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Towards a landscape scale management of pesticides: ERA using changes in modelled occupancy and abundance to assess long-term population impacts of pesticides

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Abstract

Pesticides are regulated in Europe and this process includes an environmental risk assessment (ERA) for non-target arthropods (NTA). Traditionally a non-spatial or field trial assessment is used. In this study we exemplify the introduction of a spatial context to the ERA as well as suggest a way in which the results of complex models, necessary for proper inclusion of spatial aspects in the ERA, can be presented and evaluated easily using abundance and occupancy ratios (AOR). We used an agent-based simulation system and an existing model for a widespread carabid beetle (*Bembidion lampros*), to evaluate the impact of a fictitious highly-toxic pesticide on population density and the distribution of beetles in time and space. Landscape structure and field margin management were evaluated by comparing scenario-based ERAs for the beetle. Source-sink dynamics led to an off-crop impact even when no pesticide was present off-crop. In addition, the impacts increased with multi-year application of the pesticide whereas current ERA considers only maximally one year. These results further indicated a complex interaction between landscape structure and pesticide effect in time, both in-crop and off-crop, indicating the need for NTA ERA to be conducted at landscape- and multi-season temporal-scales. Use of AOR indices to compare ERA outputs facilitated easy comparison of scenarios, allowing simultaneous evaluation of impacts and planning of mitigation measures. The landscape and population ERA approach also demonstrates that there is a potential to change from regulation of a pesticide in isolation, towards the consideration of pesticide management at landscape scales and provision of biodiversity benefits via inclusion and testing of mitigation measures in authorisation procedures.

1 Introduction

Pesticides are regulated in Europe under Regulation (EC) 1107/2009, a replacement for Directive 91/414/EEC. The pesticide regulation requires that the application of a pesticide has no unacceptable effects on the environment with particular regard to its impact on non-target species, including on the behaviour of those species, and no unacceptable impact on biodiversity and the ecosystem. The general protection goals of the pesticide regulation need to be translated into specific protection goals that define what to protect, where to protect it and over what time period it needs to be protected. The definition of these specific protection goals for the different organism groups is based on the ecosystem services concept and associated key species providing them (European Food Safety Authority 2010, Nienstedt, Brock et al. 2012). This, together with the aim of protecting biodiversity, leads, especially for mobile species, to the development of new risk assessment approaches taking into consideration not only single treated fields as in the traditional risk assessment for pesticides but also population-level impacts at landscape scales. Similar considerations have been applied to environmental risk assessment (ERA) in the USA where landscape scale ERA has already been undertaken (Landis 2003). However, the move towards larger spatial scales and population approaches requires a paradigm shift in ERA, requiring consideration of many facets of ecology not included up till now. Although it seems there is a general consensus that population models have the potential for adding value to ERA by incorporating better understanding of the links between individual responses and population size and structure and by incorporating greater levels of ecological realism, there are still many issues that require further study (Forbes, Calow et al. 2008).

One of the complexities of the real world that requires an innovative solution in regulatory pesticide ERA is the fact that the precise effect of pesticide applications in a particular landscape configuration relies on complex spatial and temporal dynamics involved in animal behaviour, ecology and exposure. For example, in ERA focus is often placed on recovery of in-field populations, utilizing the spatial dynamics of mobile agricultural land species. However, this recovery is normally based on small plot experiments that do not take into account the landscape-scale impacts of source-sink dynamics (Topping and Lagisz 2012, Topping, Kjær et al. 2013, Focks, ter Horst et al. 2014).

