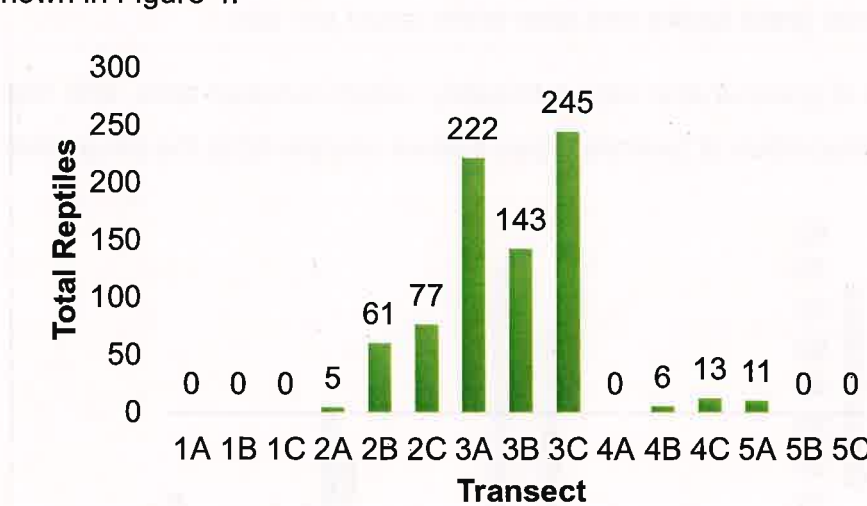


Figure 4: Total reptile counts for each transect.

Whilst grass snakes were more widespread between sites, slow worms had a higher population density concentrated within one site (Figure 5). Only two sites, Site 3 and Site 5 had multiple species with both slow worms and grass snakes found on site. Site 1 was the only site on which no reptiles were found throughout the entire survey period.

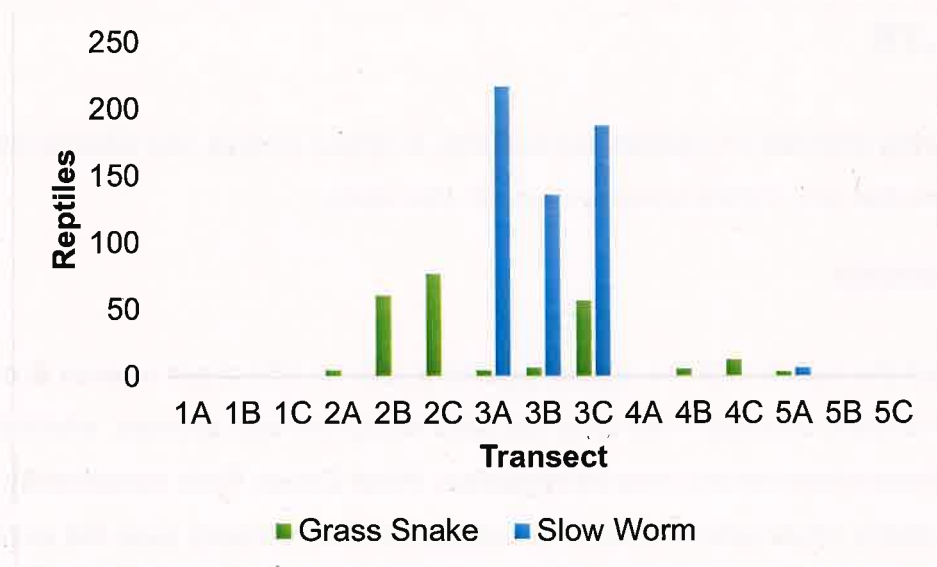


Figure 5: Total grass snake and slow worm count per site.

Age classes of grass snake were noticeably varied between sites, with Site 2 having a very high proportion of juvenile grass snakes compared to the other sites (Figure 6).

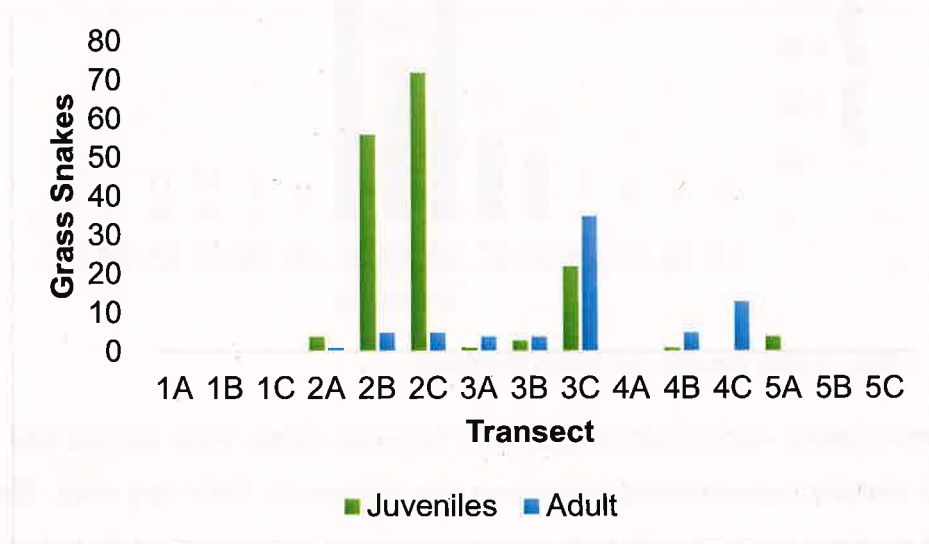


Figure 6: Grass snake age classes across transect.

Based on their size of about 25-30cm, it is estimated that the majority of juveniles on Site 2 were yearlings (Beebee and Griffiths, 2000) (Figure 7).



Figure 7: Juvenile grass snake, likely to be a yearling based on size of under 30cm.

Environmental predictors

The environmental data were generally zero inflated and highly over-dispersed. To account for this a hurdle GLM model with a specified negative binomial family (GLM.NB) was used. This model provides two outputs, one for positive-count processes (analysis of predictors of count data where reptile presence occurred), and one for zero-count processes (analysis of predictors of reptile presence/absence) (Ford, 2016).

Reptile counts

The most significant environmental predictors for total reptile presence (slow worms and grass snakes) identified by the model were temperature, UV, and the interaction of temperature and UV. Analysis of the positive-count processes indicated that the most significant environmental predictors of reptile counts were sunny conditions, sunny and cloudy conditions, and the interaction effect of cloud cover with UV (Table 1).

Table 1: Significant predictors of total reptile count, giving both the zero count and positive count outputs from a hurdle GLM.NB model (degrees of freedom=25, model AIC=1049.07).

Model output	Predictor	N	z-value	p-value
Zero count	UV and temperature interaction	290	-2.437	0.0148
	UV	290	2.407	0.0161
	Temperature	290	2.177	0.0295
Positive count	UV and cloud cover interaction	290	-2.595	0.00947
	Sun	290	-2.411	0.01589
	Cloud and sun	290	-1.982	0.04743

Inspection of the relationship between cloud cover and UV reveals that as cloud cover increased, UV decreased and thus the cause for significance is likely to be the effect of this trend (Figure 8).

