

Modelling the loss of genetic diversity in vole populations in a spatially and temporally varying environment

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Altering environmental conditions affects the genetic composition of populations via demographic and selective responses by creating of variety of population sub-structuring types. Classical genetic approaches can predict the genetic composition of populations under long-term or structurally stable conditions, but exclude factors such as animal behaviour, environmental structure, and breeding biology, all of which influence genetic diversity. Most populations are unique in some of these characteristics, and therefore may be unsuitable for the classical approach. Here, an alternative approach using a genetically explicit individual-based model (IBM) coupled to a dynamic landscape model was used to obtain measures for the genetic status of simulated vole populations. The rate of loss of expected heterozygosity (H_e) was calculated for simulated populations using two levels of spatial and temporal heterogeneity. Results showed that both spatial and temporal heterogeneity exerted an influence on the rate of loss of genetic diversity, but the precise effect was a balance between the effects of population sub-structuring, the frequency of founder effects and population size. These were in turn related to habitat availability and their influence on vole behaviour. Interaction between spatial and temporal dynamics altered the ratio of effective population size to census size. This indicates an altered reproductive potential, crucial in conservation biology applications. However, when the loss of heterozygosity was corrected for the harmonic mean of the population size, the rate of loss was almost identical in the four scenarios. Unlike classical genetic models, IBMs are flexible enough to mimic real population processes under a range of environmental and behavioural conditions. We conclude that IBMs incorporating explicit genetics provide a promising new approach to the evaluation of the effect of animal behaviour, and random and man-induced events on the genetic composition of populations. They also provide a new platform from which to investigate the implication of real world deviations from assumptions of traditional genetic models.

Introduction

Molecular and genetic techniques, together with modelling methodologies, provide promising new tools for addressing complex environmental questions. Separately these tools have provided important insight into population genetics and population ecology, but combined they may contribute greatly to the understanding of a wide range of research. Fields of applicability include conservation strategies (which require a substantial knowledge base with regard to population structure and genetics), design of environmentally acceptable technologies (e.g. in relation to bio-pesticides, bio-remediation and plant genetic engineering), and the estimation of the impact of environmental or human induced changes on the genetic condition of the populations. A small alteration of environmental conditions can affect the genetic composition via both demographic and selective responses. Thus, there is a need for methods that combine genetic and environmental processes in order to obtain knowledge of the interplay between genetics and demography. Recently *i*-state configuration models (Metz & Diekmann 1986) have become more common in ecological research. These are intrinsically attractive to geneticists since the unit of interest is the individual, however, the benefits to genetics are further-reaching due to the basic ecological concept behind the models. The *i*-state configuration model concept is that the individual responds to its own local and internal environment, resulting in the demography being simulated as the sum of the individual responses. This paper demonstrates the potential to link this type of model with genetics models, to provide realistic genetic simulations.

If the assumptions of the classical quantitative genetics models hold, a loss in expected heterozygosity (H_e) with time should be mirrored by a proportional loss in additive genetic variance (V_a) at the expected rate of $1/(2N_e)$ per generation (Falconer & Mackay 1996). V_a is the genetic variability responsible for the adaptive response of a population to environmental changes. N_e is a measure of the effective population size representing the number of individuals that actively reproduce within a population. Except in very special cases, N_e is expected to be lower than the

census size (N) (number of individuals present in a population). It is reduced by several factors such as: demographic fluctuations, environmental stochasticity, change in the population's age composition, non-random mating within the population, a non-Poisson distribution of fecundities etc. (*see* Slatkin 1989, Whitlock 1992, Caballero 1994, Whitlock & Barton 1997 for an extensive review). Experimental and field studies suggest that N_e/N can be as low as 0.1–0.2 (Frankham 1996). If H_e or the allele number is assessed by means of molecular markers and the mutation rate is known, then N_e can be inferred (Wright 1931, Chakraborty & Neel 1989). However, the mutation rate is difficult to estimate and errors result in large confidence intervals around the estimate of N_e . Furthermore, the loss of genetic variability in small populations is much higher than the gain in genetic variability caused by mutations. Thus, the amount of genetic variability maintained by natural populations exposed to fragmentation and/or demographic fluctuations will be limited (Wright 1978). When populations inhabiting individual habitat patches are stable and capable of long-term persistence, the effects of fragmentation on genetic structure are well described by standard population genetics theory (Hartl & Clark 1989). However, one likely consequence of habitat fragmentation is the formation of an array of patches supporting relatively unstable populations in space and time, subject to some extinction probability. Under these conditions populations can export dispersers destined to colonize unoccupied habitat patches. The circumstances under which such metapopulations can persist have become the focus of ecological research (Gilpin 1991). Often habitat fragmentation results not only in few remaining patches with small population sizes but also in destruction of an effective metapopulation structure (Gilpin 1991). The population genetics consequences of cycles of extinction and recolonization can be complex and have only recently been subject to much study (Hedrick 1996). Although a large body of theory has been developed to investigate the effects of population structure on genetic variation (Cockerham & Weir 1993, Slatkin 1995, Pritchard *et al.* 2000), the models are necessarily an abstraction and simplification of the complex-