Of particular importance to population spatial dynamics in reaction to stressors is the 'action at a distance' or 'source-sink' phenomenon (Pulliam 1988) (e.g. pollinators flying to insecticide treated fields to forage, or depletion of source populations by emigration to treated fields with high mortality). The consequences of these dynamics are not always easy to predict. For example, conventional wisdom would suggest that placing source habitats close to a treated area where they receive over-spray from pesticide treatments would increase the impact of the chemical at the population level, resulting in sinks. However, this may not always be so. In the case of field voles it has been shown that the rescue effects of close proximity of source populations can over-ride the higher rate of pesticide-induced impacts in the off-crop area (Dalkvist, Sibly et al. 2013). Considering this kind of complexity is in fact necessary to obtain realistic responses of the endpoint being assessed to the intended pesticide use. Importantly, this problem is not easily solved by using either simple landscape structures or small sections of landscape, which can induce heavy and unpredictable bias in the assessments. For example, in Holland, Aegerter et al. (2007) have shown that regular geometry of landscape construction results in bias in simulation results. In fact species can utilise resources from both crop and non-crop patches, and the decision to move from one place to another is

made depending on the risks associated with a particular landscape-matrix type (Macfadeyen and Muller 2013). This phenomenon makes prediction even more complicated.

As a consequence of the population properties described above, it is necessary to consider spatio-temporal dynamics in realistic landscapes when carrying out a risk assessment for mobile organisms (EFSA PPR 2015). However, when moving to landscape-scale population-level ERA we also necessarily include a number of other novel features, changing focus to a landscape ecotoxicology as called for by Cairns and Niederlehner (1996), and including social-ecological systems considerations, multiple stressors, complexity at different spatial and biological scales and variability of exposure (Artigas et al., 2012).

Assuming this landscape-scale population-level ERA is carried out, then suitable measurement endpoints need to be determined, and these must reflect the specific protection goals defined for the non-target organisms to be assessed. In the case of terrestrial organisms, where action at a distance, mobile stressors (e.g. changing spatial patterns of pesticide use) and long-term population dynamics are important, these endpoints need to encompass population viability, size, and distribution. These are new elements to regulatory risk assessment, and therefore different ways in which to carry out ERA need to be explored in order to find out which is/are most suitable.

In this paper, we consider some of the new components of landscape-scale, population-level risk assessment, and focus in particular on spatio-temporal issues, illustrated using simulations of a widespread and common European carabid beetle *Bembidion lampros* (Herbst, 1784). This species was chosen because it is a widespread predator that is common on open ground, particularly in gardens and on arable land (Tamaddoni-Nezhad et al., 2013). *B. lampros* therefore represents a typical non-target beetle species of agricultural landscapes.

We present a way to extract, in relatively simple way, the major descriptors of change in beetle population abundance and distribution from the results of complex landscape-scale models, and demonstrate the method to integrate in the assessment of a single pesticide its intended use and pesticide management at landscape scale.

2 Methods

The simulations were run using the ALMaSS system (Topping et al., 2003), a model system designed to provide answers to policy-level questions related to changing land-use or management and the resultant impacts on animal wildlife.

2.2 The model system (ALMaSS)

The ALMaSS project is an open source project hosted on CCPForge (<http://ccpforge.cse.rl.ac.uk>), from where program code can be downloaded. The ALMaSS program itself is a large system comprised of many interacting agent-based models and hence a detailed description cannot be provided here. The reader is therefore directed to the online documentation (Topping, 2009). This documentation follows ODdox format (Topping et al., 2010), combining model description with doxygen (van Heesch, 1997) code documentation. The animal models comprising ALMaSS have been tested using a pattern-oriented approach (Grimm et al.,

2005; Topping et al., 2010) to maximize confidence in their structure and function. The models are quite detailed in their behaviour and hence run times for ALMaSS can be long, usually measured in hours or even days. This is particularly the case for invertebrate model simulations, recorded as having over 40 million concurrent agents. For use in ERA, the animal models can respond to local concentrations of stressors, in this case pesticides. Pesticide stressors are simulated as changing spatial and temporal concentrations, based on spraying regimes and environmental fate of the active substances.