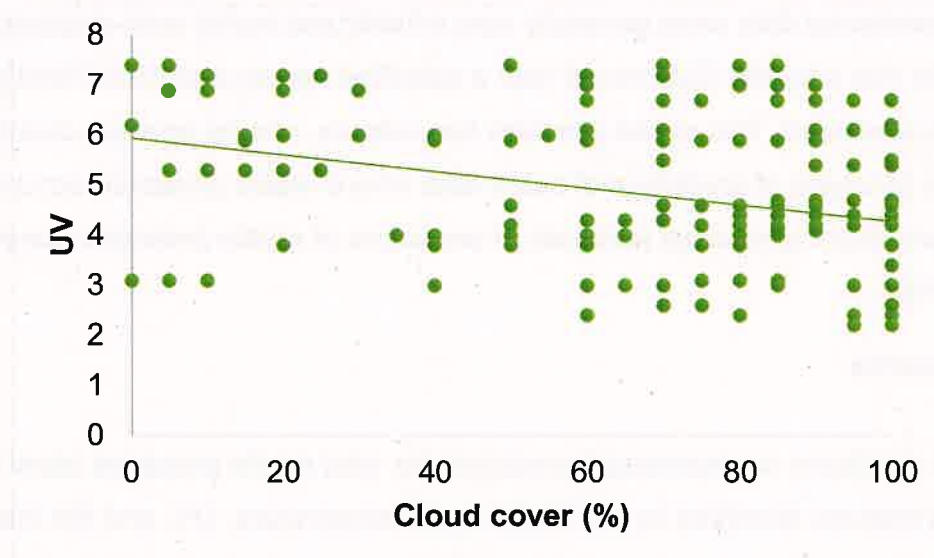


Figure 8: Relationship between UV and cloud cover.

A hurdle GLM.NB was also used to analyse total grass snake counts. The most significant predictors for grass snake presence indicated from the model (n=290, df=27, AIC=673.60) were the same as that of total reptiles: temperature (z-value = 2.422, p= 0.0154), UV (z-value=2.376, p = 0.0175), and UV interacting with

temperature (z-value= -2.50, p=0.0121). However, none of the predictors were found to be significant for positive count data.

The hurdle GLM.NB model did not fit the slow worm data, so a GLM.NB was used instead (n=290, null df=288, residual df=276, AIC=744.62). This analysis yielded similar results to the total reptile counts, with UV (z-value=1.962, p=0.04976), and the interaction between UV and temperature (z-value=-2.103, p=0.03548) identified by the model as significant predictors. The predictor with the highest significant effect on slow worm counts was sunny conditions (z-value=-3.683, p=0.00023).

Age classification

A GLM.NB model was used to analyse the significant predictors on grass snake age class. None of the environmental predictors showed any statistical significance in the model output for both juveniles (AIC=527.27) and adults (AIC=354.09).

There were several significant environmental predictors for juvenile slow worms (table 2), whereas the only significant predictor of adult slow worm count was sun (GLM.NB, n=290, z-value=-3.591, p=0.000329, null df=288, residual df=276, AIC=710.04).

Table 2: Output of a GLM.NB showing significant predictors of juvenile slow worm counts (n=290, null df=288, residual df=276, AIC=373.83).

Predictor	z-value	p-value
UV	3.489	0.000484
UV and temperature interaction	-3.454	0.000553
Temperature	3.136	0.001714
Sun	-2.949	0.003187
UV and cloud cover interaction	-2.047	0.040620

Sex classification

Two GLM.NB models were used to analyse the predictors of male and female presence. For both male and female slow worms, sun was highlighted by both models as the only variable which had a significant influence (Table 3).

Table 3: GLM.NB outputs showing the most significant environmental predictor of slow worm age class (n=290, null df= 288, residual df=276).

Sex	Predictor	z-value	p-value	Model AIC
Male	Sun	3.580	0.000344	463.51
Female	Sun	-3.168	0.00154	623.26

Habitat and landscape predictors

An appropriate model was selected using the same procedure as described in the methodology and for the environmental data. Similarly to the environmental data, the habitat data were also generally highly zero-inflated and over-dispersed, in which case a hurdle model was used. Again, two outputs were produced for the hurdle models. Depending on the response variable, a quasipoisson GLM was used if the hurdle model and GLM.NB did not fit the data.

Reptile counts

Modelling the effect of the habitat and landscape variables on total reptile count indicated that the significant predictors of reptile presence (zero count processes) were canopy, structured vegetation and dominant vegetation (herbaceous vegetation). The output for the positive count processes from the same model indicated that both the landscape and microhabitat measures of connectivity were significant predictors of reptile abundance (Table 4).

Table 4: Statistical outputs of hurdle GLM.NB giving the significant habitat predictors of reptile presence and abundance (n=187, df=23, AIC=715.76).

Model output	Predictor	z-value	p-value
Zero count	Canopy	3.168	0.00154
	Dominant vegetation type (herbaceous)	-2.371	0.01775
	Vegetation patch	2.112	0.03471
Positive count	Landscape connectivity	3.188	0.00143
	Habitat connectivity	3.087	0.00202

Further inspection of the relationship between reptile count and canopy, due to the result of canopy as the most significant predictor for reptile presence showed that it was likely to be presence of a canopy that influenced reptile presence (Figure 9).

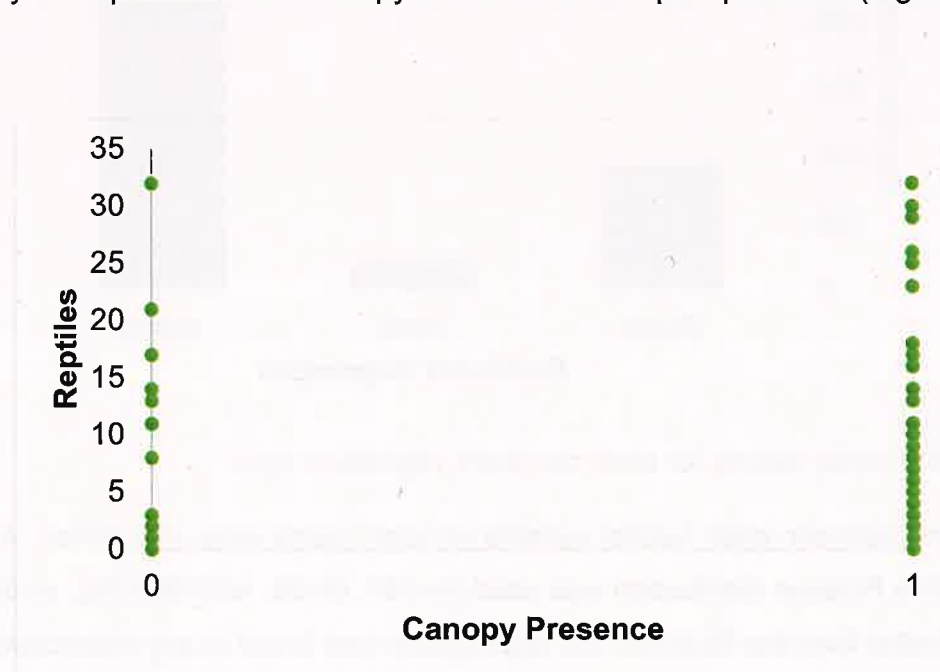


Figure 9: Number of reptile counts based on presence of a canopy (0=not present, 1=present).