ity found in nature based on metapopulation or source-sink concepts. The extent to which these models have relevance to real populations occupying perturbed habitat depends on the degree to which natural populations conform to these models (Wright 1943, Lacy 1993, Whitlock & Barton 1997, Balloux 1999, Nunney 1999). However, their applicability is limited. There is no point in developing complex models to create dynamics that are easily achievable with simple models. Nevertheless, it must be accepted that there are classes of ecological questions that involve variation in spatio-temporal conditions that cannot be investigated without models capable of handling this complexity (Wiegand *et al.* 1999, Parrott & Kok 2002). Spatial dynamics are essential in the modelling of genetics in landscapes. Whilst it has been demonstrated that the main modelling challenge for scaling up movement patterns resides in the complexity of behaviour (Morales & Ellner 2002), the drawbacks are that these behavioural models are data intensive, and that there are many more potential sources of error as the model increases in complexity.

This paper aims to characterise the development of the genetic composition of populations exposed to different environmental regimes by simulating genetic interchange in a simulated population. The modelling system used for this purpose was ALMaSS (Animal Landscape and Man Simulation System) (Topping *et al.* 2003). ALMaSS is an individual-based modelling system designed to investigate the interaction between animals and their environment, in particular the landscape structure and management. Individuals in ALMaSS react to their environment and remember past events, albeit often only physiologically. In this way animals in ALMaSS act, via sensing information from their local surroundings making behavioural decisions. The use of such multi-agent systems (Ferber 1999, Bousquet *et al.* 2001) requires not only the development of the animal models themselves, but also their environment. Moreover, if we accept that changes in environmental conditions over space and time exert an impact on our results, it is important to model these changes accurately. Thus, the challenge is to provide useful information based on the best available data and reason-

able conclusions about the relationship between biotic needs of the organism and its reaction to its surroundings, including human activities. In ALMaSS, this is achieved by the use of a dynamic landscape model. The combination of complex animal behaviour and dynamic landscape provides a comprehensive system in which to explore the behaviour of genetic measures in a realistic modelling environment. A simple genetic model (neutral genes, no mutation, etc.) was designed to investigate the applicability of ALMaSS for generating such genetic simulation data, and to investigate the intricate relationships between landscape characteristics, such as permeability and the resulting spatio-temporal dynamics of the genetic diversity.

Methods

Model Construction

ALMaSS was constructed in C++ using the OOP paradigm (Harmon 1993, Booch 1994). At present ALMaSS versions exist for PCs running the Windows based and Linux operating systems. ALMaSS consists of two models, a landscape model that simulates the structure and dynamics of the landscape and species models that simulate the behaviour and movement of the specific animal(s), which in this case is the field vole. The field vole incorporates a genetic model in order to simulate the dynamics of the genetic composition of the population.

Landscape model description

The landscape model provides the animal models with all the data they require from their surroundings, including those events triggered by man, but not including inter-animal interactions. Available landscapes are made from aerial photographs, existing digitised material and fieldwork. The 2×2 -km landscape used for these simulations was taken from a 10×10 -km area of Jutland in Denmark including the town of Bjerringbro ($56^{\circ}22'N$ $9^{\circ}40'E$). The landscape structure is stored as a raster representation of a fixed-polygon map. The resolution of the raster

is 1 m², thus fine details such as roadside verges and hedgerows can be accurately represented. Structures and elements in the landscape are represented by polygons, each covering a large number of raster elements. These polygons are classified into different habitat types, houses etc. All vegetated polygons in the landscape have vegetation growth models simulating growth in terms of total and green leaf area index and vegetation height. These models are adapted from Olesen and Heidmann (1990) and Plauborg and Olesen (1991), for crops under Danish conditions. The models are based on day-degree calculations and interactions with man's management of the crop on that polygon. Similar models were created for non-crop vegetation by estimating curves from Al-Mufti *et al.* (1977). Field polygons have an independent model for weed growth based on non-crop vegetation. The weed model is influenced by farm events such as mechanical weeding or herbicide application.

Weather data is used to create much of the dynamics of the landscape. This data is stored as daily records of mean temperature, mean wind speed, and daily precipitation. The length of the weather record is unlimited, but the current implementation uses a continuous loop containing eleven years data from Bjerringbro, Denmark for the period 1989–1999.