2.2.1 ALMaSS – short overview

ALMaSS is comprised of two main components, the environment and the animal representations. These are represented in the model by classes each forming a structured hierarchy containing smaller model representations as further classes (e.g. Landscape Class->Farm Class->Field Class->Crop Class). The environment interface is provided by the 'Landscape' class. This class contains a map of the landscape to be simulated together with individual landscape element classes such as fields, hedges, roads and woodlands. Fields are a special case. Fields are linked in groups to form farms. These groups are typically based on ownership or management information from municipal or EU farming-subsidy sources. Each farm is an instance of the Farm class, which simulates the detailed management of its fields, dependent upon its farm type, the weather, soil type, and past history of management. There is a degree of stochasticity in farmer decisions, and hence the result is a dynamic pattern of farm management across the landscape, with farmers of the same farm type, growing the same crops, making similar but not identical decisions. All vegetated landscape elements (crops and non-crops) undergo type-specific daily vegetation development based on weather and fertilizer inputs as drivers. Farm-management events (e.g. harvest or ploughing) directly interact with vegetation height and biomass, providing a dynamic picture of changing landscape conditions as a result of both environmental and anthropogenic processes and factors.

The second main ALMaSS component is the simulation of animals, represented by specific classes (e.g. *Bembidion* larva and *Bembidion* adult are two classes). All animals are agents (*sensu* agent-based modelling) and are affected by environmental variables, vegetation structure, and by direct interaction with other agents and/or farm management. Each animal represents an individual, or group of individuals, of a particular life-stage, with its own behavioural rules and interactions with its environment. Animals can sense the characteristics of their environment (habitat type, vegetation structure, temperature etc.), management events, and their own physiological condition. Hence, animals exposed to management will choose behaviour suitable for that management, their current location, and physiological state. Animals can interact with each other in a variety of ways e.g., local-density-dependent interactions. All animals share a common basic form of control simulated as a state machine. This means that they exhibit behaviour associated with a specific state, and make transitions to other behavioural states as a result of internal or external cues.

2.2.3 The *Bembidion* model

The *Bembidion* model's individuals are agents designed to simulate the ecology and behaviour of individual beetles. Due to the very high number of beetles in the real world we use the super-individual concept (Scheffer et al., 1995), using each beetle agent to represent 100 real-world beetles. Since the environment is dynamic, the resultant response of the sum of the agents' interactions with each other and their environment, through space and time, produces an emergent population response. The original model was

described in Bilde and Topping (2004), and full documentation is available in ODdox format (Topping, 2009).

Bembidion behaviour is characterised by annual dispersal and aggregation phases with aggregation linked to non-cultivated habitats and dispersal and breeding largely occurring in open areas. Primary drivers in the model are temperature-controlled developmental rates of eggs, larvae and pupae, together with adult beetle interactions with the landscape. Each super-individual beetle reacts to the local environmental drivers of beetle density within a 2-m radius and to global weather drivers for development and reproduction. Landscape management, primarily agricultural practices, affect beetles directly, e.g. ploughing causes direct mortality (Thorbek and Bilde, 2004).

The response to the pesticide is built into the model by assuming a threshold concentration above which there is a daily probability of mortality. This probability (p) is calculated from $(1-m) = (1-p)^d$, where m is the proportion assumed to die (e.g. 0.8 for 80% mortality over the test period) and d is the number of days over which the test was carried out. If the beetle finds itself in a 1-m² grid cell with an environmental concentration above the trigger, then it is assumed to die with probability p . Note there is no dose-response, so the maximum death rate is set as m over d days.

2.3 Scenario set-up

The scenarios used here were chosen specifically to evaluate impacts of spatial structure on the risk assessment. Hence some simplifications were made to reduce complexity. Specifically these were to assume zero drift and run-off of pesticide and to use a monoculture of treated crop.

Pesticide:

The pesticide properties were chosen both to highlight the issues to be addressed and to be realistic in terms of action. No drift to off-crop areas was assumed in order to isolate completely source-sink dynamics as drivers of change in off-crop areas. It was assumed that no other insecticides are applied to winter wheat and that normal herbicide and fungicide applications do not have any impact on beetles.