Habitat type was analysed separately in a GLM with a negative binomial distribution ($n=187$, null $df=185$, residual $df=183$, $AIC=683.51$), and was shown to have a highly significant impact on reptile totals, with conservation grassland habitats having the greatest influence on reptile counts (GLM.NB, $z\text{-value}=7.007$, $p\text{-value}=2.43e^{-12}$). Points along the transects where woody vegetation was dominant had the most reptiles, followed by grassy vegetation (Figure 10).

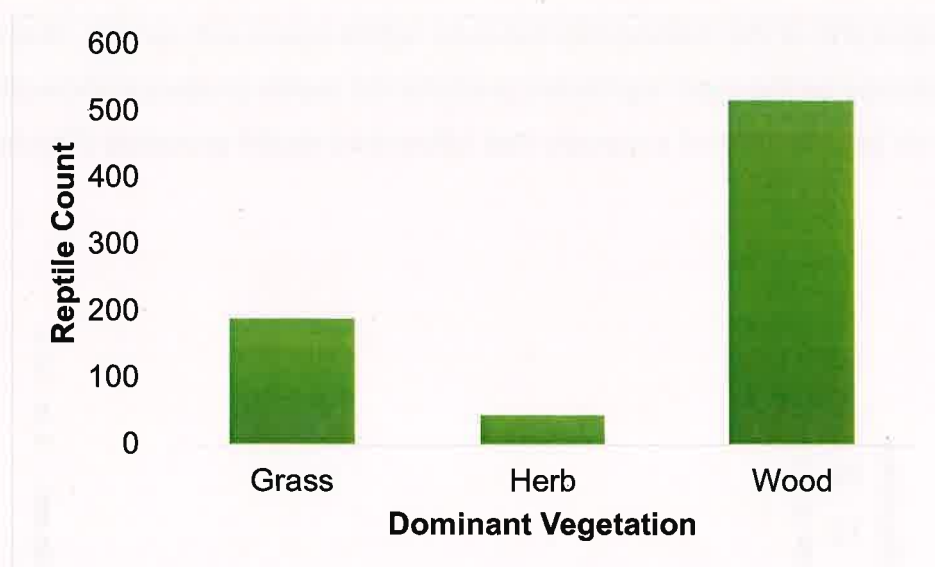


Figure 10: Reptile counts for each dominant vegetation type.

Interactions between each habitat variable on total counts were also tested. A hurdle model with a Poisson distribution was used ($n=187$, $df=20$, $AIC=909.39$), as this fit the data better than the GLM.NB. No significance was found in any interactions between the habitat variables for reptile presence, however there was found to be a significant effect on reptile abundance from the interaction between landscape and habitat connectivity ($z\text{-value}=2.060$, $p=0.039381$).

The most significant predictor of grass snake presence was presence of a vegetation patch, while the most significant predictor of count was microhabitat and landscape level connectivity (Table 5).

Table 5: Outputs from a hurdle GLM.NB ($n=187$, $df=23$, $AIC=493.31$) giving the significant habitat predictors of grass snake presence and counts.

Model output	Predictor	z-value	p-value
Zero count	Presence of suitable habitat patch	2.023	0.0431
	Landscape connectivity	-2.599	0.00936
Positive count	Habitat connectivity	2.280	0.02262

Many more reptiles were found at points along the transects which had a well-structured patch of suitable vegetation (Figure 11).

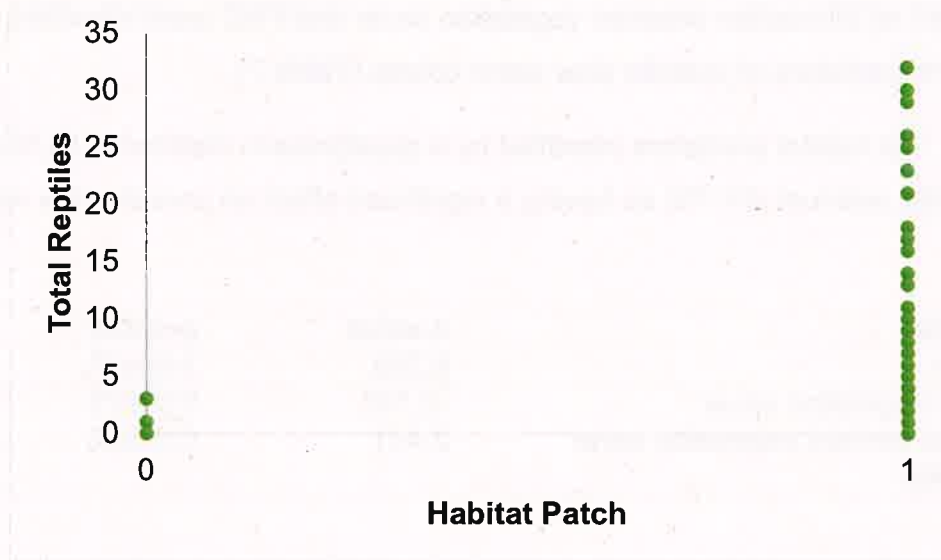


Figure 11: Relationship between patches of structured habitat and reptile counts (0=patch not present, 1=patch present).

Additionally, a GLM.NB (n=187, null df=184, residual df=175, AIC=488.18) showed that the interaction of FHD and percentage of vegetation cover had a significant impact on grass snake counts (z-value=-2.135, p=0.0327).

The most significant predictors of slow worm counts were presence of a canopy and dominant vegetation type, with herbaceous and woody vegetation also having a significant effect (Table 6).

Table 6: GLM with a quasipoisson distribution (n=187, null df=183, residual df=173) outputs showing the statistically significant habitat predictors of slow worm counts.

Predictor	z-value	p-value
Canopy	6.17	4.49e ⁻⁰⁹
Woody vegetation	-4.634	7.04e ⁻⁰⁶
Herbaceous vegetation	-3.931	0.000122

Age classification

A GLM with a quasipoisson distribution (n=187, null df=184, residual df=175) indicated presence of a canopy as the only significant predictor of adult slow worm presence (t-value=4.850, p=2.72e⁻⁰⁶). Canopy presence, percentage of vegetation

cover, and an interaction between vegetation cover and FHD were identified as significant predictors of juvenile slow worm counts (Table 7).

Table 7: The habitat predictors identified by a quasipoisson distributed GLM (n=187, null df=183, residual df=175) as having a significant effect on juvenile slow worm count.

Predictor	z-value	p-value
Canopy	6.205	3.84e ⁻⁰⁹
Percent vegetation cover	-2.742	0.00675
FHD and percent vegetation cover interaction	2.441	0.01563

Reptile counts were highest at points where vegetation cover was fairly moderate, about 15-35% (Figure 12).

