# Distribution Modelling of two Hare sub-species and Investigating the Differences

A case of the eastern and western sub-types of the European Hare in Greece

> Ellen Jessica Kayendeke February, 2010

## Distribution Modelling of two Hare sub-species and Investigating the differences

A case of the eastern and western sub-types of the European Hare in Greece

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## Abstract

The European/brown hare (Lepus europaeus) is a herbivore of medium size and an important game animal in Europe. There are two distinct sub-types (the west and east types) of the brown hare coexisting in North eastern Greece. Efficient management of these sub-species requires information on their distribution and how it is affected by different land cover types; and yet no study has been done to find out the respective home ranges of these two hare sub-types. The main objective of this study was to model suitable habitats for each hare sub-type and to find out differences in environmental variables that influence their distribution. A land cover map was created through maximum likelihood classification of a Landsat image and validated with the leave-one out cross validation (LOOCV) method. Land cover classification was extended to other areas of Greece (with an aim of including additional hare presence points to increase sample size) by relating land cover categories to NDVI (Normalised Difference Vegetation Index) classes, which were created through unsupervised classification of a MODIS NDVI time series image. The accuracy result of maximum likelihood classification was used as a baseline for evaluating the NDVI approach. An NDVI proxy map for land cover together with other environmental variables (elevation, slope, aspect, temperature, precipitation, distance to roads, distance to urban areas) were used as predictors in modelling the probability of occurrence of the east and west type hares using maximum entropy (MAXENT) method. Models were evaluated using the Area under the curve (AUC). The AUC for the model of the east sub-type was 0.68 whereas that for the west sub-type was 0.7. Distance to urban areas was the most important predictor for the east sub-type contributing 34.14% to the prediction while land cover was most important for the west sub-type prediction contributing 54.4%. The results show that the east sub-type hare has higher probability of occurrence at low altitudes whereas the probability of occurrence for the west sub-type is equally high at both low and high altitudes.

**Key words:** European hare, hare phylogeography, Leave-one out cross validation (LOOCV), NDVI, species distribution modelling, MAXENT, Greece.

## Dedication

This work is dedicated to my parents; Rose and Samuel Chalo and my siblings; Doreen, Pauline, Solomon and Derrick

Thank you all for the love and support, and for standing with me in prayer through out the study period.

God bless you

I would like to extend my sincere gratitude to all the under mentioned who have all contributed in one way or another to the successful completion of my studies;

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## Abbreviations

AUC	Area under the curve					
ETM	Enhanced Thematic Mapper					
GLCF	Global Land Cover Facility					
ISODATA	Iterative Self-Organizing Data Analysis Techniques					
LOOCV	Leave one out Cross Validation					
MAXENT	Maximum Entropy Modelling					
MODIS	The Moderate Resolution Imaging Spectroradiometer					
mtDNA	Mitochondrial DNA (Deoxyribonucleic acid)					
NDVI	Normalised Difference Vegetation Index					
SPOT	Syste`me Probatoire d'Observation de la Terre					

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## 1. Introduction

The European/brown hare (*Lepus europaeus*) is a herbivore of medium size[1], and an important game animal in Europe [2] whose population has declined drastically throughout Europe since the 1960s [3]. Lundström-Gilliéron *et al.* [4] and Smith *et al.* [3] suggest that extension of urban areas, development of roads and rail as well as agricultural intensification are among the major causes of the decline in the population; because dispersion of the brown hare is greatly hampered by habitat fragmentation[5]. In addition mechanised agriculture requires large monoculture fields and intensive use of herbicides and pesticides which results in reduced abundance of weeds on farmland [6, 7] and this in turn affects the hares adaptive feeding behaviour.

It is important that there should be diversity in areas occupied by the hare as this ensures accessibility to requirements such as food and shelter all year round [7, 8]. However, despite the negative impacts of habitat loss on biodiversity [9, 10], many European rural areas have experienced considerable changes in the landscape that have reduced habitat richness for various flora and fauna [4]. These changes reduce habitat size and increase isolation of many species [9] by rendering the remaining areas impermeable/inaccessible to wild animals like the hare [5]. For example, urban areas and road networks could act as barriers for hare dispersion into remaining habitable areas [4].

Subsequently, many countries like Germany and Switzerland have classified the hare as near threatened or threatened [7] while in Greece, restocking programmes were carried out to curb the hare population decline [11]. Additionally, the hare is also protected from being hunted as game by law in Greece from January to September to guarantee an undisturbed breeding season [12]. Further more the study of the relationship between the hare population and landscape has become increasingly important in many European countries due to the decline in hare population numbers [4]. A study on hare phylogeography revealed two distinct sub-types (the west and east types) of the brown hare in Greece, Bulgaria, Cyprus and northern Israel; these sub-types were found to co-exist in the North eastern area of Greece [2, 13] as shown in figure 1.

However, efficient management of these sub-species requires information on their abundance and distribution [14, 15] as well as how their distribution is affected by different land cover types and other anthropogenic/non-anthropogenic stress factors [10]. It is of importance therefore to model the spatial distribution of these two sub-species, because it provides information on the hare's potential habitat, which can aid in identification of additional areas that are suitable for reproduction and dispersion [5] that have not been inhabited yet due to barriers like roads/rail and urban centres which reduce connectivity between suitable habitats [5].

However, no study has been done to find out the respective home ranges of these two sub-species and if there are any differences between them[16]. This research therefore aims to use biophysical characteristics obtained from remote sensing imagery in combination with geo-referenced presence data to predict the distribution of the two sub-types of hares in north eastern Greece, and to see whether there are differences in biophysical characteristics that relate to either hare species.

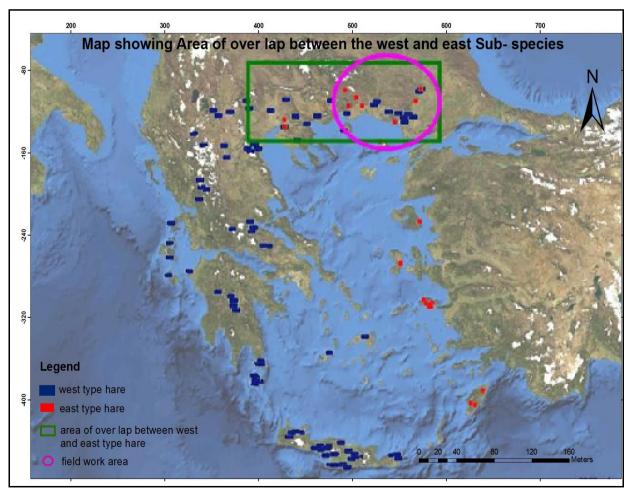


Figure 1: Map showing the distribution of the west and east type hare in Greece (Illustration provided by Antoniou Cilia (PHD student); University of Iraklion, Crete)

### 1.1. Ecology of the brown hare

The brown hare is a herbivore of medium size (3 to 5 kg) that is widely distributed in Europe. It has been observed in habitats at sea level and also at higher elevations, up to an altitude of 2200 meters [17]. It is a nocturnal animal that uses most of the day time hours for resting[6, 18], whereas feeding takes about a third of its time [6]. The brown hare likes open country with scattered shrubs for cover, but it is very adaptable and thrives well on mixed farmland [6, 11, 19] where it utilises vegetation present in its habitat as shelter [20]. The brown hares' home range varies between 38 to 330 hectares in different geographic ranges and it can travel approximately 15km when feeding in a night [6].

Common foods of the brown hare include soft greens, woody plants, root crops, grain crops and forest plants [6] that are rich in fat [21]; but its diet can be adapted according to available food resources in the habitat. For example the diet constitutes cultivated crops like winter cereal in autumn and winter [7, 19] but changes to wild herbs and grasses in spring and summer [7, 8]. In addition hares can adapt to feeding on twigs and the barks of woody trees especially during harsh winters when they are not able to access vegetation on the ground due to deep snow [21]. Since the amount of snow increases with increase in altitude, brown hares are likely to prefer lower altitude areas, except during mild winters when snow cover is less abundant even at a high altitude [22].

The brown hare's population size can be affected by landscape structure, agriculture and weather conditions [23]; but Pépin, D. *et al* [24] suggests that habitat characteristics are the most critical causes of population decline and that negative effects from climatic conditions are only exacerbated by loss of high quality year-round forage and cover. The brown hare usually prefers large highly diverse areas to small mosaics of habitat surrounded by settlements or/and high ways [25]; however with changing agriculture practices there is an increase in large fields that have a low diversity in crops [8]. In addition leveret deaths could be increased by activities like silage cutting and mechanised crop harvesting [8]. Further, intensive use of herbicides does not only affect nutrition by reducing diversity due to elimination of wild herbs and grasses, but can also bring about hare deaths from poisoning [8].

