

# A frog's-eye view of the landscape

Quantifying connectivity for  
fragmented amphibian populations

Promotor: Dr. H.H.T. Prins  
*hoogleraar in het natuurbeheer in de tropen  
en de ecologie van vertebraten*

Co-promotor: Dr. J.W. Arntzen  
*senior wetenschappelijk onderzoeker  
Universiteit van Porto, Portugal*

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# A frog's-eye view of the landscape

Quantifying connectivity for  
fragmented amphibian populations

Claire C. Vos

## PROEFSCHRIFT

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## Stellingen

1. Alle soorten ondervinden negatieve effecten van versnippering; bij welke mate van versnippering van het habitat dit gebeurt is afhankelijk van soortspecifieke eigenschappen. (*dit proefschrift*).
2. Er bestaat geen neutrale landschapsmaat waarmee het duurzaam overleven van soorten in het landschap kan worden voorspeld; dit is echter wel mogelijk wanneer landschapsmaten ecologisch worden geschaald. (*dit proefschrift*).
3. Een connectiviteitsmaat voor grondgebonden organismen dient zowel de afstand als de weerstand van het landschap tussen leefgebieden te omvatten (*dit proefschrift*).
4. De wisselwerking tussen empirisch onderzoek en modelsimulatie is binnen de landschapsecologie een sleutelfactor voor de ontwikkeling van zowel theorie als beleidsinstrumenten (*dit proefschrift*).
5. Bij de ontwikkeling naar steeds realistischer connectiviteitsmaten is het de kunst zich te beperken tot die aspecten van het landschap die de dispersie wezenlijk beïnvloeden.
6. Omdat de ruimtedruk in de toekomst alleen maar verder zal toenemen zal het Nationaal Ecologisch Meetnet moeten worden aangepast voor de toetsing van ontsnipperingsbeleid.
7. Investeren in lange tijdreeksen is onontbeerlijk om de betrouwbaarheid van modelvoorspellingen te verbeteren.
8. Hoe meer lange afstandswandelaars, hoe beter de doorlaatbaarheid van het landschap voor fauna.
9. Nu de vrouwenbeweging redelijk geslaagd is in haar streven naar een evenredige arbeidsparticipatie, wordt het tijd dat er een mannenbeweging opstaat die het recht op een evenredige verdeling van zorgtaken opeist.
10. Its better to be wrong than to be confused (*after Francis Bacon*).

*Stellingen behorende bij het proefschrift van Claire Vos:*

*A frog's-eye view of the landscape; quantifying connectivity for fragmented amphibian populations.*

*Wageningen, 5 november 1999.*

## Voorwoord

Dit proefschrift is gebaseerd op onderzoek uit de periode 1991 tot 1999. Een prettig lange periode, waarin kennisontwikkeling en toepassing van die kennis in het beleid elkaar hebben afgewisseld. Deze combinatie heb ik als zeer inspirerend ervaren. Enerzijds zijn de ontwikkelingen binnen het ruimtelijk natuurbeleid richtinggevend geweest voor het onderzoek, anderzijds hebben de onderzoekresulaten bijgedragen aan oplossingsrichtingen binnen de natuurbescherming. Ook persoonlijk spreekt deze combinatie mij aan omdat er ruimte was voor zowel mijn nieuwsgierigheid als maatschappelijke betrokkenheid. Mijn interesse voor de landschapsecologie is gewekt tijdens mijn studie aan de Universiteit van Amsterdam. Met name het holistische karakter van de landschapsecologie, waarbij de mens deel uitmaakt van het (eco)systeem en niet alleen een toeschouwer is, sprak mij aan. In de praktijk is mij echter gebleken dat een holistische onderzoekaankpak voor het ontrafelen van het functioneren van (eco)systemen niet succesvol is. Afdalen in onderdelen van het systeem, een zeker reductionisme, is m.i. onontbeerlijk om de processen te achterhalen die bepalend zijn voor patronen op een landschapsniveau. Daarbij is het echter essentieel om de vertaalslag te maken van kennis van deelprocessen naar de effecten op het hogere niveau van het landschap. Toegepast, probleemoplossend onderzoek dwingt deze 'holistische' vertaalslag naar het landschap als geheel af.

Het hier gepresenteerde onderzoek is een product van teamwork. De breedte van het onderzoekveld van de landschapsecologie vraagt om een scala van kennis en vaardigheden, die onmogelijk in één persoon verenigd kunnen worden. Daarnaast is dit proefschrift het resultaat van het functioneren binnen het inspirerende team van collega's van de afdeling Ecologie en Ruimte. Hierbij bedank ik alle mensen die hebben bijgedragen aan de totstandkoming van dit proefschrift. Er zijn een aantal mensen die ik persoonlijk wil noemen. Allereerst hebben Paul Chardon en Wim Nieuwenhuizen een belangrijke bijdrage geleverd aan het verzamelen en analyseren van de gegevens. De nauwe samenwerking met theoretisch ecologen, in de persoon van Jana Verboom en Hans Baveco, heb ik als bijzonder inspirerend ervaren. Cajo ter Braak en Paul Goedhart hebben een onmisbare rol gespeeld bij de statistische ontrafeling van soms weerbarstige data. Henk Meeuwse, Harm Houweling en René Jochem zijn onmisbaar geweest voor de modelsimulaties en de ruimtelijke analyses met GIS. Ook de mensen die betrokken waren bij het veldwerk wil ik bedanken. Met Paul Chardon, Theo van der Sluis, Wim Nieuwenhuizen, Rob Bugter, Robbert Snep, Harry Bussink en vele studenten deel ik herinneringen aan de vaak nachtelijke expedities, waarbij we luisterden naar het zachte geluid van de heikikker dan wel de gangen van boomkickers probeerden te achterhalen. Ton Stumpel wil ik bedanken voor het ter beschikking stellen van gegevens van de boomkikker in Zeeuws-Vlaanderen. De door hem en vele studenten verzamelde verspreidingsgegevens zijn zeer waardevol gebleken. Ook wil ik René Smulders van het CPRO-DLO noemen, die een stimulerende rol heeft gespeeld bij de integratie van het landschapsecologisch en genetisch onderzoek. De stimulerende discussies met mijn promotor Herbert Prins en co-promotor Pim Arntzen hebben sterk bijgedragen aan de kwaliteit van dit proefschrift. Thom van Rossum wil ik bedanken voor zijn geduld bij het corrigeren van de vele tekstuele onvolkomenheden in mijn werk. Mieke Pijfers en Janneke Schenk bedank ik voor de vormgeving van respectievelijk de binnen- en de buitenkant van dit boekwerk. Tenslotte wil ik Paul bedanken voor zijn steun en vertrouwen.

*Aan mijn moeder*

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# 1 Introduction

Ongoing intensity of land use, urbanisation and increasing mobility have put high pressure on remaining natural ecosystems. When conservation areas become increasingly fragmented, the spatial configuration of the remaining areas is an important factor for species richness. In the beginning of the nineties, the National Ecological Network (NEN) has been devised, to stop or even reverse the process of habitat fragmentation (NPP 1990). To assess whether the NEN will result in a sustained protection of species diversity, quantitative knowledge of the spatial functioning of populations is needed. In this thesis I aim to develop new ideas about the spatial functioning of amphibian populations. Amphibian species can be taken as representatives of relatively immobile ground-dwelling species. The results will contribute to the underpinning of the spatial requirements for sustainable habitat networks of amphibian species. In this thesis I have formulated spatial standards for sustainable amphibian habitat networks that can be applied in landscape management. A second aim of this study is to develop general standards for sustainable habitat networks for species groups that have comparable responses to habitat fragmentation.

The amount and spatial configuration of suitable habitat in a landscape form the basis for the distribution of species. In addition, species-specific characteristics such as dispersal ability and population dynamical aspects determine whether a species suffers from habitat fragmentation. Fragmentation is the result of the destruction of habitat suitable for a species, resulting in small fragments or patches that are separated by unsuitable habitat or barriers. Many species have a naturally disjunct distribution pattern, associated with the heterogeneity of the landscape. For amphibians that require both terrestrial and aquatic habitat during their life cycle, migrations between spatially separated habitats form a natural aspect of their life history. If, however, habitat fragmentation is the result of humanly induced changes in land use, it is the question to what extent species will be capable to cope with this fragmentation of their habitat.

Habitat fragmentation has two principal components: decrease in habitat area and increase in isolation (or decrease in connectivity) of the remaining habitat patches (Hanski & Gilpin 1991; Opdam et al. 1993). The metapopulation theory (Levins 1970) forms the basis to describe the spatial functioning of species in fragmented habitat. Metapopulation theory implies that although small populations suffer from chance extinction due to demographic stochasticity, a species can survive on a regional level if local extinctions are compensated for by recolonisations (Levins 1970). Connectivity between habitat patches depends on the landscape characteristics that determine the amount of individuals that are capable to disperse between patches. For ground-dwelling species with low dispersal capacity, such as amphibians, connectivity not only depends on the distance between suitable habitat patches but also on the relative resistance of the landscape (Forman & Godron 1986; Merriam 1991; Saunders & Hobbs 1991; Bennett et al. 1994; Wiens 1997; Bennett 1999). Especially in highly human-dominated landscapes where boundaries between habitat patches are sharp, the variation in permeability for moving organisms may be large. Since the first definition (Levins 1970) the metapopulation concept has evolved and diversified (e.g. Hanski & Simberloff 1997;

Harrison 1994; Harrison & Taylor 1997). I think that from a nature conservation point of view it makes sense to include the viability criterion into the definition, following Shaffer's (1981) definition of a viable population. A viable metapopulation (Verboom 1996) is then: '*a set of local populations distributed over a spatially discontinuous habitat network and linked by dispersal, the size of which as well as the balance between local extinction and recolonisation are such that its probability of extinction within 100 years is 5% or less*'.

Depending on the degree of fragmentation and the resistance of the landscape between suitable habitat, several stages in habitat fragmentation with corresponding species responses can be distinguished (Hanski & Gilpin 1991; Verboom et al. 1993; Reich & Grimm 1996). A first stage in fragmentation occurs when a continuous landscape breaks down into separate habitat fragments. If the habitat patches are connected by a sufficient dispersal stream of individuals, the connectivity within the network will prevent local extinctions. The species in the landscape still functions as one population. A further increase in fragmentation reduces the connectivity between populations which results in independent local population dynamics: chance extinctions of small local populations will become more frequent. As a consequence, a proportion of the suitable habitat patches will be unoccupied. If local extinctions are compensated for by recolonisation, the network of local populations will persist. The species shows the spatial dynamics characteristic of metapopulations. In a final stage of habitat fragmentation a species drops below its viability threshold. Distances between local populations can no longer be bridged by dispersing individuals and populations have become completely isolated. As recolonisations occur no longer, the species is threatened with extinction on a regional level.

The central question in this study is under what conditions (amphibian) populations can survive in fragmented habitat. I focus on two amphibian species, the tree frog (*Hyla arborea*) and the moor frog (*Rana arvalis*). Earlier studies demonstrated negative effects of habitat fragmentation for various amphibian species (Loman 1988; Sjögren 1988, 1991; Laan & Verboom 1990; Sinsch 1992; Edenhamn 1996). As distances between suitable habitat patches have become large in relation to the dispersal capacity of these relatively immobile species, emphasis in this study is put on the quantification of connectivity. The two species differ regarding the degree of fragmentation of their habitat. The habitat of the tree frog consists of remnants of (semi)natural vegetation situated in agricultural landscapes. This habitat is highly fragmented and as agricultural landscapes are often dynamic, the high 'turnover' of (semi)natural elements causes additional stress. The habitat of the moor frog is less fragmented. It consists of moorlands situated in conservation areas. In chapters 2 and 3, I analyse the distribution pattern of the two species, as a first indication whether the species show negative effects of habitat fragmentation. In chapter 3, I analyse a possible additional isolation effect of roads, as roads are known to be barriers for ground-dwelling species, among which amphibians (e.g. Van Gelder 1973; Heine 1987; Vos & Chardon 1994; Hels 1999).

A single observed distribution pattern of a species is only a snapshot of a dynamic series of possible distributions and it gives no information about presumed underlying processes of extinction and recolonisation. Therefore, in chapter 5, I analyse time series and observed dispersal events of the tree frog, to test whether it is plausible that metapopulation processes play a dominant role in explaining the

distribution of the tree frog in the landscape. This is a relevant question from a nature conservation point of view, as it decides whether improvements on the spatial configuration of suitable habitat will indeed be effective for species persistence. I analyse whether extinction, recolonisation and dispersal between local populations actually occur and if these are related to the spatial configuration of habitat in the landscape. To determine whether the habitat network as a whole is viable, a model is developed simulating local extinction and colonisation processes for all patches over time.

Chapters 4 and 6 focus on the further quantification of connectivity. As successful dispersal is essential for the long-term survival of a species in fragmented habitat, the quantification of dispersal and the relation between dispersal and landscape heterogeneity are vital prerequisites for effective conservation. Especially long distance dispersal might be very important in metapopulations but is very difficult to measure by direct methods (Crochet 1996; Turchin 1998). Therefore in chapter 4, I apply genetic techniques as indirect indicators of successful dispersal between populations of the moor frog in the present and past landscape configuration. It is determined whether genetic differentiation between local populations exists, and whether genetic similarity is related to geographical distance and the heterogeneity of the landscape. If the movement patterns of dispersing amphibians are considerably influenced by the landscape mosaic between habitat patches, the implementation of dispersal corridors or connectivity zones may be an effective nature conservation measure. In chapter 6, I study the influence of the agricultural landscape on the movement patterns of individual tree frogs by radio telemetry, to determine preference and avoidance of different habitat types. A simulation model is developed to extrapolate individual movement paths on a small scale to differences in the amount of dispersal between populations on a landscape scale.

In the last chapter I propose a general framework to predict viability for a broad array of species. Landscape managers and nature conservation planners need tools that link landscape characteristics to some critical measure of the sustainable conservation of species diversity. I formulate key characteristics that explain differences between species in their response to landscape change and translate these into ecologically-scaled landscape indices that predict species viability.

## 2 Comparison of habitat-isolation parameters in relation to fragmented distribution patterns in the tree frog (*Hyla arborea*)

### Summary

The distribution pattern of the tree frog (*Hyla arborea*) in an intensively used agricultural landscape in Zeeland Flanders, was analysed for effects of habitat fragmentation. The logistic regression models showed that the chance that a pond (potential reproduction site) was occupied by tree frogs depended on three isolation factors. The density of ponds within 750 m of the occupied pond was higher compared to ponds that remained unoccupied during the survey period. Additionally both the density of shrubs as well as the density of high herbs, two terrestrial habitat factors, was higher within 1000 m of occupied ponds. The explanatory value of two types of isolation measure was compared with logistic regression analysis. It is discussed that 'concentric isolation measures', which take size and distance of potential habitat patches in all directions into account, are expected to give a better description of isolation than the more often used 'distance from the nearest habitat patch'.

### Introduction

Habitat fragmentation is being recognised as an important cause of loss of biodiversity in intensively used agricultural landscapes (see among others Opdam et al. 1993). With an increasing loss and disintegration of habitat, local populations will become smaller, and the frequency of interpatch dispersal will decrease. Depending on species characteristics and the stage of fragmentation, populations especially in small habitat patches, may go extinct, while the degree of isolation lowers the probability that unoccupied patches become recolonised (Verboom et al. 1993). If the process of fragmentation continues, opportunities for recolonisation of empty patches will diminish and a species may eventually disappear from the region. Characteristically species whose distribution patterns are influenced by habitat fragmentation, are often absent in relatively small and isolated habitat patches. Effects of habitat size and isolation on the distribution of populations have been demonstrated for a variety of species of different species groups: woodland birds (Opdam 1991; Askins et al. 1987), small mammals (Merriam 1988; Van Apeldoorn et al. 1992), butterflies (Harrison 1991; Hanski 1994), ground-dwelling invertebrates (Mabelis 1990). Furthermore, for amphibians, a group with relatively limited mobility and often dependent on natural elements in the agricultural landscape, effects of habitat fragmentation have been determined for several species: the pool frog (Sjögren 1988, 1991), the natterjack toad (Sinsch 1992). Moreover, Laan & Verboom (1990) found an effect on species diversity.

In habitat fragmentation studies, parameters that describe the spatial distribution of habitat, the isolation parameters, are often nearest neighbour measures (here called 'linear measures') e.g.: distance from the nearest suitable habitat patch (among others: birds: Müller 1982; Van Dorp & Opdam 1987; Soulé et al. 1988; butterflies: Thomas et al. 1992; amphibians: Dickman 1987; Wederkinch 1988; Laan & Verboom 1990) or the distance from the nearest occupied habitat patch or source patch (among others: birds: Fritz 1979; Van Dorp & Opdam, 1987; Soulé et al. 1988; butterflies: Harrison et al. 1988; Thomas & Harrison 1992; Thomas et al. 1992; amphibians: Sjögren 1991). However, we suggest that the distance from the nearest patch is an inadequate descriptor of the spatial relations between local populations. Being potential sources of dispersing animals all (occupied) habitat patches within a reachable distance contribute to the probability that a patch is being reached by a dispersing individual (Verboom et al. 1991a; Hanski 1994). Isolation measures that take the total area of habitat in the patch surroundings into account are here called 'concentric isolation measures'. Few studies have considered concentric isolation measures: birds (Askins et al. 1987; Verboom et al. 1991a); butterflies (Hanski 1994); amphibians (Loman 1988). A concentric isolation measure has the advantage that size and distance of potential habitat patches in all directions are taken into account. This is probably a more realistic description of isolation in relation to actual dispersal processes, presuming that dispersing animals have no foreknowledge of the direction of the nearest habitat patch and therefore leave in a random direction.

A second reason to prefer concentric isolation measures over linear measures is that they provide quantitative and spatially explicit information on the habitat requirements of species. This is important for the development of tools for nature conservation. In recent nature conservation policy plans in The Netherlands the problem of habitat fragmentation has been recognised. This has led to spatially explicit landscape plans in which sustainable ecological networks of core areas, nature restoration areas and interconnecting corridors have been designed (NPP 1990). For the evaluation of these landscape plans, quantitative data on the amount of required habitat and the optimal spatial distribution of habitat of the target species are needed.

Our first objective in this study was to analyse whether the spatial characteristics of tree frog habitat play a role in explaining the distribution pattern of the tree frog in an intensively used agricultural landscape. The tree frog has declined considerably in number over the last decades (Borgula 1993) and it is one of the target species in nature restoration plans. There is a need for measurements by which the effects of these plans can be predicted. The chance that a habitat patch is occupied by a local population of tree frogs is related to habitat quality, size and degree of isolation of the patch. Our second objective was to determine whether concentric isolation measures give a better explanation of the distribution pattern than linear isolation measures. Finally, the translation of concentric and linear isolation measures into tools for landscape planning is discussed.

## Study area

The study area is 250 km<sup>2</sup> and is situated in the western part of Zeeland Flanders (Fig. 2.1). The landscape mainly consists of arable lands. (Semi)natural vegetations can be found on dikes, in the coastal sand dunes and in meadows with cattle drinking ponds, the aquatic habitat used by tree frogs. The terrestrial habitat required by tree frogs consists of shrubs, bushes and vegetation of high herbs (Clausnitzer 1986; Stumpel 1993). The habitat patches form approximately 1.5 % of the total landscape cover and are isolated from each other by intensively used agricultural land that is unsuitable for the species.

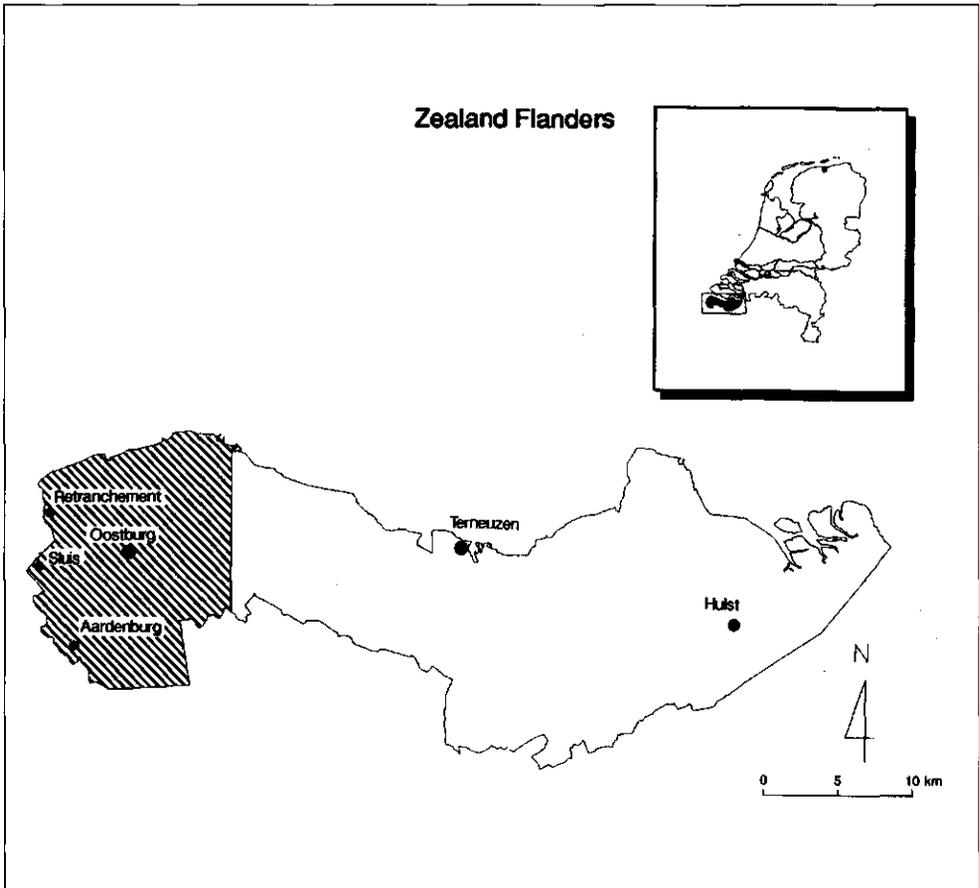


Figure 2.1  
The position of the study area.

## Methods

Presence and absence data of the tree frog were collected from 1981 to 1986 for 512 cattle drinking ponds. Tree frog presence was recorded by listening for calling males (spontaneously or in reaction to taperecorded mating calls) and by searching for eggs, tadpoles, juveniles and adults in the pond and the pond surroundings (see Stumpel 1987a for detailed information). The ponds were visited at least three times a year (in the years 1981-1983 and 1986) during the reproduction period. Since not all tree frogs call every night and because of the low chance of tracing animals in ponds with very low densities (Stumpel 1987a; Tester 1993) tree frogs may have been recorded erroneously as absent in ponds with very few animals. All occupied ponds were treated equally in further analysis, regardless of tree frog density or whether successful reproduction could be confirmed.

In 1986, habitat quality parameters and pond area were measured, in a subset of 196 ponds (Table 2.1). The subset contained all 50 ponds where tree frogs were found in at least one year. As brackish water is an unsuitable habitat for reproduction (Stumpel 1987b; Grosse 1993), ponds with a chloride concentration higher than 500 mg/l were removed from the dataset. Habitat isolation measures were calculated for the complete data set of 512 ponds in the study area. This led to a total number of 187 ponds for analysis of which both habitat quality and isolation parameters were available. To test whether the subset was a representative sample of the variation in isolation, regression analysis was also carried out with the complete dataset of 512 ponds in the study area (see Results).

In 1993 a survey of the terrestrial habitat in the study area was made. Two types of terrestrial tree frog habitat were distinguished: sites with a vegetation of high perennial herbs and sites with a developed shrub layer, such as hedgerows and wood fringes (Clausnitzer 1986; Stumpel 1993). For linear sites of high herbs only those elements with a minimum width of 5 m were taken into account. Based on a control of the vegetation map in the field from 1986 and considering the minimal width of the elements, we assumed that no substantial changes had taken place in the distribution pattern of these terrestrial habitats in the years between the tree frog inventory and the survey of the terrestrial habitat. Therefore, the data sets were analysed together. The spatial configuration of water and land habitat was put in a geographic information system Arc-Info (Fig. 2.2).

The following measures for the isolation of the ponds were calculated. Linear variables: distance from the nearest pond and distance from the nearest occupied pond. Concentric variables were calculated in circles surrounding each pond with an increasing radius from 100, 250, 500, 750, 1000, 1500 and 2000 m. In these zones the following variables were measured: total number of ponds (both occupied and unoccupied ponds), number of occupied ponds, area of high herbs and area of shrubs (Table 2.1).

The presence or absence data of tree frogs in a pond were analysed with logistic regression analysis, in which a pond was judged 'occupied' if in at least one out of six years, tree frog presence was recorded. An 'occupied' pond is also referred to as a 'tree frog pond'. Thus, we analysed the spatial pattern of occupancy. The temporal pattern was judged less informative because of potential variation in search intensity over the six years of study. The regression analysis was carried out in two

Table 2.1

Mean value and standard deviation of water quality variables, pond area and isolation variables.

| VARIABLES                           |   | OCCUPIED PONDS<br>MEAN $\pm$ SD | UNOCCUPIED PONDS<br>MEAN $\pm$ SD |
|-------------------------------------|---|---------------------------------|-----------------------------------|
| <b>Pond Variables</b>               |   |                                 |                                   |
| PH                                  | pH pond water                                       | 7.6 $\pm$ 0.8                   | 7.7 $\pm$ 0.9                     |
| COND                                | Electrical Conductivity pond water<br>( $\mu$ S/cm) | 83.2 $\pm$ 33.8                 | 88.9 $\pm$ 40.7                   |
| CHLOR                               | Chloride (mg/l)                                     | 51.9 $\pm$ 37.4                 | 65.6 $\pm$ 64.3                   |
| COVER                               | Water Vegetation cover (%)                          | 50% $\pm$ 40%                   | 20% $\pm$ 30%                     |
| SHADE                               | Shaded pond area (%)                                | 10% $\pm$ 20%                   | 20% $\pm$ 30%                     |
| AREA                                | Pond area (m <sup>2</sup> )                         | 511 $\pm$ 644                   | 359 $\pm$ 405                     |
| <b>Linear Isolation Variables</b>   |   |                                 |                                   |
| OCCDIST                             | Distance to nearest occupied pond (m)               | 390 $\pm$ 606                   | 1441 $\pm$ 1088                   |
| PONDDIST                            | Distance to nearest pond (occupied<br>or empty)     | 153 $\pm$ 124                   | 232 $\pm$ 192                     |
| <b>Circular Isolation Variables</b> |   |                                 |                                   |
| OCC100                              | Number of occupied ponds within 100 m               | 0.2 $\pm$ 0.5                   | 0.0 $\pm$ 0.2                     |
| OCC250                              | ditto 250 m   | 1.0 $\pm$ 1.1                   | 0.2 $\pm$ 0.6                     |
| OCC500                              | ditto 500 m   | 2.7 $\pm$ 1.9                   | 0.7 $\pm$ 1.5                     |
| OCC750                              | ditto 750 m   | 4.8 $\pm$ 3.2                   | 1.7 $\pm$ 3.1                     |
| OCC1000                             | ditto 1000 m  | 6.3 $\pm$ 4.3                   | 1.7 $\pm$ 3.1                     |
| OCC1500                             | ditto 1500 m  | 7.0 $\pm$ 4.5                   | 2.6 $\pm$ 3.7                     |
| OCC2000                             | ditto 2000 m  | 8.2 $\pm$ 4.7                   | 3.8 $\pm$ 4.6                     |
| POND100                             | Total number of ponds within 100 m                  | 0.5 $\pm$ 0.6                   | 0.3 $\pm$ 0.5                     |
| POND250                             | ditto 250 m   | 2.6 $\pm$ 1.7                   | 1.7 $\pm$ 1.7                     |
| POND500                             | ditto 500 m   | 7.8 $\pm$ 3.6                   | 4.8 $\pm$ 3.7                     |
| POND750                             | ditto 750 m   | 14.7 $\pm$ 6.8                  | 8.4 $\pm$ 5.3                     |
| POND1000                            | ditto 1000 m  | 20.9 $\pm$ 10.4                 | 12.7 $\pm$ 7.5                    |
| POND1500                            | ditto 1500 m  | 32.4 $\pm$ 13.8                 | 22.5 $\pm$ 11.3                   |
| POND2000                            | ditto 2000 m  | 45.0 $\pm$ 16.6                 | 35.9 $\pm$ 15.0                   |
| HERP100                             | Area of high herbs (ha) within 100 m                | 0.20 $\pm$ 0.41                 | 0.06 $\pm$ 0.18                   |
| HERP250                             | ditto 250 m   | 0.85 $\pm$ 0.93                 | 0.31 $\pm$ 0.45                   |
| HERP500                             | ditto 500 m   | 2.03 $\pm$ 1.51                 | 0.90 $\pm$ 0.95                   |
| HERP750                             | ditto 750 m   | 3.14 $\pm$ 1.85                 | 1.74 $\pm$ 1.50                   |
| HERP1000                            | ditto 1000 m  | 4.60 $\pm$ 2.07                 | 2.89 $\pm$ 1.20                   |
| HERP1500                            | ditto 1500 m  | 7.13 $\pm$ 1.98                 | 5.68 $\pm$ 2.73                   |
| HERP2000                            | ditto 2000 m  | 10.66 $\pm$ 2.70                | 9.62 $\pm$ 3.17                   |
| SHRUB100                            | Area of shrubs (ha) within 100 m                    | 0.13 $\pm$ 0.22                 | 0.11 $\pm$ 0.37                   |
| SHRUB250                            | ditto 250 m   | 0.76 $\pm$ 1.01                 | 0.39 $\pm$ 1.12                   |
| SHRUB500                            | ditto 500 m   | 1.98 $\pm$ 2.26                 | 0.82 $\pm$ 1.72                   |
| SHRUB750                            | ditto 750 m   | 3.52 $\pm$ 3.22                 | 1.40 $\pm$ 2.32                   |
| SHRUB1000                           | ditto 1000 m  | 5.08 $\pm$ 4.10                 | 2.18 $\pm$ 3.12                   |
| SHRUB1500                           | ditto 1500 m  | 7.26 $\pm$ 4.89                 | 4.09 $\pm$ 4.32                   |
| SHRUB2000                           | ditto 2000 m  | 9.75 $\pm$ 5.56                 | 6.45 $\pm$ 5.15                   |

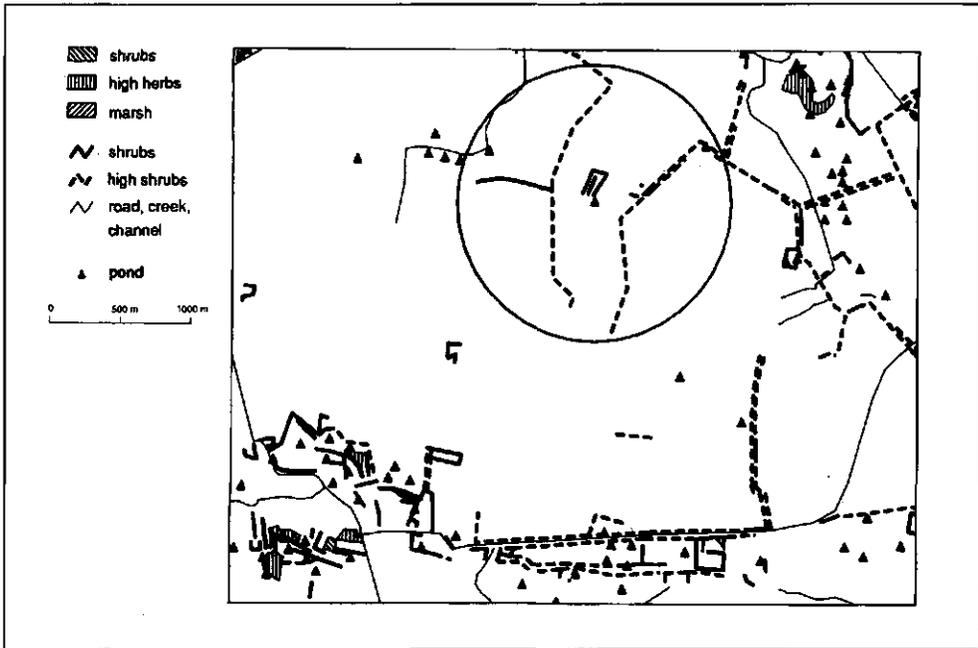


Figure 2.2

Detail of the digitised landscape elements. The circle has a radius of 1000 m.

stages. In the first stage only habitat quality parameters of the ponds were entered in the model in a stepwise fashion. In the second stage the model was extended with isolation parameters. Whether isolation parameters are additional explanatory factors to the habitat quality factors can be tested using this method (Van Apeldoorn et al. 1992). For each concentric isolation variable the optimal radius was determined by the variable with the highest explanatory value in the regression model, as judged on the basis of the deviance statistic (Jongman et al. 1995).

## Results

During the six-year survey period 50 ponds were occupied by tree frogs in at least one year. Only a few ponds were occupied during the whole period. The distribution pattern of occupied ponds is shown in Figure 2.3.

Analysis of the habitat quality variables revealed vegetation cover and electrical conductivity of the pond water as significant variables, while shading and acidity of the pond showed no effect (Table 2.1). A high coverage of the pond surface by aquatic vegetation had a positive effect on the occupation probability (COVER,  $p < 0.001$ , Table 2.2 model 1a). The vegetation mainly consisted of floating and submerged species: *Glyceria fluitans*, *Ranunculus aquatilis*, *R. baudotii*, *Lemna minor*, *Zannichellia palustris* and *Ceratophyllum submersum* (Stumpel 1987b). The second important variable was electrical conductivity, high values of which had a

Table 2.2

Selection of the logistic regression models. Significance levels of the last variable added to each model and  $R^2$  adjusted of the total model are listed (for abbreviations see Table 2.1).

| LOGISTIC REGRESSION MODELS |  | SIGNIFICANCE OF<br>THE LAST VARIABLE | $R^2$ ADJUSTED<br>TOTAL MODEL |
|----------------------------|--|--------------------------------------|-------------------------------|
| Habitat quality variables  | Linear and concentric<br>isolation variables |                                      |                               |
| PH                         |  | ns                                   |                               |
| CHLOR                      |  | ns                                   |                               |
| SHADE                      |  | ns                                   |                               |
| AREA                       |  | ns                                   |                               |
| 1a COVER                   |  | ***                                  | 10%                           |
| 1b COVER + COND            |  | **                                   | 13%                           |
| 2a COVER + COND            | + OCCDIST                                    | ***                                  | 38%                           |
| 2b COVER + COND            | + OCCDIST + HERP1000                         | ns                                   |                               |
| 2c COVER + COND            | + OCCDIST + SHRUB1000                        | ns                                   |                               |
| 3a COVER + COND            | + PONDDIST                                   | **                                   | 18%                           |
| 3b COVER + COND            | + PONDDIST + HERP1000                        | ***                                  | 27%                           |
| 3c COVER + COND            | + PONDDIST + HERP1000 + SHRUB1000            | ***                                  | 34%                           |
| 4a COVER + COND            | + OCC750                                     | ***                                  | 37%                           |
| 4b COVER + COND            | + OCC750 + HERP1000                          | ns                                   |                               |
| 4c COVER + COND            | + OCC750 + SHRUB1000                         | ns                                   |                               |
| 5a COVER + COND            | + POND750                                    | ***                                  | 29%                           |
| 5b COVER + COND            | + POND750 + HERP1000                         | **                                   | 32%                           |
| 5c COVER + COND            | + POND750 + HERP1000 + SHRUB1000             | ***                                  | 37%                           |

ns not significant, \*\*  $p < 0.01$ , \*\*\*  $p < 0.001$

negative effect on the occupation probability of a pond (COND,  $p < 0.01$ , Table 2.2 model 1b). Although electrical conductivity of the water in agricultural landscapes is generally a measure for eutrophication, in Zeeland Flanders it is highly correlated with the concentration of chloride ions in the water ( $r = 0.983$ ,  $n = 187$ ,  $p < 0.01$ ), caused by mixing with salt water from flooding and seepage (Provinciaal Bestuur Zeeland 1993).

After accounting for habitat variation in the next stage of the analysis the variables related to habitat fragmentation and area were added to the model. Although occupied ponds were somewhat larger than unoccupied ponds (occupied ponds: mean  $511 \pm 644$  m<sup>2</sup>, unoccupied ponds mean  $359 \pm 405$  m<sup>2</sup>, Table 2.1), pond area was not a powerful explanatory variable in the model.

First, the linear isolation measures were added to the model. The linear variable 'distance from the nearest occupied pond' (OCCDIST) gave the best explanation of the occupation pattern ( $p < 0.001$ , Table 2.2 model 2a). The difference between unoccupied and occupied ponds regarding their distance from a tree frog pond is shown in Figure 2.4. As is illustrated, nearly all occupied ponds were within

500 m from other occupied ponds, with a threshold value between 500 and 750 m. When the linear variable 'distance from the nearest pond' (occupied as well as unoccupied ponds, PONDDIST) was added to the model ( $p < 0.01$ , Table 2.2 model 3a), the explanatory value was lower than in model 2a. The variables OCCDIST and PONDDIST were correlated ( $r = 0.351$ ,  $n = 187$ ,  $p < 0.01$ ) and therefore exchangeable in the model (Table 2.4).

The results of the analysis of the linear isolation variables on the complete dataset of 512 ponds showed a similar explanation of the occupation pattern (OCCDIST:  $p < 0.001$ , PONDDIST:  $p < 0.001$ ). Therefore, it was concluded that the subset of 187 was a representative sample of the habitat fragmentation in the study area.

The optimal range of the concentric measures was determined by adding the concentric variables to the model separately with increasing ranges from 100 to 2000 m (Table 2.3). The optimal range varied between 750 m for ponds and 1000 m for the terrestrial habitat parts. To test whether concentric isolation measures fit better than linear measures, their contribution as explanatory variables were compared. When the distance from the nearest occupied pond (OCCDIST) was replaced by the corresponding concentric variable 'number of occupied ponds in a circle of 750 m' (OCC750) it gave about the same explanation (compare model 2a and model 4a, Table 2.2,  $R^2$  adjusted of the total model 38% respectively 37%). However, for the total number of ponds (both unoccupied and occupied) in a circle of 750 m (POND750), the concentric measure was an improvement compared to the linear variable (compare model 3a and model 5a, Table 2.2,  $R^2$  adjusted of the total model 18% respectively 29%). As was the case with the linear variables OCCDIST and PONDDIST, the variables OCC750 and POND750 were also correlated ( $r = 0.804$ ,  $n = 187$ ,  $p < 0.01$ ) and therefore exchangeable in the model (Table 2.4).

The concentric isolation measures of the two terrestrial habitat components 'area of high herbs' and 'area of shrubs' had the highest explanatory value in a circle of 1000 m surrounding each pond (HERB1000, SHRUB1000, Table 2.3). When adding these terrestrial measures to the models that already included the linear or concentric tree frog ponds variables, the terrestrial variables added no further explanation (compare model 2b and 2c for the linear tree frog variable and model 4b and 4c for the concentric tree frog variable, Table 2.2). This can be explained by the high correlation of HERB1000 and SHRUB1000 with OCCDIST (respectively  $r = 0.539$ ,  $n = 187$ ,  $p < 0.01$  and  $r = 0.374$ ,  $n = 187$ ,  $p < 0.01$ ) and of HERB1000 and SHRUB1000 with OCC750 (respectively  $r = 0.508$ ,  $n = 187$ ,  $p < 0.01$  and  $r = 0.419$ ,  $n = 187$ ,  $p < 0.01$ , Table 2.4). However, both the area of high herbs and the area of shrubs in a circle of 1000 m do add significantly to the model that already included the distance from the nearest pond (model 3b and 3c,  $R^2$  adjusted of the total model 27% and 34%, Table 2.2) and to the model with the number of ponds in a circle of 750 m (model 5b and 5c,  $R^2$  adjusted of the total model 32% and 37%, Table 2.2).

Table 2.3  
Significance levels and deviances of the concentric isolation measures, added to the model with increasing ranges from 100 to 2000 m (for abbreviations see Table 2.1).

| Habitat quality variables | Number of tree frog ponds | Number of ponds | Area of high herbs               | Area of shrubs                    |
|---------------------------|---------------------------|-----------------|----------------------------------|-----------------------------------|
| COVER + COND +            |                           |                 |                                  |                                   |
| OCC100                    | *** (12.4)                | POND100 **      | HERB100 ns ( 3.8)                | SHRUB100 ns ( 0.0)                |
| OCC250                    | *** (23.0)                | POND250 *       | HERB250 *** (16.6)               | SHRUB250 ns ( 1.3)                |
| OCC500                    | *** (33.5)                | POND500 ***     | HERB500 *** (21.4)               | SHRUB500 ** ( 6.4)                |
| OCC750                    | *** (43.7) <sup>1</sup>   | POND750 ***     | HERB750 *** (21.1)               | SHRUB750 *** (13.4)               |
| OCC1000                   | *** (39.7)                | POND1000 ***    | HERB1000 *** (21.7) <sup>1</sup> | SHRUB1000 *** (15.2) <sup>1</sup> |
| OCC1500                   | *** (31.7)                | POND1500 ***    | HERB1500 ** ( 9.8)               | SHRUB1500 *** (12.3)              |
| OCC2000                   | *** (26.0)                | POND2000 ***    | HERB2000 ns ( 3.1)               | SHRUB2000 ** (10.6)               |

ns not significant, \* p < 0.05, \*\* p < 0.01, \*\*\* p < 0.001

<sup>1</sup> highest deviance

Table 2.4  
Correlation matrix of the model variables. ns not significant, \* p < 0.05, \*\* p < 0.01 (for abbreviations see Table 2.1).

|           |           |           |           |           |          |          |          |           |  |
|-----------|-----------|-----------|-----------|-----------|----------|----------|----------|-----------|--|
| COVER     | *         |           |           |           |          |          |          |           |  |
| COND      | -0.218 ns | *         |           |           |          |          |          |           |  |
| OCCDIST   | -0.256 *  | -0.168 ns | *         |           |          |          |          |           |  |
| PONDDIST  | -0.103 ns | 0.158 ns  | 0.351 **  | *         |          |          |          |           |  |
| OCC750    | 0.143 ns  | -0.060 ns | -0.624 ** | -0.303 ** | *        |          |          |           |  |
| POND750   | 0.090 ns  | -0.168 ns | -0.546 ** | -0.490 ** | 0.804 ** | *        |          |           |  |
| HERB1000  | 0.029 ns  | -0.034 ns | -0.539 ** | -0.172 ns | 0.508 ** | 0.465 ** | *        |           |  |
| SHRUB1000 | 0.225 ns  | 0.023 ns  | -0.374 ** | -0.204 ns | 0.419 ** | 0.260 *  | 0.107 ns | *         |  |
| COVER     |           | OCCDIST   | OCC750    | PONDDIST  | OCC750   | POND750  | HERB1000 | SHRUB1000 |  |

## Discussion

The statistical models show that occupied ponds are less isolated than unoccupied ponds. The density of both aquatic and terrestrial habitat was higher in the surroundings of occupied ponds compared to ponds that remained unoccupied in the six years of the survey period. The many ponds that remained unoccupied, the few ponds that were occupied during the whole survey period (Fig. 2.3) and the fact that occupied ponds were close to other occupied ponds are indications that the distribution pattern of the tree frog in Zealand Flanders can be explained by habitat fragmentation.

When looking at the potential dispersal sources in the surroundings of a pond, the concentric isolation variable 'number of occupied ponds in a circle of 750 m' is no better explanation of the distribution pattern than the distance from the nearest occupied pond (compare model 2 and model 4,  $R^2$  adjusted of the total model 38% respectively 37%, Table 2.2). The presence of at least one dispersal source within 500-750 m seems to be the deciding factor for the occupation chance.

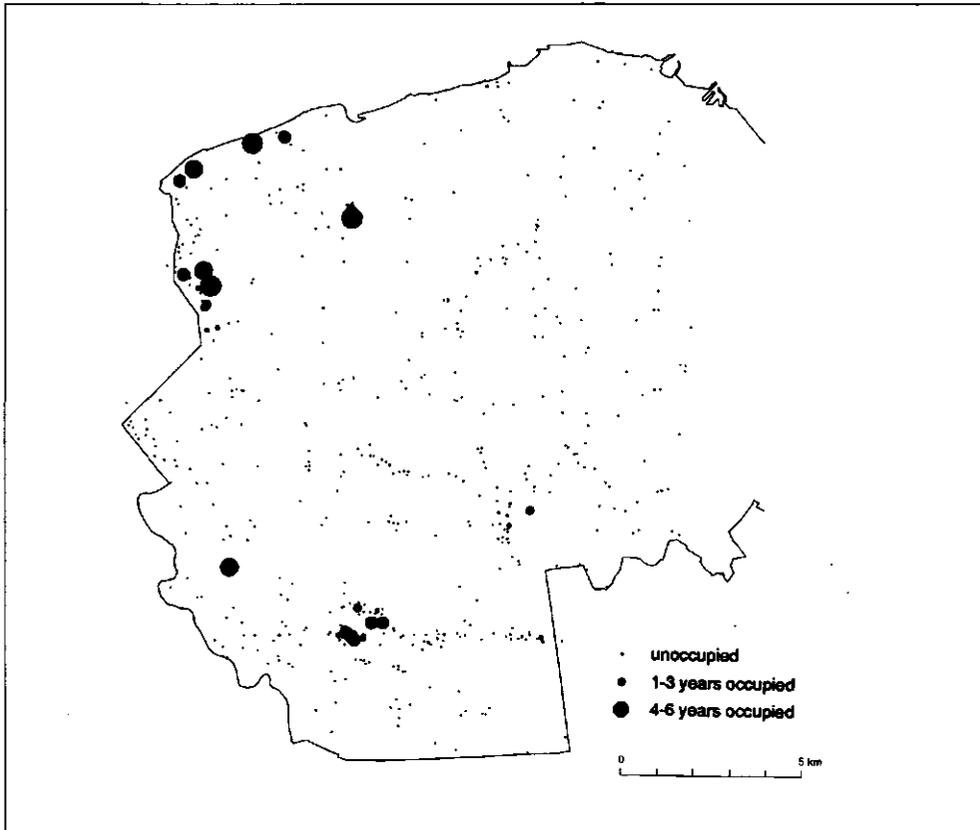


Figure 2.3

Distribution pattern of ponds in the study area. The circle size is related to the number of years of occupancy in the period 1981-1986.