Farm management

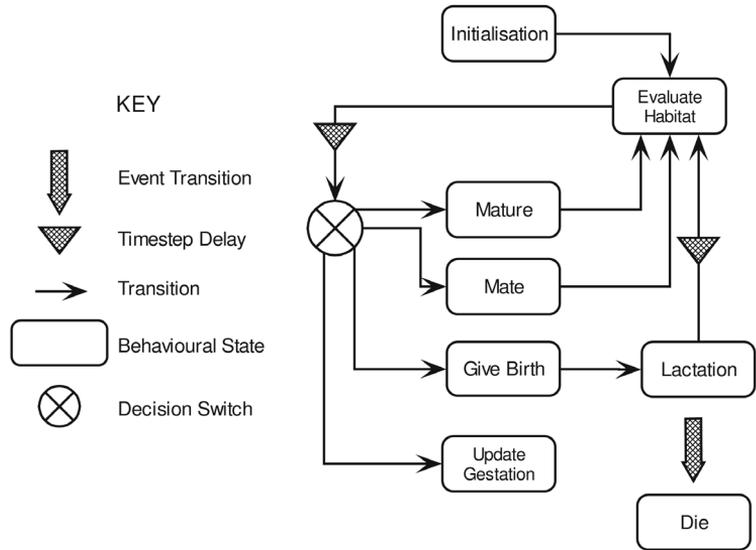
For a Western European landscape such as Denmark, agriculture is by far the largest land-use. Therefore, agricultural operations are a crucial part of a landscape simulation. ALMaSS includes a detailed farm management simulator that is capable of providing information on order, intensity and timing of farm activities. This is achieved by defining farm units, farm types with associated crop rotations, and individual crop-husbandry plans. Crop husbandry is implemented in detail since the management of crops has a profound impact on animal populations. Each crop type has its own crop husbandry plan, which consists of a series of timed events, each with its own set of dependencies and conditions. Events, such as harvest, are recorded when they take place in a field polygon. This data is avail-

able to the vole model, thus voles can respond according to their behavioural rules. An event may cause other changes in the simulation, for example, harvest will alter the vegetation biomass of a crop, and herbicide applications will affect the weed biomass. The source code for the crop management models is available from the authors.

Field vole model description

The vole model simulates *Microtus agrestis*, it is behaviour-based and built upon a state/transition principle. Thus, a vole is considered to be in a specific state when it exhibits specific behaviour. A certain condition or conditions need to be fulfilled, for transition to another state. Transition conditions may be probabilities, or internal or external events (e.g. giving birth or being eaten). The complete set of states describes the behaviour of the vole (Fig. 1). The state/transition construct, movement and territorial behaviours for the field vole require only 21 parameters. The plasticity of the model is related to the logic of combining behaviours; hence although behaviourally complex the model is structurally simple. The time-step of the model is one day. Growth is not simulated explicitly, but is assumed to relate to age. As a result territory disputes are always won by the oldest vole (*see* Evaluate and Explore). This provides for the ability of the model to generate genetic patterns impossible to generate in other ways. For example male movement or migration generates the gene-flow between different areas in the landscape matrix and thereby strongly influences the genetics as, for example, the level of heterozygosity. Such migration depends on the landscape features as well as the presence of other males in the immediate vicinity. Larger males will, as a natural consequence of their position in the dominance hierarchy, tend to disperse less than subordinate males for the simple reason that they are not forced to do so. Such behavioural mechanism increases the bias in the reproductive success among males, which in turn contributes to a lower effective population size, hence the deviation from the Poisson distributed fecundity assumed in the classical genetic models.

Fig. 1. Behavioural states modelled for female field voles. The vole moves between states via transitions. Time step delays cause a cessation of transitions for that time-step. Event transitions occur due to external (to the vole) events.



Behavioural states:

Initiation (σ^{f}) — The vole enters the simulation at the location of the female's nest. Its age is set at the weaned age (14 days, Leslie & Ranson 1940, Innes & Millar 1994), and all maturation parameters are set to their juvenile state. The individual enters the 'Evaluate and Explore' state.

Maturation (σ^{f}) — Immature voles mature when they reach the ages of 19 or 40 days for male and females respectively (Clark 1977). The vole may only breed after it is mature, and once it has a territory with sufficient quality habitat (*see* Evaluate and Explore).

Evaluate and Explore (σ^{f}) — The vole then evaluates its surroundings, assessing habitat and determining if there are enough resources to sustain it. If not, or if there is a better location nearby, the vole will move. This involves three behavioural algorithms (Movement, Dispersal and Assess Habitat).