An 80% field mortality rate (LR80) for a foliar insecticide-spray application measured over seven days was chosen. Available regulatory field data indicate that this could be considered a realistic value and is not an extreme case (see EFSA, 2015). We assumed an environmental dissipation rate (DT_{50}) of 10 days. To ensure that beetles could be exposed above the trigger threshold for at least 10 days, an application rate of twice the trigger concentration at LR80 to all winter wheat fields was used. We assumed a foliar spray twice during the activity time of the adult beetles, the first on 31st May, and the second 20 days later. This is likely to be the most sensitive period for the beetles where this species is actively breeding and population sizes are lowest.

For a subset of scenarios, toxicity was assumed to be increased by factors of x2, x5, and x10 respectively, simulating e.g. increased sensitivity, increased concentration or more toxic pesticides. This was achieved by changing the threshold concentration for effect by dividing by this factor. These increased toxicity values were used with scenarios with zero field boundaries only (see below).

Definitions of spatial elements:

To avoid confusion between terms commonly used to mean similar but different things in scientific literature we have defined our usage of spatial terms related pesticide treated and untreated areas below.

In-crop: the actual cropped area of a field, which may include unsprayed crop margins;

Unsprayed crop margin: non-sprayed crop. This is an in-crop no-spray strip managed in the same way as the crop but not exposed to pesticides either via direct spray nor via spray drift;

Off-crop: everything that is outside the in-crop area (i.e. not a cropped area and not an unsprayed crop margin), but including grassy field boundaries (off-crop is synonymous with off-field);

Field boundary: a permanent grass strip surrounding the field. Note that in the scenarios where these boundaries are added, they are created from the in-crop area, but in subsequent analysis are considered off-crop.

Crops:

All scenarios were run assuming that the landscape contained fields with a monoculture of winter wheat, which was either treated with the pesticide to be evaluated, or untreated.

Landscapes:

Two different landscapes with a size of 10x10 km² were chosen for the simulations. These landscapes differed in both composition and arrangement of landscape elements and are large enough to minimise potential edge effects (*Bembidion* only move a few m per day). The Herning landscape has a mean field size of 3.32 ha, maximum of 33.9 ha and a total of 1990 arable fields. The Præstø landscape has a mean field size of 7.77 ha, a largest field of 136.6 ha, and 905 arable fields in total. The structure of the off-crop habitats also differs, with large wooded areas in the Præstø landscape and heathland and small woodlots in the Herning (Fig. 1, Table 1).

Landscape Element Type	Herning	Præstø
Bushes/scrub	0.8	0.3
Fields (rotation)**	70.5	66.1
Heath*	3.4	0.0
Linear features (excl. hedge banks)*	3.9	2.5
Hedge bank*	0.9	0.3
Permanent pasture*	1.2	0.0
Unmanaged grassland*	2.6	2.5
Urban	4.6	6.4
Water	0.6	0.7
Wetland	2.1	1.2
Woodland	8.6	19.6
Woodland plantation	1.7	0.6
Total	100.0	100.0

Table 1: Percentage cover by area of each landscape element type in the two landscapes used in this study. The total area of each landscape is 10 x 10 km². No asterisk indicates a non-breeding habitat. * indicates suitable breeding habitat for beetles where reproductive rate of 50% of maximum possible in the model, ** optimal breeding habitat where reproductive rate is assumed to be at the maximum possible. See Topping (2009) for more details.