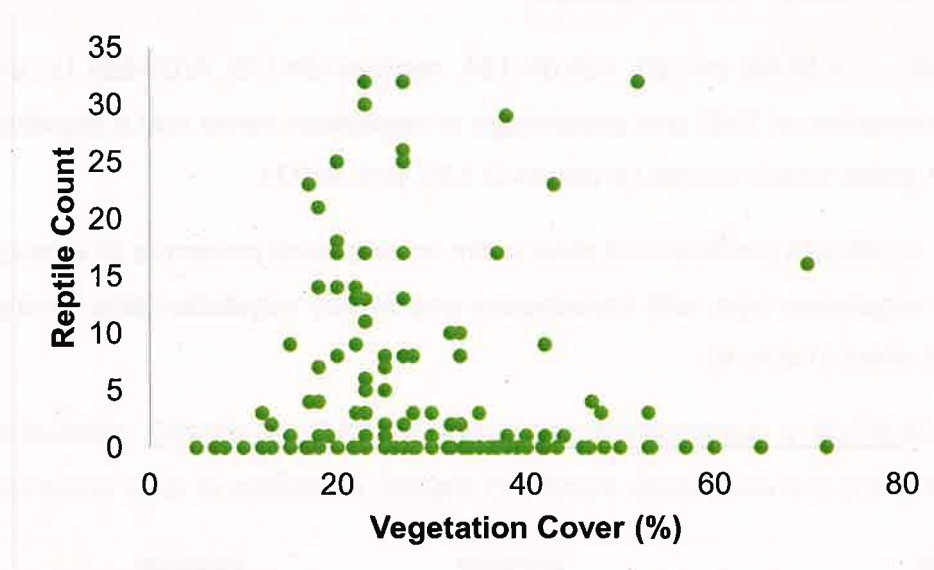


Figure 12: Relationship between vegetation cover and reptile counts.

Figure 13 shows the highly positive correlation between vegetation cover and foliage height diversity. Inspecting the data following the model results showed that greater vegetation cover and a higher FHD did not result in higher reptile totals. Instead a lower FHD index (about 0.03-0.05) and lower vegetation cover (about 15%-35%) resulted in higher reptile counts.

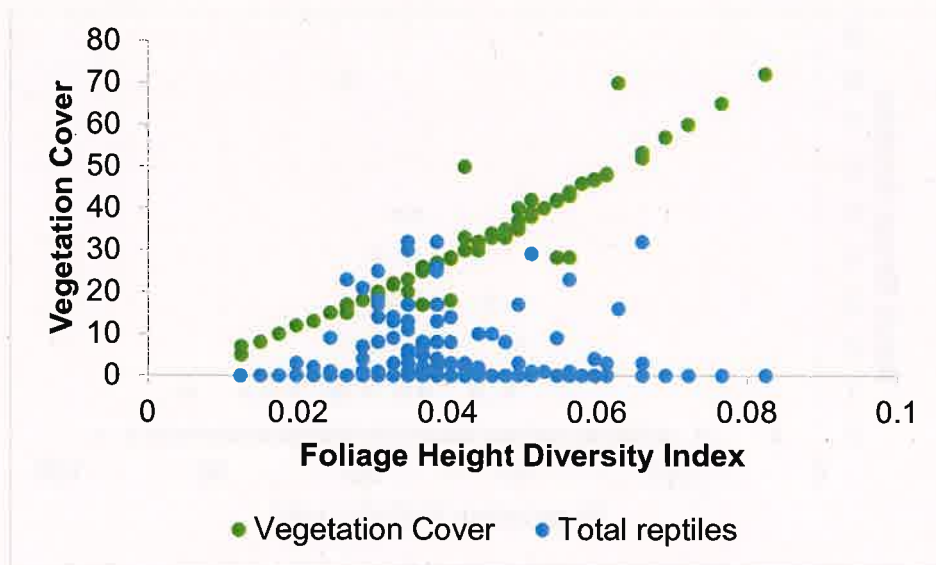


Figure 13: Reptile counts plotted against the interaction of vegetation cover and FHD.

A GLM.NB (n=187, null df=183, residual df=173, AIC=269.82) was used to highlight significant habitat predictors of adult grass snakes, and a hurdle GLM.NB (n=187, df=23, AIC=357.65) was used to analyse juveniles (Table 8). Landscape connectivity was the most significant predictor of both adult and juvenile grass snake counts. Average vegetation height had a significant effect on adults, whilst canopy presence was shown to influence juveniles.

Table 8: Habitat variables which were significant predictors of adult and juvenile grass snakes.

Age class	Predictor	z-value	p-value
Adult	Landscape connectivity	2.755	0.00588
	Average vegetation height	1.87	0.04757
Juvenile	Landscape connectivity	-3.504	0.000458
	Canopy presence	2.254	0.024196

A moderate vegetation height (about 40-80cm) yielded highest reptile counts (Figure 14).

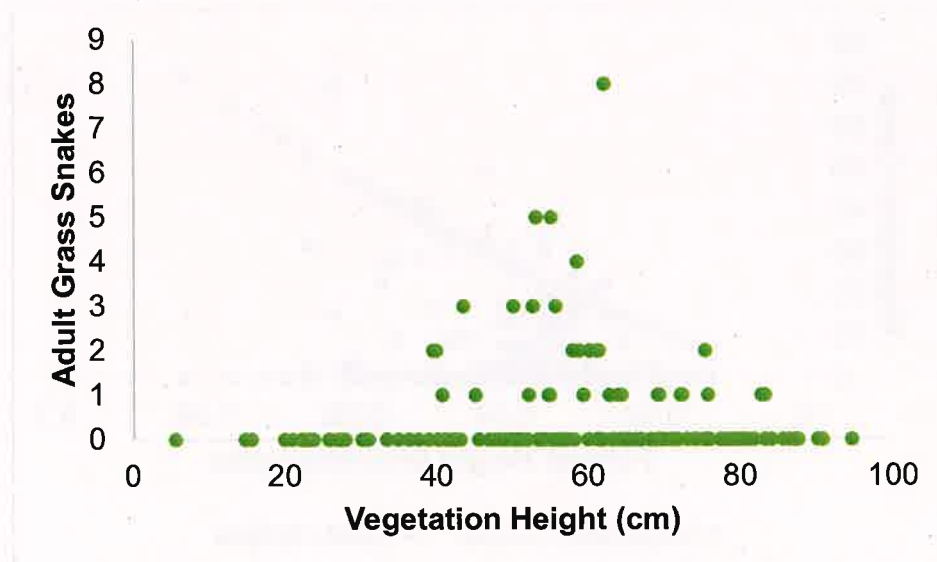


Figure 14: The relationship of average vegetation height under 1m and grass snake counts.

Sex classification

The significant predictors of male and female slow worm counts were the same as for total slow worm counts with woody vegetation, herbaceous vegetation and canopy presence all having a significant effect on count (Table 9).

Table 9: Model output from a GLM with a quasipoisson distribution (n=187, null df=183, residual df=173) of the habitat predictors of male and female slow worm counts.

Sex	Predictor	z-value	p-value
Male slow worm	Woody vegetation	-4.338	2.43e-05
	Herbaceous vegetation	-3.640	0.00036
	Canopy	2.833	0.00516
Female slow worm	Canopy	5.267	4.08e-07
	Woody vegetation	-4.506	1.21e-05
	Herbaceous vegetation	-3.789	0.000208

Connectivity within the wider landscape

As seen from the land classification map (Figure 15) there are visible barriers to biological dispersal across the landscape, mainly in the form of roads. Whilst the map also highlights several man-made paths, these are unlikely to hinder reptile movements. The map shows that there is less fragmentation between Sites 3, 4 and 5 whilst Sites 1 and 2 appear to be separated from these sites due to roads.

