The influence of landscape heterogeneity on amphibian species richness in Malaga province, Spain

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February, 2010

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by

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Thesis submitted to the International Institute for Geo-information Science and Earth Observation in partial fulfilment of the requirements for the degree of Master of Science in Geo-information Science and Earth Observation, Specialisation: (Natural Resource Management)

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“We never know the worth of water till the well is dry”

Thomas Fuller, *Gnomologia*, 1732

Dedicated to my loving wife and wonderful son.

Regina Wambui and Kyle Mwangi

Abstract

Amphibians are a good health indicator of environmental conditions due to their habitat requirements and physiological nature. They are moisture dependant ectotherms and require both aquatic and terrestrial habitats to exist. Factors that influence habitat selection are varied in species; amphibians utilize different environments during their lifetime. In this study we want to investigate whether there is a relationship between landscape heterogeneity (land cover diversity and altitude standard deviation) and the spatial variation of amphibian species richness and its relative influence in comparison to other primary determinants using regression and correlation analysis. Furthermore we also want to investigate whether the amphibian species richness varies across different land cover types using ANOVA. The study was carried out in Malaga province, Southern Spain. Land cover diversity and altitude standard deviation did not have a significant correlation to amphibian species richness variation. Climate variables such as evapotranspiration and temperature during the hottest month (july) are of more importance than landscape heterogeneity variables, with a significant correlation of 0.269 and -0.290 respectively at an R^2 of 0.214, in determining the variation of amphibian species richness. Other important determinants are slope (correlation = -0.260) and human population density (correlation = -0.10). The study highlights that the variation in species richness of amphibians is still primarily influenced by energy/water balance (measured as evapotranspiration) and energy (measured as temperature) variables which supersede landscape heterogeneity variables despite of fact that atmospheric energy is not a limitation in the Mediterranean region. The study also mentions that amphibian species richness varies at the different land cover types and illustrates that broadleaved forest cover has the highest species richness while building and infrastructure cover has the lowest species richness of amphibians. This may be due to their physiological nature and habitat requirements.

Keywords: Amphibian species richness, altitude standard deviation, landscape heterogeneity, land cover types, land cover diversity, primary determinants

Acknowledgements

My heartfelt thanks go to Nuffic for awarding me this scholarship to purse my eagerly awaited Master of Science in GIS, NRM course at ITC.

I would also like to send my thanks and gratitude to my supervisors, Dr. Thomas Groen and Dr. Bert Toxopeus, for mentoring me through my research work and appreciating how spatial information can be used as a tool to understand nature.

I would also like to thank my lectures in NRM department for teaching me various GIS and Remote sensing skills and techniques that are of importance in conserving and management of our God given natural resources. If it wasn't for you Eddie, creating land cover maps could still be a mystery.

To my fellow ITC students who have braved the tough times and enjoyed the good times, it has been a pleasure and good experience working with such a diverse group, from all parts of the world. I will definitely miss these times. Special regards goes to the NRM students that I have worked and partied hard with.

My heart full gratitude also extends to the ITC Christian fellowship who have facilitated my spiritual nourishment, Kenyan society that has made me feel at home, the run for fun team and off course the awesome football team that has kept me physically on toes, and not forgetting DJ Ronnie who has always kept us entertained with music from all corners of the world.

Out of the ITC realm, I like to gratefully thank my wife and kid for their love, support and understanding, my Dad and Mom and my sister for their great support and definitely my brother and his family for their accommodative support and my grandmother for blessing me during my stay at ITC.

Finally my out most thanks goes to Almighty God for providing me my strength and wisdom.

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1. Introduction

One of the fundamental purposes of ecology is to understand the factors that affect and regulate the geographic variation in species richness of organisms for effective conservation and management of wildlife species (Currie 1991; Dunning, Danielson et al. 1992; Hawkins, Field et al. 2003; Ricklefs, Qian et al. 2004; Tews, Brose et al. 2004; Qian, Wang et al. 2007).

The distribution in faunal species richness has been associated with various environmental variables including:

- Ambient energy inputs such as temperature and solar radiation facilitates species richness because ectotherms regulate their body temperature by directly absorbing heat from their environment, and the metabolic costs of homeotherms decrease with increasing ambient temperature (Turner J. R. G. 1987; Currie 1991).

- Precipitation and atmospheric humidity facilitates species richness of amphibians due their high dependency on moisture for survival and reproduction (Hawkins, Field et al. 2003; Rodríguez, Belmontes et al. 2005).

- Primary productivity: Animal diversity depends primarily on the conversion of energy from plant production. This energy is passed on to different trophic levels hence facilitating species diversity (Hutchinson 1959).

- Habitat heterogeneity: The higher the diversity of natural resources in an ecosystem the greater it's potential to accommodate more animal species (Tews, Brose et al. 2004).

- Climatic variability: It predicts that the less variable the climate the more stable the ecosystem and in turn support more species (Currie 1991).

The importance of environmental variables to species richness varies geographically depending on the environmental constraints of the region. In the tropics, sub tropics and warm temperate zones water variables are usually the strongest predictors while energy variables dominate in the higher latitudes were ambient energy is a constrain (Hawkins, Field et al. 2003).

The species richness of different animal groups maybe influenced differently, by various environmental factors. Ectotherms highly depend on the environment to regulate their body heat therefore are predominately influenced by water and energy input variables while endotherms distribution may be dictated by the structure of the habitat and/or ecosystem interactions (Hawkins, Field et al. 2003; Tews, Brose et al. 2004). For example Moreno-Rueda (2007; 2009) mentions that the geographic distribution of endothermic species such as mammals and birds are highly influenced by the heterogeneity of the habitat but on the other hand ectotherms such as amphibian and reptiles are significantly related to climatic variables of precipitation and temperature respectively.

The spatial resolution (grain size) and scale (extent of analysis) that one considers has a strong influence on the processes driving richness. At small grain size (few meters squared to tens of kilometre squared) and in local extents a wide range of biotic and abiotic factors are important whereas at large grain size and continental or global extents climatic factors dominate (Whittaker 2001; Willis 2002).

1.1. Landscape Heterogeneity

Most studies reviewed by Tews (2004) show a strong relationship between species richness and the structure of the habitats such as landscape heterogeneity – which refers to habitat diversity or resource richness of an ecosystem. This is also known as environmental or habitat heterogeneity (Atauri and de Lucio 2001; Tews, Brose et al. 2004; Rodríguez, Belmontes et al. 2005; Moreno-Rueda and Pizarro 2007) . The effect of landscape heterogeneity is one of the fundamental determinants of vertebrate species richness distribution (Tews, Brose et al. 2004). On a local scale it would provide a better explanation to species diversity than climate variables (Atauri and de Lucio 2001) which are better determinants on a continental or global scale. Climate (as such as temperature and precipitation) variables are more homogeneous at smaller scales and hence less important than landscape heterogeneity (Böhning-Gaese 1997)) in determining the variation in species richness. Furthermore at a regional and local scale, variation in landscape heterogeneity and its effects to species richness is more detectable using remote sensing technologies.

Landscape heterogeneity is the habitat diversity of the environment, which offers resources to species thus facilitating ecological niches for species (Pulliam 2000; Wiens and Donoghue 2004). In case of resource constrains ecological niches are limited and therefore dominant species tend to exclude other species sharing the same niche, thereby reducing the species richness (Pulliam 2000). The more resources are available within an ecosystem, the more species can coexist, each occupying their respective ecological niche thus facilitating species richness.

Elevation range and land cover diversity have been frequently used as measurable proxies to landscape heterogeneity (Atauri and de Lucio 2001; Moreno-Rueda and Pizarro 2007; Moreno-Rueda and Pizarro 2009) . The presumption for elevation range is that the greater the elevation variability of an area, the greater the spatial variability of its climate and therefore the more likely the area will have a larger number of habitats. For land cover diversity, it is assumed that the greater the land cover diversity the more natural resources are available to satisfy the needs of different species. Many studies have found a positive correlation between landscape heterogeneity and species richness (Atauri and de Lucio 2001; Tews, Brose et al. 2004; Moreno-Rueda and Pizarro 2007; Moreno-Rueda and Pizarro 2009).

The response to landscape heterogeneity by different groups of animals is not the same shows the importance of the ideal scale at which different species perceive their landscape (Keith 1997; Mazerolle and Villard 1999). The correlation between species diversity to landscape heterogeneity may vary depending on the type of species group and the spatial scale where landscape heterogeneity is measured. Scale dependence is caused by the species operation scale which is dictated by their home ranges and dispersal capacity. The operational scale is defined as the distinct spatial or keystone structure (as mentioned by Tews (2004)), that provides resources, shelter, goods and services for a particular species or group of species, *Figure 1-1*.

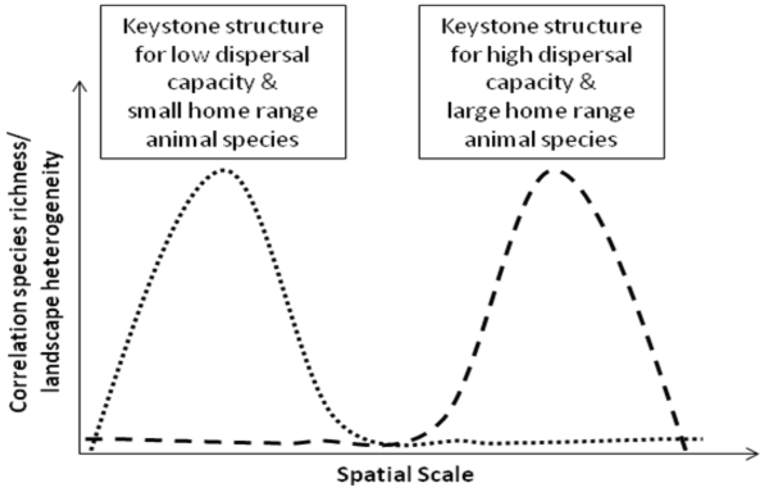

Figure 1-1 The operational scale also known as "keystone structure" varies between animal groups (Adapted from* Tews, Brose et al. 2004*)

The relationship between landscape heterogeneity to animal groups with high dispersal capacity such as birds and most mammals have been studied at a course spatial resolution, show a high positive correlation. For example, Moreno-Rueda (2007) carried out a study in Granada province, Spain at 100 km^2 resolution grids; found that the primary determinant of the distribution of mammals and birds species was landscape heterogeneity compared to other climatic variables and human population. On the contrary low dispersal capacity animal groups such as reptiles and amphibians were not determined by the landscape heterogeneity.

On the other hand landscape heterogeneity can influence species richness variation at a finer spatial resolution for species with lesser dispersion capacity such as reptiles and amphibians as suggested by Atauri (2001). Brose (2003) indicated that effects of habitat heterogeneity for ground beetles assemblages were positive at small grain size of 0.25 to 1000 m^2 while they were non-significant at a larger grain size of 10 $km^{2.}$

Habitat diversity is favoured by animals that use different ecosystems for breeding and foraging or during different stages of their life (Tews, Brose et al. 2004). A classical example are amphibian species which exhibit complex life cycles within a complex landscape matrix of aquatic and terrestrial habitats (Joly, Miaud et al. 2001).

1.2. Amphibians and their Environments

Amphibians (Class: Amphibia) such as frogs, toads (Order: Anura), salamanders and newts (Order: Caudata) are among the few terrestrial vertebrates that highly depend on environmental moisture. Their geographic range, ecologies, behaviours and life histories are strongly influenced by the distribution and abundance of water (Heyer W.R. 1994). They are cold-blooded (ectotherms) and mostly egg-laying animals (oviparous). They play an important role in the natural ecosystem as predators of invertebrates and even some vertebrates and as prey for small mammals, birds and snakes (Pearman 1997).

Amphibians are considered to be bio-indicators of the general health of the environment due to their ecological requirements and physiological nature. They have moist sensitive skin which plays an active role in water balance, respiration and protection from desiccation. They use both the aquatic and terrestrial component of their habitat (Collins and Storfer 2003; Rodríguez, Belmontes et al. 2005). Loss of either habitat component could result to their population decline. One third of global amphibian species have recently been classified as threatened, exceeding the numbers of birds and mammals (Stuart and Chanson 2004**.** Susceptibility to habitat alteration has made amphibians a target for conservation efforts (Browne, Paszkowski et al. 2009).

Several scientific studies done in different geographic regions have shown that the spatial distribution of amphibians are influenced by various environmental factors as shown in Figure 1-2 (Guerry and Hunter 2002; Rodríguez, Belmontes et al. 2005; Dayton and Fitzgerald 2006; Browne, Paszkowski et al. 2009; Moreno-Rueda and Pizarro 2009).

According to Denoël (2006), the terrestrial landscape significantly affected the abundance of palmate newts (*Triturus helveticus*), with the largest population found in forest areas in Southern Larzac limestone plateau, France (Figure 1-2 box a).

Altitude range and temperature positively correlates with amphibian species richness in Spain although the relationship diminishes at high altitude and temperature (Moreno-Rueda and Pizarro 2009) (Figure 1-2 box c and h).

Abundance of particular land cover types (broad leaved forest, pine forest and water reservoirs) increased the spatial distribution of amphibian species in Madrid region in Spain (Atauri and de Lucio 2001) (Figure 1-2 box b).

According to Browne (2009), in the Lake Utikuma region of Alberta, Canada, there is a positive relationship between amphibian abundance and pond dissolved oxygen, surrounding deciduous forest, mixed forest cover and urban cover while a negative relationship to pond depth, total dissolved solids, conductivity, aquatic plant diversity and surrounding low shrub and conifer cover (Figure 1-2 box b and e).

Elevation and slope is an important factor limiting the distribution of many amphibian species. The majority of desert amphibians in Big Ben National park, USA inhabit areas with elevations between 550 and 1000 meters above sea level and flat regions provide more opportunities for ponds to form than steeper slopes (Dayton and Fitzgerald 2006) (Figure 1-2 box a and c).

Actual evapo-transpiration (used as a measurable proxy of energy and water availability) has a strong positive correlation with amphibian species richness across Europe and China due to its reliance on water for survival and reproduction (Rodríguez, Belmontes et al. 2005; Qian, Wang et al. 2007) (Figure 1-2 box h).

A high positive correlation was also found in China and Europe between plant productivity, measured as EVI (Enhanced Vegetation Index) and NDVI (Normalised Difference Vegetation Index), and amphibian species richness, due to the shade provision from forest trees that protects amphibians from desiccation and abundance of prey (mainly insects) in under story vegetation (Rodríguez, Belmontes et al. 2005; Qian, Wang et al. 2007) (Figure 1-2 box d).

Precipitation shows a positive correlation with amphibian species richness both at a continental scale in China and local scale in Granada province, South Eastern Spain (Moreno-Rueda and Pizarro 2007; Qian, Wang et al. 2007) (Figure 1-2 box h).

Soil properties influence the distribution of many amphibian species (Bradford, Neale et al. 2003; Dayton, Jung et al. 2004; Dayton and Fitzgerald 2006). Coarse-rocky soil has low water capacity and drains quickly while soils with fine texture and high water capacity are more important because they facilitate surface moisture for water breeding amphibians and soil moisture for burrowing amphibians (Figure 1-2 box g) (Shoemaker 1988).

Human presence influences the amphibian species either positively or negatively depending on human distribution and activities. Urbanised and densely populated areas are a threat to amphibian population while moderately populated farmland areas have a positive correlation to amphibian species richness in Spain due to the presence of irrigation pools (Moreno-Rueda and Pizarro 2009) (Figure 1-2 box f).

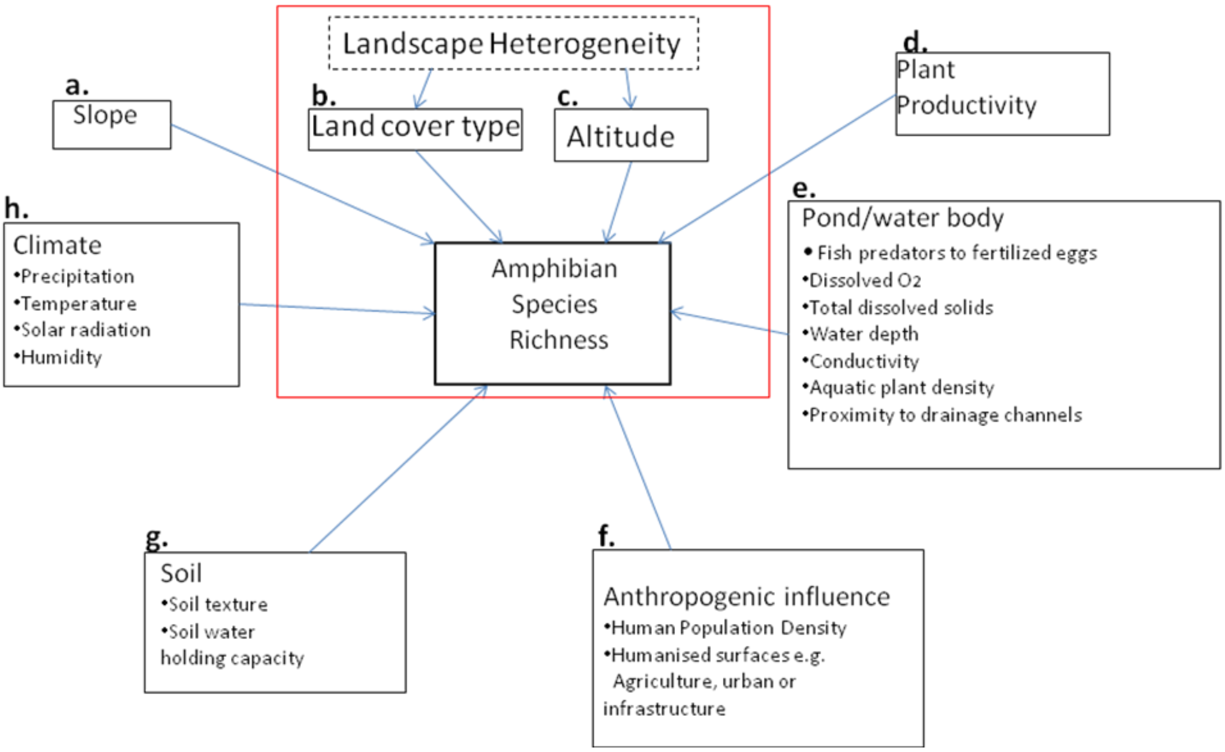

Figure 1-2 ***Conceptual diagram showing factors that determine the spatial distribution of amphibians based on various research studies. The red box indicates the focus of the research study.***

1.3. Research Problem and Justification

Climatic variables are mentioned to be primary determinants in the geographic variation of amphibian species richness across various geographic scales. On a continental scale in Europe actual evapotranspiration has a strong association with amphibian species richness (Rodríguez, Belmontes et al. 2005). On a national scale in Spain, temperature strongly correlates with amphibian species richness (Moreno-Rueda and Pizarro 2009). On a more local scale in Granada province, South East Spain, Amphibian species richness was primarily influenced by climate (temperature and precipitation) which explained up to 27% of its variance (Moreno-Rueda and Pizarro 2007). The main reason for these findings was due to the reliance of amphibian species to moisture for its survival and reproduction.

On the other hand it is still not clear whether landscape heterogeneity influences the variation of amphibian species richness in the Mediterranean region on a regional or local scale. In Madrid region, Spain, spatial variability of amphibian species richness positively correlated with the landscape heterogeneity (Atauri and de Lucio 2001). On the contrary at a similar geographic scale in Granada province, Spain landscape heterogeneity did not correlate with amphibian species richness (Moreno-Rueda and Pizarro 2007). (Atauri and de Lucio 2001), also mentions that variation of species richness for amphibians not only depends on the landscape heterogeneity but also on the land cover composition (land cover group types). He mentions that less amphibian species were found in xerophytic habitats such as scrubland and un-irrigated cropland while more species were found in mesophytic and hydrophytic habitats such as broadleaved forest, pine forest, and marshland.

Another important factor to consider is the grain size of the study, also known as the spatial resolution in other studies (Moreno-Rueda and Pizarro 2007). Most studies that investigated the relationship of amphibian species richness to landscape heterogeneity, at the Mediterranean region, were carried out on a course sample size of $100km^2$ and did not consider at finer grain sizes (Atauri and de Lucio 2001; Moreno-Rueda and Pizarro 2007; Moreno-Rueda and Pizarro 2009).

Atauri and de Lucio (2001) mentions that at a smaller grain size, landscape heterogeneity may relate better to species richness of animal groups with low dispersal capacity such as amphibians and reptiles. Various researchers in North America have investigated that most amphibian activities occur within 1 km of their breeding sites (Knutson, Sauer et al. 1999; Browne, Paszkowski et al. 2009). Therefore it can be assumed that the operational scale for amphibians in the Mediterranean region can also be $1km^2$.

In the light of aforementioned matters the focus of the study was geared to determining the relationship between landscape heterogeneity and species richness of amphibians at a spatial resolution of 1 km^2. The study used predictive distribution models, biodiversity indices, GIS and remotely-sensed data and tools to come up with values that can be used to find out their relationship through regression and correlation analysis. The study was carried out in Malaga Province, Southern Spain. On a regional scale, Southern Spain has been identified as one of the most important eco-regions in Europe. It is at the Mediterranean Basin and is considered a biodiversity hotspot with endemic faunal species that are under threat where amphibians top the list with 29 % of the species being threaten (IUCN 2008).

1.4. Objectives

The main objective of this study was to determine whether there is a relationship between landscape heterogeneity (altitude range and land cover diversity) and species richness of amphibians based on 1 km^2 resolution, landscape grids in Malaga province, Spain. This resulted in the following sub objectives:

1.4.1. Specific Objectives

- To determine whether the variation in species richness of amphibians relates to altitude range.
- To determine whether the variation in species richness of amphibians relates to land cover diversity.
- To determine whether species richness of amphibians is the same across all land cover types.
- To determine the relative significance of altitude range and land cover diversity in relation to the significance of other primary determinants that influence the variation of species richness of amphibians.

1.4.2. Research Questions

1. Does altitude range significantly correlate to the species richness of amphibians at 1 km^2 resolution?
2. Does land cover diversity significantly correlate to the species richness of amphibians at 1 km^2 resolution?
3. Is the species richness of amphibians the same across all land cover types in the study area?
4. Are altitude range and land cover diversity important determinants to the variation in species richness of amphibians at 1 km^2 resolution?

1.4.3. Research Hypothesis

1.4.3.1. Hypothesis 1

Ho: Altitude range does not significantly correlate to the species richness of amphibians at 1 km^2 resolution.

Ha: Altitude range does significantly correlate to the species richness of amphibians at 1 km^2 resolution.

1.4.3.2. Hypothesis 2

Ho: Land cover diversity does not significantly correlate to the species richness of amphibians at 1 km^2 resolution.

Ha: Land cover diversity does significantly correlate to the species richness of amphibians at 1 km^2 resolution.

1.4.3.3. Hypothesis 3

Ho: Amphibian species richness does not vary among land cover types.

Ha: Amphibian species richness does vary among land cover types.

1.4.3.4. Hypothesis 4

Ho: Altitude range is not an important determinant to the species richness of amphibians at 1 km^2 resolution.

Ha: Altitude range is an important determinant to the species richness of amphibians at 1 km^2 resolution.

1.4.3.5. Hypothesis 5

Ho: Land cover diversity is not an important determinant to the species richness of amphibians at 1 km^2 resolution.

Ha: Land cover diversity is an important determinant to the species richness of amphibians at 1 km^2 resolution.

2. Materials and Methodology

2.1. Study Area

The research study was carried out in the province of Malaga, Spain (Coordinates 36^0 43'North 4^0 25'West), as shown in Figure 2-1. It is located at the southern coast of Spain, in the region of Andalusia. It is bordered at the South by the Mediterranean Sea and by the provinces of Cadiz, Sevilla and Granada. Its area is 7,308 km2 with a human population of approximately 1.3 million persons. The climate is warm Mediterranean with dry, warm long summers and wet, mild short winters. The geographic relief varies greatly ranging from 0 to approximately 2000 meters above sea level (Wikipedia 2009a).