Presence of open areas in forested areas could encourage adaptation of the brown hare in forest ecosystems, where they can still access food and are also less likely to be spotted [22] by predators and hunters. This is especially true for areas with trees where there is enough space in the undergrowth for hares to travel through to feeding places as opposed to shrubs that are entangled making movement

difficult (Petros Platis, October 2009; personal communication [26]). Vegetation cover/density is affected by the orientation of slopes (i.e. in Greece, North facing slopes have more vegetation than south facing slopes) and in turn influence suitability for the brown hare in terms of cover and forage (Petros Platis, October 2009; personal communication [26]).

Although the brown hare has a prolonged breeding season from January to September [27], local variations in climate may lead to changes in beginning and end (length) of this season in different years [12, 22, 28]. High precipitation increases the rate of bacterial and parasitic infections which in turn lead to high mortality of hare offspring (leverets). On the other hand, higher temperature increases the brown hare's reproductive rate, and in addition warmer temperatures during mild winters improve availability of forage because grasses are able to grow nearly all year round [19].

Many species' adaptations to local conditions could be due to mitochondrial composition; for example *L. timidus*'s (mountain hare) mtDNA is expected to have contributed to its successful adaptation to cold weather compared to other hare species [29]. Similarly, the phylogeographic distribution of the subspecies of *L.europeaus* could be attributed to adaptations linked to their specific mtDNA make-up [13].

### 1.2. Relevant factors

The conceptual diagram (figure 2) shows the factors that influence distribution of the hare; changes in agricultural practices leading to larger monoculture fields and intensive use of pesticides/herbicides all reduce diversity, expansion of urban areas and road networks result in landscape and land cover changes that eventually lead to degradation and loss of habitat [30]. Different climatic conditions (as explained in section 1.1 above) also affect hare distribution.

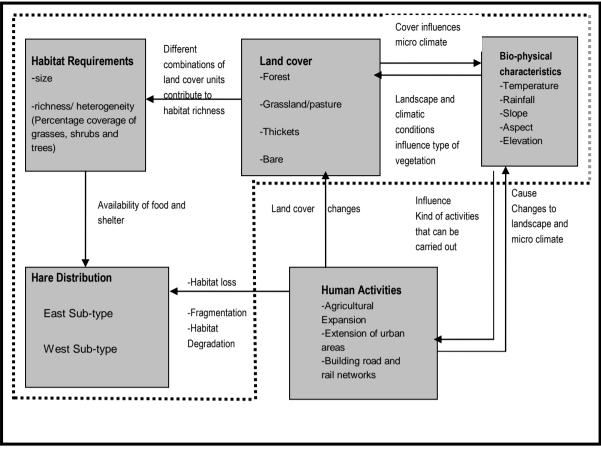


Figure 2: Conceptual diagram illustrating factors that influence distribution of the brown hare

## 1.3. Species Distribution Modelling

Information about the range of a species distribution is important because it can be utilised for conservation purposes [31-36]. As a consequence, more research is being done on obtaining reliable distribution maps from incomplete species data [37] and a variety of statistical modelling techniques is in use to simulate the spatial distribution of terrestrial animal species [32, 33]. A model is a simplification of reality and it can aid in providing more understanding of physical, and biological systems [14]; for example a model can be utilised to understand factors relevant for the two sub-species of the hare.

Hengl, T., *et al* defines a species distribution model (SDM) as "as a statistical and/or analytical algorithm that predicts (either actual or potential) distribution of a species, given field observations and auxiliary maps, as well as expert knowledge"[38]. Predictive modelling can be static or dynamic, where static models provide time-independent equilibrium predictions and dynamic models predict time-dependent response in relation to changes in the environment [39]. Static models are more feasible for use in

species distribution modelling because a large number of species have not been studied in detail in relation to their responses to changes in the environment [33].

Modelling techniques use species presence/absence data in combination with environmental conditions to identify factors controlling the observed distribution of species [33, 37, 40-42]. However, species absence data is difficult to obtain [31, 37, 43] due to the elusive behaviour of animals and inaccessibility of some habitats [43] and on top of that false absences can reduce the accuracy of predictive models [37]. Alternative methods that use only presence data have therefore been developed [37, 43]. Examples of such modelling techniques include; Ecological niche factor analysis (ENFA), Genetic algorithm for rule-set prediction (GARP), and Maximum entropy method (Maxent) [38].

Maxent is a statistical model that was defined by Phillips et al [44] as "a general-purpose method for making predictions or inferences from incomplete information". This modelling approach has other advantages in addition to the ability of making predictions basing on only presence data; for example it can use both continuous and categorical data and is suitable for use when the number of training samples is small because it is generative rather than discriminative [44]. Generative approaches fully exploit information available (in this case presence points and environmental variables) which is not the case with discriminative approaches [45]. Finally, the output is continuous which makes it easy to differentiate modelled suitability of different areas [44].

Maxent predicts the distribution of species by comparing environmental conditions in a given geographic range to conditions in areas where the species has been observed [37, 44]. Existing knowledge about biophysical processes that govern the relationship between species and their environment can be utilised to select relevant variables to be used in the distribution modelling [46].

However, species distribution modelling can be affected by multi-collinearity, which refers to situations where explaining variables are correlated [47]. This can be checked through use of the variance inflation factor (VIF) [33, 48]; which measures the impact of collinearity among variables [49]. Values of VIF greater than 10 indicate presence of collinearity [48, 49] and variables with a high VIF are usually eliminated from the model. This can however lead to removal of important environmental variables and retaining those that are less important [50]. Therefore a researcher should not depend entirely on such statistical methods to select variables to use in the model but also on prior knowledge/theory [50].

### 1.4. Research Problem

The genetic analysis of brown hare populations revealed the existence of two distinct clades/sub types, which are co-existing in the North eastern part of Greece [2]. Genetic diversity as in the case of the hare, is strongly linked to a species ability to adapt to changes in the environment [11, 51] and is therefore recognised by the IUCN as a level of diversity that should be conserved [11]. Kasapidis et al [2] suggested that these hare sub-populations qualify as Evolutionary Significant Units (ESUs), which are suitable for conservation.

Effective management of these sub species requires information on their respective distribution and habitat requirements [52] and yet this information is not available [16]. There is a need therefore to identify environmental variables that are important for the two sub-species in order to model the spatial distribution of suitable areas. However the number of observations of the two sub-species in the study area (Northern Macedonia and Thrace) is small; due to the fact that they need genetic verification to be included in either the east or west type sub-populations. This insufficient sample size hampers statistical testing.

There are other presence observations/points in other areas of Greece (central Macedonia, west Macedonia, Epirus and Thessaly) that could be included in the distribution modelling. However since the field work on land cover types was carried out only in Eastern Macedonia and Thrace, these land cover observations are not adequate for making a land cover map of the whole of Greece. In spite of this, if it can be established that there is a relationship between land cover and NDVI classes in the field work area, land cover types can be related to NDVI classes and the land cover classification can be extended to the rest of Greece.

This can make it possible to expand the extent of the distribution modelling to a wider area of Greece since NDVI and other environmental variables are available for the rest of Greece. Results from the modelling would not only provide information on the respective distribution of the two sub-species but also give an improved understanding of environmental conditions that are most important for each of them, as well as differences between them. Such information could help inform management decisions concerning the west and east type hares in future.

## 1.5. Objectives

### 1.5.1 General Objective

To determine the distribution of each of the hare sub types, as well as differences in environmental variables that influence the distribution

### 1.5.2 Specific Objectives

1a) To map different land cover types present in the study area using two approaches (supervised classification and NDVI based method); and to compare the accuracy of the two approaches

1b) To expand land cover classification to a wider area of Greece using NDVI classes as a proxy for land cover

2) To model suitable habitats for each hare sub-type, and to find out if there is a difference in environmental variables that influence their distribution

3) To identify environmental variables/predictors that have the most contribution in modelling the habitat for each sub-species type

## 1.6. Research Questions

- 1. Is the accuracy of NDVI based mapping comparable to that of supervised land cover classification?
- 2. Is there a difference in the set of environmental variables that determine hare sub type distribution for the east and west type hares?
- 3. What are the most important environmental variables determining each hare sub type distribution?