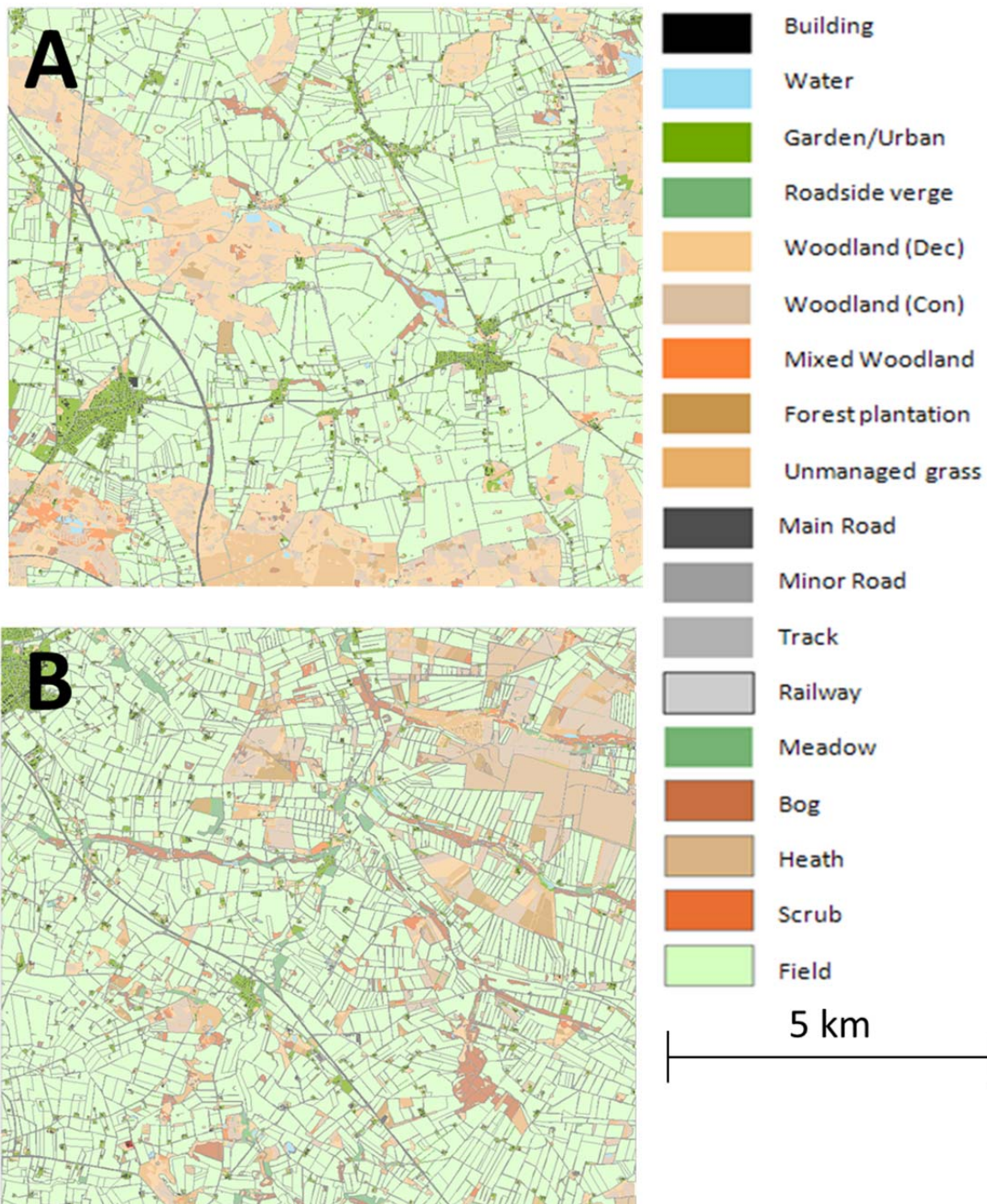


Figure 1. A - Herning landscape; B - Præstø landscape. Key denotes major landscape elements.

Each landscape was used in two artificially manipulated forms. The first was fields that were completely covered by the crop, the second was to create a grassy field boundary in-field around the crops. Such grassy field boundaries could be an option to mitigate the risk from intended pesticide use (i.e. strips which are created in-field, managed as permanent grassy strips and not subjected to the same agricultural practice as the crop itself such as ploughing or harvesting or pesticide application). These boundaries were applied to all fields in three widths, 1 m, 5 m or 10 m. The resulting area cover for field boundaries in the two

landscapes was markedly different, from 1.0 to 17.5% cover as a proportion of the arable field area (Table 2).