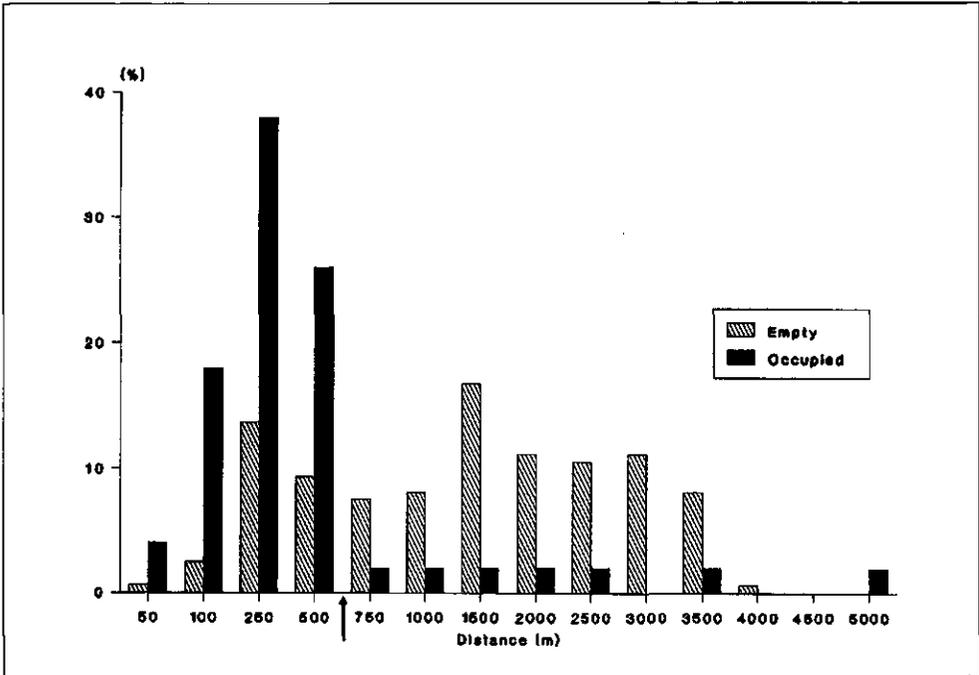


Figure 2.4

Frequency distribution of the distance from the nearest pond for occupied ( $n = 50$ ) and unoccupied ponds ( $n = 137$ ).

The fact that there can be more than one source in the pond's surroundings does not improve this chance. Sjögren (1988, 1991) also found that the distance from the nearest occupied pond was an important explanatory factor for the distribution pattern of the pool frog (*Rana lessonae*). The distances between ponds were larger in the Swedish study area and the landscape between ponds mainly consisted of pine forest instead of arable land.

When the actual sources are ignored and all ponds are taken into account, the concentric compared to the linear measure is an improvement (compare model 3 and model 5,  $R^2$  adjusted of the total model 18% and 29% respectively, Table 2.2). Here the concentric measure is a standard for pond density. In population networks where continuous occupancy of ponds is rare, the overall pond density may be an important measure of the mean presence of dispersal sources over time.

The area of high herbs and bushes is regarded as an isolation variable. However, especially in the direct surroundings of the pond, it will also have a function as the terrestrial habitat part of a local population. In agricultural landscapes the terrestrial and aquatic part of the tree frog habitat are often separated, and the terrestrial habitat is spread over several linear elements. In this respect increasing distances between terrestrial and aquatic habitat will increase the probability of higher mortality rates during the terrestrial phase and seasonal migrations and therefore lower the survival probability of a local population. The

amount of suitable terrestrial habitat surrounding the pond can also be regarded as a connectivity measure, which will increase the colonisation probability of a pond. It is plausible that dispersal will be more successful in a landscape with a high density of suitable terrestrial habitat. The fact that the concentric measure over a distance as far as 1000 m, rather than 100, 250 or 500 m ranges, gives the highest explanation in the model (Table 2.3), is an indication of this last function. The range of the concentric isolation measures (750 m for ponds and 1000 m for terrestrial habitat) is an indication of the distance over which regular exchange among populations is possible.

The terrestrial habitat area has no significant effect when added to a model that includes the number of occupied ponds. This can be explained as follows: because the complete habitat of amphibians consists of an aquatic as well as a terrestrial part, the presence of occupied ponds in the neighbourhood automatically implies that suitable terrestrial habitat must be present. This is also indicated by the relatively strong correlation between these parameters (Table 2.4).

Pond area was only weakly correlated with tree frog presence. However, in a fragmented landscape one would expect a correlation between patch size and occupation change (Levins 1970; Goodman 1987; Verboom et al. 1993). A positive effect of pond area on the occupation probability has indeed been demonstrated for several amphibian species (Loman 1988; Laan & Verboom 1990; Sjögren 1991). One possible explanation for the absence of such an effect is that the variation in pond size in the study area was too small (Table 2.1). Brönmark and Edenhamn (1994) found a negative correlation between the presence of fish and tree frogs, especially in ponds larger than 1000 m<sup>2</sup>. Relatively strong predation pressure in large ponds could reduce the positive effect of pond size on tree frog presence. However, this is not a plausible explanation for the lack of an area effect in this study because only 13 ponds were larger than 1000 m<sup>2</sup>, of which a relatively large number (50%) was occupied by tree frogs. An alternative explanation is that population size is determined not only by the aquatic habitat part but also by the suitable terrestrial habitat area. Although generally the size of the reproduction site is thought to be a limiting factor in amphibian populations (Wilbur 1987; John-Alder & Morin 1990), in an intensively used agricultural landscape the terrestrial habitat might be more determining. If the mortality in the terrestrial phase is high, due to unfavourable conditions, the carrying capacity of ponds will never be reached.

Each pond was treated as a separate potential site for a population. This simplification seems justified by the fact that even in clusters of ponds that are within 100 m of each other, in two-thirds of these clusters only one pond was occupied during the survey period. Still it is not possible to determine exactly the limits of a local population. In an extreme stage of habitat fragmentation a phase can be reached when habitat patches become so small that they can no longer contain a local population. If these small patches are close to each other, several patches will be used by one 'local population' and effects of habitat fragmentation will play a role on the individual level (Haila et al. 1993; Andrén 1994). This is probably the case in the metapopulation study on the natterjack toad (*Bufo calamita*) by Sinsch (1992). Based on the movements of individuals, clusters of 6 to 20 ponds were regarded as one local population. However, in this (Sinsch 1992) study the distances between ponds were much smaller.

### **Potentials for application in landscape planning and evaluation**

Based on the regression model, predictions can be made of the occupation probability of a pond given the amount of suitable terrestrial habitat and number of ponds in the pond's surroundings. Using these predictions, guidelines for the design of an 'optimal tree frog landscape' can be developed and different landscape planning scenarios can be compared (Vos 1993). Concentric isolation measures have better potentials for application compared to linear measures, because they can be translated into habitat density requirements such as 'pond density or shrub area per km<sup>2</sup>'. For application purposes, it is preferable to focus on the models with pond density (Table 2.2, model 5) rather than on the models with occupied pond density (model 4). New ponds can be created easily, while it is not possible to create 'occupied ponds', unless one considers the introduction of the target species. Of course it is essential that the target species already be present in the planning area, and that restoration plans are designed around these potential dispersal sources (Vos 1993). For the same reason, models including both terrestrial and aquatic habitat isolation variables (Table 2.2, model 5b and 5c) have better potential for application because in these models both essential habitat parts can be quantified. As discussed above, the variable 'presence of occupied ponds' is a combination of both suitable terrestrial and aquatic habitat in the pond's surroundings, in which the contribution of the two components cannot be separated.

### **Towards further quantification of isolation parameters**

The concentric measure of pond density in a zone of 750 m is a more powerful explanatory variable than the linear variable 'distance from the nearest pond'. A limitation of the concentric isolation measure is that all ponds in the circle contribute equally to the connectivity of a pond, regardless of their actual distance. It is, however, a simplification to assume that potential dispersal sources at a distance of 750 m have the same impact as sources near the target pond. In addition, one would like to take into account the actual size of dispersal sources. A measure that takes these spatial characteristics into account is the isolation or connectivity measure used by Verboom et al. (1991a) and Hanski (1994). In this measure, all (occupied) habitat patches are treated as potential dispersal sources, corrected for habitat size (as a measure of population size) and distance from the target patch. However, for ground-dwelling species not only the distance but also the character of the intermediate landscape influences the isolation of a pond, as was indicated by the contribution to the model of the amount of suitable terrestrial habitat components in a circle of 1000 m surrounding a pond. For several species groups it has been demonstrated that corridors do have a positive effect on the dispersal of species (Bennett et al. 1994). In a 'complete' isolation measure, not only the distance but also the relative resistance of the landscape between ponds should contribute to the connectivity of a habitat patch.

### 3 Effects of habitat fragmentation and road density on the distribution pattern of the moor frog *Rana arvalis*

#### Summary

1. The effects of habitat fragmentation on the distribution pattern of the moor frog *Rana arvalis* were investigated. Also the possible isolation effects of the road network were taken into account.

2. Indications were found that habitat fragmentation partly explains the distribution pattern of the moor frog. The statistical models showed a positive effect of pond size (or marsh area) and a negative effect of road density on the probability of occupation of a moorland pond.

3. Because of the strong correlation between habitat quality variables and isolation variables, no unambiguous effects of isolation, described as the amount of suitable terrestrial habitat (moorland) in the surroundings of a moorland pond in a radius of 100 to 2000 m, could be demonstrated.

4. Spatial differences in road density can play a role in the selection of optimal locations for nature protection areas. The regression model used in this study predicts a reduced occupation probability in 55% of the study area. In the part of the study area adjacent to a motorway, occupation probability is lowered to less than 30%.

5. European studies of habitat fragmentation on amphibian species revealed a mean distance between occupied ponds of < 1 km in all studies. This could be a general rule of thumb for persistent amphibian populations. Effects of pond size on the probability of occupation were more variable.

6. When discussing the effects of habitat fragmentation on amphibians and other ground-dwelling species, the negative effects of roads are often underestimated.

#### Introduction

Recent discussions on a presumed worldwide decline of amphibian species led to speculation on its causes. Pollution from pesticides, acid precipitation, increased levels of ultraviolet radiation, climate change and habitat destruction have all been suggested as possible causes (Blaustein & Wake 1990; Blaustein et al. 1994; Pounds & Crump 1994). However, it is becoming clear that not all species are declining and that not one 'global' factor but various complexes of factors are responsible for the decline in different parts of the world. Habitat destruction leading to increased habitat fragmentation is probably one of the most important factors causing amphibian decline in industrialised regions (Blaustein et al. 1994). In densely populated countries, amphibians often depend on small habitat patches separated by

intensively used agricultural landscapes. Effects of habitat fragmentation, expressed by the more frequent absence of species in small or isolated habitat patches, have been demonstrated for several amphibian species including the pool frog *Rana lessonae* (Sjögren 1988, 1991), the common frog *Rana temporaria* (Loman 1988) and the tree frog *Hyla arborea* (Edenhamn 1996; Vos & Stumpel 1996). So far, the evidence that habitat fragmentation causes (part of) the amphibian decline is biased towards extremely fragmented landscapes. The remaining habitats are distributed as small remnants separated by agricultural fields or production forests (Sjögren 1988, 1991; Edenhamn 1996; Vos & Stumpel 1996). In addition, agricultural landscapes are often dynamic, with a high 'turnover' of (semi)natural elements. The changing distribution of habitat over time causes additional stress for surviving populations (Opdam et al. 1993). Remarkably, additional isolation effects of roads, which are known to be extreme barriers for ground-dwelling species (Bennett 1991), have not been taken into account in the aforementioned studies. As most amphibian species are ground-dwelling and slow moving, the crossing of roads is expected to be problematic.

Schematically several thresholds in habitat fragmentation with corresponding species responses can be distinguished. (See for an extensive discussion Hanski & Gilpin 1991; Verboom et al. 1993; Hastings & Harrison 1994; Reich & Grimm 1996.) A first fragmentation threshold occurs when a continuous landscape breaks down into separate habitat fragments. With frequent exchange between habitat patches, the species suffers no negative effects of habitat fragmentation, and in principle all suitable habitat patches are occupied.

In the second stage, caused by increasing fragmentation, habitat patches become smaller and more isolated. Chance extinctions of small local populations will become more frequent. If local extinctions are compensated for by recolonisation, the network of local populations will persist (Verboom et al. 1993). The species shows the spatial dynamics characteristic of metapopulations (Levins 1970; Opdam et al. 1993).

In the third stage of habitat fragmentation, populations have become completely isolated. As extinctions can no longer be compensated for by recolonisation, the species is threatened with extinction on a regional level. The results of the above-mentioned studies on amphibians (Loman 1988; Sjögren 1988, 1991; Edenhamn 1996; Vos & Stumpel 1996) suggest habitat fragmentation of the second or the third stage. For nature conservation policy, to be effective, it is relevant to determine if suitable patches are found empty and if the number of empty patches threatens the persistence of the metapopulation as a whole.

For ground-dwelling species isolation can be regarded as a combination of distance to other habitat patches (Verboom et al. 1991a; Hanski 1994) and the resistance of the landscape between patches (Forman & Godron 1986; Merriam 1991; Saunders & Hobbs 1991; Bennett et al. 1994). The road network will probably contribute considerably to the resistance between habitat patches (Bennett 1991). Amphibian species may encounter roads during seasonal migrations between reproduction sites, summer habitat and hibernation sites or during dispersal to new habitat patches. Mortality on roads can have a considerable impact on amphibian populations and eventually can lead to extinction (Vos & Chardon 1994). In several studies, a negative correlation was found between traffic density and the probability

of survival of amphibians crossing a road (Van Gelder 1973; Berthoud & Müller 1983; Heine 1987; Kuhn 1987; Münch 1989). A model by Heine (1987) predicts almost zero survival probability for individuals that cross roads with more than 26 cars per hour. Fahrig et al. (1995) found lower amphibian densities in areas with high traffic intensity, which might suggest both increased mortality as well as increased isolation.

In order to formulate effective nature conservation policy, quantitative data on the required reduction in habitat fragmentation are needed. The stage at which habitat fragmentation becomes a problem for amphibian survival needs to be determined. For this purpose, effects of habitat fragmentation on the distribution pattern of the moor frog *Rana arvalis* Nilsson were studied. The moor frog habitat is less fragmented, with relatively large and stable habitat patches, in comparison to the above-mentioned studies. In addition, in this study the road network as an extra isolation factor was considered.

## Study area and methods

The study area covers 40 000 hectares in the south-west of the province of Drenthe, The Netherlands (Fig. 3.1). The boundaries consist of four canals with upright sheet piling, which are considered to be severe but not absolute barriers for amphibians. The moor frog is found in moist, nutrient-poor habitats, often on peat or sandy soils with impermeable layers or high groundwater tables (Hartung 1991; Podloucky 1987; Wijnands 1987). The aquatic part of the moor frog habitat consists of moorland ponds and swamps and covers 0.5% of the area. Moorlands, dominated by *Molinia caerulea* and *Erica tetralix*, form the main terrestrial part of the habitat and cover 6% of the area. Also hedgerows and mixed woods with well developed herb-layer play a role as terrestrial habitats. Although mixed woods and hedgerows cover 5 % of the area, the proportion of suitable wood parts is only small. Therefore it was not taken into account in further analysis. The habitats in the study area are remnants of a (semi)natural ecosystem that covered the whole study area until the beginning of this century (Historische Atlas Drenthe 1989). In the 1930s large parts were cultivated and the moorlands became fragmented. The agricultural land consists of arable fields and grasslands. The intensive agricultural exploitation, characterised by eutrophication and lowered groundwater tables, makes the habitat unsuitable for the moor frog.

In 1993, an inventory was made of the presence of moor frogs in a selection of 109 potentially suitable reproduction sites. Ponds or marshes in moorland and remnants of moorland in the agricultural area, with some adjacent terrestrial habitat, were considered suitable reproduction sites. Each site was visited at least twice in early spring during the reproduction period. The occurrence of moor frogs was established by counting calling males (spontaneously or in reaction to tape-recorded mating calls) and by searching for egg clumps, tadpoles, juveniles and adults in the ponds and immediate pond surroundings. Suitable calling conditions were tested using a reference pond. Egg clumps that were too old to distinguish them with certainty from those of the common frog (Büchs 1987; Rahmel & Eikhorst 1988) were raised in captivity and identified in their tadpole stage. Numbers were recorded in

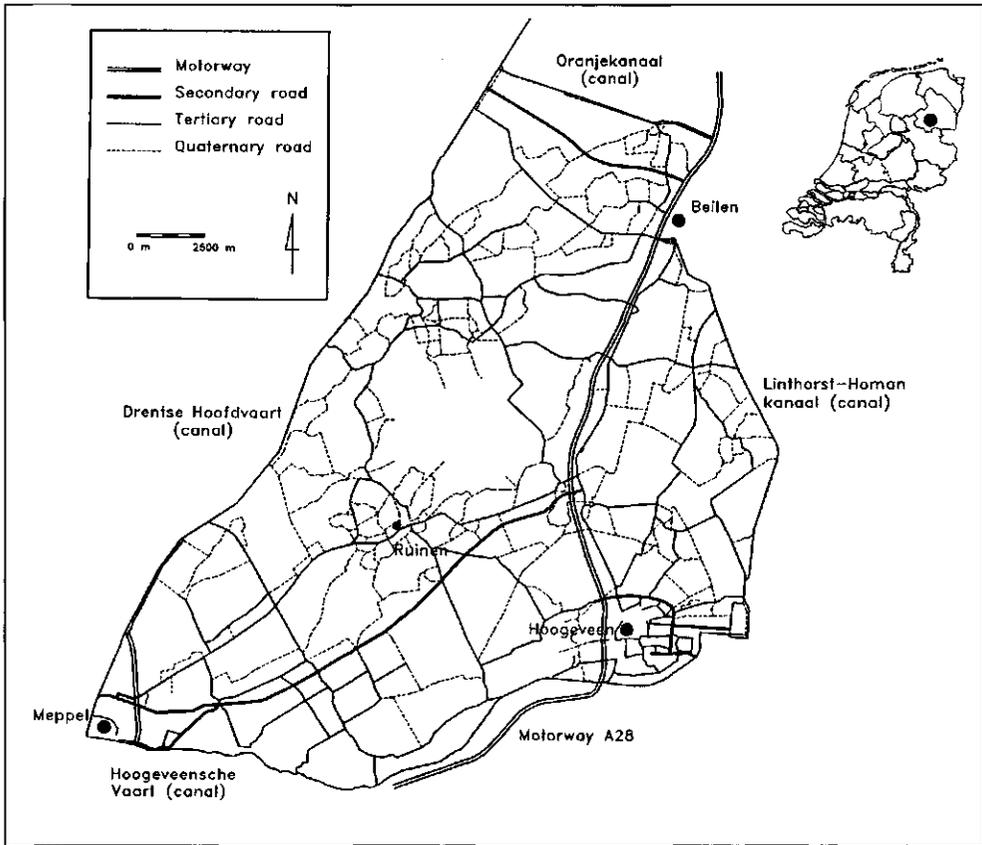


Figure 3.1

The study area in the south-west of the province of Drenthe, The Netherlands. The distribution pattern of the road network is indicated.

classes. A relative measure of the population size was derived from the number of calling males and egg clumps. In 'very small populations' no more than 2-5 calling males or 2-5 egg clumps were recorded; in 'small populations' 6-25 calling males or 6-25 egg clumps; in 'moderate populations', 26-50 calling males or 26-100 egg clumps; and in 'large populations' more than 100 egg clumps were recorded. Estimating the number of calling males in choruses larger than 50 animals was not possible, but it was possible to estimate the number of egg clumps. Since females lay only one egg clump, this is a widely used method for estimating the size of a population (Schlöpman 1988; Griffiths & Raper 1994), assuming equal sex ratio. The moor frog is an explosive breeder, so calling and oviposition take place in a short time. Therefore, two visits during this period were considered sufficient to estimate population size. Note that this method does not reveal the absolute population size but it can be used as a relative measure. Pond size, shore area, area of marsh vegetation in the pond, water acidity, and water conductivity were measured in the field, early in the reproduction period (Table 3.1). Dominant species in the marsh vegetation were *Molinia caerulea*, *Juncus effusus* and *Eriophorum*

Table 3.1

Overview of the habitat quality, pond area and isolation variables; the mean value and the standard deviation (sd) are listed for ponds that were occupied and not occupied by moor frogs in 1993.

| VARIABLES   |  | MEAN ± SD<br>OCCUPIED<br>PONDS | MEAN ± SD<br>UNOCCUPIED<br>PONDS |
|---|--|--------------------------------|----------------------------------|
| <b>Habitat Quality Variables</b>                          |  |                                |                                  |
| pH  | Water Acidity                            | 5.2 ± 0.8                      | 6.6 ± 0.8                        |
| CONDUCTIVITY  | Water Conductivity                       | 126.5 ± 55.0                   | 284.9 ± 174.7                    |
| <b>Area Variables Aquatic Habitat</b>                     |  |                                |                                  |
| Area  | Pond area (ha)*                          | 0.61 ± 1.36                    | 0.21 ± 0.22                      |
| Shore   | Pond area with water depth < 1m (ha)*    | 0.30 ± 0.68                    | 0.95 ± 0.18                      |
| Marsh   | Pond area with marsh vegetation (ha)*    | 0.15 ± 0.25                    | 0.03 ± 0.04                      |
| <b>Linear Isolation Variables Aquatic Habitat</b>         |  |                                |                                  |
| Pond distance   | Distance to nearest pond (km)            | 0.50 ± 0.48                    | 0.46 ± 0.26                      |
| Occupied pond distance                                    | Distance to nearest occupied pond        | 0.63 ± 0.62                    | 1.18 ± 1.27                      |
| <b>Concentric Isolation Variables Aquatic Habitat</b>     |  |                                |                                  |
| Occupied ponds 250 m                                      | Area (ha) of occupied ponds within 250 m | 0.07 ± 0.15                    | 0.01 ± 0.02                      |
| Occupied ponds 500 m                                      | " 500 m                                  | 0.39 ± 0.70                    | 0.22 ± 0.57                      |
| Occupied ponds 750 m                                      | " 750 m                                  | 0.85 ± 1.19                    | 0.65 ± 1.11                      |
| Occupied ponds 1000 m                                     | " 1000 m                                 | 1.31 ± 1.54                    | 1.21 ± 1.81                      |
| Occupied ponds 1500 m                                     | " 1500 m                                 | 2.11 ± 2.34                    | 2.41 ± 3.19                      |
| Occupied ponds 2000 m                                     | " 2000 m                                 | 3.31 ± 3.18                    | 3.51 ± 3.69                      |
| Ponds 250 m   | Area (ha) of all ponds within 250 m      | 0.07 ± 0.15                    | 0.03 ± 0.69                      |
| Ponds 500 m   | " 500 m                                  | 0.45 ± 0.71                    | 0.32 ± 0.60                      |
| Ponds 750 m   | " 750 m                                  | 0.98 ± 1.27                    | 0.80 ± 1.12                      |
| Ponds 1000 m  | " 1000 m                                 | 1.56 ± 1.64                    | 1.40 ± 1.79                      |
| Ponds 1500 m  | " 1500 m                                 | 2.53 ± 2.46                    | 2.81 ± 3.28                      |
| Ponds 2000 m  | " 2000 m                                 | 4.09 ± 3.35                    | 4.11 ± 3.81                      |
| <b>Concentric Isolation Variables Terrestrial Habitat</b> |  |                                |                                  |
| Moorland 250 m  | Area (ha) of moorland within 250 m       | 6.71 ± 9.03                    | 1.11 ± 1.90                      |
| Moorland 500 m  | " 500 m                                  | 15.39 ± 19.05                  | 4.28 ± 6.69                      |
| Moorland 750 m  | " 750 m                                  | 25.08 ± 28.00                  | 8.49 ± 14.02                     |
| Moorland 1000 m   | " 1000 m                                 | 35.15 ± 35.50                  | 13.94 ± 21.41                    |
| Moorland 1500 m   | " 1500 m                                 | 55.45 ± 43.82                  | 27.83 ± 31.56                    |
| Moorland 2000 m   | " 2000 m                                 | 77.63 ± 51.19                  | 49.41 ± 43.49                    |
| <b>Concentric Isolation Variables Road Density</b>        |  |                                |                                  |
| Weighted roadlength 250 m                                 | Weighted roadlength within 250 m         | 628 ± 951                      | 1472 ± 2415                      |
| Weighted roadlength 500 m                                 | " 500 m                                  | 2633 ± 3100                    | 4461 ± 6161                      |
| Weighted roadlength 750 m                                 | " 750 m                                  | 5526 ± 5734                    | 9089 ± 10226                     |
| Weighted roadlength 1000 m                                | " 1000 m                                 | 9948 ± 8799                    | 14360 ± 13497                    |
| Weighted roadlength 1500 m                                | " 1500 m                                 | 25465 ± 17640                  | 30668 ± 21313                    |
| Weighted roadlength 2000 m                                | " 2000 m                                 | 50457 ± 29533                  | 52934 ± 31396                    |

\* Because of skewed distribution log-transformation was used in further analysis.

*angustifolium*. Water acidity and water conductivity are important indicators for nutrient-poor conditions (Büchs 1987). Quality variables for the terrestrial habitat were not taken into account. The spatial configuration of water and land habitat and road types was recorded from a topographical map 1 : 25 000 using a Geographic Information System (Fig. 3.2).

Each moorland pond was treated as a separate potential site for a population. However, ponds in the same moorland, within 200 m from each other, were joined and treated as one site in further analysis. The following measures for the isolation of the moorland ponds were estimated. Linear variables: distance from the nearest pond and distance from the nearest pond occupied by moor frogs. Concentric

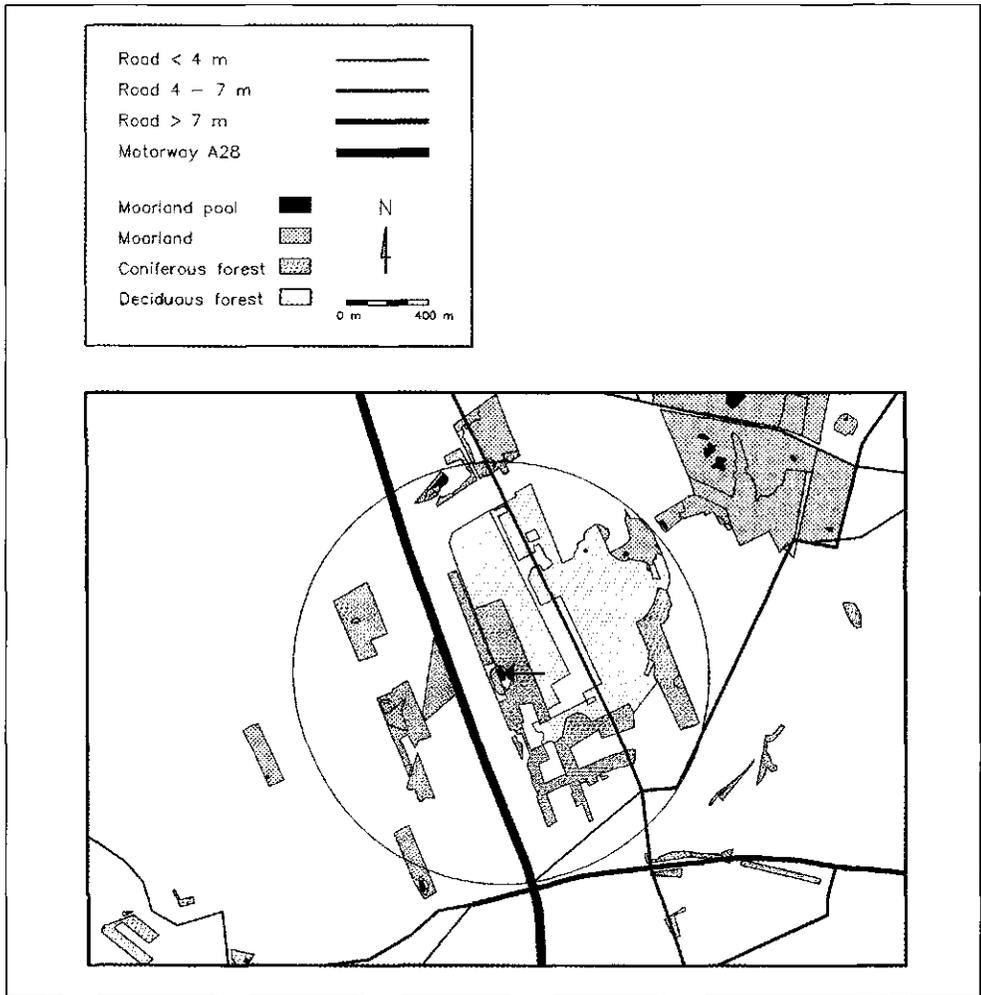


Figure 3.2

Example of the digitised landscape types and of a circle with a radius of 1000 m surrounding a moorland pond. In this circle, the concentric connectivity measures 'total area of aquatic and terrestrial habitat' and 'total road length' were calculated. The arrow indicates the position of the moorland pond.

variables were calculated in circles surrounding each pond with increasing radius of 250, 500, 750, 1000, 1500, and 2000 m (Fig. 3.2; see also Vos & Stumpel 1996). Within these zones, the following variables were measured: total area of ponds (the sum of both occupied and unoccupied ponds), the area of occupied ponds, the area of moorland and the road length per road type (Table 3.1).

Four road types were distinguished: four lane motorways, secondary roads (wider than 7 m), tertiary roads (between 7 and 4 m width) and quaternary roads (paved roads smaller than 4 m; Fig. 3.1). To aggregate effects of all road types in the surroundings of a pond, road lengths were added after a correction for traffic intensity. Based on the mean traffic volume per road type (Provincie Drenthe 1994; Provincie Overijssel 1993), the road length of motorways, secondary and tertiary roads was multiplied by a factor 20, 4 or 2 respectively as compared to quaternary roads.

Effects of habitat quality and fragmentation on the presence of moor frogs in a pond were analysed with logistic regression analysis (Jongman et al. 1995) using the statistical programme Genstat (Genstat 5 Committee 1993). Whether habitat fragmentation factors contribute significantly to the regression model in addition to the habitat quality factors can be tested using a two-step regression method (Van Apeldoorn et al. 1992; Fahrig et al. 1995; Vos & Stumpel 1996). In the first step, differences in habitat quality between ponds were accounted for by entering only habitat quality variables to the model in a stepwise fashion (pH and conductivity, Table 3.1). In the second step, the model was extended with fragmentation variables (area and isolation variables, Table 3.1). For each concentric isolation variable, the optimal radius was determined by the variable with the highest explanatory value in the regression model, as judged on the basis of the deviance statistic (Jongman et al. 1995). To reduce the number of variables describing road density, only the weighted variable for all road types was tested (Table 3.1).

To give an indication of the effects of habitat fragmentation on abundance, the relative measure for population size was analysed by regression. In this linear regression the population size classes were coded with integer values: 0 (unoccupied ponds), 1 (very small populations), 2 (small populations), 3 (moderate populations), and 4 (large populations). The more advanced method of ordinal regression was not applied since the potential gain of this method is small if the number of classes exceeds three (Ter Braak pers. com.). The independent variables and stepwise analysis were the same as in the first analysis.

## Results

Moor frogs were found in 73 of the 109 ponds. The distribution pattern and density classes are given in Figure 3.3. In the first step of the logistic regression analysis, the habitat quality variables were entered to the model. Both water acidity (pH) and water conductivity were important explanatory variables for the presence of moor frogs. The occupation probability increased with lower pH and water conductivity values (model 1, pH  $p < 0.001$ , conductivity  $p < 0.01$ , Table 3.2).

After accounting for habitat quality variation, in the next step of the analysis the variables related to habitat fragmentation were added to the model (area and

Table 3.2

Selection of the multiple regression models obtained with the two-step approach. Significance levels of the last variable added to each model and  $R^2$  adjusted of the total model are listed. The logistic models 1 and 2 are tested with the deviance test. The linear models 3 and 4 are tested with the F-test.

| VARIABLES OF MULTIPLE REGRESSION MODEL                       | SIGN AND SIGNIFICANCE OF THE LAST VARIABLE | $R^2$ ADJUSTED OF THE TOTAL MODEL |
|--|--|-----------------------------------|
| MODEL 1 BASED ON PRESENCE-ABSENCE DATA                       |  |                                   |
| pH   | - ***                                      | 36%                               |
| pH + Conductivity  | - **                                       | 43%                               |
| pH + Conductivity + Marsh                                    | + ***                                      | 58%                               |
| pH + Conductivity + Marsh + Weighed Roads 750 m              | - **                                       | 64%                               |
| MODEL 2 BASED ON PRESENCE-ABSENCE DATA                       |  |                                   |
| Moorland 250 m   | + ***                                      | 12%                               |
| Moorland 250 m + Conductivity                                | - ***                                      | 36%                               |
| Moorland 250 m + Conductivity + Marsh                        | + ***                                      | 52%                               |
| Moorland 250 m + Conductivity + Marsh + Weighed Roads 750 m  | - *  | 55%                               |
| MODEL 3 BASED ON DENSITY CLASSES                             |  |                                   |
| pH   | - ***                                      | 43%                               |
| pH + Conductivity  | - *  | 46%                               |
| pH + Conductivity + Marsh                                    | + ***                                      | 53%                               |
| pH + Conductivity + Marsh + Tertiary Roads 250 m             | - *  | 55%                               |
| MODEL 4 BASED ON DENSITY CLASSES                             |  |                                   |
| Moorland 250 m   | + ***                                      | 18%                               |
| Moorland 250 m + Conductivity                                | - ***                                      | 32%                               |
| Moorland 250 m + Conductivity + Marsh                        | + ***                                      | 42%                               |
| Moorland 250 m + Conductivity + Marsh + Tertiary Roads 250 m | ns   |                                   |

Significance level: \*  $p < 0.05$ , \*\*  $p < 0.01$ , \*\*\*  $p < 0.001$ , ns not significant

isolation variables, Table 3.1). All variables describing pond size (pond area, shore area and area of marsh vegetation of the pond) contributed significantly to the model. The variable 'area of marsh vegetation of the pond' had the highest explanatory value (model 1, marsh  $p < 0.001$ , Table 3.2). The larger the area of marsh vegetation, the higher the occupation probability.

None of the isolation variables describing the amount of aquatic or terrestrial habitat in the surroundings of the pond added significantly to the explanatory value of the models, yet significant effects were shown in single variable models (Table 3.3). Their failure in the multiple model could be explained by the correlation between these isolation measures and the habitat quality variables. The correlations with water acidity were especially high (ranging from  $r = -0.207$   $p < 0.05$  to  $r = -0.578$   $p < 0.001$ ). Without the acidity variable in the model, 'area of moorland within 250 m' was the best explanatory variable (model 2, moorland-250 m  $p < 0.001$ , Table 3.2). All other ranges up to 2000 m were also significant (Table 3.3). Model 2 explained frog occupancy less than model 1 ( $R^2$  adjusted of 55% and 64%, respectively, Table 3.2).

Table 3.3

Significance levels of the concentric isolation measures 'moorland area' and 'area of occupied ponds' are added to the model with increasing ranges from 250 to 2000 m. For the logistic model deviances and for the linear model F-values are listed.

| CONCENTRIC ISOLATION VARIABLES | LOGISTIC MODEL BASED ON PRESENCE-ABSENCE DATA | LINEAR MODEL BASED ON POPULATION DENSITY CLASSES |
|--------------------------------|---|--|
| Moorland 250 m                 | 17.1 ***                                      | 33.2 ***   |
| Moorland 500 m                 | 12.2 ***                                      | 27.4 ***   |
| Moorland 750 m                 | 12.0 ***                                      | 26.9 ***   |
| Moorland 1000 m                | 11.2 **                                       | 24.8 ***   |
| Moorland 1500 m                | 11.3 **                                       | 22.1 ***   |
| Moorland 2000 m                | 12.4 ***                                      | 19.0 ***   |
| Occupied ponds 250 m           | 7.9 **  | 9.2 **   |
| Occupied ponds 500 m           | 6.2 *   | 3.2 ns   |
| Occupied ponds 750 m           | 0.9 ns  | 11.5 ***   |
| Occupied ponds 1000 m          | 2.5 ns  | 6.3 *  |
| Occupied ponds 1500 m          | 3.0 ns  | 8.6 **   |
| Occupied ponds 2000 m          | 5.3 *   | 10.0 **  |

Significance level: \*  $p < 0.05$ , \*\*  $p < 0.01$ , \*\*\*  $p < 0.001$ , ns not significant

Due to high correlations between the terrestrial and aquatic isolation variables (from  $r = 0.262$   $p < 0.01$  to  $r = 0.832$   $p < 0.001$ ) the aquatic isolation variable 'area of occupied water' added no further explanation to the model.

Finally, the isolation measure describing the road density weighted for traffic intensity within 750 m of the pond added significantly to the model (model 1, weighted roads 750 m  $p < 0.05$ , Table 3.2). Road density had a negative effect on the occupation probability of a pond. Substituting water acidity by moorland area within 250 m did not alter the contribution of road density to the model (model 2, weighted roads 750 m  $p < 0.05$ , Table 3.2).

The results of the linear regression analysis with the population size classes as the dependent variable were to a great extent in accordance with analysis of the presence - absence data. The population size in a pond increased with low pH and low water conductivity values (model 3, pH  $p < 0.001$ , conductivity  $p < 0.05$ , Table 3.2). Also an increase in the area of marsh vegetation in a pond had a positive effect on the population size (model 3, marsh  $p < 0.001$ , Table 3.2).

Similar to the analysis of the presence - absence data, the isolation measure 'moorland area within 250 m' contributed significantly only to a model without the acidity variable (model 4, moorland 250 m  $p < 0.001$ , Table 3.2). Again the isolation variable added significantly for all ranges up to 2000 m; the range 0-250 m was the most significant (Table 3.3).

Also the aquatic isolation variable 'area of occupied water' did not contribute to the model already including the terrestrial isolation variable 'area of moorland'.

The effect of road density on the population size, however, differed from the presence - absence model results. None of the weighted road measures, ranging from 250 to 2000 m, added significantly to the explanatory value of the models. However,

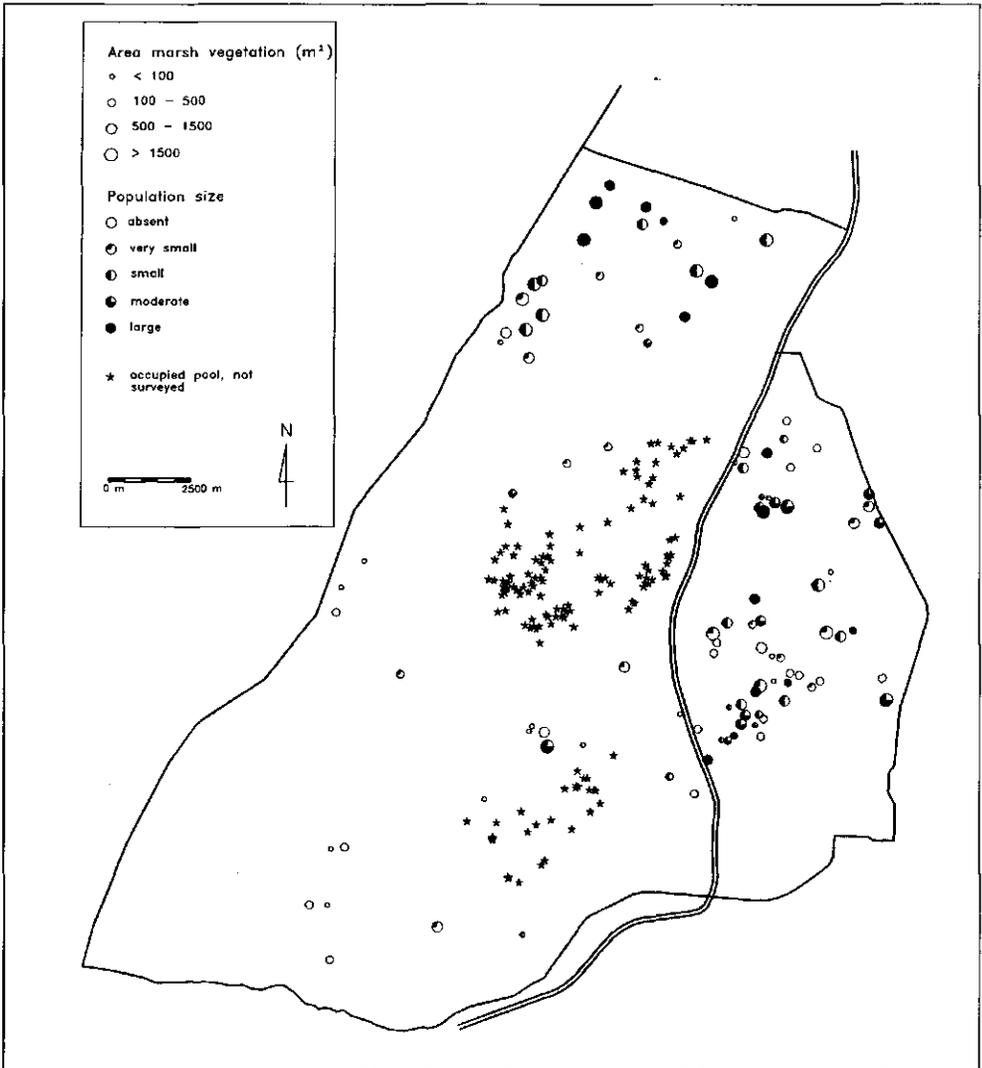


Figure 3.3

Distribution pattern of the moor frog in 1993 in south-west Drenthe. The area of marsh vegetation in a moorland pond and the estimated population size are indicated.

for exploratory purposes the variables for the separate road types were also tested. This revealed a significant negative effect of roads on the population size for tertiary roads in a circle of 250 m surrounding the pond (model 3, tertiary roads 250 m  $p < 0.05$ , Table 3.2). After substituting the acidity variable by the isolation measure 'area moorland within 250 m' no effect of road density on population size could be determined (model 4, tertiary roads 250 m ns, Table 3.2).

An illustration of the predicted effects of road density on the occupation probability based on model 1 (Table 3.2) is given in Figure 3.4. The study area was

divided in grids of 50 by 50 m. For each grid, the weighted road density was calculated by determining the total weighted road length in a radius of 750 m surrounding each grid cell. According to the model, 45% of the study area is free of negative road effects. In 30% of the study area, the occupation probability of ponds is between 70 and 85%. The third zone encompasses 15% of the area and in this part the occupation chance is lowered to 30 to 70%. Finally in 10% of the area, situated along the motorway, ponds have an occupation probability of less than 30%.

### A comparison of species

To gain an impression of whether trends can be detected between the degree of habitat fragmentation and the effects on amphibian distribution, the results of several studies were investigated. The degree of habitat fragmentation in a landscape can be described by a combination of mean patch size and mean interpatch distance (Gustafson & Parker 1992; Schumaker 1996). In Figure 3.5 these fragmentation measures are plotted for occupied and empty habitat patches (ponds) derived from studies on several amphibian species (Sjögren 1988, 1991; Edenhamn 1996; Vos & Stumpel 1996; this study). In Table 3.4, information essential for comparison of the studies is summarised. As species characteristics such as dispersal capacity and site tenacity also determine effects of habitat fragmentation in a given landscape (Sinsch 1990), a comparison can only be rough. As is illustrated in Figure 3.5 and Table 3.4, in all studies a negative effect of isolation on the distribution pattern of the different species was found. The positive effect of pond size on occupation probability is less clear and was found for the moor frog and the tree frog in Sweden. For the pool frog, with the relatively large ponds a negative effect of pond size was found. This could probably be explained by the occurrence of fish predation in the relatively large ponds (Sjögren 1991). Generally the size of the reproduction site is thought to be the

Table 3.4

Summary of some studies on the effects of habitat fragmentation. Explanation of abbreviations: sign = significant in regression, ns = not significant, (-) or (+) = positive or negative effect in regression.

| SPECIES                     | DISTANCE NEAREST OCCUPIED POND | POND AREA POND (%) | OCCUPIED VARIABLE | DEPENDENT                       | REFERENCE          |
|-----------------------------|--------------------------------|--------------------|-------------------|---------------------------------|--------------------|
| Pool frog                   | sign (-)                       | sign (-)           | 52% **            | Persistence                     | Sjögren 1988, 1991 |
| Tree frog (Sweden)          | sign (-)*                      | sign (+)           | 30%               | Persistence/<br>Population size | Edenhamn 1996      |
| Tree frog (The Netherlands) | sign (-)                       | ns                 | 10%               | Occupation                      | Vos & Stumpel 1996 |
| Moor frog                   | sign (-)*                      | sign (+)           | 67%               | Occupation /<br>Population size | this study         |

\* only significant in single regression model  
 \*\* based on permanent waters only

limiting factor determining population size in amphibians (Wilbur 1987; John-Alder & Morin 1990). However, in extremely fragmented situations as is the case for the tree frog in the Dutch landscape, there are indications that the distribution of suitable terrestrial habitat may be the limiting factor, due to high mortality in the terrestrial phase (Loman 1988; Vos & Stumpel 1996). Also, in most cases a positive effect of the distribution of the terrestrial habitat could be demonstrated (not investigated for pool frog, Sjögren 1991). The variables, however, differed from case to case so the degree of isolation could not be compared.

## Discussion

The statistical models showed a positive effect of pond size and a negative effect of road density on the probability of occupation of a moorland pond by moor frogs. We interpret this as an indication that habitat fragmentation plays a role in explaining the distribution pattern of the moor frog. No unambiguous effects of isolation, described as the amount of suitable habitat in the surroundings of a pond, could be demonstrated. The results of the analysis with presence - absence data as the dependent variable and the analysis of population size classes were in good accordance, except for the effect of road density. The population size models have the advantage that more precise predictions of effects on abundance are possible.

The habitat quality variables explain the probability of moor frog presence better than the area and isolation variables. Typical moor frog habitat consists of moorlands of nutrient-poor sandy areas and peat moorlands with high water tables or impermeable ground layers (Wijnands 1987; Podloucky 1987; Nettmann 1987; Hübner & Sennert 1987; Hartung 1991). A low water acidity and a low water conductivity characterise such a habitat. Although the moor frog has a relatively high acid tolerance, a pH that is too low may be detrimental in areas suffering from acidification (Andrén *et al.* 1988; Clausnitzer 1987).

### Pond size

The positive effect of pond size on the probability of occupation and the abundance of moor frogs is in accordance with effects that would be expected in fragmented landscapes. Compared to small habitat patches, large patches will generally contain larger populations with lower extinction probabilities and will therefore be occupied more often (Levins 1970; Goodman 1987; Verboom *et al.* 1993). The predictive effect of marsh area is strongest, presumably because this is the most important part of the water habitat. Shallow water with water vegetation is used as oviposition sites (Nöllert 1987). The marsh vegetation will probably also function as terrestrial habitat, when summer water levels are low. Loman (1988) found a positive effect on probability of occupation by the common frog for a comparable variable, namely the length of shallow unshadowed water bank (i.e. the part where marsh vegetation will develop optimally).