The typical vegetation of Malaga province is Mediterranean woodland, characterized by leafy xerophilic perennials, adapted to the long, dry summers, for example, Holly Oak (Quercus ilex), Cork Oak (Quercus suber), various pines and Spanish Fir (Abies pisapo). The dominant understory vegetation is composed of thorny and aromatic woody species, such as Rosemary (*Rosmarinus officinalis*), Thyme (*Thymus*), and Cistus. In the wettest areas with acidic soils, the abundant vegetation are the Oak Eucalyptus forest (Ibarra 2003).

The primary cultivation is dryland farming of cereals without artificial irrigation of barley, oats and wheat. The most important tree crops are olives. Others include fruits such as citrus and avocado mainly grown under irrigation (Naranjo 2003).

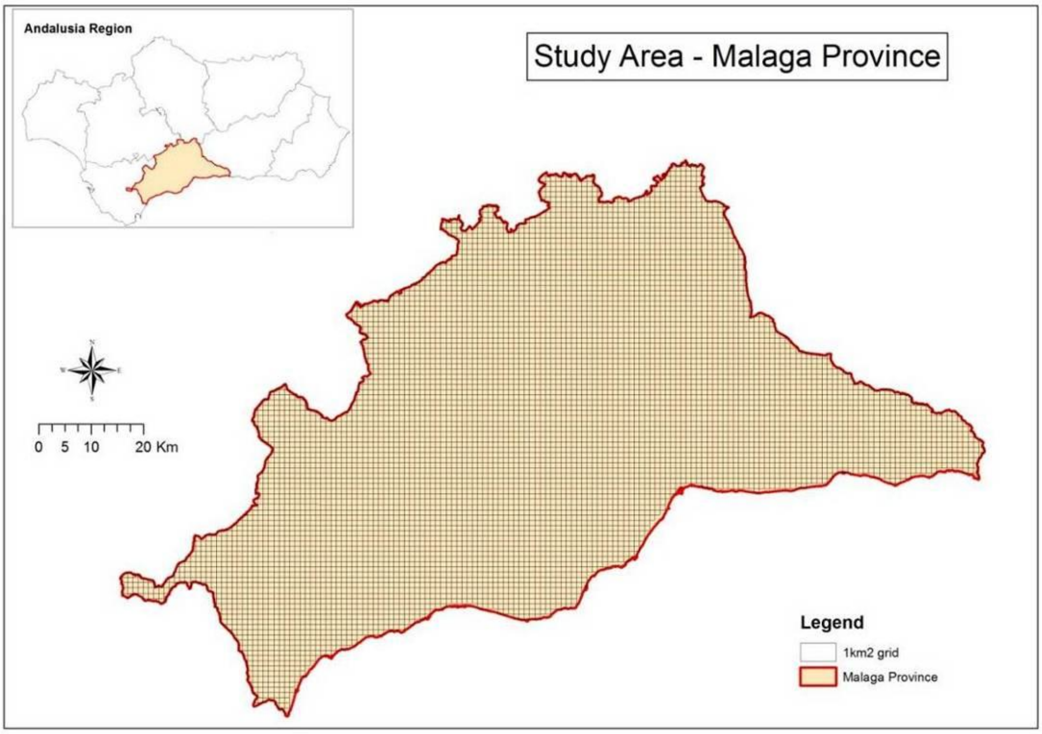

Figure 2-1 ***Study area***

2.2. Methodology

Overview

To be able to investigate and answer the four research questions required in the study the methodology was divided into 5 phases as shown in Figure 2-2.

1st Phase

Each of the eleven amphibian species data of the study area obtained from the 10 by 10 km grid herpetological atlas data were up scaled to 1 by 1 km resolution grid data using its $1km^2$ resolution, environmental data set determinants with a niche based model software called maxent (Phillips, Anderson et al. 2006). The $1km^2$ grid data for each species were then summed up in ArcGIS 9.3 (ESRI 2009) software to obtain an amphibian species richness data of the study area.

2nd Phase

Land cover data of the study area sourced from the Environmental ministry of Andalusia/Spain was used to derive the land cover types using ArcGIS 9.3 (ESRI 2009) software and land cover diversity per $1km^2$ sample unit using fragstats 3.3 software (McGarigal and Marks 1995).

3rd Phase

The digital elevation data sourced from ASTERDEM (METI 2009) was used to calculate the altitude standard deviation per $1km^2$ sample unit using ArcGIS 9.3 (ESRI 2009).

4th Phase

The relationship between land cover diversity and altitude standard deviation to amphibian species richness was analysed using simple linear regression in Spss 16 (IBMco. 2008).

The variation in amphibian species richness at different land cover types was analysed using ANOVA in Spss 16(IBMco. 2008).

5th Phase

The relationship between Altitude standard deviation, land cover diversity and other primary determinants derived from the maxent model to the amphibian species richness was analysed using stepwise multiple regression in Spss 16 (IBMco. 2008).

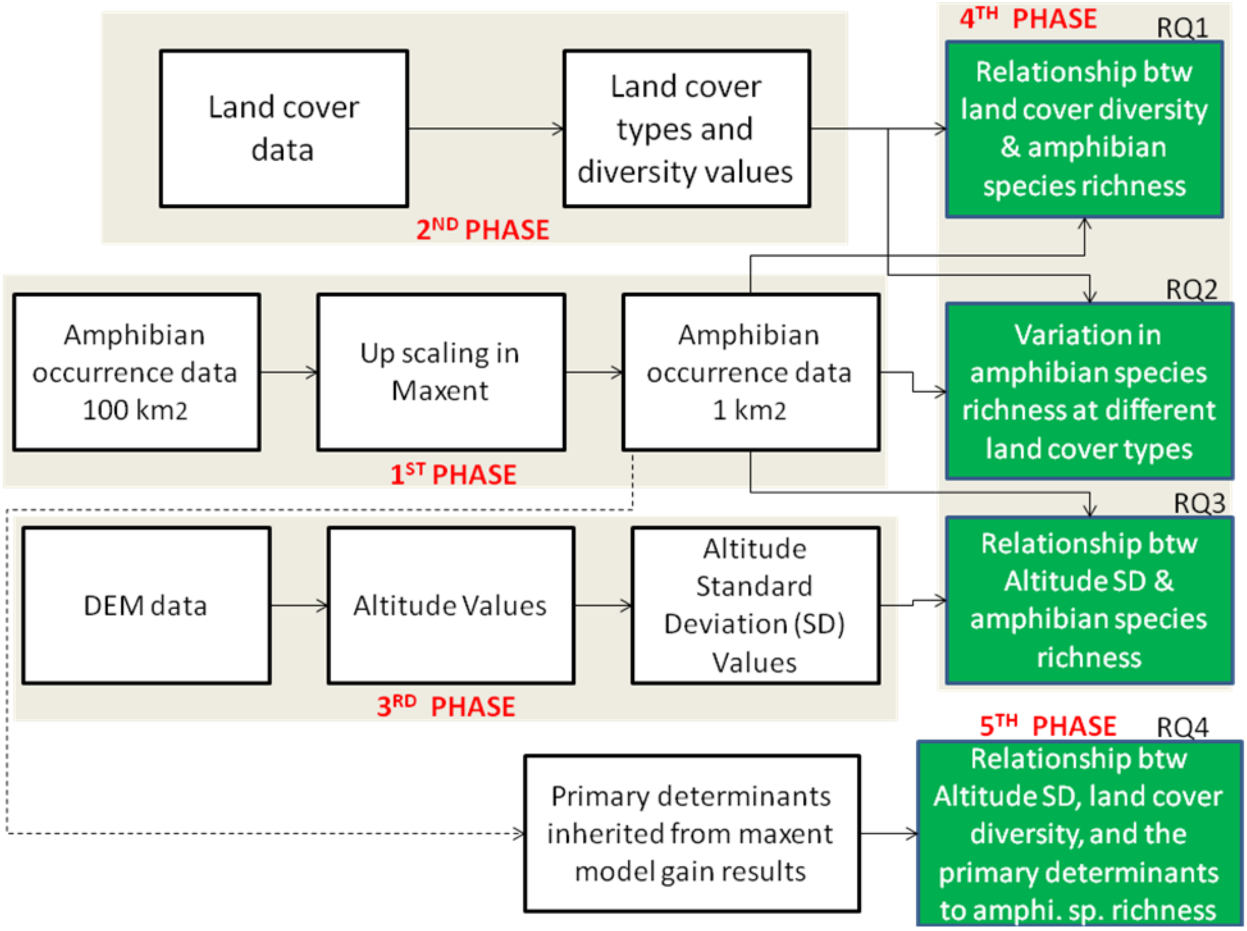

Figure 2-2 ***Methodological flow chart***

2.2.1. Phase 1: Up scaling to 1km2 grids using maxent modelling

Predictive distribution Modelling

The development of predictive habitat distribution models has rapidly increased in ecology, using powerful statistical techniques and GIS tools. These models are static and probabilistic in nature since they relate the geographical distribution of species to their present environment. Models have been developed to understand the bio geographical patterns of species in relation to their environmental needs (Guisan and Zimmermann 2000).

Plant species diversity can be directly mapped using various remote sensing and GIS techniques (Nagendra 2001) while the elusive and secretive nature of animals requires proxies to map their distributions (Leyequien, Verrelst et al. 2007).

The 100 km^2 herpetological atlas data from the Ministry of Environment of Spain was used to model 1 km^2 grids of probability of occurrence (P *occ*) of each amphibian species in the study area using a species distribution (niche based) modelling software called maxent (Phillips, Anderson et al. 2006).

2.2.1.1. Herpetological atlas data

Data on the presence of amphibian species at any location in the study area was derived from the herpetological atlas data of Spain. The data is in vector format and includes the absence (0) and presence (1) occurrence of herpetological species on $100km^2$ (10 km by 10 km) UTM grids, throughout Spain, sourced from ministry of environment of Spain (Pleguezuelos, Marquez et al. 2004). The occurrence data was collected from 1997 to 2001 and compiled by various collaborative groups e.g. University Museum of Florence (DRA), Department of animal biology of the University of Salamanca and other herpetological institutes (Pleguezuelos, Marquez et al. 2004). According to the atlas data eleven amphibian species occurred in Malaga province, which were used to derive the species richness of the study area. A brief explanation of these eleven amphibian species used in the research study, are shown in Table 2-1 and depicted in Figure 2-3.

Name	Habitat	IUCN threat status
Alytes dickhilleni (Betic midwife toad)	Found in pine and holm oak forests and open land with little or no vegetation. Altitude ranges from 340 to 2100 meters and can be found in very steep slopes. Adults are near clean water bodies in rock crevices or under rocks.	Vunerable (VU)
Bufo bufo (Common Toad)	Occupies all type of habitats, from deciduous forests to conifers areas, bushes, riparian vegetation, cultivated areas or gardens and parks. Prefers coniferous forest with marshes, fairly wet sites with dense vegetation.	Least concerned (LC)
Bufo Calamita (Natterjack Toad)	Found in many habitats, from coastal, arid, and humid to mountainous areas above 2500 meters. Also thrive in altered areas such as crops and gravel pits. Preferably breed in freshwater bodies but supports brackish conditions.	Least concerned (LC)
Discoglossus jeanneae (Spanish painted frog)	Usually found in open spaces such as pastures, meadows and crops and breed in ponds, small streams and also in ditches, water holes, springs and fountains.	Near Threatened (NT)
Hyla meridionallis (Mediterranean tree frog)	Adults occupy meadows and dense bush land near ponds and rivers. Prefer places with good vegetation coverage, with seasonal or permanent ponds, flooded meadows also inhabit small dams and reservoirs.	Least concerned (LC)
Pelobates cultripes (Western spade foot)	Lives in open landscapes, such as dunes, cultivated fields and meadows. They are especially associated with sandy soil. Reproduction occurs in stagnant water bodies. Commonly found in altitudes below 1000m	Near Threatened (NT)
Pelodytes ibericus (Iberian Parsley frog)	Altitudinal distribution ranges from lowlands, including areas close to the sea, to the mountainous medium altitude (900 m). Prefers open exposed areas. Found in a variety of aquatic media, mainly in ponds, shallow lagoon and streams	Least Concerned (LC)
Pleurodales waltl (Iberian ribbed newt)	Found in Mediterranean forest, crops and wetlands, in deep (1m and above) natural and artificial water bodies. Mainly aquatic. Common at altitudes from sea level to 1000 m.	Near Threatened (NT)
Rana perezi (Iberian green frog)	Present in a wide range of environmental conditions, can be found from sea level to elevations to 2000 m, and occupies all type of biotopes in the Mediterranean with permanent or temporary water sources, even with intense human activities. Found also in saline and polluted stagnant water	Least concerned (LC)
Salamandra salamandra (Fire salamander)	Found in medium to high mountains up to 2500, inhabits mainly deciduous and mixed, sometimes conifer forests. Prefers wet areas with peat bog or meadows and with abundant rainfall.	Least concerned (LC)
Triturus pygmaeus (Pygmy newt)	Often found in Cork and holm Oak forest, breed in temporary ponds, fountains, canals and creek with little current. Also found in abundant quarries and mines. Present in coastal areas up to 1450m.	Near Threatened (NT)

Table 2-1 The *Amphibian name, their habitat and threat status according to IUCN red list of threatened species 2009. (Pleguezuelos, Marquez et al. 2004; Bosch J. and 2009; IUCN 2009).*

Alytes dickhellini
(Betic midwife toad)

Bufo Calamita
(Natterjack toad)

Bufo bufo
(Common toad)

Discoglossus jeanneae
(Spanish painted frog)

Pelobates cultripes
(Western spade foot)

Pelodytes ibericus
(Iberian parsley frog)

Hyla Merridionallis
(Mediterranean tree frog)

Rana perezi
(Iberian green frog)

Perodales waltl
(Iberian ribbed newt)

Salamandra salamandra
(Fire salamander)

Triturus pygmeaus
(Pygmy newt)

Figure 2-3 ***Photographs of the amphibian species of the study area (Pleguezuelos, Marquez et al. 2004)***

2.2.1.2. Species distribution (Niche based) modelling

Niche based models represent an approximation of species ecological niche given various environmental conditions. One of the current niche based model tools is maxent, others include GARP (Genetic Algorithm for Rule-Set Prediction), GLM (Generalised Linear Model) and GAM (Generalised additive model) (Phillips, Anderson et al. 2006).

Maxtent is based on the principle of maximum enthropy (Phillips 2006). Entropy according to Shannon (1948) is “a measure of how much choice is involved in the selection of an event”. This means that species distribution with higher entropy has more choices thus less constrained (Phillips, Anderson et al. 2006). Therefore the higher the resilience of a species the more it is adaptable to extreme environmental conditions hence the greater its spatial prevalence.

Maxent uses spatial presence data of a species and its pertaining environmental data variables as input for making predictions of occurrences in the form of a continuous surface. The presence data of all amphibians in Andalusia region were acquired from the 10 by 10 km atlas grid data in conjunction with the 1 km^2 environmental data variables that influenced the geographic distribution of amphibians were used to obtain 1 km^2 continuous surface of the probability of occurrence of each amphibian species. This was carried out through the following steps:

1. **Environmental Variables**

There are a wide variety of environmental variables that influence the distribution of amphibians. The preselected variables that were used for modelling the distribution of amphibian species were based on literature review. These were:

Temperature

It is widely used as a measure of ambient energy input required for organisms (Currie 1991; Rodríguez, Belmontes et al. 2005). Minimum temperature of the year indicates the frost or freezing tolerance, while maximum temperature of the year indicates the heating tolerance of amphibians (Qian, Wang et al. 2007). Arunans face high desiccation due to their permeable skin and therefore are mostly nocturnal when temperatures are lower (Adler 2004). The variables selected for temperature were, the lowest temperature of the coldest month (January), the highest temperature in the hottest month (July) in temperate and Mediterranean regions and the Temperature variation (Maximum minus Minimum annual temperature).

Precipitation

Atmospheric water is one of the essentials for any living organisms. Amphibians are one of the few vertebrates that highly dependent on moisture and part of their life cycle is usually aquatic (Heyer W.R. 1994). The variables used were, precipitation in spring, summer, autumn and winter. In Andalusia, Southern Spain the summer is usually dry and the onset of rain starts in autumn with a wet winter sweeping from the Atlantic Ocean (AEMET 2008).

Evapotranspiration

Evapotranspiration combines temperature and water availability and in turn measures the energy water balance (Bini, Diniz et al. 2004). Actual evapotranspiration (AET) is one of the primary predictors in determining amphibian species richness in Europe; it is of special importance to amphibians which require moisture for its survival and reproduction (Rodríguez, Belmontes et al. 2005). AET in winter, summer, autumn and spring were the variables used in this study.

Primary Productivity

Plant productivity influences the geographic variation of species richness for many plants and animals at a wide range of scales (Mittelbach, Steiner et al. 2001)(Mittelbach , 2001). NDVI (Normalised difference vegetation index), EVI (Enhanced Vegetation Index) and GVI (Global Vegetation Index) are variables that estimate plant primary production that represents realised energy captured. Plant productivity indices have been used as measures to determine the geographic distribution herpetological species in Europe (Rodríguez, Belmontes et al. 2005). The variables selected were classified NDVI (unsupervised classified temporal NDVI profiles), NDVI values in spring, summer, winter and autumn.

Terrain (Elevation, Slope, West and South exposure)

Topographic elevation influences the climatic variation and vegetation composition and structure of an ecosystem; this in turn influences the distribution of faunal species. The topographical slope of landscape influences the local drainage system. Flat areas tend to accumulate water in form of wetlands or ponds especially during rainy seasons which favour amphibians breeding (Dayton and Fitzgerald 2006).

The aspect of a slope affects the local climate. The sun rays are in the west at the hottest time of the day, therefore west side of a slope tend to be warmer than the shadowed east facing side of the slope. Similarly in the Northern hemisphere the southern facing slopes are more open to sunlight thus warmer and drier than the northern facing slopes (Bennie 2006).

The moist Westerly current sweeps into the Southern part of Spain (Andalusia) from the Atlantic Ocean during autumn and winter. As a result the wind ward (West sloping) side of mountains are wet as opposed to dry lee ward, east sloping side (AEMET 2008).

The variables used to determine terrain were West exposure, South exposure, Altitude and Slope.

Soil type

Soil properties influence the distribution of many amphibian species (Bradford, Neale et al. 2003; Dayton, Jung et al. 2004). Different soil types have different water holding capacity. Soils with relatively high available water capacity tend to be more important for pond breeding amphibians and/or burrowing amphibians because they provide an aquatic media for their eggs and tadpoles and a moist refuge site to avoid desiccation respectively. Some frogs such as *Pelobates cultripes* (Western spade frog) and *Alytes obstetricians* (Common midwife toad) are especially associated with sandy soil as opposed to most frogs which prefer water holding capacity soils that retain surface water such as clay or silt (Pleguezuelos, Marquez et al. 2004).

Water availability

Amphibians are one of the few terrestrial vertebrates that are highly dependent on surface water especially for breeding (Heyer W.R. 1994). The variable used to determine surface water availability were, distance to large water bodies and rivers dams, weir, springs and small rivers/streams.

Human Population

Humans tend to establish themselves in high productive zones were species richness is high (Gaston and Evans 2004; Luck 2007). Most amphibians are threatened by human population and activities (Real, Barbosa et al. 2003; Lee, Ding et al. 2004). The determining variable used was human population density which has a negative impact on the amphibian population.

Humidity index

The humidity index is a combination of air temperature and relative humidity and is based on the ratio of precipitations and potential evapotranspiration (UNEP/GRID 1994) . Most arunans are active at night when temperatures are lower and the atmospheric humidity is high (Halliday et al, 2004).

Solar Radiation

Solar radiation is a measure of the heat intensity in kWh m-2 day-1 from the sun. It is energy that drives various natural systems such as climate, ecosystem and hydrological systems. Arntzen (2008) used mean annual solar radiation as one of the important explanatory variables to determine the species geographic range of two species of marble newt (*Triturus marmoratus* and *Triturus pygmeaus*). The variables preselected to determine radiation were, summer, spring, winter and autum radiation.

Landcover

Land cover type divides the biosphere layer into discriminatory classes that describe the nature of an ecosystem. It is used in many ecological studies (Atauri and de Lucio 2001; Rodríguez, Belmontes et al. 2005; Moreno-Rueda and Pizarro 2007; Moreno-Rueda and Pizarro 2009) as a fundamental explanatory variable in determining the spatial variation of faunal species. According to Pleguezuelos (2004) most amphibians are found in a wide range of land cover types i.e. Oak forests, Coniferous forest , bush land to grassland. Agricultural surfaces with moderate amount of farm lands tend to favour amphibians due to the creation of irrigation pools (Diaz-Paniagua 2001).

The behaviour and geographic distribution of amphibians in temperate (including Mediterranean) regions are influenced by the climatic conditions across the different seasons (winter, summer, autum and spring). For example Heyer (1994)and Adler (2004) mention that amphibians breeding season in temperate regions are during spring, when there it is warmer and more surface water availability from melting ice, but hibernate during cold winters under the soil frost line or plant litter. In the light of this, it was more insightful to use the possible environmental variables per season. The preselected environmental variables used for modelling are as shown in Table 2-2.

Category	Variable	Data Format	Source
Climate	Seasonal mean precipitation in millimetres Temperature in coldest month (January) and hottest month (July) and temperature varability in degrees centigrade	Raster	Worldclim (Graham 2006)
	Seasonal mean radiation in kWh/m^2/season Seasonal mean evapo-transpiration in millimetres per season (3 months)	Raster	Local Database (ITC-NRM Department)
	Humidity Index (preticipation/PET)	Point Data	UNEP/GRID (UNEP/GRID 1994)
Land cover	Reconstructed Corine landcover data, 1999	Vector	Ministry of Environment Andalusia/Spain
Primary (plant) productivity	Seasonal NDVI Classified NDVI (Unsupervised classified NDVI temporal)	Raster	Local Database (ITC-NRM)
Topography	Altitude/Elevation in meters Slope in percentage (Developed using "Slope tool" Spatial analyst tool box ArcGIS 9.3) Aspect (West and South exposure in degrees i.e. a Minimum of 0^0 and a maximum of 180^0 .The higher the degrees the closer the aspect to the west or south direction respectively)	Raster	ASTER GDEM (METI 2009)
Soil	Soil type	Raster	FAO (FAO 2003)
Human Population	Population Density of people/km2	Raster	Local Database (ITC-NRM Department)
Proximity to water	Distance to Spring, wells, dams in meters (Developed using "Euclidean distance" Spatial analyst tool box ArcGIS 9.3) Distance to large water bodies and rivers in meters (Developed using "Euclidean distance" in Distance, Spatial analyst tool box ArcGIS 9.3)	Raster	Local Database (ITC-NRM Department)

Table 2-2 ***Summary of the environmental data variables used in maxent modelling***

2. Multicollinearity

Multi-collinearity is the correlation between independent variables i.e. the continuous environmental variables. It is of little consequence if it is not large, but large multi-collinearity (VIF ≥ 10) may result in highly unstable performance of the Least Square Estimator, which will affect the distribution modelling performance (Phillips, Anderson et al. 2006).

The Variance Inflation factor (VIF) measures the strength of the relationship between the independent variables (covariates). The VIF is calculated as follows:

$$\mathbf{VIF = 1/\,(1\text{-}R^2\,)}$$

Where: $\mathbf{R^2}$ is the regression coefficient. If $\mathbf{R^2}$ increases **VIF** also increases, a large **VIF** indicates that the covariates are highly correlated.

Multicollinearity was performed among the continuous environmental variables in SPSS 16 and those with a VIF below 10 were selected:

3. Modelling Extent

The modelling spatial extent was done on Andalusia region instead of the study area -Malaga province and then clipped to the study area extent. This is because of the prevalence of data for rare species such as *Alytes dickhellini, Pelobate cultripes and Triturus pygameus*, are much higher on an Andalusia extent as compared to the study area extent. This is an important factor when acquiring training data to develop a robust model (Pearson and Dawson 2003; Peterson and Holt 2003). For example *Alyte dickhellini* has only 4 presence data at the study area extent while on an Andalusia extent it has 109 presence data.

4. Modelling preparation

Raster to ASCII file format

The environmental variables used for maxent modelling were clipped to the extent of Andalusia region then converted to ascii format in ArcGIS 9.3 “raster to ascii conversion tool”. Maxent only accepts environmental variable layers in ascii format with the same projection, bounding extent and cell size.