Movement — the movement algorithm uses four parameters (1) a vector, randomly chosen, except if barriers; (2) a weight determining how strongly the vole will adhere to the direction vector, (3) the number of steps to take. For the male the maximum movement m day⁻¹ with age in months was 10, 40, 70, 110 m, for

the female a fixed value of 50 m was used (fitting with telemetry data from Jensen & Hansen 2001); (4) the probability of not choosing the optimal choice when presented with a range of legal moves (w). This parameter simply allows the vole a chance of moving across non-optimal habitats, without this roads and arable fields would become barriers. The parameter value of 0.001 results in the vole crossing roads with the same frequency as recorded by Hammershøj and Jensen (1998). Movement occurs by taking a stochastically chosen number of steps from ten to the maximum movement allowed (above). Model voles always choose the most optimal habitat based on the Assess Habitat function, unless the probabilistic parameter ' w ' results in a chance of a 'poor choice', in which case the choice is random between all legal moves.

Dispersal — Dispersal uses the same basic model as movement except that the weighting used is higher (4 cf., 2 for standard movement), and unless dispersal is completed, the direction vector, once chosen, is held constant between time-steps.

Assess Habitat — This is implemented differently for different ages, and sexes, but relies on two basic evaluations. These are the number of voles present in an area around the individual, and the quality of the habitat. Quality is adapted

from Hansson (1977) as categorizing habitat into five categories depending on cover and height of grassy vegetation: 1 — cover > 80% and height > 40 cm; 2 — cover > 40% and height > 10 cm; 3 — cover < 40% or height < 10 cm, 4 — no grass, and 5 — no vegetation (buildings etc.). These habitats are arbitrarily scored as 3, 2, 1, 0, -1 respectively. Assessment of single squares or areas returns the sum of the individual scores for each 1-m² area. The habitat assessment used for evaluation and exploration of the vole is based on territory size: maximum male territory radius of 20 m (Agrell *et al.* 1996), maximum female territory radius of 16 m (Erlinge *et al.* 1990), minimum male territory radius of 12 m (Erlinge *et al.* 1990), and minimum female territory radius of 8 m (Jensen & Hansen 2001). In order for a territory to be acceptable the score for the area must be above a minimum threshold value of 1.0. This value is calculated by dividing the score obtained by the minimum male or females territory area multiplied by the score for sub-optimal habitat (category 3). A value of less than 1.0 is below the survival level and will result in immediate dispersal. If the vole is territorial (i.e. adult with a territory in the breeding season), then the habitat score is modified by the number of voles of the same sex, but older, sharing that space. Hence if two older voles are positioned inside the territory the value for acceptance is score/3, therefore, 3.0 is the minimum value, which will not initiate dispersal. This method of implementing territoriality simulates the seasonally territoriality of voles described by Erlinge *et al.* (1990) and Agrell *et al.* (1996). For males in the breeding season, the presence of territorial females is also a factor in the habitat assessment. Where no females are present in the male's territory, the territory is deemed unsuitable.

Mating (♀) — If the female is mature with a territory, she will mate if there is a male with his territory overlapping the female's position. Gestation will last for 21 days (Leslie & Ranson 1940, Innes & Millar 1994), after which the Giving Birth behaviour will follow.

Giving Birth (♀) — The number of young produced (3–5), is determined according to a variable parameter. This parameter includes adjustment

for the time of year and age of the female and is estimated from Anděra (1981). Sex ratios of young are assumed equal (Myllymäki 1977). The model restricts the females, which can produce young to those occupying good habitat (category 2), for at least three days prior to giving birth.

Lactation (♀) — Initially the female places her nest in the best area of habitat available to her in her territory. For the rest of the lactation period (14 days; Leslie & Ranson 1940, Innes & Millar 1994), the female stays within 6 m of the nest (Jensen & Hansen 2001). When the young are weaned, the female does the 'Evaluate and Explore' behaviour.

Dying (♂♀) — Three factors can kill a vole. It may reach the end of its natural lifespan (450, Myllymäki 1977), it may be killed by a predator, here assumed to be a background mortality chance of 1% per day, or it may die because it spends too long in habitats with a quality lower than 1.0 (due largely to increased risk of predation). The vole is arbitrarily allowed to spend 8 days in such habitats before dying. Female voles that die with non-weaned young result in the mortality of the young. Incorporation of habitat related mortality gives the model the ability to simulate different selective advantages of different voles in different habitats. However, this is a coarse approximation to the actual impact of habitat, which may alter movement ability and mortality in a more complicated way.

Infanticide (♂) — If, as a result of dispersal, the male moves outside the bounds of his original territory, he will initiate infanticide behaviour. This behaviour is assumed to aid reproductive access to females (Agrell *et al.* 1998). The chance of him successfully killing the young depends on their age, which in turn determines how aggressively the female guards them. Figures were not available for *M. agrestis*, hence values were used from *Microtus arvalis* (Heise & Lippke 1997).