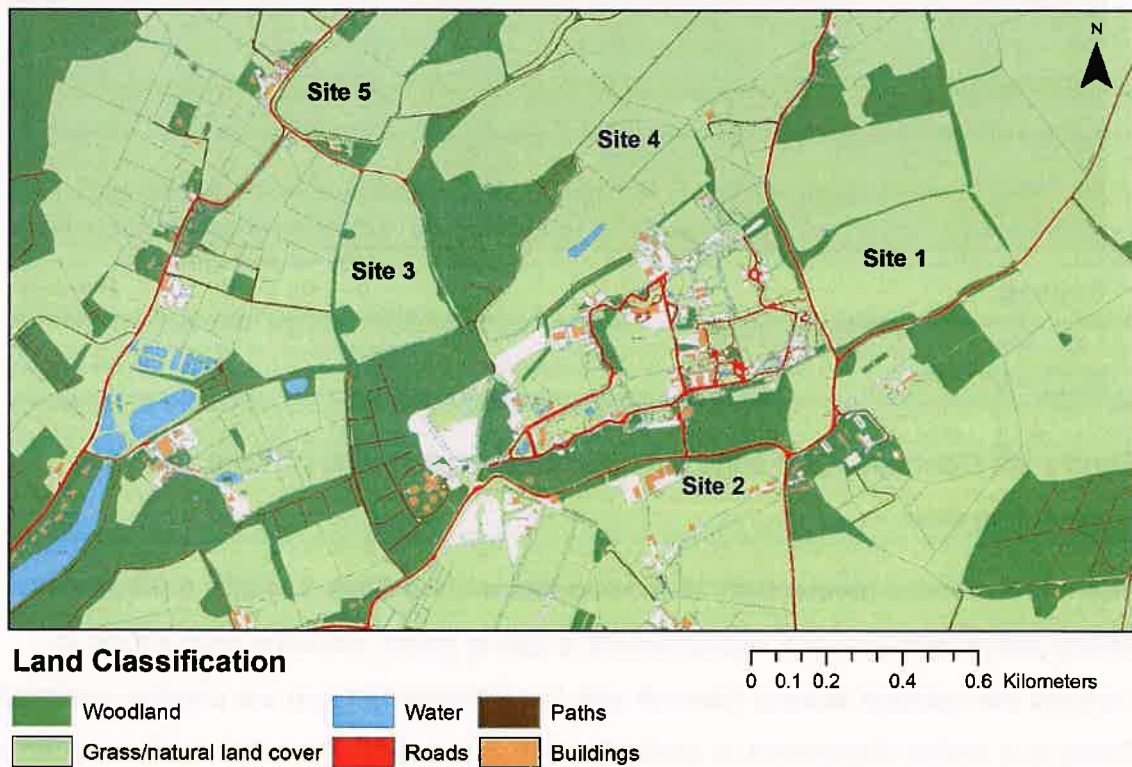


Figure 15: Land classification of key features, man-made and natural, within the study area.

From the large, connected areas of woodland and grassland and lack of roads Sites 3, 4 and 5 appear far more connected than 1 and 2. This is supported by the map of edge habitat, which was used to generate a connectivity index used in the GLMs (Figure 16). This map shows all edge habitat in the area, which in this case is the boundary between woodland and grassland habitats. Therefore, this shows the potential corridors for likely reptile movement around the sites.



Legend

- ☆ Sites
- 1500m radius
- Edge habitat

0 0.5 1 2 Kilometers

Figure 16: Connectivity of edge habitats between sites and with the wider surrounding area.

There is extensive connectivity of the habitats around Sites 3, 4 and 5 (Figure 17), where active conservation management is taking place, showing how efforts to connect the habitats around Marwell with the wider landscape are proving successful. There is a visible disconnect in particular with Site 1, both from the roads as seen in Figure 14 and from the lack of connective habitat corridors that reptiles are likely to use for movements.



Legend

- ☆ Sites
- 1500m radius
- Largest habitat connection

0 1 2 Kilometers

Figure 17: The largest extension of connective habitat.

DISCUSSION

General findings

Slow worm presence only in Site 3 reflects slow worm behaviour and movements. Slow worms prefer areas of woodland and thick cover and are rarely known to bask in open areas as they prefer to thermoregulate via surface refugia and ACOs (Beebee and Griffiths, 2000). The dense woodland surrounding Site 3 provides this cover for them, whilst the structured edge between woodland and grassland provides them with basking opportunities within dense cover. Slow worms are not wide-ranging whereas grass snakes are: slow worms have an average home range of about 200m, whilst the home range of grass snakes has been estimated at 2.4ha per year (Langton and Beckett, 1995; Beebee and Griffiths, 2000; Reading and Jofré, 2009). Grass snakes have the largest home range of all British reptiles and forage the most widely, which is likely why they were found most widely between sites (Beebee and Griffiths, 2000). Slow worms generally exhibit higher population sizes than grass snakes as they feed only on invertebrates (Beebee and Griffiths, 2000), explaining the high counts at Site 3 but low dispersal. As Site 3 has been more actively managed than the other sites (Wilkie, 2017) these findings indicate effective management. It is hypothesised that one reason for success is the presence of wide field margins as Sites 2 and 3 were the only sites with this feature and were shown to be most suitable for reptiles. Field margins consist of a marginal strip which runs between the agricultural crop and field boundary, providing access and other functions for wildlife (Marshall and Moonen, 2002). These findings meet the study aims as it suggests effective management of Site 3, which can be applied to other sites.

The high number of juvenile grass snakes at Site 2 suggests high reproductive successes in recent years. This could be attributed to climate, as the winters of 2015/16 and 2016/17 were very mild, with 2015/17 being the warmest winter in England since 1910 (Met Office, 2017). Another possible explanation for such a high juvenile to adult ratio could be proximity to nesting sites, which other studies have attributed high juvenile counts to (Mertens, 1995). Further habitat assessments are

necessary to determine where these could be. Juveniles generally tend to disperse away from where they hatch (Ciesiołkiewicz, 2006), so it is likely that there is a nearby habitat acting as a population source for this site. For example, the rough grassland directly adjacent to Site 2 (Figure 15) is ideal habitat for grass snakes and could have suitable nesting sites.

Environmental predictors

The environmental predictors which had a significant influence on the response groups were temperature, UV, sunny conditions, sunny and cloudy conditions, and the interaction effect of UV and temperature, and UV and cloud cover. These findings make biological sense and fit well within the literature on UK reptiles. All these variables and the interactions appear to relate to reptiles need to thermoregulate. Due to their reliance of external heat sources for energy (Edgar *et al.*, 2010), the greatest predictor of reptile richness is generally temperature (Qian, 2010). In this case, other factors with relate to basking such as UV, sun and cloud have also been found to be significant predictors. These findings, mainly of the significance of temperature, confirm Hypothesis 1 which predicted that temperature would be the greatest environmental predictor of reptile groups.