## Pond isolation and pond quality

Habitat isolation variables and habitat quality variables were highly correlated and therefore it was not possible to separate between these effects. This can be explained by the spatial configuration of the landscape in the study area. The habitat consists of moorlands of variable size in which several moorland ponds are often present. Ponds in moorland form optimal (nutrient-poor) conditions for moor frogs. This is illustrated by the two significant habitat quality variables: a high water acidity and low water conductivity. However, ponds in moorlands also have low isolation which is described by the two variables: high frequency of (occupied) ponds and a large proportion of moorland in the surroundings. On the other hand, the ponds situated in the agricultural landscape are more isolated but also have a lower habitat quality, because of more direct agricultural disturbances. To distinguish clearly between these effects the study should be repeated in a landscape with a different spatial configuration of habitat or with different land use between habitat patches.

In the models without water acidity, the amount of moorland had a positive effect on occupation probability up to 2000 m. This indicates that the variable moorland is not merely a substitute for pond quality variables. The moorland in the first few hundred metres surrounding a pond will function as the terrestrial habitat part of the local population (Hartung 1991). But the proportion of moorland over larger distances can be regarded as a connectivity variable, which will increase the colonisation probability of a pond. It is plausible that dispersal will be more successful in a landscape with a large proportion of suitable terrestrial habitat (Vos & Stumpel 1996; Merriam 1991). Hartung (1991) found that habitat preferences influence the movements of moor frogs where unsuitable habitat types, such as open fields, were avoided by adults and especially by newly metamorphosed juveniles. 'Area of moorland' was a better explanatory factor than 'area of occupied ponds'. This indicates not only the importance of distance to sources of dispersing animals (occupied ponds) but also of the connectivity of the surrounding landscape (area of moorland).

## Roads and isolation

The road density adjusted for traffic intensity in the surroundings of a moorland pond had a negative effect on the occupation probability. This was interpreted as an indication that roads increase the isolation between ponds and therefore contribute to habitat fragmentation. The exchange of individuals in the landscape matrix between habitat patches will decrease due to the extra mortality on roads, which will lower colonisation rates. Furthermore, more mortality of individuals will increase the extinction risk of local populations. Also the results of Fahrig *et al.* (1995) indicate a negative effect of roads, with a higher proportion of dead frogs and toads on roads with high traffic intensity and lower abundances of amphibians near intensively used roads. When comparing the highest traffic intensity class of Fahrig *et al.* (1995) of 8500 - 13 000 cars per 24 hours with car densities in the study area (secondary roads 6 713 and motorways 30 821 cars per 24 hours; Provincie Drenthe 1994; Provincie Overijssel 1993), we expect the negative effects of roads on amphibian distribution to be even higher.

There was an indication that roads in the direct surroundings of a reproduction site (250 m) affect the population size negatively. This is in accordance with the expected extra mortality on roads. However, the effect was only weak and probably the population size classes were too coarse to measure such an effect.

### Implications for nature conservation

The negative effects of road density on the probability of occupation of a pond by moor frogs is important information for nature conservation policy. In the study area the occupation probability of ponds in areas with high road density (adjusted for traffic intensity) was strongly reduced (Fig. 3.4). The regression model used in this study predicts a reduced occupation probability in 55% of the study area. In the part of the study area adjacent to a motorway, occupation probability is lowered to less than 30%. Spatial differences in road density should play a role in the selection of optimal locations for nature protection areas.

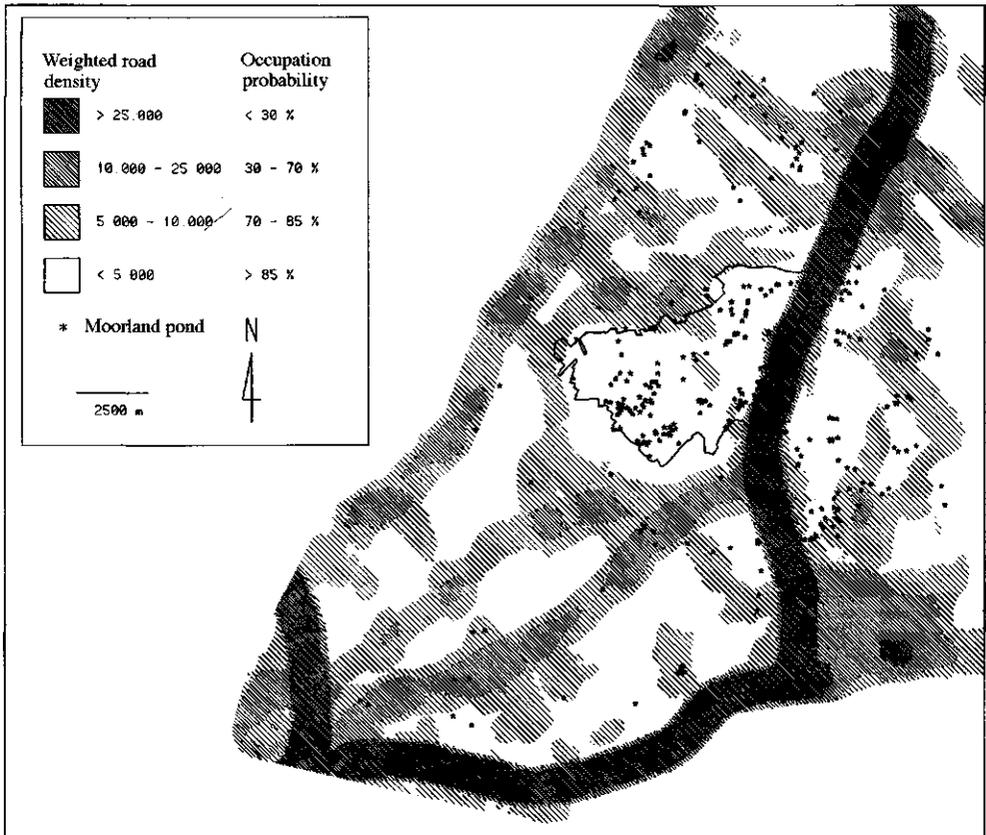


Figure 3.4

Zones of weighted road density in the study area. The weighted road density is calculated for each grid cell of 50 by 50 m. For each zone, the occupation probability of the moorland ponds is indicated based on the regression model.

### Thresholds in habitat fragmentation?

The comparison of several studies of effects on habitat fragmentation on amphibian distribution revealed negative effects of habitat fragmentation for all species in all landscapes studied. As the mean distance between occupied patches was below 1 km for all species in study (Fig. 3.5) this could provide a general rule of thumb for amphibian population networks. Habitat patches with low isolation were found occupied more often, which indicates that probably the degree of fragmentation is still linked with a metapopulation phase in which habitat patches are connected by dispersing animals. However, the low degree of occupancy of habitat patches on a regional level, especially for the tree frog (Sweden 28%, Edenhamn 1996; The Netherlands 10%, Vos & Stumpel 1996), indicates that at least part of the habitat network in the studied landscapes is already disintegrated. Simulation models are needed in order to predict whether the distribution patterns will persist in future (Verboom *et al.* 1993; Hanski 1994).

Based on this study on the moor frog, it can be concluded that even in less fragmented habitat, negative effects of habitat fragmentation could be detected. The stage of fragmentation at which amphibians do not show negative effects is becoming increasingly rare in our cultural landscapes. Furthermore, the additional isolation effect of roads for amphibians and other ground-dwelling species is often underestimated. As road density and traffic intensity will increase with urbanisation, roads will become an important factor in habitat fragmentation.

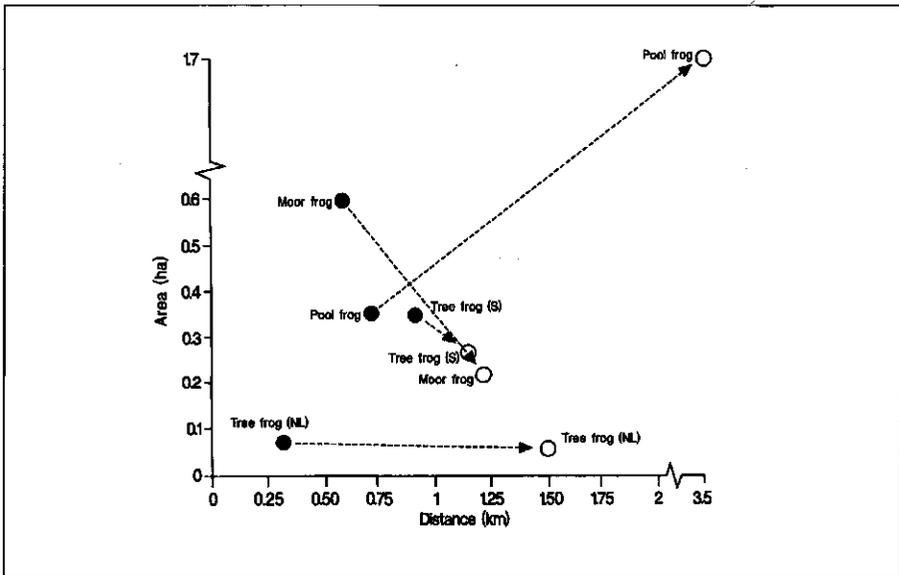


Figure 3.5

Mean pond size and mean distance to the nearest occupied pond for occupied and empty ponds (filled and open circles) for several amphibian species: pool frog (Sjögren 1988, 1991); tree frog in Sweden (S) (Edenhamn 1996); tree frog in The Netherlands (NL) (Vos & Stumpel 1996) and moor frog (this study).

## 4 Habitat fragmentation and genetic structure in the moor frog (*Rana arvalis*)

### Summary

The genetic structure of ten *Rana arvalis* populations was determined in a fragmented study area in the north of The Netherlands, using five microsatellite loci. The number of alleles per locus ranged from 2 to 8.  $F_{IS}$  was 0.049 across loci, and most populations were in HW equilibrium. Despite the fact that the populations were separated by agricultural land, roads and railways, the degree of population subdivision was relatively low ( $F_{ST} = 0.052$ ), possibly due to the limited number of generations passed since cultivation of the continuous moorland. Nevertheless, there was a significant positive correlation between genetic distance and geographical distance between populations. The resistance variable for the fraction of negative linear elements (roads and railroads) gave a higher explanatory value than geographical distance. The correlation between genetic distance and geographical distance indicates that dispersal rate between populations decreases with distance. In particular, the amount of barriers (roads and railroads) between populations seem to reduce exchange between populations.

### Introduction

Small populations in fragmented landscapes run the risk of going extinct. However, if local extinctions are compensated for by recolonisations, a species may survive on a regional level. Local extinction and recolonisation are characteristic processes in metapopulations (Verboom et al. 1993; Hanski & Gilpin 1991, 1997). Dispersal of individuals, which allow for the occurrence of recolonisation events, is vital for long-term survival of a species in subdivided habitat. The quantification of dispersal and the understanding of the relation between dispersal and landscape heterogeneity is a prerequisite for effective species conservation (Wiens 1997). Guidelines for the spatial distribution of habitat require knowledge about the distance over which regular exchange between local populations is still possible. Quantifying dispersal distances has high research priority, but these studies tend to be very time-consuming and methodological problems are manifold. Especially long distance dispersal is difficult to measure directly (Crochet 1996; see Turchin 1998, for a review on dispersal study methods).

The demographic processes that define metapopulation dynamics also have impact on genetic structure (McCauley 1991). An effect of limited dispersal in fragmented landscapes is the reduction of gene flow between local populations. Especially in small and isolated populations lack of gene flow among populations in combination with genetic drift may increase differentiation between populations. With decreasing population size inbreeding will occur. Loss of heterozygosity may reduce individual fitness, and the next phase, loss of genetic variation, will reduce the species' flexibility towards environmental changes (Frankham 1995, 1998; Booy 1998).

The genetic structure of populations is described by the spatial variation of allele frequencies at the polymorphic loci. There are many studies where a correlation between genetic dissimilarity and geographical distance of populations was demonstrated (see Avise 1994 for examples of different taxa; Crochet 1996 for a review on birds). The use of highly polymorphic mini- and microsatellite DNA markers may reveal differences between populations on a small spatial and temporal scale (Scribner *et al.* 1994; Waser & Strobeck 1998).

In this paper a pilot study is described on the moor frog (*Rana arvalis*) to determine whether or not genetic differentiation between local populations is associated with predicted connectivity of populations. As the moor frog is a ground-dwelling, slow moving species, connectivity between habitat patches can be regarded as a combination of distance and the resistance of the landscape between patches (Forman & Godron 1986; Merriam 1991; Saunders & Hobbs 1991; Bennett *et al.* 1994; Wiens 1997). A negative effect of road density on the distribution pattern of the moor frog was demonstrated in an earlier study (Vos & Chardon 1998). Influence of the landscape mosaic on the movement patterns of moor frogs was demonstrated by Hartung (1991): moor frogs preferred ditches and hedgerows and avoided dry and open areas. Therefore, the impact of distance corrected for the relative resistance of different habitat types and linear landscape elements will also be considered. To determine genetic similarity, microsatellite loci are employed. We generate markers for the moor frog, using microsatellite-containing sequences from other *Rana* species present in the EMBL and Genbank databases. Since the sequences flanking the repeats tend to be conserved, PCR-based detection of repeat length variation produces microsatellite markers in related species (Smulders *et al.* 1997). Five microsatellite primer pairs were chosen based on amplification results and used to determine genetic variation among frogs and frog populations across a fragmented habitat.

## Material and methods

### Study area

The pilot study was carried out in the eastern part of the area where in 1993 an inventory was made of the moor frog in all potential reproduction sites in the south-west of the province of Drenthe, The Netherlands (Fig. 4.1; Vos & Chardon 1998). The moor frog is found in moist, nutrient-poor habitats, often on peat or sandy soils with impermeable layers or high groundwater tables (Podloucky 1987; Wijnands 1987; Hartung 1991). The aquatic part of the moor frog habitat consists of moorland ponds and swamps. The terrestrial habitat part consists mainly of moorlands dominated by *Molinia caerulea* and *Erica tetralix*. Also hedgerows and mixed woods with a well-developed herb layer play a role as terrestrial habitat. The habitats in the study area are remnants of a (semi)natural ecosystem until the beginning of this century (Historische Atlas Drenthe 1989). In the 1930s, in a short time, large parts of Drenthe were cultivated and the moorland became fragmented. The present fragmentation of moorland therefore exists for approximately 60 years. As moor frogs become adult in their third year (Hartung 1991), this would imply that 30

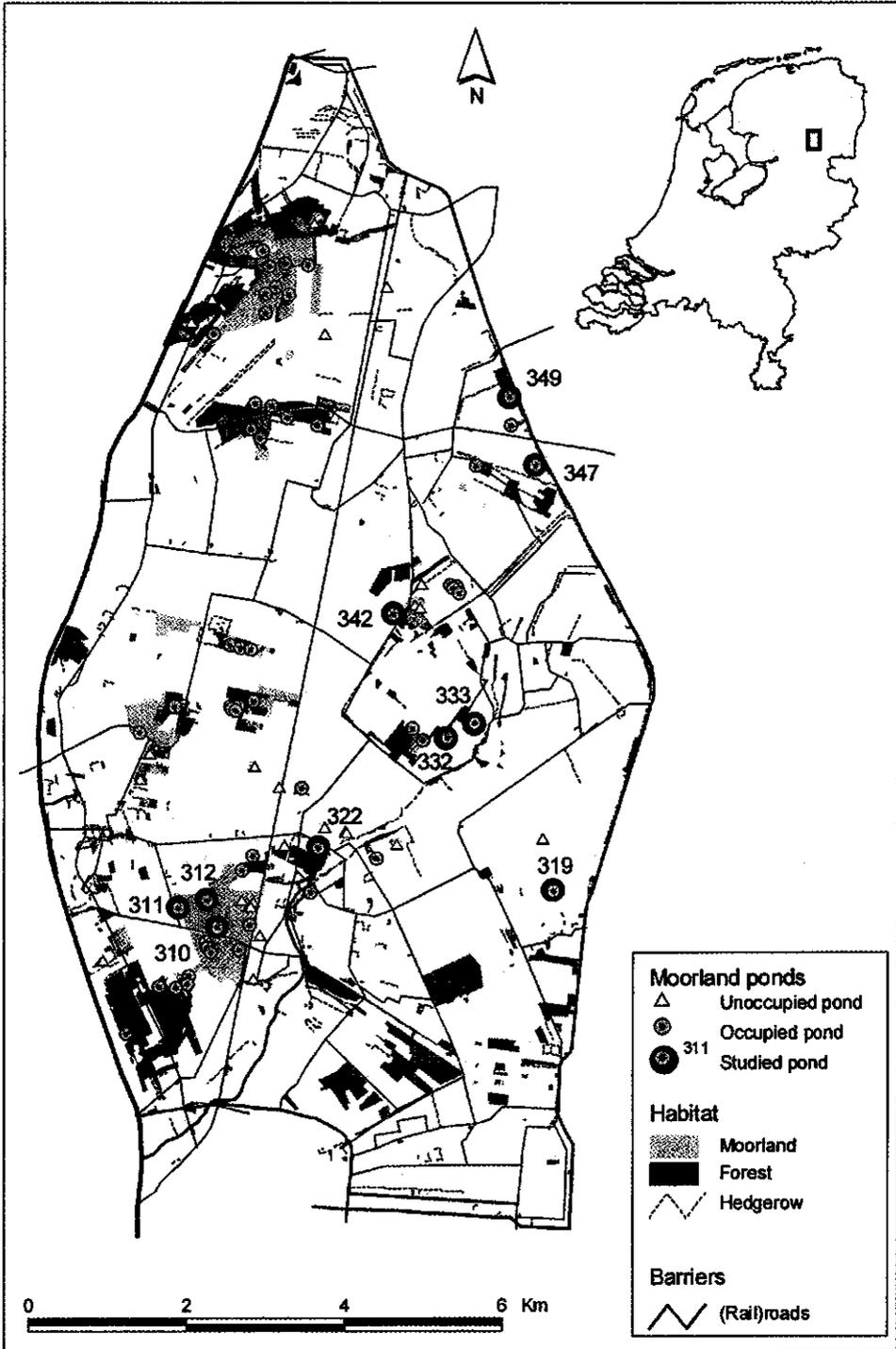


Figure 4.1  
The study area with the ten moorland ponds sampled. Ditches are not indicated.

generations have developed since the fragmentation of the area. Overlapping generations do occur, which will reduce the effective number of generations. Estimations of the annual adult mortality are between 40% (Loman 1987) and 60% (Ryser 1988).

### Geographical distance and landscape resistance

The spatial configuration of the moor frog habitat, the habitat types in the landscape matrix between suitable habitat patches, roads, railroads and ditches were recorded from a topographical map 1: 25 000 using a Geographic Information System (Fig. 4.1). Distances between all moorland ponds were calculated. To correct for the relative resistance of the landscape, length and area were calculated of habitat types and linear landscape elements in a 200 m wide strip between ponds (Table 4.1). Habitat types that are being avoided by moor frogs such as dry and open areas (Hartung 1991), or that form relative barriers, such as (rail)roads, were considered negative landscape elements that increase the resistance of the landscape. Habitat types that are preferred by moor frogs (Hartung 1991) and that resemble moor frog habitat were regarded as positive landscape elements that reduce resistance (Forman 1995). So deciduous forest, mixed forest, hedgerows and ditches were classified as positive landscape elements, while agricultural land, built-up areas, roads and

Table 4.1

The calculation of the resistance variables:  $D \cdot R_L$ , distance weighted for the fraction of negative linear elements;  $D \cdot R_A$  distance weighted for the fraction of negative area;  $D \cdot C_L$  distance corrected for the fraction of positive linear elements.

|  |  |
|--|--|
| <b><math>D \cdot R_L</math></b><br>Distance weighted for the fraction of barriers<br>- negative linear elements  | where $R_L = (NL + D) / D$<br>being the length of negative linear elements relative to the distance between two ponds.<br>$R_L$ is a figure between 1 and infinity, without dimension. It increases with NL.                         |
| <b><math>D \cdot R_A</math></b><br>Distance weighted for the fraction of barriers<br>- negative area             | where $R_A = (NA + SA) / SA$<br>being the negative area relative to the total area in a strip of 200 m wide between two ponds.<br>$R_A$ is a figure between 1 and infinity, without dimension. It increases with NA.                 |
| <b><math>D \cdot C_L</math></b><br>Distance weighted for the fraction of corridors<br>- positive linear elements | where $C_L = D / (PL + D)$<br>being the length of positive linear elements relative to the distance between two ponds.<br>$C_L$ is a figure between 1 and 0, without dimension. It decreases asymptotically to 0 with increasing PL. |

Habitat types and linear landscape elements were measured in a strip of 200 m width between moorland ponds.

D = Distance between ponds (m)

SA = Strip Area ( $D \cdot 200$ ) ( $m^2$ )

NL = Negative Linear elements: roads (weighted for car density; Vos & Chardon 1998), railroads (m)

NA = Negative Area : agricultural fields, built-up areas ( $m^2$ )

PL = Positive Linear elements: hedgerows, ditches (m)

railroads were regarded as negative landscape elements. Three resistance variables were calculated. In  $D \cdot R_L$  the distance ( $D$ ) is corrected with  $R_L$ , the fraction of negative linear elements: roads (weighted for car density, Vos & Chardon 1998) and railroads. In  $D \cdot R_A$  the distance is corrected with  $R_A$ , the fraction of negative areas: agricultural land and built-up area. In  $D \cdot C_L$  the distance is corrected with  $C_L$ , the fraction of positive linear elements: hedgerows and ditches (see Table 4.1 for details).

### Sampling

Ten populations were sampled in the study area in 1995 (Fig. 4.1). Three samples were taken in ponds in the same moorland at close range from each other (pond 310, 311, and 312, Fig. 4.1). The distances between the three sites varied from 0.28 to 0.51 km and the landscape between the three breeding sites consisted of continuous moorland, which is suitable terrestrial habitat for the moor frog. Seven populations were sampled from moorland ponds on distances ranging from 0.5 to 8 km from the main moorland. These ponds were separated by agricultural landscape (Fig. 4.1). To get an impression of the overall genetic diversity of the moor frog, one population (Beuven) was sampled at 20 km distance from the study area within the province of Drenthe. A second population (Heerenven) was sampled in the province of Limburg at approximately 150 km from the study area. An indication for the population size was derived from the number of egg clumps based on estimates from 1993 (Vos & Chardon 1998; Table 4.2). Since females lay a single egg clump, the number of egg clumps is an estimate for the reproductive population size (Schlupmann 1988;

Table 4.2

Details of the sampled populations. The egg-clump classes are an indication for population size.

|                  | MOORLAND<br>POND | NUMBER OF<br>SAMPLED<br>INDIVIDUALS | NUMBER OF<br>EGG<br>CLUMPS | POND AREA<br>(HA) |
|------------------|------------------|-------------------------------------|----------------------------|-------------------|
| Study area       | 310              | 19                                  | 100-250                    | 0.511             |
|                  | 311              | 6                                   | 5-15                       | 0.008             |
|                  | 312              | 17                                  | 50-100                     | 0.190             |
|                  | 319              | 17                                  | 100-250                    | 0.320             |
|                  | 322              | 16                                  | 100-250                    | 0.202             |
|                  | 332              | 18                                  | 100-250                    | 0.296             |
|                  | 333              | 12                                  | 25-50                      | 0.121             |
|                  | 342              | 24                                  | 25-50                      | 0.643             |
|                  | 347              | 7                                   | 25-50                      | 0.181             |
|                  | 349              | 9                                   | 25-50                      | 0.329             |
| Study area total |                  | 145                                 |                            |                   |
|                  | Beuven           | 20                                  | > 1000                     | 0.581             |
|                  | Heerenven        | 18                                  | 500-1000                   | 5.750             |
| Total            |                  | 183                                 |                            |                   |

Griffiths & Raper 1994). For the two reference populations, the number of egg clumps in 1995 was taken as an estimate for population size. In each population 20 egg clumps were sampled, taking several eggs from each clump. If less than 20 egg clumps were found, all available clumps were sampled. As moor frogs are explosive breeders with a very short breeding season, it is unlikely that males participate more than once in reproduction, although this cannot be ruled out completely. Therefore, each clump was regarded to represent one reproducing couple. The eggs were raised in captivity and one of each clump was harvested in its tadpole stage. As old egg clumps cannot be distinguished with certainty from those of the common frog (*Rana temporaria*; Büchs 1987; Rahmel & Eikhorst 1988), specific characteristics were checked at the tadpole stage. Due to small population size, mortality during raising and some tadpoles that were identified to be common frogs, the number of samples for some populations dropped under 20. In Table 4.2 the number of samples per moorland pond was given. Tadpoles were frozen in liquid nitrogen and stored at -80 °C until DNA extraction. DNA was extracted from nuclei as described by Bernatzky and Tanksley (1986) modified after Vosman *et al.* (1992).

### Microsatellite development

A search for *Ranidae* sequences in the EMBL and Genbank databases using SRS (Sequence Retrieval System, version 3.1) yielded 167 sequences up to October 29, 1996. The sequences retrieved were subsequently screened for homology to an artificial sequence containing all mono-, di-, and trinucleotide motifs, plus two tetranucleotide repeats (ATCT and ATGT; Smulders *et al.* 1997) using FastA (Pearson & Lipman 1988). The first selection was for those sequences that had at least 12 mononucleotide repeat units, 6 repeat units for di- and trinucleotide repeats, or 4 repeat units for the tetranucleotide repeats, allowing for 1 base pair mismatch internally. Some compound repeats were also included. Primer pairs were designed flanking microsatellite loci using PRIMER (version 0.5). Upon amplification in a small number of test samples, five loci proved to be useful for microsatellite analysis in the moor frog (Table 4.3). A check whether the fragments amplified were alleles of the same microsatellite locus was done by purification of fragments from gel, reamplification, and direct sequencing using a Taq DyeDeoxy Terminator Cycle Sequencing kit on a ABI 370 sequencer.

### Amplification reactions and detection of microsatellite polymorphisms

Each 25 µl amplification reaction contained: 10 ng genomic DNA, 0.32 µM of each primer (Isogen, Maarssen, The Netherlands), 100 µM deoxy-ribonucleo-tides, 50 mM KCl, 20 mM Tris-HCl (pH 8.4), 1.5 mM MgCl<sub>2</sub>, 0.05% (v/v) polyoxyethylene ether (W-1), and 0.5 U Taq DNA polymerase (Life Technologies). Amplifications were performed in microtiter plates using a Hybaid Omni Gene thermal cycler. Basically, the amplification conditions were: 1 cycle of 94 °C for 3 min; 30 cycles of 55 °C for 45 s, 72 °C for 1 min 45 s, and 94 °C for 45 s. After the final cycle, 1 cycle of 55 °C for 45 s and 72 °C for 3 min was added (Bredemeijer *et al.* 1998). If necessary, the annealing temperature was lowered to 50 °C, and the number of cycles increased to 35 (details in Table 4.3).

Samples were prepared for PAGE electrophoresis by adding an equal volume of formamide, containing 10 mM NaOH and 0.05% bromphenol blue, to the reaction mixtures. After denaturation at 80 °C for 5 min followed by quenching on ice, samples were analysed on vertical gels (6% polyacrylamide, 8 M urea, Trisborate buffer) using a Model S2 sequencing gel electrophoresis apparatus (Life Technologies). The DNA bands were visualized by silver staining according to the Silver Sequence DNA sequencing system (Promega). The sizes of the PCR products were determined by comparison to an accompanying sequence reaction using pGEM-3Zf(+) control DNA (Promega).

### Statistical analysis

The fraction of observed and expected heterozygotes, the mean number of alleles per locus and Wright's (1965) inbreeding coefficient ( $F_{is}$ ) were calculated as measures for gene diversity within populations (Cockerham 1973). Hardy-Weinberg equilibrium was tested using a Markov Chain test (with Arlequin 1.0). The degree of differentiation between populations was estimated with Wright's (1965) index of population subdivision ( $F_{st}$ ). Genetic similarity between populations was estimated by Nei's (1972) index for genetic distance. As a measure for the amount of gene flow between populations within the study area correlations between genetic distance and geographical distance were tested with the Mantel test (Mantel 1967) using the statistical program Genstat (Genstat 5 Committee 1993). In a first step of analysis, geographical distance (D) and the distance variables weighted for relative resistance ( $D \cdot R_L$ ,  $D \cdot R_A$ , and  $D \cdot C_L$ ) were tested in a single Mantel test. In a second step, a multiple Mantel test was conducted. First the geographical distance variable was put into the model. Subsequently the model was extended with the relative resistance variables ( $R_L$ ,  $R_A$ , and  $C_L$ ; note that the variables are without a geographical distance component) to test whether resistance variables contribute significantly to the model in addition to geographical distance.

## Results

To generate STMS markers for use in the moor frog, microsatellite-containing sequences from *Rana* species were extracted from the EMBL and Genbank databases. In total, 167 sequences yielded 11 microsatellite sequences in 9 accessions. For these 11 microsatellites, primer pairs were designed and amplification was tested in a small number of moor frogs, and also in one common frog. After amplification, five loci (Table 4.3) were found to produce fragments that were polymorphic among the moor frogs, with two to eight alleles per locus (Table 4.4). The polymorphism information content (PIC) ranged from 0.23 to 0.60 (Table 4.4). All microsatellite loci also amplified a product in the common frog. Some fragments produced by loci RRD590, RNTYR2, and RC08604 were purified from the gel, reamplified, and sequenced (not shown). In all cases they were found to be alleles of different repeat lengths, but with some point mutations throughout the fragment, when compared to the database sequence from other *Rana* species used for designing primers. The primers for RC08604 amplified two sets of bands, four fragments between 174 and

Table 4.3  
Database-derived microsatellite loci and designed primer pairs.

| DATABASE ENTRY | ACCESSION NUMBER | RAWA SPECIES OF ORIGIN   | REPEAT <sup>1</sup>        | REPEAT LOCATION <sup>2</sup> | PREDICTED FRAGMENT SIZE (bp) | PRIMER SEQUENCES (FORWARD, REVERSE)                              | AMPLIFICATION CONDITIONS USED |
|----------------|------------------|--------------------------|----------------------------|------------------------------|------------------------------|--|-------------------------------|
| RRD590         | D78590           | <i>R. rugosa</i>         | (GAA)3(GAG)13 <sub>2</sub> |                              | 176bp                        | 5'-GATTAGTAAACAATAAAGGCACATG-3'<br>5'-CTTTGGAGCAGGAGGTG-3'       | 55°C 30 cycles                |
| RECALQ         | X64324           | <i>R. esculenta</i>      | (GAT/C)42                  | m                            | 208bp                        | 5'-GGAGGCTGAAGTCAACACT-3'<br>5'-ACATTTGAATATATAAACTAGTTAGATGC-3' | 50°C 35 cycles                |
| RNTYR2         | D37779           | <i>R. nigro-maculata</i> | (T)22 <sub>1</sub>         | i                            | 195bp                        | 5'-TGGGACTTTTGTAGCTG-3'<br>5'-GCCATAGGCCATAGAGCG-3'              | 50°C 35 cycles                |
| RCIDI          | L42815           | <i>R. catesbeiana</i>    | (TA)9 <sub>2</sub>         | m                            | 173bp                        | 5'-GCTGAATTTTGGAAAGATGAG-3'<br>5'-CAGGAAGAGCAAAAGTTCC-3'         | 50°C 30 cycles                |
| RC08604        | U08604           | <i>R. catesbeiana</i>    | (C)15                      |                              | 174bp                        | 5'-TTGGAGACCCTCAGGG-3'<br>5'-TCATGGGAAGTGCTCTTATCAC-3'           | 50°C 33 cycles                |

<sup>1</sup> - nr, indicates mismatch    <sup>2</sup> m = mRNA; i = intron

Table 4.4  
Population genetic statistics of the moor frog for the ten populations in the study area and for all twelve populations (including Beuven and Heerenven). The number of alleles and the polymorphism information content (PIC) are listed for each locus. The observed heterozygosity ( $H_o$ ) and the expected heterozygosity ( $H_e$ ) are indicated for the loci separately and for all loci together. The relative inbreeding within populations ( $F_{is}$ ) and the interpopulation variation ( $F_{st}$ ) are listed.

| LOCUS                                | ALLELES FOUND <sup>1</sup>      | PIC   | $H_o$ | $H_e$ | $F_{is}$ | $F_{st}$ |
|--------------------------------------|---------------------------------|-------|-------|-------|----------|----------|
| POPULATIONS IN THE STUDY AREA (N=10) |                                 |       |       |       |          |          |
| RRD590                               | 181-189-205-209-223             | 0.225 | 0.248 | 0.230 |          |          |
| RECALQ                               | 195-201-217-226-229-232-235-241 | 0.239 | 0.213 | 0.205 |          |          |
| RNTYR2                               | 136-139                         | 0.366 | 0.338 | 0.355 |          |          |
| RCIDI                                | 170-171-172                     | 0.598 | 0.532 | 0.598 |          |          |
| RC08604                              | 311-314-315-317-320             | 0.483 | 0.366 | 0.495 |          |          |
| all loci                             |                                 |       | 0.340 | 0.378 | 0.049    | 0.052    |
| ALL POPULATIONS (N=12)               |                                 |       |       |       |          |          |
| all loci                             |                                 |       | 0.337 | 0.383 | 0.052    | 0.065    |

<sup>1</sup> most frequent allele underlined

180 bp (the expected fragment size was 174 bp), and five fragments between 311 bp and 320 bp. During the reamplification of the larger fragments, both a large and one of the small fragments were amplified. Upon sequencing, it became clear that the primer binding site was present at the expected position, but apparently also some 160 bp downstream, one that was also used in the PCR reaction. In the original sequence from *Rana catesbeiana*, a duplication of the primer binding site was present at this downstream site, with only one bp mismatch. Polymorphisms among the fragments were present both upstream and downstream of the first primer binding site, hence the two groups of fragments did not reflect exactly the same variation, and we subsequently used the large fragments as representing the polymorphisms of this locus.

Using these five primer pairs, genetic variation was measured among the ten populations in the study area in Drenthe ( $n = 145$  samples, Table 4.2), and in the Beuven ( $n = 20$ , Table 4.2) and the Heerenven ( $n = 18$ , Table 4.2). Although the primers amplified between two and eight alleles per locus, mostly one or two alleles were present in higher frequencies (underlined in Table 4.4). The other alleles were either very rare, or had low but similar frequencies in most populations, so that they provided hardly any discriminating power. The only exception was locus RECALQ, where alleles 232 bp and 235 bp amounted to 15% and 18% of the alleles in the population of Heerenven, which was double the frequency of any other population.

The observed heterozygosity ( $H_o$ ) and the expected heterozygosity ( $H_e$ ) for the five loci separately and for all loci together are listed in Table 4.4, followed by the relative inbreeding within populations ( $F_{is}$ ).  $F_{is}$  for all ponds (0.052) and  $F_{is}$  for the ten ponds within the study area (0.049) were low, giving no indication of loss of heterozygosity. Indeed, most populations were in HW equilibrium (except for 312 and 342, exact  $p$  value (Markov chain) of HW equilibrium 0.0003 and 0.0055, respectively).

There was a limited degree of interpopulation genetic variation, based on the  $F$  statistic ( $F_{st}$  among all populations 0.065, Table 4.4). Also among the ten populations within the study area the population subdivision was low.  $F_{st}$  within the study area was 0.052 (Table 4.4).

With the Mantel test a possible correlation between genetic distance and the geographical distance between pairs of ponds was analysed within the study area. Nei's estimate for genetic distance showed a significant correlation with geographical distance (Table 4.5). Genetic distance increased with geographical distance ( $D$ ) between moorland ponds ( $D$  p (Mantel)  $< 0.05$ , Table 4.5). Also the resistance variables in which distance was corrected for the fraction of negative linear elements ( $D \cdot R_L$ ), the fraction of negative area ( $D \cdot R_A$ ) and the fraction of positive linear elements ( $D \cdot C_L$ ) showed a significant correlation with genetic distance ( $D \cdot R_L$  p (Mantel)  $< 0.05$ ;  $D \cdot R_A$  p (Mantel)  $< 0.05$ ;  $D \cdot C_L$  p (Mantel)  $< 0.05$ , Table 4.5). To test whether resistance variables also gave an explanation of genetic distance in addition to geographical distance, a multiple Mantel test was conducted. In a model with geographical distance, the variable for the fraction negative linear elements ( $R_L$ ) showed a significant correlation with genetic distance ( $R_L$  p (Mantel)  $< 0.05$ , Table 4.5). Geographical distance did not add significantly to the explanatory value of the model, although there was a positive correlation ( $D$  p (Mantel)  $< 0.1$ , Table 4.5). The models with geographical distance and the variables for the fraction

Table 4.5

Mantel test for the relation between genetic distance (Nei 1972) and geographical distance between pairs of moorland ponds in the study area. Geographical distance (D) and the distance variables weighted for relative resistance ( $D \cdot R_L$ ,  $D \cdot R_A$ , and  $D \cdot C_L$ ) were tested in a single Mantel test. In a multiple Mantel test the geographical distance variable was added first and subsequently the model was extended with the relative resistance variables ( $R_L$ ,  $R_A$ , and  $C_L$ ; note that these variables are without a geographical distance component). See Table 4.1 for further details on the calculation of the resistance variables. Statistical significance is based on randomisation values.

| SINGLE MANTEL TEST |              |  |
|--------------------|--------------|--|
| VARIABLE           | SIGNIFICANCE | DESCRIPTION OF VARIABLE  |
| D                  | $p < 0.05$   | D = distance between pond pairs  |
| $D \cdot R_L$      | $p < 0.05$   | $D \cdot R_L$ = distance weighted for the fraction of negative linear elements (roads and railroads) |
| $D \cdot R_A$      | $p < 0.05$   | distance weighted for the fraction of negative area (agricultural fields and built-up areas)         |
| $D \cdot C_L$      | $p < 0.05$   | distance weighted for the fraction of positive linear elements (hedgerows and ditches)               |

| MULTIPLE MANTEL TEST |                              |  |
|----------------------|------------------------------|--|
| D<br>and<br>$R_L$    | $ns$ $p < 0.1$<br>$p < 0.05$ | $R_L$ = fraction of negative linear elements (roads and railroads)         |
| D<br>and<br>$R_A$    | $ns$<br>$ns$                 | $R_A$ = fraction of negative area (agricultural fields and built-up areas) |
| D<br>and<br>$C_L$    | $ns$<br>$ns$                 | $C_L$ = fraction of positive linear elements (hedgerows and ditches)       |

Table 4.6

Correlation matrix of the model variables, \*\*  $p < 0.01$ , for abbreviations see Table 4.5.

|       |           |           |           |       |
|-------|-----------|-----------|-----------|-------|
| D     | -         |           |           |       |
| $R_L$ | 0.267 **  | -         |           |       |
| $R_A$ | 0.538 **  | 0.348 **  | -         |       |
| $C_L$ | -0.356 ** | -0.493 ** | -0.685 ** | -     |
|       | D         | $R_L$     | $R_A$     | $C_L$ |

of negative area ( $R_A$ ) or positive linear elements ( $C_L$ ) showed no significant relation with genetic distance, yet significant effects were shown in single variable models (Table 4.5). Their failure in the multiple model could be explained by the correlations between these relative resistance variables and the geographical distance variable ( $D$  and  $R_A$   $r = 0.538$   $p < 0.01$ ;  $D$  and  $C_L$   $r = -0.356$   $p < 0.01$ , Table 4.6).

## Discussion

Using five microsatellite loci, a low level of genetic population subdivision was found for the moor frog, in a landscape that became fragmented 60 years ago. Also the levels of heterozygosity within the populations were not much below the expected values. These results indicate that at least within the studied landscape and time scale, habitat fragmentation has not seriously affected the genetic structure of the local populations. However, even though the genetic similarity between populations was high, there was a correlation between genetic distance and geographical distance within the study area. Scaling the genetic similarity on the actual spatial distribution of the local populations, populations that are close to each other were more alike than more distant populations. The relative resistance variable for the fraction of negative linear elements (roads and railroads) gave a higher explanatory value than geographical distance. The correlation between genetic distance and geographical distance indicates that dispersal rate between populations decreases with distance. In particular, the amount of barriers (roads and railroads) between populations seem to reduce exchange between populations.

### Comparison of $F_{st}$ with other species

Ward et al. (1992) compared  $F_{st}$  values based on allozymes for several taxa and found a relatively high mean  $F_{st}$  value for amphibians ( $F_{st} 0.315 \pm 0.040$ ). They concluded that species mobility is a strong factor influencing the magnitude of population structure, amphibians being relatively immobile species. Scribner et al. (1994) compared  $F$  statistics derived from allozymes, minisatellites and one microsatellite. They found that mini- and microsatellites, which are much more variable, give lower levels of inbreeding and larger genetic distances compared to allozymes. Apparently, the  $F_{st}$  values found in this study were very low, compared to the mean  $F_{st}$  values for amphibians, especially as microsatellites tend to give larger genetic distances. This is probably related to the fact that the fragmentation in the study area is too recent to have seriously affected population structure.

### Geographical distance and landscape resistance

Genetic distance increased with geographical distance. A correlation between genetic distance and geographical distance has been demonstrated for many species in different taxa (Avice 1994). In a multiple Mantel test the fraction of (rail)roads gave a better explanation of genetic distance than geographical distance. Geographical distance was no longer significant in a model with the fraction of (rail)roads. However, one would expect an effect of geographical distance as the number of

dispersers will decrease with distance. In the study area the largest distance between populations is 8 km, which exceeds the dispersal capacity of the species. Although data on the dispersal capacity of moor frogs are scarce, distances will probably not exceed 1 to 3 km (Hartung 1991). Still, some gene flow between these populations might be possible, when intermediate populations function as stepping stones. The fact that the amount of (rail)roads in the landscape lower genetic similarity is an indication that some exchange between populations takes place and that this is hampered by (rail)roads.

In an earlier study in the same area, a negative effect of road density was demonstrated on the distribution pattern of the moor frog (Vos & Chardon 1998). The probability that a pond was occupied by the moor frog lowered with increasing road density in a radius of 750 m. Hitchings and Beebee (1997) also found a stronger correlation between genetic distance and geographical distance weighted for relative resistance. In an urban environment a correction for the amount of post 1932 development improved the correlation with genetic distance (Hitchings & Beebee 1997).

The corrections for landscape resistance are based on the assumption that habitat types resembling suitable habitat will decrease resistance (Forman 1995) and therefore increase dispersal success. On the other hand, extra mortality on roads and in other hostile habitats will decrease the probability of successful dispersal. The mortality risk of amphibians crossing roads is well known (Van Gelder 1973; Berthoud & Müller 1983; Heine 1987; Kuhn 1987; Münch 1989; Hels 1999). Avoidance of unsuitable habitat types such as open fields and dry areas by moor frogs was found by Hartung (1991). Three measures describing landscape features that might contribute to landscape resistance were analysed separately. By this method it is possible to get an indication which habitat type or linear landscape elements most strongly determine landscape resistance. To incorporate all features into one resistance variable, a better understanding would be required of the interaction between moving organisms and the configuration of negative and positive landscape elements. Dispersal models (e.g. Tischendorf & Wissel 1997; Vos et al. submitted) based on individual movement patterns in heterogeneous landscapes, might be a better alternative. With dispersal models one can predict the influence of preference and avoidance of habitat types and the effect of absolute barriers on the connectivity between habitat patches (Wiens 1992).

### **Genetic structure and dispersal**

The distribution of barriers in the present landscape plays a role in explaining the genetic distance between populations (Hitchings & Beebee 1997; this article). This suggests that dispersal between populations is reduced as a result of habitat fragmentation. In the study area the moor frog habitat has become fragmented fairly recently, through wide-scale cultivation in the 1930s. The maximum number of generations since the fragmentation of the habitat is 30, but much less if we take overlapping generations into account. Therefore, the present genetic structure will to a large extent still reflect the former situation.

Especially when the genetic similarity between populations is high, differences can only be demonstrated with many loci and large sample sizes. In this

study five microsatellites were tested and sample size per population varied. The effects of the five loci were quite variable, indicating that using more loci could make the results more stable. For a better estimate of population subdivision by  $F_{st}$  more loci will have to be tested. Three microsatellites and six repeat-containing genomic fragments have recently been reported for *Rana luteiventris* (Call & Hallett 1998). Otherwise, no more microsatellites are available in the EMBL and Genbank databases, so that it will be necessary to clone microsatellite-containing sequences from this frog species directly, e.g. using an enrichment procedure (Van Der Wiel et al. 1998).

An approach to get a better insight to what extent genetic differentiation is related to reduced dispersal, is to compare genetic structure in landscapes with different degrees of habitat fragmentation. To test this method a second study is being carried out in a landscape with less suitable habitat and higher traffic intensity of the road network. Still for a proper interpretation of the present genetic structure information about the historic distribution pattern is essential. To better understand processes of drift and gene flow in fragmented landscapes, it may be useful to monitor the genetic structure at regular time intervals.

## 5 Empirical evidence of metapopulation dynamics; the case of the tree frog (*Hyla arborea*)

### Summary

In nature conservation many recommendations for the protection of species in fragmented landscapes are based on metapopulation concepts for optimal spatial habitat configuration. However, conservation measures on a landscape or regional scale will only be effective if fragmentation of habitat is a major cause for decline. In this paper it is discussed which empirical evidence is ideally needed to conclude that metapopulation processes play a dominant role in explaining the distribution of a species in a landscape. In a case study of the tree frog (*Hyla arborea*) four aspects of the distribution pattern are analysed. The results seem to justify the conclusion that the tree frog in the particular study area functions as a metapopulation. 1. Extinctions do take place regularly and are influenced by spatial features of the landscape: extinction probability decreases with patch size. 2. Empty patches are recolonised and this process is influenced by the configuration of habitat: colonisation probability increases for large patches with high connectivity. 3. The observed dispersal distances in comparison with the distances between patches and the observed recolonisations, show that the habitat network is still connected by dispersing individuals. The observed dispersal events indicate that dispersal distances towards occupied patches are larger. The possible role of conspecific attraction in unpredictable habitats and the implications for metapopulation survival are discussed. To answer the question if the habitat network is still viable, an extended incidence function model, *Wink*, is developed. Simulations indicate that the metapopulation is still viable. However, several aspects that will influence viability negatively, are not incorporated in the model such as the high turnover of suitable habitat by habitat destruction and natural succession.

### Introduction

Species in fragmented landscapes are often found absent in apparently suitable habitat. When these absences are correlated with landscape structure, i.e. the small and isolated patches are empty relatively often, the species is concluded to suffer from habitat fragmentation. Effects of habitat fragmentation have been demonstrated in empirical studies for a wide range of species - landscape combinations (see for reviews Harrison 1991, 1994; Opdam 1991; Reich & Grimm 1996, Hanski & Gilpin 1997). Recommendations for the protection of species in fragmented landscapes are often based on metapopulation concepts. Metapopulation theory implies that although small populations suffer from chance extinction due to demographic stochasticity, the species can survive on a regional level, if local extinctions are compensated for by recolonisations (Levins 1970).