Vole genetics

The genes carried by voles consisted of 16 unlinked *loci* and four potential alleles for each

locus. Thus, being diploid each vole carries 32 individual alleles in 16 pairs. At mating, an equal amount of genetic material is passed on to the offspring by each parent. The allele passed on at each *locus* is selected randomly from the two paternal alleles they carry at each *locus*. The inheritance process occurs independently for each newly born vole. Thus, all voles are potentially genetically unique. Linkage equilibrium, no mutation and neutral genes are assumed. During simulations the population was censused at the end of each year, and the population size, number of alleles present, allele frequencies and expected heterozygosity (H_{es} , calculated from the allele frequencies) were recorded.

Scenarios

Four scenarios were created and compared. All scenarios shared a common landscape structure, but differed in the farm management. Two types of crop allocation were defined. The first, a 'Standard', which simulates the actual crops (ca. 20 crop types) and their coverage found on farms in the landscape, and the second, a 'Simple' consisting of three crops (Spring Barley, Rye Grass, Winter Rape). These two cropping schemes were incorporated into the four scenarios by using crop rotation and no rotation versions. Thus, there were two degrees of spatial heterogeneity (Standard and Simple) and two of temporal heterogeneity (Rotation and No Rotation). The combination of the two degrees of spatial and temporal heterogeneity produce four different scenarios: standard rotation (StdR+), standard no rotation (StdR-), simple rotation (SimR+) and simple no rotation (SimR-).

In all cases the initial vole population was 2000 randomly positioned individuals. Allele frequencies were varied among *loci*, but were standardised between initial populations in the different simulations (starting allelic frequencies for each *locus* are available from the first author on request). The resulting heterozygosity at year zero was 0.56, and was the result of random mating between the initial 2000 voles. Forty replicates of each scenario were conducted. Simulations were run for 200 simulated years during which the simulated yearly allele frequencies,

and expected heterozygosity (H_{es}) were calculated. Thus, H_{es} was calculated from these allele frequencies observed in the simulation output, according to the equation:

$$H_{es} = 1 - \sum_{i=1}^k p_i^2 \quad (\text{Hartl \& Clark 1989}) \quad (1)$$

where p_i is the frequency of the i 'th allele. Changes in N_e drive changes in H_{es} . In fact, in equilibrium condition, N_e can be inferred from H_e if the mutation rate is known (Chakraborty & Neel 1989). The theoretically expected heterozygosity (H_{eT}) after t generations in a population of changing size, where random genetic drift and changing N_e are reducing heterozygosity, was calculated from equation:

$$H_{eT} = H_{e0} \left(1 - \frac{1}{2N_e} \right) \quad (2)$$

where H_{e0} is the expected heterozygosity, calculated from allele frequencies in year 0 and N_e (the effective population size in year t) which has been assumed to be equal to the harmonic mean (HN) of the census size N calculated for each time step. This would provide an estimate of H_{eT} that incorporates drift and changes in N_e . Although unlikely it is possible that a spatial dynamic pattern could emerge, which creates sub-structuring and hence leads to (or results in) a change in N_e , but not in HN. To check for this possibility, we compared the expected heterozygosity values detected in the simulation (H_{es}) for each year with the theoretical loss of heterozygosity H_{eT} . Hence, the compared heterozygosity (H_{ec}) is simply the difference between H_{es} and H_{eT} :

$$H_{ec} = H_{es} - H_{eT} \quad (3)$$

Clearly, in the simulation $N_e/N < 1$ and is not a constant (C) among scenarios, $N_e/N \neq C$.

Statistical analyses

A linear regression analysis was conducted for all the \log_{10} -transformed values of H_{ec} and H_{es} versus the year of observation. The mean of census size $N \pm SE$, the harmonic mean $HN \pm SE$ and the ratio $(HN/N) \pm SE$ were calculated for every scenario, and the values calculated for each year graphed.

Results

Mean population sizes ($N \pm SE$) for the four scenarios were in decreasing ranking order: (SimR– > SimR+ > StdR+ > StdR–). The Harmonic mean (HN) ranking order was different than the average census size (SimR+ > SimR– > StdR+ > StdR–) (*see* Table 1). The HN/N (harmonic mean/census size) ranking order was neither concordant with N or HN ranking order: (StdR– > SimR+ > StdR+ > SimR–) (*see* Table 1).

For each scenario, the regression line and the 95% confidence interval (c.i.) of H_{es} and H_{ec} for the forty replicates of population size were transformed using \log_{10} and graphed (Figs. 2 and 3). StdR+ had the steepest drop in the \log_{10} -transformed values of H_{es} during the simulation, followed by StdR–, SimR+ and SimR– (*see* Fig. 2 and Table 1).

The picture changed when looking at \log_{10} -transformed values of H_{ec} as all the four scenarios showed nearly the same slope (Fig. 3 and Table 1). The HN and N values for each plotted for each year showed qualitative differences among scenarios of N , HN and the ratio HN/N (Fig. 4).