Another option to mitigate the risk arising from pesticide use is to leave parts of the crop untreated. In order to investigate the efficiency of such a measure, unsprayed crop margins were added to the fields in the simulations. Unsprayed crop margins of 2 m, 5 m or 10 m were added to the model versions which had fields with 1 m grassy field boundaries around the crop of both landscapes (see Table 3).

Field Boundary Width (m)	Landscape	
	Herning	Præstø
1	1.8%	1.0%
5	8.8%	5.2%
10	17.5%	10.4%

Table 2. Percentage by area of arable field for manipulated non-cropped field boundaries (FB) for the Herning and Præstø landscapes.

Temporal effects

To determine the extent to which year-on-year application of the pesticide resulted in population impacts, the impact relative to baseline was used and compared over time. However, due to the large annual fluctuations caused by the weather, it was necessary to eliminate weather effects for this analysis. Since the weather cycle was repeated after 10 years, comparing the impacts between like weather years was necessary. Therefore the ratio of impact relative to baseline from each year following pesticide application to the corresponding year, but 10 years later, was taken. The analysis was carried out for Herning and Præstø landscapes without field boundaries, with increased toxicity by factors of 2, 5 and 10 (scenarios designated X2, X5, X10).

Other settings and replicates

All landscapes described above were simulated with beetles for baselines and runs with pesticide application. Starting number of beetles were the same in all cases and were randomly distributed across the landscape at a density of 1000 super-individuals per km². Baseline conditions were identical to the product run except that no insecticide was applied to the winter wheat fields. In pesticide-application runs, the pesticide was applied from year 11 until year 30. Data were extracted from the simulations only after the first 10 years of simulation to allow the populations to equilibrate with the landscape (burn-in period), i.e. all data sets consist of 20 simulated years. Ten years was determined to be adequate in initial tests.

Weather conditions were selected to represent the decade 1990-1999 from central Denmark. Each simulation was run for a total of 30 simulation years, looping the 1990-1999 sequence of weather data three times.

2.4 Simulation data extraction

Two main sets of data were extracted from all the simulation runs and gave two measurement endpoints: the Abundance:Occupancy Relationship Index (AOR) and data on numbers of extant female beetles annually over the 20-year simulation period.

2.4.1 Overall population impact

To compute the overall impact of the pesticide use as stressor, statistical analysis was based on the mean differences between baseline and treatment runs with time, resulting in an estimate of mean population depression during the second 10-years of pesticide application. The procedure was to use the raw beetle abundance output from ALMaSS and average for each month over all replicates, then to average these values within each simulation year. The ratio of with-pesticide treatment scenario to appropriate baseline (i.e. baselines for the same landscape map as for the treatment), was computed for each year, and the average over the final 10 years was taken. This was then converted to percentage loss of the population. This method provided an estimate of impact relative to baseline, and controls for year to year variation caused by weather-driven processes within the simulation.

2.4.2 The AOR-Index

Results from a comprehensive simulation model are often themselves complicated and difficult to handle in a management or policy context. To alleviate this problem, ALMaSS output was used to create an index developed from the Abundance to Occupancy Relationship, AOR (Gaston et al., 2000), often studied in macro-ecology. The AOR-index as measurement endpoint has the advantage that it provides a clear picture of the changes in range and density of animals relative to a baseline condition (Hoye et al., 2012). Previously, ALMaSS results have been expressed as changes in local abundance and spatial distribution as described by the univariate Ripley's $K(r)$ (Jepsen et al., 2005). However, this approach is both statistically difficult and results in relatively complex outputs. The AOR-Index was designed to ease both calculation and communication, and works by comparing changes in occupancy and abundance to a baseline scenario. The baseline acts as a reference against which the impacts of scenario changes can be evaluated.