Presence data for Amphibian species

The occurrence atlas grid data of Spain was short listed to only the 11 amphibians found in Malaga province and clipped to the extent of Andalusia region in ArcGIS 9.3. The atlas grid data was then converted to points using the “convert feature to point tool” in ArcGIS 9.3 and its XY coordinates added.

The presences points for each species were filtered and combined in MS excel and converted to the species name in the first column, the second and third column were reserved for the XY UTM coordinates. The spread sheet was converted to a comma separated values (csv) format – Maxent can only work with the presence point of species with its XY location points as csv format.

5. Modelling process

Maxent is a general-purpose method for making predictions from incomplete information (Phillips, 2006). The species model is determined from a set of environmental layer variables in the of grid cells of the study area, together with a set of sample locations where the species has been observed. The model expresses the probability of occurrence (if logistic output is used) of each grid cell as a function of the environmental variables at that grid cell. The computed model is a probability distribution of species occurrence over all the grid cells.

The presence data (csv format) of all the 11 amphibian species and its pre-processed environmental variables data (ascii format)- 3 categorical (Soil, Land cover types and classified NDVI data) and 22 continuous data layers were introduced into maxent GUI. The presence data was split into test data, 30% and training data, 70% as recommended by (Phillips, Anderson et al. 2006). All the other parameters were left at default i.e. the maximum number of iterations, 500; maximum number of background points 10000; regularisation multiplier, 1; and convergence threshold, 0.00001.

When the model is executed a progress window appears, indicating the amount of gain acquired by each species from the environmental variables.

The gain is closely related to deviance, a measure of goodness of fit used in generalized additive and generalized linear models. It starts at 0 and increases towards an asymptote during the run. During this process, maxent is generating a probability distribution over pixels in the grid, starting from the uniform distribution and repeatedly improving the fit to the data. The gain is defined as the average log probability of the presence samples, minus a constant that makes the uniform distribution have zero gain. At the end of the run, the gain indicates how closely the model is concentrated around the presence samples (Phillips, Anderson et al. 2006); for example if the final gain is 1.5 the average likelihood of the presence samples is exp(1.5) = 4.5 times better than the random background pixel.

The output data is a continuous raster surface (ascii format) of the probability of occurrence of each species at a 1 km^2 grid resolution.

2.2.1.3. Model validation

Validation is an important process that assesses the performance and quality of the species distribution model.

The evaluation techniques that used to validate the predictive power of the 1km^2 probability of occurrence modelled raster layers of each species were; one threshold independent and two threshold dependent techniques.

1. Threshold independent evaluation

A common approach in evaluating the performance of a model is using receiver operating characteristics (ROC) curves (Zweig and Campbell 1993; Fielding and Bell 1997; Phillips, Anderson et al. 2006). It measures the improvement of a model from the random performance, through the analysis of the area under the ROC curve (AUC). ROC plot is obtained by plotting all sensitivity values (true positive values) on the y axis against the equivalent 1-specificity values (false positive values) of all available threshold in the x-axis (Fielding and Bell 1997; Phillips, Anderson et al. 2006).

The AUC gauges "the probability that a random positive instance and a random negative instance are correctly ordered by a classifier"(Phillips, Anderson et al. 2006). AUC values are typically from 0.5 to 1.0 (perfect model). At 0.5, the model performs the same as a random model meaning there is a 50 % chance of obtaining a true positive value. For example an AUC of 0.75 means that if a random selection is done, 75% of the time such a selection from the positive group will be encountered than a random selection from the negative group (Deleo 1993).

The AUC from the test data for each species was obtained from the ROC/AUC graph in the maxent results.

It is important to note that due to the fact that Maxent uses only presence data the ROC/AUC analysis is done using pseudo-absence (background pixels) in place of true absence (Phillips, Anderson et al. 2006). Pseudo-absence means false absence values which tends to be overestimated by presence only data (Anderson, Lew et al. 2003).

Secondly the maximum achievable AUC with presence data is less than 1. If the presence distribution of a species covers a fraction ***A*** of the total pixels, the maximum achievable AUC will be ***(1-A)/2***. This means that AUC values for rare species tend to be higher than that of common species, which is an artefact of the AUC statistic(Phillips, Anderson et al. 2006).

To counter the aforementioned artefacts a kappa and an omission error rate threshold dependent evaluation were carried out.

2. Threshold dependent evaluation

Threshold dependent measures divide modelled data into classes that can be used to evaluate a model with the observed/referenced data using a contingency matrix. The main problem of dividing continuous data into classifier data is the loss of information when grouping the data values into classes (Fielding and Bell 1997).

Most habitat associated studies have used accuracy techniques such as overall percentage accuracy (Capen 1986; Verbyla and Litvaitis 1989; Donazar, Hiraldo et al. 1993) and Kappa analysis (Fielding and Bell 1997; Couto 2003) which are based on the contingency matrix.

Kappa (Cohen 1960) is based on the difference between the actual agreement (the difference between the modelled classified data and the reference data, indicated by the major diagonal) and the chance agreement that is indicated by the total rows and columns in a contingency matrix. Kappa is a more holistic approach than percentage overall accuracy which only measures the actual agreement of the model based on the major diagonal of the contingency table. Kappa analysis requires both presence and absence data of the species in case it is used to evaluate species distribution models (Fielding and Bell 1997; Anderson, Lew et al. 2003)

For the case study the binary threshold for absence (0) and presence (1) data of the model for each species were obtained at "equal sensitivity and specificity, test, of the model", as proposed by (Phillips, Anderson et al. 2006) - where the number of true presence is equals to the number of true absence according to the test data, through the following steps:

- All the ascii data of probability of occurrence for each species were converted to raster grid format data in ArcGIS 9.3 using the "Convert from ascii to raster tool".
- The raster grid data were then reclassified in ArcGIS 9.3 using the logistic threshold value mentioned above to obtain binary (absence (0)/presence (1)) data of each species.

The accuracy of the binary modelled data was done using the Kappa analysis test in Spss 16.0 with reference data from the centroid points of the 100km^2 atlas grids occurrence (absence/presence) data as follows:

- The centroid points were linked to the corresponding modelled value using the "extract raster values to points" tool in ArcGIS 9.3.
- The values of the centroid points were cross tabulated with the modelled values in Spss 16.0 and the Kappa value for each species derived.

The Kappa analysis uses the pseudo absence from the model results and also the same data used to train the species distribution models (self validation). To counter this problem another threshold dependent evaluation was carried out (omission error rate) based on independent presence data source from the University Of Malaga, Spain (Prof. Rimundo unpublished data). The presence 1km^2 grid data was only available at the study area (Malaga province) extent and therefore the omission error rate analysis was only done at this extent using ArcGIS 9.3.

2.2.1.4. 1 km^2 Species Richness raster Layer

Species richness is the number of species in a defined area (McGarigal and Marks 1995). Species richness index has been used as an indicator in various ecological studies to investigate the relationship between species richness and other bio-climatic or anthropogenic factors (Atauri and de Lucio 2001; Rodríguez, Belmontes et al. 2005; Moreno-Rueda and Pizarro 2007; Qian, Wang et al. 2007; Moreno-Rueda and Pizarro 2009). To obtain the amphibian species richness layer, the summation of all amphibian species was done per 1km2 grid in ArcGIS 9.3, based on the absence and presence of each species, through the following steps:

- The maxent output raster data in Ascii format for each species probability of occurrence were converted to raster grid format floating data using the “convert Ascii to Raster tool”.
- The absence (0) and presence (1) binary values for each species were reclassified using the “reclass tool” in ArcGIS spatial analyst, based on the “equal sensitivity and specificity, test” binary threshold values derived for the maxent results.
- All the species raster layers were summed using the raster calculator in spatial analyst, to obtain a species rich raster layer of the Andalusia extent.
- The Malaga province administrative boundary shapefile (sourced for the Nrm – bio fragmentation database) was used to clip out the species rich raster layer of the study area using the ‘’extraction by mask tool” in ArcGIS.
- To avoid edge area effect error, caused by the study area administrative boundary, only pixels that are completely within the study area were extracted.

2.2.2. Landscape sampling grid layer

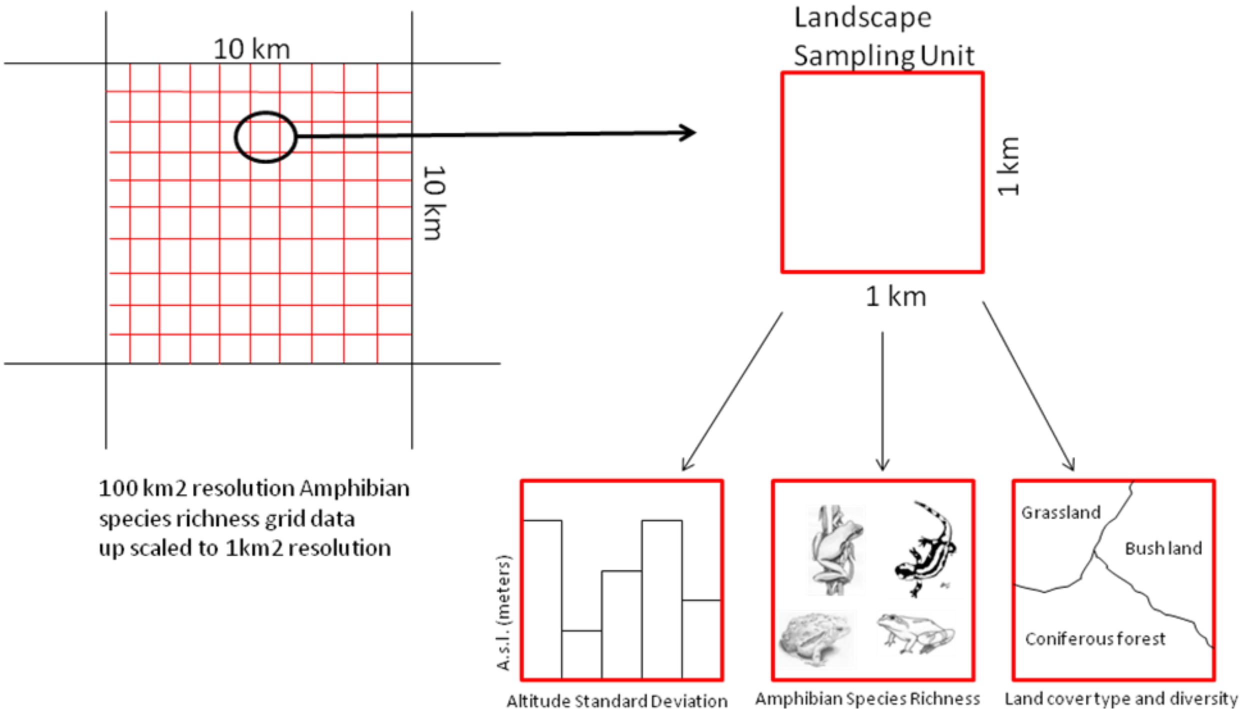

Figure 2-4 ***Landscape sampling grid layer from the up scaled amphibian species richness raster layer***

To be able to derive the altitude range (calculated as standard deviation) values and land cover type and diversity values based on 1km2 sampling units of the study area, as shown in Figure 2-4, the amphibian species richness/diversity raster layer had to be converted to a landscape vector grid layer as follows (and illustrated in Figure 2-5):

- The 1 km^2 species richness raster layer grid (from section 2.2.1.4) was converted to centroid points using the “convert raster to point tool” in ArcGIS 9.3 and the species richness values preserved in the attribute table.
- The centroid points were then converted to 1 km^2 vector grid mesh using the “create to plot tool” in Hawths tool, ArcGIS 9.3. The options specified were 500 meters radius and a square plot type.

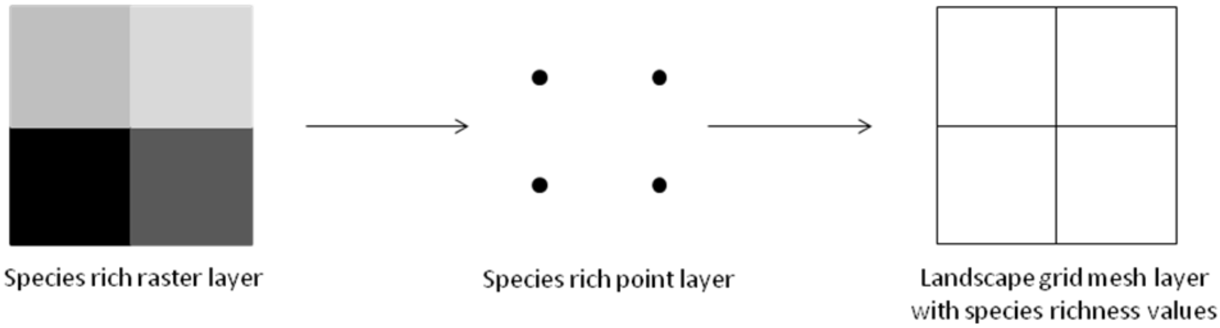

Figure 2-5 ***Amphibian species richness raster converted to a vector Landscape sampling grid layer***

2.2.3. Phase 2: Land cover Data

The spatial land cover data was an important component for the study to derive the land cover type and land cover diversity, as shown in Figure 2-4.

The land cover data (reconstructed land cover corine data) used in the study was sourced from the Ministry of Environment of Andalusia/Spain. It is a 50 meter resolution (i.e. the smallest width of a geographic feature is 50 meter) land cover dataset derived from landsat TM 1999 merged with IRS-PAN imagery in the summer of 1998 and 1: 50,000 topographic maps published by the geographical service of the army and verified using aerial photographs scale 1/20,000-1/30,000.

The land cover data was developed by the Ministry of Environment in collaboration with Environmental agencies of Andalusia, for the intension of predicting the land cover / use change in Andalusia from 1991, 1995 to 1999. The methodology used for classification was similar to that used to develop the Corine land cover data (EEA 2005). Some of the distinct differences are as follows:

- The increase in scale from 1:100,000 to 1:50,000.
- The use of aerial photos scales 1/20,000 to 1/30,000 as an essential document of support.

2.2.3.1. Land cover consolidation

The land cover data was consolidated into a 16 land cover classes, as shown in the Table 2-1 and Appendix 1. The land cover reclassification was based from other amphibian literature studies that utilized land cover data (Atauri and de Lucio 2001; Pleguezuelos, Marquez et al. 2004; Moreno-Rueda and Pizarro 2007; Browne, Paszkowski et al. 2009; Moreno-Rueda and Pizarro 2009).

Category	Land cover Type	Original land cover type
Natural vegetation	Bareland	Burnt area, dunes & beaches, bare rock, land with little vegetation.
	Grassland	Pasture, grassland, leisure places with grass
	Bushland	Open and closed bushland
	Eucalyptus Forest	Open and closed (dense) Eucalyptus forest
	Broad leaved forest	Open(with grass/bushes) and closed (dense) Broadleaved forest
	Coniferous forest	Open (with grass/bushes) and closed (dense) Coniferous forest
	Mixed forest	Mixture of the above 3 or either 2 forest types
	Riverine vegetation	Riparian vegetation, rivers and streams
Water Bodies	Sea	Sea, Estuaries, Coastal lagoons
	Wetlands	Inland marshes, salt marshes, peat bogs
	Lakes	Dams, natural lakes and other large water bodies
Agriculture	Olive grooves	
	Non irrigated crops	Mainly Wheat and also hay
	Irrigated crops	Vineyards, horticulture crops, green house crops, rice field
Others	Buildings or Infrastructure	Residential, Urban, Industrial, transport buildings such as sea and airport, Roads, Highways
	Extractive sites or Dumpsites	Mines, Quarry, Dumpsites, other extractive activities

Table 2-3 ***Reclassification of Land cover types used in the research study***

2.2.3.2. Land cover diversity indices

Biodiversity is a quantitative term used to describe the diversity of living organisms. Diversity measures, in form of indices, have been used extensively in a variety of ecological applications to quantify animal and plant diversity (McGarigal and Marks 1995) .

The indices commonly utilized in land cover diversity measurement combine two separate aspects of diversity: richness (the number of land cover types) and evenness (distribution of area among different land cover types). The more the land cover types the higher the diversity in terms of richness. The more equitable the distribution, the higher the diversity in terms of evenness (Nagendra 2002).

Studies use land cover diversity measured as a count of land cover types for discrete variables (structural richness) or as the extent for continuous variables (structural diversity). Structural diversity will yield better insight in the land cover diversity and species richness/diversity relationship if the species need subareas of different structural groups (i.e. land cover types). For example amphibians and birds require different habitats for breeding and foraging in addition amphibians require different habitats at different stages of life (Tews, Brose et al. 2004).

The most popular applied indices in community ecology to measure plant and animal diversity are Shannon and Simpson's indices (Forman 1995; McGarigal and Marks 1995; HainesYoung and Chopping 1996) .

1. **Shannon diversity indices (SHDI)**

Shannon and Wiener independently derived the function which has become known as Shannon index of diversity (Magurran 1988) . It is defined as:

$$SHDI = -\sum_{i=1}^{N} pi \times \ln pi$$

Where: **N** - is the number of land cover types
pi - is the proportional abundance of the ith type of land cover

In theory this index ranges from 0 to infinity, but it is usually found to fall between 1.5 and 3.5 (Magurran 1988) . It increases with increase in diversity and versa visa. Shannon diversity index is more sensitive to richness than evenness, thus rare land cover types have a disproportionately large influence on the index. The absolute magnitude of SHDI is not particularly meaningful therefore it is used as a relative index for comparing different landscapes (McGarigal and Marks 1995).

2. **Simpson diversity indices (SIDI)**

Simpson diversity indices is relatively less sensitive to richness and therefore place more weight on common land cover types and is less sensitive to rare types of land cover. It has an interpretation that is much more intuitive than Shannon diversity index. The value of Simpsons indices represents the probability that two randomly selected patches are of two different land cover types (McGarigal and Marks 1995). It is defined as:

$$SIDI = 1 - \sum_{i=1}^{N} pi^2$$

It ranges from 0 to 1 and increases with increase in diversity.

For the research study the Shannon diversity indices was preferred because it lays more emphasis on the small and rare land cover types which may be of importance to amphibians than the larger and common land cover types. A wetland ecosystem such as ponds are usually a small patch size relative to other land cover patches in the study grain size of $1km^2$, but it highly influences the variation of amphibian species richness due to their reliance on surface moisture. For example a landscape grid of the study area may contain a pond amongst other usually larger land cover types, SHDI will elaborate more on the richness of the landscape and lay more emphasis on the pond ecosystem which is of greater importance to amphibians. On the other hand SIDI, which is less sensitive to rarity/smallness, will lay less importance in small patches such as pond ecosystems.

2.2.3.3. Validation

To assess the quality of the re-classed land cover data into 16 land cover classes, a validation process was carried out through the following steps:

1. **Fieldwork data collection**

Field work study was carried out to sample the 16 re-classed land cover types for qualitative validation and also its land cover diversity per 1km2 grid of the study area for quantitative validation. In order to sample and appreciate the two mentioned aspects a stratified random systematic sampling was used (adapted from (Alexander and Millington 2000)). It is important to note that this sampling strategy does not consider rare land cover types such as Eucalyptus forest and mixed forest, therefore a purposive sampling strategy was predetermined but could not be done due to lack of time in the field.

Stratification was done according to the amphibian species richness i.e. Low (0 to 3 species), Medium (4 to 7 species) and High (8 to 11 species) classes. The stratification element enabled sampling of the land cover types and diversity across the whole range of amphibian species richness.

Twelve random points were selected per each strata i.e. Low, Medium, and high species richness, totalling to 36 random points. The main reason for random selection was to avoid bias in the sampling exercise. Unfortunately only 33 of the 36 points could be visited due to various constrains (time, accessibility) experienced in the field.

At each random point a 1 km^2 plot was created and 16 systematic points (250 m apart) were created within each plot, as shown in Figure 2-6. All (528) systematic points were visited and the land cover composition and structure were observed, recorded and the land cover type (according to the 16 re-classed land cover types) deduced (as shown in appendix 2). The land cover sampling method at each point was adapted from the Land Cover Classification System 2 of FAO (Di Gregorio 2005) and a field sample record is elaborated in appendix 2. The main purpose of sampling 16 systematic points per 1km2 plots was to develop reference data required to validate the type and diversity of the reclassified land cover data as mentioned in the following sub-sections.

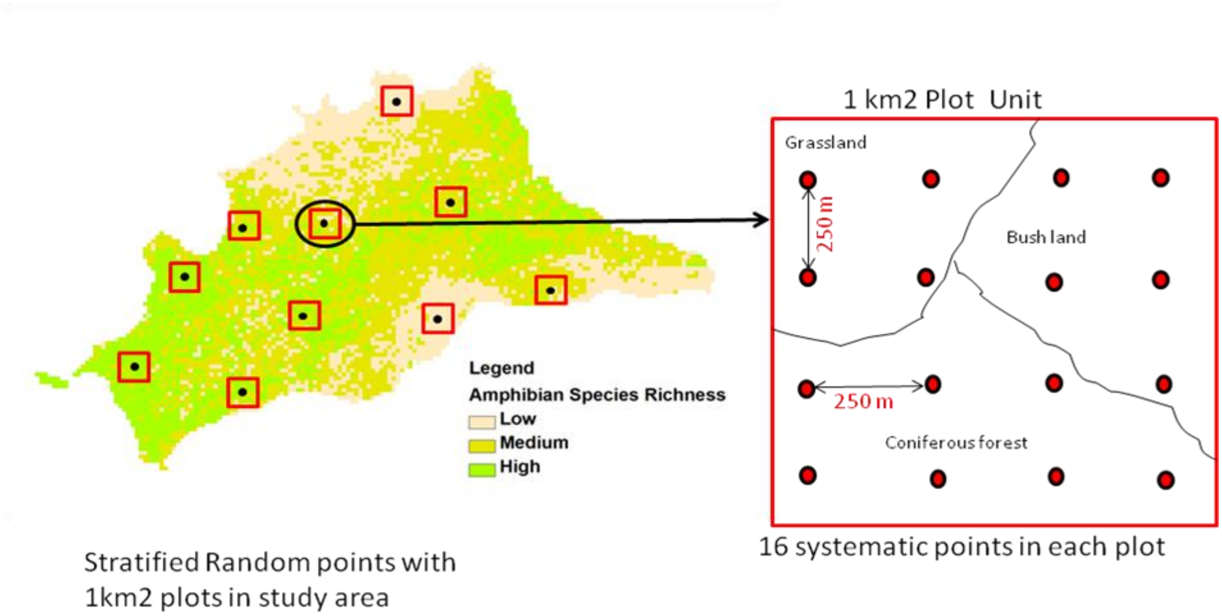

Figure 2-6 ***The stratified random and systematic sampling carried out in the field***

2. Land cover type Validation

One of the most common methods used to validate the accuracy of nominal data such as coded land cover classes is the percentage overall accuracy. It measures the accuracy of the land cover data using the major diagonal of a contingency table. However, this doesn't measure the chance agreement or error of the matrix, which is measured with the Kappa statistic. Therefore, to enhance the validation analysis a kappa test is normally required for accuracy assessment (Fielding and Bell 1997). Kappa analysis is based on the difference between the actual agreement and chance agreement (Cohen 1960).
.

The main steps required to carry out the land cover validation were as follows:

- The reference (field) data (528 XY points) containing the derived land cover types (numerically coded) were used to extract the corresponding land cover type (numerically coded) of the land cover data using the "extract to point" tool in ArcGIS 9.3 spatial analyst.

- The field data values were cross tabulated with the land cover data values in SPSS 16.0 and the Kappa value obtained.

- The percentage overall accuracy of the land cover data was also calculated using the contingency table with reference data (as columns) and the land code data (as rows) values in microsoft excel 2007.