Since the first definition (Levins 1970) the metapopulation concept has evolved and diversified. The definitions range from narrow, involving a strict extinction-colonisation balance (Harrison 1994; Harrison & Taylor 1997) to broad, such as 'a set of local populations within some larger area, where typically migration from one local population to at least some other patches is possible' (Hanski & Simberloff 1997). The latter definition emphasises the spatial structure of a metapopulation and disregards the matter of persistence. Following Shaffer's (1981) definition of a viable population (Verboom 1996), we prefer to include the viability criterion into the definition, because such a definition is useful for nature conservation: a viable metapopulation is a set of local populations distributed over a spatially discontinuous habitat network and linked by dispersal, the size of which as well as the balance between local extinction and recolonisation are such that its probability of extinction is 5% or less over 100 years. Fragmentation processes characteristically lead to a large variation in patch size and patch clustering (Gardner *et al.* 1987). This can result in mainland-island (Boorman & Levitt 1973) or source-sink (Pulliam 1988) patterns of fragmentation. Note that large or source patches with a major impact on the survival probability of the metapopulation (Harrison 1991; Murphy *et al.* 1990) fall within our definition. With this interpretation of a metapopulation, each species will show metapopulation characteristics at some degree of landscape fragmentation and at some spatial scale. The key parameter is the minimum required amount of suitable habitat, below which a species will go extinct, the so-called extinction threshold (Lande 1987; Bascompte & Solé 1996; Fahrig 1997; Vos *et al.* submitted). It depends on the spatial scale at which a species operates and the amount to which its habitat is fragmented. Below the threshold local extinctions are no longer sufficiently compensated for, and a species will go extinct regionally in the foreseeable future. For nature conservation the issue at stake is which species in habitat networks are or are not able to cope with, often rapid and humanly induced, landscape changes or already have crossed the extinction threshold.

How can we assess if species, for which effects of fragmentation can be demonstrated, indeed function as a metapopulation? So that it is plausible that improvements on the spatial structure of suitable habitat indeed be effective for species persistence? For the conservation of a species it is important to know whether or not processes of extinction and recolonisation take place, and if so at what time scale. A single observed distribution pattern of a species is only a snapshot of a dynamic series of possible distributions. It provides no information about presumed underlying processes. Unfortunately, there are few empirical studies in which effects of landscape fragmentation on the processes of local extinction and recolonisation were demonstrated (see for a review Harrison 1991, 1994; Opdam 1991; Reich & Grimm 1996; Hanski & Gilpin 1997). What empirical evidence do we need to conclude from a species distribution that metapopulation processes play a dominant role? Ideally the following questions should be answered:

- 1 Does extinction of local populations take place, if so at what rate do extinctions occur and are local extinctions correlated with patch size?
- 2 Are empty patches being recolonised, if so at what rate do colonisations occur and are local colonisations correlated with patch connectivity?
- 3 Does dispersal of individuals between habitat patches actually occur, as an

indication that the habitat network has not disintegrated? This depends on the dispersal distance and the number of dispersers in relation to the distance between patches and local populations.

If metapopulation processes seem to be relevant, a fourth question should be answered, which is the most important question from a conservation point of view:

- 4 Do the size of the habitat network and the ratio between average extinction and recolonisation frequencies indicate that the metapopulation is still above its extinction threshold and will be viable in time?

The aim of this paper is to answer these questions for a concrete case: the tree frog (*Hyla arborea*) in Zeeland Flanders. We have analysed three consecutive annual distribution patterns to determine if local extinction and recolonisation probability are affected by habitat fragmentation. The third question has been answered by the analysis of data of individual movements registered with capture-recapture techniques. The possible influence of conspecific attraction on dispersal has been taken into account by analysing whether dispersal is more often directed towards occupied patches. The occurrence of conspecific attraction can be a relevant factor in the metapopulation process, as it could influence colonisation and extinction probabilities. The influx of individuals into a population could lower the extinction probability, the so-called rescue effect (Brown & Kodric-Brown 1977). However, by consequence, empty patches will receive less dispersers, which will lower the colonisation probability (Ray et al. 1991; Smith & Peacock 1990). Conspecific attraction therefore may influence processes at the metapopulation level (Ray et al. 1991).

Finally to predict the viability of a metapopulation a model is needed in which local extinction and colonisation probabilities are simulated for all patches over time. For this purpose the incidence function model *Wink* was developed (Verboom et al. 1991a; Hanski 1994; Ter Braak et al. 1998). *Wink* is a patch occupancy model (Hanski & Simberloff 1997), a spatially explicit metapopulation model in which patches can be either occupied or empty. We developed a new method to fit the incidence function model to data, based on Ter Braak et al. 1998. The extinction and recolonisation functions are estimated from both the turnover events between years and the spatial distribution pattern of the first year of survey. The advantage of this method is that the available information in the occupancy data is optimally applied, which will improve the quality of estimated functions. To make the model more realistic, *Wink* has been extended to include habitat quality variables, that proved to be important in predicting extinction and colonisation probabilities (Vos & Stumpel 1996; this article).

## Methods

### Analysis of extinction and colonisation

The study area is 250 km<sup>2</sup> and is situated in the western part of Zeeland Flanders (Fig. 5.1). The habitat of the tree frog in this mainly agricultural landscape consists of cattle drinking ponds and shrubs, bushes and vegetation of tall herbs. The suitable

habitat covers approximately 1.5 % of the total landscape. Census data of tree frog distribution were collected in three successive years (1981-1983) and in 1986. The ponds were visited at least three times a year during the reproduction period. See Stumpel 1987a and Vos & Stumpel 1996 for details on survey methods for tree frogs presence, the collection of habitat quality parameters and the inventory of the suitable terrestrial habitat. For the analysis we selected a subset of 187 ponds of which both habitat quality and isolation variables were available out of approximately 500 ponds. This subset was a representative sample of the variation in isolation in the study area (Vos & Stumpel 1996). Arc-Info was used to obtain spatial habitat parameters. The size of a suitable patch for a local population was determined by taking into account both the aquatic habitat part (the pond) and the terrestrial habitat part (the area of suitable terrestrial habitat in a radius of 250 m surrounding the pond). The suitable terrestrial habitat within a distance of 250-300 m from the reproduction site is regarded as the main terrestrial habitat of a local population (Blab 1986; Stumpel 1987a; Tester 1990). For convenience, a patch, which is a combination of a pond and suitable terrestrial habitat, will in the next sections be referred to as a 'pond'.

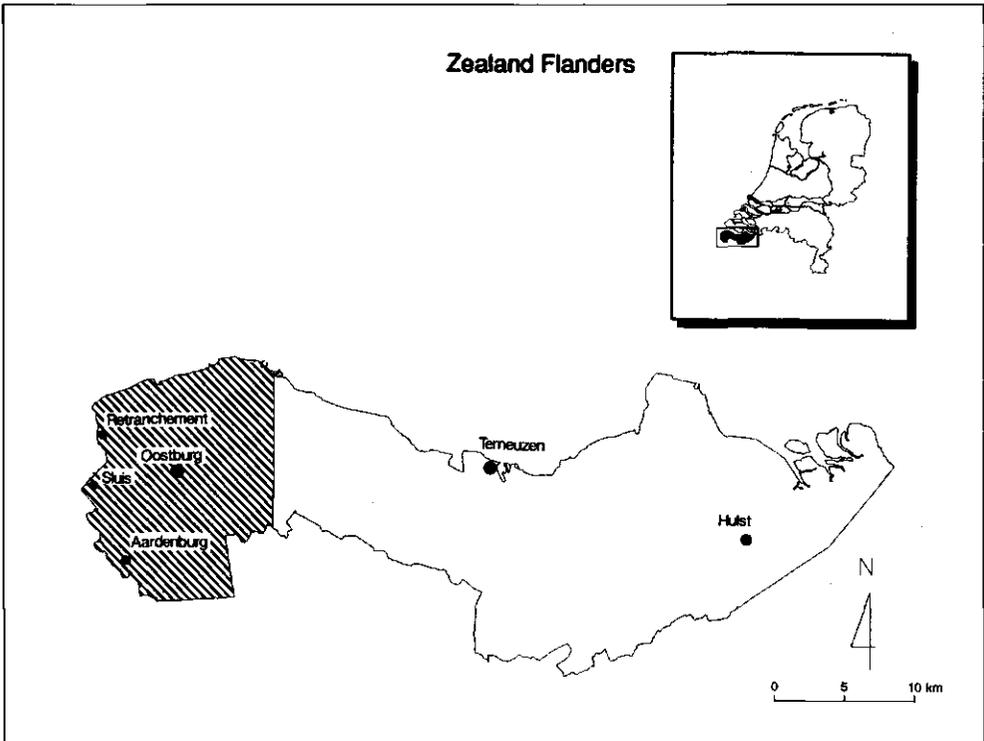


Figure 5.1  
The position of the study area in The Netherlands.

The connectivity of a pond  $i$  ( $S_i$ ) is based on an estimation of the number of migrants arriving in pond  $i$ , taking into account the distance and size of dispersal sources. The number of migrants is a weighted sum over all occupied ponds  $j$  unequal to  $i$  namely (Verboom et al. 1991a; Hanski 1994; Ter Braak et al. 1998):

$$S_i = \sum_{j=1}^n y_j A_j^b B_{ij} \exp(-\alpha d_{ij}) \quad (j \neq i) \tag{1}$$

where  $y_j$  equals 1 for occupied and 0 for empty patches,  $A_j^b$  is the area of pond  $j$ ,  $B_{ij}$  is an indicator for the presence of road barriers between pond  $i$  and  $j$  and  $\alpha$  is a constant setting the survival rate of migrants over distance  $d_{ij}$ , the distance between pond  $i$  and  $j$ . Since most tree frogs will disperse a distance of 2 km or less (Fog 1993; Figure 5.4b), occupied ponds within a maximum radius of 2 km from pond  $i$  were taken into account to calculate the connectivity. As the exact relation between dispersal distance and survival is not known for tree frogs an  $\alpha$  of 2 was used, which corresponds reasonably with the maximum dispersal distance of 2 km (Fig. 5.2). The area  $A^b$  consisted of pond area and the amount of suitable terrestrial habitat in a radius of 250 m surrounding the pond. When the pond area was unknown, the median value (250 m<sup>2</sup>) was taken. Three different connectivity measures were tested. It was tested whether area of source patches plays a role in connectivity. In the connectivity variable 'S' the pond area of the source patches was not taken into account ( $b = 0$ , Table 5.1). In the connectivity variables 'S-area' and 'S-area-barrier'

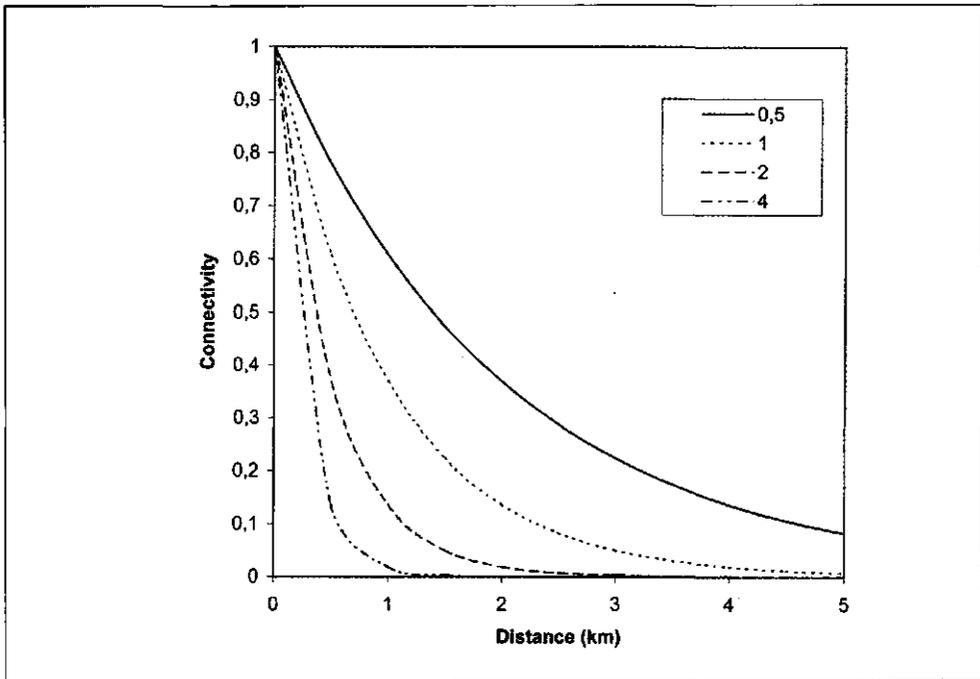


Figure 5.2  
The contribution of source patches to the connectivity of a target patch given the distance between source and target patch for different values of  $\alpha$  (see equation 1).

**Table 5.1**  
**Overview of the pond quality variables and habitat fragmentation variables. The mean value and the standard deviation (sd) are listed for ponds that remained occupied, went extinct, were colonised or remained unoccupied during two successive years in the survey period.**

| VARIABLES                 | DESCRIPTION  | OCCUPIED PONDS *<br>(n= 25)<br>t = 1 / t + 1 = 1<br>Mean ± sd | EXTINCTION PONDS<br>(n = 10)<br>t = 1 / t + 1 = 0<br>Mean ± sd | COLONISED PONDS<br>(n= 16)<br>t = 0 / t + 1 = 1<br>Mean ± sd | UNOCCUPIED PONDS<br>(n = 336)<br>t = 0 / t + 1 = 0<br>Mean ± sd |
|---------------------------|--|---|--|--|---|
| Cover                     | Coverage (%) of the pond surface by aquatic vegetation   | 48.3 ± 36.6<br>(48.2 ± 37.6)                                  | 49.1 ± 39.8  | 57.47 ± 41.3   | 21.20 ± 34.2  |
| Water-Cond                | Electrical conductivity pond water (µS/cm)   | 56.0 ± 25.8<br>(59.7 ± 26.9)                                  | 80.9 ± 38.7  | 74.5 ± 41.7  | 98.1 ± 40.4   |
| Area**                    | Pond area and area of suitable terrestrial habitat in a radius of 250 m surrounding the pond.  | 2.21 ± 1.92<br>(1.97 ± 1.80)                                  | 1.23 ± 1.13  | 1.48 ± 1.49  | 0.66 ± 1.3  |
| Connectivity measures (S) | $S_i = \sum y_j A_j^b B_{ij} \exp(-2d_{ij})^{***}$<br>$y_j$ = pond occupancy 0/1; $A_j^b$ = area pond $j$ ; $B_{ij}$ = road barriers between pond $i$ and $j$ ; $d_{ij}$ = distance between pond $i$ and $j$ | 0.49 ± 0.97<br>(-0.37 ± 2.00)                                 | 0.27 ± 0.72  | -0.77 ± 1.44   | -2.53 ± 1.66  |
| S**                       | Connectivity based on distance to all occupied ponds: $S_i = \sum y_j \exp(-2d_{ij})$<br>ditto including size of sources:<br>$S_i = \sum y_j A_j^{1/2} \exp(-2d_{ij})$                                       | 0.65 ± 1.19   | 0.46 ± 1.06  | -0.92 ± 1.78   | -2.95 ± 2.00  |
| S-area-barrier**          | ditto including size of sources and 50% of road barriers:<br>$S_i = \sum y_j A_j^{1/2} 0.5 \exp(-2d_{ij})$   | 0.33 ± 1.35<br>(-0.73 ± 2.51)                                 | 0.11 ± 1.20  | -1.13 ± 1.76   | -3.27 ± 2.10  |

\* In brackets values including three ponds that were removed from further analysis (see Table 5.2 and text for explanation); \*\* Because of skewed distribution log transformation was used; \*\*\* cf. Verboom et al. 1991a; Hanski 1994, Ter Braak et al. 1998.

the area of the source patches was incorporated ( $b = 1/2$ , Table 5.1). To test the influence of roads as barriers for crossing individuals, in one of the connectivity variables a barrier effect  $B_{ij}$  was considered. A negative effect of roads on amphibian survival and distribution has been demonstrated (Van Gelder 1973; Heine 1987; Fahrig et al. 1995; Vos & Chardon 1998). In the connectivity variable 'S-area-barrier' a barrier effect of 50% was presumed ( $B_{ij} = 1/2$ , Table 5.1), which means that the contribution of patches that are divided by a road from pond  $i$  contribute only 50% to the connectivity of pond  $i$ .

The presence-absence data of the successive years 1981-1983 were used for analysis of extinction and colonisation. An extinction of a local population was recorded when a pond was found occupied in a year  $t$  and unoccupied in year  $t+1$ . The reverse will be referred to as a colonisation. A one-year absence was chosen to include all possible recolonisations. However, in some cases a one-year absence of individuals may not necessarily indicate an extinction followed by a recolonisation. For instance juveniles, still present in the pond surroundings, could take part in reproduction the next year. Extinction and colonisation probability were analysed separately with logistic regression analysis (Jongman et al. 1995; Sjögren-Gulve & Ray 1996). Transitions between year 1981-1982 and 1982-1983 were analysed simultaneously (Verboom et al. 1991a; Ter Braak et al. 1998). Possible differences between years in colonisation and extinction probability were tested by adding a factor 'year of survey' to the regression model. Two habitat quality variables were tested: the vegetation cover of the pond surface (Cover, Table 5.1) and electrical conductivity of the pond water (Water-Cond, Table 5.1). These variables had proved to be important in predicting the distribution pattern of the tree frog in an earlier study (Vos & Stumpel 1996). In the regression analysis first the variable 'year of survey' and the pond quality variables were entered in the model in a stepwise fashion. Subsequently the model was extended with fragmentation variables to test whether these variables contribute significantly to the regression model in addition to the quality variables (Van Apeldoorn et al. 1992; Fahrig et al. 1995; Vos & Chardon 1998).

### Analysis of dispersal data

Individuals were captured in the ponds and their direct surroundings from 1981 to 1989. During capture activities all sighted frogs were captured and marked individually (Stumpel 1987a). Time between captures, the transition period, was recorded and the sex (male, female or juvenile) was registered. In the period 1981-1986 all ponds were visited at least two times, but especially ponds with large populations were visited more than ten times (Stumpel 1987a). In the period 1987-1989, searches were incidental. A disperser was defined as an animal that was recaptured in a pond (immigration pond) other than the pond where it was first captured (emigration pond). An exception was made for animals that were recaptured in a different pond but had returned to their original pond in a next observation. This was interpreted to be exploratory behaviour rather than dispersal. Individuals that were captured more than once and had not moved to another pond were defined as non-dispersers or residents. To determine if conspecific attraction played a role in pond choice, immigration ponds were categorised depending on the

probability that a pond was actually occupied when a disperser arrived. Immigration ponds were regarded 'occupied' when during the first observation of the dispersed animal also other individuals were present and more than five animals were recorded during the transition period. If that period was one day, only the number of animals during the first observation was taken into account. Ponds were regarded 'unoccupied' if the disperser was the only individual present during the first observation and less than five observations were done during the transition period. All other ponds were categorised 'probably occupied'. These ponds could have been occupied during first arrival but if so only by a small population.

The possible role of conspecific attraction was analysed with an ordinal regression analysis, using the proportional-odds model (McCullagh & Nelder 1989). The category of occupancy of the immigration pond being the response variable and dispersal distance, transition period and sex as possible explanatory variables. Possible difference in dispersal probability between males, females and juveniles was analysed with logistic regression. To correct for differences in (re)capture probability, especially calling males will have a higher probability to get caught, only animals that were captured more than once were taken into account.

### Metapopulation viability

The incidence function model with rescue effect (Hanski 1994) was used to model the metapopulation dynamics. In this model, the occupancy of each patch (here called pond) is modelled by a discrete Markov chain with two states, occupied or vacant. The matrix of transition probabilities of this chain is determined by extinction and colonisation probability functions and varies in space (among ponds) and in time (among years). In the original incidence function model (Hanski 1994) the extinction probability ( $E$ ) is a function of pond area ( $A$ ) only, namely

$$E = \min(e A^{-x}, 1) \quad (2)$$

while the colonisation probability ( $C$ ) depends only on the connectivity ( $S$ ) of the pond by the function

$$C = 1 / (1 + y / S^z) \quad (3)$$

where  $e$ ,  $x$ ,  $y$  and  $z$  are unknowns. Our logistic regression of the extinction and colonisation events suggested however that the extinction and colonisation also depended on two habitat quality variables (Vos & Stumpel 1996; this article). Extinction was related to water conductivity (Water-Cond,  $H_1$ ), and colonisation to percentage cover of the water vegetation of the pond (Cover,  $H_2$ ). *Wink* can be extended to include these habitat variables by assuming that they modify the area and the connectivity that are effective in determining  $E$  and  $C$ . Let  $A_{\text{eff}} = AH_1^{q_1}$  and  $S_{\text{eff}} = SH_2^{q_2}$ , where  $q_1$  and  $q_2$  are additional unknown constants. On inserting  $A_{\text{eff}}$  and  $S_{\text{eff}}$  for  $A$  and  $S$  in (2) and (3) and reparametrising to  $q_1 = -x H_1$  and  $q_2 = z H_2$ , we obtain the extended incidence model

$$E = \min(e A^{-x} H_1^{q_1}, 1) \quad (4)$$

and

$$C = 1 / (1 + y / S^2 H_2 q_2) \quad (5)$$

where  $e$ ,  $x$ ,  $y$ ,  $z$ ,  $q_1$  and  $q_2$  are unknown parameters that need to be estimated from data. With the rescue effect, immigrants have increased the local population size, which is assumed to decrease the probability of extinction from  $E$  to  $E(1-C)$ . The resulting incidence probability is (Hanski 1994)

$$J = 1 / (1 + E ((1-C)/C)) \quad (6)$$

The unknown parameters of the extended incidence function model were estimated from the 1981-1983 data using a new method that extracts information from both the spatial occupancy pattern as well as the turnover events (colonisations and extinctions) between years. Our new method is based on the observation in Appendix B of Ter Braak et al. (1998) that the full likelihood of the data can be divided in three parts, namely

- 1 a spatial part, consisting of the occupancy pattern in the first year (1981),
- 2 an extinction part, consisting of the potential and realised extinctions in subsequent years (1981-1982 and 1982-1983) and
- 3 a colonisation part, consisting of the potential and realized colonisations in subsequent years (1981-1982 and 1982-1983).

Previous approaches used either only the spatial part (Hanski 1994) or the extinction and colonisation parts (Verboom et al. 1991a; Sjögren-Gulve & Ray 1996). For convenience, in all models of turnover events logistic regression was applied. Thus implicitly a logistic function for the extinction probability was used instead of equations (2) or (4). Our method extends Hanski's (1994) pseudo-likelihood, which is based on the spatial part only, with the likelihoods of the extinction and colonisation parts, thus combining the previous approaches in a consistent manner. In accordance with Hanski (1994) a binary non-linear regression is used. The data set of this regression consists of the three parts. The spatial data set of presence/absence of the ponds in the first year (1981) is modelled by  $J$ , equation (6). The extinction data set with potential and realised extinctions in subsequent years (1981-1982 and 1982-1883) is modelled by  $E(1-C)$ , as described below equation (5). Finally, the colonisation data set with potential and realised extinctions in subsequent years is modelled by  $C$ , equation (3). For the spatial data set the connectivity  $S$  of a pond is calculated from the occupancies of the remaining ponds in the same year (as in Hanski's method), whereas for the other data set the occupancies of the remaining ponds in the preceding year are used (as in Verboom et al. 1991a). The connectivity measure that we used in the incidence model calculations is  $S$ -area-barrier as described earlier, because this measure takes all elements into account that are considered to be important. The binary non-linear regression for the extended incidence function model was programmed using the procedure 'Fitnonlinear' of Genstat 5 release 3.1 (Genstat 5 Committee 1993).

To test the predictive value of *Wink*, the actual distribution pattern from 1986 and the distribution predicted by the model for year 1986 by equation (6) were

compared. To predict the viability of the metapopulation, the fraction of occupancy during the next 100 years was simulated. For this purpose, all 500 ponds in the study area were taken into account. However, the habitat quality (Water-Cond and Cover) of 60% of the ponds was unknown. Therefore, the missing values were assigned to ponds by the following method. If the values of both variables were missing, the values were randomly drawn from a bivariate lognormal distribution that was estimated from ponds without missing data (Rubin 1987; Bradley 1994). If only one of the values was missing, the missing value was drawn from a univariate lognormal distribution that was estimated from the non-missing data by regression on the other variable (Rubin 1987; Bradley 1994). Each simulation of 100 years started with the occupancy pattern from 1986. In total, 100 simulation runs were carried out.

## Results

### Extinction probability

During the 1981-1983 period ten local populations went extinct, while 31 ponds remained occupied during two successive years (Table 5.1, Fig. 5.3). Three ponds, indicated by arrows in Figure 5.3, remained occupied during the survey period and had a high leverage value (0.26) in the regression analysis of extinction probability (Table 5.2a). In regression models including these three ponds the extinction probability increased with high connectivity (S, S-area, S-area-barrier, Table 5.2a). The three ponds were already occupied in 1977 long before the survey period (Burny 1976). However, because of no available sources in the present landscape, the connectivity of these ponds is zero. This may be explained by a time lag effect in which the occupancy is no longer correlated with the present landscape but to a landscape in the past (Tilman et al. 1994). Therefore, it was decided to remove the three ponds from further analysis (Table 5.2b). In the first step of the logistic regression analysis, 'year of survey' and the habitat quality variables were entered to the model. Only water conductivity added significantly to the model predicting a higher extinction probability with increasing conductivity (Water-Cond, Table 5.2b). In the second step, fragmentation variables were added to the model. Pond size was a significant explanatory variable, predicting a higher extinction probability in small ponds (Area, Table 5.2b). Now the positive effect of the connectivity variables on extinction probability had disappeared and the connectivity variables no longer contributed significantly to the model (S, S-area, S-area-barrier, Table 5.2b).

### Colonisation probability

There were 16 ponds colonised, while 336 ponds remained unoccupied during two successive years (Table 5.1, Fig. 5.3). In the first step of regression analysis, 'year of survey' and percentage of water vegetation cover contributed significantly to the model (Year, Cover, Table 5.2c). The colonisation probability was higher from year 1982 to 1983 than from 1981 to 1982. A higher vegetation cover in the pond increased the colonisation probability. In the second step, the fragmentation

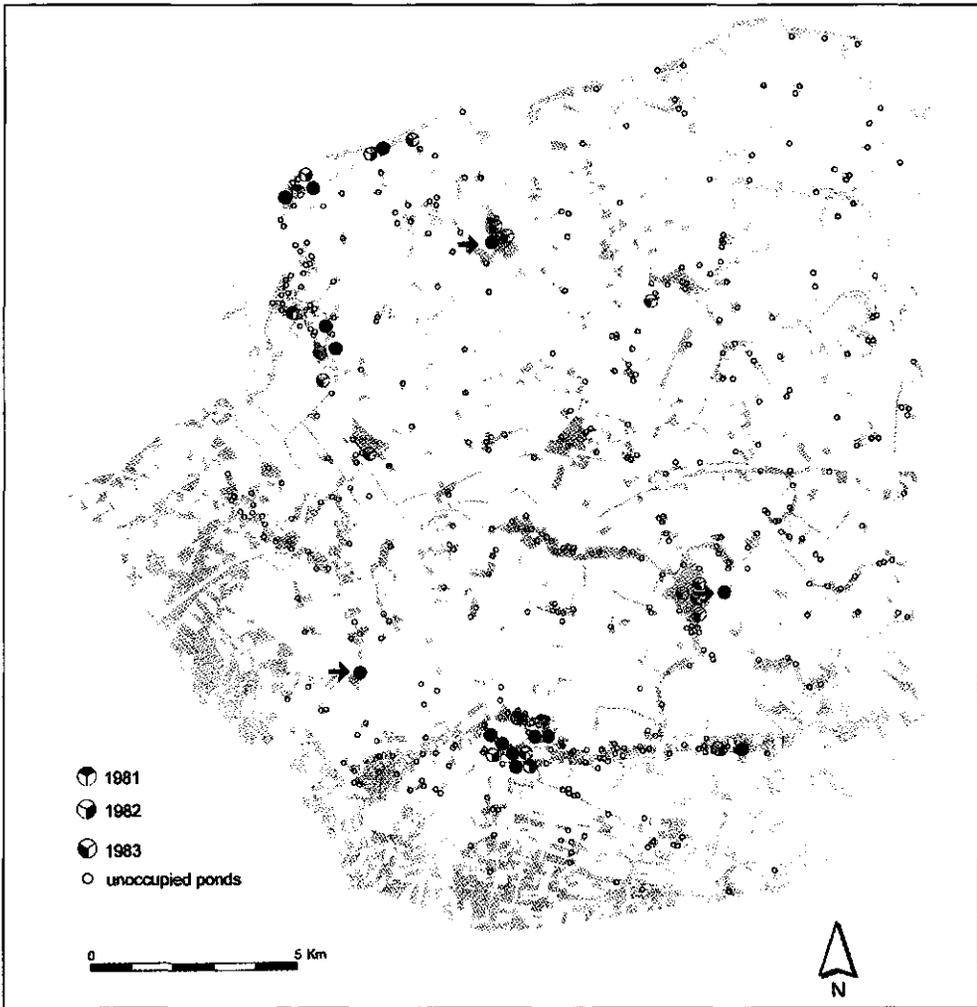


Figure 5.3

Presence and absence of tree frogs in Zeeland Flanders during 1981-1983. An extinction of a local population was recorded when a pond was occupied in year  $t$  and unoccupied in year  $t+1$ . A colonisation is recorded when a pond was unoccupied in year  $t$  and occupied in year  $t+1$ . The arrows indicate three ponds that were removed from the extinction analysis.

variables were added to the model. Pond size added significantly to the model, predicting increasing colonisation probability for larger ponds (Area, Table 5.2c). All connectivity variables contributed significantly to the model, predicting a higher colonisation probability with increasing connectivity (S, S-area, S-area-barrier, Table 5.2c). Judged on the deviance statistic there was not much difference in explanatory power between the connectivity variables (deviance values were 7.56, 6.21 and 6.52 respectively, Table 5.2c). The most complex connectivity variable, which includes

**Table 5.2**  
Logistic regression models of extinction and colonisation probability.

| MODEL CONSTANT                                   | YEAR                          | COVER                            | WATERCOND                      | AREA                           | CONNECTIVITY (S) WITH AREA (S-AREA) | CONNECTIVITY WITH AREA AND BARRIERS (S-AREA-BARRIER) |
|--|-------------------------------|----------------------------------|--------------------------------|--------------------------------|-------------------------------------|--|
| <b>2a. Extinction analysis</b>                   |                               |                                  |                                |                                |                                     |  |
| -2.79 ± 1.11                                     | ns                            | ns                               | 0.028 ± 0.015<br>(dev 3.13 ns) | -1.476 ± 0.644<br>(dev 4.04 *) | 0.554 ± 0.330<br>(dev 3.95 *)       | -  |
| -2.78 ± 1.11                                     | ns                            | ns                               | 0.027 ± 0.015<br>(dev 3.13 ns) | -1.587 ± 0.677<br>(dev 4.04 *) | 0.480 ± 0.283<br>(dev 4.08 *)       | -  |
| -2.59 ± 1.09                                     | ns                            | ns                               | 0.027 ± 0.015<br>(dev 3.13 ns) | -1.613 ± 0.685<br>(dev 4.04 *) | -                                   | 0.461 ± 0.268<br>(dev 4.12 *)                        |
| <b>2b. Extinction analysis minus three ponds</b> |                               |                                  |                                |                                |                                     |  |
| -2.68 ± 1.15                                     | ns                            | ns                               | 0.033 ± 0.017<br>(dev 4.02 *)  | -1.312 ± 0.643<br>(dev 6.32 *) | -                                   | -  |
| -2.68 ± 1.16                                     | ns                            | ns                               | 0.036 ± 0.017<br>(dev 4.02 *)  | -1.265 ± 0.672<br>(dev 6.32 *) | ns                                  | -  |
| -2.73 ± 1.18                                     | ns                            | ns                               | 0.033 ± 0.017<br>(dev 4.02 *)  | -1.311 ± 0.676<br>(dev 6.32 *) | -                                   | ns   |
| <b>2c. Colonisation analysis</b>                 |                               |                                  |                                |                                |                                     |  |
| -123.3 ± 52.4                                    | 0.016 ± 0.006<br>(dev 5.35 *) | 0.020 ± 0.007<br>(dev 13.93 ***) | ns                             | 0.335 ± 0.237<br>(dev 8.51 **) | 0.476 ± 0.178<br>(dev 7.56 **)      | -  |
| -130.2 ± 52.3                                    | 0.015 ± 0.006<br>(dev 5.35 *) | 0.020 ± 0.007<br>(dev 13.93 ***) | ns                             | 0.328 ± 0.242<br>(dev 8.51 **) | -                                   | 0.352 ± 0.146<br>(dev 6.21 *)                        |
| -128.9 ± 52.5                                    | 0.015 ± 0.006<br>(dev 5.35 *) | 0.020 ± 0.007<br>(dev 13.93 ***) | ns                             | 0.288 ± 0.247<br>(dev 8.51 **) | -                                   | 0.373 ± 0.153<br>(dev 6.52 *)                        |

In the extinction analysis 'minus three ponds' three ponds were removed from the data (see text for explanation). Estimate and standard error are indicated. In brackets: level of significance based on the deviance statistic (dev) \* p < 0.05; \*\* p < 0.01; \*\*\* p < 0.001, ns not significant. Not significant variables were not incorporated in the total model; in each model one connectivity variable is incorporated.

both the area of sources and the barrier effect of roads on connectivity (S-area-barrier, Table 2c), gave no better explanation of colonisation events.

### Dispersal

In total 89 dispersal events were registered involving 79 different tree frogs. Mean dispersal distance between ponds was 1469 m with a maximum distance of 12 570 m. Figure 5.4a gives an overview of the observed dispersal events. 2878 residents were registered involving 858 different tree frogs that were recaptured in the same pond. Thus 9% from the recaptured individuals dispersed successfully to another pond. Logistic regression analysis showed that males had a significantly higher probability to disperse ( $p < 0.001$ ), whereas females had the lowest probability, and juveniles took an intermediate position.

The analysis of the possible role of conspecific attraction on the choice of immigration ponds showed the following results. Most registered movements were directed towards occupied ponds (49) and probably occupied ponds (34), while only six movements were made towards probably empty ponds (Fig. 5.4b). In the study area approximately 500 potential immigration ponds were available of which only 10% were occupied at least once during the period 1981-1989, many empty ponds being available at close ranges (Fig. 5.4a). The ordinal regression analysis showed that dispersal distances towards occupied ponds were significantly larger than towards empty ponds, while probably occupied ponds took an intermediate position ( $p < 0.01$ ). As is illustrated in Figure 5.4c the predicted fraction of dispersers that immigrate into an occupied pond increases with dispersal distance. Although females and juveniles never dispersed to empty ponds, this was not significant. The transition period was positively correlated with dispersal distance ( $r = 0.333$   $p < 0.01$ ) and added no further explanation to the model.

### Metapopulation viability

In Table 5.3 the parameter values of the extended incidence function model estimated from the 1981-1983 data by a non-linear binary regression are listed. In Figure 5.5 an overview is given how extinction and colonisation processes are incorporated in the model, given the pond area, pond connectivity, the water conductivity and the vegetation cover of the pond. Figure 5.5a illustrates how the colonisation probability of a pond increases with increasing connectivity and how this relation is modified by the level of vegetation cover of the pond. The decrease of the extinction probability of a pond with increasing pond size and the effect of the water conductivity value of the pond on this relation is illustrated in Figure 5.5b. The extinction probability decreases slightly with increasing connectivity of the pond, which is caused by the rescue effect that is built in the model (Fig. 5.5c). The predictive power of the model is tested by comparing the occupation probability predicted by the model with the actual distribution pattern in 1986 (Fig. 5.6). On the horizontal axis, the ponds are divided into classes, based on the predicted occupation probability of the model. Note that the number of observations per class differ, with 34% of the ponds in the class with the lowest predicted occupation. When comparing the predicted occupation of the ponds for each occupation class

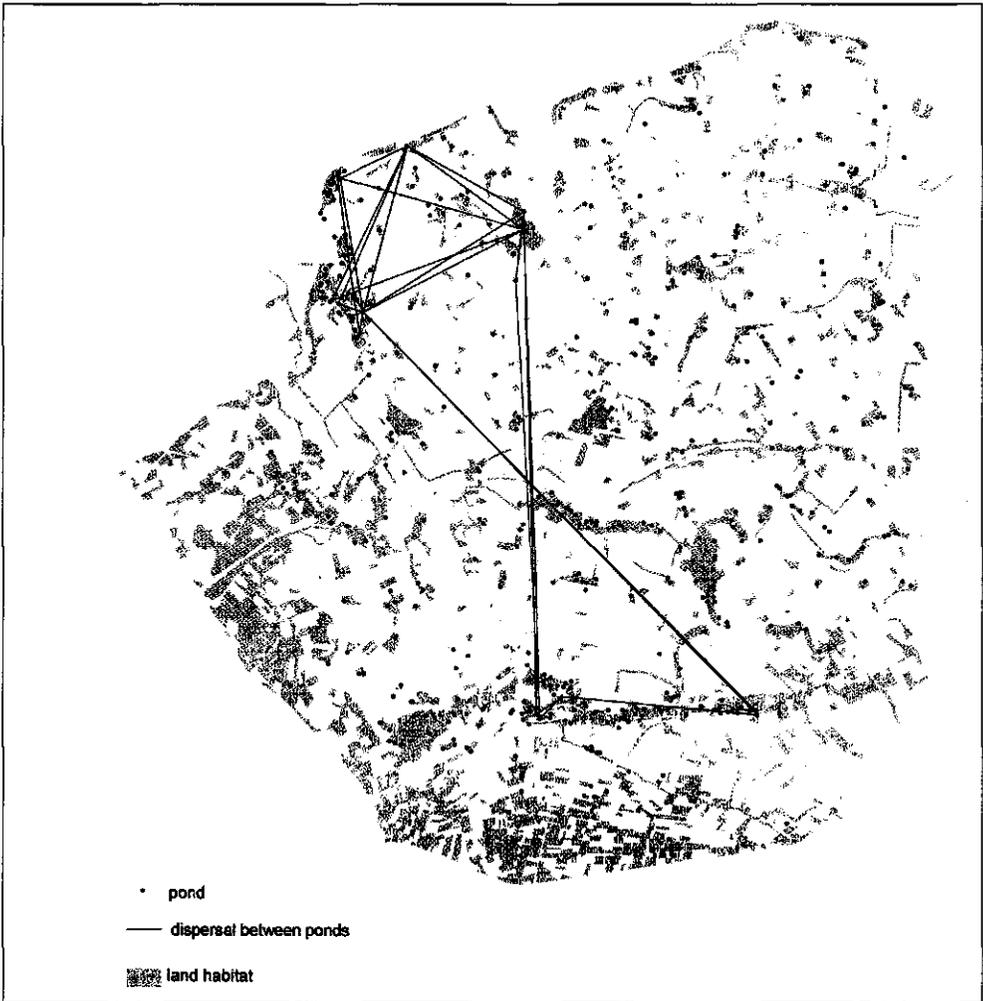


Figure 5.4a

Overview of the dispersal events registered by capture-recapture techniques in Zeeland Flanders during the period 1981-1989.

Table 5.3

Parameter estimates and standard error (s.e.) of the extended incidence function model estimated from the 1981-1983 data by non-linear binary regression (see equations 4 and 5 for explanation of the model parameters).

| MODEL PARAMETERS | ESTIMATE | S.E.   |
|------------------|----------|--------|
| $e$              | 0.0086   | 0.0154 |
| $z$              | 0.338    | 0.118  |
| $x$              | 0.429    | 0.227  |
| $q_1$            | 0.895    | 0.412  |
| $q_2$            | 0.398    | 0.157  |

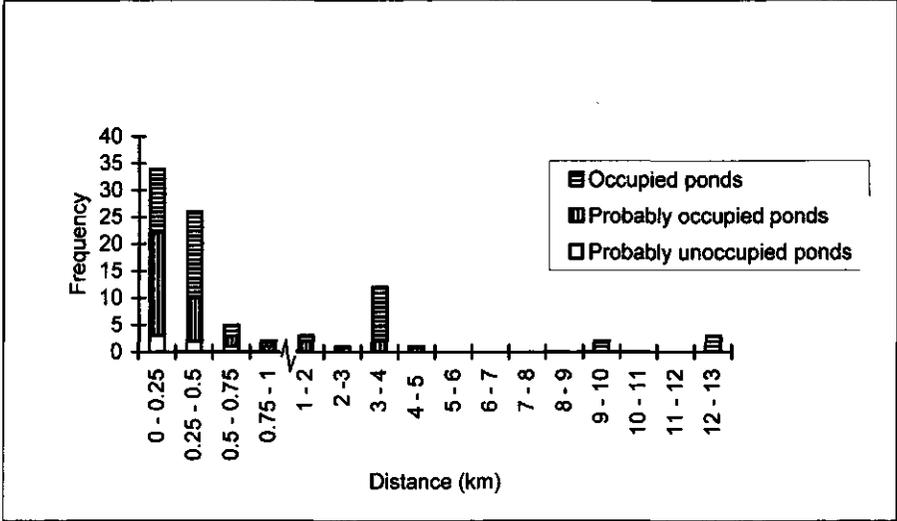


Figure 5.4b  
 Frequency distribution of dispersal distances. The shading indicates the immigration pond type: occupied ponds, probably occupied ponds and probably unoccupied ponds.

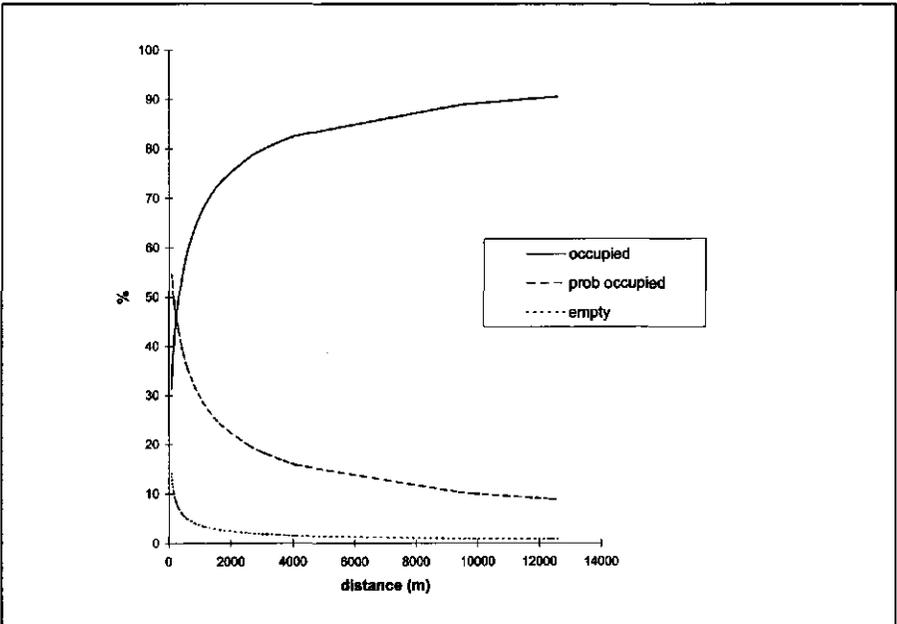


Figure 5.4c  
 Predicted fractions of pond types in relation to dispersal distance of ponds that received an immigrant, which were already occupied, probably occupied or unoccupied, when the immigrant arrived.

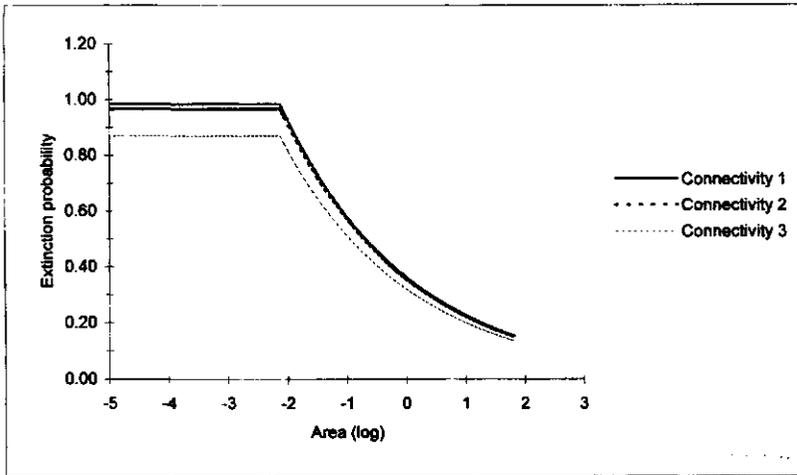
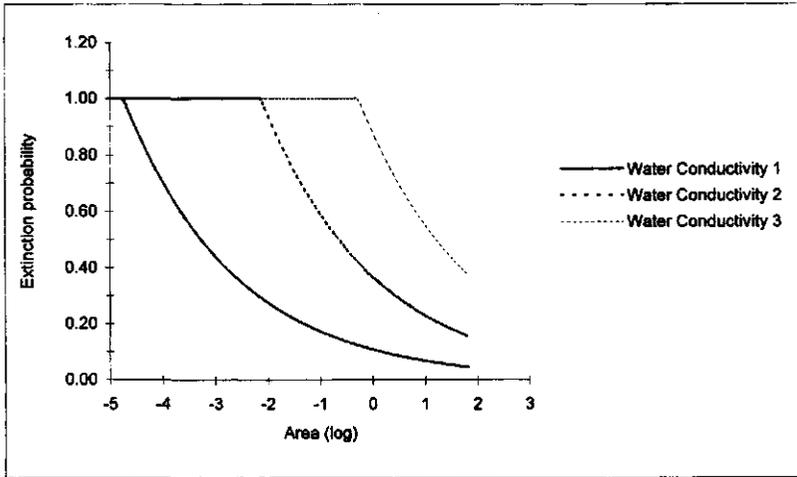
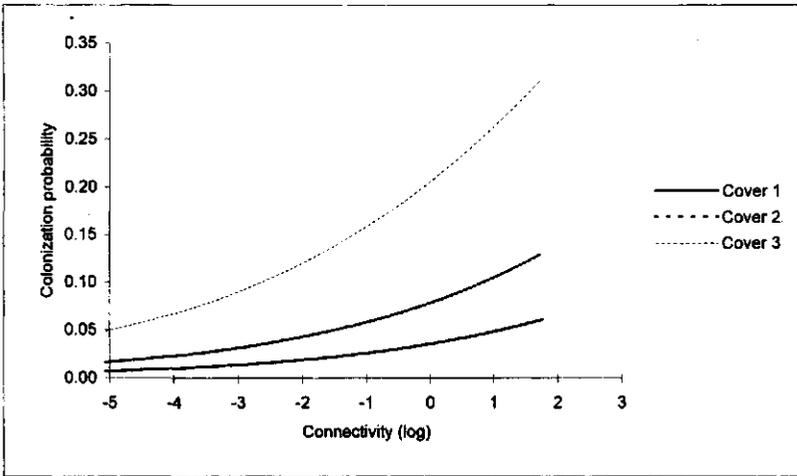


Figure 5.5

The extended incidence function model.