## 1.7. Hypothesis

- 1.  $H_0$ : NDVI based mapping is less accurate than supervised classification
  - H<sub>1</sub>: The accuracy of NDVI based mapping is comparable or better than that of supervised classification

- 2. H<sub>0</sub>: There is no difference in environmental variables determining the distribution of each of the Sub-species
  - H<sub>1</sub>: The distribution of the sub-species is influenced by different combinations of environmental variables
- 3. H<sub>0</sub>: All environmental variables are equally important in modelling distribution of the two sub-Species
  - H<sub>1</sub>: Some environmental variables like land cover are more important than climatic variables in modelling the distribution of the two sub-species

## 2. Materials and Methods

## 2.1. Study area

Greece is a mountainous country located in south Eastern Europe that shares borders with Albania, Macedonia, Bulgaria, in the north and Turkey in the east; it also borders the Aegean and Ionian seas to the east, south and west respectively [53]. It is located between 39 00 N and 22 00 E and is approximately 131,957 square kilometres, 130,647 of which are on land (including islands) and 1,310 on water [54]. Greece has three climate types; Mediterranean, alpine and temperate [53].

The field work area was in north eastern Greece in the regions of East Macedonia and Thrace (figure 3), which cover approximately 14,157 square kilometers [53]. This area is characterised by plains and a high altitude mountain range (Rhodope range) that is covered by thick forests; and has a temperate climate with cold damp winters and hot dry summers [53]. This area was chosen because the two sub-types (West and East type) of the brown hare are co-existing here [2] and there have been no studies to find out if the two types have the same habitat requirements or not [16].

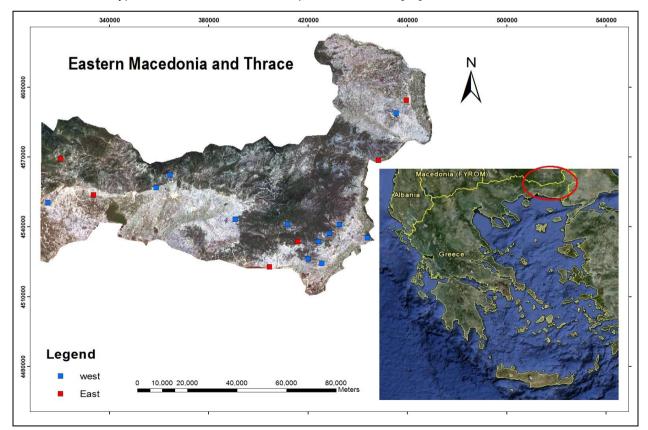


Figure 3: Map of Eastern Macedonia and Thrace; with east and west sub-type presence points (in blue and red respectively)

## 2.2. Data collection

### 2.2.1. Secondary data

Secondary data used included geo-referenced (decimal degrees) hare presence points that were recorded with the help of the hunters' association of Greece and provided by PHD student Ms. Antoniou Cilia and Professor Kotoulas Georgios of the University of Iraklion, Crete. There are a total of 315 points covering East Macedonia and Thrace, central Macedonia, west Macedonia, Epirus and Thessaly; but of these only 63 have been genetically analysed so far, and results show that 52 samples belong to the west type hare and 11 to the East type. Within the field work area alone, there are 12 and 6 samples of the west and east type hare respectively (table 1).

Region		Total		
	East	West	Unknown	
			( genetic testing not yet done)	
Whole of Greece	11	52	252	315
Field work area (East	6	12	57	75
Macedonia and Thrace)				

Table 1: Showing number of east and west type observations

### 2.2.2. Field work preparation

The presence points were re-projected to UTM 35N in ArcGIS, and together with the Greece boundary saved on an Ipaq SD card for navigation during fieldwork. In addition, the presence points were also exported to a Google earth KLM file; in Google earth every point was zoomed up to 3508 feet (areas surrounding each point were clearly visible), saved as an Image file, and then printed to use as a navigation back up in case of Ipaq malfunction in the field.

Further more, 30 random points were created within the field work area using ArcGIS "create random points" tool. The points were also saved on the Ipaq in UTM 35N projection and an image was printed for each random point as with the presence points above. The random points were created because visiting as many random points as possible would help in removing bias when drawing conclusions([55, 56]; pgs 14 and 341) about the land cover types present in the area; as it improves the possibility of visiting other cover types that are not represented by the presence points.

Finally, a topographic map of Greece was scanned, geo-referenced, and saved as an image file in UTM 35N projection. In ArcGIS, both the presence and random points were projected on the topographic map (in different colours) and the image was printed to help the driver with navigation in the field.

#### 2.2.3. Field work

Field work was carried out to collect information about percentage cover of different vegetation layers, and a total of 58 points were visited. At each point, a 50 meter line intercept was laid out using a measuring tape. Line intercept sampling is a method that was originally developed to estimate shrub coverage[57], it is widely used in the estimation of density and coverage of plant communities [58]. Implementation can be by use of fixed length transects whose direction can be decided either randomly or purposively prior to sampling [57]. To estimate cover, the length over which plants intercept the line are recorded; but this should be done separately for plants of different height categories especially if vegetation layers overlap ([59]; pgs 90-92). The layers were recorded separately because different combinations/percentages of vegetation layers are expected to affect suitability of areas for the brown hare.

This sampling method was chosen as opposed to plot sampling methods because it reduces subjective error in estimating percentage canopy cover [60]. Due to the heterogeneity of the landscape, the direction of the transect was selected purposively so that recordings made of the land cover would be representative of the area surrounding the point.

Three vegetation layers were defined (before field work) according to height classes [59];

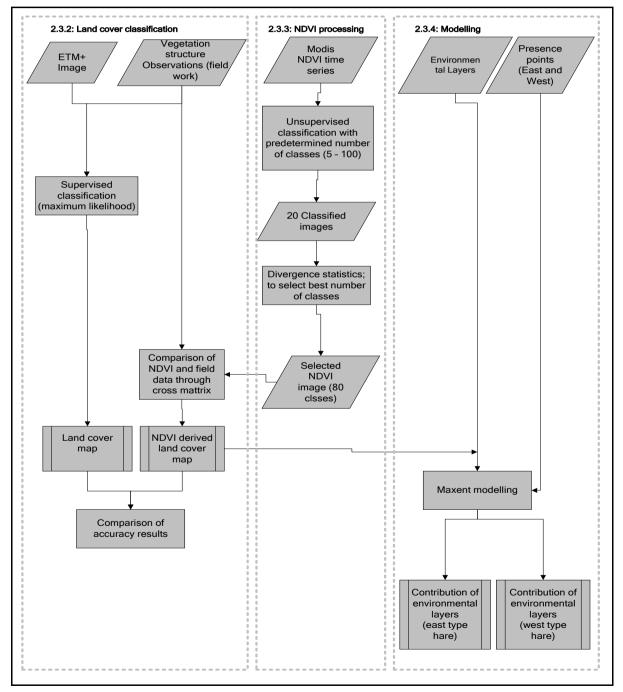
- Low layer; which included grasses and herbs with a height less than 1 meter
- Medium layer; shrubs with height between 1 and 3 meters
- High layer; trees with height above 3 meters

The length of each layer along the intercept was recorded separately, and the percentage cover of each of these layers was calculated as the accumulated length divided by the total length of the line intercept.

## 2.3. Data Analysis

#### 2.3.1. Methodological flow chart

The flow chart (figure 4) is an illustration of the steps taken in data analysis. Two mapping approaches were applied; supervised classification described in section 2.3.2, and an NDVI approach involving unsupervised classification of an NDVI time series image (section 2.3.3). The resultant land cover map together with other selected environmental variables, were used as predictors in modelling the probability of occurrence of the east and west hare sub-types (section 2.3.4).





#### 2.3.2. Land cover classification

The percentage cover of the three vegetation layers (recorded at each of the 58 points) was converted to land cover types by comparing it to a classification key (see Appendix 2) that had been made prior to field work based on FAO country guidelines for Greece [61, 62] and later adapted according to the conditions observed in the field. The points were then used as training samples in the supervised classification of a landsat (ETM+) image using maximum likelihood classification (MLC), to create a land cover map of the study area.

Although spot images have a higher spatial resolution, landsat data was selected because it has more spectral bands than Spot images and also because the landsat scene is larger [14] therefore fewer images were needed to cover the study area. Two ETM + images (30x30 meters) taken in July 2007 downloaded from the Global land Facility (GLCF were cover website: http://glcfapp.umiacs.umd.edu:8080/esdi/index.jsp), and using the Erdas Imagine "Mosaic Tool" a single image of the study area (Northern Macedonia and Thrace) was created. The 58 points collected in the field were used to classify the image (figure 6) with land cover types that had been observed; Dense forest, open forest, Dense shrub land, Open shrub land, farm land and grassland.