3. Land cover diversity Validation

The land cover diversity validation was based on the Shanon diversity index (SHDI). This was carried out through the following steps:

- The Shannon Diversity index of each 33, 1km2, sample plot was calculated using the land cover types derived for the 16 systematic data points (as shown in figure 2.6), in MS. Excel 2007. For example a sample plot with, 5 of land cover type 1, 10 of land cover type 2 and 1 of land cover type 3, the SHDI will be calculated as:

$$SHDI= 5/16*\ln(5/16) + 10/16*\ln(10/16) + 1/16*\ln(1/16) = 0.8306$$

- The SHDI of the land cover data (raster-grid format) within the corresponding sample plots were also done using fragstats 3.3 software.
- The SHDI values from the sampled field data (x axis) were then regressed with the SHDI values derived from the corresponding land cover data (y axis). The R^2 was assessed and the significance of their association tested using the p-value.

2.2.4. Phase 3: Altitude (Elevation) Data

Elevation data was an important component for the research to derive the altitude standard deviation, as shown in Figure 2-4, to investigate the relationship between amphibian species richness and altitude range. For the study the Advanced Spaceborne Thermal Emission and Reflection Radiometer Global Digital Elevation Model (ASTER GDEM) was used.

The ASTER GDEM was developed jointly by the Ministry of Economy, Trade, and Industry (METI) of Japan and the United States National Aeronautics and Space Administration (NASA). It is a GeoTIFF format raster data with 30 meter resolution grids and georeferenced to the WGS84/EGM96 geoid. The accuracy of the elevation product is 20 meter with 95% confidence for the horizontal data and 30 meter with 95% confidence for the vertical data (METI 2009).

2.2.4.1. Altitude data layer

Six 1 by 1 degree tiles of the ASTER GDEM that covered the study area- indexed as ASGTM_N36W004, ASGTM_N36W005, ASGTM_N36W006, ASGTM_N37W005 and ASGTM_N37W006, were electronically downloaded from the Earth Remote Sensing Data Analysis Center (ERSDAC) of Japan (METI 2009).

The tiles were mosaiced in ArcGIS 9.3 and converted to a grid raster format with a WGS84 30N projection system. The mosaiced elevation data was then clipped using the “extraction tool”, in ArcGIS spatial analyst with the study area (Malaga province Administrative) boundary to produce an altitude data of the study area.

2.2.5. Phase 4: Simple Linear Regression/Correlation

To determine the relationship between the landscape heterogeneity determinants, land cover diversity (using SHDI) and altitude range (as standard deviation) to amphibian species richness at 1 km2 resolution the following steps were preferred, due to the tedious and time consuming option of analysing the whole study area using the landscape grid layer:

1. 5% sampling grid units were randomly selected from the landscape sampling grid layer population (see section 2.2.2). To effectively capture to the whole range of amphibian species richness across the study area a stratified random selection was carried out based on five classes of species richness i.e. Very low, Low, Medium, High and Very high species richness. The stratified random selection was done using the Hawths tool extension, "Select random option", of ArcGIS 9.3.

2. The pearson correlation and simple linear regression analysis was then done in SPSS 16 on the corresponding values.

3. A 2nd run of step 1 and 2 was done to confirm the simple linear regression/correlation analysis results.

2.2.5.1. Relationship between Amphibian species richness and Altitude Standard deviation

It is presumed that the wider the altitude ranges the more the range of environments in that landscape (Morena- Rueda, 2005 and 2007). Altitude standard deviation is a better measurable index (Rodriguez, 2005) than altitude range (Maximum-Minimum altitude) used in some studies (Morena-Rueda, 2005 and 2007) because it is more intuitive in determining the altitude diversity.

- The 5% of the sample grid units were used to clip out the elevation data using the extract by mask tool in ArcGIS 9.3, as shown below:

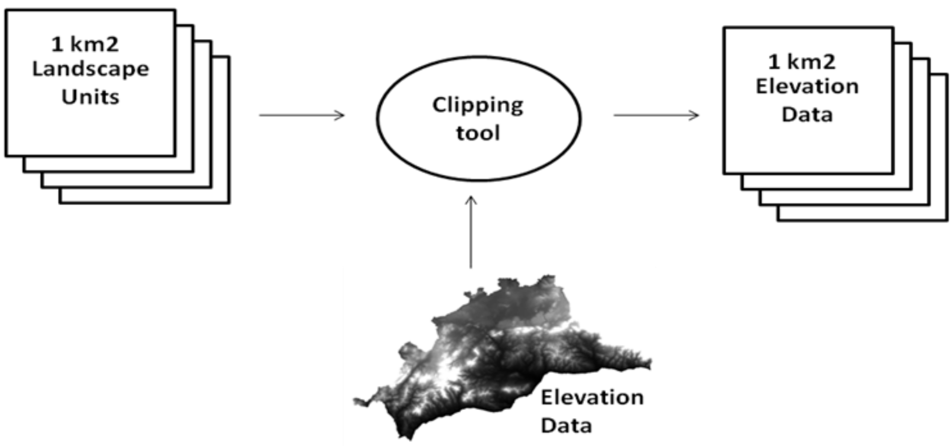

Figure 2-7 ***Extracting 1 km2 elevation data using the landscape units with the clipping tool in ArcGIS 9.3***

- The altitude standard deviation values for each landscape unit were derived from the attribute table of the 1km2 elevation data in ArcGIS 9.3.

- The corresponding values of species richness were also derived from the attribute table of the sample grid units in ArcGIS 9.3.

A scatter plot with a trend line and a simple linear correlation/regression analysis was done in SPSS 16 for amphibian species richness as the response variable against the altitude standard deviation as the explanatory variable. The significance of the correlation slope determined using the p value at a critical value of 0.05, and the R^2 was assessed.

The procedure was repeated to confirm the results.

2.2.5.2. Relationship between Amphibian species richness and land cover diversity

The selected 5 % sampling grid units were used to clip out the corresponding subset of 1 km^2 land cover data from the land cover data (in raster format) of the study area, using the "extraction by mask tool", in ArcGIS 9.3, as shown below:

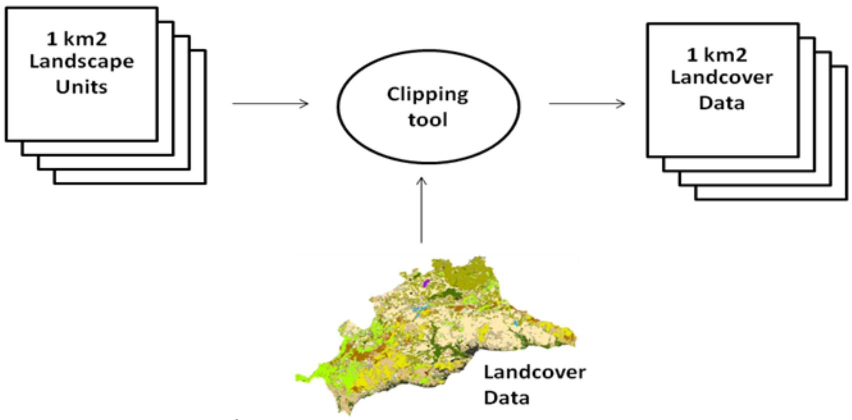

Figure 2-8 ***Extracting the 1km^2 land cover data using the landscape sampling units with the clipping tool in ArcGIS 9.3***

The SHDI for each 1 km2 land cover data unit were derived on the basis of the land cover type and its relative distribution using fragstats 3.3 as follows:

- All the 348 land cover data units were introduced as a batch file grid format; the class properties file, which mentions the land cover type and respective codes, were also introduced as a text file format.
- The inputs were processed and the SHDI values for each sampling grid unit recorded in a landscape metrics table.
- The corresponding values of species richness were obtained from the attribute table of the sampling grid units in ArcGIS 9.3.

A scatter plot with a trend line and a simple linear correlation/regression analysis was done in SPSS 16 for amphibian species richness as the response variable against the SHDI value of land cover diversity as the explanatory variable. The significance of the correlation slope was determined using the p value at a critical value of 0.05, and the R^2 assessed.

The procedure was repeated to confirm the results.

2.2.6. Phase 4: Testing the equality of Amphibian species richness at different Land cover types

(Atauri and de Lucio 2001) mentions that amphibian species richness differs depending on the nature of the land cover type. Land cover of Xerophytic nature tends to have low species richness while land cover of Mesophytic nature tends to have high species richness of amphibians. This in turn provokes a question as to whether species richness of amphibians in the study area are equal across all land cover types.

To answer the question an ANOVA test was carried out on the mean of species richness for each land cover type as follows:

- The species richness point layer (obtained from section 2.2.2) was used to extract the land cover type values (as land cover codes) from the land cover data- as raster format, using the extraction tool in ArcGIS 9.3.
- 20 % of the points representing each land cover type were randomly selected using stratified random sampling in "Hawth sampling tools", ArcGIS 9.3. The stratification was based on the land cover type (code).
- The selected random values with the appending land cover code values were used to test the species richness mean equality per land cover type with ANOVA and post hoc (pairwise comparison) test in SPSS 16.
- A descriptive analysis (mean and standard deviation) was also derived on the random values and a bar graph was used to illustrate the mean and the mean standard error of species richness across the different land cover types. Land cover types with significantly equal mean of species richness were classified together.

2.2.7. Phase 5: Stepwise regression analysis

Stepwise regression analysis factors in more than one explanatory variable as functions to a response variable. It allows us to eliminate variables that do not provide significant information and select those that best explain the variability in the response variable (in this case amphibian species richness) and evaluate their relative importance (Atauri and de Lucio 2001; Rodríguez, Belmontes et al. 2005) . To measure the degree of significance of altitude standard deviation and land cover diversity variables with other important variables that influence the variation of amphibian species richness a stepwise regression was carried out as follows:

- The most important continuous explanatory variables from the results of the maxent model (i.e. the variable with the highest gain when alone and that that will reduce the overall gain most if omitted(Phillips, Anderson et al. 2006)) for each species were considered.
- All the considered raster layer values were compiled together using the combine tool in ArcGIS 9.3 spatial analyst.
- 5% random points of species richness were selected and used to extract the combined values using the “extract values to points” tool in ArcGIS 9.3.
- The attribute table of the species richness was exported as a data base file (dbf) to microsoft excel 2007 and joined with the corresponding altitude standard deviation and land cover diversity (SHDI) values that were obtained using the landscape grid units (as mentioned in section 2.2.5.1 and 2.2.5.2 respectively).
- The explanatory variables and species richness (response) variable were exported to SPSS 16 where a stepwise regression analysis was carried out.
- A second confirmatory run was also done.

3. Results

3.1. Amphibian species occurrence data

3.1.1. Multicollinearity test results

The selected explanatory environmental variables, with a VIF =< 10, after running a multicollinearity test in SPSS 16 are shown on Table 3-1. These continuous variables and the classified NDVI, Land cover and Soil categorical data were used to model the distribution of the 11 amphibian species of the study area, using the maxent software.

Environmental Variable	VIF	Environmental Variable	VIF
Distance to stream	1.074	NDVI spring	7.151
Distance to spring, well and dam	4.769	NDVI summer	6.386
Distance to large rivers and water bodies	1.494	Population density	1.050
Evapotranspiration in winter	8.160	Precipitation autumn	3.414
Evapotranspiration in summer	5.992	Precipitation spring	5.104
Humidity index	1.394	Radiation autumn	6.084
NDVI autumn	5.874	Radiation spring	7.040
South exposure	1.983	Radiation summer	3.020
West exposure	2.026	Radiation winter	3.448
Temperature variation	11.149	Altitude	9.143
Temperature July	8.922	Slope	1.573

Table 3-1 ***Multicollinearity results of the explanatory variables used in the maxent modelling software.***

Despite temperature variation being above 10 (11.15) it was still used in the model because amphibians are sensitive to temperature extremes. Amphibians are ectotherms and have permeable skin and therefore they are prone to freezing during low frost temperatures and desiccation during high summer temperatures (Adler 2004; Qian, Wang et al. 2007).

3.1.2. Species distribution maps

Based on the maxent software, as explained in section 2.2.1.2, the niche based species distribution, modelled maps of the probability of occurrence at 1km^2 resolution for each amphibian species in the Andalusia region were as follows:

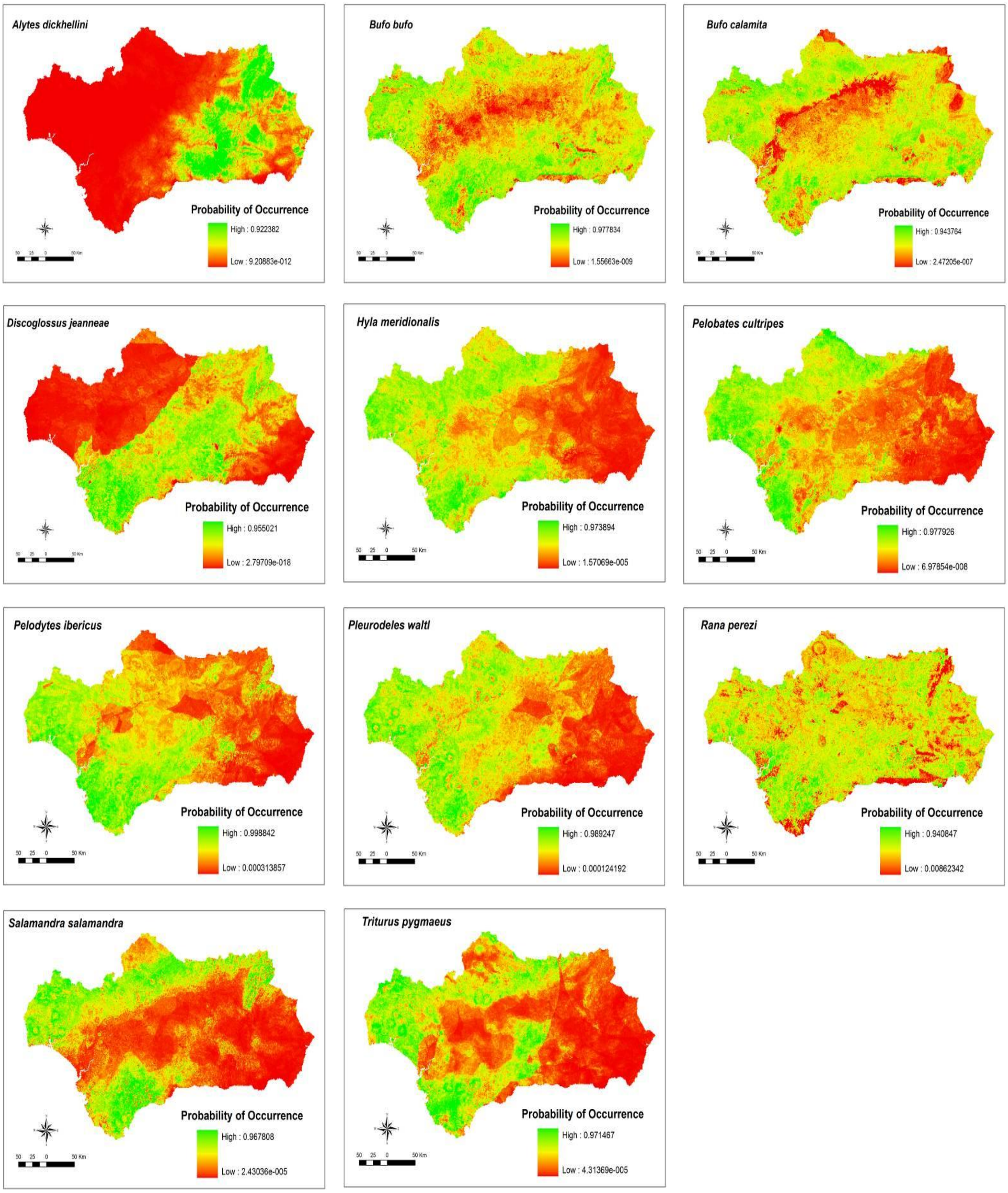

Figure 3-1 ***Probability of occurrence for each of the 11amphibian species in Andalusia region***

3.1.3. Validation of Amphibian species occurrence data

The evaluation of each species occurrence data model using both the threshold dependent (AUC) and an independent (Kappa) technique, as explained in section 2.2.1.3, are shown in Table 3-2

Amphibian Species	Maxent test data AUC	Maxent Logistic Threshold	Kappa values	Number of presence points obtained from the independent 1 by 1 km grid data	Omission error rate
Alytes dickhellini	0.848	0.255	0.642	3	0.667
Bufo bufo	0.563	0.460	0.459	48	0.208
Bufo calamita	0.502	0.461	0.359	49	0.265
Discoglossus jeanneae	0.689	0.333	0.609	33	0.375
Hyla meridionalis	0.667	0.395	0.650	10	0.4
Pelobates cultripes	0.687	0.358	0.520	4	0.0
Pelodtyes ibericus	0.685	0.387	0.592	26	0.192
Pleurodeles waltl	0.645	0.390	0.590	6	0.333
Rana perezi	0.436	0.469	0.11	72	0.222
Salamandra salamandra	0.715	0.336	0.698	22	0.273
Triturus pygmaeus	0.738	0.352	0.562	3	0.333

Table 3-2 ***Validation results: Self validation at the Andalusia region extent, from the maxent test data AUC and the threshold dependent, Kappa; and Independent validation using independent presence points to derive the omission error rate at the study area (Malaga province) extent.***

The average AUC for all species is 0.652. This means that on average if a random selection is done, 65.2% of the encounters will be from the positive group and the remaining 34.8 % will be from the negative group (Deleo 1993) . On average the kappa was 0.527. The average omission error rate was 0.297.

Alytes dickhellini has the the best AUC of 0.848 compared to *Rana perezi* with the lowest (0.436) this is mainly due to the rareness and commonness of the species respectively. On carrying out the kappa evaluation which is more holistic (it also involves absence values),*Salamandra salamandra* has the highest kappa of 0.698 and still *Rana perezi* with the lowest of 0.11. On carrying out an omission error rate using independent data *Pelobates cultripes* has the lowest omission rate of 0 while *Alytes dickhellini* has, ironically the highest of 0.667, but the number of presence data for this two species were very low.

3.1.4. Amphibian Species richness data

The amphibian species richness data layer was obtained from summation of the presence and absence occurrence of all species is as shown in Figure 3-2.

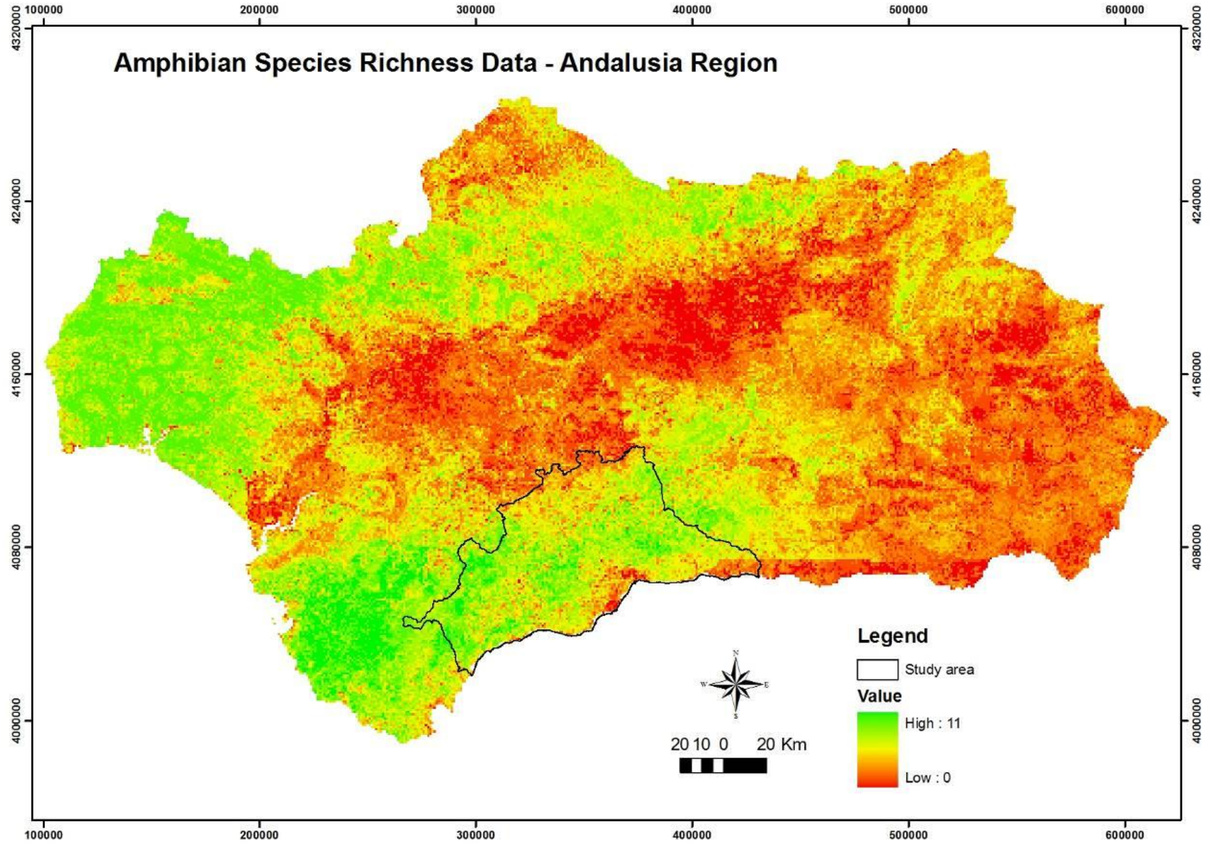

Figure 3-2 ***Amphibian species richness based on the summation of the presence and absence occurrences of the 11 amphibian species of the study area at an Andalusia region extent***

The species richness layer was then clipped with the administrative boundary of the study area (Malaga province) to obtain the species richness layer data of the study area, as shown in Figure 3-3.

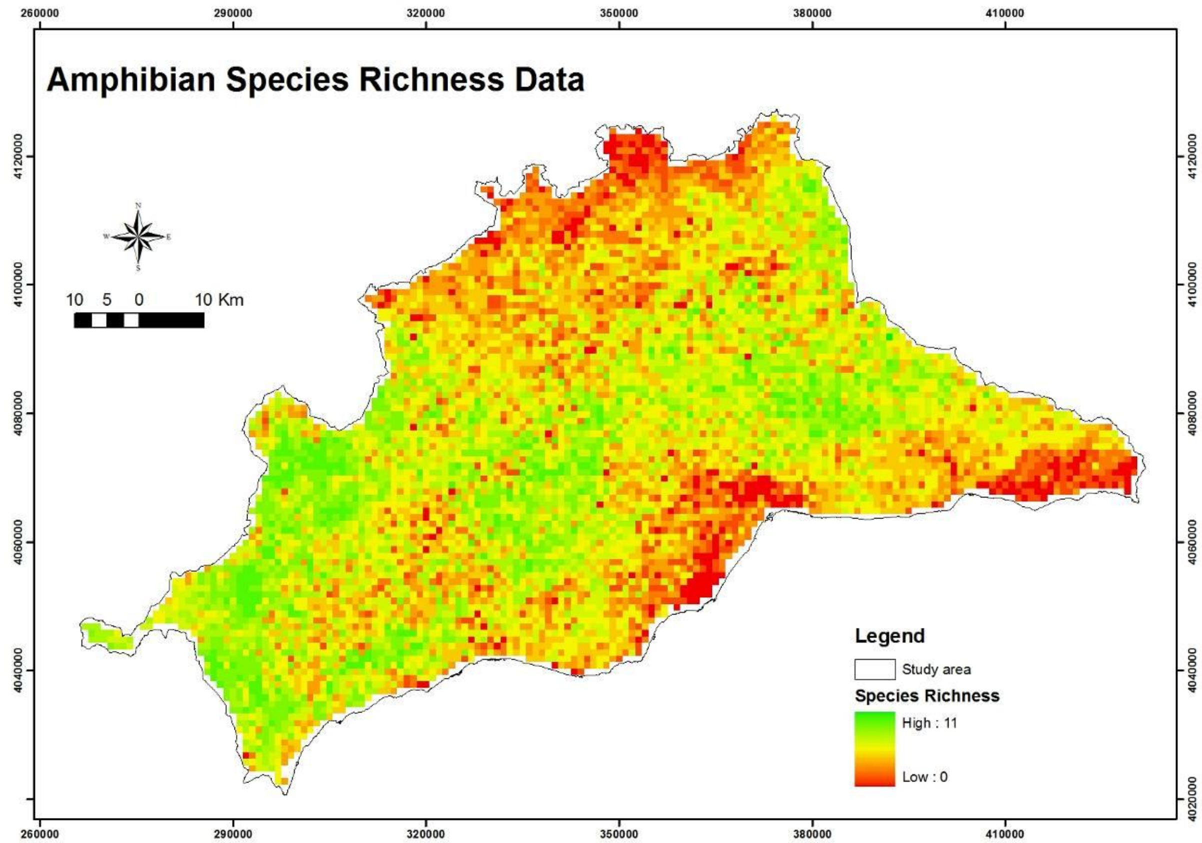

Figure 3-3 ***Amphibian species richness of the study area (Malaga province)***

The amphibian species richness ranges from 0 to 11 species. Amphibian species richness are evidentially low at the mid South and South East part of the study area along the large coastal towns such as Malaga, found at the mid South and Torrox, found at the South East of the study area. High amphibian species richness is found at the inland areas of the study area especially to the West, which is predominantly broadleaved forest.