Discussion

Model results

The H_{es} levels obtained from running the simulations gave the impression, that the two simple scenarios (SimR+ and SimR–) represented

situations where genetic variation was better preserved as compared to StdR+ and StdR–. The findings indicate that a high level of spatial heterogeneity (StdR+ and StdR–) leads to loss of genetic variation, at a much higher rate than in scenarios with low spatial heterogeneity (Sim+ and Sim–) (Fig. 2). In the Simple scenario, addition of temporal heterogeneity (SimR+) increased the rate by which heterozygosity was lost as compared with SimR–. The greater demographic fluctuations (due to agricultural disturbances), of the rotational landscape contributed to a quicker drop in genetic variability. However, where spatial heterogeneity was sufficiently high (Standard scenarios) the addition of temporal heterogeneity (StdR+) seemed to decrease the rate of loss of genetic variability. This is almost certainly due to the temporary increase in permeability between points when suitable crops are adjacent, thus facilitating population success. In the SimR+ and SimR– scenarios, these factors still operate, but are considerably weakened by the restricted crop choice, which means these landscapes already have decreased landscape heterogeneity but increased permeability (Topping *et al.* 2003).

The SimR+ and SimR– scenarios sustained large populations, diminishing the rate by which heterozygosity (determined by H_{es}) was lost as compared to the lower population sizes in StdR+ and StdR–.

When correcting H_{es} with H_{eT} to obtain H_{ec} , the relationship between spatial and temporal variation was no longer clear. In fact, in contrast to H_{es} the rate of loss of H_{ec} was nearly the same in all the four scenarios (Fig. 3). A more thorough

Table 1. Slopes (b) and SE of fitted regressions for the landscape scenarios measured as the average expected heterozygosity detected in the simulation $\log_{10}H_{es}$ and the compared expected heterozygosity $\log_{10}H_{ec}$ (both $\times 1000$ for clarity). The mean of the census size $N \pm SE$, the harmonic mean of the census size $HN \pm SE$ and the ratio N/HN in the four scenarios are shown.

Scenario	StdR+	StdR–	SimR+	SimR–
Mean N (census size) $\pm SE$	809 \pm 17.72	590 \pm 8.34	1224 \pm 16.92	1276 \pm 13.53
HN (harmonic mean of N) $\pm SE$	557 \pm 3.71	494 \pm 1.21	962 \pm 6.57	863 \pm 9.32
HN/ N $\pm SE$	0.74 \pm 0.02	0.867 \pm 0.01	0.794 \pm 0.01	0.699 \pm 0.01
H_{es} (slope of the regression) $\pm SE$ ($\times 1000$)	–4.74 \pm 5.4E–05	–5.00 \pm 5.8E–05	–4.15 \pm 4.0E–05	–3.75 \pm 4.5E–05
H_{ec} (slope of the regression) $\pm SE$ ($\times 1000$)	–0.566 \pm 7.0E–06	–0.582 \pm 1.0E–05	–0.563 \pm 7.0E–06	–0.572 \pm 6.0E–06

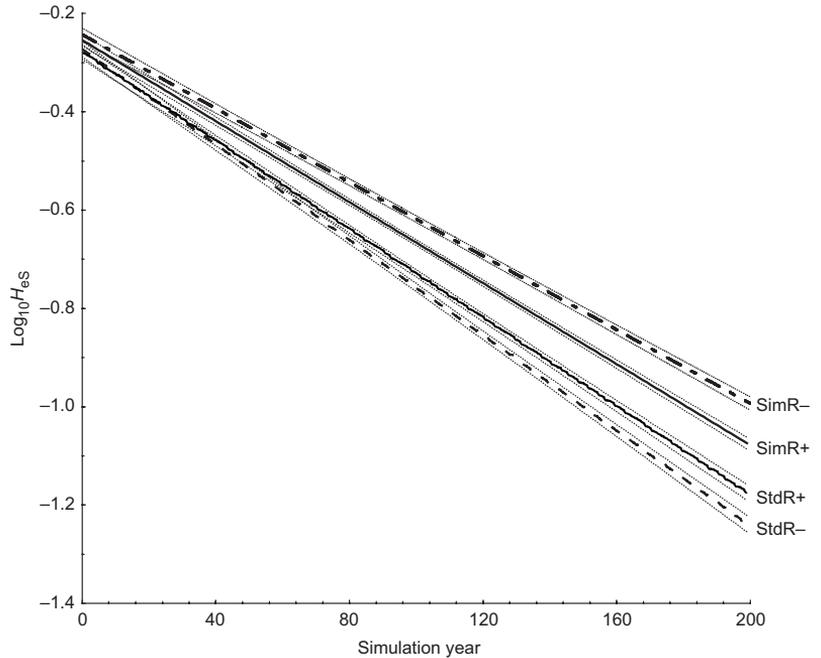


Fig. 2. Log_{10} expected heterozygosities (H_{es}) (95% c.i.) calculated as the mean of 40 scenarios for the four scenarios *versus* year. Values plotted as $\text{log}_{10} H_{es} \times 1000$ for clarity.