However, MLC requires a number of samples that is between 10 to 30 per class [63, 64] for the training stage of classification [63]. For that reason, the leave-one out cross validation (LOOCV) method (see [65, 66]) was used in the classification because it is helpful when there is a low number of samples [65-67], as well as in situations where there is an unequal number of samples for each class [67]. This method also makes good use of available data since each sample point is used as both a training and test point [66].

The LOOCV method involves running the classification many times each time leaving out one sample which is later used as the test point, and the overall accuracy calculated as the average of the accuracy values obtained with each test sample [65].

A model (figure 5) was built using ArcGIS model builder to carry out maximum likelihood classification on the landsat image. For each classified image, the accuracy assessment was done by overlaying the test point over the image to check if it had been correctly classified, and overall accuracy was obtained by calculating average of all the accuracies.

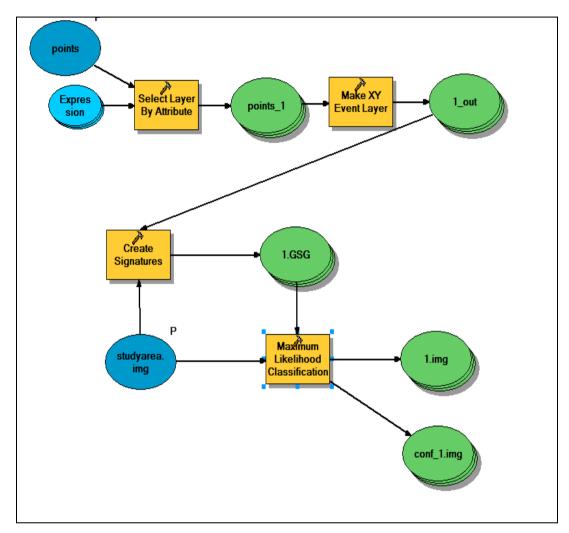


Figure 5: Leave-one out classification model

The resultant land cover map should have been used as an environmental layer in modelling the distribution of the brown hare, however due to the small number of presence points of the hares (12 west, and 6 east) in the study area; there was a need to expand the area so as to include presence points in the areas of central Macedonia, west Macedonia, Epirus and Thessaly. In order to expand the land cover classification to the rest of Greece, an NDVI mapping approach explained in detail in section 2.3.3 was used.

#### 2.3.3. NDVI Data Processing

#### 2.3.3.1. Data down load and pre-processing

NDVI is a biophysical parameter that has a correlation with vegetation's photosynthetic activity; it can therefore give information on the greenness of vegetation [68]. It is calculated as;

$$NDVI = \frac{NIR - RED}{NIR + RED}$$

Although NDVI does not give exact land cover types, an NDVI time series can be used to separate different land cover classes based on their phenology [68, 69]. NDVI values vary between -1 and 1, where values greater than 0.1 indicate vegetated areas and those greater than 0.5 normally represent areas with dense vegetation [70].

MODIS NDVI 16-day composite grid data at a spatial resolution of 250 metres were downloaded in HDF format from a NASA website (<u>https://wist.echo.nasa.gov/api/</u>) for a time period between January 2001 and December 2008; see ([71]: pgs: 13 - 46) for details about the data compositing and algorithms used in producing the NDVI products. There were 23 images per year, totalling up to 184 images for the entire time period. MODIS time series were used in this research because they have a higher spatial resolution than the SPOT NDVI data which is at a resolution of 1km.

The data was first imported to the imagine format using Erdas Imagine software, and then re-projected from a sinusoidal to UTM projection using the nearest neighbour resampling method. The images were then stacked to create an NDVI time series in one single image consisting of 184 bands.

#### 2.3.3.2. Unsupervised classification

Unsupervised classification runs were carried out on the resultant NDVI image using the ISODATA clustering algorithm; the classification was repeated with a pre-determined number of classes (from 5, 10, 15 e.t.c up to 100 classes). With each classification run, the maximum number of iterations was set to 50 and convergence threshold set to 1 [72].

Unsupervised classification is based on the natural grouping of pixels of an image when plotted in feature space, and the ISODATA clustering algorithm (a self organising data analysis technique) is a

sequential method that uses spectral distances and iteratively classifies pixels and repeats the classification after redefining the criteria for each class [73].

The classified images were compared by computing divergence statistics for each image; which is a measure of distance between the generated cluster signatures [73]; the divergence was expressed in form of minimum and average separability; where minimum separability refers to the similarity between the two most similar classes and average separability refers to the similarity amongst all the classes [72]. According to C.A de Bie *et al* [72], both the minimum and average divergence should be as high as possible. An image of 80 classes had the highest values for minimum and average separability and it was therefore selected for use in further analysis.

#### 2.3.3.3. Comparing NDVI classes to field data

The NDVI image was compared with the land cover types that had been identified in the field; and NDVI classes that were related to each land cover type were identified. This was done through visual inspection in ArcGIS, where the field data points were overlaid on the NDVI image. A cross matrix of NDVI classes and land cover types was made to visualise the co-occurrence of vegetation classes with NDVI classes and to relate the two.

After demonstrating that there is a relationship between NDVI and land cover, the NDVI image was reclassified into land cover categories;

- 1. farm land
- 2. shrub land
- 3. Open forest
- 4. Dense forest
- 5. "other"; representing land cover classes existing in other areas of Greece but not sampled in the study area.

#### 2.3.4. Modelling distribution of east and west type hare

Environmental variables that were used in the modelling included; land cover, aspect, slope, elevation, distance to roads, distance to urban areas, precipitation (mean monthly values for January up to December) and temperature (average values per month for January up to December); these were a total of 30 variables which were selected based on the description of hare ecology in sub section 1.1 above.

The environmental layers were all re-projected to UTM 35N projection, and cell size resampled to 30 x 30 meters (to fit the DEM resolution) using the ArcGIS software. All layers were clipped using the boundary of the extended study area. Using the combine function of ArcGIS, the information in all the environmental layers was combined into a single table; and the table was imported into SPSS software for multi-collinearity diagnostics using the Variance Inflation Factor (VIF).

Environmental layers with a VIF greater than 10 were eliminated, one variable was removed at each step and the calculation repeated until all remaining layers had a VIF less than 10. The selected images were then converted to ASCII format because the Maxent software works with data in this format. Two separate tables with X Y coordinates were created for the west and east type hare in an excel worksheet; the tables were saved in CSV comma delimited format after which they were ready for use in the modelling.

The maxent model was run for both the east and west type hare, using 45% of the presence points as a test percentage, for use in calculating the Area under the curve/receiver operating characteristic (AUC/ROC). Lobo *et al* defines AUC as a "discrimination index that represents the likelihood that a presence will have a higher predicted value than an absence" [74], and it gives a measure of model performance across all thresholds [44, 74].

The ROC curve is obtained by plotting sensitivity on the *y* axis and 1–specificity on the *x* axis for all possible thresholds, where sensitivity refers to all positive instances that are classified as present and specificity represents all negative instances that are classified as absent [44, 75]. Sensitivity is also known as the true positive rate and therefore 1–specificity represents false positive rate [44].

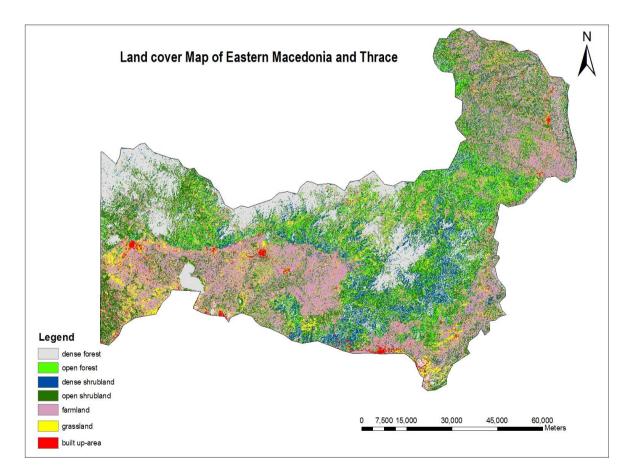
There were 11 and 52 presence points for the east and west hare respectively. For the east hare, the model was run 11 times, each time with a different random test/train set partition; and averages of AUC and statistics of variable contribution over all the models were computed. For the west hare, 5 of the samples that were within the land cover type "other" were excluded form the model; therefore 47 presence samples were used. This is because it would be problematic to make conclusions about the effects of this cover type since there was no information on vegetation structure or type within this class.