3.2. Landscape sampling grid data

The 1 km^2 landscape vector grid layer and its appending attribute table, containing the amphibian species richness values, of the study area is as shown in Figure 3-4

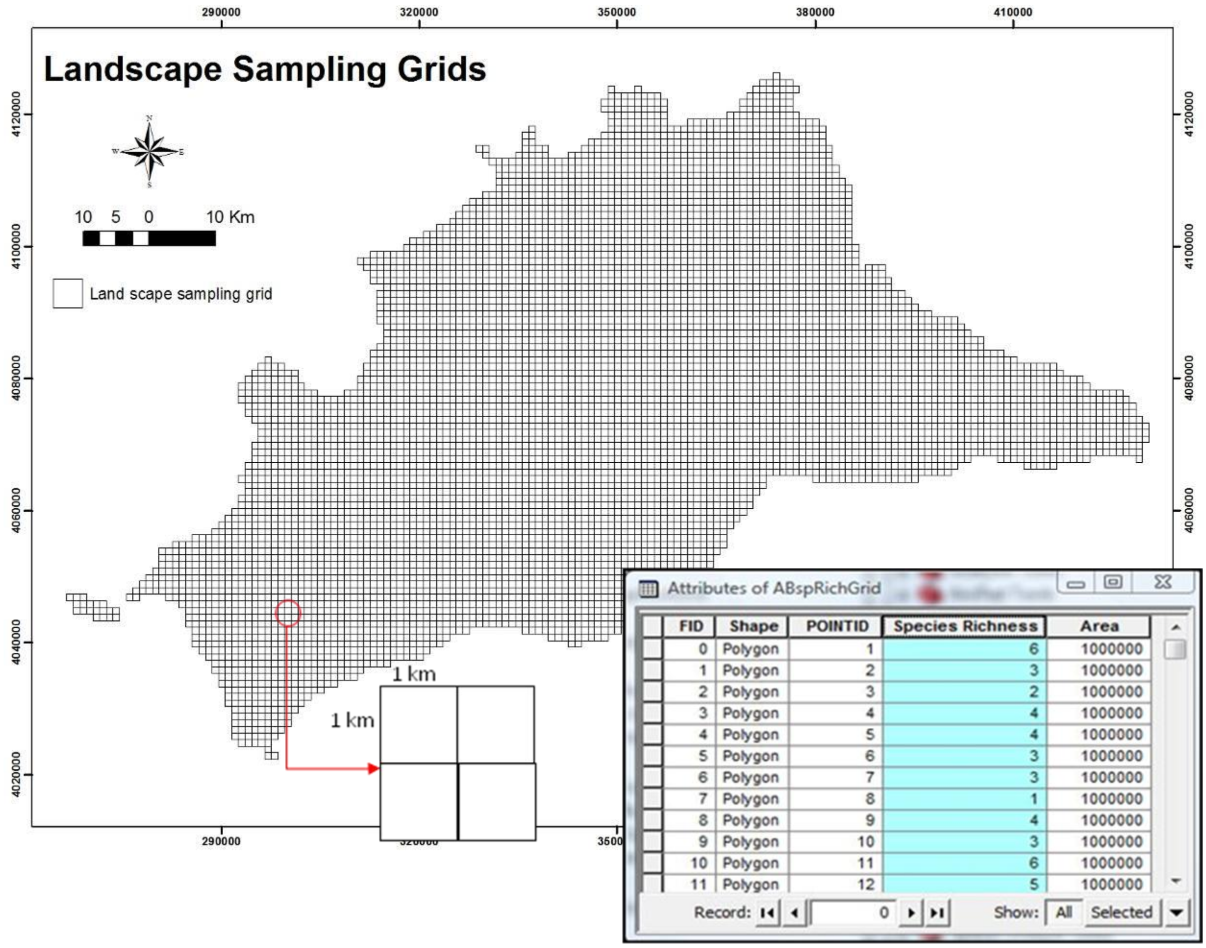

Figure 3-4 ***Landscape vector grids of the study area (Malaga province)***

3.3. Land cover data

The land cover data of the study area (Figure 3-5) showing the geographic distribution of the 16 land cover types-15 land cover types (minus the Sea) were used in the analysis:

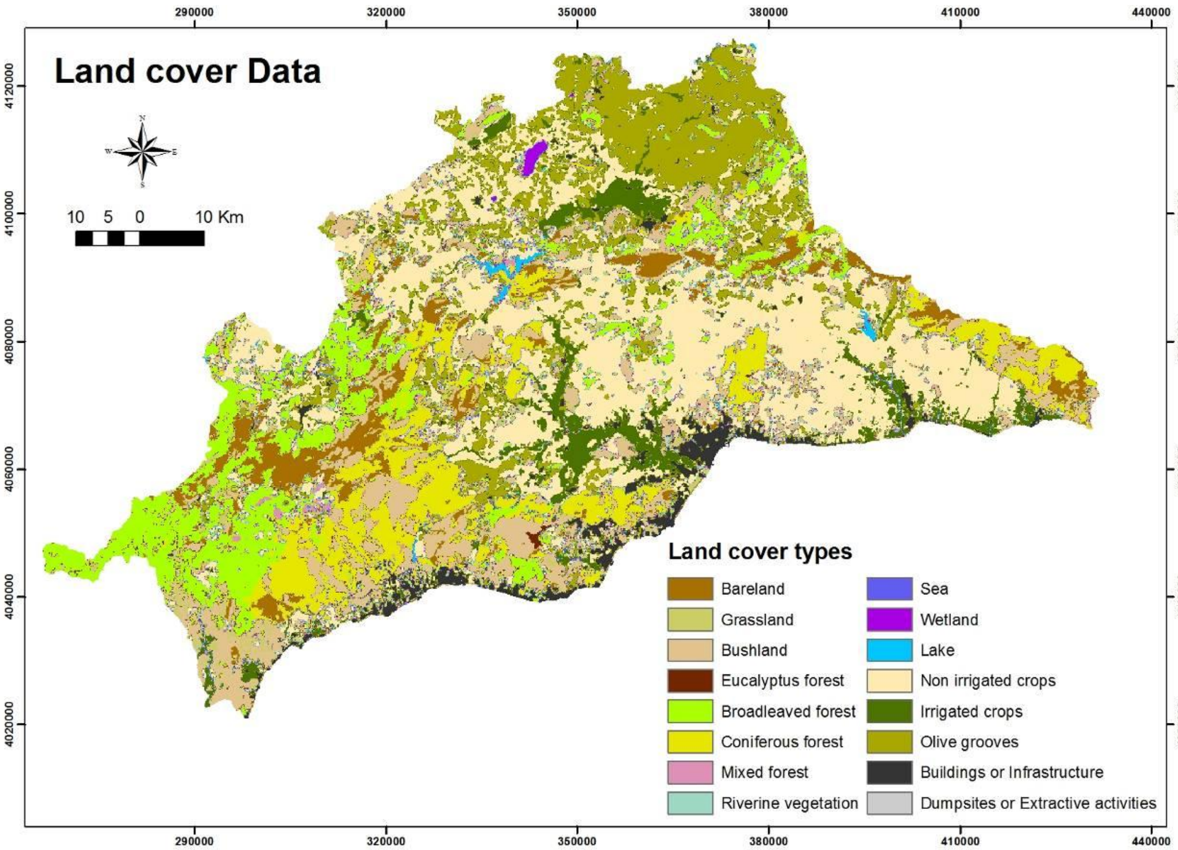

Figure 3-5 ***Land cover type data of the study area***

3.3.1. Land cover type Validation

With reference to section 2.2.3.3, the contingency table of the land cover data and the respective percentage overall accuracy and kappa values are as follows:

Land cover types		Field Observation											Total	
		BaLa	GrLa	BuLa	BrFo	CoFo	WeLa	NoIrCr	IrCr	OlGr	BuIn	ExDu		User Accuracy
Data	BaLa	15	7	16	4	0	2	1	0	0	2	0	47	**31.91**
	GrLa	0	11	0	0	0	0	0	0	0	13	0	25	**44.00**
	BuLa	6	1	46	2	12	0	0	0	3	15	0	87	**52.87**
	BrFo	1	4	4	57	0	1	0	0	0	0	0	68	**83.82**
	CoFo	3	3	2	0	31	0	1	0	0	1	0	41	**75.61**
	WeLa	0	0	2	0	0	2	0	0	0	0	0	4	**50.00**
	NoIrCr	0	1	1	1	3	2	79	2	36	12	2	141	**56.03**
	IrCr	0	0	0	0	0	0	0	6	0	1	0	7	**85.71**
	OlGr	0	0	1	0	0	0	8	0	46	1	0	57	**80.70**
	BuIn	0	1	0	1	0	0	0	0	1	42	0	46	**91.30**
	ExDu	0	0	1	0	0	0	0	0	0	1	3	5	**60.00**
Tota l		25	28	73	65	46	7	89	8	86	88	5	**528**	
Producer Accuracy		**60**	**39.29**	**63.01**	**87.69**	**67.39**	**28.57**	**88.76**	**75**	**53.49**	**47.73**	**60**		

Figure 3-6 ***The validation contingency table of the field observation versus data land cover types (BaLa = Bare land, GrLa = Grassland, BuLa = Bush land, BrFo = Broadleaved Forest, CoFo = Coniferous Forest, WeLa = Wetland, NoIrCr = Non Irrigated crops, IrCr = Irrigated crops, OlGr = Olive groves, BuIn = Buldings and Infrastructure, ExDu = Extractive sites and Dump sites)***

Percentage Overall Accuracy = 64.02 %
Kappa = 0.60

Three rare land cover types out of the fifteen required land covers types were not validated through purposive sampling due to time constrains, as mentioned in section 2.2.3.3. These were riverine vegetation, mixed forest and Eucalyptus forest.

Bare land, grassland and bush land cover have a low producer and user accuracy. This may be due to the construction boom that has occurred in Spain over the past decade, and that is why the Building and Infrastructure cover has high user accuracy but a low producer accuracy (this will be explained further under the discussion chapter section 4.3.1).

3.3.2. Land cover diversity Validation

With reference to, section 2.2.3.3, the relationship between the land cover diversity values (as SHDI) of the reference/field data and its corresponding land cover data were shown using a scatter graph and its regression line and analysis derived as follows:

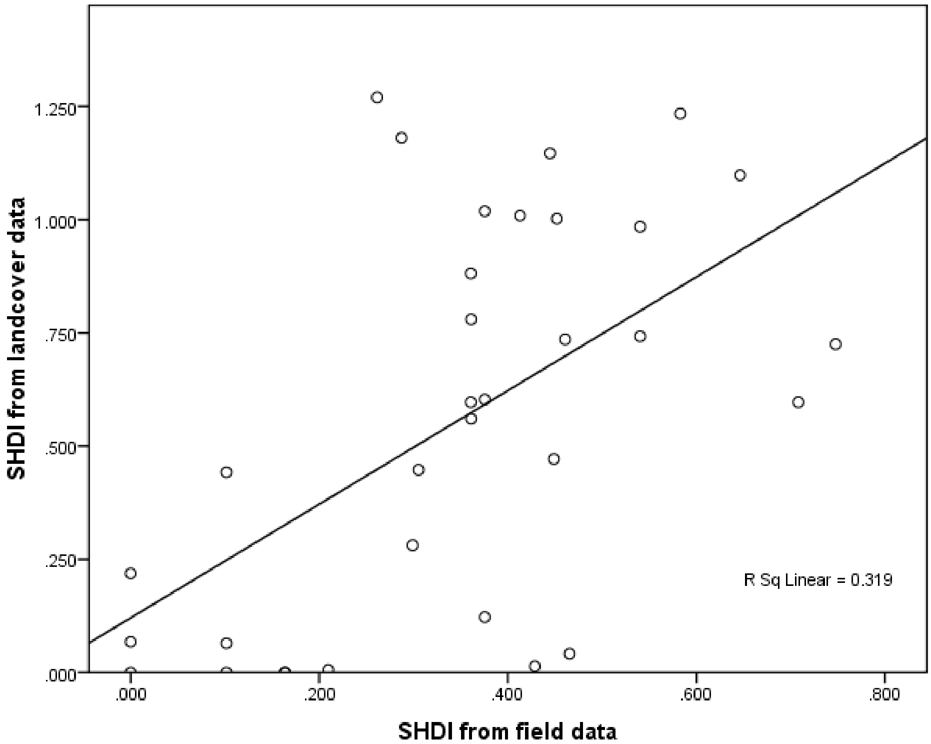

Figure 3-7 ***Scatter graph with regression line relating the land cover diversity values (measured with SHDI) collected from the field and that from the land cover data***

Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.565[a]	.319	.297	.3645787

Coefficients

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
1	(Constant)	.121	.131		.924	.363
	SHDI from field data	1.255	.329	.565	3.811	.001

Dependent Variable: SHDI from landcover data

Table 3-3 ***Regression Model summary output (SHDI land cover data values against SHDI field values)***

There was a low fitness of the regression model (R^2 =0.297). The slope (b= 1.255) shows that the diversity from the land cover data is overestimated, compared to the field observation values. The p value is 0.001, which is below the critical value of 0.05. This in turn means there is a significant correlation between the land cover diversity found in the field and that derived from the land cover data.

3.4. Altitude Data

The mosaic and clipped altitude raster data of the study area, sourced from ASTERDEM, is as shown below.

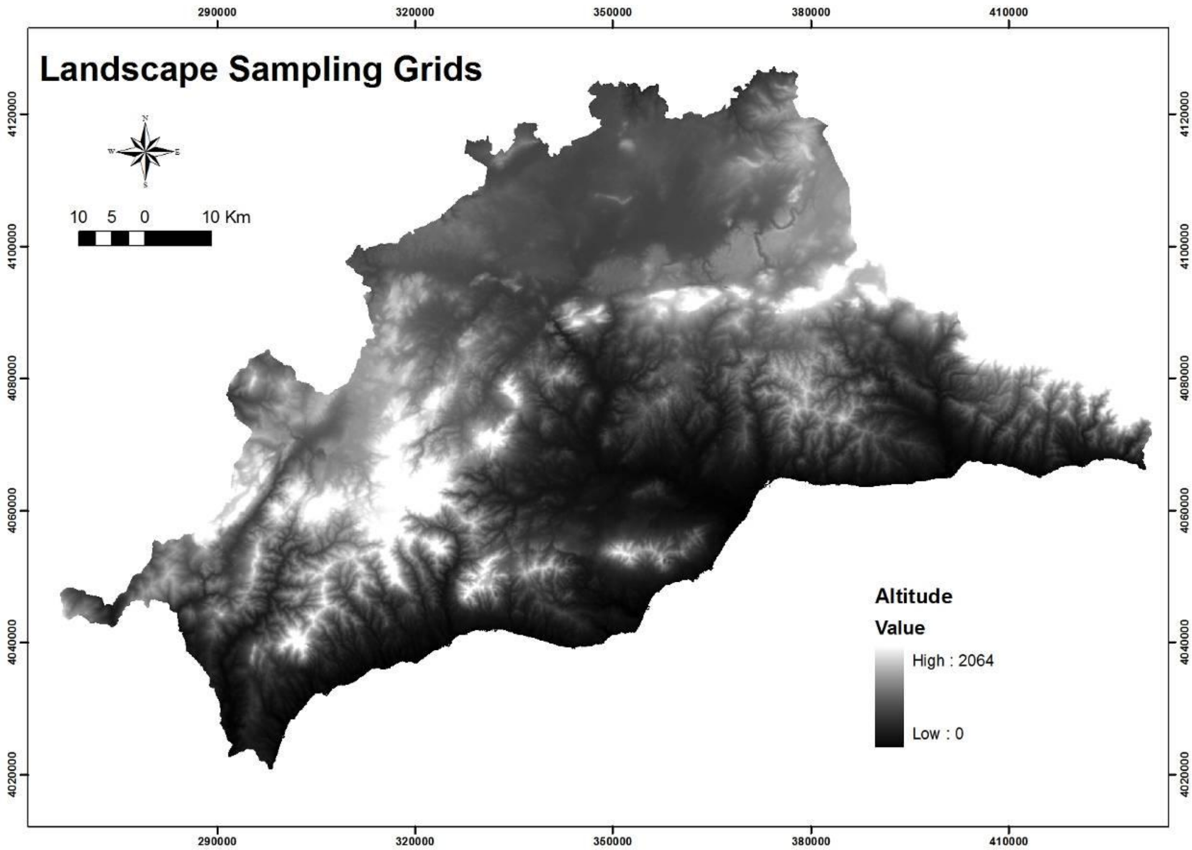

Figure 3-8 ***Altitude data of the study area (Malaga province)***

3.5. Simple linear regression/correlation analysis

3.5.1. Amphibian Species richness and Altitude Standard deviation relationship

With reference to section 2.2.5.1, the correlation and regression analysis between the amphibian species richness (response variable) and altitude standard deviation (explanatory variable) were as follows:

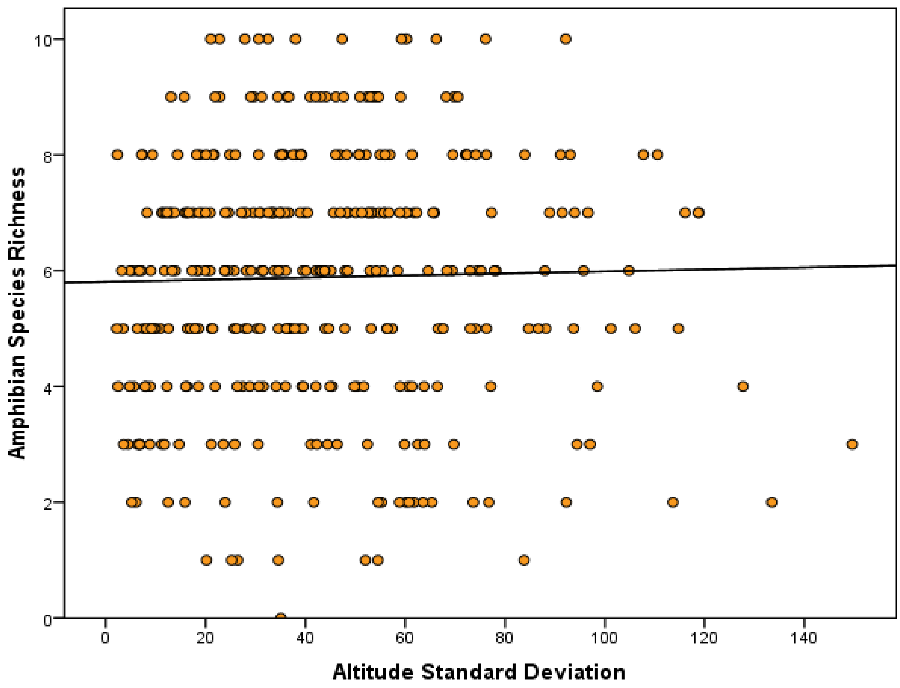

Figure 3-9 ***Scatter plot and regression line model of Amphibian Species richness against the altitude standard deviation***

Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.022[a]	.000	-.002	2.169E0

Coefficients

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
1	(Constant)	5.810	.216		26.891	.000
	Altitude Standard Deviation	.002	.004	.022	.413	.680

Dependent Variable: Amphibian Species Richness

Table 3-4 ***Regression model summary analysis of Amphibian species richness and Altitude Standard Deviation***

As shown in Figure 3-9 and Table 2-1, there is no correlation (slope =0.022) between altitude standard deviation (explanatory variable) and amphibian species richness (response variable). The p value (0.680) is higher than the critical value of 0.05 and it has a very low R^2 of 0.002. This means that the regression model explains only 0.2 % of the population analysed. Therefore the ***Ho: b=0* was not rejected**; this means that there is no significant correlation between the aforementioned variables.

The confirmatory results of the above analysis were similar as shown in Appendix 3

3.5.2. Amphibian species richness and Land cover diversity relationship

With reference to section 2.2.5.2, the correlation and regression analysis between the amphibian species richness (response variable) and land cover diversity (explanatory variable) were as follows:

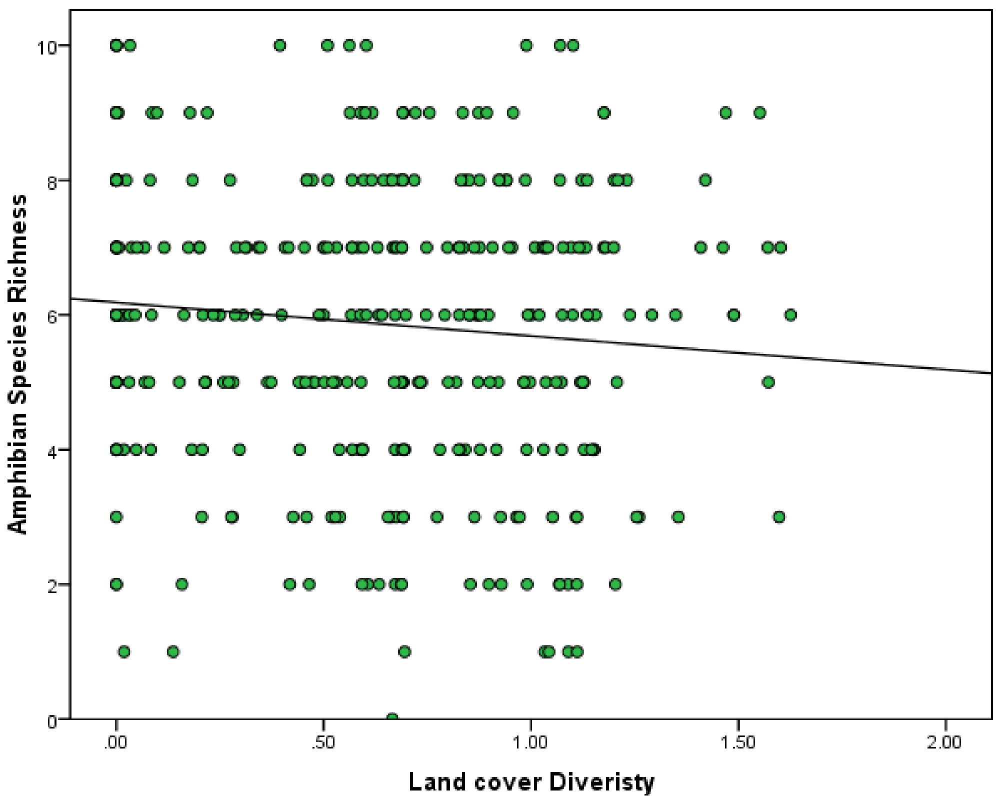

Figure 3-10 ***Scatter graph and regression line model of amphibian species richness against land cover diversity***

Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.099	.010	.007	2.159E0

Coefficients

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
1	(Constant)	6.185	.199		31.064	.000
	Land cover Diveristy	-.498	.269	-.099	-1.853	.065

Dependent Variable: Amphibian Species Richness

Table 3-5 ***Regression model summary analysis of Amphibian species richness and Land cover diversity***

As shown in the analysis (Figure 3-10 and Table 3-5) there is a weak negative correlation with a slope of -0.99 between amphibian species (response variable) richness and land cover diversity (explanatory variable). Only 0.7% of the analysed population was explained by the regression model (R^2 = 0.007). Furthermore the p value was 0.065, which is above the critical value of 0.05. This in turn means the ***Ho: b=0 was not rejected***; therefore no significant correlations between the land cover diversity and amphibian species richness.