5.5a

Relation between colonisation probability and connectivity with minimum (1), mean (2) and maximum (3) levels of vegetation cover of the pond.

5.5b

Relation between extinction probability and patch area with minimum (1), mean (2) and maximum (3) values of the water conductivity of the pond.

5.5c

Relation between extinction probability and patch area for patches with minimum (1), mean (2) and maximum (3) connectivity.

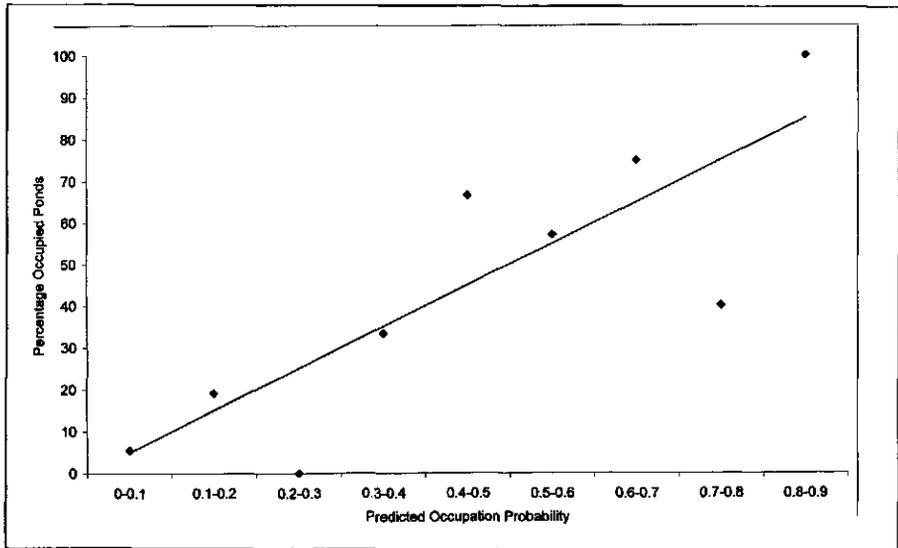


Figure 5.6

Comparison of the actual occupation of the tree frog in Zeeland Flanders in 1986 and the occupation probability of a pond predicted by the model. The predicted occupation probability is divided into classes. For each class the percentage of actual occupied patches based on the distribution pattern of 1986 is given. The solid line indicates a 100% fit between predicted and observed occupation pattern.

with the percentage of ponds that were actually occupied in 1986, a clear relation between predicted and actual occupation emerges. The solid line indicates a 100% fit between predicted and observed occupation.

To test the viability of the metapopulation the predicted fraction of occupation during the next 100 years was simulated in 100 runs. The mean number of occupied ponds increased in the first few years from 29 ponds, the number of occupied ponds in year 1986, to 36 ponds and stayed stable between 35 and 40 ponds until year 100. The metapopulation never went extinct during the 100 years of simulation and can therefore be regarded viable as the extinction probability is below the extinction threshold of 5% for the next 100 years.

## Discussion

In the introduction four questions were posed that ideally should be answered to infer from empirical data that a habitat network in a landscape supports a (viable) metapopulation. The results seem to justify the conclusion that the tree frog in Zeeland Flanders does function as a metapopulation. Extinctions take place regularly and are influenced by spatial features of the landscape: extinction probability decreases with patch size. Empty patches are recolonised and this process is influenced by the configuration of habitat, with increasing colonisation probability for large patches with high connectivity. The observed dispersal distances in comparison with the distances between patches and the observed recolonisations show that the habitat network is still connected by dispersing individuals. These results seem to justify a landscape or regional approach in conservation planning and the application of metapopulation concepts for optimal spatial habitat configuration. Finally the fourth question if the metapopulation is still above its extinction threshold should be answered positively. Simulations with *Wink* predict that the metapopulation is still viable, at least in the landscape of the eighties. However, this optimistic prediction should be handled with some reservation as will be discussed below.

### Is local extinction related to landscape structure?

Pond size, encompassing both pond area and suitable terrestrial habitat in a radius of 250 m around the pond, was negatively correlated with the extinction probability of a local population. This is in accordance with the metapopulation theory, if one assumes that small patches can sustain only small populations that can go extinct by chance processes of demographic stochasticity (Levins 1970; Richter-Dyn & Goel 1972). Generally the size of the reproduction site is thought to be a limiting factor in amphibian populations (Wilbur 1987; John-Alder & Morin 1990). However, in an earlier study (Vos & Stumpel 1996) patch size described by pond area only did not explain occupation probability. The extension of pond area with terrestrial habitat is likely to be a better approximation of patch size, since suitable terrestrial habitat is scarce and often spatially separated from the reproduction site by unfavourable habitat types.

In many studies connectivity is found to be inversely related to extinction probability (Stacey et al. 1997), this is often attributed to the rescue effect (Brown & Kodric-Brown 1977). We found no significant effect of connectivity on the extinction probability. On the other hand, the dispersal data indicated a preference of the dispersers for occupied ponds. In a tree frog population in the south of Sweden, a rescue effect was found. The extinction probability decreased with the number of calling males in a radius of 500 m surrounding a pond (Edenhamn 1996). However, the population in Sweden is less fragmented and much larger with an occupied fraction of 0.30 of a total of 1500 ponds compared with an occupied fraction of 0.1 of a total of 500 ponds in Zeeland Flanders (see for further comparison Vos & Chardon 1998). In Zeeland Flanders the number of immigrants might be too small to lower extinction probability. We do not believe that the absence of a rescue effect can be attributed to the choice of radius of 2 km for the calculation of the connectivity

measures. Although larger dispersal distances were observed, the large majority of recorded dispersal distances was well below 2 km (Fig. 5.4b).

### Is recolonisation related to landscape structure?

The colonisation probability of a pond increased with connectivity. This is in accordance with the metapopulation theory, where a higher number of potential sources of dispersers will increase colonisation probability (Verboom et al. 1991a; Hanski 1994). There was no difference in level of significance between the three connectivity measures, based on the deviance statistic. Theoretically it is expected that a connectivity measure, which takes an area component of the potential dispersal source into account, as well as a reduction of this source when roads have to be crossed, would give the most accurate estimation of connectivity. A negative effect of roads on the density and occupation probability of ponds has been demonstrated for amphibians (Fahrig et al. 1995; Vos & Chardon 1998). However, a barrier effect of roads is only one of the aspects of the relative resistance of the landscape between ponds. For instance the presence of corridors was not taken into account. Landscape elements were claimed to facilitate (dispersal) movements for several species, among which amphibians (Forman & Godron 1986; Merriam 1991; Saunders & Hobbs 1991; Bennett et al. 1994; Wiens 1997; Vos & Chardon 1997). Although the results clearly indicate that the presence of source ponds within a radius of 2 km is an important factor explaining colonisation probability, these results do not underpin a choice for complexity of the connectivity measure. Still in *Wink* we used the connectivity measure that corrects for both source size and barriers, as we think these factors do influence dispersal success.

A positive effect of pond area and of the vegetation cover of the pond on the colonisation probability was found. We interpret this as an indication that the larger the patch, the higher the probability that the patch is detected by the dispersing animal. As the area of suitable terrestrial habitat in a radius of 250 m surrounding the pond is an important component of the total size, it is plausible that dispersal to the pond is facilitated by these landscape elements as well (see also Vos & Stumpel 1996). But this does not explain why also the vegetation cover of the pond, a habitat quality variable, increased the colonisation probability. If one assumes that better habitat quality will improve reproductive success, it will increase population establishment probabilities of a starting population. However, it is unlikely that such differences already occur in the first season after colonisation. Increasing colonisation probability with pond size and vegetation cover could also imply that dispersers choose to stay only when circumstances seem unfavourable and otherwise move on. This is supported by the recapture data where individuals cross large distances, even though many ponds are available at much shorter ranges (Fig. 5.4a).

In the analysis every pond was regarded as a separate potential site for a local population. This simplification seems justified, as was pointed out in Vos and Stumpel (1996) by the fact that even in clusters of ponds that are within 100 m of each other, in two-thirds of these clusters only one pond was occupied during the survey period. The delimitation of local populations and hence local extinction and recolonisation are to some extent arbitrary (Harrison & Taylor 1997). Not every

presence of frogs in a pond that was unoccupied in the previous year will be a real colonisation, while not every abandonment of a pond will be a real extinction. There will be a continuum between distance and independent behaviour of local populations. In an extreme case of habitat fragmentation where several fragments are used by one individual, the so-called patchy population (Harrison 1994; Harrison & Taylor 1997), presences followed by absences may be the result of foraging behaviour or responses to conspecifics. However, the fact that 90% of the recaptured individuals had remained resident at the site where they were first sighted does imply a reasonable independence between ponds. Also it is known that in some cases tree frogs were found to occupy a single pond over several decades (Stumpel 1987a). There is a possibility of misclassification of turnover events in amphibian populations and other species that have a juvenile phase that exceeds the yearly census frequency. One-year absence of individuals would erroneously be regarded as an extinction followed by a recolonisation, if juveniles were still present in the pond surroundings. However, these wrong classifications will not be correlated with landscape structure and will only weaken effects of habitat fragmentation in explaining extinction and recolonisation events.

### **Is the habitat network connected by dispersers?**

The observed dispersal distances in relation to the distances between ponds indicate that the populations are still interconnected by dispersal. The dispersal distances observed are large in comparison with the maximum distances of tree frogs mentioned by other authors: 3750 m (Clausnitzer & Clausnitzer 1984) and 5000 m (Fog 1993). The capture-recapture data show that males and juveniles have a higher probability to disperse than adult females and that the majority of displaced individuals moved to occupied or probably occupied ponds. Sjögren-Gulve (1998) also found target-oriented dispersal, directed towards occupied ponds, for the pool frog (*Rana lessonae*). The distance covered was significantly larger for animals that had moved to occupied ponds compared with empty ponds and probably occupied ponds, although many ponds were available at shorter ranges (Fig. 5.4a). This result implies that conspecific attraction plays a role in the choice of immigration pond. The probability to move on is higher when the immigration pond is empty or probably occupied, resulting in larger distances covered before a disperser finally reaches an occupied pond. Search intensity was higher in the large populations, which belonged to the category 'occupied immigration ponds'. As higher search intensity will increase the probability of finding dispersed individuals, the delimitation between fractions is probably biased towards the occupied category. However, this does not influence the result that the ratio between the three categories changes with distance.

### **Conspecific attraction in metapopulations**

In amphibians male calls play an important role to advertise their presence to reproductive females over a long range (Forrest 1994; Cocroft & Ryan 1995). However, as most displaced animals were males or juveniles (unspecified) it seems that also males are attracted to calls of conspecifics. Could this behaviour be

interpreted in relation to the metapopulation structure of the species? Several benefits for conspecific attraction have been mentioned: increased mating success, predator protection, defence against intruders and information about habitat quality (Kiestler 1979; Stamps 1988; Prins 1996). For anurans increased mating success has been suggested as a possible explanation for the evolution of reproductive grouping. An aggregation of calling males would be an advantage because: the mating signals produced by a large chorus travel over a greater distance than from a small chorus (Otte 1974; Alexander 1975); it would facilitate female mate choice (Alexander 1975; Bradbury 1981; Schwartz 1994) and less effective advertisers could exploit the display efforts of more attractive males (Arak 1983). Joining a large chorus would only increase mating success of an individual male if the number of attracted females would increase disproportionately with chorus size. There are some field observations that a large chorus attracted more females per male in *Physalaemus pustulosus* (Ryan 1985; Wagner & Sullivan 1992). But in other studies no difference was found (Tejedo 1993; Schwartz 1994). The loud calls of the tree frog, which can be heard over a distance of 1000 m (by humans) could have an orientation function for dispersing individuals, as was also suggested for the chorus frog (*Pseudacris triseriata*) and the natterjack toad (*Bufo calamita*) (Sinsch 1990). A second benefit of conspecific attraction that could play a role for tree frogs is that conspecifics are good indicators of habitat quality (Stamps 1987, 1988; Kiestler & Slatkin 1974; Shields et al. 1988; Alatalo et al. 1982). Because of natural succession, reproduction waters of amphibians will be suitable for only a limited period (Joly & Grolet 1996). In addition, the quality and location of the tree frog habitat are unpredictable because of environmental fluctuations. An important part of the natural habitat of the tree frog is situated along river flood plains (Creemers 1991). Flooding can make an area unsuitable for years but can at the same time create new habitat on another location. So the network of suitable habitat patches is dynamic both in space and time. Therefore, a species in such unpredictable habitat must have strong colonisation ability and the mechanism to find conspecifics must be well developed. The loud mating call of the tree frog could be an adaptation to the survival in unpredictable habitat, where the location of suitable reproduction sites can change between years.

What is the impact of conspecific attraction on the survival in fragmented landscapes? Conspecific attraction will decrease the extinction probability of a local population, due to the rescue effect (Brown & Kodric-Brown 1977). By consequence, empty ponds will receive fewer colonisers and will have a lower colonisation probability (Ray et al. 1991; Smith & Peacock 1990). Model simulations (Ray et al. 1991) predict a lowered level of occupation within the metapopulation even with a small attraction value. Ray et al. (1991) point out that conspecific attraction may be evolutionarily stable, but it may prove to be a quite unstable strategy in increasingly fragmented landscapes. If landscapes become too fragmented, the populations will become smaller and the probability of extinction will increase. On the other hand, dispersing animals will have to cover larger distances. As the relatively small choruses can be heard over smaller distances, the dispersal is less directed and search time will increase (Ray et al. 1991). Thus the mortality during dispersal will increase, and individuals will be lost for the metapopulation. Model simulations with a more detailed mechanistic model may indicate at which stage of fragmentation conspecific attraction becomes a handicap for a species.

### The extended incidence function model *Wink*

The fit between the predictive value of the simulation model and the actual distribution pattern of 1986 is quite high, as is illustrated in Figure 5.6. This result is an indication that the metapopulation model describes relevant processes in the spatial population dynamics of the tree frog in Zeeland Flanders. However, does it imply that the predictions of the simulation model are reliable? Some comments should be made here.

Some causes of local extinction are not taken into account, which makes the model predictions too optimistic. The extinction and colonisation probabilities are based on a three-year data set. These years are not necessarily representative years and definitely do not incorporate the total range of possible fluctuation of population numbers between years. As strong fluctuation of population size will increase extinction probability (Gilpin & Soulé 1986), predictions based on relative 'good years' might be overoptimistic. A second complication is that local extinctions are not only due to stochastic processes in small populations. Extinction may also be deterministic, due to habitat destruction and the loss of suitable habitat caused by natural succession. The turnover of especially ponds is high in agricultural landscapes. These aspects form an extra threat to extinction, which is not incorporated in the model. Longer time series (10-20 years) would certainly improve the estimation of extinction and colonisation probabilities. For species that occur in dynamic habitat networks, the incorporation of habitat succession (Lindenmayer & Possingham 1995) and changes in habitat distribution over time would improve the predictive value of incidence models.

## 6 The role of landscape heterogeneity on the movement paths of the tree frog (*Hyla arborea*) in an agricultural landscape

### Summary

When the effects of habitat fragmentation are considered, interest in landscape ecology grows for an approach in which the complete landscape mosaic is studied. Especially in highly human-dominated landscapes not only distance but also landscape resistance may have large impact on connectivity. In this paper the effect of the landscape mosaic on the movements of displaced tree frogs is studied in an agricultural landscape. The movement model *SmallSteps* is developed, to extrapolate individual movements to differences in connectivity. The movement paths of displaced tree frogs indicate a preference for hedgerows and an avoidance of arable land, while pastures take an intermediate position. The spatially explicit calibration of the model shows the best fit between observed and predicted movement paths, when transition probabilities are strongly biased towards hedgerows. It is concluded that the resistance of the landscape mosaic is an important element to determine connectivity for tree frogs and that hedgerows may function as corridors that will enhance dispersal. Prerequisites are discussed for the extrapolation of the model from an individual level on a local scale towards estimates of dispersal between populations on a regional scale.

### Introduction

A successful approach in studies on the effect of habitat fragmentation is to characterise the landscape as an archipelago of suitable habitat islands, separated by a 'sea' of inhospitable habitat, the landscape matrix (e.g. Levins 1970; Gilpin & Hanski 1991; Opdam et al. 1993; Hanski & Gilpin 1997). In these studies the colonisation probability of a patch depends on its connectivity, which is often described by a combination of the distance towards neighbouring (occupied) patches and the size of potential sources (e.g. Verboom et al. 1991a; Hanski 1994; Vos et al. submitted). In this definition of connectivity, the role of the landscape matrix in controlling the movement pattern is neglected. For ground-dwelling species with small dispersal capacity this disregard is a serious oversimplification (Forman & Godron 1986; Henein & Merriam 1990; Merriam 1991; Saunders & Hobbs 1991; Bennett et al. 1994; Wiens 1997; Bennett 1999). In landscape ecology the necessity of a landscape approach, in which not only the suitable habitat patches but also the complete mosaic of habitat types are considered, receives a growing interest (Wiens 1997; Bennett 1999). Especially in highly human dominated landscapes, where boundaries between habitat patches are sharp and the permeability of boundaries and matrix mosaic for moving organisms will vary greatly, landscape resistance cannot be neglected. In nature conservation practice, the protection of zones with

high permeability, so-called corridors or linkages, are recognised to enhance successful dispersal of organisms (e.g. NPP 1990; Bonner 1994; IUCN 1995; Jongman & Troumbis 1996; Bennett 1999; Vos et al. 1999). The corridor concept has its roots in island biogeography theory. A higher conservation value was predicted for fragments that are linked by corridors compared to isolated fragments (Diamond 1975; Wilson & Willis 1975). The corridor concept was also incorporated in metapopulation theory. Fahrig and Merriam (1985), for example, predicted that metapopulation survival increased, the more patches of the network were connected by corridors.

There are many empirical studies in which some positive effect of corridors has been demonstrated, although empirical evidence is often incomplete (Beier & Noss 1998; Bennett 1999; Vos et al. 1999). An adequate understanding of the role of corridors in species conservation requires a combination of different research approaches. A first step is to explore, by pattern studies on a landscape level, whether linear landscape elements are likely to contribute to connectivity. In many studies a positive relation has been found between the probability of occupation or colonisation of a suitable habitat patch and the density of linear elements (e.g. Pahl et al. 1988; Bright & Morris 1990; Laurance 1990; Verboom & Van Apeldoorn 1990; FitzGibbon 1993; Bright et al. 1994; Vos & Stumpel 1996; Grashof-Bokdam 1997). In several studies a higher colonisation probability or a higher frequency of visiting individuals was found for habitat patches connected by linear elements compared to unconnected patches (e.g. Dmowski & Kozakiewicz 1990; Saunders & De Rebeira 1991; Haas 1995; Dunning et al. 1995; Riffell & Gutzwiller 1996). In a second research step, detailed studies at the individual level provide quantitative information to what extent movements of individuals are influenced by the landscape mosaic and which habitat types function as corridors. For instance in several studies conducted in agricultural landscapes, preference for hedgerows and avoidance of open fields were registered (e.g. Bright & Morris 1990; Lindenmayer et al. 1994a/b). A third essential step in studying the effects of landscape resistance is the extrapolation from individual movements patterns towards differences in connectivity between local populations on a landscape level. This requires spatially explicit movement models, taking into account the composition and configuration of the landscape matrix. These models obtain their empirical underpinning from individual-level studies of movement on microscale, to make predictions on connectivity on the larger landscape scale (Vermeulen 1995; Tischendorf & Wissel 1997).

In this article we investigate whether and how the landscape mosaic influences the movement paths of tree frogs in an agricultural landscape. We assume that landscape elements which function as relative corridors, enhance the dispersal between habitat patches and thus will increase connectivity within the population network. In an earlier study on the distribution pattern of the tree frog (Vos & Stumpel 1996) a positive correlation was found between occupation probability of a pond and the amount of suitable terrestrial habitat in a radius of 1000 m surrounding the pond. The terrestrial habitat mainly consisted of linear hedgerows and dikes with rough growth vegetation in a matrix of agricultural fields. It was assumed that the amount of suitable terrestrial habitat would increase the survival probability of a local population but also would improve the connectivity.

In this study we explore the second and third steps, mentioned above, to quantify the influence of the landscape mosaic on connectivity. The study consists of a field experiment to register individual movements and a modelling part in which the individual movements are extrapolated to differences in connectivity. The following questions are addressed:

- 1 Are there differences in speed and directness of movement in different habitat types, comparing hedgerows, arable land and pastures?
- 2 Does the tree frog show preference for or avoidance of particular habitat types when a boundary between habitat types is encountered? We have registered the movement paths of radio-tracked tree frogs in displacement experiments in an agricultural landscape. Dispersal patterns may differ from movement paths within the home range (e.g. Merriam & Lanoue 1990; Rufenacht & Knight 1995; Mauritzen et al. 1999). By displacing individuals into various landscape configurations that resemble the landscape matrix, the behaviour of dispersers in unfamiliar landscapes is simulated.
- 3 If movement paths are influenced by the configuration of different habitat types, how would this affect connectivity? We have explored how the impact of the landscape matrix on the movement paths would result in an uneven distribution of animals. We developed a movement model *SmallSteps* to extrapolate observed individual movements to differences in connectivity within the studied landscape. The movement model is vector-based and spatially explicit. Some parameters are directly estimated from the radio-tracking observations. Other parameters are tested with a new calibration method, based on the observed movement paths and the configuration of habitat types.

## Radio-tracking experiment

### Methods

The study was conducted in 1994, 1995 and 1997 from May to July in an agricultural landscape near the 'Vildersveen' in the province of Gelderland (Fig. 6.1). The region mainly consists of arable land, pastures and hedgerows, often accompanied by ditches. Some swamps and woodlots are present as remnants of the former natural vegetation. The habitat of the tree frog is composed of small swamps (the aquatic reproduction habitat) with adjacent shrubs, bushes and rough growth (the terrestrial habitat part). Also cattle drinking ponds that are situated in pastures in combination with hedgerows and rough growth are suitable habitats (Stumpel 1987a; Vos & Stumpel 1996; Vos et al. submitted). Suitable habitat patches are separated by a matrix of intensively managed agricultural fields. Hedgerows are expected to facilitate dispersal through these agricultural fields.

The Vildersveen, which is occupied by a population of tree frogs, consists of a reproduction water and adjacent terrestrial habitat: hedgerows, shrubs in forest edges and rough growth vegetation (Fig. 6.1). To study movements patterns in arable land, pastures and hedgerows, tree frogs were released at five locations in the agricultural landscape near the Vildersveen. Locations 1 and 3 were situated in

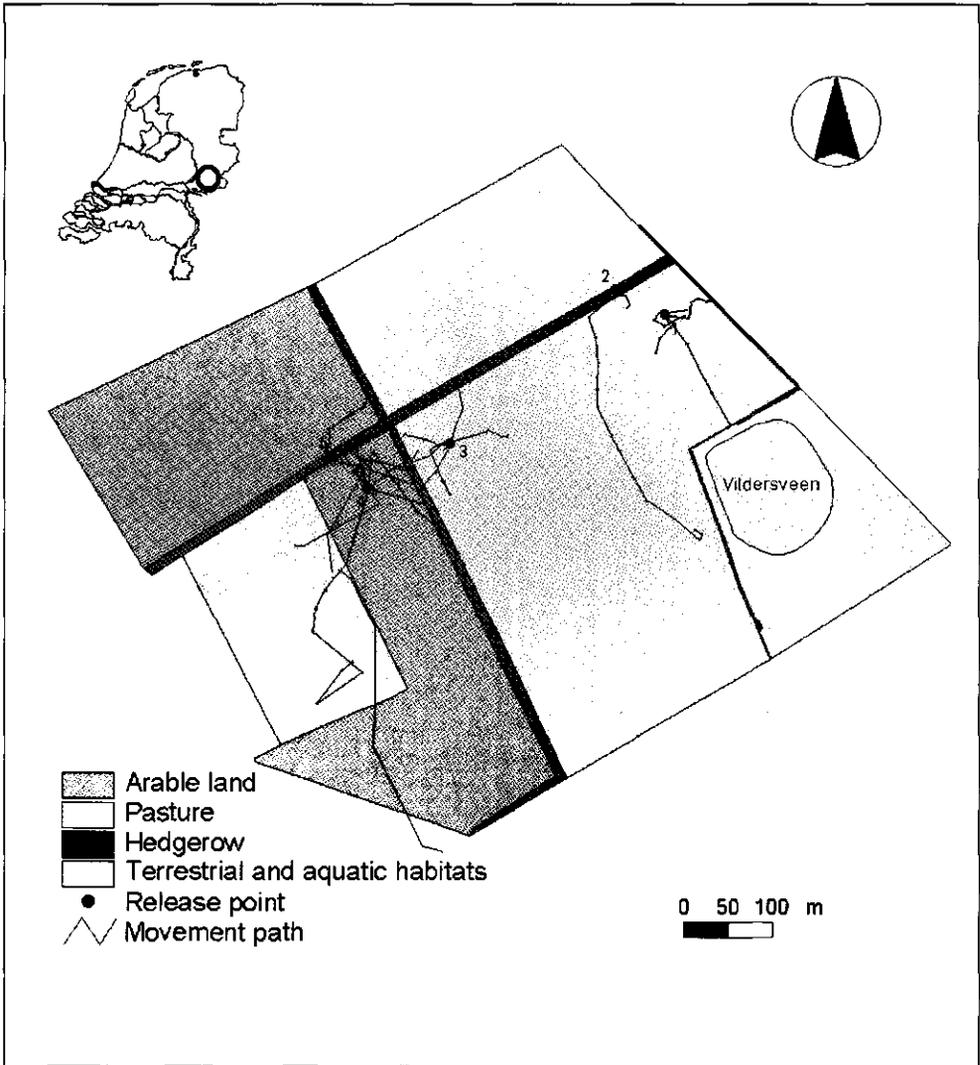


Figure 6.1

The study area in the vicinity of the Vildersveen. The five release locations of the radio-tracking experiment and the observed movement patterns are indicated.

pasture at 50 m distance from two hedgerows. Location 4 was situated in arable land, also 50 m from two hedgerows. Locations 2 and 5 were in a hedgerow, 75 m from a hedgerow junction and surrounded by pasture (2) or arable land (5). The distance to the Vildersveen from the release locations varied from 240 to 520 m. As suitable terrestrial habitat is abundant adjacent to the reproduction water in the Vildersveen, it was assumed that the landscape in the vicinity of the release locations was not familiar to the frogs. The animals were radio-tracked with transmitters of 0.72 g (Holohil systems Ltd, type BD-2Ard) that were glued onto a cotton waist band. This method was already successfully applied on tree frogs in Sweden (pers. comm.

Edenhamn). Hand-held dipole antennas in combination with a small receiver (type YEASU FT 290rll) were used. This receiver had a variable resistance for assessing the exact location of the radio signal at close ranges.

To test whether transmitters hindered the animals, individuals were observed in an enclosure. They appeared able to climb and move around freely, showing comparable behaviour to animals without transmitter. For instance one radio-tracked individual jumped to a height of 40 cm, which is considered high for a tree frog. To determine the activity period three tree frogs were followed continuously for several days. As the frogs never moved during daylight, subsequent observations started at sunset (21.00 hrs) and terminated at dawn (6.00 hrs). Each animal was tracked in two successive nights. To avoid double participation of the same individual in the experiment, each animal was photographed before it was released. Tree frogs can be identified by their unique lateral black lines (Tester 1990).

Adult males were collected from the Vildersveen, weight and length were determined and the transmitter was adjusted. The animals were transported separately to the release sites in a blinded aquarium. At each site one animal was released, so a maximum of five animals participated in the experiment simultaneously. To avoid influencing the initial direction of movement the aquarium was put on the release site and the animals had to climb out. The animals were tracked every hour and a pole with reflecting tape was put in the ground a few metres from the estimated position of the frog. The position of the poles was measured during daylight, using a 10-m grid along the field fences as X and Y axes. The (Amersfoort) co-ordinates were calculated to enable further analysis in GIS.

For all animals the total distance covered during the experiment and the relative time spent per habitat type were measured. The distance covered between observations (step size) was converted into velocity (m/h) and the landcover type was registered. The change in direction between observations (turning angle) was calculated ( $\pm 180^\circ$ ), assuming that the animal had moved in a straight line between observations. Step size and turning angle per habitat type were used to simulate the movement of individuals in the movement model *SmallSteps*.

The probability to cross a boundary between habitat types was estimated in two ways. In the first approach, the transition probability between habitat types was estimated by counting the times that a transition actually occurred, taking into account all observations in which a transition was possible. For example, in Figure 6.2, the probability to move from habitat type A to B would be 0.5, based on movements II and III. In the second approach, all transition probabilities are derived from hedgerow observations. As animals in linear habitat elements are on a boundary between habitat types permanently, they have a probability to cross a boundary regardless of the moving distance. Transitions between all habitat types can be derived from hedgerow observations, by assuming that boundary reactions between habitat types are symmetrical. The probability  $q_2$  to stay in type A is similar to the probability  $p_2$  to move from type B to A (Fig. 6.3).

The experiment was conducted during the reproductive season, in the vicinity of an occupied patch, the Vildersveen. It was expected that the animals would be motivated to move during the reproduction period. As there are indications that the dispersal of tree frogs is directed towards conspecifics (Vos et al. submitted), it was tested if the released animals preferably moved towards the

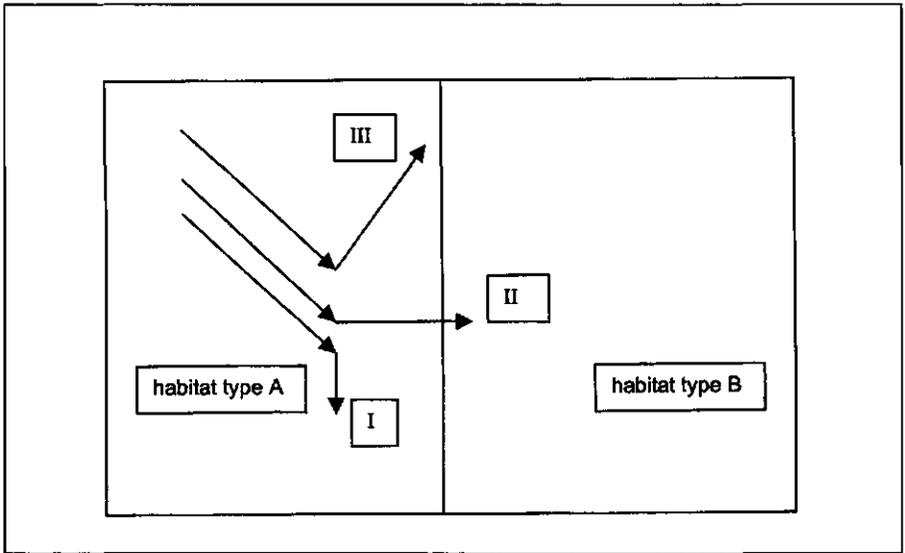


Figure 6.2  
 First approach to estimate transition probabilities between habitat types. The transition probability is estimated by counting the times that a transition actually occurred, taking into account observations in which a transition is possible.

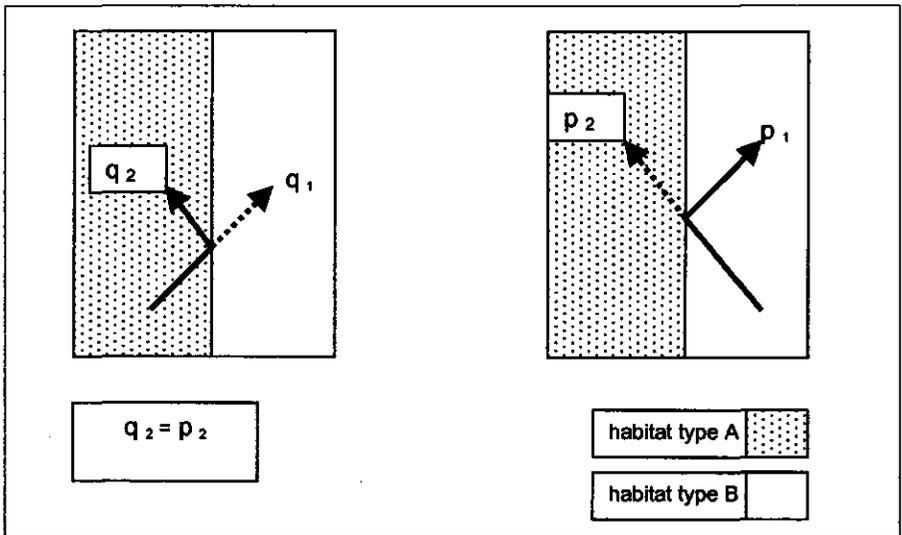


Figure 6.3  
 Second approach to estimate transition probabilities between habitat types. All transition probabilities are derived from hedgerow observations, assuming that boundary reactions between habitat types are symmetrical.

Vildersveen. Therefore, the observed movement paths were also analysed as directed walk, by calculating differences between the observed direction of movement and direct movement towards the Vildersveen. To get an impression of behaviour in an area far away from a reproduction pond, in 1997 some animals were released in the 'Kolkman'. This second study area is situated in the same region as the Vildersveen but has no occupied reproduction sites within a radius of 3 km. The composition of the habitat types and the position of the release locations is similar to the Vildersveen site.

## Results

In total 46 animals were radio-tracked in the Vildersveen area: 5 animals in 1994, 21 animals in 1995 and 20 animals in 1997. The mean weight of the animals was  $5.9 \text{ g} \pm 1.0$  and the mean length was  $42 \text{ mm} \pm 3$ . Three trials could not be used for analysis because the movement was hampered by the transmitter that had moved to a belly position, or animals had lost their transmitter, ten trials did not provide relevant information as the animals had not moved during the experiment, leaving 33 movement paths to be analysed. As is shown in Figure 6.1 variation in covered distances is large. When the distances are corrected for the total time spent in the field, mean distance covered during the experiment is  $223 \text{ m} \pm 138$ .

Mean velocity on arable land was highest ( $36.9 \text{ m} \pm 28.9$ ), compared to pasture ( $14.3 \text{ m} \pm 15.7$ ; Table 6.1). The highest speed was observed on arable land, where one individual moved 120 m in one hour. On arable land animals had moved in 93% of the cases between observations, as compared to 73% on pasture (Table 6.1). Movements in hedgerows were only small (mean  $4.5 \text{ m} \pm 8.3$ ), while in 33% of the observations animals had moved in hedgerows. Leaving out the non-movements, differences in speed between habitat types were still considerable:  $39.5 \text{ m} \pm 28.9$ ,  $19.7 \text{ m} \pm 15.2$  and  $13.5 \text{ m} \pm 9.3$  for arable land, pasture, and hedgerow respectively (Table 6.1).

Table 6.1

Step size (m/h) and turning angle ( $\phi$ ), the change in direction between observations, in pasture, arable land and hedgerows, based on 33 analysed movement paths. Mean values and standard deviation (sd) are indicated.

| HABITAT TYPE | STEP SIZE       |  | TURNING ANGLE  |
|--------------|-----------------|--|--|
|              | Velocity (m/h)  | mean $\pm$ sd                              | Angle ( $\phi$ ) mean $\pm$ sd                             |
| Pasture      | $14.3 \pm 15.7$ | (n = 109)<br>( $19.7 \pm 15.2$ ; n = 79) * | $8.17 \pm 59.34$ (n = 75)                                  |
| Arable land  | $36.9 \pm 28.9$ | (n = 30)<br>( $39.5 \pm 28.9$ ; n = 28) *  | $4.24 \pm 76.50$ (n = 21)                                  |
| Hedgerow     | $4.5 \pm 8.3$   | (n = 66)<br>( $13.5 \pm 9.3$ ; n = 22) *   | $P_{\text{forward}} 0.28$<br>$P_{\text{turn around}} 0.02$ |

\*excl. non-moving observations

The change of direction between observations in pasture ( $8.17 \pm 59.34$ ) or arable land ( $4.24 \pm 76.50$ ; Table 6.1), the turning angle, did not differ significantly (Wilcoxon signed-ranks test, at  $p < 0.05$ ). Lateral movements within the hedgerow fell below the detection limit of 5 m, therefore hedgerows were regarded as linear elements. Movements in the hedgerow were divided into a probability to move forwards (0.28) and to turn around (0.02).

At the end of the experiment, 56% of the animals were found in hedgerows, whereas 24% were released in hedgerows at the start. Only 6% of the animals ended in arable land, compared to 29% released on arable land, indicating an avoidance of arable land. Pasture takes in an intermediate position with 38% present at the end and 47% released on pasture. The presence of frogs in hedgerows at the end of the experiment was larger and the presence in arable land was lower than expected, when assuming similar presence in all habitat types (chi-square test,  $p < 0.01$ ). As the actual cover of hedgerows in the landscape is much lower compared to arable land and pasture (2.6%, 34% and 64% respectively), this indicates a preference for hedgerows. The habitat type at the end of the experiment is only an indication for preference, as this is also determined by the habitat configuration at the release sites and by differences in velocity and turning angle in the different habitat types (Turchin 1998).

The two methods to estimate the transition probability between habitat types render different results. In the first approach, based on all occasions that transitions between habitat types might have occurred (Fig. 6.2), the probability to cross a pasture-hedgerow boundary is 0.86, while the estimated probability to cross a arable land-hedgerow boundary is smaller, i.e. 0.53 (Table 6.2). The probability to cross an arable land-pasture boundary is estimated 0.75, while observations on (potential) transitions from pasture to arable land are lacking. The estimated probability to cross a hedgerow-pasture boundary is 0.11, while the probability to cross a hedgerow-arable land boundary is 0.02 (Table 6.2). In the second approach, all transition probabilities are derived from the observed probability to cross a hedgerow-pasture boundary ( $P_{hp}$  0.11) or a hedgerow-arable land boundary ( $P_{ha}$  0.02), by assuming that boundary reactions between habitat types are symmetrical (Fig. 6.3). The two methods render similar estimates for pasture-hedgerow crossings (0.86 and 0.89 respectively). The estimates for an arable land-hedgerow are quite different (0.53 and 0.98 respectively). As the number of observations in hedgerows are most abundant, the transition probabilities based on hedgerow observations (second approach, Table 6.2) are regarded the best possible estimate from the field data.

Of the tracked animals 47% had reduced their distance to the Vildersveen with 10 m or more (mean reduction 84 m), 35% had increased their distance towards the Vildersveen with more than 10 m (mean increase 43 m), while for 18% of the animals the distance to the Vildersveen between start and end position differed less than 10 m. These results give no clear indication that animals were attracted towards the Vildersveen. To determine if attraction was correlated with distance from the Vildersveen, angles between the observed direction of movement and a direct movement towards the Vildersveen were calculated. All angles below  $90^\circ$  decrease the distance towards the Vildersveen, while angles between  $90^\circ$  and  $180^\circ$  will increase the distance (Fig. 6.4). The movement angle became significantly smaller with decreasing distance to the Vildersveen (linear regression,  $p < 0.001$ ). Under

Table 6.2

Estimated transition probabilities between pasture, arable land and hedgerows. The estimates are based on the Vildersveen data, including four extra observations in the Vildersveen area, where the animals were released in grassland at 30 or 75 m from two hedgerows. Transition probabilities are calculated by two approaches. See text and Figures 6.2 and 6.3 for explanation.

|                        | TO HEDGEROW          | TO PASTURE                  | TO ARABLE LAND              |
|------------------------|----------------------|-----------------------------|-----------------------------|
| <b>First approach</b>  |                      |                             |                             |
| FROM HEDGEROW          | -                    | $P_{hp}$ 0.11 (n=91)        | $P_{ha}$ 0.02 (n=91)        |
| FROM PASTURE           | $P_{ph}$ 0.86 (n=7)  | -                           | no observations             |
| FROM ARABLE LAND       | $P_{ah}$ 0.53 (n=15) | $P_{ap}$ 0.75 (n=4)         | -                           |
| <b>Second approach</b> |                      |                             |                             |
| FROM HEDGEROW          | -                    | $P_{hp}$ <b>0.11 (n=91)</b> | $P_{ha}$ <b>0.02 (n=91)</b> |
| FROM PASTURE           | $P_{ph}$ 0.90        | -                           | $P_{pa}$ 0.17               |
| FROM ARABLE LAND       | $P_{ah}$ 0.98        | $P_{ap}$ 0.83               | -                           |

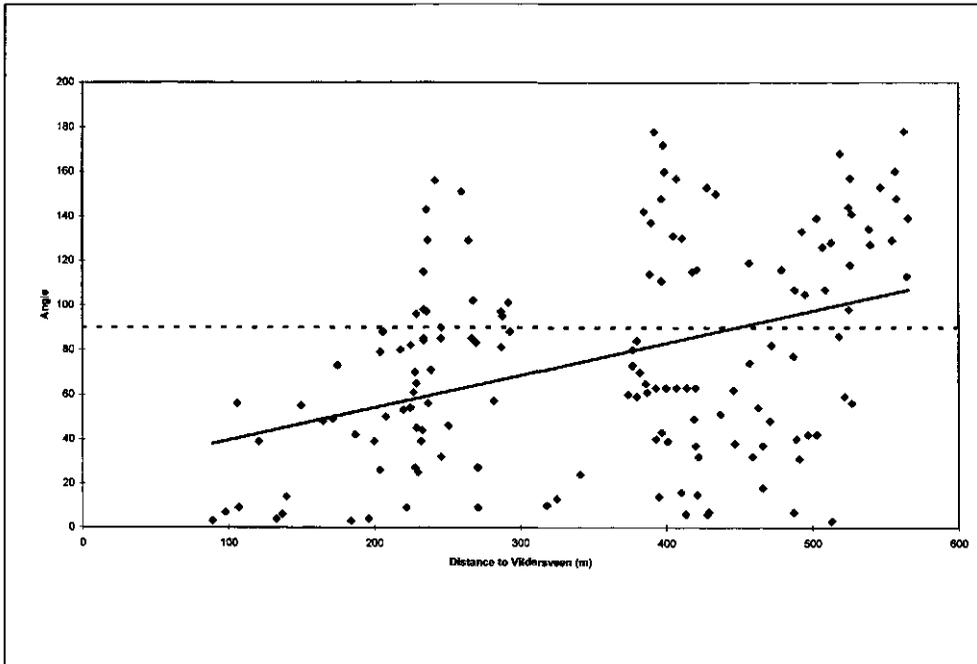


Figure 6.4

The Vildersveen angle, the angle between the observed direction of movement and a direct movement towards the Vildersveen. The Vildersveen angle is significantly smaller with decreasing distance to the Vildersveen (linear regression,  $p < 0.001$ ). Under 200 m all movement angles were smaller than 90° which decreased the distance towards the Vildersveen.

200 m all movement angles were smaller than  $90^\circ$ , thus reducing the distance towards the Vildersveen (Fig. 6.4). However, these observations under 200 m were based on the movement paths of four animals, only. This suggests that an influence of the breeding pond cannot be ruled out for the movement paths of these individuals.

In 1997, seven animals were radio-tracked in the second study area (Kolkman), to get an impression of behaviour in an area without a reproduction site in the vicinity. Four out of seven animals had not moved during the experiment. The small number of observations in the Kolkman area permits a qualitative comparison only. Velocity was slow compared to the Vildersveen ( $7.7 \text{ m} \pm 5.0$  in arable land,  $5.0 \text{ m}$  in pasture and  $1.0 \text{ m} \pm 3.3$  in hedgerows). The two animals that were released in arable land, left the arable land and ended in a hedgerow. The third animal was released in a hedgerow and ended in the pasture, close to the hedgerow. These movement paths seem to be in accordance with observed patterns in the Vildersveen area.

## The movement model *SmallSteps*

### Methods

Based on the empirical data obtained from the field experiments, a movement model is constructed. This model is individual- and vector-based, simulating movement of individuals as a correlated random walk: CRW (Kareiva & Shigesada 1983; Fisher 1993; Turchin 1998). The CRW is an appropriate formulation of movement for organisms that show some degree of directional persistence (Turchin 1998). For use in heterogeneous habitats, in addition to basic CRW formulations, rules are included representing behaviour at boundaries in the habitat. The actual model can be considered a species-specific exponent of the generic *SmallSteps* model (Baveco & Vos in prep.).

The simulation landscape is based on a vector-based GIS map of the Vildersveen area. Standard Arc-Info cover files are imported, defining polygons and arcs (linear habitat elements), with their attributes. Relevant habitat elements through which individuals may move, can be represented in the model both as polygons and arcs. In the simulation habitat for the tree frog, three habitat types are distinguished: arable land and pasture (polygons) and hedgerows (arcs).

A simulation started on one of the five release sites. In each simulation 10 000 hypothetical animals were released. The initial direction for the simulation animals was random, as was the case for the field data (Rayleigh test, Fisher 1993). The animal was allowed to move 20 time steps, which corresponds with two nights from 21.00 to 6.00 hrs.

Habitat-specific estimates of step size and turning angle are directly based on the field observations (Table 6.1). Step size, the distance covered between two observations, is assumed to have an exponential distribution. Turning angle, the change in direction between subsequent movements, is assumed to be normally distributed with a turning angle of  $0^\circ$  as the mean direction. This implies that for each distribution a single parameter is relevant. A similar turning angle was taken for arable land and pasture, as the observed angle distribution in the field data did

not differ significantly. For movement on linear habitat elements (hedgerows) represented as arcs, instead of a turning angle distribution, a probability of turning-around is defined (Table 6.1).

Individual decision-making at boundaries involves two aspects: deciding whether to cross a border, and determining the direction of the continued movement. By assuming that boundary reactions between habitat types are symmetrical, transition probabilities are interpreted as the reserve of preferences, with a value of 0.5 implying indifference (Fig. 6.3). This assumption implies that the history of the last move does not affect the probability to move to either side. With this simplification the number of required transition parameters in the model is reduced by half. The direction of the continued movement is based on 'reflection', as a default reaction for individuals deciding not to cross a boundary. Individuals that cross a boundary maintain their current angle.

#### Individual variation

The individual-based approach opens up the possibility to take into account differences between individuals (DeAngelis & Gross 1992). Early simulation results suggested that the initial assumption of all individuals being equal did not hold, as the variation in generated path lengths was much less than observed. Re-analysis of the data showed that indeed there was a large between-individuals variation in mean individual velocity (step size), at least in grassland. The log of the individual means of velocity was approximately normally distributed. Corresponding variation could be reproduced in the model by defining for each individual an intrinsic velocity, as a multiplication factor for step size, with a log-normal distribution over the population.