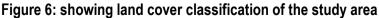
## 3. Results

## 3.1. Land Cover Classification

The percentage cover of vegetation layers recorded in the field were reclassified using a classification key (appendix 2) into the following land cover types; dense forest, open forest, dense shrub land, open shrub land, farm land and grass land (appendix 3). These land cover types were used in the supervised classification of the ETM image of the study area. In the LOOCV accuracy assessment, 36 (out of 58) of the test points were classified correctly; therefore Overall accuracy was 62%.

In the land cover map produced (figure 6), farm land, grassland and built-up areas are generally in the low altitude areas whereas areas of dense and open forest are located at higher altitudes within the study area. There are mosaics of dense shrub land and open forest occurring on lower slopes of the mountains, as well as mosaics of open shrubs and cultivated areas in some of the low altitude areas.





### 3.2. NDVI Data Processing

#### 3.2.1. Unsupervised Classification

The divergence plot (figure 7) showed that images with 80 and 100 classes had a high average separability; but the image with 80 classes was selected because it corresponded to a higher value of minimum separability than the later.

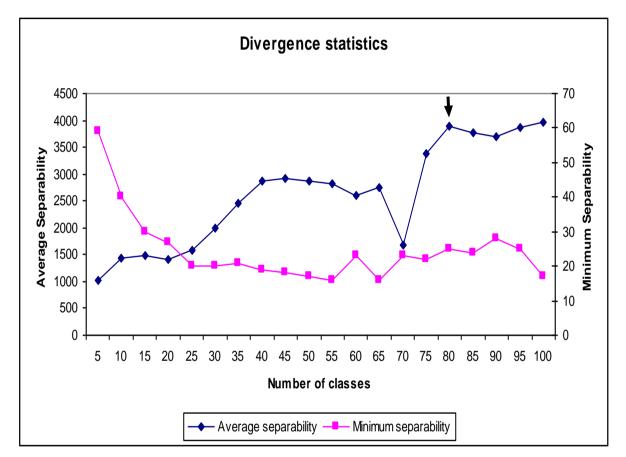


Figure 7: graph showing average and minimum divergence against number of classes

#### 3.2.2. Comparing NDVI image to Field Data

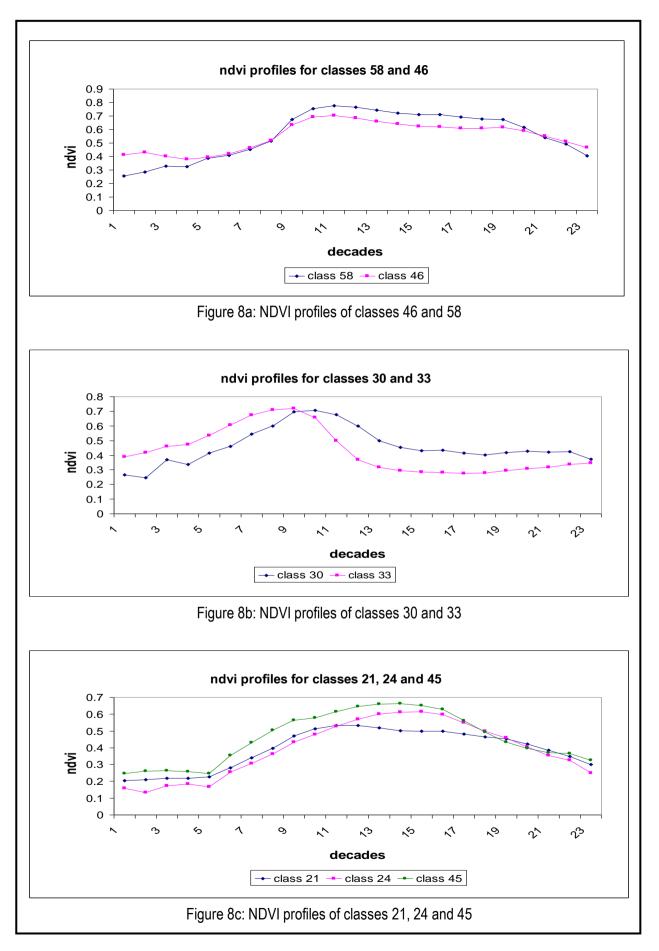
The selected NDVI image was compared with field data points in a cross matrix (table 2) to ascertain whether there was a correlation between NDVI and land cover. The grey cells indicate NDVI classes that had a correlation with each of the land cover types identified in the field. For example NDVI class 80 was in the category dense forest while classes 31, 35 and 58 were representative of open forest. In the same way farmland was represented by 5 NDVI classes (11, 17, 19, 30, and 21); and this is probably due to differences in crop calendars for winter and summer crops as well as differing management practices (monocultures versus mixed cropped system or cereals versus olive plants).

There was confusion within NDVI classes 19 and 31 because the field points are spread out in more than one land cover category. They were matched to farmland and open forest respectively by selecting the land cover type that had the most number of field points, and further through visual inspection of the respective NDVI class profiles (which give information on seasonal changes/phenology).

Some NDVI classes (for example 45 and 58) were combined because they had almost similar profiles [72] which indicates comparable phenology (figure 8). On the other hand, some classes were not matched to any land cover categories; for example class 32 had only two data points that were spread between open forest and dense shrub land, and this was insufficient to make a correlation with any of the two land categories.

land cover	80	31	35	46 +58	78	13	11	17	19	30+33	21 + 24 + 45	32	34	Total
Dense forest	3		1	1				1	2					8
open forest		5	3	3				1	1			1		14
dense shrub land		1			5				1			1		8
open shrub land		2				2				1			1	6
grassland							1							1
farm land		1				1	3	4	5	3	3			20
Total	3	9	4	4	5	3	4	6	9	4	3	2	1	57
correct	3	5	3	3	5	2	3	4	5	3	3	-	-	39
														68.4%

Table 2: cross matrix showing relationship between NDVI and land cover classes





## 3.3. Modelling Distribution of the east and west type hare

The following environmental variables (table 3) were selected to be used in the modelling after the multicollinearity diagnostics;

Environmental variable	VIF
Precipitation-august	4.3
Pre-February	1.6
Precipitation-June	3.9
Temperature-April	8.2
Temperature-January	7.3
Distance to roads	1.4
Distance to urban areas	1.1
Aspect	1.6
Elevation	11.3
Slope	3.4
Land cover	-

Table 3: environmental variables selected for species distribution modelling

Land cover was not included in the test because categorical variables can not be tested for multicollinearity. After the VIF test, elevation was retained despite it having a value greater than 10 because it is expected to have a big influence on the hare's distribution as described in the ecology section 1.1 above.

#### Distribution of the East type hare

The average AUC value for the east type model runs was 0.68 with a standard deviation of 0.1. The probability map (figure 9) shows that there is a higher probability of occurrence for the east hare in low altitude areas than in the mountain areas. The jack-knife test (figure 10) for the east hare shows that elevation has the highest model gain when used in isolation and therefore it has the most useful information by itself, on the other hand distance to urban reduces model gain the most when omitted and this indicates that it has information which is not present in the other variables.