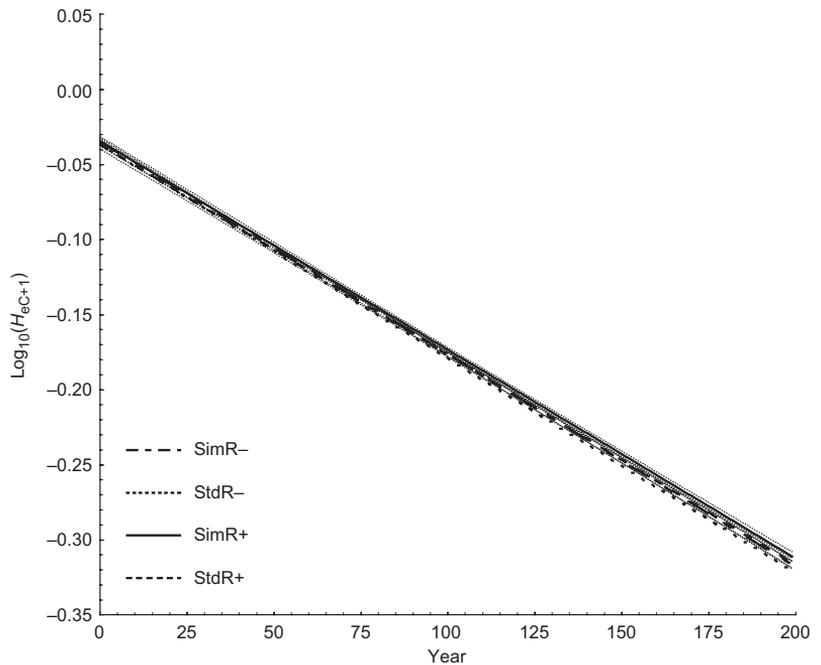


Fig. 3. Log_{10} corrected heterozygosities (H_{ec}) (95% c.i.) calculated as the mean of 40 scenarios for the four scenarios *versus* year. Values plotted as $\text{log}_{10} H_{es} \times 1000$ for clarity.

investigation of the results including behavioural aspects is needed to provide the explanation.

Generally, heterozygosity is expected to be reduced by a rate of $1/(2N_e)$ per generation, in a panmictic population. In a fluctuating population N_e is mainly determined by the harmonic mean

of the population size over time. Since the harmonic mean tends to be dominated by the smallest terms (Caballero 1994), the degree of loss of genetic variability is mainly determined by the lowest population size values observed during the simulations (*see* Table 1). However, an exact