#### Attraction to the Vildersveen

In the model, we assumed the probability of being attracted, to be inversely related to the distance (*dist*) to the attraction source, the Vildersveen, according to the sigmoid relationship

$$P(\text{dist}) = 1 - (\text{dist}^2 / \text{par}^2 + \text{dist}^2)$$

where *par* denotes the distance at which *P* is 0.5 is set to 150 m in the calibration runs. When a disperser is attracted, it will make its next step in the direction of the attraction source, irrespective of its current habitat type.

#### Calibration

The transition probability matrix estimated from the field data suggests large differences in preferences for habitat types. In heterogeneous landscapes involving the habitat types of our study area, we may expect a large impact of the landscape mosaic on movement patterns. At the same time, as shown above, transition probabilities are hardest to estimate with certainty from field data. Therefore they form the main focus of a procedure that may produce parameters with an overall better fit to the data (calibration) as well as an indication of just how sensitive the observed movement pattern is to changes in the extent of the individual preferences. By varying transition probabilities in a stepwise fashion, several 'frog types' are tested. A strict 'corridor' frog type ( $P_{hp}$  0.1,  $P_{ha}$  0.01) is strongly bound to hedgerows

and the landscape mosaic will strongly affect movement paths. While on the other hand the choice of direction of the most 'indifferent' frog type ( $P_{hp}$  0.5,  $P_{ha}$  0.30) will hardly be influenced by the landscape mosaic.

As the telemetric data showed that movements within a radius of 200 m may be directed towards the Vildersveen, we calibrated the model with and without an attraction zone towards the Vildersveen (Fig. 6.4). So we tested whether an attraction zone would give a better explanation of the observed movement paths.

We compared the observed and simulated movement paths, to estimate which transition parameters describe the observed movement paths best. The compass direction (0-360°) and the transition time of first passage were registered, when crossing an imaginary circle at 25, 50, 75, 100, and 150 m distance from the release sites. This can be visualised as the intersection between a movement path and a circle with variable radius  $d$  drawn around the release sites. Time-angle pairs when a circle was crossed were calculated from simulations and empirical data, by linearly interpolating the first circle passage, assuming constant speed and straight direction between observations. We classified the compass angles in 24 classes of 15° and classified the time in five classes of approximately equal size. Animals that did not reach  $d$  within the simulation time, were classified in a separate class. Per simulation, results were summarised in a table of probabilities classified by compass angle and time, for each release location separately. A simulated multinomial log-likelihood was calculated for every parameter combination by comparing the empirical data with the simulated probabilities. Maximum likelihood, employing the summed log-likelihoods for the five release sites, was then used to estimate the transition parameters. Parameter combinations which did not differ significantly from the maximum likelihood estimates were determined by a likelihood ratio test (see e.g. Silvey 1975).

### Model results and calibration

We estimated the transition parameters by means of maximum likelihood for first passage at 25, 50, 75, 100, and 150 m distance. However, as the simulated multinomial distributions are not independent between distances, only one distance can be used for calibration. Release sites 1, 3 and 4 are situated in homogeneous landscapes, on 50 m distance from hedgerows (Fig. 6.1). These sites are non-informative to estimate transition probabilities in the first 50 m, as no boundaries are encountered. The release sites 2 and 5 are directly situated on a habitat boundary, and heterogeneity continues with increasing distance (Fig. 6.1). To incorporate maximum spatial differentiation of all release sites, we decided to calibrate on a circle distance of 75 m. For larger distances, a growing fraction of animals did not reach the distance at all within the simulation time, which decreases the power to discriminate between transition probabilities concomitantly.

Figure 6.5 shows the distribution patterns of individuals for different parameter values at release site 4 at 75 m. It illustrates how the predicted spatial distribution depends on the different transition probabilities. The compass direction where the imaginary circle intersects pasture is indicated by a single circle line, an intersection with arable land is indicated by a double circle line, while an intersection with hedgerow is indicated by a small line perpendicular to the circle

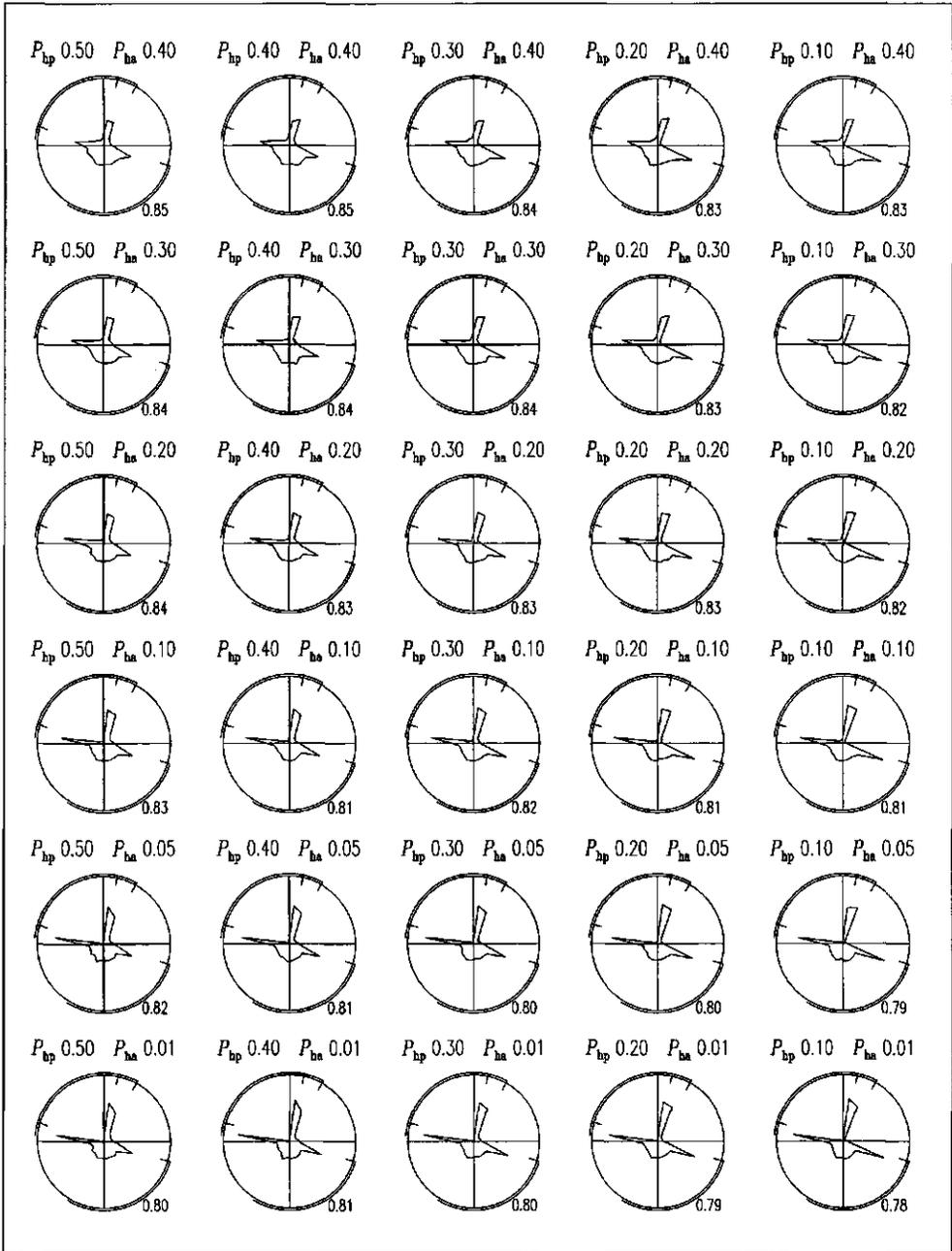


Figure 6.5

The distribution of simulated movement patterns at release location 4 at 75 m. Transition probabilities from hedgerow to pasture vary from 0.50 left to 0.10 right and from hedgerow to arable land from 0.30 top to 0.01 bottom. An intersection with pasture is indicated by a single circle line, an intersection with arable land by a double circle line and an intersection with hedgerow by a small line segment, perpendicular to the circle line. The numbers at the circle bottom indicate the fraction of animals that reached the circle distance within the simulation time.

line (Fig. 6.5). The transition probabilities vary from the most 'indifferent' frog type ( $P_{hp}$  0.5,  $P_{ha}$  0.4, Figure 6.5 top left) to the most strictly 'corridor bound' frog type ( $P_{hp}$  0.1,  $P_{ha}$  0.01, Figure 6.5 bottom right). In all cases frogs are found in hedgerows relatively often compared to arable land, as is illustrated by the uneven distribution of animals, in favour of the sections with hedgerows (Fig. 6.5, small lines perpendicular to the circle line). In absence of pronounced preference, this is caused by the relatively slow frog movement in hedgerows, compared to arable land and pastures. This effect gets stronger when transition probabilities become more biased towards the hedgerows (bottom right). Also note that the probability of reaching the 75-m circle distance within the simulation time, declines from top left to bottom right, due to differences in frog velocity per habitat type.

The influence of the landscape mosaic on the spatial distribution of individuals is illustrated in Figure 6.6a which shows the distribution at successive first-passage distances for release site 4. At 25 and 50 m distance, the landscape consists of homogeneous arable land and frogs are evenly distributed in all directions. From 50 to 75 m, the landscape becomes heterogeneous, and frogs encounter boundaries with hedgerows followed by pasture. This is illustrated by the skewed distribution in favour of the encountered hedgerows. At 100 and 150 m, the distribution pattern does not change as no new habitat types are encountered, although the fraction of animals that reach 100 and 150 m within the simulation time becomes smaller.

Figure 6.6b illustrates differences between the release sites by comparing the predicted circular patterns for  $P_{hp}$  0.1 and  $P_{ha}$  0.05 and first passage at 75 m. At release site 2 and 5, which are situated in hedgerows, the distribution peaks coincide with hedgerow positions. At release sites 1, 3 and 4 the landscape becomes heterogeneous after 50 m in part of the compass directions (Fig. 6.1). The two encountered hedgerows are reflected in the distributions (Fig. 6.6b).

Figure 6.6c illustrates the effect of incorporating an attraction zone surrounding the Vildersveen on the distribution pattern. The fact that the attraction is inversely related to distance is illustrated by the distribution patterns at release sites 1, 2 and 3, which are more strongly directed towards the Vildersveen, compared to release sites 4 and 5 (compare Figures 6.6b and 6.6c, see Figure 6.1 for the distances between release sites and the Vildersveen).

#### Determining the best parameter values

In Table 6.3 the results of the model calibration are listed, for the model with and without an attraction zone towards the Vildersveen, respectively. In Table 6.3a the summed log-likelihood is given for the first-passage distance of 75 m for the model without Vildersveen attraction. The maximum value attained by the summed log-likelihood (-83.76, Table 6.3a) is given by  $P_{hp}$  0.10 and  $P_{ha}$  0.05. The light grey area ( $p < 0.1$ ) and the dark grey area ( $p < 0.05$ ) indicate the parameter values that differ significantly from the maximum likelihood estimates, using a likelihood ratio test. At the  $p < 0.1$  level of significance, 12 parameter combinations do not significantly differ from the maximum likelihood estimates. At the 0.05 level of significance only seven parameter combinations have a significantly worse fit.

In Table 6.3b the summed log-likelihoods are listed when an attraction zone towards the Vildersveen is incorporated in the model. The maximum likelihood

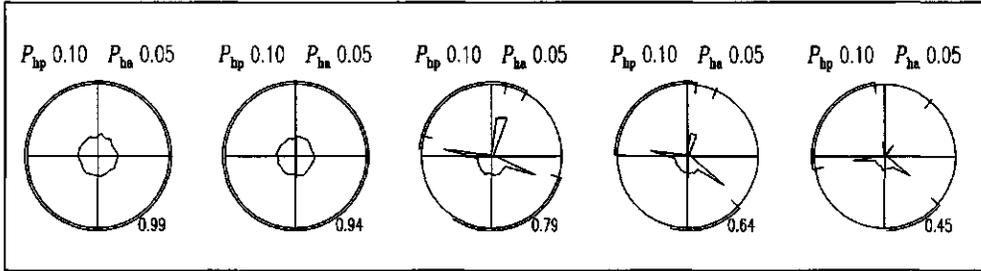


Figure 6.6a

The distribution of simulated movement patterns at release location 4, at a circle on 25, 50, 75, 100, and 150 m distance from the release location. The transition probability between habitat types is  $P_{hp}$  0.10 and  $P_{ha}$  0.05.

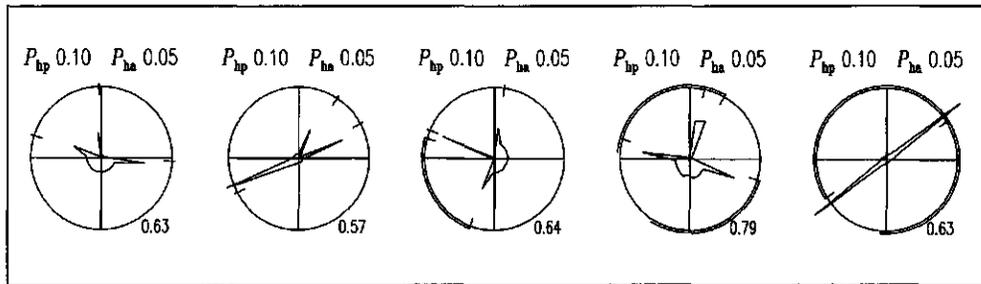


Figure 6.6b

The distribution of simulated movement patterns at the five release locations, at 75 m circle distance. The transition probability between habitat types is  $P_{hp}$  0.10 and  $P_{ha}$  0.05.

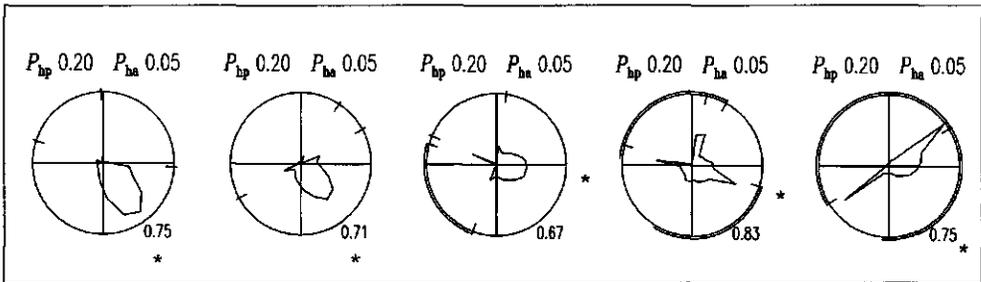


Figure 6.6c

The distribution of simulated movement patterns at the five release sites, at 75 m circle distance. An attraction zone towards the Vildersveen is incorporated. The transition probabilities between habitat types are  $P_{hp}$  0.20 and  $P_{ha}$  0.05. \* indicates the direction towards the Vildersveen.

estimates (-86.24, Table 6.3b) are given by  $P_{hp}$  0.2 and  $P_{ha}$  0.05, which is similar to the first calibration, except for a small increase in the probability to leave the hedgerow towards pasture from  $P_{hp}$  0.1 to  $P_{hp}$  0.2. The second calibration is slightly less discriminating between parameter values as compared to the first calibration. Also note that the first calibration, i.e. the model without Vildersveen attraction, has

Table 6.3

The calibration results. The summed log-likelihood for the circle crossing at 75 m are listed. The light grey area ( $p < 0.1$ ) and the dark grey area ( $p < 0.05$ ) indicate the parameter values that differ significantly from the maximum likelihood estimates, using a likelihood ratio test. The rows represent transition probabilities from hedgerow to pasture increasing from  $P_{hp}$  0.10 to  $P_{hp}$  0.50. In the columns the transition probability from hedgerow to arable land increases from  $P_{ha}$  0.01 to  $P_{ha}$  0.40. In Table 6.3a the results of the model without attraction are listed. The maximum value attained by the summed log-likelihood is -83.76. The maximum value is attained for  $P_{hp}$  0.10 and  $P_{ha}$  0.05.

In Table 6.3b the results of the model with an attraction zone towards the Vildersveen are listed. The maximum value attained by the summed log-likelihood is -86.24. The maximum value is attained for  $P_{hp}$  0.20 and  $P_{ha}$  0.05.

| $P_{hp}$ | 0.10   | 0.20   | 0.30   | 0.40   | 0.50   |
|----------|--------|--------|--------|--------|--------|
| 0.01     | -87.45 | -85.60 | -86.37 | -87.07 | -87.78 |
| 0.05     | -83.67 | -83.85 | -84.97 | -86.22 | -86.39 |
| 0.10     | -84.12 | -84.49 | -84.73 | -85.24 | -85.47 |
| 0.20     | -84.65 | -85.34 | -85.62 | -86.52 | -87.68 |
| 0.30     | -85.99 | -86.29 | -86.53 | -86.35 | -86.63 |
| 0.40     | -86.02 | -86.14 | -87.01 | -87.33 | -88.13 |

| $P_{hp}$ | 0.10   | 0.20   | 0.30   | 0.40   | 0.50   |
|----------|--------|--------|--------|--------|--------|
| 0.01     | -87.57 | -86.83 | -87.37 | -88.00 | -88.77 |
| 0.05     | -86.28 | -86.24 | -88.14 | -87.84 | -87.54 |
| 0.10     | -87.84 | -87.88 | -87.72 | -87.82 | -88.38 |
| 0.20     | -88.05 | -88.33 | -89.25 | -89.12 | -89.11 |
| 0.30     | -89.26 | -89.08 | -89.42 | -89.39 | -89.83 |
| 0.40     | -89.23 | -89.66 | -89.21 | -89.37 | -91.30 |

3b  $p < 0.1$   
 $p < 0.05$

a higher maximum log-likelihood compared to the second calibration (-83.76 and -86.24 respectively), indicating that the model without an attraction zone towards the Vildersveen is more plausible.

Both calibrations approximate the transition values that were estimated directly from the field data. The transition probabilities that were regarded the best estimate from the field data (Table 6.2, second approach),

gave a probability to cross a hedgerow-pasture boundary from  $P_{hp}$  0.11 and a probability to cross a hedgerow-arable land boundary from  $P_{ha}$  0.02. These probabilities are almost similar to the parameter values that gave the best fit for the model without attraction:  $P_{hp}$  0.10 and  $P_{ha}$  0.05 (Table 6.3a).

## Discussion

Displaced tree frogs showed in their movement paths a preference for hedgerows and an avoidance of arable land, while pastures took in an intermediate position. At the end of an experiment tree frogs were relatively often found in hedgerows. The probability to move from hedgerows to arable land, and to a lesser extent to pasture, was low. The calibration of the movement model *SmallSteps* showed the best fit between observed and simulated movement paths, when transition probabilities were strongly biased towards hedgerows. The predicted distribution patterns of this strict 'corridor' frog type was strongly skewed in favour of hedgerows. Although observed velocity in hedgerows was low especially when compared to arable land, the strong preference for hedgerows, makes hedgerows function as corridors in agricultural landscapes. Based on these results it is concluded that the resistance of the landscape mosaic between habitat patches should be incorporated to determine connectivity for tree frogs.

### The radio-tracking experiment

The displacement of individuals into a variation of different landscape configurations is based on the assumption that behaviour is similar to real dispersal behaviour. A similar approach was conducted in several other studies (Merriam & Lanoue 1990; Rufenacht & Knight 1995; Mauritzen et al. 1999). Merriam and Lanoue (1990) compared the behaviour of displaced animals, the so-called transients, with resident animals. Although they found a different behaviour between transients and residents, this still gives no information if transients resemble real dispersers. The decision to follow displaced animals instead of real dispersers is a practical one. As it is unknown which individuals will disperse, it would require disproportionately large numbers of tracked individuals and long tracking periods for only a small amount of observations of real dispersers. So displacement is the second-best option. Consequently, our model is based on and calibrated with field data collected from displaced animals. An option to test if the model also holds for real dispersal is to predict differences in connectivity between ponds and to compare these with observed dispersal events of marked individuals.

The tracking experiment was carried out during the reproduction season in the vicinity of the population in the Vildersveen. Movement patterns may thus have been influenced by social attraction. We have no unequivocal evidence that animals were attracted. We suggest that social attraction will decrease the impact of landscape heterogeneity on movement pattern. As we found outspoken preferences, this implies that without attraction the influence of landscape heterogeneity could even be larger.

As hedgerows form part of the terrestrial habitat of the tree frog, it is possible

that movements of individual frogs that are not motivated to disperse are directed towards hedgerows, while 'real dispersers' show less preference. However as hedgerows provide food and shelter against predation and drying out, which are lacking in arable land, it is plausible that also 'real dispersers' will show similar preference.

### *SmallSteps*

In the calibration method, we projected imaginary circles on the landscape and compared the compass direction of first-passage events between observed and simulated movement paths. This is a spatially explicit method, which permits to calibrate on a landscape level. The distribution of animals over the landscape is the result of individual decisions taken in reaction to local landscape properties. First-passage events are the result of cascading decisions, involving landscape-specific velocity, turning angle and transition parameters. An imaginary circle was required as, on the scale of the experiment, the intrinsic structure of the landscape itself was not sufficiently fine-grained. For instance it was not possible to calibrate on exchange between suitable habitat patches.

The calibration results indicate that the model without social attraction is a bit more plausible than the model with attraction towards the Vildersveen. In such a case the less complex model should be preferred. In their estimates of transition probabilities, the models show little difference. Both models indicate that an outspoken 'corridor' frog type fits best. This is in accordance with the transition probabilities that were estimated from the field data directly (Table 6.2).

The discriminating power of the calibration method was not strong, as many alternative transition probabilities differed not significantly from the best estimate. More empirical data would greatly improve the vigour of the test to point out the best fit.

GIS-based movement models often have many parameters and are not calibrated (Wennergren et al. 1995; Kareiva & Wennergren 1995). The availability of empirical data to calibrate and test the model appears the most serious void in application of spatially realistic modelling studies (Wennergren et al. 1995; Kareiva & Wennergren 1995). One should carefully balance the complexity and realism of model formulations against the empirical data available (Hartway et al. 1998; Ruckelshaus et al. 1997), and incorporate only those aspects and relationships that appear firm enough. This asks for a close link between data collecting in the field, and the development of predictive models. We, as well as others (Vermeulen 1995; Tischendorf & Wissel 1997), try to provide a sound empirical base to movement modelling on a landscape scale, by using observed movement paths as a starting point, and by extrapolating movements to a limited extent across spatial and temporal scales. We think our study is an example of an approach, where model development and the collection of empirical data are so closely linked, that parameter values of the model can directly be tested in the field. To test the reliability of the model we will use a dataset of observed dispersal events in a study area in Zeeland Flanders (Stumpel 1987a; Vos *et al.* submitted). We will analyse whether the predicted probability of dispersal between populations and the observed dispersal events are in accordance.

### Extrapolating to landscape connectivity

Connectivity does not solely depend on the relative resistance to movement of the matrix between habitat patches. Life-history characteristics that determine dispersal ability, such as the duration of the dispersal period, will pose a limit on the covered dispersal distance as well. Mortality during dispersal is a second aspect that should be considered. Little is known about habitat type-specific mortality during dispersal. Mortality risk when crossing arable land will be relatively high, due to absence of shelter and food. However, as the probability to move into arable lands is low, the incorporation of mortality risk in arable land as an extra model parameter may not be necessary. This is obviously not the case for the mortality risk when crossing roads. Amphibians do cross roads and have a considerable risk to die (e.g. Van Gelder 1973; Heine 1987; Kuhn 1987; Münch 1989; Vos & Chardon 1994; Hels 1999). As the survival probability when crossing roads is relatively well studied, estimates could be incorporated in the model (Van Gelder 1973; Heine 1987; Hels 1999).

In nature conservation, there is a need for general standards to incorporate the impact of landscape resistance on the connectivity within population networks. Also knowledge of effective corridors is required. The movement model *SmallSteps* can play a role to further underpin standards for optimal corridor design.

## 7 Towards ecologically-scaled landscape indices

### Summary

Nature conservation is increasingly based on a landscape approach rather than a species approach. Landscape planning that includes nature conservation goals requires integrated ecological tools. However, species differ widely in their response to landscape change. We propose a framework of ecologically-scaled landscape indices which takes into account this variation. Our approach is based on a combination of field studies of spatially structured populations (metapopulations) and model simulations in artificial landscapes. From these we seek for generalities in the relationship between species features, landscape indices and metapopulation viability.

The concept of ecological species profiles is used to group species according to characteristics that are important in metapopulation response to landscape change: individual area requirements as the dominant characteristic of extinction risk in landscape patches, and dispersal distance as the main determinant of the ability to colonise patches. The ecological profiles and landscape indices are then integrated into two ecologically-scaled landscape indices (ESLI): 'average patch carrying capacity' and 'average patch connectivity'. The field data show that the fraction of occupied habitat patches is correlated with the two ESLI.

To put the ESLI into a perspective of metapopulation persistence, we determine the viability for each of the six ecological profiles at different degrees of habitat fragmentation, using a metapopulation model and computer-generated landscapes. The model results show that the fraction of occupied patches is a good indicator for metapopulation viability. We discuss how ecological profiles, ESLI and the viability threshold can be applied for landscape planning and design in nature conservation.

### Introduction

In landscapes where natural habitats have been severely fragmented by intensive farming and urbanisation, biodiversity conservation is a growing issue in landscape planning and management. Networks of nature reserves are being proposed as a solution where the degree of fragmentation is considered to endanger the long-term persistence of target species (NPP 1990; European Union 1992; Opdam et al. submitted). Landscape managers and conservation planners need tools that link landscape characteristics to some critical measure of the sustainable conservation of biodiversity values. The biggest problem is how to integrate habitat network requirements of an array of species which greatly differ in their response to landscape pattern and change, due to different spatial requirements and different movement capacities (Lord & Norton 1990; Opdam 1990; Opdam et al. 1993; Noon et al. 1997).

Four approaches to this problem have been used. (1) Landscape indices, quantitative measures of landscape pattern, have been widely proposed (e.g. Franklin

& Forman 1987; O'Neill et al. 1988a; Turner 1989; Ripple et al. 1991; Baker & Cai 1992; McGaral & Marks 1995; Gustafson 1998). The strength of landscape indices is their straightforwardness, but their weakness is the lack of any explicit relationship to ecological processes. Such indices do not take into account differences in the scale at which species respond to landscape structure. (2) Strategic models (May 1973) have been used, that produce simple rules of thumb, such as extinction thresholds (e.g. Levins 1970; Lande 1987; Quin & Hastings 1987; Fahrig 1990; Wissel & Stoecker 1991). Although these rules, based on general ecological mechanisms, provide a sound framework for addressing the problem, they are of no practical use because they lack an explicit link to real landscapes and often neglect species-specific responses to landscape scale. (3) Empirical studies were conducted (see for reviews: Harrison 1991, 1994; Opdam 1991; Reich & Grimm 1996; Harrison & Taylor 1997), in most of which species attributes are linked to landscape pattern using single-year distribution or turnover patterns. These produce regression models that usually are hard to extrapolate to other landscape areas and also to long-term chance of persistence (Ter Braak et al. 1998). (4) A modelling approach to extend the empirical studies with has been used. Here empirical data are used to calibrate tactical models, and the models are used to link the landscape pattern to population viability (Lande 1987; Doak 1989; Verboom et al. 1991a; Hanski 1994; Lindenmayer & Lacy 1995; Lindenmayer & Possingham 1995; Sjögren-Gulve & Ray 1996; Thomas & Hanski 1997; Vos et al. submitted). Although ecologically sound, the weakness of the approach is the large amount of required data and its focus on single species. Furthermore, these tactical models yield no general results (May 1973).

A new approach, combining the strengths and overcoming the weaknesses of these earlier mentioned methods is needed. In such an approach it is essential to focus on the variation among species, by classifying species according to their response to habitat fragmentation. Although this has been attempted by using model simulations to group species according to their sensitivity to fragmentation (Andrén 1996), and by proposing 'ecological profiles for colonisation ability' (Grimm et al. 1996), *no empirical data were presented to underpin these model results.* Grimm et al. (1996) ignored the fact that some species have strong responses to changes in patch area, while others are more susceptible to patch connectivity (*sensu* Fahrig & Merriam 1985).

In this paper, we attempt to bridge the gap between empirical data (approach 3), single species models (approach 4) and general indices for habitat configuration at the landscape level (approach 1). Indicator species are distinguished that represent clusters of species (ecological profiles) differing in their spatial requirements for long-term persistence. We propose ecologically-scaled landscape indices (ESLI) as an alternative to general landscape indices. Results are then generalised using indicator species matrices and model simulations in computer-generated landscapes. We use the models to predict metapopulation viability from occupation patterns of habitat networks. We test the usefulness of the ESLI with empirical data from the highly human-dominated landscapes in The Netherlands.

## Introducing ESLI

Habitat fragmentation has two principal components: decrease in habitat area and increase in the isolation of the remaining habitat patches (Hanski & Gilpin 1991; Opdam et al. 1993). The incidence of a species in a patch ( $J$ ) can be described as a function of colonisation rate  $C$  and extinction rate  $E$ :

$$J = C / (C+E) \quad (1)$$

(Hanski 1994). The probability that a patch is occupied increases with growing colonisation rate and decreases with increasing extinction rate. Since colonisation rate is a function of patch isolation and extinction rate of patch size, relatively small or isolated habitat patches will more often be empty (Opdam et al. 1993; Verboom et al. 1993; Reich & Grimm 1996; Noon et al. 1997). The survival of a metapopulation (Levins 1970) depends on the equilibrium proportion of occupied patches and is a function of the colonisation rate/extinction rate ratio of the patches (equation 1). To determine a species' sensitivity to fragmentation the key factors which determine the (area-dependent) extinction and (isolation-dependent) colonisation probabilities must be known (Hanski 1994). Although several factors determine extinction risk, it is generally accepted that extinction risk is closely linked to population size (MacArthur & Wilson 1967; Richter-Dyn & Goel 1972; Den Boer 1981; Shaffer 1981; Gilpin & Soulé 1986; Goodman 1987; Mangel & Tier 1994; Foley 1997). For a species in a fragmented landscape, average population size is determined by the individual area requirements in relation to patch size. In this paper we use 'individual area requirements' (IAR, the area required for a reproductive unit, e.g. the territory of a pair of birds) as the key factor that differentiates between the sensitivity of species to patch size related extinction risk. In relation to IAR we define the carrying capacity  $K$  of a species  $s$  in a patch  $i$ ,  $K_{si}$  as

$$K_{si} = AREA_i / IAR_{si} \quad (2)$$

$AREA_i$  is the area of patch  $i$  and  $IAR_{si}$  is the individual area requirements of species  $s$  in patch  $i$ . We propose the following index 'average patch carrying capacity', being

$$ESLI \text{ 'average patch carrying capacity'} = \sum_{i=1}^n K_{si} / n \quad (3)$$

the arithmetic mean of the  $K_{si}$  values in a landscape. The index combines the average patch size, a neutral (not ecologically-scaled) landscape index, and the species-specific individual area requirement, and is therefore an ecologically-scaled landscape index (ESLI). In the same landscape, a species with small individual area requirements reaches a higher ESLI for average carrying capacity than a species with large individual area requirements.

To colonise habitat patches in a fragmented landscape, the prime condition is that dispersing individuals are able to reach habitat patches. Dispersal distance is highly variable amongst species (Hansson 1991; Stenseth & Lidicker 1992). For patch

connectivity we use the index  $C$  (Verboom et al. 1991a; Hanski 1994):

$$C_{si} = \sum_{j=1}^n A_j \exp(-\alpha_s D_{ij}) \quad (j \neq i) \quad (4)$$

where  $C_{si}$ , the connectivity of a species  $s$  in a patch  $i$ , is the sum of all contributions of patches  $j$  weighted both by their area ( $A_j$ ) and the distance between patches  $i$  and  $j$  ( $D_{ij}$ ). The contribution of a patch at distance  $D_{ij}$  declines exponentially with a species-specific parameter  $\alpha_s$  (Verboom et al. 1991a; Hanski 1994). The species-specific parameter  $\alpha_s$  can be derived from knowledge about the dispersal distance distribution of the species:  $\alpha$  can be set to a value that yields close to zero contributions at distances beyond the maximum observed dispersal distance (Fig. 7.1). As index we propose

$$\text{ESLI 'average patch connectivity'} = \sum_{i=1}^n C_{si} / n \quad (5)$$

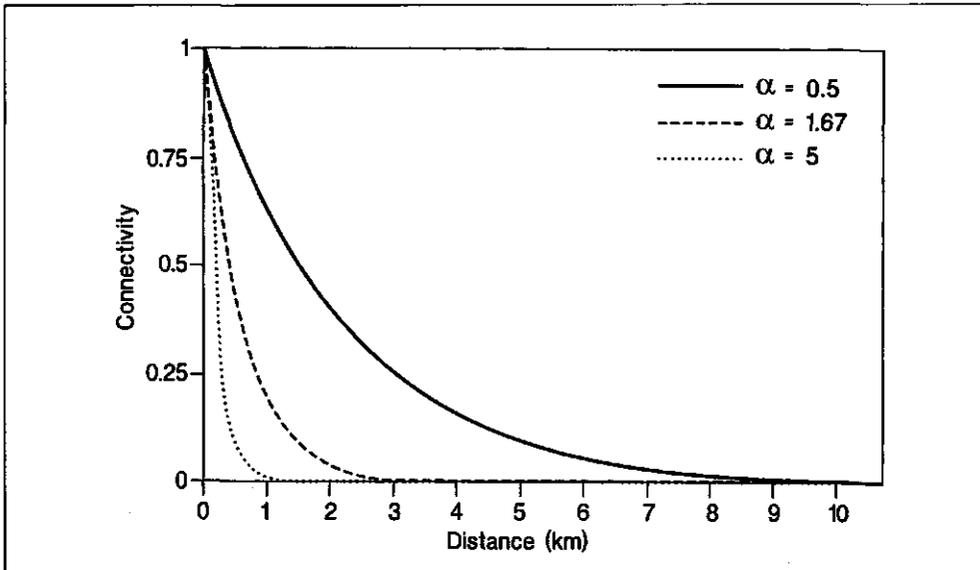


Figure 7.1

The influence of the value of the species specific parameter  $\alpha$  on the contribution of source habitat at a certain distance to the overall connectivity of a target patch. The limit cases are that all habitat contributes equally ( $\alpha = 0$ ), corresponding to unlimited dispersal, and that no patches contribute ( $\alpha \rightarrow \infty$ ), corresponding to zero dispersal distance. The  $\alpha$  value 0.5 corresponds to a maximum dispersal distance of 10 km,  $\alpha = 1.67$  to a maximum distance of 3 km and  $\alpha = 5.0$  to a maximum distance of 1 km. These values are used in the model simulations.

the arithmetic mean value of  $C_{si}$  in a landscape. In contrast to other neutral (not ecologically-scaled) connectivity indices, this index combines both species and landscape characteristics. In the same landscape it will be higher for species with a large dispersal distance (small  $\alpha_s$ ) than for species with small dispersal distance (large  $\alpha_s$ ). Throughout this paper we will use log-transformed ESLI in graphs and statistical analyses.

Table 7.1 gives an example of six model species that represent six ecological profiles for fragmentation sensitivity, combining different levels of individual area requirements and dispersal distances. The species with the largest individual area requirements and shortest dispersal distances is the most sensitive to fragmentation (Table 7.1, top left). The most fragmentation-tolerant species is able to reach high densities and to disperse over large distances (Table 7.1, bottom right). Note that all factors influencing extinction risk (growth rate, patch size etc.) also indirectly influence colonisation ability and vice versa (rescue effect).

Table 7.1

Six examples of ecological profiles for fragmentation sensitivity (a to f). Each ecological profile is a different combination of individual area requirements (IAR) and dispersal distance. IARs is a species (group) specific parameter for patch size related extinction risk (see equation 2).  $\alpha_s$  is a species (group) specific parameter for patch connectivity related colonisation ability (see equation 4). The six ecological profiles correspond with the six model species used in the simulations.

| Indicator species for fragmentation sensitivity  | Short Dispersal Distance*<br>0.1 - 1 km<br>( $\alpha_s = 5$ , equation 4)                                | Middle Dispersal Distance*<br>1 - 3 km<br>( $\alpha_s = 1.67$ , equation 4) | Large Dispersal Distance*<br>3 - 10 km<br>( $\alpha_s = 0.5$ , equation 4)                              |
|--|--|---|---|
| Large individual area requirements<br>1 r.u. **/ha<br>(IAR <sub>s</sub> = 1, equation 2)   | <b>a</b><br>high extinction risk<br>low colonisation ability<br><b>highest fragmentation sensitivity</b> | <b>b</b><br>high extinction risk<br>medium colonisation ability             | <b>c</b><br>high extinction risk<br>good colonisation ability   |
| Small individual area requirements<br>50 r.u. /ha<br>(IAR <sub>s</sub> = 0.02, equation 2) | <b>d</b><br>low extinction risk<br>low colonisation ability  | <b>e</b><br>low extinction risk<br>medium colonisation ability              | <b>f</b><br>low extinction risk<br>good colonisation ability<br><b>lowest fragmentation sensitivity</b> |

\* maximum dispersal distance covered by the main body of dispersers

\*\* r.u. = reproductive unit

## Model simulations

### Methods

ESLI have significance for the conservation of biodiversity if they can be linked to species viability for a variety of species and landscapes. We define a metapopulation in a landscape viable if the habitat configuration combined with the species' characteristics yield an extinction probability of less than 5% over a 100-year period (*sensu* the definition of a viable population Shaffer 1981; Shaffer & Samson 1985). As a consequence of our definition of metapopulation viability, which includes both a large time span and a probabilistic character, we have chosen a stochastic simulation model that captures the complexity of both the landscape and the species dynamics. The modelling has a strategic goal (*sensu* Metz & De Roos 1992), using hypothetical species in artificial landscapes. The main objective is to compare the performance of the ecological profile characteristics under increasingly fragmented conditions.

The simulations were carried out with the metapopulation model *Metaphor* (Verboom 1996; Verboom et al. 1999), developed in 1995 and used since in several theoretical and applied studies (Reijnen et al. 1995; Bugter & Vos 1997). *Metaphor* describes the spatial dynamics of a metapopulation in discrete time. The metapopulation is monitored once a year and changes in the metapopulation are calculated on a yearly basis. Individuals have a chance to reproduce and to die (dynamics within a patch) or to disperse (dynamics between patches; see the appendix for a detailed model description). The model species modules were derived from calibrated modules for two bird species: European bittern (*Botaurus stellaris*) and reed warbler (*Acrocephalus scirpaceus*). They were tuned to represent the six ecological profiles of Table 7.1, by incorporating the dispersal characteristics and area requirements. All model species are territorial and monogamous. The model species with large individual area requirements has been given a relatively low density-dependent growth rate and low mortality. The species with small individual area requirement has been given a higher density-dependent growth rate, higher number of dispersers and higher mortality. Some environmental stochasticity for this small species is incorporated as a higher standard deviation for birth and death rates. An overview of all parameter values is given in the appendix (Table A1).

The simulations were carried out in computer-generated landscapes of 100 km<sup>2</sup>, with suitable habitat percentages of 0.5%, 1%, 2%, 4% and 8%, distributed over habitat patches of 1 ha, 2 ha, 4 ha, 8 ha and 16 ha (Table A2). The number of patches per simulation landscape varied from 7 to 50 (Table A2). The low percentage of suitable habitat in the simulation landscapes reflects the habitat coverages in the empirical studies (Table 7.4). The low fraction of suitable habitat is also determined by the focus on viability thresholds in this study. The patches were distributed randomly by a landscape generator, keeping a minimal distance between patches of 1.5 times the home range size of the species with the largest area requirements. For each combination of habitat percentage and patch size, one landscape was generated. To keep the number of landscape-species combinations at a manageable level, patch size was kept constant in a simulation landscape. 100 simulation runs were conducted for each landscape-species combination. A run was

**Table 7.2**  
 The average fraction of occupied patches during the period the metapopulation was extant in relation to patch size and habitat percentage of the simulation landscapes for the six model species (a to f).

| a. Large individual area requirements |     |                          |      |      |                           |      |      |                          |      |      |      |  |  |
|---------------------------------------|-----|--------------------------|------|------|---------------------------|------|------|--------------------------|------|------|------|--|--|
|                                       |     | Short dispersal distance |      |      | Middle dispersal distance |      |      | Large dispersal distance |      |      |      |  |  |
|                                       |     | patch size               |      |      | patch size                |      |      | patch size               |      |      |      |  |  |
|                                       |     | 1                        | 2    | 4    | 8                         | 16   | 1    | 2                        | 4    | 8    | 16   |  |  |
| habitat (%)                           | 0.5 | 0.00                     | 0.00 | 0.00 | 0.07                      | 0.16 | 0.00 | 0.01                     | 0.04 | 0.34 | 0.71 |  |  |
|                                       | 1   | 0.00                     | 0.01 | 0.15 | 0.55                      |      | 0.00 | 0.07                     | 0.36 | 0.71 |      |  |  |
|                                       | 2   |                          | 0.01 | 0.18 | 0.63                      |      | 0.05 | 0.53                     | 0.77 |      |      |  |  |
|                                       | 4   |                          |      | 0.24 | 0.86                      |      | 0.84 | 0.89                     |      |      |      |  |  |
|                                       | B   |                          |      |      | 0.77                      |      |      |                          |      |      |      |  |  |

| b. Large individual area requirements |     |                           |      |      |                          |      |      |      |      |      |    |  |  |
|---------------------------------------|-----|---------------------------|------|------|--------------------------|------|------|------|------|------|----|--|--|
|                                       |     | Middle dispersal distance |      |      | Large dispersal distance |      |      |      |      |      |    |  |  |
|                                       |     | patch size                |      |      | patch size               |      |      |      |      |      |    |  |  |
|                                       |     | 1                         | 2    | 4    | 8                        | 16   | 1    | 2    | 4    | 8    | 16 |  |  |
| habitat (%)                           | 0.5 | 0.00                      | 0.01 | 0.04 | 0.34                     | 0.71 | 0.00 | 0.07 | 0.36 | 0.71 |    |  |  |
|                                       | 1   | 0.00                      | 0.00 | 0.07 | 0.36                     | 0.71 | 0.00 | 0.05 | 0.53 | 0.77 |    |  |  |
|                                       | 2   |                           | 0.05 | 0.53 | 0.77                     |      | 0.89 |      |      |      |    |  |  |
|                                       | 4   |                           |      | 0.84 | 0.89                     |      |      |      |      |      |    |  |  |
|                                       | B   |                           |      |      | 0.96                     |      |      |      |      |      |    |  |  |

| c. Large individual area requirements |     |                          |       |       |                           |    |       |                          |   |   |    |  |  |
|---------------------------------------|-----|--------------------------|-------|-------|---------------------------|----|-------|--------------------------|---|---|----|--|--|
|                                       |     | Short dispersal distance |       |       | Middle dispersal distance |    |       | Large dispersal distance |   |   |    |  |  |
|                                       |     | patch size               |       |       | patch size                |    |       | patch size               |   |   |    |  |  |
|                                       |     | 1                        | 2     | 4     | 8                         | 16 | 1     | 2                        | 4 | 8 | 16 |  |  |
| habitat (%)                           | 0.5 | 0.00                     | 0.028 | 0.069 | 0.667                     | 1  | 0.00  | 0.051                    | 0 | 0 | 0  |  |  |
|                                       | 1   | 0.008                    | 0.106 | 0.366 | 0.704                     | 1  | 0.00  | 0.01                     | 0 | 0 |    |  |  |
|                                       | 2   |                          | 0.29  | 0.777 | 1                         |    | 0.982 | 0.99                     | 1 | 1 |    |  |  |
|                                       | 4   |                          |       | 0.873 | 1                         |    |       |                          | 1 | 1 |    |  |  |
|                                       | B   |                          |       |       | 1                         |    |       |                          |   | 1 |    |  |  |

| d. Small individual area requirements |     |                          |       |       |                           |    |       |                          |   |   |    |  |  |
|---------------------------------------|-----|--------------------------|-------|-------|---------------------------|----|-------|--------------------------|---|---|----|--|--|
|                                       |     | Short dispersal distance |       |       | Middle dispersal distance |    |       | Large dispersal distance |   |   |    |  |  |
|                                       |     | patch size               |       |       | patch size                |    |       | patch size               |   |   |    |  |  |
|                                       |     | 1                        | 2     | 4     | 8                         | 16 | 1     | 2                        | 4 | 8 | 16 |  |  |
| habitat (%)                           | 0.5 | 0.008                    | 0.028 | 0.069 | 0.667                     | 1  | 0.00  | 0.051                    | 0 | 0 | 0  |  |  |
|                                       | 1   | 0.008                    | 0.106 | 0.366 | 0.704                     | 1  | 0.00  | 0.01                     | 0 | 0 |    |  |  |
|                                       | 2   |                          | 0.29  | 0.777 | 1                         |    | 0.982 | 0.99                     | 1 | 1 |    |  |  |
|                                       | 4   |                          |       | 0.873 | 1                         |    |       |                          | 1 | 1 |    |  |  |
|                                       | B   |                          |       |       | 1                         |    |       |                          |   | 1 |    |  |  |

| e. Small individual area requirements |     |                           |      |      |                          |      |      |      |      |      |    |  |  |
|---------------------------------------|-----|---------------------------|------|------|--------------------------|------|------|------|------|------|----|--|--|
|                                       |     | Middle dispersal distance |      |      | Large dispersal distance |      |      |      |      |      |    |  |  |
|                                       |     | patch size                |      |      | patch size               |      |      |      |      |      |    |  |  |
|                                       |     | 1                         | 2    | 4    | 8                        | 16   | 1    | 2    | 4    | 8    | 16 |  |  |
| habitat (%)                           | 0.5 | 0.00                      | 0.01 | 0.04 | 0.34                     | 0.71 | 0.00 | 0.07 | 0.36 | 0.71 |    |  |  |
|                                       | 1   | 0.00                      | 0.00 | 0.07 | 0.36                     | 0.71 | 0.00 | 0.05 | 0.53 | 0.77 |    |  |  |
|                                       | 2   |                           | 0.05 | 0.53 | 0.77                     |      | 0.89 |      |      |      |    |  |  |
|                                       | 4   |                           |      | 0.84 | 0.89                     |      |      |      |      |      |    |  |  |
|                                       | B   |                           |      |      | 0.96                     |      |      |      |      |      |    |  |  |

| f. Small individual area requirements |     |                           |      |      |                          |      |      |      |      |      |    |  |  |
|---------------------------------------|-----|---------------------------|------|------|--------------------------|------|------|------|------|------|----|--|--|
|                                       |     | Middle dispersal distance |      |      | Large dispersal distance |      |      |      |      |      |    |  |  |
|                                       |     | patch size                |      |      | patch size               |      |      |      |      |      |    |  |  |
|                                       |     | 1                         | 2    | 4    | 8                        | 16   | 1    | 2    | 4    | 8    | 16 |  |  |
| habitat (%)                           | 0.5 | 0.00                      | 0.01 | 0.04 | 0.34                     | 0.71 | 0.00 | 0.07 | 0.36 | 0.71 |    |  |  |
|                                       | 1   | 0.00                      | 0.00 | 0.07 | 0.36                     | 0.71 | 0.00 | 0.05 | 0.53 | 0.77 |    |  |  |
|                                       | 2   |                           | 0.05 | 0.53 | 0.77                     |      | 0.89 |      |      |      |    |  |  |
|                                       | 4   |                           |      | 0.84 | 0.89                     |      |      |      |      |      |    |  |  |
|                                       | B   |                           |      |      | 0.96                     |      |      |      |      |      |    |  |  |