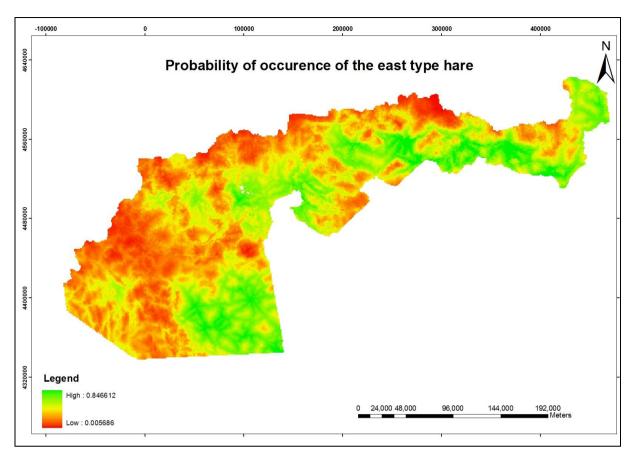


Figure 9: map showing probability of occurrence for the east type hare

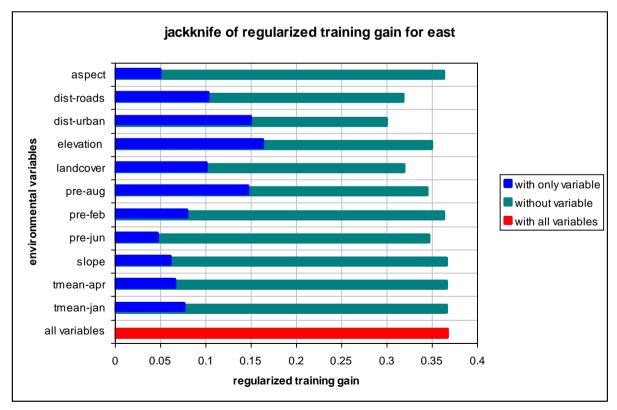


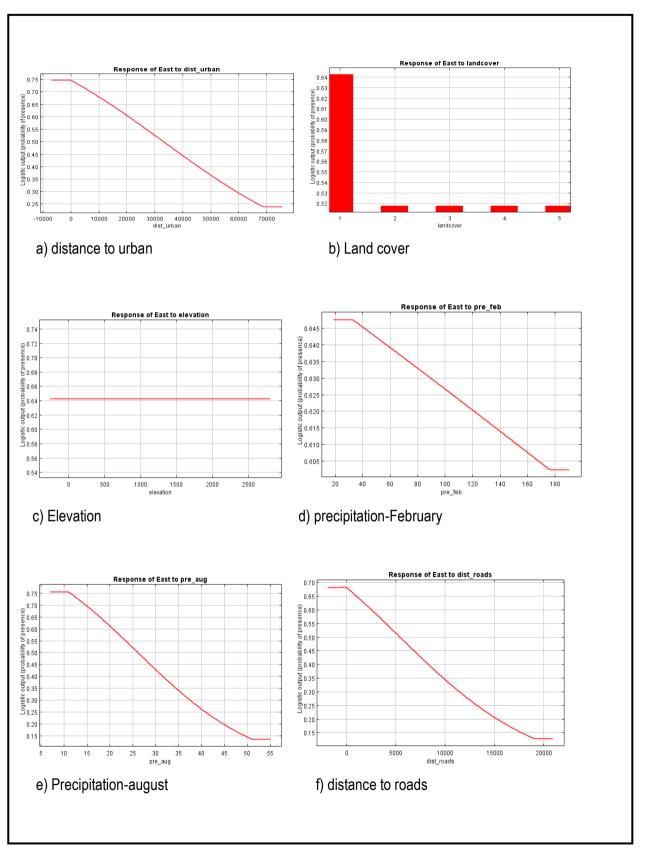
Figure 10: jack-knife test of variable importance for east hare

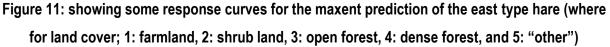
When all the eleven environmental variables were included in the modelling, two of these (temperature-April and temperature-January) did not make any contribution in the model predictions (table 4). The highest contributions were by distance to urban areas, land cover and elevation while the remaining variables contributed less than 10% each.

Number	variable	percent contribution
1	Distance to urban	34.14
2	Land cover	23.80
3	elevation	15.85
4	precipitation-august	9.84
5	Distance to roads	9.21
6	precipitation-February	3.64
7	precipitation-June	2.98
8	aspect	0.49
9	slope	0.05
10	temperature-April	0.00
11	temperature-January	0.00

Table 4: showing percentage contribution of variables in maxent model for east hare

Figure 11 shows the response curves for six of the environmental variables that contributed most in the model prediction. The probability of occurrence for the east hare reduced with increasing values of distance to urban, precipitation-February precipitation-August, and distance to roads. For the categorical variable (land cover), the probability of occurrence was less than 0.5 for all land cover classes except class farm land; however probability did not change at varying levels of altitude.





#### II. Distribution of the West Type Hare

For the west type hare the AUC value was 0.7, and the map (figure 12) shows that there is a high probability of occurrence both in low land areas as well as in mountainous areas. The jack-knife test (figure 13) shows that land cover is the most important variable since it decreased the gain most when excluded from the model. On the other model gain was zero when distance to roads was used in isolation and removing it did not affect model gain.

The response curves of the most important environmental variables (figure 14) show that probability of occurrence for the west hare reduced with increases in temperature-January and precipitation-February; the probability reduced with increase of precipitation-august but started increasing after a precipitation value of 40mm. Probability increased as distance to urban increased but reached a peak at 39000 meters and started declining with further increases in distance from urban areas. On the other hand, probability of occurrence reduced as elevation increased up to 500m where it remained constant at 0.5 but started increasing after 2000m. Finally, there was a higher probability of occurrence (0.66) in shrub land followed by farm land (0.49) whereas both open and dense forest had a probability of occurrence of 0.34.

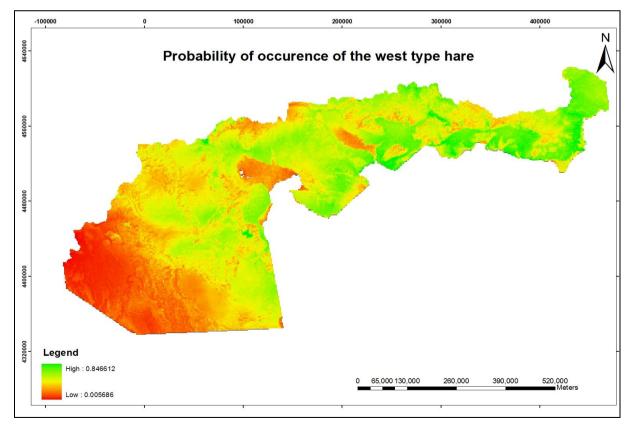


Figure 12: Map showing probability of occurrence for the west type hare

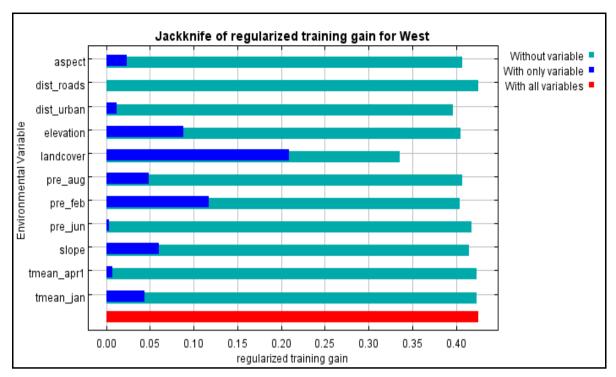


Figure 13: Jack-knife test of variable importance for the west hare

The environmental variables when used in combination did not have equal contribution; for example land cover contributed more than 50% to model predictions (table 5) followed by precipitation-august with a 11.1%. All other variables contributed less than 10% whereas distance to roads did not have any contribution in modelling the occurrence of the west hare.

Number	Variable	Percent contribution
1	Land cover	54.4
2	Precipitation-august	11.1
3 Temperature-January		8.6
4	Precipitation-February	6.4
5	elevation	6
6	Distance to urban	5.2
7	aspect	5
8	slope	1.9
9	Precipitation-June	1.1
10	Temperature-April	0.4
11	Distance to roads	0

Table 5: showing percentage contribution of variables in maxent model for west hare

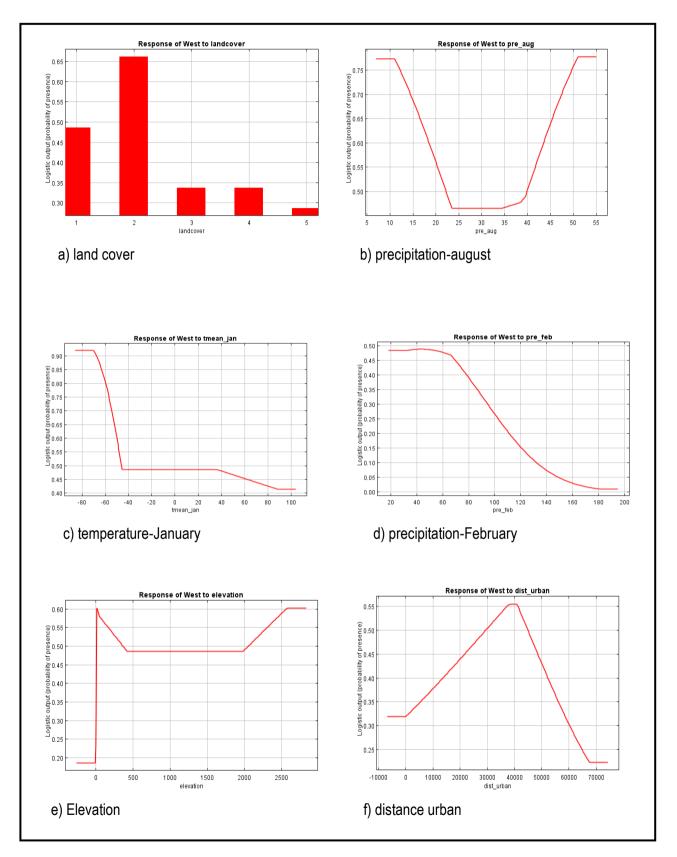


Figure 14: showing some response curves for the maxent prediction of the west type hare (where for land cover; 1: farmland, 2: shrub land, 3: open forest, 4: dense forest, and 5: "other")