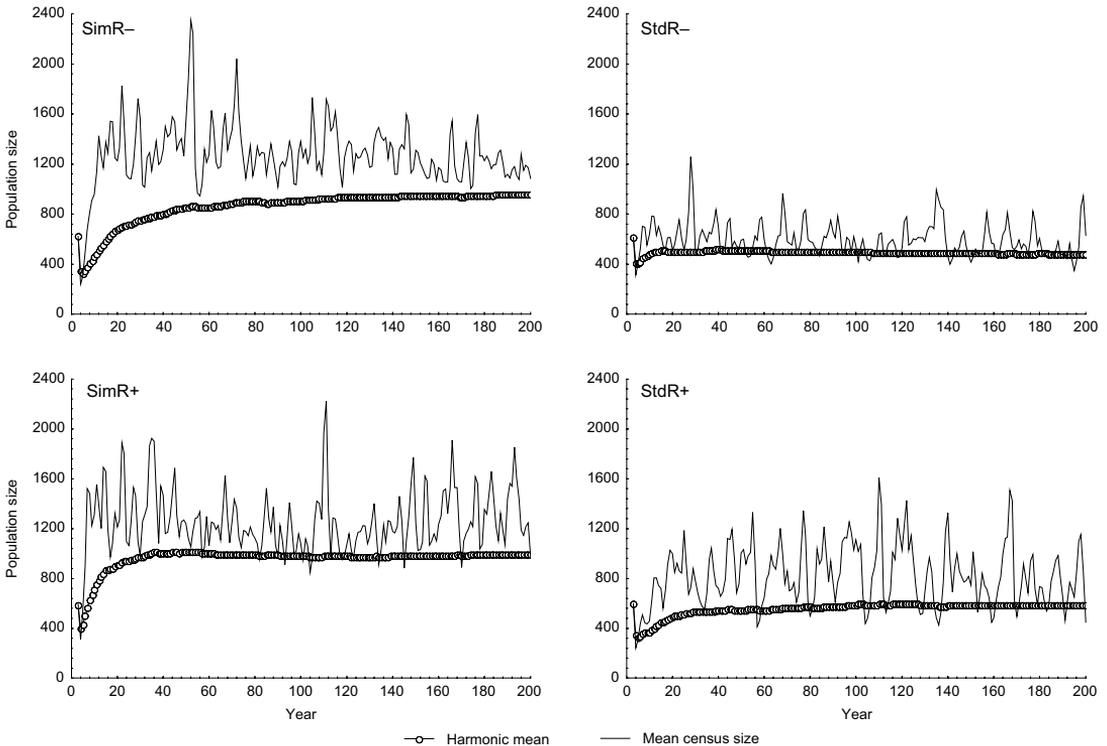


Fig. 4. Census size (N) and harmonic mean of census size (HN) calculated as the mean of 40 scenarios for the four scenarios *versus* year.

relationship between the harmonic mean and the N_c can be derived exclusively for discrete non-overlapping generations (Wright 1938). In our case, as in many others, the organisms reproduce in continuous overlapping generations and hence no equivalent analytically-derived relation can be applied. This only stresses the need for explicit approaches in modelling spatio-temporal fluctuations and their effect on the loss of genetic diversity. However, since H_{ec} is a term that is corrected for the harmonic mean of the population and since the difference of loss of heterozygosity among scenarios becomes minimal when correcting for the harmonic mean we can conclude that the amplitude of oscillations of the population in a given scenario is the dominating factor for the preservation of genetic variability. Although, as stated above, an exact relationship between the harmonic mean and N_c can only be derived definitively for discrete generations, in the scenarios the harmonic mean was a reliable estimator of N_c , despite the generation overlap. Thus, the slope of loss of H_{ec} can be used to

determine the extent to which other factors, not visible from the census statistics, are driving genetic change.

The model shows that two different pictures are obtained depending on whether H_{es} was corrected with H_{et} or not. It is therefore important to note that the effect of population size on the genetic composition is important, and that information can be misleading without considering this, hence comparisons of scenarios could give misleading results. The HN/N ratio can also be misleading; in fact, despite the fact that StdR- had the highest ratio of HN/N it was the scenario which produced the quickest drop of H_{es} , whereas SimR-, which had the lowest HN/N ratio, showed the slowest drop of H_{es} .

Neither the spatial heterogeneity, nor the temporal heterogeneity was the dominating factor that determined the ratio HN/N . In fact, the ranking order of this ratio: (StdR- > SimR+ > StdR+ > SimR-), showed no clear patterns, indicating the complexity and unpredictability of the interaction between the spatial and the

temporal dynamics (*see*, for example, Whitlock & Barton 1997 and Whang & Caballero 1999 for recent theoretical developments in the calculation of N_e in subdivided population, and Whang & Pollak 2002 for the calculation of N_e in temporally variable populations). Since the temporal fluctuations in population size are however closely interlinked with the specific spatial substructure it is crucial that spatial features are accounted for with a degree of explicitness sufficient to accurately describe population, and thus genetic structure.

Model applicability to genetic studies

This paper attempts to present an example of the potential for IBM models to be used for genetic investigation. To that extent, many details of the vole model are superfluous to this message. However, the fact that the model can be built at all is significant, therefore every attention was paid to detail in its development. Nevertheless, the model is still an abstraction of reality, and many other features could be added to the model to improve it. Stephens *et al.* (2002) show that complex behavioural models can be successfully developed for higher species, even those with complex social behaviour. They show that incorporation of this detailed behaviour will have significant implications for the results. The importance of animal decision-making on the genetic variation cannot be determined from field and molecular studies alone, hence IBMs can be a useful approach. The individual-based/dynamic landscape methodology simulates the real world situation rather closely, in this case in terms of the rate of change in genetic composition being affected by the spatio-temporal dynamics of the agricultural landscape and vole behaviour. The details in the behaviour of the animals allow the effects of these behaviours to be implicitly taken into account. This is essential because behaviour which leads to despotic resource allocation results in a variety of spatial effects, such as the creation of source-sink dynamics, and the attempt to colonise even unsuitable areas. To gain insight to this type of problem, Stephen and Wissel (1999) suggest the use of 'narrowly defined IBMs' as defined by Uchmanski and Grimm

(1996) (very much like the vole model described here). Environmental fluctuations are a feature of real landscapes, and unequal resource partitioning is probably the norm for most species, hence there is a need to model the environment sufficiently accurately, which should be added to Uchmanski and Grimm's definition (Topping *et al.* 2003). These additional steps towards spatial realism open the possibility for more accurate predictions, and sensitivity analyses of various spatial factors on genetic measures.

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