**Table 7.3**  
 The extinction probability of the metapopulation in relation to patch size and habitat percentage of the simulation landscapes for the six model species (a to f).

| a. Large individual area requirements |     |                          |      |      |                           |      |      |                          |      |      |      |  |  |
|---------------------------------------|-----|--------------------------|------|------|---------------------------|------|------|--------------------------|------|------|------|--|--|
|                                       |     | Short dispersal distance |      |      | Middle dispersal distance |      |      | Large dispersal distance |      |      |      |  |  |
|                                       |     | patch size               |      |      | patch size                |      |      | patch size               |      |      |      |  |  |
|                                       |     | 1                        | 2    | 4    | 8                         | 16   | 1    | 2                        | 4    | 8    | 16   |  |  |
| habitat (%)                           | 0.5 | 1.00                     | 1.00 | 1.00 | 0.98                      | 0.92 | 1.00 | 1.00                     | 0.97 | 0.90 | 0.80 |  |  |
|                                       | 1   | 1.00                     | 1.00 | 1.00 | 0.98                      | 0.92 | 1.00 | 1.00                     | 0.96 | 0.91 | 0.80 |  |  |
|                                       | 2   |                          | 1.00 | 1.00 | 0.93                      | 0.80 | 1.00 | 1.00                     | 1.00 | 0.90 | 0.80 |  |  |
|                                       | 4   |                          |      | 1.00 | 0.90                      | 0.80 | 1.00 | 1.00                     | 1.00 | 0.90 | 0.80 |  |  |
|                                       | B   |                          |      |      | 1.00                      | 0.90 | 1.00 | 1.00                     | 1.00 | 0.90 | 0.80 |  |  |

| b. Large individual area requirements |     |                           |      |      |                          |      |      |      |      |      |      |  |  |
|---------------------------------------|-----|---------------------------|------|------|--------------------------|------|------|------|------|------|------|--|--|
|                                       |     | Middle dispersal distance |      |      | Large dispersal distance |      |      |      |      |      |      |  |  |
|                                       |     | patch size                |      |      | patch size               |      |      |      |      |      |      |  |  |
|                                       |     | 1                         | 2    | 4    | 8                        | 16   | 1    | 2    | 4    | 8    | 16   |  |  |
| habitat (%)                           | 0.5 | 1.00                      | 1.00 | 1.00 | 0.98                     | 0.92 | 1.00 | 1.00 | 0.97 | 0.90 | 0.80 |  |  |
|                                       | 1   | 1.00                      | 1.00 | 1.00 | 0.98                     | 0.92 | 1.00 | 1.00 | 0.96 | 0.91 | 0.80 |  |  |
|                                       | 2   |                           | 1.00 | 1.00 | 0.93                     | 0.80 | 1.00 | 1.00 | 1.00 | 0.90 | 0.80 |  |  |
|                                       | 4   |                           |      | 1.00 | 0.90                     | 0.80 | 1.00 | 1.00 | 1.00 | 0.90 | 0.80 |  |  |
|                                       | B   |                           |      |      | 1.00                     | 0.90 | 1.00 | 1.00 | 1.00 | 0.90 | 0.80 |  |  |

| c. Large individual area requirements |     |                          |      |      |                           |      |      |                          |      |      |      |  |  |
|---------------------------------------|-----|--------------------------|------|------|---------------------------|------|------|--------------------------|------|------|------|--|--|
|                                       |     | Short dispersal distance |      |      | Middle dispersal distance |      |      | Large dispersal distance |      |      |      |  |  |
|                                       |     | patch size               |      |      | patch size                |      |      | patch size               |      |      |      |  |  |
|                                       |     | 1                        | 2    | 4    | 8                         | 16   | 1    | 2                        | 4    | 8    | 16   |  |  |
| habitat (%)                           | 0.5 | 0.97                     | 0.94 | 0.89 | 0.17                      | 0.00 | 0.00 | 0.00                     | 0.00 | 0.00 | 0.00 |  |  |
|                                       | 1   | 0.97                     | 0.27 | 0.10 | 0.00                      | 0.00 | 0.00 | 0.00                     | 0.00 | 0.00 | 0.00 |  |  |
|                                       | 2   |                          | 0.10 | 0.00 | 0.00                      | 0.00 | 0.00 | 0.00                     | 0.00 | 0.00 | 0.00 |  |  |
|                                       | 4   |                          |      | 0.00 | 0.00                      | 0.00 | 0.00 | 0.00                     | 0.00 | 0.00 | 0.00 |  |  |
|                                       | B   |                          |      |      | 0.00                      | 0.00 | 0.00 | 0.00                     | 0.00 | 0.00 | 0.00 |  |  |

| d. Small individual area requirements |     |                          |      |      |                           |      |      |                          |      |      |      |  |  |
|---------------------------------------|-----|--------------------------|------|------|---------------------------|------|------|--------------------------|------|------|------|--|--|
|                                       |     | Short dispersal distance |      |      | Middle dispersal distance |      |      | Large dispersal distance |      |      |      |  |  |
|                                       |     | patch size               |      |      | patch size                |      |      | patch size               |      |      |      |  |  |
|                                       |     | 1                        | 2    | 4    | 8                         | 16   | 1    | 2                        | 4    | 8    | 16   |  |  |
| habitat (%)                           | 0.5 | 0.97                     | 0.94 | 0.89 | 0.17                      | 0.00 | 0.00 | 0.00                     | 0.00 | 0.00 | 0.00 |  |  |
|                                       | 1   | 0.97                     | 0.27 | 0.10 | 0.00                      | 0.00 | 0.00 | 0.00                     | 0.00 | 0.00 | 0.00 |  |  |
|                                       | 2   |                          | 0.10 | 0.00 | 0.00                      | 0.00 | 0.00 | 0.00                     | 0.00 | 0.00 | 0.00 |  |  |
|                                       | 4   |                          |      | 0.00 | 0.00                      | 0.00 | 0.00 | 0.00                     | 0.00 | 0.00 | 0.00 |  |  |
|                                       | B   |                          |      |      | 0.00                      | 0.00 | 0.00 | 0.00                     | 0.00 | 0.00 | 0.00 |  |  |

| e. Small individual area requirements |     |                           |      |      |                          |      |      |      |      |      |      |  |  |
|---------------------------------------|-----|---------------------------|------|------|--------------------------|------|------|------|------|------|------|--|--|
|                                       |     | Middle dispersal distance |      |      | Large dispersal distance |      |      |      |      |      |      |  |  |
|                                       |     | patch size                |      |      | patch size               |      |      |      |      |      |      |  |  |
|                                       |     | 1                         | 2    | 4    | 8                        | 16   | 1    | 2    | 4    | 8    | 16   |  |  |
| habitat (%)                           | 0.5 | 0.97                      | 0.94 | 0.89 | 0.17                     | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |  |  |
|                                       | 1   | 0.97                      | 0.27 | 0.10 | 0.00                     | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |  |  |
|                                       | 2   |                           | 0.10 | 0.00 | 0.00                     | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |  |  |
|                                       | 4   |                           |      | 0.00 | 0.00                     | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |  |  |
|                                       | B   |                           |      |      | 0.00                     | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |  |  |

| f. Small individual area requirements |     |                           |      |      |                          |      |      |      |      |      |      |  |  |
|---------------------------------------|-----|---------------------------|------|------|--------------------------|------|------|------|------|------|------|--|--|
|                                       |     | Middle dispersal distance |      |      | Large dispersal distance |      |      |      |      |      |      |  |  |
|                                       |     | patch size                |      |      | patch size               |      |      |      |      |      |      |  |  |
|                                       |     | 1                         | 2    | 4    | 8                        | 16   | 1    | 2    | 4    | 8    | 16   |  |  |
| habitat (%)                           | 0.5 | 0.97                      | 0.94 | 0.89 | 0.17                     | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |  |  |
|                                       | 1   | 0.97                      | 0.27 | 0.10 | 0.00                     | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |  |  |
|                                       | 2   |                           | 0.10 | 0.00 | 0.00                     | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |  |  |
|                                       | 4   |                           |      | 0.00 | 0.00                     | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |  |  |
|                                       | B   |                           |      |      | 0.00                     | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |  |  |

started with all patches occupied up to carrying capacity level. The simulations ran for 150 time steps, but only the last 100 were used in the analyses. After 50 time steps the pattern of occupancy is presumed to be the result of an equilibrium between species and landscape characteristics and no longer dependent on the initial distribution.

Metapopulation viability was determined by calculating the extinction probability of the metapopulation during the 100 runs. The average fraction of occupied patches, during the period the metapopulation was extant, was calculated as an ecological measure of fragmentation effects (Levins 1970). The percentage of suitable habitat is given as a neutral landscape index of fragmentation, so linking our approach to previous literature (O'Neill et al. 1988b; Gustafson & Parker 1992; Gustafson 1998). To explore the viability of the metapopulations in the model species-simulation landscape combinations, the extinction probability was analysed with logistic regression analysis (Jongman et al. 1995), using the average fraction of occupied patches and the model species as explanatory variables. In addition, the average fraction of occupied patches was analysed by logistic regression analysis with the ESLI as explanatory variables.

### Model results

Simulation results are presented in Tables 7.2 and 7.3. As expected, the mean fraction of occupied patches increased for each model species with increasing percentage of suitable habitat and increasing patch size (Table 7.2). Differences between the model species were as expected. Larger dispersal distances increased the fraction of occupation for both species with large individual area requirements (compare Table 7.2 a, b and c) and for species with small individual area requirements (compare Table 7.2 d, e and f). The species with small individual area requirements had a higher degree of occupation in similar landscapes, with similar dispersal distance compared to species with large individual area requirements (compare Table 7.2 a with d, b with e and c with f).

Similarly the extinction probability of the metapopulation decreased with increasing habitat percentage and increasing patch size (Table 7.3). Small individual area requirements and increasing dispersal distance had a positive effect on the survival probability. The model species with the highest fragmentation sensitivity, large individual area requirements and small dispersal distance, became extinct in most simulation landscapes. The most fragmentation-tolerant model species, with small individual area requirements and large dispersal distance, had the highest survival probability in most landscapes (compare Table 7.3a and 7.3f). Hence our assumption holds that the ecological profiles characteristics (Table 7.1) are related to fragmentation sensitivity and that the fragmentation-tolerant species can survive in a wider range of landscape structures than fragmentation-sensitive species.

In the logistic regression analysis of the extinction probability, both the fraction of occupied patches and the factor 'model species' were significant ( $p < 0.001$ ). The fraction occupied patches explained 340 times more variance per degree of freedom than the factor 'model species' and was therefore kept as the only predictor for the extinction probability. The extinction probability decreased with increasing fraction of occupied patches (Fig. 7.2) and is less than 5% in 100 years

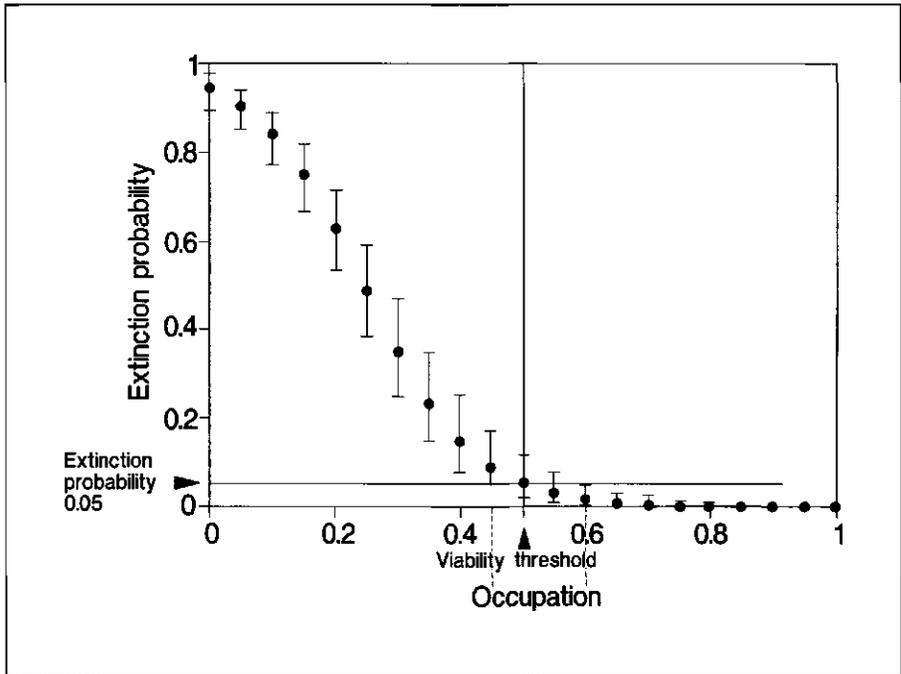


Figure 7.2

The viability of the metapopulations in the model simulations was analysed with logistic regression analysis. The predicted relationship between the average fraction of occupied patches and the extinction probability of the metapopulation is shown. The extinction probability is less than 5% in 100 years when the fraction of occupation is  $> 0.5$ . The viability threshold at 0.5 fraction of occupied patches is indicated. The 95% confidence interval is 0.4-0.6.

when the fraction of occupied patches is 0.5 or higher. Thus the metapopulation is viable when the fraction of occupied patches is 0.5 or higher, which can be regarded as a viability threshold (Fig. 7.2). The 95%-confidence interval for this threshold is 0.4-0.6. The model thus predicts that a metapopulation is viable if the fraction of occupied patches is larger than 0.6 and nonviable when it is less than 0.4. This prediction rule gave five mismatches (6%) in the simulation data. In three simulations the metapopulation was wrongfully judged nonviable, and in two simulations the metapopulation was wrongfully judged viable

The rows and columns of Table 7.2 were then re-scaled from neutral landscape indices into ecologically-scaled landscape indices. The ESLI 'average patch carrying capacity' and 'average patch connectivity' were calculated for all model species-simulation landscape combinations using equations (3) and (5). The combinations were graphed with the two ESLI as axes (Fig. 7.3). For each model species the mean fraction of occupied patches increased with increasing values of the ESLI (Fig. 7.3). The different position of the six model species in the graph illustrates the dependence of the ESLI on the species characteristics. Any particular



landscape has different ESLI values for each species, the more fragmentation-tolerant the species, the higher the ESLI values (compare Figure 7.3a and 7.3f). If we consider a certain landscape and look at the performance of the six model species, we see as expected that the occupation and viability increase with both increasing dispersal distance and decreasing individual area requirements.

The isoline of 0.5 fraction of occupation, the viability threshold, is estimated with logistic regression analysis with the two (log-transformed) ESLI as explanatory variables (Fig. 7.3). The landscape space in which metapopulations are viable (on the right side of the 0.5 isoline) increases from the most fragmentation-sensitive ecological profile (large individual area requirements and short dispersal distance; Figure 7.3a) to the most fragmentation-tolerant ecological profile (small individual area requirements and large dispersal distances; Fig. 7.3f). So the ESLI are useful predictors of metapopulation viability when a comparison is made for one species in landscapes with different degrees of habitat fragmentation.

If the species-specific characteristics are effectively incorporated into the ESLI, one would expect that the positive correlation between fraction of occupation and the ESLI values will hold regardless of the model species. The ESLI for average patch connectivity does show this expected increase in occupation irrespective of the model species (compare the fraction of occupation and the position along the ESLI for average patch connectivity axis for all species, Fig. 7.3). However, the ESLI for average patch carrying capacity does not correlate with occupation, when a comparison is made between species. There seems to be a discrepancy between the three model species with large individual area requirements (a, b and c, Fig. 7.3) and the species with small individual area requirements (d, e and f, Fig. 7.3).

## Testing ESLI to empirical data

The empirical studies were carried out in the extremely fragmented landscapes of The Netherlands - fragmentation due mainly to human activities - with percentages of suitable habitat ranging from 0.2 to 25 (Table 7.4). Empirical data come from a wide range of species: birds, mammals, insects, amphibians and plants. Data collection methods - except for species-specific inventory methods - were similar and so results are valid for between-species comparison. Details of the studies are available as follows: birds: nuthatch (*Sitta europea*) Verboom et al. 1991a; reed warbler (*Acrocephalus scirpaceus*) Foppen & Chardon 2000; mammals: root vole (*Microtus oeconomus*) Bergers et al. 1994; insects: ants: trunk ant (*Formica truncorum*) Mabelis 1987; grasshoppers: meadow grasshopper (*Chorthippus parallelus*) and bog bush cricket (*Metrioptera brachyptera*) Mabelis & Mekenkamp 1996; butterflies: green hairstreak (*Callophrys rubi*), silver studded blue (*Plebejus argus*) and grayling (*Hipparchia semele*) Verspui (submitted); amphibians: tree frog (*Hyla arborea*) Vos & Stumpel 1996, Vos et al. (submitted), moor frog (*Rana arvalis*) Vos & Chardon 1998 and plants: honeysuckle (*Lonicera periclymenum*) and bird cherry (*Prunus serotina*) Grashof-Bokdam 1997. Each study made an inventory of all potential suitable habitat patches and collected presence and absence data summarised in Table 7.4. Both occupied and unoccupied habitat patches were found in all studies.

Table 7.4

A summary of the empirical data. For each species the % coverage of suitable habitat and the fraction of occupied patches in the study areas are listed with the results of logistic regression analysis on patch area and connectivity (levels of significance: \*  $p < 0.05$ , \*\*  $p < 0.01$ , \*\*\*  $p < 0.001$ , ns not significant). References for more species-specific details are given. Note that for the root vole the results from two different study cases are given separately

| SPECIES                | HABITAT % | OCCUPIED FRACTION | REGRESSION ANALYSIS |              | Reference                |
|------------------------|-----------|-------------------|---------------------|--------------|--------------------------|
|                        |           |                   | PATCH SIZE          | CONNECTIVITY |                          |
| nuthatch               | 1.5       | 0.42              | ***                 | ***          | Verboom et al. 1991a     |
| reed warbler           | 0.2       | 0.67              | ***                 | ***          | Foppen & Chardon 2000    |
| root vole - Goeree (1) | 2.18      | 0.71              | ns                  | ns           | Bergers et al. 1994      |
| - Schouwen (2)         | 0.36      | 0.59              | ns                  | *            | Bergers et al. 1994      |
| trunk ant              | 0.9       | 0.24              | **                  | *            | Mabelis 1987             |
| bog bush cricket       | 2.5       | 0.51              | ***                 | *            | Mabelis & Mekenkamp 1996 |
| meadow grasshopper     | 7.4       | 0.72              | ***                 | ns           | Mabelis & Mekenkamp 1996 |
| grayling               | 18        | 0.57              | ***                 | ns           | Verspui submitted        |
| green hairstreak       | 25        | 0.89              | *                   | ns           | Verspui submitted        |
| silver studded blue    | 18        | 0.34              | ***                 | ns           | Verspui submitted        |
| tree frog              | 1.4       | 0.10              | ns                  | ***          | Vos & Stumpel 1996       |
| moor frog              | 11.2      | 0.67              | ***                 | ns           | Vos & Chardon 1998       |
| honeysuckle            | 16        | 0.09              | ***                 | ***          | Grashof-Bokdam 1997      |
| bird cherry            | 16        | 0.09              | ***                 | ns           | Grashof-Bokdam 1997      |

To test effects of habitat fragmentation, the distribution pattern was analysed by logistic regression. In the regression analysis habitat quality factors were first entered in the model. Subsequently, to test whether fragmentation variables were additional explanatory factors (Van Apeldoorn et al. 1992; Fahrig et al. 1995; Vos & Stumpel 1996), the model was extended with an area variable 'patch size' and a connectivity variable depending on the dispersal distance of the species (see equation 2). Except for the root vole on the island of Goeree, logistic regression analysis of the distribution pattern revealed significant effects of habitat fragmentation in all studies. However, in only 5 of the 14 studies was the probability of occupancy related to both patch size and connectivity (Table 7.4).

Although the fraction of occupation differed widely between species (Table 7.4) there seemed to be no relation with the neutral landscape indices 'mean patch size' and 'percentage of suitable habitat' (Fig. 7.4). For example there was a very low degree of occupancy (0.09) in honeysuckle and bird cherry, although the percentage of suitable habitat (16%) is relatively high. In contrast, the reed warbler was found in a much higher fraction (0.67) in a landscape with only 0.5% suitable habitat (Fig. 7.4). So these neutral landscape indices cannot be applied as predictors of metapopulation viability if different species are compared.

What picture emerges if the neutral landscape indices are re-scaled into ESLI, in which species differences for fragmentation sensitivity are incorporated? In Table 7.5 the species are grouped according to their estimated individual habitat area requirements and their maximum dispersal distance, the distance that will be

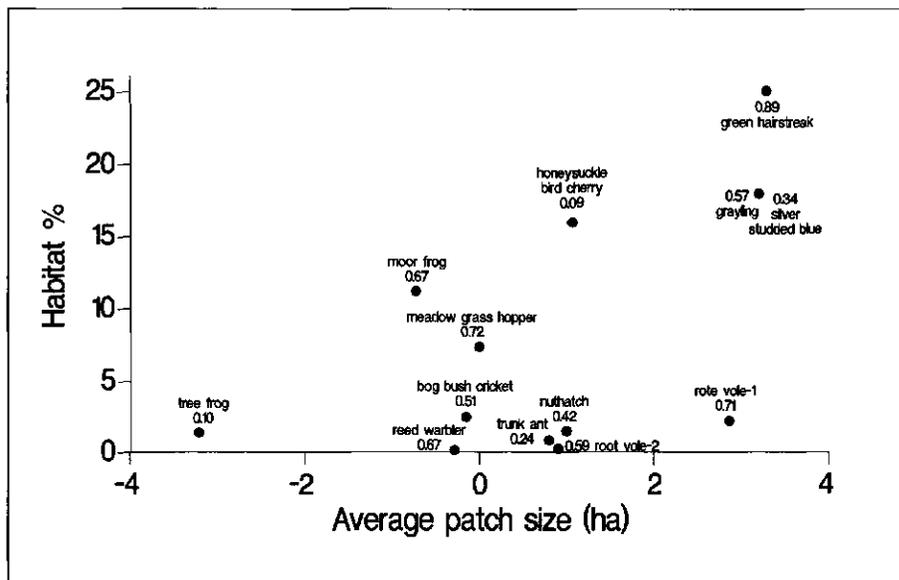


Figure 7.4

Arrangement of the empirical data according to two neutral landscape indices: the percentage of suitable habitat and the (log-transformed) average patch size of the study areas. Each point in the graph represents a species-landscape combination. For each species the observed fraction of occupied patches is indicated.

Table 7.5

Ecological profiles and ecologically-scaled landscape indices for species in the empirical dataset. Classes of individual area requirements (IAR<sub>s</sub>), the species (group) specific parameter for patch size related extinction risk, are listed (see also equation 2 in text ). The species (group) specific dispersal distance and the dispersal distance related  $\alpha_s$  the parameter for patch connectivity related colonisation ability, are listed (equation 4). Species are listed in order of fragmentation sensitivity, starting with the most fragmentation-sensitive profile. Nine different ecological profiles are distinguished. Model species with similar ecological profiles are indicated. The ESL 'average patch carrying capacity' (equation 3) and 'average patch connectivity' (equation 5) are given

| SPECIES<br>INDIVIDUAL AREA | MAXIMUM                        | ECOLOGICAL PROFILE                          |                          |            |  | ESL                                       |  |
|----------------------------|--------------------------------|---|--------------------------|------------|--|---|--|
|                            |                                | REQUIREMENTS IAR <sub>s</sub><br>(R.U.*/HA) | DISPERSAL<br>DISTANCE ** | $\alpha_s$ | AVERAGE PATCH<br>CARRYING<br>CAPACITY $\pm$ SD | AVERAGE PATCH<br>CONNECTIVITY<br>$\pm$ SD |  |
| 1 honeysuckle              | <i>Lonicera periclymenum</i>   | 1-10  | < 0.1 km                 | 50         | 17.4 $\pm$ 197.00                              | 0.07 $\pm$ 4.40                           |  |
| 2 trunk ant                | <i>Formica truncorum</i>       | 1 nest                                      | 0.1 - 1 km               | 5          | 2.2 $\pm$ 5.85                                 | 0.80 $\pm$ 0.67                           |  |
| 3 bird cherry              | <i>Prunus serotina</i>         | 1-10  | 0.1 - 1 km               | 5          | 17.4 $\pm$ 197.00                              | 0.28 $\pm$ 8.52                           |  |
| 4 root vole - Goeree (1)   | <i>Microtus oeconomus</i>      | 10-100                                      | 1 - 3 km                 | 1.67       | 983.136 $\pm$ 3973.50                          | 36.11 $\pm$ 72.07                         |  |
| - Schouwen (2)             | <i>Microtus oeconomus</i>      | 10-100                                      | 1 - 3 km                 | 1.67       | 136.864 $\pm$ 506.21                           | 2.55 $\pm$ 6.62                           |  |
| grayling                   | <i>Hipparchia semele</i>       | 10-100                                      | 1 - 3 km                 | 1.67       | 1372 $\pm$ 8330.36                             | 138.10 $\pm$ 267.56                       |  |
| green hairstreak           | <i>Callophrys rubi</i>         | 10-100                                      | 1 - 3 km                 | 1.67       | 1504.72 $\pm$ 10022.75                         | 117.59 $\pm$ 284.91                       |  |
| 5 silver studded blue      | <i>Plebejus argus</i>          | 100-1000                                    | 0.1 - 1 km               | 5          | 13499.5 $\pm$ 81964.79                         | 84.44 $\pm$ 245.26                        |  |
| bog bush cricket           | <i>Metrioptera brachyptera</i> | 100-1000                                    | 0.1 - 1 km               | 5          | 469.3418 $\pm$ 2259.03                         | 0.25 $\pm$ 0.91                           |  |
| tree frog                  | <i>Hyla arborea</i>            | 100-1000 ***                                | 1 - 3 km                 | 1.67       | 22.68 $\pm$ 65.52                              | 0.28 $\pm$ 0.18                           |  |
| moor frog                  | <i>Rana arvalis</i>            | 100-1000 ***                                | 1 - 3 km                 | 1.67       | 268.8 $\pm$ 90.31                              | 1.05 $\pm$ 0.99                           |  |
| 7 meadow grasshopper       | <i>Chorthippus parallelus</i>  | 1000-10.000                                 | 0.1 - 1 km               | 5          | 5413.5341 $\pm$ 26018.43                       | 6.98 $\pm$ 7.45                           |  |
| 8 nuthatch                 | <i>Sitta europea</i>           | 1   | 3 - 10 km                | 0.5        | 2.7 $\pm$ 6.44                                 | 64 $\pm$ 25.50                            |  |
| 9 reed warbler             | <i>Acrocephalus scirpaceus</i> | 10-100                                      | 3 - 10 km                | 0.5        | 42 $\pm$ 200.89                                | 3.43 $\pm$ 4.85                           |  |

\* R.U. = reproductive unit

\*\* covered by the main body of dispersers

covered by the main body of dispersers. These estimations are based on field observations, literature and expert knowledge. The 13 species can be grouped into 9 different ecological profiles based on their individual area requirements and dispersal distance (Table 7.5). Six species have ecological profiles that coincide with those of the model indicator species (Table 7.5, model species a, c, d, and f). The profiles of the other seven species have either a smaller individual area requirement, (with a maximum of 1000-10 0000 reproductive units per hectare of the meadow grasshopper), or a smaller dispersal distance (e.g. honeysuckle:  $< 0.1$  km,  $\alpha = 50$ , Table 7.5).

ESLI of 'average patch carrying capacity' and 'average patch connectivity' were calculated for each species landscape combination (equations (3) and (5), Table 7.5). The standard deviation of the values that are averaged to obtain the ESLI are very high, notably for the silver studded blue and the meadow grasshopper (Table 7.5). This can be explained by the presence of a large 'mainland patch' or source patch in the landscape (Harrison 1991), a feature that was not incorporated in our simulation models, where patch size per simulation landscape was held constant. When arranging the species in a graph with the ESLI on the axes, the fraction of occupied patches positively correlates (Fig. 7.5). The silver studded blue deviates from this general trend, by having a small occupied fraction (0.34) in contrast to the high connectivity and carrying capacity of its landscape. In a logistic regression of the fraction of occupied patches on the ESLI, without this outlier, both indices are significant ( $p < 0.05$ ). In contrast to the model simulation results this positive correlation also holds for the ESLI 'average patch carrying capacity'. Species in the left bottom side of the graph are those that will suffer most from habitat fragmentation: their low connectivity results in low colonisation chances and their small populations have a high extinction risk. These species have very low values for patch occupancy. The fraction of occupied patches gradually increases with increasing connectivity and carrying capacity per patch, with the highest occupancy in the right top side of the graph.

Since the ESLI account for species-specific dispersal distance and individual area requirements, species can have quite different values with similar amounts of habitat: compare for example the figures of the nuthatch and the tree frog (Table 7.4, Fig. 7.5). The reason for relatively high patch occupation in the reed warbler (0.67) in a landscape with only 0.2% of suitable habitat now becomes clearer. The reed warbler has the most fragmentation-tolerant profile, and so has relatively high ESLI values even in an extremely fragmented landscape. In general, however, the relationship of increasing values of the ESLI and increasing patch occupation holds for a broad range of species in the empirical studies. The empirical data do suggest that differences in fragmentation sensitivity between species are sufficiently incorporated in the ESLI. The ESLI can therefore be applied for comparisons between species.

Our prediction rule for metapopulation viability, that was derived from the simulation data, predicts that the metapopulations of five of the species (trunk ant, silver studded blue, tree frog, honeysuckle and bird cherry) used in the empirical analyses are nonviable and that the metapopulation of one species (nuthatch) is on the edge of nonviability.

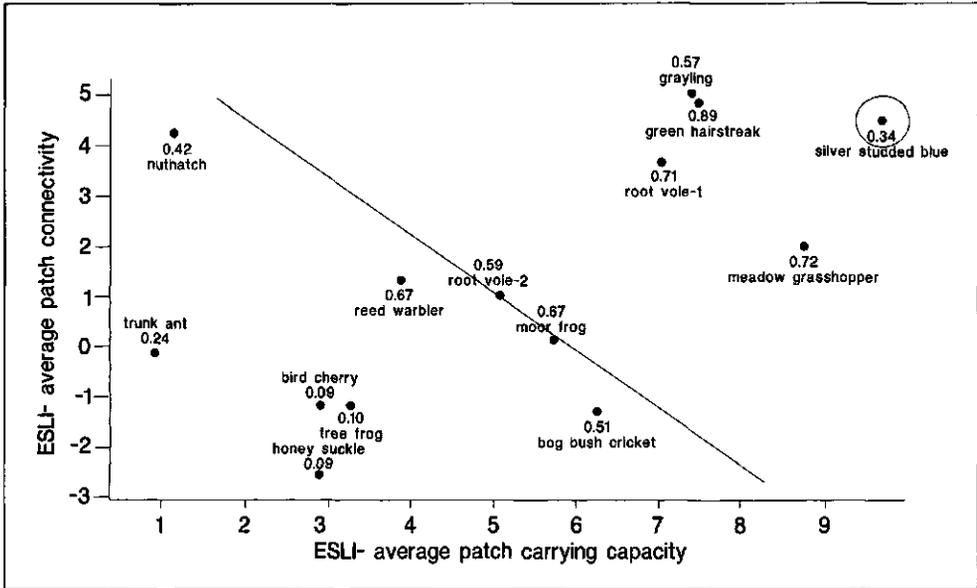


Figure 7.5

Arrangement of the model empirical data according to the (log-transformed) ESLI, for average patch carrying capacity and average patch connectivity, using equations 3 and 5 (see text and Table 7.5). Each point on the graph represents a species-landscape combination and the observed fraction of occupied patches is indicated. The isoline of 0.5 fraction of occupation, the viability threshold, is estimated with logistic regression analysis with the two ESLI as explanatory variables.

## Discussion

### *When do landscape indices have ecological significance?*

Both the empirical data and the model results illustrate that species have different scale-dependent responses to landscape fragmentation. It follows that any landscape index which fails to account for this scale-dependent variation has no ecological significance. These include the neutral measures proposed by among others Franklin and Forman (1987), O'Neill et al. (1988a), Turner (1989), Ripple et al. (1991), McGarrigal and Marks (1995) and Gustafson (1998). Similarly there is no universally applicable viability threshold at a fixed percentage of suitable habitat. Therefore, it is not valid to apply the commonly used landscape index 'fraction of suitable habitat' (Hanski 1994; Andr n 1994, 1996; O'Neill et al. 1988b; Gustafson & Parker 1992; Gustafson 1998) as an universal predictor of metapopulation persistence.

Landscape indices can be indicators for the fraction of occupied habitat, and indirectly for metapopulation viability, but *only* if these indices are ecologically-scaled. Our empirical data show a positive correlation between the fraction of occupied habitat patches (an important predictor of metapopulation viability) and the ecologically-scaled landscape indices (ESLI) proposed in this paper. Therefore,

we conclude that ecological profiles and ESLI are useful tools for integrated fragmentation assessment and for making predictions about the spatial conditions of the landscape for sustainable conservation of biodiversity.

By distinguishing ecological profiles for extinction risk and colonisation ability we have shown that the ecological variation encompassed by species in landscapes can be effectively reduced. We have shown that, using the model species, these ecological profiles can be linked to viability thresholds at different levels of fragmentation. The resulting matrix of ecological profiles, classified according to individual area requirements and dispersal distance, has described relevant elements of this specific scale-dependent fragmentation sensitivity. Andrén (1996) has demonstrated that fragmentation and viability thresholds vary across species and landscapes, using an incidence function metapopulation model (Hanski 1994; Ter Braak et al. 1998) which is spatially explicit but neglects local dynamics.

### **Fraction of occupied patches as a universal predictor for the effect of fragmentation?**

Our model results corroborate the suggestion by Hanski (1995) that the fraction of occupied patches is a useful predictor of metapopulation viability. In nonviable metapopulations (*sensu* Shaffer 1981) the fraction of occupied patches drops below 0.5 with confidence limits of 0.45 - 0.60 (Fig. 7.2). The fraction of occupied patches in a landscape results from interactions between landscape characteristics and species characteristics. One would therefore expect that species characteristics also influence metapopulation viability. However, species characteristics explained little extra variation in the logit regression of the simulation results. We therefore propose the fraction of occupied patches as a universal yardstick for fragmentation effects. The 50% point of occupation might be a universal empirical indicator for a serious conservation threat due to habitat fragmentation. It is important to stress that the landscapes in which species reach this occupation fraction differ widely, and that the effect is the combined result of the interplay of species and landscape characteristics (as expressed in the ESLI).

A direct extrapolation from the relationship between the fraction of occupied patches and the extinction probability in the model simulations suggests that at least 5 of the 14 species in the empirical studies would be considered nonviable. A universal viability threshold linked to the fraction of occupied suitable patches would be a very strong (and useful) tool for conservation planning. Its robustness should therefore be further tested for other combinations of landscape configurations and other ecological profiles. Nevertheless it seems likely that metapopulations with occupied fractions far below the viability threshold of 0.5 are seriously threatened.

There are, however, some qualifications to this general rule. Firstly, the simulation landscapes in this study had patches of equal size. Thus the fraction of occupied patches is strongly correlated with the fraction of occupied habitat. In real landscapes, however, patches vary in size. Large patches tend to be occupied more often than small patches. If a habitat network consists of a very large occupied patch and several small and unoccupied ones, the fraction of occupied patches may be low (one out of many) but the one large population may be viable on its own. In general, variation in patch size will increase metapopulation viability, and the rule of

thumb based on the fraction of occupied patches will become more complex if patches of unequal size are incorporated.

Second, the total number of patches in the simulation landscape (7 to 50 patches) was small. As the probability of chance extinctions increases with decreasing number of occupied patches (Verboom et al. 1991b), simulations should be extended to variable metapopulation size.

Third, the interpretation of occupancy data in empirical studies can lead to overoptimistic viability predictions because of the time lag or extinction debt problem (Nee 1994; Tilman et al. 1994; Harrison & Taylor 1997). Figure 7.3 is based upon the average occupancy over 100 years of simulation, starting from year 51, with the assumption that over this period the metapopulation was in dynamic equilibrium with the landscape. However, real landscapes will not remain the same over such long periods and usually in the past 100 years they have witnessed a progressive fragmentation (Vos & Zonneveld 1993). Therefore, patterns of occupancy may relate more to the landscape of the past than to the landscape of the present. This has been shown for the plant species honeysuckle and bird cherry (Grashof-Bokdam 1997). In a fragmenting landscape with slowly reacting species, the fraction of occupied patches will always be higher than would be the case if the landscape was not changing. This 'ghost of the landscape past' problem is especially likely to occur where turnover rates are low, such as invertebrates in large patches (De Vries 1996) and plants with large generation times (Grashof-Bokdam 1997).

Given the low fraction of occupancy found in our empirical studies, it is remarkable that in only five cases the distribution pattern could be explained by both an area and a connectivity effect in regression analysis. The six studies in which an area effect but no connectivity effect was found need careful evaluation, since we cannot rule out a passive sampling effect as an alternative explanation of the distribution pattern (Verboom et al. 1991a; Andrén 1994). It is possible that a species suffers from fragmentation but for statistical or ecological reasons this is not detectable (e.g. too small field samples, too little variation in the explanatory variables, correlation between habitat quality and fragmentation variables, species interaction). Our results imply that caution must be exercised in drawing conclusions based on a regression analysis of only a one-year distribution pattern. A regression analysis relating extinction and colonisation events to landscape patterns should be more robust (Kareiva & Wennergren 1995; Reich & Grimm 1996).

### **Improving ecological profiles and ESLI**

We consider that developing ecological profiles and ESLI is a valuable step towards establishing general rules for the relationship between biodiversity and landscape pattern. Our framework now needs to be more thoroughly tested, both with more empirical data and more extensive exploration of model species and landscape configurations.

The ESLI levels at the isoline of 0.5 fraction of occupation were different for the model species and the empirical data, and even the six model species yielded different 0.5 isoline levels. This can be explained by the fact that model results such as mean occupancy are extremely sensitive to the exact value of input parameters like mortality and amount of variation (Goodman 1987; Verboom et al. 1993). In real

species, parameters are linked through complex evolution-based relations (e.g. Comins et al. 1980). In our selection of parameter values for the model species, we did not take these relations into account quantitatively; the exact values are rather arbitrary. As a consequence, merely the relative model outcome should be regarded (the performance of one species in different landscapes, or different species in one landscape). Also the relations between model results such as patch occupancy and extinction probability can be compared. The latter was shown to depend little on specific parameter values.

For species, such as insects and to some extent amphibians and reptiles that are very sensitive to environmental stochasticity, even large populations have a real extinction risk (Gilpin & Soulé 1986; Goodman 1987). Sensitivity to environmental stochasticity will have to be incorporated into the ecological profiles. Another aspect that requires incorporation is the mode of locomotion used during dispersal and the interaction with landscape pattern. In ground-dwelling species, the landscape mosaic influences the connectivity (Forman & Godron 1986; Opdam 1990; Merriam 1991; Saunderson & Hobbs 1991; Bennett et al. 1994; Wiens 1997). For these species a correction for matrix resistance should be incorporated in the average connectivity index.

### Implications for nature conservation

The empirical data and model results indicate that general rules for species and landscape characteristics can be developed in an ecologically sensible way. The definition of viability thresholds of indicator groups for fragmentation sensitivity would be an especially valuable tool for maintaining optimal species diversity in natural areas. Although the proposed tools for landscape managers and conservation planners are still in their infancy, we can now give an outline how the indicator groups and ESLI can in principle be applied. Even without exact information of the species distribution, a landscape manager can detect fragmentation problems as follows:

- 1 create as basis a GIS map of habitat types and the interpretation of habitat suitability for characteristic species or target species;
- 2 link the target species to indicator groups for fragmentation sensitivity, based on knowledge about individual area requirements and dispersal distances (see for instance Table 7.5);
- 3 calculate the ESLI of the landscape for each indicator group-suitable habitat combination;
- 4 look up the position of the managed landscape for each indicator group (and corresponding target species) in the matrix of ESLI (for instance for the six indicator groups in Figure 7.3) and determine if the species in the landscape is above or below its viability threshold.

This system also shows potential for landscape design, through determining the domain that is above the viability threshold for relevant species profiles, for instance expressed as average patch size and percentage of habitat coverage (e.g. Table 7.4). Where complete census data are available, the percentage of occupation of suitable habitat patches can be used as an indicator of potential conservation problems of target species due to habitat fragmentation.

## Appendix: the simulation model Metaphor

*Metaphor* is a spatially explicit individual-based model that simulates the dynamics of a metapopulation. A simulation starts with a number of individuals of different age classes and sex categories in a number of patches. The default initiation is with each patch filled to carrying capacity. Mortality, reproduction and dispersal determine the magnitude and structure of subpopulations and thus direct metapopulation dynamics. Carrying capacity is a linear function of patch area, truncated to discrete numbers.

At the start of a year, a census is taken. One could say that this is a pre-breeding census, implying that individuals in the first age class are already almost one year old. The first event in the new year is reproduction. Next, mortality occurs. Then, individuals move to the next age class. Finally, dispersal takes place. Mortality and recruitment are density-dependent as shown below.

Mortality risk is calculated on an annual basis. Mortality probability in patch  $i$  is made up of a background mortality component, affected by environmental variability, and a density-dependent component, depending on local population density (per sex). In this application all patches have equal quality and the environment is equal for all patches in any year: the environmental variation is perfectly correlated for all patches, age classes and both sexes.

Mortality e.g. of males ( $m$ ) in patch  $i$  with e.g.  $N$  male individuals is the result of  $N$  Bernoulli trials with probability  $\mu_{m,i}$ . The result has a Binomial distribution with an expected value of

$$N_{m,i} \cdot \mu_{m,i}$$

The expected individual mortality  $\mu_i$  in patch  $i$  consists of the following components:

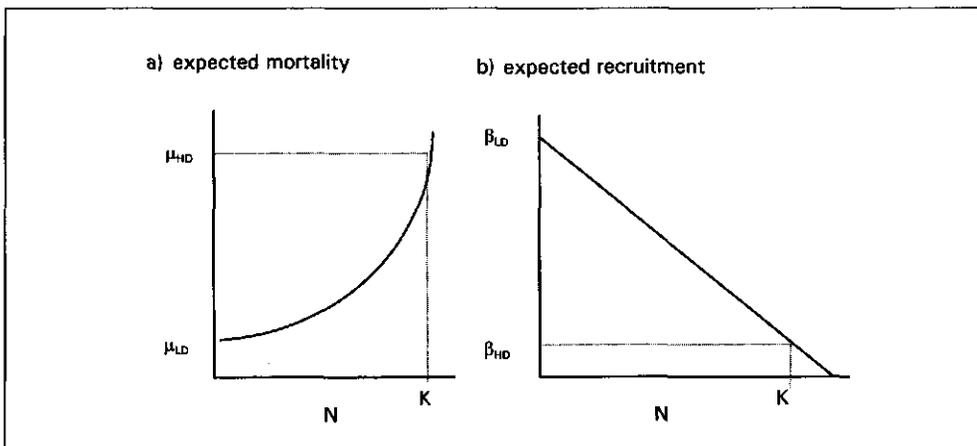


Figure A1

Expected mortality (a) and recruitment (b).  $K$  = carrying capacity;  $\mu_{LD}$ ,  $\mu_{HD}$  = expected mortality at low and high density;  $\beta_{LD}$ ,  $\beta_{HD}$  = expected recruitment at low and high density, respectively. High density refers to carrying capacity.

$$\mu_i = \mu_{dd,i} + \mu_0$$

where  $\mu_0$  refers to the annual background mortality in optimal habitat, in absence of density dependence, while  $\mu_{dd,i}$  refers to the density-dependent component.

$\mu_0$  is a Normally distributed random variable, with mean  $\mu_{LD}$  and standard deviation  $sd_\mu$ . The standard deviation corresponds to the observed natural variation in mortality rate, excluding the impact of demographic stochasticity. It is obtained from a random variable  $\epsilon$ , with a standard Normal distribution, in the following way:

$$\mu_{0(t)} = \mu_{LD} + \epsilon_t \cdot sd_\mu$$

$\mu_{dd,i}$  is the density-dependent component of mortality: the expected extra death rate in patch  $i$  with a density  $N_i$  (per sex), with optimal quality, and in absence of environmental variation. Density-dependent mortality increases exponentially (with coefficient  $z$ ) with the ratio of density  $N_i$  to carrying capacity  $K_i$ :

$$\mu_{dd,i} = (\mu_{HD} - m_{LD}) \cdot \left[ \frac{N_i}{K_i} \right]^z$$

Reproduction parameters in *Metaphor* refer to recruitment instead of birth. Birth is the number of offspring per female, measured (shortly) after the reproductive season. Recruitment indicates offspring that survive their first year of life, or reproduction on a yearly basis. Thus,

$$recruitment = birth \cdot (1 - mortality)$$

Recruitment (male offspring in this example) per reproductive female in patch  $i$  is calculated from the parameters  $\beta_{LD}$  (recruitment at low density) and  $\beta_{HD}$  (recruitment at high density) as follows:

$$\beta_{m,i} = \beta_{LD,m} - (\beta_{LD,m} - \beta_{HD,m}) \cdot \left[ \frac{N_{rf,i}}{K_{fi}} \right]$$

where  $N_{rf,i}$  refers to the number of reproductive females in patch  $i$  and  $K_{fi}$  to the carrying capacity for females. In this application with territorial, monogamous animals, the expected number of 'nests' (females reproducing) is the minimum of number of males, number of females, and carrying capacity (=number of territories).

Recruitment is subject to environmental variability and is influenced both by the annual fluctuation in mortality and the annual fluctuation in reproduction; a year can be a good reproduction year and a low mortality year etc. To incorporate the impact of environmental stochasticity the expected recruitment number is first transformed into birth number (using the equation above). A random variable (Normally-distributed with mean of zero and a standard deviation of  $sd_\beta$ ) is added to the birth number; the result is transformed back to recruitment by multiplication with the survival of that year. Negative numbers are truncated to zero. The final value - recruitment adjusted for environmental noise in both recruitment and

mortality - becomes the parameter of a Poisson distribution from which the number of recruits is obtained.