## 4. Discussion

### 4.1. Land cover mapping

The accuracy of the supervised classification of the ETM+ image (leave-one out classification method) was 62% whereas that from the NDVI approach was 68%. Since the accuracy for the supervised classification was used as a baseline to gauge the performance of the NDVI approach, this shows that the two approaches give reasonably similar results. The low accuracy levels could have been as a result of pixels with mixed spectral responses[76] because the study area is heterogeneous with changes in land cover at relatively short distances; this is especially true for the NDVI image at a spatial resolution of 250 meters. Such pixels are normally excluded from the training set to retain only those that are good representatives of the relevant land cover categories [76] but in this case since the sample size was small, all points were utilised in both the training and validation process.

During field work, the different vegetation layers were recorded in order to gain further insight on which vegetation structure types are important for the brown hare, however there are differences in appearance of vegetation structure when viewed at the ground level (during field work) as opposed to outer space [77] as shown in figure 15. Some of the lower vegetation layers (shrubs and grasses) may not be visible on satellite imagery especially if trees have dense canopies. This difference in detail between field observations and the ETM image could also have had an effect on the accuracy of the land cover classification.

Understory vegetation is important for the brown hare which feeds mainly on grasses/herbs; therefore the hare distribution is likely to be affected by different vegetation structure characteristics. However landsat images may not accurately map lower vegetation layers but classification could be improved by integrating information from radar imagery. This is because radar backscatter depends on the structural properties of vegetation and also due to the ability of long off-nadir wavelengths to penetrate vegetation canopies [78-80].

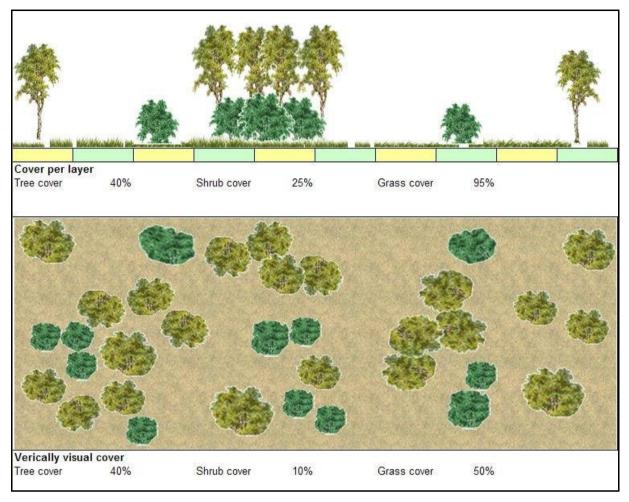


Figure 15: showing differences in canopy cover when viewed at ground level and outer space (Illustration from Rasheed Khwarahm's thesis; ITC 2010 [77])

## 4.2. Distribution of the east and west type hare

The AUC values for the east (0.68) and west (0.7) models showed that the predictions are better than random; they were both above 0.5 which indicated that the models have the ability to discriminate between areas of high and low probability of occurrence [81]. These values though below 0.8; a value above which models are judged to be excellent [33] are not un expected since the brown hare is an adaptable animal that thrives in a variety of habitats [6].

The area under the curve (AUC) is used as a measure of how much species' distributions are restricted to a given range of the predictor variables; and for a generalist species the AUC value would be low because probability is high through out the whole range of the variables [74]. Fulgione, D. *et al* [82] also reported a low evaluation index (0.61) for a habitat suitability model that had been developed for the

European hare. In such cases low AUC values do not imply that the model is inaccurate but rather show that a model has been able to depict the generalist nature of the species [74].

Nevertheless, since it is inherent in modelling that perfect truth can not be obtained [33], a model should not be categorised as correct or incorrect but should rather be assessed within the context of its application [33]. The maxent models used give the probability that a species occurs in an area and since the predictions are not random, the results are useful because they can give an insight on potentially suitable areas for the two sub-species of the hare in Greece and any differences between them.

The results from the modelling show that there is a difference in the combination of variables that are useful to predict the probability of occurrence of the two sub-species. For example for the east hare, temperature-April and temperature-January did not contribute anything to the model prediction, while for the west hare all variables apart from distance to roads contributed in the prediction. In addition, whereas the most important predictor variables for the east hare were distance to urban areas (34.14%) followed by land cover (23.8%); for the west hare land cover contributed more than 50% followed by precipitation- august with 11%. This corroborates with Pépin, D. *et al* proposition that land cover may have a greater influence on the brown hare population than other environmental factors [24].

However since there is a correlation between distance to urban areas, distance to roads and elevation; the results of variable importance have to be interpreted with care [47]. For the case of the east hare, the high importance of distance to urban areas could be due to preference for more open areas which are closer to cultivated as well as built up/urban areas. Indeed the bar plot of land cover for the east hare shows that there is a higher probability of occurrence in cultivated areas than in shrubs and forested areas. Such areas are ideal habitat for the hare because of the presence of shrubs at field edges/boundaries as was observed during field work; and this relates to literature on hare ecology where hares have been observed to prefer open areas with a few shrubs for cover [6, 83].

Another explanation for the trend observed with the east hare could be due to the small number of samples that have been collected so far with out a pre-defined sampling strategy. Therefore, samples could have been collected in more accessible areas that are nearer to urban areas than in other areas. This indicates that other areas further away could also be equally suitable for the east hare but they have just not been sampled yet.

On the other hand land cover was the most important variable for the west hare, and the bar plot shows that there is a higher probability of occurrence within shrub land (0.66) than in agricultural areas (0.49). This could mean that the west hare is more adapted to areas of higher elevation compared to the east hare since shrubs were observed to occur mostly on higher ground than the cultivated areas.

Further more, the response curve for distance to urban areas shows that probability of occurrence increases up to 3900 meters where it starts decreasing; whereas probability reduced with increasing elevation but remained constant at approximately 0.5 from 490 to 2000 meters after which it started increasing again. This increase in probability of occurrence with increase in distance from urban areas could also be due to the fact that shrub land (which is preferred), is at longer distances from built up areas as compared to cultivated land.

Both sub-species had similar responses to precipitation; where probability of occurrence reduced with increasing precipitation. This could be due to the fact high precipitation levels increase the rate of bacterial and parasitic infections which leads to high mortality especially for leverets [19] during the breeding season.

The probability maps show that the there is a higher probability of occurrence for the east type hare in low altitude areas whereas the probability of occurrence of the west type hare is equally high at both low and high elevations. This difference could however be due to the fact that there are more samples of the west hare that are spread out through the study area whereas the east hare has fewer samples that are mostly within north eastern Greece.

Finally, agricultural land seems to be an important habitat for both sub-species; this could be because the fields have diverse vegetation due to the presence of shrubs and trees in most cultivated areas. Further, land cover types within the landscape change at relatively short distances (as observed during field work) which provide a diversity that is advantageous for the brown hare. However if there is increased intensification and mechanisation leading to larger monoculture fields in the future, this could adversely affect the hare's habitat[3].

# 5. Conclusions and Recommendations

### 5.1. Conclusions

The accuracy derived from the NDVI mapping approach at 68% is comparable to that of the conventional supervised classification method, and it was therefore a useful approach to expand land cover classification to other areas of Greece. However, classification could be improved by integrating information from both landsat and radar imagery.

Results from the analysis above suggest that the low lying areas with cultivated land as well as pockets of shrub land occurring at gentler slopes are potentially suitable for both the sub-species of the brown hare. However, the west type hare is more wide spread compared to the east type hare and has a higher probability of occurrence also at high elevations.

There are differences in environmental factors that determine distribution of the east and west type hares. For example while temperature was useful in determining probability of occurrence of the west hare this was not the case for the east hare; and in the same way distance to roads was important for the east hare but not for the west type hare.