Reptiles regulate their body temperature through external heat sources, which in turn governs their physiological functions and is vital in allowing them to function at peak efficiencies (Beebee and Griffiths, 2000). To do this, reptiles actively seek warm sites or direct sunlight to raise their body temperatures in order to maintain essential functions such as movement and digestion (Isaac and Gregory, 2004; Edgar *et al.*, 2010). Warm temperatures are associated with higher light levels (Greenberg, 2001), and basking allows reptiles to absorb solar radiation directly from the sun or allows them to use external substrate to heat their bodies via convection (Beebee and Griffiths, 2000). This behaviour supports the significant finding of the effect of UV, sun and cloud effects, as these variables are representative of solar radiation, and its importance for reptiles. The significance of the interaction between UV and cloud cover supports this further, as the statistical significance as well as inspection of Figure 8 indicates that the interaction between high UV and low cloud cover

significantly predicted reptile counts. Cloud cover has a significant effect on UV within short timescales (Calbo and González, 2005) which could be why it was highlighted within the model.

An interesting finding was that sunny conditions were shown to significantly predict total reptile counts and all slow worm groups, but not grass snakes. As both species thermoregulate it was surprising to see such a high statistical significance of sun for slow worm counts but not the same for grass snakes. A potential reason behind this could be that slow worms are known to bask in direct sunlight or under sun-warmed objects (Capula and Luiselli, 1993), whereas a study on the thermoregulatory behaviour of grass snakes showed that during sunny, warm periods grass snake activity such as foraging or searching for mates increased (Isaac and Gregory, 2004). The study by Isaac and Gregory (2004) showed that grass snakes were limited in their activities at low temperatures due to slowed physiological processes, but were more active in periods of full sun, when temperature was higher. This is supported by Ciesiołkiewicz *et al.*, (2006) which also showed that grass snakes are much more active and mobile under warm, sunny conditions.

Therefore, it is likely that grass snakes were more active during sunny conditions and therefore not using the ACO for heat, particularly as British reptiles invest time in behavioural thermoregulation due to restricted sunshine availability and low optimal temperatures (Beebee and Griffiths, 2000). Furthermore, grass snakes rely on direct solar radiation for thermoregulation, whilst slow worms thermoregulate via contact with warm surfaces (Beebee and Griffiths, 2000). Grass snakes have a much less rigid basking behaviour than other British reptiles hence it is highly possible they utilised the ACO less than the slow worms, particularly in sunny conditions when they were likely basking elsewhere in direct sunlight or being active.

These findings are important to the study aims as they show that the greatest predictors of reptile counts are based on thermoregulation, which in turn governs their habitat preference (Singh *et al.*, 2002). From this, effective management techniques for reptile specific conservation purposes can be determined.

Habitat predictors of reptile groups

Dominant vegetation type

The results showed that areas where herbaceous vegetation was dominant significantly predicted overall reptile counts and slow worm counts (Figure 10). It is likely that lack of herbaceous vegetation was the predicting effect, as points of dominantly herbaceous vegetation had the lowest reptile counts out of all three vegetation types. This is likely because woody and grassy vegetation patches are more suitable to reptiles as they offer shelter, are less damp and let in more light than areas of predominantly herbaceous vegetation.

Woody vegetation was identified as a significant predictor of slow worm counts. Slow worms are known to favour woodland habitats as well as grassland and heathland (Platenberg and Griffiths, 1999), and use structured vegetation patches, generally consisting of woody vegetation, to thermoregulate via their surroundings rather than direct sunlight (Beebee and Griffiths, 2000). Grass snakes tend to avoid woodland, which is potentially why woody vegetation was not a predictor of grass snake counts (Reading and Jofre, 2009).

Canopy

Canopy was shown to have a significant effect on four of the modelled response groups: total reptile counts, adult slow worms, juvenile slow worms and total slow worm counts. Clearly canopy presence was significant for slow worms rather than grass snakes. Azor *et al.*, (2015) highlight that reptile response depends on habitat and species type, and a study by Todd and Andrews (2008) showed that in many cases reptiles benefitted from presence of a canopy, as it provides habitat structure and leaf litter which can be used for cover. However, this was an interesting finding as removal of canopy cover is used as a tool in reptile conservation to restore reptile assemblages (Pike *et al.*, 2011). Studies have suggested that gaps in a canopy result in higher light levels and therefore influence reptile abundance (Greenberg, 2001), whilst presence of a continuous canopy blocks solar radiation and result in less favourable habitat (Azor *et al.*, 2015). Reduced canopy and tree density results in

increased light availability for plants underneath, increasing plant diversity and structural diversity (Azor *et al.*, 2015), and changes the quality of the thermal habitat (Dodd, 2016).

This finding could also however be a result of bias, as the sites with highest reptile counts (Site 2 and 3) were bordered by dense woodland which the other sites lacked. Furthermore, as transects were selected based on southern-facing locations which catch the sun, it could be that the canopy had less effect on the amount of light that reached the ground than in locations that are not south-facing.

Habitat type

Habitat type was shown to significantly influence total reptile counts, with conservation grassland being a key predictor. This result supports Hypothesis 3, as it was expected that areas of actively managed grassland would be more suitable to reptiles especially in comparison to the two agricultural sites. This is because agricultural landscapes account for 74% of the threats faced by reptiles, as they disturb and alter the natural landscape, change microhabitat structure and disrupt connectivity (Dodd, 2016). Several studies on grass snakes have shown that they experience high levels of disturbance in arable farmland areas from crop management and avoid fields used for grazing, explaining the lack of reptiles on Site 1 and 5 (Reading and Jofré, 2009). However, there have been conflicting findings on how grass snakes utilise agricultural areas. Madsen (1984) showed that grass snakes only used arable land for movements between edge habitats, whereas Wisler *et al.*, (2008) showed that grass snakes used monoculture habitats for basking and avoiding predators. However, even whilst they used agricultural land they still demonstrated a clear preference for edge habitat (Wisler *et al.*, 2008). It was unexpected that no reptiles whatsoever were found on Site 1 for numerous reasons. The microhabitat was suitable for slow worms, and it was highly proximal to areas where grass snakes have been found, on the very right edge of Marwell Zoo. It is likely that the road which separates Site 1 from the zoo (Site 1) prevented movements to this site (Spellerberg, 1975). The same conclusion was made about dispersal from Site 2, which is disconnected from surrounding habitat by roads (Figure 15). These findings are in

line with the study aims as they give insight to how landscape connectivity affects reptiles.

Vegetation height and cover

Vegetation height and percentage cover were both highlighted in the model as having statistical significance on adult grass snakes and juvenile slow worms, respectively. As both the wider literature and results from this study show, reptiles are thermally limited in their activity by their environments (Isaac and Gregory, 2004). All UK reptiles prefer well-structured habitat which provides foraging opportunities and shelter whilst also meeting thermal requirements (Jofré *et al.*, 2016). Their thermal requirements affect their habitat preference, meaning that vegetation structure is a key influence in reptile presence (Singh *et al.*, 2002). In addition to needing open areas to bask, reptiles also need structured cover proximal to basking sites so that they can forage, shelter and avoid overheating during thermoregulation, which can be fatal (Edgar *et al.*, 2010). To precisely maintain correct body temperatures throughout the day, reptiles actively switch between open areas for heat and areas of vegetation cover to cool themselves, particularly along edge habitat (Edgar *et al.*, 2010). A study by Pettersson (2014) revealed that grass snakes were found to spend much of their time in areas with high vegetation because of this behaviour, and slow worms generally have a preference for microhabitats with high vegetation cover (Jofré and Reading, 2012).