The confirmatory results of the above analysis were similar as shown in Appendix 3

3.6. The equality of Amphibian species richness per land cover type

3.6.1. Descriptive statistic

With reference to methodology section 2.2.6 the descriptive statistics of species richness per land cover type were obtained as follows:

Mean species richness in:	N	Range	Mean	Std. Deviation
Bare land	92	8	6.05	1.706
Grassland	29	6	7.31	1.713
Bush land	179	10	5.16	2.435
Eucalyptus forest	20	5	6.80	1.436
Broadleaved forest	151	7	7.38	1.649
Coniferous forest	153	9	5.86	2.026
Mixed forest	22	8	6.20	2.093
Riverine vegetation	20	6	6.45	1.849
Wetlands	20	8	3.30	2.536
Lakes	23	9	4.91	2.661
Non Irrigated crops	474	10	6.13	1.922
Irrigated crops	70	9	5.36	2.525
Olive groves	183	9	5.73	1.987
Building & Infrastructure	38	7	3.05	1.785
Extractive & Dump sites	24	5	6.25	1.410

Table 3-6 ***Mean and standard deviation of amphibian species richness per land cover type***

As a preliminary visualization of the equality of the means of amphibian species richness per land cover type (as shown in Table 3-6), it is possible that the means are not equal across different land cover types, this in turn prompt for an ANOVA test.

Broadleaved forest cover had the highest number of amphibian species richness, with a mean of 7.38 while Building and Infrastructure had the lowest amphibian species richness, with a mean of 3.05.

3.6.2. Testing normality

Testing the normality distribution of the sample data is a requirement before ANOVA test can be effectively carried out.

	Kolmogorov-Smirnov[a]			Shapiro-Wilk		
Amphibian Species Richness in:	Statistic	df	Sig.	Statistic	df	p value
Bare land	.144	20	.200*	.950	20	.372
Grassland	.239	20	.004	.884	20	.021
Bush land	.169	20	.135*	.923	20	.115
Eucalyptus forest	.155	20	.200*	.943	20	.268
Broad leaved forest	.131	20	.200*	.968	20	.719
Coniferous forest	.172	20	.122*	.937	20	.209
Mixed forest	.155	20	.200*	.953	20	.410
Riverine vegetation	.167	20	.146*	.936	20	.198
Wetlands	.218	20	.014	.883	20	.020
Lakes	.139	20	.200*	.924	20	.118
Non Irrigated crops	.176	20	.105*	.946	20	.308
Irrigated crops	.149	20	.200*	.958	20	.511
Olive groves	.205	20	.027	.925	20	.124
Buildings & Infrastructure	.242	20	.003	.918	20	.089
Extractive & Dump sites	.197	20	.040	.905	20	.051

Table 3-7 ***Test for normality of data species richness samples per land cover type***

As shown in Table 3-7 most of the samples per land cover type were normally distributed (at a critical p value of 0.05) according to Kolmogov-Smirnov and Shapiron-Wilkinson test. This in turn warrants an ANOVA test to be carried out.

3.6.3. Test for homogeneity of variance

With reference to the standard deviation obtained in Table 3-6 it seems the sample data variance is heterogeneous therefore a homogeneous test of variance was done using levene statistics, and the results are as shown on Table 3-8.

Species Richness

Levene Statistic	df1	df2	p value
5.324	14	1477	.000

Table 3-8 ***Test of Homogeneity of Variances using levene statistics***

The variance of the sample data is not homogeneous this means that an ANOVA and a counter ANOVA test (Welch and forsythe test) were carried out to confirm whether the amphibian species richness means are equal per land cover type.

3.6.4. ANOVA test with Post hoc pair wise test

Species Richness	Sum of Squares	df	Mean Square	F	p value
Between Groups	1033.362	14	73.812	18.291	.000
Within Groups	5960.231	1477	4.035		
Total	6993.592	1491			

Table 3-9 ***ANOVA test results of equality of means of amphibian species richness per land cover type***

Robust Tests of Equality of Means

Species Richness	Statistic[a]	df1	df2	p value
Welch	19.965	14	185.738	.000
Brown-Forsythe	18.088	14	414.890	.000

Table 3-10 ***Welch and Brown-Forsythe test results of the equality of means of amphibian species richness per land cover type, it was required as a confirmatory test of ANOVA because the variance and sample size from each land cover were different***

With reference to the ANOVA results (Table 3-9) and the Welch and Brown-Forsythe results (Table 3-10) the p value was much lower than critical 0.05 p value therefore the ***Ho: μ of land cover type1 = μ land cover type2 = μof land cover type3... μ th*** **was rejected**. This means that the mean of the species richness per land cover type are not equal.

3.6.4.1. Pair wise comparison, post hoc test of the mean of species richness between land cover types

ANOVA test is an omnibus test and it only mentioned that the amphibian species richness per land cover types are not equal but didn't elaborate which land cover types have an equal mean species richness and which ones don't. Therefore a post hoc, pair wise comparison test was carried out to elaborate more on the variation/equality of the amphibian species richness per land cover type.

The sample size and variance are not equal as shown in Table 3-6 and Table 3-8, therefore a post hoc, Games -howels test was preferred to tukey test. The results are as shown in (appendix 4)

The mean amphibian species richness per land cover types were graphically represented using bar graphs with the standard error from the mean (Figure 3-11). The bar graphs were arranged in order of their increasing mean from left to right.

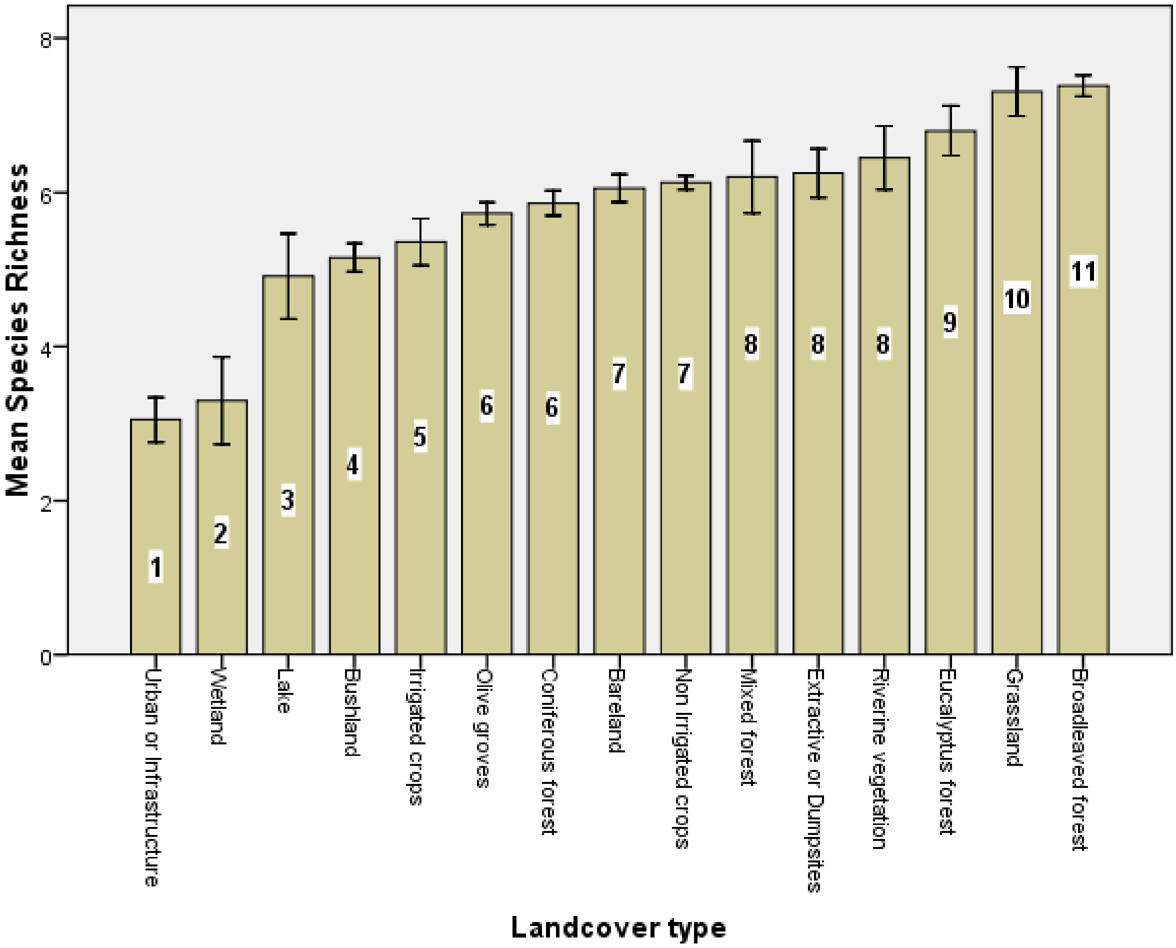

Figure 3-11 ***Bar graph of the mean Amphibian Species richness per the land cover type with the standard error of the mean and their grouped classes***

As shown in Figure 3-11 the land cover types were classified into eleven classes in an ascending order i.e. 1 to 11, based on the significant (at a critical p value of 0.05) mean equality of amphibian species richness per land cover type. The classification was derived using the post hoc, Games-Howel, pair wise comparison test results (appendix 4) and the amphibian species richness per land cover type classification matrix in appendix 5

The olive grove and coniferous forest had the same mean of amphibian species richness at an intermediary level compared to other classes. Other classes with the equal amount of amphibian species richness were Bareland and non irrigated crop covers and also riverine vegetation with extractive/dump sites and mixed forest.

3.7. Significance of explanatory variables to Amphibian Species richness using stepwise regression

The most important explanatory variables that influence the distribution of each amphibian species according to the maxent model gain were as follows:

Amphibian Species	Total Gain	Variable with highest gain if alone	Variable with highest influence on total gain
Alytes dickhellini	1.4	Radiation in spring	Radiation in spring
Bufo bufo	0.14	Temperature in July	West exposure
Bufo calamita	0.12	NDVI classified	Population density
Discoglossus jeanneae	0.78	Radiation in spring	Radiation in spring
Hyla meridionalis	0.50	Evapotranspiration in winter	NDVI classified
Pelobates cultripes	0.60	Evapotranspiration in winter	South exposure
Pelodtyes ibericus	0.53	Precipitation in autumn	NDVI classified
Pleurodeles waltl	0.48	Distance to spring, wells or dams	Population density
Rana perezi	0.07	NDVI classified	NDVI classified
Salamandra salamandra	0.76	Evapotranspiration in winter	Slope
Triturus pygmaeus	0.76	Evapotranspiration in winter	Distance to spring, wells or dams

Table 3-11 ***Maxent "gain" results for each amphibian species***

The results of the stepwise regression using species richness as the response variable and the land cover diversity, altitude standard deviation and other primary variables inherited from the maxent "gain" as explanatory variables were as shown:

Adjusted R squared (4^{th} model)	Primary variables	Beta (Regression Coefficient)	Significance at 0.05 critical p value
	Temperature in july	-0.290	0.000
0.214	Evaporation in winter	0.269	0.000
	Slope	-0.262	0.001
	Human Population Density	-.100	0.040

Table 3-12 ***Stepwise regression results of the last (4th) model***

With a R^2 regression fit of 0.214 (explaining 21.4 % of the data analysed), the primary variables with a significant correlation to amphibian species richness are shown in Table 3-12., in order of their importance in determining the variation in amphibian species richness. The explanatory variable with the highest influence to the variation in amphibian species richness was temperature in July with a negative correlation of -0.290.

The stepwise regression models summary and its confirmatory run are shown in Appendix 7.

4. Discussion

4.1. Introduction

a) To kick off with this chapter a review of the pre-processing and validation of the two important data components (Amphibian species richness data and land cover data) used in the analysis were discussed (section 4.2)
b) Discussion on the relationship of amphibian species richness to landscape heterogeneity (section 4.4)
c) Discussion on the important/primary determinants of amphibian species richness (section 4.6).
d) Uncertainty and limitations that may have obscured the excepted results (section4.7).

4.2. Amphibian species distribution (niche based) modelling

4.2.1. Overview

Niche based modelling represents an approximation of a species realised niche (subset of species fundamental niche where it actually occupies) of a study area in relation to the environmental variables considered (Hutchinson 1959; Phillips, Anderson et al. 2006). For the study the approximation of the realised niche of each of the 11 amphibian species was modelled in maxent based on the occurrence data of the amphibian species (i.e. their realised niche) and the environmental variables that determine their spatial distribution. The extent of modelling was larger than that of the required study extent i.e. Andalusia region instead of Malaga province.

Training species distribution models beyond the required extent is a recommended tact to improve the robustness of a species distribution model (Pearson and Dawson 2003; Peterson and Holt 2003). For the study case, training the models on the Andalusia extent was an ideal technique used to develop robust amphibian species distribution models for the subset study extent of Malaga province. This is because a larger extent improves the chances of the training data (species presence data) to encounter greater variations of the environmental variables that are used to develop the model and as a result improve the robustness of the output model. This is especially true for the rare species such as *Alytes dickhellini* and *Triturus pygmaeus* which had only a few presence data at the study area extent of Malaga province.

Finally the species distribution models for each species were divided into absence and presence data base on “equal specificity and sensitivity, test, threshold value ”, then summed up to derive the amphibian species richness data that was clipped into the study area (Malaga province) extent.

4.2.2. Modelling Validation

The predictive power of the species distribution model for each of the 11 amphibian species was evaluated using three known evaluation techniques, (Fielding and Bell 1997; Anderson, Lew et al. 2003; Phillips, Anderson et al. 2006), these are ROC/AUC, kappa statistics and omission error rate each improving the short comings of the other. For example the test data AUC for rare species such as *Alytes dickhellini* (Table 3-2) was the highest with 0.848 but on the other hand common species such as *Rana perezi* and *Bufo Calamita* had a low AUC of 0.436 and 0.502 respectively. This may be due to the fact that AUC only works with presence data and the background data of the models are considered absent (pseudo absence) thus inflating the maximum achievable AUC for rare species (thus low presence data) and reducing the maximum achievable AUC for the common species as mentioned by (Wiley E. O. 2003; Phillips, Anderson et al. 2006).

To counter this problem a Kappa analysis was done using the presence absence data from the herpetological atlas data (same data used to develop the species distribution models). Furthermore kappa is a more holistic evaluation technique because it asses both the commission and omission error rate and therefore avoids the model agreement by chance (Fielding and Bell 1997). As a result relatively common species such as *Salamandra salamamdra* and *Hyla meridionalis* had the highest kappa of 0.698 and 0.650 but *Rana perezi* still maintained a low kappa of 0.11.

The short coming of the aforementioned evaluation techniques is that they use the same data that was used to develop the model and they include false absence values from the models (pseudo absence). According to Anderson (2003) species distribution models based on only presence data have an over estimation of their pseudo absence due to the prevalence of background data. To counter this challenges an omission error rate was also done using independent presence data, although Anderson (2003) mentions that it is a necessary condition to evaluate a good model, because it measures the type I error of a model, but not sufficient because it only evaluates the presence occurrence of the model. According to the results (Table 3-2) *Pelobates cultripes* had the lowest omission error rate of 0.00 while *Alytes dickhellini* had the highest with 0.667 but on counter checking the number of independent presences points they were very low i.e. 4 and 3 respectively thus reducing the evaluation confidence of these species.

4.3. Land cover reclassification

The land cover data used for the research was obtained from the Environmental ministry of Andalusia Spain as mentioned earlier (section 2.2.3). It originally consisted of 44 land cover classes that were consolidated to 16 classes (as shown in Table 2-3and appendix 1) according to how various literature studies on amphibians have discriminated the land cover surface. For example to investigate the relationship of amphibian abundance to habitat features across spatial scales in the Boreal plains, Alberta, Canada, the land cover types used were 15 classes, for example, Coniferous forest, deciduous forest, mixed forest, shrubs, herbaceous, urban, agricultural areas, burnt areas and wetland (Browne, Paszkowski et al. 2009). This highly concurs with most of the land cover types used in the study. Another study example is Moreno-Rueda (2007) who investigated the influence of environmental heterogeneity to vertebrates (including amphibian) species richness, in Granada province, Spain, they used 11 land cover types that pertain to the land cover types used in the study.

Browne (2009) mentions that deciduous forest and mixed forest cover provide more understory vegetation for insect foraging and shade to avoid desiccation thus having more amphibians abundance than coniferous forest and shrub land. Atauri (2001) mentioned that in Madrid region more amphibian species are found in mesophytic vegetation such as broadleaved forest and eucalyptus forest while less species at xerophytic vegetation such as scrubland and non-irrigated crops.

4.3.1. Land cover Validation

The land cover type evaluation results were not very strong i.e. percentage overall accuracy of 64%, a kappa of 0.6 and a low R^2 (0.297) of the land cover diversity evaluation. This is may be due to the land cover and land use changes that have taken place in the study area over the past 10 years since the production of the land cover data in 1999. The contingency table (Table 4-1) show some of the significant land cover changes (highlighted in grey) that have taken place in the past ten years.

Land cover types		Field Observation												
		BaLa	GrLa	BuLa	BrFo	CoFo	WeLa	NoIrCr	IrCr	OlGr	BuIn	ExDu	Total	**User Accuracy %**
Data	BaLa	15	7	16	4	0	2	1	0	0	2	0	47	**31.91**
	GrLa	0	11	0	0	0	0	0	0	0	13	0	25	**44.00**
	BuLa	6	1	46	2	12	0	0	0	3	15	0	87	**52.87**
	BrFo	1	4	4	57	0	1	0	0	0	0	0	68	**83.82**
	CoFo	3	3	2	0	31	0	1	0	0	1	0	41	**75.61**
	WeLa	0	0	2	0	0	2	0	0	0	0	0	4	**50.00**
	NoIrCr	0	1	1	1	3	2	79	2	36	12	2	141	**56.03**
	IrCr	0	0	0	0	0	0	0	6	0	1	0	7	**85.71**
	OlGr	0	0	1	0	0	0	8	0	46	1	0	57	**80.70**
	BuIn	0	1	0	1	0	0	0	0	1	42	0	46	**91.30**
	ExDu	0	0	1	0	0	0	0	0	0	1	3	5	**60.00**
Total		25	28	73	65	46	7	89	8	86	88	5	**528**	
Producer Accuracy %		**60**	**39.29**	**63.01**	**87.69**	**67.39**	**28.57**	**88.76**	**75**	**53.49**	**47.73**	**60**		

Table 4-1 ***Contigency table; highlighting the land covers types that have undergone significant changes over the past 10 years, hence affecting the producer and user accuracy percentages.***
(BaLa = Bare land, GrLa = Grassland, BuLa = Bush land, BrFo = Broadleaved Forest, CoFo = Coniferous Forest, WeLa = Wetland, NoIrCr = Non Irrigated crops, IrCr = Irrigated crops, OlGr = Olive groves, BuIn = Buldings and Infrastructure, ExDu = Extractive sites and Dump sites)

Noticeable is a lot of bush land and grassland has been converted to buildings or infrastructure. This in turn reduces the user accuracy of the grassland and bush land cover types i.e. grassland 44 % and bush land 52.87 % but increases the user accuracy of buildings and infrastructure to 91.30 % and in turn reduces the producer accuracy of building and infrastructure to 47.73%. This is consistent with the high increase in road construction and urbanisation in Malaga, Spain during the construction boom over the past decade (Gonzalez 2007; Wikipedia 2009b).

Bare land and a substantial amount of bush land cover have been converted to broad leaved and coniferous forest plantation respectively. This in turn results to a low user accuracy of 52.87 % for bush land cover. Furthermore to a small extent, non-irrigated crops (mainly wheat) have been converted to coniferous forest plantation. This concurs with the nature rehabilitation strategies of the reforestation of coniferous and broadleaved forest in the past 3 decades. Moreover farmers through the Common Agricultural Policy (CAP), benefit from government subsidies by reforesting part of their farmland (*Ministerio de Medio Ambiente* 2006).

Due to natural dynamics and vegetation succession a substantial amount of bare land has changed to grassland and bush land. This in turn results to a low user accuracy of bare land (31.91 %) and a low producer accuracy of grassland (39.29 %).

Farmers in the past decade have changed their land use from non irrigated crops to olive groves (but less verse visa) and to smaller extent to buildings and infrastructure. These results to a low user accuracy for non-irrigated crops (56.3 %) and a low producer accuracy for olive groves (53.49 %).

This is consistent with the ever expansion of olive cultivation in Andalusia, devoting around 17% of its land to olive groves hence contributing 25% to the global agrarian production of olive oil (Gómez, Sobrinho et al. 2009).

In the light of the aforementioned changes that have occurred over the past decade, since the production of the land cover data to the validation period of its reclassified data, the kappa and overall accuracy and the R^2 of the land cover data may be better than what was obtained.

4.4. Relationship of amphibian species richness variation to Landscape heterogeneity

Recall that landscape heterogeneity was assessed using two measurable proxies, land cover diversity and altitude standard deviation, both calculated at a 1km spatial resolution (grain size) and related to the variation of amphibian species richness which is the summation of the 11 species occurrence (presence and absence).

4.4.1. Altitude range to Amphibian species richness variation

It is assumed that the greater the elevation variability of an area, the greater the spatial variability of its climate and therefore the more likely that the area will have larger number of habitats (Rodríguez, Belmontes et al. 2005) . Moreno-Rueda (2009) study in Spain mentions that amphibian species richness increases with Altitude range although flattens at high values of altitude range. On the contrary the study analysis shows there is no correlation between amphibian altitude standard deviation and amphibian species richness.

The grain size of 1 km^2 may still be too course to detect the relationship between the altitude range and amphibian species richness. Some of the studied amphibian species may have an operational scale of less than 1 km^2 due their pond dwelling nature thus hardly moving away from their pond proximity. Good examples are *Hyla meridionallis, Discoglossus jeanneae, Triturus pygmaeus*, and *Pleurodales waltl* which are pond dwelling and/or pond breeding amphibians (Pleguezuelos, Marquez et al. 2004).This species may in turn dilute the effect of altitude range to amphibian species richness.

4.4.2. Land cover diversity to Amphibian species richness variation

The greater the land cover diversity or number of habitats the more the natural resources available to satisfy the needs of different species which in turn reduces competition and favours species coexistence (Berg 1997; Farina 1997). When resources are limited, competition becomes more prevalent and dominant species tend to exclude others thus reducing the number of species (Pulliam 2000). On the contrary according to the study results there was no significant correlation between land cover diversity and species richness variation of amphibians. This agrees with similar studies done both in Spain and nearby Granada province, South East of Spain (Moreno-Rueda and Pizarro 2007; Moreno-Rueda and Pizarro 2009) in spite of having reduced the grain size of the study area to 1 km^2.

Atauri (2001) found a significant positive correlation between landscape heterogeneity and species richness of amphibians exists in Madrid region, Spain. He further mentioned that it depends with the land cover composition. Xerophytic habitats such as unirrigated crops and scrubland land covers had low amphibian species richness while more mesophytic habitats such as broad-leaved forest and Eucalyptus forest had high amphibian species richness. For Atauri (2001) case the study area had less xerophytic habitats.

On a critical look at land cover composition characteristics of the study area (shown in Table 4-2), and literature (Ibarra 2003) xerophyitic habitats such as Non Irrigated crops and bush land predominate and mesophytic and hydrophytic habitats such as Eucalyptus forest, wetlands and riverine vegetation are less prevalent. This in turn means that the study area is predominantly a water scarce environment thus limiting favourable areas where most amphibian species can survive due to their high water dependency nature and as a result diluting the effect of land cover diversity to amphibian species richness.

Land cover type	Area in km2
Non irrigated crops	2409.1507
Bushland	978.6278
Olive groves	970.7172
Broadleaved forest	823.0606
Coniferous forest	755.0118
Bareland	482.7823
Irrigated crops	389.7667
Building or Infrastructure	242.1902
Grassland	160.7711
Water reservoirs	26.6798
Extractive site or Dumpsite	18.4438
Mixed Forest	17.2457
Wetlands	14.0540
Riverine Vegetation	11.5095
Eucalyptus forest	8.3229

Table 4-2 ***The area covered by the different land cover types. Xerophytic habitat composition predominates the study area with non irrigated crops and bush land cover types being the most common***

4.5. Land cover type and amphibian species richness variation

The ANOVA results (Table 3-9) show that the species richness of amphibians differs among land cover types. Building and infrastructure cover type showing the lowest species richness (with a mean of 3.05) and broadleaved forest cover have the highest species richness (mean of 7.38) as shown on Table 3-6 and Figure 3-11.