Lastly, the environmental variables when used in combination were not equally important in contributing to model predictions. Distance to urban contributed 34% of the model gain for the east type predictions whereas slope contributed only 0.05%. In the same way for the west type hare, land cover contributed 54% followed by precipitation-august with 11% and all other variables had a contribution less than 10%.

## 5.2. Recommendations

It is recommended that in future, information from radar images should be integrated in mapping land cover types relevant for the hare to find out if this could improve classification of vegetation structure types that have differing composition of understory vegetation.

Future research could look at faecal pellet analysis to find out if the two sub-species have different dietary preferences/breadth because such additional information can also improve on understanding the ecology of the two sub-species.

Another recommendation is that more samples should be collected, with sampling effort concentrated in areas where samples have not been collected before to reduce the bias in the sample points, for example more samples of the east hare should be collected in the mountainous region of East Macedonia and Thrace. In addition, genetic testing of samples already collected should be carried out since this can significantly increase the sample size of both sub-species.

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	Data Sheet For Ve	getation Co	Data Sheet For Vegetation Cover Types In Alexandroupolis, Greece	polis, Greece	
Date:	Coordinates		Observer Name:	Length of Line intercept:	Sub-species Type:
Sample No:	X: Y:				
Elevation (tick):	Slope Steepness (tick):	Aspect		Dominant species	
Hill(top) Slope	Flat 0-5 Gently sloping 5-10		High layer >5m (trees) 1	Medium layer 3-5m (shrubs) 1	Low layer <3m (grasses/weeds)
- Flat	Moderately sloping 10- 20		1 2	2	2
	Steep >20		3	3	3
Expected Land cover types (tick)			Recordings along line intercept	intercept	
Grassland/Pasture					
Shrub land/woodland					
Cereal Jarms Multinle cronned systems					
Crop and natural vegetation					
mosaic					
Bare areas/sparse vegetation					
Natural Forest					
Plantation forest					
		R	Remarks:		

## Appendix II: Land cover Classification key

Tree	Shrub	Grass	Crops	Vegetation structure classes
>=50%				Dense Forest
10-50%				Open forest
<10%	>=50%			Dense shrub land
	10-50%			Open shrub land
	<10%	>=50%		Grassland
		<50%	>=50%	Cropped system
			<10%	bare

### Appendix III: land cover classification

NUMBER	code	cover type	% tree	% shrub	% grass	% bare	Dominant species	Description
64	1	Dense forest	90	0	Ő	10	Carpinus petulus	
17	1	Dense forest	90	0	0	10	Carpinus petulus	
76	1	Dense forest	88.8	11.2	0	0	Quercus species	
74	1	Dense forest	97.4	0	1.8	0.8	Pinus Brutia	Dense forest: tree cover ranging from 50 - 100%
43	1	Dense forest	64	30	6	0	Fraxinus ornus	
28	1	Dense forest	100	0	0	0	Fagus orientalis	
27	1	Dense forest	70	28	0	2	Pinus Brutia	
30	1	Dense forest	56	44	0	0	Pinus Brutia	
6	2	Open forest	34	63	0	3	Carpinus petulus	
56	2	Open forest	16.4	0	69.8	13.8	Quercus species	
77	2	Open forest	49.2	36.8	0	14	Quercus species	
40	2	Open forest	35	41	4	20	Qercus pubescerns	
41	2	Open forest	38	32	0	30	Pinus Brutia	
4	2	Open forest	18	10	72	0	Quercus penduncularis	
31	2	Open forest	22	2	66	6	Carpinus petulus	Open forest: tree cover ranging from 10 - 50%
3	2	Open forest	48	0	37.4	14.6	Pinus Brutia	
19	2	Open forest	30	36	34	0	Pinus Brutia	
78	2	Open forest	30	34	34	2	Quercus Sessilifrova	
42	2	Open forest	28	0	68.6	3.4	quercus species	
38	2	open forest	22	6	42	30	Quercus penduncularis	
47	2	Open forest	40	8	38	14	Quercus pubescens	
2	2	Open forest	36	48	0	16	Carpinus petulus	
44	2	Open forest	44	26	15	5	Juniperus oxycedrus	
66	3	Dense shrubland	0	96.6	0	3.4	Quercus coccifera	
50	3	Dense shrubland	0	81	0	19	Quercus coccifera	
13	3	Dense shrubland	0	70	30	0	Quercus coccifera	
1	3	Dense shrubland	0	100	0	0	Quercus coccifera	dense shrub land: shrub layer ranging from 50 - 100%
79	3	Dense shrubland	0	100	0	0	Quercus coccifera	
70	3	Dense shrubland	10	89	0	1	Quercus coccifera	
37	3	dense shrubland	0	78	22	0	Quercus coccifera	
71	3	Dense shrubland	0.7	72.6	23.3	3.4	Quercus coccifera	
69	4	Open shrubland	2.6	38	27	32.4	Erica arborea	
36	6	Open shrubland	0	6	0	94	Paliurus spina-christi	
51	4	Open shrubland	8.6	26	0	65.4	Paliurus spina-christi	Open shrub land: shrub layer ranging from 10 - 50%
75	4	Open shrubland	0	46	10	44	Paliurus spina-christi	
55	4	Open shrubland	0	48	52	0	olea europea	
68	4	open shrubland	0	40	60	0	Quercus coccifera	
29	5	grassland	0	0	100	0	Tobacco	
14	5	grassland	0	0	71	29	poa species	
53	5	grassland	0	0	100	0	Cereal	
62	5	grassland	0	0	100	0	Cereal	
65	5	grassland	0	0	100	0	Cereal	
18	5	grassland	0	0	100	0	Cereal	
23	5	grassland	0	0	100	0	Cereal	
80	5	grassland	0	0	100	0	Cereal	
16	5	grassland	0	0	100	0	Cereal	Grassland: grass layer ranging from 50 - 100%
20	5	grassland	0	0	100	0	Cereal	
59	5	grassland	1.2	0.8	91.6	6.4	Cereal	
81	5	grassland	0	0	100	0	Cereal	
48	5	grassland	0	0	100	0	Cereal	
12	5	grassland	0	0	100	0	Cereal	
5	5	grassland	0	0	100	0	Cereal	
24	5	grassland	0	0	100	0	Cereal	
26	5	grassland	0	0	100	0	Cereal	
67	5	grassland	0	0	100	0	Cereal	
73	5	grassland	0	0	100	0	Cereal	
61	5	grassland	0	0	100	0	Maize	

## Appendix IV: Table showing secondary data used

Data	Description	year	Resolution	source
ETM+ Images	ETM+ Images Mosaic of two		30 x 30 m	GLCF
	scenes of Landsat 7			
NDVI Images MODIS 16 day		January 2001 to	250 x 250 m	https://wist.echo.nasa.gov/api/
	composites	December 2008		
Environmental	Temperature,	2009	1000 x 1000 m	Biofrag <sup>1</sup> database
layers				
	distance to roads,			
	distance to urban			
DEM Aster derived DE		2009	30 x 30 m	METI/NASA
Greece Shape file in UTM		2009	N/A	Biofrag database
boundary 35N projection				
Map of Greece	Map of Greece Topographic map of		1:700,000 cm	Book shop
	Greece			

### Appendix V: Results of multi-collinearity Test

	Coefficients <sup>a</sup>										
		Unstandardized Coefficients		Standardized Coefficients			Collinearity Statistics				
M	odel	В	Std. Error	Beta	t	Sig.	Tolerance	VIF			
1	(Constant)	4.000	1.026		3.897	.000					
	PRE_AUG	034	.010	625	-3.360	.001	.231	4.327			
	PRE_FEB	003	.002	168	-1.495	.139	.634	1.577			
	PRE_JUN	003	.010	053	298	.766	.254	3.935			
	TMEAN_APR	010	.004	636	-2.483	.015	.122	8.201			
	TMEAN_JAN	020	.006	850	-3.515	.001	.137	7.312			
	DIST_ROAD	2.053E-5	.000	.117	1.094	.277	.699	1.430			
	DIST_URBAN	-6.196E-6	.000	153	-1.632	.107	.906	1.103			
	ASPECT	.000	.001	084	751	.455	.633	1.579			
	ELEVATION	001	.000	875	-2.911	.005	.089	11.286			
	SLOPE	.015	.011	.236	1.428	.157	.293	3.408			

**Coefficients**<sup>a</sup>

a. Dependent Variable:

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<sup>1</sup> Biodiversity in Fragmenting Landscapes; an ITC research theme