In *Metaphor*, dispersal takes place once a year after mortality and reproduction. Dispersal in *Metaphor* involves several steps: to determine (1) which (age, sex) individuals leave (emigration), (2) in which direction they move (towards which patch), and (3) what their chances are of arriving alive and settling in a target patch.

The number of emigrants, per age class and sex, leaving patch *i* is obtained from a Binomial distribution with parameters  $E_{i,a}$  and  $N_{i,a}$  (ignoring the indices for male/female). The average (age-specific) individual emigration probability  $E_a$  is calculated for each patch as follows:

$$E_{a(A)} = \begin{cases} \delta_a \cdot \left[ 2 \cdot \sqrt{\frac{A_{mxd}}{A}} - \frac{A_{mxd}}{A} \right] & \text{for } A > A_{mxd} \\ \delta_a & \text{for } A \leq A_{mxd} \end{cases}$$

where *A* refers to the area of the source patch, and  $A_{mxd}$  represents the 'area of maximum dispersal' and  $\delta_a$  is the dispersal probability for age class *a*. It is assumed that, moving from the border of a patch inwards, individual dispersal probability will decrease.  $A_{mxd}$  defines the maximum outer edge area of patches, where dispersal is unhampered. If the total area of a patch is smaller than  $A_{mxd}$ , the entire patch belongs to the outer edge area. When patch area is larger than  $A_{mxd}$ , average probability of emigration decreases with increasing patch area.

In this application of *Metaphor*, a simple dispersal direction algorithm is used in which only the distance to, and the area of, the target patch determine transition probability between patches. More sophisticated methods are available within *Metaphor*. Let  $P_{i,j}$  denote the transition probability between two patches, source patch *i* and target patch *j*. The algorithm is used to produce a matrix of transition probabilities from patch *i* to patch *j*:

$$P_{ij}$$

Survival probability may be associated with the transition from source patch *i* to target patch *j*. This depends on the distance between the two patches  $D_{ij}$  (in metres):

$$e^{-\mu_\delta \cdot D_{ij}}$$

Here,  $\mu_\delta$  denotes the mortality probability ( $m^{-1}$ ) while dispersing.

The probability of selecting patch *j* as a target patch is proportional to the distance between source and target patch  $D_{ij}$ , and the size of the target patch  $A_j$ :

$$P_{ij} = A_j \cdot e^{-\lambda \cdot D_{ij}}$$

The importance of distance is indicated by the value of  $\lambda$ . If  $\lambda$  equals 0, distance has no effect and transition probabilities are directly proportional to target patch size. For large values of  $\lambda$ , all dispersers will move to the nearest patch. Patches are considered to be circles and by calculating the distances the center or border of patch *i* and *j* are used. Transition probabilities are normalized, in order to sum to one:

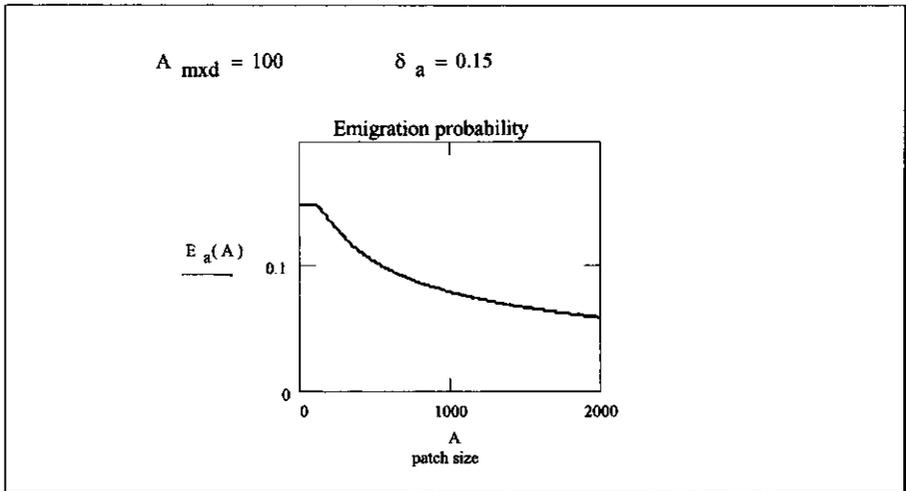


Figure A2  
The emigration probability in relation to patch size.

$$P_{ij} = \frac{A_j \cdot e^{-\lambda \cdot D_{ij}}}{\sum_j A_j \cdot e^{-\lambda \cdot D_{ij}}}$$

Transitions from a patch to itself are not allowed ( $j \neq i$ ). In this application the disperser always settles in the arrival patch, independent of density or the presence of a partner.

In Table A1 the parameter settings of the six model species are listed. The model species modules were derived from calibrated modules for two bird species: European bittern (*Botaurus stellaris*) and reed warbler (*Acrocephalus scirpaceus*). They represent the six ecological profiles (Text, Table 7.1) by incorporating the dispersal characteristics and area requirements.

The simulations were carried out in computer-generated landscapes of 100 km<sup>2</sup>. Patch size was kept constant. The patches were distributed randomly by a landscape generator, keeping a minimal distance between patches of 1.5 times the home range size of the species with the largest area requirements. 14 different landscapes were used for each model species (Table A2).

Table A2  
Number of patches in relation to patch size and habitat percentage in the 14 simulation landscapes.

| HABITAT PERCENTAGE | PATCH SIZE → |      |      |      |       |
|--------------------|--------------|------|------|------|-------|
|                    | 1 ha         | 2 ha | 4 ha | 8 ha | 16 ha |
| ↓ 0.5              | 50           | 25   | 13   | 7    |       |
| 1                  |              | 50   | 25   | 13   | 7     |
| 2                  |              |      | 50   | 25   | 13    |
| 4                  |              |      |      | 50   | 25    |
| 8                  |              |      |      |      | 50    |

Table A1  
Parameter values of the six model species.

| PARAMETER                              | SYMBOL       | A   |  |   | B  |  |  | C  |  |  | D  |  |  | E  |  |  | F  |  |  |
|--|--------------|---|--|---|--|--|--|--|--|--|--|--|--|--|--|--|--|--|--|
|  |              | LARGE IAR<br>SHORT<br>DISPERSAL<br>DISTANCE | LARGE IAR<br>MEDIUM<br>DISPERSAL<br>DISTANCE | LARGE IAR<br>LARGE<br>DISPERSAL<br>DISTANCE | monogamous<br>yes<br>2<br>1 ha<br>0.1<br>0.2<br>0.0<br>4 | monogamous<br>yes<br>2<br>1 ha<br>0.1<br>0.2<br>0.0<br>4 | monogamous<br>yes<br>2<br>1 ha<br>0.1<br>0.2<br>0.0<br>4 | monogamous<br>yes<br>2<br>1 ha<br>0.4<br>0.5<br>0.1<br>4 |  |
| sex structure                          |              |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| territoriality                         |              |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| number of age classes                  |              |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| area of maximum dispersal              | $A_{maxd}$   |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| mortality at low density               | $\mu_{LD}$   |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| mortality at carrying capacity         | $\mu_{HD}$   |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| standard deviation of mortality        | $sd_{\mu}$   |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| exponent of density dependence         | $z$          |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| maximum mortality allowed              |              |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| recruitment at low density             | $\beta_{LD}$ |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| recruitment at carrying capacity       | $\beta_{HD}$ |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| standard deviation of recruitment      | $sd_{\beta}$ |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| dispersal probability (juveniles)      | $\delta_j$   |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| dispersal probability (adults)         | $\delta_a$   |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| number of territories per ha           |              |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| dispersal distance dependence          | $\lambda$    |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| dispersal mortality (m <sup>-1</sup> ) | $\mu$        |   |  |   |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |

## 8 General discussion and synthesis

### Habitat fragmentation and the metapopulation approach

Can the conclusion be drawn that the metapopulation approach is useful to explain the distribution pattern of the species studied, the tree frog and the moor frog? In other words, do the results justify a landscape or regional approach in conservation planning and the application of metapopulation concepts for recommendations on optimal spatial habitat design? There are different aspects to this question that I will discuss separately for the two species in the following sections. However, in short the metapopulation approach seems to be a useful approach for the tree frog, whereas the picture of the moor frog in the studied habitat configuration is less clear.

#### Tree frog

In chapter 1, I identified several research phases that ideally should be followed to infer from empirical data that a species in a particular habitat network functions as a metapopulation. The results seem to justify the conclusion that the tree frog in Zeeland Flanders does function as a metapopulation. In the study on the tree frog both pattern and process studies were conducted. In the pattern analysis of chapter 2, the occurrence of apparently suitable empty patches and the fact that the occupation probability increased with connectivity are indications that the distribution pattern is affected by habitat fragmentation. In chapter 5, I analysed the processes of local extinction and recolonisation, the underlying metapopulation dynamics. Extinctions took place regularly and were influenced by spatial features of the landscape. Pond size, encompassing both pond area and suitable terrestrial habitat in a radius of 250 m around the pond, was negatively correlated with the extinction probability of a local population. This is in accordance with the metapopulation theory, if one assumes that small patches can sustain only small populations that can go extinct by chance processes of demographic stochasticity (Levins 1970; Richter-Dyn & Goel 1972). Empty patches were recolonised and this process was influenced by the configuration of habitat. The colonisation probability of a pond increased with increasing connectivity. This is in accordance with the metapopulation theory, which predicts that colonisation probability increases with the number of potential sources of dispersers (Verboom et al. 1991a; Hanski 1994). The observed dispersal distances in comparison with the distances between patches and the observed recolonisations, as shown in chapter 5, demonstrate that the habitat network is still connected by dispersing individuals.

To answer the question if the tree frog metapopulation is still viable I conducted model simulations. In chapter 1, I defined a viable metapopulation as: *'a set of local populations distributed over a spatially discontinuous habitat network and linked by dispersal, the size of which as well as the balance between local extinction and recolonisation are such that its probability of extinction within 100 years is 5% or less'*. In this thesis I used two metapopulation models: *Wink* (Verboom et al. 1991a; Hanski 1994; Ter Braak et al. 1998) in chapter 5 and *Metaphor* (Verboom

1996; Verboom et al. 1999) in chapter 7. *Wink*, an incidence function model, or 'winking patch' model describes metapopulation dynamics on a local population level, with patches being either empty or occupied. Turnover estimates are based on the observed probabilities of extinction and recolonisation. An advantage of this method is that all model parameters can be estimated directly from field observations, as I did in chapter 5. The drawback of this method is that only long time series, which incorporate the range of possible fluctuations of population numbers between years, render reliable predictions. The metapopulation model *Metaphor* is a mechanistic model based on processes on an individual level. The advantage of this method is that relevant processes in a metapopulation context such as local population dynamics and dispersal are described explicitly. However, this renders many parameters and requires profound autecological knowledge of the species. The two models yielded different predictions of the metapopulation viability. *Wink* predicted that the metapopulation was still viable. I consider the predictions of *Wink* to be overoptimistic; a three-year data set is not enough to incorporate all variation between good and bad years. The predictions of viability with *Metaphor* were based on imaginary model species that represented different ecological profiles for fragmentation sensitivity. Results indicated that species with a fraction of occupied patches below 0.5 were below the viability threshold. The tree frog with a fraction of 0.1 occupied patches is far below this threshold, and would therefore be not viable. A second drawback that holds for both modelling approaches is that the simulation landscape is held constant during the simulation period, while realistic landscapes change considerably during a period of 100 years. Habitat destruction and the loss of suitable habitat by natural succession will cause deterministic extinctions. The destruction of suitable habitat forms an extra threat to metapopulation viability. For species in dynamic habitat networks, the incorporation of habitat succession (Lindenmayer & Possingham 1995) and changes in habitat distribution over time would improve the predictive value of both models.

### The moor frog

In the study on the moor frog I conducted a first step to detect effects of fragmentation by analysing a single-year distribution pattern in chapter 3. However, as no information is available on extinction and recolonisation processes related to landscape structure, empirical evidence whether the species functions as a metapopulation is incomplete.

The analysis of the distribution pattern showed a positive effect of pond size on the probability of occupation and the abundance of moor frogs which would be expected in fragmented landscapes. I could demonstrate no unambiguous effects of connectivity, described as the amount of suitable habitat in the surroundings of a pond. As I pointed out in chapter 3, habitat connectivity variables and habitat quality variables were highly correlated and therefore it was not possible to choose between these effects. This was a result of confounding variables in the study area, where high habitat quality coincided with high connectivity. To clearly distinguish between connectivity and quality the study should be repeated in a landscape with a different spatial configuration of habitat. However, the fact that road density in addition to habitat quality, lowered occupation probability (chapter 3) does imply

effects of isolation. If potential colonisers are killed on roads, the colonisation probability of a patch with a high road density in its surroundings is reduced. Still a lowered occupation probability in patches could also be caused by an increased extinction probability as a result of extra mortality. It is not possible to distinguish between these two factors, based on a pattern study. This illustrates clearly that a study based on an analysis of patterns only, at best can provide a first indication of fragmentation effects. A time series and spatial analysis of local extinctions and recolonisations yield much stronger evidence whether a species functions as a metapopulation.

One of the unanswered questions is whether dispersal between local populations still occurs or that the moorland fragmentation has progressed to a stage where (clusters of) local populations have become completely isolated. I found a slight decrease in genetic similarity with geographical distance in chapter 4. This is an indication for some gene flow but does not answer this question satisfactorily. The genetic similarity between populations is still very high. The fragmentation in the study area, that consisted of continuous moorland with scattered moorland ponds until the 1930s, is probably too recent to affect population structure. Still the fact that distance weighted for (rail)road density, which is a feature of the present landscape only, slightly improved the correlation is an indication that some exchange between local populations takes place and that this is hampered by (rail)roads. The monitoring of actual dispersal events with capture-mark-recapture techniques or by following gene flow of (unique) alleles would further underpin the spatial range at which the species operates. An alternative approach to quantify dispersal would be the monitoring of colonisation of newly created habitat at different distances from existing populations.

## Differences between tree frog and moor frog

When comparing amphibian species with the total range of variation between species, both the tree frog and the moor frog belong to the same 'ecological profile' (chapter 7) of relatively immobile species with small individual area requirements. However, when looking more in detail there are some differences between the species that affect their sensitivity to habitat fragmentation. Probably the most important differences in life history traits between moor frog and tree frog are caused by their choice of habitat. The tree frog is a species of early succession stages in dynamic habitat (Creemers 1991), while the moor frog is a species of relatively stable habitat (Hartung 1991). An important part of the natural habitat of the tree frog is situated along river flood plains (Creemers 1991). Flooding can make an area unsuitable for years but can at the same time create new habitat at another location. So the network of suitable habitat patches is dynamic both in space and time. A species in unpredictable habitat must have strong colonisation ability. The relative large dispersal distances of the tree frog and the loud mating calls could be an adaptation to the survival in unpredictable habitat. The habitat of the moor frog consists of nutrient-poor habitats, often on peat or sandy soils with impermeable layers or high groundwater tables (Podloucky 1987; Wijnands 1987; Hartung 1991). As these systems are more stable, a high population persistence probably is a more

effective strategy for long-term survival than being a good coloniser. This is illustrated by the special adjustments of the species to nutrient-poor conditions such as its high tolerance for acid water conditions (Hartung 1991). Although, to my knowledge, quantitative studies on dispersal of the moor frog are lacking, reported dispersal distances of the moor frog are smaller compared to the tree frog. In this respect the very soft mating call also suggests no special adaptations to improve its colonisation ability.

Based on these differences one would expect that the tree frog is better adapted to fragmentation of its habitat than the moor frog. This is illustrated by the presence of the tree frog in a more fragmented habitat situation compared to the moor frog. The habitat of the tree frog in Zeeland Flanders consists of small remnants in an agricultural landscape with a total cover of 1.4%, where even within one local population aquatic and terrestrial habitat parts are spatially separated (chapter 2). The turnover of suitable habitat as a result of agricultural activities causes additional stress. On the other hand the studied moor frog landscape consists of 11.2% suitable habitat that is situated in larger habitat patches and aquatic and terrestrial habitats are mostly adjacent (chapter 3). As the habitat is situated in conservation areas, there is no turnover of habitat and succession in nutrient-poor conditions is slow. However, taking patch occupancy as an indication for (meta)population viability, the tree frog is probably not viable with only 10% of the habitat occupied (chapter 7). Even for species that are less sensitive to fragmentation, the degree and speed of fragmentation in the Dutch agricultural landscapes may have exceeded a critical threshold. High resistance of the landscape between ponds as a factor that decreases successful dispersal within the habitat network, has an additional negative impact on survival. Based on the fraction of 0.67 occupied patches, the moor frog seems to be still above the viability threshold of 0.5 fraction of occupation (chapter 7). If the turnover rate of moor frog populations is very low, the present distribution may be still linked to the former continuous landscape. In this respect it is essential to know if recolonisations are still possible. If dispersal between moorlands is no longer possible, the viability of the pond clusters within separate moorlands should be the management focus point.

## Research strategy and implications for landscape management

Although the conclusions are mainly based on experiences with amphibian research, I pose that these conclusions also hold for research on the effect of habitat fragmentation in general.

### Delimitation of local populations

A complication is encountered when studying the distribution of species that require different types of habitat, within their life cycle. This is especially the case for amphibians in agricultural landscapes where terrestrial and aquatic habitat parts are often separated by agricultural fields. This makes the exact delimitation between habitat patches impossible. In this thesis every pond was regarded as a separate

potential site for a local population (except for ponds in the same moorland within 200 m from each other). In most cases the area of the local population was based on the aquatic habitat part. In chapter 5 the amount of terrestrial habitat in a radius of 250 m was incorporated. In general, the aquatic habitat part is probably a good estimate for population size, as the size of the reproduction site is thought to be the limiting factor in amphibian populations (Wilbur 1987; John-Alder & Morin 1990). However, when the terrestrial habitat is scarce and highly fragmented, as is the case in the study area of the tree frog, population size is probably also determined by the amount of terrestrial habitat. The delimitation of local populations is to some extent arbitrary (Harrison & Taylor 1997). There will be a continuum between distance and independent dynamics of local populations. If both terrestrial and aquatic habitats are highly fragmented and spatially separated from each other, the common approach of suitable 'habitat islands' in a sea of inhospitable habitat (Levins 1970; Gilpin & Hanski 1991; Opdam et al. 1993; Hanski & Gilpin 1997) is less adequate. A landscape approach taking the whole landscape mosaic of habitat types into account would be a more suitable method of addressing this problem. Also, when considering the large impact of landscape resistance on connectivity, a landscape approach seems appropriate. In landscape ecology the necessity of a landscape approach receives a growing interest (Wiens 1997; Bennett 1999).

### How to measure connectivity?

Connectivity is determined by the complex of landscape characteristics that influence dispersal and therefore the amount of exchange between habitat patches. In chapter 6, I demonstrated that the landscape mosaic does influence the movement patterns of tree frogs. Displaced tree frogs showed a preference for hedgerows and an avoidance of arable land, while pastures took an intermediate position. The calibration of the movement model *SmallSteps* showed the best fit between observed and simulated movement paths, when transition probabilities were strongly biased towards hedgerows. The predicted distribution patterns of this strict 'corridor' frog type was strongly skewed in favour of hedgerows. Based on these results I concluded that the resistance of the landscape mosaic between habitat patches should be incorporated to determine connectivity for tree frogs and that hedgerows function as 'relative corridors' in agricultural landscapes.

In this thesis, a growing complexity in the measurement of connectivity can be observed, with the simple linear variable 'distance to the nearest pond' (chapter 2) on the one hand and the complex prediction of dispersal with *SmallSteps* on the other. In between, connectivity was expressed as the number of patches within a certain distance (chapters 2 and 3), which has the limitation that all patches within the circle contribute equally to the connectivity, regardless of their actual distance. In subsequent chapters, a connectivity measure was applied that was based on an estimation of the number of migrants arriving in a pond, taking into account the distance and size of dispersal sources (chapters 5 and 7; Verboom et al. 1991a; Hanski 1994). As a first attempt to incorporate landscape resistance into this connectivity measure in chapter 5, the barrier effect of roads was incorporated, while in chapter 4 an attempt was made to correct distance between ponds for the relative resistance of the landscape between ponds.

The increasing complexity is instigated by growing insight that connectivity is a key factor in metapopulation dynamics and a general trend towards spatial explicitness (Wiens 1997; Opdam et al. submitted). In addition, the simulation models revealed that predictions are very sensitive to assumptions concerning the dispersal process, so more spatially realistic measures were required. Emphasis became more heavily on connectivity as the reduced connectivity seems to be the bottleneck in our highly human-dominated landscapes, at least for small-sized species. Small species do not need extensive areas to maintain local populations, although they tend to be badly buffered against environmental variation, but distances between habitat patches become a problem quite soon. For ground-dwelling species, an extra motivation to concentrate on connectivity is that in our landscapes with sharp boundaries between habitat types, the large variation in permeability and the presence of barriers cannot be ignored in realistic estimates of connectivity. The trend towards more complex spatially explicit connectivity measures asked for a better understanding of the underlying processes of the interaction between moving organisms and landscape heterogeneity. This is necessary to test the realism of the numerous assumptions that are made in these measures. Field experiments were needed in which actual movement patterns in heterogeneous landscapes are registered and mechanistic movement models that simulate the dispersal process, such as *SmallSteps* (chapter 6). A movement model is required to extrapolate individual decisions on a local level to exchange between populations on a landscape scale. The field experiments in combination with observed dispersal events in interaction with simulation models are essential prerequisites to test assumptions on how dispersal is governed in complex landscapes. Movement models can be applied on a species level when detailed spatially explicit recommendations on optimal corridors are required. However, the model also forms the basis for the underpinning of more general corrections for landscape resistance in connectivity.

### **Implications for landscape management**

The results of this study can contribute to various stages in nature conservation policy. I developed tools that can help to reveal fragmentation problems in an early phase of problem detection. The spatial standards for sustainable habitat networks can be applied in landscape evaluation as well as landscape design.

I used several types of predictive model: regression models to predict the probability of patch occupancy, simulation models to predict metapopulation viability, and a movement model that focuses on the aspect of connectivity in heterogeneous landscapes. These models are necessary to extrapolate in time and space. As nature conservation is aiming at the conservation of species in the long run, recommendations are necessarily based on predictions about survival probability.

Based on the regression models developed in chapter 2, the occupation probability of a pond can be predicted, given the amount of suitable terrestrial habitat and the number of ponds nearby. Using these predictions, guidelines for the design of an 'optimal tree frog landscape' can be developed and different landscape planning scenarios can be compared (Vos 1993; Grashof 1997). As regression models

are based on correlations in a particular landscape, one should be reluctant with extrapolation to other areas, as this may require some adjustments to landscape specific characteristics. Another limitation of regression models is that no predictions can be made about the viability of the metapopulation as a whole. The negative impact of road density on the probability of occupation of a pond by moor frogs holds relevant information for nature conservation policy. The regression model in chapter 3 predicted an occupation probability of less than 30% in the part of the study area adjacent to a motorway. In landscape design, these spatial differences in road density should be incorporated in the selection of optimal locations for nature protection areas.

I derived a general rule for the spatial requirements of amphibian habitat networks in chapter 3 from comparison of amphibian studies on several species and in different landscapes. As the mean distance between occupied patches was below 1 km in all studies, this was proposed as a general rule of thumb for amphibian populations.

Metapopulation models are needed to predict if a population is still viable within the habitat network of a landscape. The application of simulation models on a species-specific level in realistic landscapes was illustrated in chapter 5, where I applied the *Wink* model to predict viability of the tree frog population in Zeeland Flanders. The ecologically-scaled landscape indices (ESLI) from chapter 7 are an example how a metapopulation model (*Metaphor*), in combination with empirical data, can render general principles for habitat network requirements. Both the empirical data and the model results illustrated that species have different scale-dependent responses to landscape fragmentation. The ecological profiles that describe sensitivity for local extinction and colonisation ability seem to capture relevant differences between species concerning sensitivity for habitat fragmentation. When translating ecological profiles for species groups into ESLI, a tool is developed to detect fragmentation problems or to design landscapes for optimal species diversity. Also the strong correlation between the fraction of occupied patches and viability, predicting viability in metapopulations if 50% or more of the patches are occupied, is a yardstick to detect fragmentation problems if the distribution of a species is known. Although the proposed ESLI are still in their infancy and should be further tested, they are in potential a powerful tool for more global species richness assessments.

For many species (groups) basic knowledge of dispersal behaviour, dispersal distances and the influence of landscape heterogeneity on dispersal direction is incomplete or lacking totally. The continuation of movement studies on species behaviour in heterogeneous landscapes can identify which species are sensitive to barriers and what kind of corridors might be effective, for a broader range of species. As it is impossible to study all species separately, there is a great need for the translation of knowledge towards general principles for corridor requirements. More effort should be put in determining the key characteristics of species that predict corridor effectiveness in landscapes with different degrees of habitat fragmentation and in the grouping of species with comparable corridor requirements. The movement model *SmallSteps* can play a role to further underpin standards for optimal corridor design.

## Summary

In the beginning of the nineties, the National Ecological Network (NEN) has been devised, to put a halt to the continuing process of habitat fragmentation. This study contributes to the underpinning of the spatial requirements for sustainable habitat networks of amphibian populations. Amphibian species are representatives of relatively immobile ground-dwelling species. The central question in this study is under what conditions species can survive in landscapes in which the habitat is fragmented. Results are translated into spatial standards that can be applied in landscape management.

Fragmentation is the result of the destruction of habitat suitable for a species, resulting in small fragments or patches that are separated by unsuitable habitat or barriers. Metapopulation theory implies that although small populations suffer from chance extinction due to demographic stochasticity, a species can survive on a regional level if local extinctions are compensated for by recolonisations. Connectivity between habitat patches depends on the landscape characteristics that determine the amount of individuals that are capable to disperse between patches. For ground-dwelling species with low dispersal capacity, such as amphibians, connectivity not only depends on the distance between suitable habitat patches but also on the relative resistance of the landscape. In landscapes with moderately fragmented habitat, the distribution of species is governed by the density and spatial configuration of habitat. Characteristically species whose distribution is affected by fragmentation are often absent in relatively small habitat patches with low connectivity. If habitat destruction continues, distances between local populations can no longer be bridged by dispersing individuals and populations become completely isolated. As extinctions can no longer be compensated for by recolonisation, the species is threatened with extinction on a regional level. For nature conservation the important question is whether species are still capable to cope with habitat destruction or already have crossed the viability threshold.

The study is focused on two amphibian species: the tree frog (*Hyla arborea*) and the moor frog (*Rana arvalis*). The two species differ regarding the degree of fragmentation of their habitat. The habitat of the tree frog consists of remnants of (semi)natural vegetation in agricultural landscapes. This habitat is highly fragmented and the 'turnover' of habitat is high. The habitat of the moor frog consists of moorlands with ponds situated in conservation areas and is stable and less fragmented.

In chapter 2 the distribution pattern of the tree frog in Zeeland Flanders is analysed, as a first indication whether the species shows negative effects of habitat fragmentation. The occurrence of apparently suitable but unoccupied patches and the fact that the occupation probability increases with higher connectivity are indications that the distribution pattern is affected by habitat fragmentation. The occupation probability of a pond increases with pond density within 750 m radius of the occupied pond. Additionally, both the density of shrubs and that of tall herbs, two terrestrial habitat factors, are higher within 1000 m of occupied ponds. Contrarily to what is expected, no correlation is found between pond size and probability of occupation.

In chapter 3 the distribution pattern of the moor frog in south-west Drenthe is

analysed. It shows a positive effect of pond size on the probability of occupation and the abundance of moor frogs, which is an indication that the distribution pattern is affected by habitat fragmentation. No unambiguous effects of connectivity, described as the amount of suitable habitat in the surroundings of a pond, could be demonstrated. This is because in the study area high habitat quality coincides with high connectivity. However, the fact that road density in addition to habitat quality, lowers occupation probability does imply effects of isolation. If potential colonisers are killed on roads, the colonisation probability of a patch with a high road density in its surroundings is reduced.

The role of dispersal in connecting the moor frog habitat network is further investigated by applying genetic techniques in chapter 4. Genetic similarity between populations is studied, as an indirect indicator of successful dispersal between populations in the present and past landscape configuration. The degree of population subdivision, based on five microsatellite loci, is relatively low ( $F_{st} = 0.052$ ), possibly due to the limited number of generations, since cultivation of the formerly continuous moorland in the 1930s. Still, genetic distances between populations correlate positively with geographical distances between moorland ponds. When geographical distance is weighted for (rail)road density, this slightly improves the correlation. This is an indication that some exchange between local populations takes place and that the connectivity between ponds is reduced by (rail)roads.

To test the assumption that metapopulation processes play a dominant role in the distribution of the tree frog in Zeeland Flanders, in chapter 5, a time series of distribution patterns and observed dispersal events of the tree frog are analysed. The results seem to justify the conclusion that the tree frog in the particular study area functions as a metapopulation.

- 1 Extinctions take place regularly and are influenced by spatial features of the landscape: extinction probability decreases with patch size.
- 2 Empty patches are recolonised and this process is influenced by the configuration of habitat: colonisation probability increases for large patches with high connectivity.
- 3 The observed dispersal distances, in comparison with the distances between patches and the observed recolonisations, show that the habitat network is still connected by dispersing individuals.

To answer the question if the tree frog metapopulation is viable, simulations are conducted with *Wink*, an incidence function model, or 'winking patch' model. Although the model predicts that the tree frog metapopulation is still viable, it is pointed out that several aspects that will influence viability negatively, are not incorporated in the model such as the high turnover of suitable habitat by habitat destruction and natural succession.

Chapter 6 further focuses on connectivity. Especially in highly human dominated landscapes not only distance but also landscape resistance may have a large impact on connectivity. Radio-tracking experiments with displaced tree frogs show a preference for hedgerows and an avoidance of arable land. The calibration of the movement model *SmallSteps* shows the best fit between observed and simulated movement paths, when transition probabilities are strongly biased towards hedgerows. It is concluded that the resistance of the landscape mosaic is an

important element to determine connectivity for the tree frog and that hedgerows may function as corridors that will enhance dispersal.

Landscape managers and nature conservation planners need tools that link landscape characteristics to some critical measure of the sustainable conservation of species diversity. In chapter 7 a general framework is developed to predict viability for a broad array of species. The approach is based on a combination of field studies in fragmented landscapes and model simulations in artificial landscapes. The concept of ecological profiles is developed to group species according to characteristics that are important in responses to landscape fragmentation: individual area requirements as the dominant characteristic of extinction risk in habitat patches, and dispersal distance as the main determinant of the ability of species to colonise patches. The ecological profiles are transformed into two ecologically-scaled landscape indices (ESLI): 'average patch carrying capacity' and 'average patch connectivity'. Simulations with model species, representing different ecological profiles, indicate that the fraction of occupied patches is a useful predictor of metapopulation viability. In nonviable metapopulations the fraction of occupied patches drops below 0.5 (confidence limits 0.4 - 0.6). Both the empirical data and the simulations with different ecological profiles illustrate that species have different scale-dependent responses to habitat fragmentation. The empirical data show a positive correlation between the fraction of occupied habitat patches (an important predictor of metapopulation viability) and the ESLI. The conclusion is that ESLI are useful tools for integrated fragmentation assessment.

In the general discussion of chapter 8 it is discussed whether the metapopulation approach is a useful approach and justifies a landscape or regional approach in conservation planning. Based on the results in chapters 2 and 5 it is concluded that a metapopulation approach seems to be a useful approach for the tree frog. For the moor frog, empirical evidence for a metapopulation is still incomplete. Only a first step to detect effects of fragmentation, by analysing a single-year distribution pattern, is conducted. However, no information is available on extinction and recolonisation processes related to landscape structure. One of the unanswered questions is whether dispersal between local populations still occurs or that the moorland fragmentation has progressed to a stage where (clusters of) local populations have become completely isolated. Comparing the two species it is concluded that the species differ in their sensitivity to habitat fragmentation. The tree frog, a species of unpredictable habitat, is better adjusted to habitat fragmentation. However, the tree frog is probably no longer viable in Zeeland Flanders and the degree and speed of fragmentation in the Dutch agricultural landscapes may have exceeded a critical threshold. The habitat of the moor frog is less fragmented and the species seems to be still viable. However, the present distribution may be still linked to the former continuous landscape. In this respect it is essential to know if recolonisations are still possible.

Subsequently the different methods by what connectivity is measured in this study are discussed. The trend throughout the subsequent chapters towards more complex spatially explicit connectivity measures is instigated by a growing insight that connectivity is a key factor in metapopulations. For ground-dwelling species the large variation in permeability of highly human dominated landscapes is an extra motivation to focus on spatially explicit connectivity.

The last section focuses on implications for landscape management. Several tools and their potentials for application are discussed. Statistical models give guidelines for the design of optimal amphibian landscapes. Simulation models render standards for viable population networks and optimal corridor design that can play a role in landscape evaluation and the comparison of planning scenarios.

## Samenvatting

In het begin van de jaren negentig is de Ecologische Hoofdstructuur (EHS) ontworpen om een halt toe te roepen aan het voortschrijdende proces van habitatfragmentatie (versnippering). Deze studie draagt bij aan de onderbouwing van de ruimtelijke voorwaarden voor duurzame netwerken van amfibiepopulaties. Amfibieën zijn representatief voor relatief immobiele grondgebonden soorten. De centrale vraag in deze studie is onder welke omstandigheden soorten kunnen overleven in landschappen met versnipperd leefgebied. De resultaten worden vertaald in ruimtelijke normen, die kunnen worden toegepast in de ruimtelijke planvorming.

Versnippering is het resultaat van de vernietiging van het habitat van een soort, resulterend in kleine habitatfragmenten, die van elkaar gescheiden zijn door ongeschikt habitat of barrières. Kleine populaties hebben een kans om uit te sterven als gevolg van demografische stochasticiteit. Volgens de metapopulatietheorie kan een soort overleven op regionaal niveau, wanneer lokale extinctions worden gecompenseerd door rekolonisaties. De connectiviteit tussen habitatplekken hangt af van die eigenschappen van het landschap die bepalend zijn voor de mate van uitwisseling van individuen tussen habitatplekken. Voor grondgebonden soorten met een beperkte dispersiecapaciteit, zoals amfibieën, hangt de connectiviteit niet alleen af van de afstand tussen leefgebieden maar ook van de relatieve weerstand van het landschap. In matig versnipperde landschappen wordt de distributie van soorten bepaald door de dichtheid en ruimtelijke configuratie van habitat. Kenmerkend voor soorten die te lijden hebben van versnippering, is dat zij vaak ontbreken in de relatief kleine en geïsoleerde habitatplekken. Wanneer de versnippering voortschrijdt, kunnen dispergerende individuen de afstanden tussen leefgebieden niet langer overbruggen en raken populaties volledig geïsoleerd. Omdat extinctions niet langer worden gecompenseerd door kolonisaties, wordt de soort bedreigd met regionale extinctie. Een belangrijke vraag voor het natuurbeleid is of een soort de mate van versnippering van haar leefgebied aankan of dat de drempel van duurzaamheid reeds is overschreden.

De studie richt zich op twee amfibiesoorten: de boomkikker (*Hyla arborea*) en de heikikker (*Rana arvalis*). De mate van versnippering van het habitat van de twee soorten is verschillend. Het habitat van de boomkikker bestaat uit resten (semi)natuurlijke vegetatie in agrarische landschappen. Dit habitat is sterk versnipperd en heeft een hoog verloop. Het habitat van de heikikker is minder versnipperd en meer stabiel. Het is gesitueerd in natuurgebieden en bestaat uit heide met vennen.

Als eerste indicatie voor een effect van versnippering wordt in hoofdstuk 2 het verspreidingspatroon van de boomkikker in Zeeuws-Vlaanderen geanalyseerd. Het feit dat de soort afwezig is in ogenschijnlijk geschikte plekken en dat de kans op voorkomen toeneemt met hogere connectiviteit, zijn indicaties dat de soort te lijden heeft van versnippering. De kans op voorkomen neemt toe met de poelendichtheid in een straal van 750 m rond de poel. Daarnaast is de dichtheid van struiken en ruigte, twee componenten van het terrestrische habitat, hoger in een straal van 1000 m rond bezette poelen. In tegenstelling tot de verwachting wordt geen correlatie gevonden tussen de grootte van de poel en de kans op bezetting.

In hoofdstuk 3 wordt het verspreidingspatroon van de heikikker in het zuidwesten van Drenthe geanalyseerd. Er bestaat een positief effect tussen de grootte van

het ven en de kans op voorkomen en dichtheid van heikikkers. Dit is een indicatie dat het verpaidinspatroon van de heikikker negatief wordt beïnvloed door versnippering. Er zijn geen ondubbelzinnige effecten van connectiviteit gevonden, beschreven als de hoeveelheid geschikt habitat in de omgeving van een ven. Dit wordt veroorzaakt doordat in het studiegebied een hoge habitatkwaliteit en een hoge connectiviteit samen voorkomen. Het feit dat de kans op voorkomen van heikikkers afneemt bij een hoge wegendichtheid, is een aanwijzing voor effecten van isolatie. De kolonisatiekans van een ven met een hoge wegendichtheid in de omgeving neemt af wanneer potentiële dispersers worden doodgereden.

In hoofdstuk 4 is de rol van dispersie in het verbinden van het habitatnetwerk van de heikikker verder onderzocht met behulp van genetische technieken. De genetische similariteit tussen populaties is geanalyseerd als indirecte indicator voor succesvolle dispersie tussen populaties in de huidige en historische configuratie van het landschap. De genetische verschillen tussen de populaties, gebaseerd op vijf microsatellieten, blijkt gering te zijn ( $F_{st} = 0.052$ ). Dit is waarschijnlijk een gevolg van het gering aantal generaties sinds de heide in de jaren dertig werd gecultiveerd. Toch blijkt de genetische afstand tussen populaties gecorreleerd te zijn aan de geografische afstand tussen vennen. Deze correlatie wordt sterker wanneer de geografische afstand wordt gewogen voor de dichtheid aan (spoor)wegen. Dit is een indicatie dat enige uitwisseling tussen populaties plaatsvindt en dat wegen de connectiviteit tussen vennen reduceren.

In hoofdstuk 5 wordt nagegaan of het aannemelijk is dat metapopulatieprocessen een dominante rol spelen in de verspreiding van de boomkikker in Zeeuws-Vlaanderen. Hiervoor worden een tijdreeks van verspreidingsgegevens en waargenomen dispersie tussen poelen geanalyseerd. De resultaten lijken de conclusie te rechtvaardigen dat de boomkikker in het desbetreffende gebied functioneert als een metapopulatie.

- 1 Extinctie vindt regelmatig plaats en wordt beïnvloed door de ruimtelijke configuratie van het landschap: de kans op extinctie neemt toe met de grootte van de plek.
- 2 Lege plekken worden gekoloniseerd en dit proces wordt beïnvloed door de configuratie van habitat: de kans op kolonisatie neemt toe voor grote plekken met een hoge connectiviteit.
- 3 De geobserveerde dispersie in relatie tot de afstanden tussen leefgebieden en de geobserveerde kolonisaties, laten zien dat het habitat netwerk nog steeds verbonden is.

Om de vraag te beantwoorden of de metapopulatie van de boomkikker duurzaam is, zijn simulaties uitgevoerd met *Wink*, een model waarin plekken leeg of bezet kunnen zijn. Het model voorspelt dat de metapopulatie duurzaam is. Er wordt echter op gewezen dat verschillende aspecten die de duurzaamheid negatief zullen beïnvloeden niet zijn opgenomen in het model, zoals het hoge verloop in geschikt habitat als gevolg van habitat destructie en natuurlijke successie.

Hoofdstuk 6 is gericht op connectiviteit. Met name in sterk gecultiveerde landschappen zal niet alleen de afstand maar ook de weerstand van het landschap bepalend zijn voor de connectiviteit. Zenderexperimenten met verplaatste boomkikkers tonen een voorkeur voor houtwallen aan en het vermijden van akkers. Aan het einde van een experiment werden de boomkikkers relatief vaak in houtwallen aan-

getroffen. De calibratie van het bewegingsmodel *SmallSteps* geeft de beste overeenkomst tussen geobserveerde en gesimuleerde routes, wanneer de overgangskansen tussen landschapstypen sterk in het voordeel van houtwallen zijn. De conclusie wordt getrokken dat het landschapsmozaïek een belangrijk element is bij het bepalen van de connectiviteit voor boomkickers. Houtwallen kunnen de dispersie bevorderen en hebben daarmee een corridorfunctie in het agrarisch gebied.

Natuurbeheerders en landschapsplanners hebben behoefte aan instrumenten, waarmee landschapspatronen kunnen worden verbonden aan een kritische maat voor het duurzaam behoud van biodiversiteit. In hoofdstuk 7 is een algemeen raamwerk ontwikkeld waarmee de duurzaamheid kan worden voorspeld voor zeer diverse soorten. De methode is gebaseerd op een combinatie van veldstudies in versnipperde landschappen en modelsimulaties in kunstmatige landschappen. Het concept 'ecologische profielen' wordt geïntroduceerd, waarin soorten worden gegroepeerd volgens eigenschappen die belangrijk zijn voor de respons op versnippering. De individuele oppervlaktebehoefte van een soort is de dominante eigenschap die de gevoeligheid voor extinctie bepaalt, terwijl dispersieafstand de belangrijkste factor is voor het kolonisatievermogen van een soort. De ecologische profielen worden getransformeerd in twee ecologisch geschaalde landschapsindices (ESLI): 'de gemiddelde draagkracht van een plek' en 'de gemiddelde connectiviteit van een plek'. Simulaties met modelsoorten, die representatief zijn voor verschillende ecologische profielen, laten zien dat de fractie bezette plekken een sterke indicator is voor de duurzaamheid van een metapopulatie. Metapopulaties zijn niet langer duurzaam wanneer de fractie bezette plekken kleiner is dan 0.5 (betrouwbaarheidsinterval 0.4-0.6). Zowel de empirische data als de simulaties met verschillende ecologische profielen laten zien dat soorten verschillende schaal-afhankelijke reacties hebben op versnippering. De empirische data vertonen een positieve correlatie tussen de fractie bezette plekken (een belangrijke indicator voor duurzaamheid) en de ESLI. De conclusie is dat ESLI een bruikbaar instrument zijn voor een geïntegreerde bepaling van duurzaamheid op landschapsniveau.

In hoofdstuk 8, de algemene discussie, wordt besproken of de metapopulatiebenadering zinnig is en een landschaps- of regionale benadering voor het natuurbeheer rechtvaardigt. Op basis van de resultaten van hoofdstuk 2 en 5 wordt geconcludeerd dat de benadering inderdaad zinnig lijkt voor de boomkikker. Voor de heikikker is het empirisch bewijs nog incompleet. Het analyseren van een eenjarig verspreidingspatroon is een eerste stap om effecten van versnippering aan te tonen. Er is echter geen informatie beschikbaar of extinctie en rekolonisatie plaatsvinden en of deze gerelateerd zijn aan de habitatconfiguratie in het landschap. Een van de onbeantwoorde vragen is of dispersie tussen populaties nog plaatsvindt of dat de versnippering van het habitat zodanig is dat (clusters) val lokale populaties volledig geïsoleerd zijn geraakt. Er wordt geconcludeerd dat de soorten verschillen in hun gevoeligheid voor versnippering. De boomkikker, een soort van onvoorspelbaar habitat, is beter aangepast aan versnippering. Toch is de boomkikker in Zeeuws-Vlaanderen waarschijnlijk niet langer duurzaam. De mate en snelheid van de versnippering van het Nederlandse agrarische landschap hebben de kritische grens voor de boomkikker reeds overschreden. Het habitat van de heikikker is minder versnipperd en de soort lijkt nog duurzaam voor te komen. De huidige verspreiding kan echter nog steeds gerelateerd zijn aan het vroegere aaneengesloten landschap. In dit ver-

band is het essentieel om te weten of kolonisaties nog mogelijk zijn.

Vervolgens worden de in deze studie gebruikte methoden om connectiviteit te meten besproken. De trend in de opeenvolgende hoofdstukken naar complexe ruimtelijk expliciete maten is het gevolg van een groeiend inzicht dat connectiviteit een sleutelfactor is in metapopuaties. Voor grondgebonden soorten is de grote variatie in de doorlaatbaarheid van sterk door mensen gedomineerde landschappen een extra motivatie voor ruimtelijk expliciete connectiviteitsmaten.

De laatste paragraaf richt zich op de implicaties voor landschapsplanning. Verschillende instrumenten en hun bruikbaarheid voor de toepassing worden besproken. Statistische modellen geven richtlijnen voor het optimaal inrichten van landschappen voor amfibieën. Simulatiemodellen leveren normen op voor duurzame populatienetwerken en de optimale inrichting van corridors. Deze normen kunnen worden toegepast bij de evaluatie van landschappen en bij het vergelijken van inrichtingsscenario's.

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## Curriculum vitae

Claire Christine Vos werd geboren op 17 september 1959 te Johannesburg, Zuid-Afrika. In 1977 behaalde zij het diploma Gymnasium- $\beta$  aan het Vossiusgymnasium te Amsterdam. Zij studeerde biologie aan de Universiteit van Amsterdam en deed in 1985 haar doctoraal. Voor haar hoofdvak bij de vakgroep Landschapsoecologie en Natuurbeheer van de Universiteit Utrecht leverde zij een bijdrage aan een vergelijkend onderzoek naar de nutriëntenhuishouding van *Erica tetralix* en *Molinia caerulea* op een vergraste heide. Daarnaast bracht zij een half jaar op Schiermonnikoog door - met Marieke Kuipers - waar zij de effecten van secundaire duinvorming op de successie van het landschap onderzocht, onder begeleiding van Henk Doing verbonden aan de vakgroep Vegetatiekunde, Plantenoecologie en Onkruidkunde van de Landbouwniversiteit Wageningen. Verder deed zij een bijvak bij de Intrafacultaire vakgroep Milieukunde en bij de vakgroep Wetenschapsdynamica van de Universiteit van Amsterdam. In 1986 nam zij deel aan een internationale cursus Landschapsecologie, georganiseerd door het Institut für Geographie und Geo-Oekologie te Leipzig. Het onderzoek dat zij in 1987 op vrijwillige basis verrichtte voor NBLF Overijssel aan de boomkikker in de Zuid-Eschmarke, is richtinggevend geweest voor haar latere loopbaan. In 1988 volgde zij de postdoctorale universitaire beroepsopleiding Milieukunde. Zij brak deze opleiding af, om van 1988 tot 1991 werkzaam te zijn als secretaris van de Werkgemeenschap Landschapsecologisch Onderzoek (WLO). Sinds 1991 werkt zij als onderzoeker bij de afdeling Ecologie en Ruimte van het Instituut voor Bos- en Natuuronderzoek (IBN-DLO) te Wageningen.