According to Browne (2009) at landscape levels of 500 to 5000 meters resolution deciduous forest showed the strongest correlation with the abundance of the studied arunans. This concurs with the study result of the similar land cover of broad leaved forest (assuming that deciduous forest are mainly broadleaved) which has the highest species richness of amphibians.

Terrestrial invertebrate density is positively related to density of understory vegetation furthermore, broadleaved stands have greater understory than coniferous stands (Comet 1996; Ferguson and Berube 2004). Invertebrates are the primary source of food for amphibians therefore the density of understory vegetation increases the number of invertebrates which in turn increase the diversity of amphibians (Browne, Paszkowski et al. 2009). We found that the species richness of amphibians was intermediate in the coniferous forest cover. This may be due to the fact that the foraging quality for amphibians maybe low due to their scarce understory vegetation.

Olive groves may have a similar scarce amount of understory vegetation hence a significantly equal amount of amphibian species richness to coniferous forest at intermediate level.

The foraging quality for grassland cover is quite ideal due to the relatively high presence of flowering herbaceous plants that attract insects which are prey to amphibians. This might explain the very high species richness of amphibians in grassland cover.

Another scenario to consider is that habitats with higher understory offer protection from predation and desiccation especially during summer when temperatures can be well above 30^0C (Atauri and de Lucio 2001; Browne, Paszkowski et al. 2009). This may explain why multilayered canopy covers with abundant understory such as broadleaved and Eucalyptus forest have high to very high species richness of amphibians. As a matter of fact broadleaved forest offers more shed due to the large surface area of the leaves, thus relating to its highest accommodation of amphibian species according to the study.

Atauri (2001) mentions that xerophytic vegetation such as Olive groves and shrub land found in dry areas have low species richness while areas with mesophytic vegetation cover such as broadleaved forest, Eucalyptus forest and have high amphibian species richness due to the abundance of moisture. This concurs with our study findings where Broadleaved, Eucalyptus forest and riverine vegetation had a relatively high species richness of amphibians compared to the xerophytic land cover of bush land and Olive groves had low species richness.

Mixed forest and riverine vegetation had equally high amount of species richness indicates that they the structure and abundance of water for both cover types may be the same. On the other hand dumpsite and extractive sites also have a significantly equal amount of amphibian species despite having a different structure. This cover may be predominated by species such as *Triturus pygmaeus* that thrive in abandoned quarries and mines and those adapted to temporal/seasonal ponds, such as *Bufo calamita* and *Bufo bufo*, that form in these excavated sites during the rainy seasons (Pleguezuelos, Marquez et al. 2004).

Fahirig (1995) mentions that urban areas have high traffic volumes which are a serious threat to the amphibian population. Further more natural habitats are altered to concrete slabs which reduce the amphibian population (Pleguezuelos, Marquez et al. 2004). This explains why buildings and infrastructure covers- that are predominantly in urban areas, have the lowest amphibian species.

Surprisingly the wetland land cover type hosts relatively low species richness of amphibians despite the fact that it is a water abundant ecosystem. This concurs with (Browne, Paszkowski et al. 2009) study on amphibian abundance. Browne (2009) mentioned that amphibian abundance may be limited by the pond condition and the amount of suitable terrestrial habitat for foraging and hibernation. On critically reviewing the land cover map it was confirmed that one large wetland (Laguna de Fuente de Piedra) has high salinity which is not tolerated by amphibians (Adler 2004), although *Rana perezi* can exist in even saline water conditions (Pleguezuelos, Marquez et al. 2004). This in turn can also explain why wetland cover has the one of the highest standard deviations (see Table 3-6), with species richness being very low in and around the saline wetland and moderately high in most of the other “assumed” freshwater wetlands (see also appendix 6). Furthermore the wetlands are mainly surrounded by olive and non irrigated crops which are unfavourable terrestrial habitats for foraging and hibernation due to lack of permanent understory vegetation and litter.

Lake and water reservoirs show low species richness according to the results. This may be due to the water depth of these large water bodies which discourage amphibians from breeding. Shallower water bodies tend to be warmer during the day and warm temperatures facilitate growth of tadpoles(Moran 1985; Collins and Storfer 2003). Furthermore most lakes and water reservoirs consist of fish which are the prime predators of amphibians because they feed on their larva (tadpoles) and eggs (Denoël and Lehmann 2006).

Humanised surfaces such as agricultural fields show an intermediate amount of species richness for both non irrigated crops and Olive groves. This agrees with Moreno-Rueda (2009) findings that at intermediate values of humanised surfaces, primarily at cropland areas, amphibian species richness positively correlates. This may be explained by the fact that high productivity of the land attracts both wildlife species and agricultural activities and creation of irrigation pools may favour amphibians (Moreno-Rueda and Pizarro 2009). Over the past few decades olive groves have been irrigated in order to boost it production during dry seasons ((*Ministerio de Medio Ambiente* 2006) and field observation). This irrigation pools may attract amphibian’s species which are water loving animals. Ironically irrigated crops have low species richness. This may be due to the condition of the irrigation water that may contain high amounts of fertilizer or other chemical components used to improve the crop production. This is a fact yet to be established.

4.6. Primary Variables that determine Amphibian species richness variation

The importance of environmental variables to faunal species varies geographically depending on the condition of the region (Hawkins, Field et al. 2003). Water variables are primary determinants in species richness variation in the tropics subtropics and up to warm temperate zones such as the Mediterranean region (Hawkins, Field et al. 2003). In the Mediterranean region, energy is less important in determining species richness because it is not a limitation factor. In the higher latitudes where energy availability is low such as Northern Europe, climate (in this case ambient energy e.g. temperature) becomes a constraint and important in determining species richness geographic variation (Whittaker 2001; Hawkins, Field et al. 2003).

Another important condition to consider is the study extent or scale. Climate (as energy and water/energy variables) is more homogeneous at smaller scales hence less important than landscape heterogeneity in determining the variation in species richness (Böhning-Gaese 1997; Moreno-Rueda and Pizarro 2009). On the other hand climate supersedes landscape heterogeneity at larger scales such as continental and global scales due to its pronounced variation at such extents (Hawkins, Field et al. 2003; Rodríguez, Belmontes et al. 2005).

According to this the study analysis, performed in the Mediterranean region, at a local scale extent, the variation in amphibian species richness is still primarily determined by climatic variables which even supersede the landscape heterogeneity variables (land cover diversity and altitude standard deviation). With regression model fit of 0.214 (R^2), temperature in the hottest month (July) and Actual evapo-transpiration (AET) are the highest determinants of amphibian species richness with correlation slope of -0.290 and 0.269 respectively (see Table 3-12). This concurs with studies carried out at a continental scale for example in Europe and China (Rodríguez, Belmontes et al. 2005; Qian, Wang et al. 2007) where water/energy variables (measured as actual evapotranspiration) are the primary determinants of amphibian species variation. Furthermore at even a similar scale in South East Spain (Moreno-Rueda and Pizarro 2007) precipitation superseded other variables including landscape heterogeneity variables.

The importance of environmental variables can also depend on the group of species under consideration (Atauri and de Lucio 2001; Hawkins, Field et al. 2003; Moreno-Rueda and Pizarro 2007). On a local scale extent endotherms are strongly influenced by the habitat structure but at this scale, ectothermic species such as reptiles and amphibians are still influenced by climate (Moreno-Rueda and Pizarro 2007). Ectotherms are primarily influenced by climate conditions; even at small scales due to their physiological nature (Qian, Wang et al. 2007) i.e. they entirely depend on their environmental heat to maintain homeostasis (metabolism within their body).

Further down the ectothermic animal groups, water variables explain more variance in amphibians' species richness than in reptile species richness while energy variables are important for the solar ectotherms, such as reptile species (Rodríguez, Belmontes et al. 2005; Qian, Wang et al. 2007). According to Qian (2007) water variables explain 50% of amphibian species richness and Rodríguez (2005) found that AET had the highest positive correlation with amphibian species richness. This concurs with the study results where AET is the one of the strongest determinant of amphibian species richness variation with a positive correlation of 0.269. Amphibians usually require water for reproduction (their eggs and larvae require water to survive). Adults require environmental moisture and cooler temperatures because they have sensitive moist skin that makes them vulnerable to desiccation in hot and dry environments. Therefore measures of atmospheric water and energy condition describe amphibian species richness gradient best (Rodríguez, Belmontes et al. 2005; Qian, Wang et al. 2007).

In spring amphibians require heat from the sun to warm up and become active hence explaining the positive correlation of amphibian species richness to mean annual temperature in some studies (Moreno-Rueda and Pizarro 2009). On the contrary during mid summer, when the temperatures in the Mediterenean region are high sometimes exceeding 30^0C, amphibians seek for cooler regions to avoid desiccation or overheating. This in turn explains why mid-summer (July) temperature is a strong primary determinant, with a negative correlation (-0.290), to amphibian species richness. During the mid-summer ambient energy may be over abundant at the Mediterranean region which can be overwhelming for the amphibian species that has sensitive moist skin which is prone to desiccation.

According to the results (Table 3-12), slope has also a significant influence on amphibian species richness variation, ranked third according to the stepwise multiple regression with a negative correlation with a slope of -0.262. This concurs with Dayton (2006) study on habitat suitability for desert amphibians in Big Ben National Park, USA, where flat areas were regarded as high likelihood of amphibian presence while steep slopes were regarded as low likelihood areas. The steepness of a region significantly influences water runoff. Flat regions provide more opportunities for water accumulation hence formation of permanent or temporal ponds required for amphibian breeding unlike areas with steeper slopes. In the Mediterranean region water bodies in the form of ponds, especially during summer, are limited due to semi arid condition. Furthermore the Iberian Peninsula is a mountainous region with water runoff from highlands flowing to lowlands or intermediary plateaus where water accumulates into temporal or permanent ponds (Moreno-Rueda and Pizarro 2007). This in turn may explain why slope has a negative association with amphibian species richness.

The slope analogy to water availability has also been emphasised by Moreno-Rueda (2009). He mentions that precipitation is not tightly related to water availability due to the mountainous nature of the Iberian Peninsula. It might rain in the steep slope highlands while water accumulates in the flat lowlands. Therefore due to the fact that water is a constraint in the Mediterranean region (Hawkins, Field et al. 2003) amphibian's species may have adapted to living in flat low lying regions where ponds can form, thus excluding their high dependency on rainfall as a source of water.

Human population, measured as population density per km^2 is another factor that significantly influences the distribution of amphibian species richness with a negative correlation of -0.100. This is in agreement with other studies which explain that people may negatively affect species richness distribution due to their harmful activities to species, such as urbanisation and infrastructure (Real, Barbosa et al. 2003). On the contrary a positive correlation might also be found due to the high primary productivity which attracts both faunal species and human activities through agriculture (Gaston and Evans 2004; Moreno-Rueda and Pizarro 2009).

The sign of the relationship between human population and amphibian species richness can differ depending on the grain size in which the study was done, with a positive correlation at large grain size and a negative correlation at smaller grain size. For the study case there was a negative correlation because the negative impacts of human presence can be detected at a small grain size of $1km^2$ (Pautasso 2007).

4.7. Uncertainties and Limitations

4.7.1. Uncertainties

Amphibian Species richness concept

The concept of utilizing species richness as an indicator of amphibian diversity, despite being adapted by various journals (Atauri and de Lucio 2001; Moreno-Rueda and Pizarro 2007; Moreno-Rueda and Pizarro 2009), may not be very holistic because it ignores a very important component which is the number of individuals per species (abundance). Shannon and simpson indices are more robust indicators of diversity because they factor in the abundance of each species. These indices can be more intuitive in explaining the diversity of amphibians; unfortunately data on the abundance of species was not available.

The scale perception: A conflict of interest

According to Browne (2009) the operational scale of amphibians may vary from their breeding sites, depending on the availability of resources and also the type of species. Browne (2009)study mentions that the chorus frog (*Lithobates sylvaticus*) responded most strongly to environmental variables at 1000 meters while the west toad (*Anaxyrus boreas*) at 100 meters. For our study case we only focused on a grain size of 1 km^2, which was presumed to be the ideal operational scale for all the amphibian species. 1 km2 may still be a course scale for some of the amphibians used in the study, especially the pond dwelling species such as the Iberian newts, *Pleurodales waltl* and *Triturus pygmaeus* which only utilize the adjacent terrestrial habitats for foraging (Pleguezuelos, Marquez et al. 2004).

On the other hand the principle behind altitude standard deviation as an indicator of the ecosystem diversity (as mentioned in section 4.4.1), may be diluted by the fact that the study focuses on a relatively small grain size of $1km^2$. At a larger sample size the variation in climate may be higher hence; more elaborate ecosystem diversity may be evident.

Land cover diversity indices: The missing link

Land cover diversity, calculated with Shannon diversity index, does not provide information on the land cover types used for the calculation (McGarigal and Marks 1995). This in turn obscures the study from vital information about the land cover composition i.e. whether the index was calculated predominantly in xerophytic, mesophytic or hydrophytic type of vegetation or whether the index was calculated predominantly in natural land cover (e.g. broadleaved forest) or land use (e.g. buildings or infrastructure), that may be underlying factors that determine the species richness of amphibians.

The course $100km^2$ presence data of amphibians

The 100km2 presence data used to develop amphibian species richness data was too course. The large area covered per grid cell increases the uncertainty of the exact position of XY presence points where the different amphibian species were actually found, during the survey. This may in turn reduce the predictive power of the niche based species distribution models, and as a result affect robustness of the derived amphibian species richness data. This may in turn lead to the very low R^2 (a lot of noise in the scatter plot) obtained when carrying out the correlation regression analysis with the landscape heterogeneity variables.

4.7.2. Limitations

The first and foremost limitation is that, the land cover data does not discriminate the wetland types into saline and freshwater wetlands. This is an important discriminatory factor when it comes to amphibian habitat preference. This limitation is also reflected on the results (Table 3-6 and Figure 3-11) where the wetland cover had a very low amphibian species richness and high standard deviation probably due to the saline and freshwater conditions of the different wetlands.

There was a limitation on the availability of independent data for model validation, using the omission error rate. Rare/specialised species such as *Alytes dickhellini* and *Pelobates cultripes* had very few presence data (3 and 4 points respectively) which in turn resulted to exaggerated omission rates of lows 0.00 (for *Pelobates cultripes*) to highs of 0.667(for *Alytes dickhellini*).

As mentioned in section 4.4.2, the study area was predominated by xerophytic habitats which repulse the existence of amphibian species. Therefore, the relationship between amphibian species richness and landscape heterogeneity maybe nonexistent due to the scarcity of water abundant habitats.

When carrying out the stepwise regression analysis only continuous variables can be considered leaving out the analysis on the relative importance of categorical data – in the study case classified NDVI (see Table 3-11).

5. Conclusion

In summary amphibian species richness variation is mainly determined by the climatic condition of the environment which supersedes other determinants including landscape heterogeneity due to their physiological nature (they have moist sensitive skin) and habitat requirements (they require both terrestrial and aquatic habitats for foraging and breeding).

Secondly, amphibian species feel more at home in ecosystems with tree canopies which offer shade for protection from desiccation, that have a good amount of under story vegetation for foraging and ample amount of moisture required for their survival and reproduction. Therefore broadleaved forest was found to be an ideal ecosystem that accommodated the highest amount of amphibian species in the study area. On the other hand human presence such as urbanization and infrastructure are a treat to the amphibian population. Therefore building and infrastructure land cover was found to have the lowest amount of amphibian species richness.

The main objective of the study was to determine whether there is a relationship between landscape heterogeneity (altitude range and land cover diversity) and species richness of amphibians at 1 km^2 resolution in Malaga province, Spain. With respect to this the following conclusions were drawn:

- There is no relationship between amphibian species richness and altitude range (measured as standard deviation) at a spatial resolution of $1km^2$.
- There is no relationship between amphibian species richness and land cover diversity (measured with Shannon diversity index) at a spatial resolution of $1km^2$
- Amphibian species richness is not equal among the different land cover types. Building and Infrastructure land cover have the lowest amount amphibian species with a mean of 3 species while Broadleaved forest land cover had the highest with a mean of 7 species.
- Altitude range is not an important determinant on the variation of amphibian species richness
- Land cover diversity is not an important determinant on the variation of amphibian species richness.

6. Recommendation

There are other important underlying factors that are vital to the distribution of amphibian species richness that must be considered before investigating its relationship to the habitat structure (for the study case, landscape heterogeneity), a major one being water or moisture availability. All the research studies that found a relationship between the spatial distribution of species richness and abundance of amphibian pre-empt the constraint of water. For example Browne (2009) only looked at the relationship of amphibian abundance to the habitat structure at the proximity of the pond breeding sites. Atuari (2001) could only find a positive relationship of amphibian species richness to land cover diversity at mesophytic and hydrophytic habitats. This is also re-emphasised in the study, where climatic components such as evapotranspiration and temperature which express the energy/water balance and energy of the environment respectively were of higher priority to the distribution of amphibian species richness than the landscape heterogeneity variables, altitude range and land cover diversity.

One of the shortcomings that the study experienced was the acquisition of an ideal land cover data that is classified into the appropriate land cover types according to an amphibian perception. One of the important divisions of classes that the land cover data should have is the wetland types, such as salt marshes and freshwater marshes. This is an important discriminatory factor that should be put into consideration prior to further analysis, such as relating land cover types to the geographic variation of amphibian species richness, in future studies.

It is recommended to use XY presence point data or a finer resolution presence data for the development of the niche based species distribution models for each species so as to obtain a more robust amphibian species richness data. For the study a course resolution occurrence data (10 by 10 km herpetological atlas data) was preferred to develop the models due to the high prevalence of presence data for all 11 amphibian species across the Andalusia region.

The abundance of species is an important component in elaborating the diversity of species. The study was only limited to using the richness index as an indicator of amphibian diversity due to lack of data on the abundance per species. Therefore it is recommended that data on the abundance of each species be acquired to develop a more robust index of amphibian diversity for future studies.

On a biodiversity perception, specialist species prefer continuous homogeneous habitats while generalist species can exist in highly habitat diverse landscapes (Andren 1994; Edenius and Sjoberg 1997). Therefore for future studies pertaining to the habitat structure of amphibians it is recommended to analyse the generalist and the specialist species separately.

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Appendices

Appendix 1: Land cover reclassification Table

English version of original land cover types	Reclassified land cover types	Code
Burnt Areas	Bareland	1
Sparsely vegetated areas	Bareland	1
Dunes and beaches	Bareland	1
Bare rocks	Bareland	1
Pastureland	Grassland	2
Green leisure places	Grassland	2
Dense bushland	Bushland	3
Sparse bushland	Bushland	3
Eucalyptus forest with bushes	Eucalyptus forest	4
Dense Eucalyptus forest	Eucalyptus forest	4
Hardwood or Oak forest with bushes	Broadleaved forest	5
Oakwood forest with herbecious vegetation	Broadleaved forest	5
Oakwood forest with bushes	Broadleaved forest	5
Dense Oakwood forest	Broadleaved forest	5
Coniferous forest with bushes	Coniferous forest	6
Dense Coniferous forest	Coniferous forest	6
Dense mixed forest	Mixed Forest	7
Mixed forest with herbecious vegetation	Mixed Forest	7
Raparian Vegetation	Riverine Vegetation	8
Rivers and streams	Riverine Vegetation	8
Sea	Sea	9
Wetlands	Wetlands	10
Water reserviors	Water reservoirs	11
Crops with natural vegetation spaces	Non irrigated crops	12
Non irrigated crops herbecious	Non irrigated crops	12
Mosaic of Non irrigated with irrigated crops	Non irrigated crops	12
Herbous and wooded non irrigated crops	Non irrigated crops	12
Irrigated crops herbecious	Irrigated crops	13
Irrigated crops wooden	Irrigated crops	13
Greenhouse crops	Irrigated crops	13
Vineyards	Irrigated crops	13
Olives	Olive grooves	14
Industrial service areas	Building or Infrastructure	15
Urban and residential areas	Building or Infrastructure	15
Roads and railways	Building or Infrastructure	15
Airports	Building or Infrastructure	15
Construction sites	Building or Infrastructure	15
Sea ports	Building or Infrastructure	15
Extractive sites	Extractive site or Dumpsite	16
Dumpsites	Extractive site or Dumpsite	16

Appendix 2:

Field Data Recording Sheet Sample

At each systematic sampling point an imaginary 5 meter plot was created to observe and record the structure and composition of the ground vegetation (0.5 meters and below e.g. grass), 30 meters for mid story vegetation (0.5 to 3 meters e.g. bushes) and 40 meters for upper story vegetation (above 3 meters e.g. trees). Quickbird 2004 orthophotos displayed in a hand held GPS enabled Ipaq were used as support material to located the points and classify the land cover type.