The relationship between reptiles and vegetation cover was that most reptiles were found in areas of moderate cover, about 20%-40% (Figure 12) and moderate height, about 40%-80%. Whilst this does not necessarily agree with Pettersson (2014), low vegetation cover and strong radiation are features of the general habitat preferred by reptiles according to Azor *et al.*, (2015). These results indicate levels of vegetation cover and height which seem to provide adequate levels of light and shelter for reptiles, achieving the study aim of quantifying habitat suitability.

FHD and vegetation cover

The interaction between FHD and vegetation cover was statistically significant in predicting grass snake totals and juvenile slow worms. This interaction was based on a fairly low FHD and vegetation cover resulting in higher grass snake and juvenile slow worm counts (Figure 13).

The finding of significance from this interaction makes sense based on reptiles' need for cover and structural diversity. It is particularly understandable that it was significant for juvenile slow worms, which are at high risk of predation by both small and larger animals and require more cover (Beebee and Griffiths, 2000). However, the significance based on lower values for both cover and FHD was interesting as vegetation cover and diversity is such a driving factor in reptile presence, and has been shown to determine survivorship in reptiles (Jofré *et al.*, 2016). Due to the importance of structural diversity as specified by the literature on reptiles, is also surprising that FHD was not a significant predictor for more reptile groups, and that FHD was not linked to higher reptile numbers. This could be a result of the methodology and survey technique for measuring FHD, as FHD was based only on general vegetation groups rather than individual plant species, which would give a much more accurate representation of structural diversity.

Areas of high native vegetation cover are important for reptiles (Dodd, 2016), and a study by Lindenmayer *et al.*, (2005) showed that reptile species richness increased with native vegetation cover. Grass snakes need suitably dense vegetation for cover from predators (Reading and Jofré, 2009). Additionally, reptiles receive varying cooling effects (e.g. rapid or gradual) based on vegetation structure, and therefore high plant heterogeneity is widely cited as a desirable feature for reptiles (Edgar *et al.*, 2010). It is interesting therefore that FHD did not account for more significance on reptile counts. These findings refute Hypothesis 2, which predicted that higher heterogeneity and structural variability in vegetation would lead to greater reptile counts. The results indicate there is a threshold above which cover becomes a limiting factor, possibly due to reliance on solar energy for thermoregulation (Qian,

2009). These findings are important as they reveal a need for further research to better explain the result.

Vegetation patch

British reptiles require habitats which provide warmth, structural complexity, and habitat connectivity (Edgar *et al.*, 2010). The presence of a patch of well-structured vegetation of about 0.5m³ was a significant predictor of reptile totals, grass snake totals, and juvenile slow worms, with very few reptiles found in areas which had no patch (Figure 11). This finding is supported by the literature which outlines the importance of microhabitat features such as cover for reptiles (Dodd, 2016; Jofré *et al.*, 2016). British reptiles use patches of dense vegetation for basking and shelter (Edgar *et al.*, 2010) and have been shown to be found close to dense vegetation (Pettersson, 2014).

In addition to presence, the connectivity of these patches was significant for total reptile counts and grass snakes. Dispersal abilities of reptiles are limited, hence why connectivity is so important for reptiles (Edgar *et al.*, 2010). The findings support this, as results show reptiles responded well to well-connected areas of suitable cover. The results clearly specify the importance of connectivity at multiple scales, as both patch and landscape connectivity as well as their interaction were statistically significant.

Importance of landscape connectivity

Landscapes are mosaics of habitats which vary in heterogeneity (Dodd, 2016). Movement between habitat patches is dependent upon the landscape, as well as the organism which is moving through it (Tischendorf and Fahrig, 2000). Figure 16 illustrates how the study site consists of a matrix of connected and unconnected edge habitat. Assessing landscape connectivity is crucial in any form of conservation and is highly relevant to reptiles (Dodd, 2016). The measure of landscape connectivity used in this study takes corridors into account, which are continuous strips of habitat which connect two habitat patches structurally (Tischendorf and Fahrig, 2000). These

corridors contribute to connectivity in varying extents, dependent on the nature of the corridors and animal response to it (Tischendorf and Fahrig, 2000).

One of the key hypotheses of this study was that sites which were well-connected and proximal to well-structured vegetation such as woodland edge would yield significant populations. However, from comparing Sites 3 and 4, this does not appear to be the case. The woodland adjacent to Site 3 provides a highly well-connected pathway for reptile movement between Site 3 and 4, yet the lack of slow worms at Site 4 proves that it is not being used as such (Figure 17).

Conversely, from looking at Site 1 which lacks dense woodland yet has a small connective strip of woodland running along it, it is clear that reptiles are not moving between Sites 2 and 1. This raises the question, what type of connective habitat do slow worms and grass snakes require to move from one place to another? Reptiles generally move through well-structured habitat which provides safe passage from predation, and use corridors such as hedgerows, ditches, stone walls, meadows, orchards, field margins, ponds and manure heaps (Dodd, 2016). In this instance, it could be that the woodland adjacent to Site 3 is too dense to provision for the basking needs of slow worms, whilst the connective strip along Site 1 is too small to provide adequate shelter and foraging opportunities. To examine this further, more variables relating to slow worm and grass snake presence would have to be measured, in this instance prey availability and light levels.

Landscape effects and fragmentation

It is hypothesised that the southern edge of Site 1 could harbour grass snake populations, due to its proximity to relatively dense woodland and Site 2 which has a confirmed population. If this were so, it would envisage that the snakes are limited in their movements by the roads, and that the connected route of woodland on the western and eastern edges of the site does not meet the species requirements for movement or presence. Road networks are responsible for much of the fragmentation of reptile habitats (Spellerberg, 1975), and distance to roads have been shown to negatively correlate with snake presence (Wisler *et al.*, 2008). Whilst it may appear

that the lack of species on Site 1 is due to the presence of a busy road on the western edge of the site, this observation should be treated with caution as the same road cuts through the woodland north of Site 2, which had a substantial grass snake population (Figure 15).

The importance of connectivity in predicting reptile assemblages helps explain why Sites 1 and 5 had hardly any reptiles. Natural habitat connectivity is disrupted by agricultural landscapes and ranges of grass snakes in an agricultural landscape are different than in an undisturbed landscape (Reading and Jofré, 2009; Dodd, 2016). The ranges of snakes in these landscapes depend on cover availability, and they tend to follow hedged field edges and banks and avoid open fields and woodland (Reading and Jofré, 2009), meaning that the hedgerows and connecting features between these landscapes did not provide adequate cover for reptiles.

Recommendations for conservation

Since canopy cover was a significant predictor of reptile counts, it is recommended that the canopy be retained. However, as temperature, sun and UV were significant predictors of reptile counts, opening the woodland to create areas of sun-exposed habitat within the woodland could be beneficial (Pike *et al.*, 2011). This adds structural complexity to the woodland and benefits invertebrates, amphibians, and mammals as well as reptiles (Pike *et al.*, 2011).

Based on the findings of this study, it is recommended that to enhance the suitability of the landscape for reptiles, conservation efforts should focus on maximising connectivity. A habitat must be suitable at a multitude of scales, at the microhabitat level as well as the landscape level, to appropriately meet species needs (Mayor *et al.*, 2009), so conservation should focus on both wider landscape connectivity and connectivity within a site. There must be high connectivity both within and between sites for reptile dispersal (Edgar *et al.*, 2010). Connectivity facilitates gene flow and acts as a buffer for catastrophic events, reducing risk of populations becoming extinct (Dodd, 2016). This is particularly important as doing so incurs an mitigative approach

to biodiversity loss, rather than adaptive measures of dealing with biodiversity loss such as attempting to restore a threatened population (Novacek and Cleland, 2001).