Date	Area	Sampling Code	Species Rich	Rand Sampling No	Sampling Point	Trees%	Bush%	Herb%	Bare%/Type	Euca%	Brod%	Coni%	Waterbody	Agriculture	Other activities	Remarks	Picture	Landcover type
22/09/2009	Sierra de los Pinos	301224	3	1	224	65	30	20	80/litter	0	65	0	0	0	0		123	Broadleaved Forest
22/09/2009	Sierra de los Pinos	301223	3	1	223	40	30	10	90/rock	0	40	0	0	0	0		124	Broadleaved Forest
22/09/2009	Sierra de los Pinos	301228	3	1	228	5	80	50	50/soil+rock	0	5	0	0	0	0		125	Bushland
22/09/2009	Sierra de los Pinos	301231	3	1	231	0	0	70	30/soil+rock	0	0	0	0	0	0		126	Grassland
22/09/2009	Sierra de los Pinos	301232	3	1	232	35	60	20	80/litter	0	35	0	0	0	0	Red Oak	127	Broadleaved Forest
22/09/2009	Sierra de los Pinos	301230	3	1	230	40	40	60	40/soil	0	40	0	0	0	0	Red Oak	128	Broadleaved Forest
22/09/2009	Sierra de los Pinos	301229	3	1	229	80	20	80	40/soil	0	80	0	0	0	0	Red Oak	129	Broadleaved Forest
22/09/2009	Sierra de los Pinos	301227	3	1	227	50	20	70	30/soil	0	50	0	0	0	0	Red Oak	130	Broadleaved Forest
22/09/2009	Sierra de los Pinos	301226	3	1	226	60	30	60	40/soil	0	30	30	0	0	0		131	Broadleaved Forest
22/09/2009	Sierra de los Pinos	301225	3	1	225	70	35	20	80/litter	0	70	0	0	0	0	Red Oak	132	Broadleaved Forest
22/09/2009	Sierra de los Pinos	301222	3	1	222	30	20	30	70/litter	0	15	15	0	0	0		133	Mixed Forest
22/09/2009	Sierra de los Pinos	301221	3	1	221	75	25	60	40/litter	0	75	0	0	0	0	Red Oak	134	Broadleaved Forest
22/09/2009	Sierra de los Pinos	301220	3	1	220	20	5	80	20/soil	0	20	0	0	0	0	Red Oak	135	Broadleaved Forest
22/09/2009	Sierra de los Pinos	301219	3	1	219	65	30	20	80/litter	0	65	0	0	0	0	Red Oak	136	Broadleaved Forest
22/09/2009	Sierra de los Pinos	301218	3	1	218	20	0	30	70/soil		10	10	0	0	0		137	Broadleaved Forest
22/09/2009	Sierra de los Pinos	301217	3	1	217	65	20	40	60/soil	0	65	0	0	0	0	Red Oak	138	Broadleaved Forest
22/09/2009	Sierra de los Pinos	302206	3	2	206	0	0	80	20/soil	0	0	0	0	0	0		139	Grassland

Appendix 3:
2nd run results of Simple linear regression model summary of Amphibian species richness to:

1. Altitude Standard deviation

Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.093[a]	.009	.006	2.176

a. Predictors: (Constant), Altitude SD

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
1	(Constant)	5.478	.212		25.861	.000
	Altitude SD	.007	.004	.093	1.720	.086

a. Dependent Variable: SpRich

2. Land cover diversity (SHDI)

Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.014[a]	.000	-.003	2.185

a. Predictors: (Constant), SHDI

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
1	(Constant)	5.735	.210		27.328	.000
	SHDI	.082	.307	.014	.266	.790

a. Dependent Variable: SpRich

Appendix 4:
Games howels test of pair wise comparison of land cover type's amphibian species richness

Multiple Comparisons

Species Richness

Games-Howell

(I) Landcover type	(J) Landcover type	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval Lower Bound	95% Confidence Interval Upper Bound
Bareland	Grassland	-1.256	.365	.070	-2.56	.05
	Bushland	.898*	.254	.036	.03	1.77
	Eucalyptus forest	-.746	.367	.766	-2.09	.60
	Broadleaved forest	-1.330*	.223	.000	-2.10	-.56
	Coniferous forest	.192	.242	1.000	-.64	1.02
	Mixed forest	-.146	.501	1.000	-2.02	1.73
	Riverine vegetation	-.396	.450	1.000	-2.07	1.28
	Wetland	2.754*	.594	.008	.51	5.00
	Lake	1.141	.583	.804	-1.03	3.31
	Non Irrigated crops	-.072	.199	1.000	-.76	.61
	Irrigated crops	.697	.350	.800	-.52	1.91
	Olive groves	.328	.231	.985	-.46	1.12
	Urban or Infrastructure	3.002*	.340	.000	1.80	4.20
	Extractive or Dumpsites	-.196	.362	1.000	-1.52	1.13
Grassland	Bareland	1.256	.365	.070	-.05	2.56
	Bushland	2.154*	.367	.000	.84	3.46
	Eucalyptus forest	.510	.452	.998	-1.11	2.13
	Broadleaved forest	-.074	.345	1.000	-1.32	1.18
	Coniferous forest	1.448*	.358	.015	.16	2.73
	Mixed forest	1.110	.566	.806	-.95	3.17
	Riverine vegetation	.860	.522	.936	-1.03	2.75
	Wetland	4.010*	.650	.000	1.62	6.40
	Lake	2.397*	.640	.038	.07	4.72
	Non Irrigated crops	1.184	.330	.060	-.03	2.39
	Irrigated crops	1.953*	.439	.002	.42	3.49

	Olive groves	1.584*	.350	.004	.32	2.85
	Urban or Infrastructure	4.258*	.430	.000	2.74	5.78
	Extractive or Dumpsites	1.060	.448	.549	-.54	2.67
Bushland	Bareland	-.898*	.254	.036	-1.77	-.03
	Grassland	-2.154*	.367	.000	-3.46	-.84
	Eucalyptus forest	-1.644*	.369	.007	-2.99	-.29
	Broadleaved forest	-2.228*	.226	.000	-3.00	-1.45
	Coniferous forest	-.706	.245	.209	-1.54	.13
	Mixed forest	-1.044	.502	.736	-2.92	.84
	Riverine vegetation	-1.294	.452	.274	-2.97	.39
	Wetland	1.856	.596	.183	-.39	4.11
	Lake	.243	.584	1.000	-1.93	2.42
	Non Irrigated crops	-.970*	.202	.000	-1.66	-.28
	Irrigated crops	-.201	.352	1.000	-1.42	1.02
	Olive groves	-.570	.234	.488	-1.37	.23
	Urban or Infrastructure	2.104*	.342	.000	.90	3.31
	Extractive or Dumpsites	-1.094	.364	.203	-2.42	.24
Eucalyptus forest	Bareland	.746	.367	.766	-.60	2.09
	Grassland	-.510	.452	.998	-2.13	1.11
	Bushland	1.644*	.369	.007	.29	2.99
	Broadleaved forest	-.584	.348	.922	-1.88	.71
	Coniferous forest	.937	.361	.408	-.39	2.27
	Mixed forest	.600	.568	.999	-1.47	2.67
	Riverine vegetation	.350	.524	1.000	-1.55	2.25
	Wetland	3.500*	.652	.001	1.10	5.90
	Lake	1.887	.641	.227	-.45	4.22
	Non Irrigated crops	.673	.333	.767	-.59	1.94
	Irrigated crops	1.443	.441	.100	-.12	3.01
	Olive groves	1.073	.353	.200	-.24	2.38
	Urban or Infrastructure	3.747*	.432	.000	2.20	5.29

	Extractive or Dumpsites	.550	.450	.995	-1.08	2.18
Broadleaved forest	Bareland	1.330*	.223	.000	.56	2.10
	Grassland	.074	.345	1.000	-1.18	1.32
	Bushland	2.228*	.226	.000	1.45	3.00
	Eucalyptus forest	.584	.348	.922	-.71	1.88
	Coniferous forest	1.521*	.212	.000	.80	2.25
	Mixed forest	1.184	.487	.518	-.66	3.03
	Riverine vegetation	.934	.435	.694	-.71	2.57
	Wetland	4.084*	.583	.000	1.86	6.31
	Lake	2.471*	.571	.014	.33	4.61
	Non Irrigated crops	1.258*	.161	.000	.71	1.81
	Irrigated crops	2.027*	.330	.000	.88	3.18
	Olive groves	1.657*	.199	.000	.98	2.34
	Urban or Infrastructure	4.331*	.319	.000	3.20	5.47
	Extractive or Dumpsites	1.134	.343	.120	-.14	2.41
Coniferous forest	Bareland	-.192	.242	1.000	-1.02	.64
	Grassland	-1.448*	.358	.015	-2.73	-.16
	Bushland	.706	.245	.209	-.13	1.54
	Eucalyptus forest	-.937	.361	.408	-2.27	.39
	Broadleaved forest	-1.521*	.212	.000	-2.25	-.80
	Mixed forest	-.337	.496	1.000	-2.20	1.53
	Riverine vegetation	-.587	.445	.988	-2.25	1.08
	Wetland	2.563*	.590	.015	.33	4.80
	Lake	.950	.579	.933	-1.21	3.11
	Non Irrigated crops	-.264	.186	.985	-.90	.37
	Irrigated crops	.506	.343	.978	-.69	1.70
	Olive groves	.136	.220	1.000	-.62	.89
	Urban or Infrastructure	2.810*	.333	.000	1.64	3.98
	Extractive or Dumpsites	-.387	.355	.998	-1.69	.92
Mixed forest	Bareland	.146	.501	1.000	-1.73	2.02

	Grassland	-1.110	.566	.806	-3.17	.95
	Bushland	1.044	.502	.736	-.84	2.92
	Eucalyptus forest	-.600	.568	.999	-2.67	1.47
	Broadleaved forest	-1.184	.487	.518	-3.03	.66
	Coniferous forest	.337	.496	1.000	-1.53	2.20
	Riverine vegetation	-.250	.624	1.000	-2.51	2.01
	Wetland	2.900*	.735	.023	.23	5.57
	Lake	1.287	.726	.895	-1.33	3.91
	Non Irrigated crops	.073	.476	1.000	-1.75	1.90
	Irrigated crops	.843	.557	.966	-1.18	2.87
	Olive groves	.473	.490	.999	-1.38	2.33
	Urban or Infrastructure	3.147*	.550	.000	1.14	5.16
	Extractive or Dumpsites	-.050	.564	1.000	-2.11	2.01
Riverine vegetation	Bareland	.396	.450	1.000	-1.28	2.07
	Grassland	-.860	.522	.936	-2.75	1.03
	Bushland	1.294	.452	.274	-.39	2.97
	Eucalyptus forest	-.350	.524	1.000	-2.25	1.55
	Broadleaved forest	-.934	.435	.694	-2.57	.71
	Coniferous forest	.587	.445	.988	-1.08	2.25
	Mixed forest	.250	.624	1.000	-2.01	2.51
	Wetland	3.150*	.702	.006	.59	5.71
	Lake	1.537	.692	.649	-.96	4.04
	Non Irrigated crops	.323	.423	1.000	-1.29	1.94
	Irrigated crops	1.093	.512	.705	-.75	2.94
	Olive groves	.723	.439	.929	-.93	2.37
	Urban or Infrastructure	3.397*	.505	.000	1.57	5.23
	Extractive or Dumpsites	.200	.520	1.000	-1.69	2.09
Wetland	Bareland	-2.754*	.594	.008	-5.00	-.51
	Grassland	-4.010*	.650	.000	-6.40	-1.62
	Bushland	-1.856	.596	.183	-4.11	.39
	Eucalyptus forest	-3.500*	.652	.001	-5.90	-1.10

	Broadleaved forest	-4.084*	.583	.000	-6.31	-1.86
	Coniferous forest	-2.563*	.590	.015	-4.80	-.33
	Mixed forest	-2.900*	.735	.023	-5.57	-.23
	Riverine vegetation	-3.150*	.702	.006	-5.71	-.59
	Lake	-1.613	.793	.768	-4.47	1.25
	Non Irrigated crops	-2.827*	.574	.005	-5.03	-.62
	Irrigated crops	-2.057	.642	.141	-4.42	.31
	Olive groves	-2.427*	.586	.024	-4.65	-.20
	Urban or Infrastructure	.247	.637	1.000	-2.10	2.60
	Extractive or Dumpsites	-2.950*	.649	.006	-5.34	-.56
Lake	Bareland	-1.141	.583	.804	-3.31	1.03
	Grassland	-2.397*	.640	.038	-4.72	-.07
	Bushland	-.243	.584	1.000	-2.42	1.93
	Eucalyptus forest	-1.887	.641	.227	-4.22	.45
	Broadleaved forest	-2.471*	.571	.014	-4.61	-.33
	Coniferous forest	-.950	.579	.933	-3.11	1.21
	Mixed forest	-1.287	.726	.895	-3.91	1.33
	Riverine vegetation	-1.537	.692	.649	-4.04	.96
	Wetland	1.613	.793	.768	-1.25	4.47
	Non Irrigated crops	-1.214	.562	.687	-3.33	.91
	Irrigated crops	-.444	.632	1.000	-2.74	1.85
	Olive groves	-.814	.574	.977	-2.96	1.33
	Urban or Infrastructure	1.860	.626	.215	-.42	4.14
	Extractive or Dumpsites	-1.337	.638	.729	-3.66	.99
Non Irrigated crops	Bareland	.072	.199	1.000	-.61	.76
	Grassland	-1.184	.330	.060	-2.39	.03
	Bushland	.970*	.202	.000	.28	1.66
	Eucalyptus forest	-.673	.333	.767	-1.94	.59
	Broadleaved forest	-1.258*	.161	.000	-1.81	-.71
	Coniferous forest	.264	.186	.985	-.37	.90
	Mixed forest	-.073	.476	1.000	-1.90	1.75

	Riverine vegetation	-.323	.423	1.000	-1.94	1.29
	Wetland	2.827*	.574	.005	.62	5.03
	Lake	1.214	.562	.687	-.91	3.33
	Irrigated crops	.769	.314	.489	-.33	1.87
	Olive groves	.400	.171	.566	-.19	.99
	Urban or Infrastructure	3.074*	.303	.000	1.99	4.16
	Extractive or Dumpsites	-.123	.327	1.000	-1.37	1.12
Irrigated crops	Bareland	-.697	.350	.800	-1.91	.52
	Grassland	-1.953*	.439	.002	-3.49	-.42
	Bushland	.201	.352	1.000	-1.02	1.42
	Eucalyptus forest	-1.443	.441	.100	-3.01	.12
	Broadleaved forest	-2.027*	.330	.000	-3.18	-.88
	Coniferous forest	-.506	.343	.978	-1.70	.69
	Mixed forest	-.843	.557	.966	-2.87	1.18
	Riverine vegetation	-1.093	.512	.705	-2.94	.75
	Wetland	2.057	.642	.141	-.31	4.42
	Lake	.444	.632	1.000	-1.85	2.74
	Non Irrigated crops	-.769	.314	.489	-1.87	.33
	Olive groves	-.370	.336	.999	-1.54	.80
	Urban or Infrastructure	2.305*	.418	.000	.85	3.76
	Extractive or Dumpsites	-.893	.436	.763	-2.44	.65
Olive groves	Bareland	-.328	.231	.985	-1.12	.46
	Grassland	-1.584*	.350	.004	-2.85	-.32
	Bushland	.570	.234	.488	-.23	1.37
	Eucalyptus forest	-1.073	.353	.200	-2.38	.24
	Broadleaved forest	-1.657*	.199	.000	-2.34	-.98
	Coniferous forest	-.136	.220	1.000	-.89	.62
	Mixed forest	-.473	.490	.999	-2.33	1.38
	Riverine vegetation	-.723	.439	.929	-2.37	.93
	Wetland	2.427*	.586	.024	.20	4.65
	Lake	.814	.574	.977	-1.33	2.96

	Non Irrigated crops	-.400	.171	.566	-.99	.19
	Irrigated crops	.370	.336	.999	-.80	1.54
	Urban or Infrastructure	2.674*	.325	.000	1.52	3.82
	Extractive or Dumpsites	-.523	.348	.965	-1.81	.77
Urban or Infrastructure	Bareland	-3.002*	.340	.000	-4.20	-1.80
	Grassland	-4.258*	.430	.000	-5.78	-2.74
	Bushland	-2.104*	.342	.000	-3.31	-.90
	Eucalyptus forest	-3.747*	.432	.000	-5.29	-2.20
	Broadleaved forest	-4.331*	.319	.000	-5.47	-3.20
	Coniferous forest	-2.810*	.333	.000	-3.98	-1.64
	Mixed forest	-3.147*	.550	.000	-5.16	-1.14
	Riverine vegetation	-3.397*	.505	.000	-5.23	-1.57
	Wetland	-.247	.637	1.000	-2.60	2.10
	Lake	-1.860	.626	.215	-4.14	.42
	Non Irrigated crops	-3.074*	.303	.000	-4.16	-1.99
	Irrigated crops	-2.305*	.418	.000	-3.76	-.85
	Olive groves	-2.674*	.325	.000	-3.82	-1.52
	Extractive or Dumpsites	-3.197*	.428	.000	-4.73	-1.67
Extractive or Dumpsites	Bareland	.196	.362	1.000	-1.13	1.52
	Grassland	-1.060	.448	.549	-2.67	.54
	Bushland	1.094	.364	.203	-.24	2.42
	Eucalyptus forest	-.550	.450	.995	-2.18	1.08
	Broadleaved forest	-1.134	.343	.120	-2.41	.14
	Coniferous forest	.387	.355	.998	-.92	1.69
	Mixed forest	.050	.564	1.000	-2.01	2.11
	Riverine vegetation	-.200	.520	1.000	-2.09	1.69
	Wetland	2.950*	.649	.006	.56	5.34
	Lake	1.337	.638	.729	-.99	3.66
	Non Irrigated crops	.123	.327	1.000	-1.12	1.37
	Irrigated crops	.893	.436	.763	-.65	2.44
	Olive groves	.523	.348	.965	-.77	1.81

	Urban or Infrastructure	3.197*	.428	.000	1.67	4.73

*. The mean difference is significant at the 0.05 level.

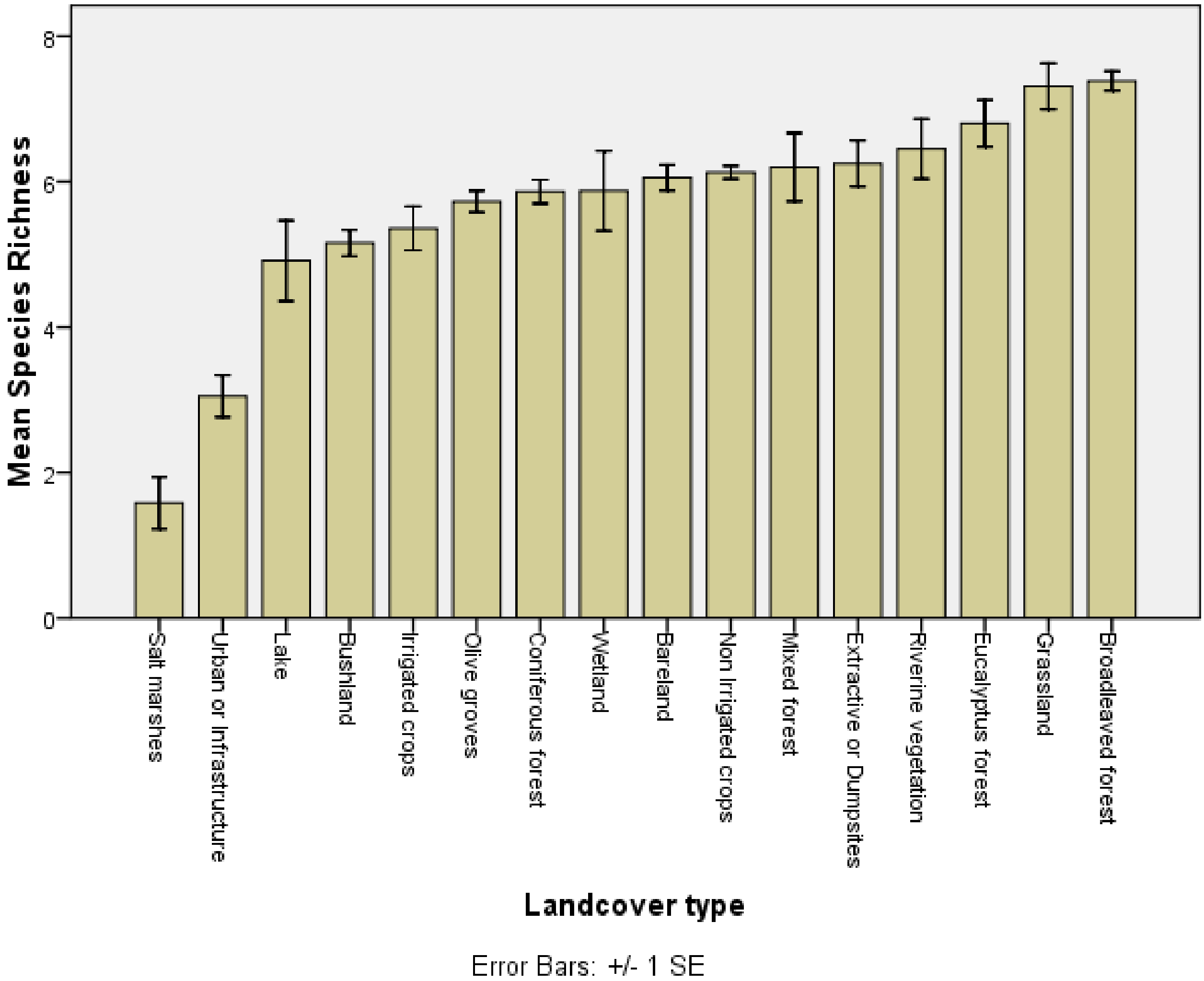
Appendix 5

The classification matrix table of assessing the equality of the means of amphibian species richness per land cover type, with reference to the games-howel test in appendix 4. (A = no significant equality of mean of amphibian species richness: P = significant equality of mean of amphibian species richness)

		REFERENCE OF MEAN AMPHIBIAN SPECIES RICHNESS ON LAND COVER TYPE														
	LANDCOVER	BuIn	WeLa	La	BuLa	IrCr	OlGr	CoFo	BaLa	NoIrLa	MiFo	ExDu	RiVe	EuFo	GrLa	BrFo
TESTED MEAN EQUALITY OF AMPHIBIAN SPECIES RICHNESS ON LAND COVER TYPE	BaLa	A	A	P	A	P	P	P	P	P	P	P	P	P	P	A
	GrLa	A	A	A	A	A	A	A	P	P	P	P	P	P	P	P
	BuLa	A	P	P	P	P	P	P	A	A	P	P	P	A	A	A
	EuFo	A	A	P	A	P	P	P	P	P	P	P	P	P	P	P
	BrFo	A	A	A	A	A	A	A	A	A	P	P	P	P	P	P
	CoFo	A	A	P	P	P	P	P	P	P	P	P	P	P	A	A
	MiFo	A	A	P	P	P	P	P	P	P	P	P	P	P	P	P
	RiVe	A	A	P	P	P	P	P	P	P	P	P	P	P	P	P
	WeLa	P	P	P	P	P	A	A	A	A	A	A	A	A	A	A
	La	P	P	P	P	P	P	P	P	P	P	P	P	P	A	A
	NonIrCr	A	A	P	A	P	P	P	P	P	P	P	P	P	P	A
	IrCr	A	P	P	P	P	P	P	P	P	P	P	P	P	A	A
	OlGr	A	A	P	P	P	P	P	P	P	P	P	P	P	A	A
	BuIn	P	P	P	A	A	A	A	A	A	A	A	A	A	A	A
	ExDu	A	A	P	P	P	P	P	P	P	P	P	P	P	P	P
	CLASSES	**1**	**2**	**3**	**4**	**5**	**6**	**6**	**7**	**7**	**8**	**8**	**8**	**9**	**10**	**11**

Appendix 6:

The bar graphs of mean of amphibian species richness per land cover type with the standard error of the mean when separating the saline Laguna de Fuente de Piedra wetland from the other wetlands. It is important to note that the separation was only based what was coincidentally found in the field and on scientific sampling was done to discriminate the wetlands into saline and freshwater types.



Appendix 7

Stepwise regression model summary:

1st Run

Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.343[a]	.118	.115	2.03747493602108E0
2	.367[b]	.135	.130	2.02047742714604E0
3	.403[c]	.163	.155	1.99079267882424E0
4	.419[d]	.176	.166	1.97825194563956E0

a. Predictors: (Constant), EVAPOWIN

b. Predictors: (Constant), EVAPOWIN, SLOPE

c. Predictors: (Constant), EVAPOWIN, SLOPE, TEMPJULY

d. Predictors: (Constant), EVAPOWIN, SLOPE, TEMPJULY,POPDEN

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients		
		B	Std. Error	Beta	t	Sig.
1	(Constant)	3.528	.364		9.690	.000
	EVAPOWIN	.118	.017	.343	6.786	.000
2	(Constant)	3.628	.363		9.993	.000
	EVAPOWIN	.133	.018	.385	7.314	.000
	SLOPE	-.038	.015	-.138	-2.613	.009
3	(Constant)	13.681	3.007		4.549	.000
	EVAPOWIN	.109	.019	.317	5.690	.000
	SLOPE	-.058	.016	-.209	-3.725	.000
	TEMPJULY	-.399	.118	-.201	-3.367	.001
4	(Constant)	13.164	2.996		4.393	.000
	EVAPOWIN	.109	.019	.317	5.736	.000
	SLOPE	-.060	.016	-.216	-3.881	.000
	TEMPJULY	-.374	.118	-.189	-3.170	.002
	POPDEN	.000	.000	-.115	-2.316	.021

a. Dependent Variable: Amphibian Species Richness

2nd Run

Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.356[a]	.127	.124	2.042
2	.399[b]	.160	.155	2.006
3	.462[c]	.213	.206	1.944
4	.472[d]	.223	.214	1.935

a. Predictors: (Constant), EVAPOTRWIN

b. Predictors: (Constant), EVAPOTRWIN, TEMPJULY

c. Predictors: (Constant), EVAPOTRWIN, TEMPJULY, SLOPE

d. Predictors: (Constant), EVAPOTRWIN, TEMPJULY, SLOPE, POPDEN

Coefficients[a]

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
1	(Constant)	3.391	.357		9.500	.000
	EVAPOTRWIN	.120	.017	.356	7.041	.000
2	(Constant)	13.503	2.805		4.814	.000
	EVAPOTRWIN	.084	.020	.249	4.319	.000
	TEMPJULY	-.400	.110	-.210	-3.634	.000
3	(Constant)	18.131	2.883		6.290	.000
	EVAPOTRWIN	.092	.019	.272	4.840	.000
	TEMPJULY	-.572	.113	-.300	-5.080	.000
	SLOPE	-.072	.015	-.254	-4.816	.000
4	(Constant)	17.769	2.874		6.182	.000
	EVAPOTRWIN	.091	.019	.269	4.816	.000
	TEMPJULY	-.553	.112	-.290	-4.924	.000
	SLOPE	-.075	.015	-.262	-4.983	.000
	POPDEN	.000	.000	-.100	-2.059	.040

a. Dependent Variable: SpRich