Habitat retention, particularly of linear strips, and increasing the connective corridors across the landscape is highly recommended, particularly in the agricultural landscapes. Edge habitats, particularly those with structural vegetation attributes, should be managed and protected (Dodd, 2016). It is also recommended that where possible, hedgerows should be established or enhanced. Hedgerows are a critical element of the landscape and hedgerows composed of trees and shrubs best facilitates animal movement and creates connective corridors (Forman and Baudry, 1984). A key recommendation for the agricultural sites in particular is to establish wide field margin strips of at least 6m along arable fields (Kleijn *et al.*, 2006). Wide field margins provide refugia for many species and are a highly effective conservation tool in arable landscapes (Moonen and Marshall, 2001). Field margins hold high wildlife value and are often adopted on agricultural land as part of agri-environment schemes in the UK (Vickery *et al.*, 2009). Whilst species translocation can be used as a conservation technique (Spellerberg, 1975; Platenberg and Griffiths, 1999) this is not recommended based on the study findings, as increased connectivity from Site 3 could result in a natural increased distribution of slow worm populations across the landscape.

Another management technique for reptile-based conservation is the addition of certain habitat features desirable for reptiles to the wider landscape. These include potential nesting or oviposition sites for egg laying, such as manure piles on agricultural land, and shelter sites/hibernacula such as coarse woody debris, leaf litter or artificial burrows (Dodd, 2016). A key recommendation by Beebee and Griffiths (2000) for managing reptile populations within agricultural landscapes is to revert to traditional farming techniques where possible. There is no reason for grass snakes not to be present in farmlands which use less intensive agricultural management practices, and traditional agricultural features such as a manure heap are highly beneficial as they provide oviposition sites (Beebee and Griffiths, 2000). A collaborative approach to conservation between Marwell Wildlife and the surrounding

landowners could be highly beneficial for this landscape. From Figure 17 it is clear that the conservation actions of Marwell Wildlife are responsible for much of the connectivity and applying this to the wider landscape would have significant positive effects for reptiles and other groups. Whilst these recommendations are based on the study area, they can be applied at multiple scales and used in the context of wider reptile conservation.

Study limitations

This study provides only a snapshot of reptile population assemblages due to the very short-term nature of data collection, meaning no long-term trends can be revealed as to population size or health of these species (Gibbon *et al.*, 2000). Duration of research can have a crucial impact on the findings of a study, with longer term studies often drawing different conclusions from shorter term research (Gibbon *et al.*, 2000).

There were many limitations surrounding the collection of environmental variables. Data recorded using the handheld environment metre was representative of current weather conditions only, not the earlier conditions which determine reptile movement. Whilst using data from external sources was considered, it was decided that the temperature readings taken would act as an indicator of the conditions that day, as any other data would not take temperature differences between sites into account. The data were also biased based on sampling conditions as surveying did not take place on days with poor weather conditions.

Another limitation arises from the fact that habitat features were only recorded once, due to time constraints of the project. This means that only a snapshot of the structural environment across the transects was taken, rather than a more accurate representation of vegetation over three months.

This study was further limited by not incorporating certain variables measured in similar studies (Guisan and Hofer, 2003). Important predictors which were unable to fit into the scope of this project were topographical variables and additional climatic predictors such as rainfall. It was decided that rain would not be included as a

predictor variable in this study as surveying generally took place in dry, warm conditions and therefore would incur too much bias. Topography has been shown to more effectively predict species distribution than climate in some instances, as topographical predictors often depend on climatic predictors. However, this was not so much of a problem in this study as it focused on a local scale where climate was generally the same (Guisan and Hofer, 2003).

Recommendations for further study

This study focused on edge habitats, meaning certain habitat types suitable to reptiles were overlooked. A follow up study could survey a more diverse range of locations, which could potentially lead to finding other reptiles in the area such as common lizard or adders. Another recommendation is that data loggers (e.g. iButtons) or a similar instrument be used to collect live data such as temperature be deployed to obtain the most accurate environmental conditions possible.

It is also recommended that a follow up study measure FHD more intricately, perhaps based on number/percent cover of plant species rather than groups. This could examine the threshold at which FHD, vegetation height, and vegetation cover stop predicting reptile counts. Additionally, if the study is replicated habitat features should be measured at least once a month to account for changes in vegetation during this time, and its possible effect on reptiles.

The key recommendation for another, longer term study is to examine reptile movements across the landscape in more detail to build upon and better understand the inferences made about landscape connectivity and reptiles within this study. Mark-recapture techniques are recommended to determine inter-individual variability to determine how individuals use the landscape (Dodd, 2016). Additionally, a radio tagging approach similar to those used by Reading and Jofré (2009 and Pettersson (2014) could be used to measure reptile movements. Tracking reptile movement would provide additional data on their use of and movements through the landscape whilst further quantifying habitat requirements, foraging strategies, interspecies interactions and physiological tolerance (Dodd, 2016).

Conclusion

The findings of this study show that temperature and its related effects highly predict reptile assemblages, and seem to determine their habitat use. Areas of tall vegetation and high plant diversity did not yield the greatest reptile counts, likely due to the balance of light/heat and structural diversity required by reptiles. Connected habitats appear to yield more reptiles yet more research is needed to further quantify reptile movements, perhaps through radio tagging. Many more reptiles were found in areas of managed conservation grassland than in agricultural areas, which is likely to be due to disturbance from land use practice and from lack of sufficient connective corridors across the agricultural landscapes. Suggestions for reptile-focused conservation management include increasing connectivity through wide field margins and hedgerows, managing areas of preferred microhabitat such as achieving a balance between structural heterogeneity and light levels, and collaborating with land managers so that landscape-scale conservation success can be achieved.

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APPENDIX

Appendix 1: The environmental predictors analysed in a GLM to determine their effect on the response variable, reptile counts.

Environmental predictor	Description	Units
Cloud cover	Estimation of percentage of cloud cover	%
Condition	Description of general weather condition	C = cloud, R= rain, S = sun
Humidity		%
Temperature		Degrees Celsius
UV	Maximum UV over 24 hours	UV total index
Wind speed	Wind speed	Miles per second

Appendix 2: The habitat predictors analysed in a GLM to determine their effect on the response variable, reptile counts.

Habitat predictor	Description	Units
Canopy	Presence of a canopy greater than 3m high	Yes/No
Dense canopy	Presence of a dense vegetated canopy greater than 3m high	Yes/No
Foliage height diversity (FHD) index	An index of the structural diversity of vertical vegetation	Index
Landscape connection links	A measure of landscape connectivity, the number of potential links to suitable edge habitat used by reptiles	Index
Microhabitat connection links	Whether the structural vegetation at the ACO is connected to other vegetation patches	Yes/No
Structured vegetation patch	Presence of a vegetation patch providing structural cover greater than 0.5m ³ , which provides potential cover	Yes/No
Vegetation cover	The percentage of total vegetation cover	%
Vegetation height	Average vegetation height of vegetation under 1m high	Centimetres (cm)