Occupancy was quantified by overlaying the landscape by a regular grid and quantifying the proportion of grid cells containing (super-) individual female beetles using the procedure described by Hoye et al. (2012). The aim of this procedure is to obtain a grid-cell size large enough to allow more than one (super-) individual to be present in each grid cell but small enough also to avoid occupancy and abundance being identical. Two rules were used to identify the grid cell size: 1) in the baseline scenario approximately 50% of the cells should be occupied; 2) if possible within the above constraints the grid size chosen should result in a mean occupancy of >5 . The resulting grid size for all simulations in this study was $50 \times 50 \text{ m}^2$. Occupancy was quantified by the proportion of grid cells occupied by at least one model adult female (i.e. 100 real-world females) for each annual recording of the locations of super-individuals averaged across a 10-year simulation period from year 20 to year 30. Abundance was calculated as the mean number of super-females in grid cells where super-individuals were present. The result can be recorded and translated into a plot of AOR indicating changes in abundance and occurrence relative to a baseline condition expressed as a 2-D plot.

2.4.3 Spatially indexed data

Data were collected on the first day of each month on the number of female beetles present both in the arable fields (in-crop) and in the rest of the landscape (off-crop). Note that beetles in an unsprayed crop margin were classed as being in-crop. These data were collected at the full 10x10 km landscape scale, but also for each 1-km² area based on a regular 1-km grid. Using the same procedure as for the overall impact, the 1-km² grid provided the opportunity to assess how representative of the 10x10 km landscape an impact assessment would be if carried out on the 1-km² scale.

3 Results

The impact of pesticide use on the population of *B. lampros* in different landscape setting scenarios (see Table 3) was simulated with ALMaSS. For each scenario, standard errors for the simulation endpoints 'reduction in beetle population size in in-crop and off-crop areas' (Table 3) were computed from 10 replicate runs and were in all cases less than 0.17%, giving a margin of error less than 0.40% for any single scenario and a margin of error less than 0.55% for comparing any two scenarios. Simulation replicates were therefore very similar and no more than 10 replicates were needed.

Landscap	Grassy Field Boundary Width (m)	Unsprayed Crop Margin Width (m)	Total unsprayed zone width (m)	Toxicity increase	Impact in-Crop (% reduction)	Impact off-Crop (% reduction)	Impact Ratio
He	0 (absent)	0	0	-	35	26	0.74
He	1	0	1	-	26	20	0.77
He	5	0	5	-	23	17	0.74
He	10	0	10	-	19	15	0.79
He	1	2	3	-	25	19	0.76
He	1	5	6	-	24	18	0.75
He	1	10	11	-	22	16	0.73
Pr	0 (absent)	0	0	-	39	22	0.57
Pr	1	0	1	-	29	18	0.63
Pr	5	0	5	-	25	16	0.64
Pr	10	0	10	-	21	14	0.64
Pr	1	2	3	-	28	17	0.62
Pr	1	5	6	-	26	16	0.62
Pr	1	10	11	-	24	15	0.61
Pr	0 (absent)	0	0	X2	55	32	0.59
Pr	0 (absent)	0	0	X5	88	58	0.66
Pr	0 (absent)	0	0	X10	97	68	0.70
He	0 (absent)	0	0	X2	49	37	0.76
He	0 (absent)	0	0	X5	82	66	0.80
He	0 (absent)	0	0	X10	97	84	0.87

Table 3: Scenario definitions and impacts in in-crop and off-crop areas as relative proportion reduction of mean annual beetle population size of all scenarios relative to their respective baseline when pesticide is applied. Impact ratio is the ratio of off-crop to in-crop impacts on population size. Landscapes: He= Herring, Pr= Præstø; X2, X5, X10: toxicity increased 2, 5, 10 fold. See text for more details.

The annual variation in beetle numbers measured as the mean abundance over each 12 month period showed considerable variation. This variation was related to changes in weather, with repeating 10-year cycles being clearly visible (Fig. 2). Total beetle populations were comprised of approximately two-thirds in-crop and one-third off-crop beetles in both Herring and Præstø landscapes.

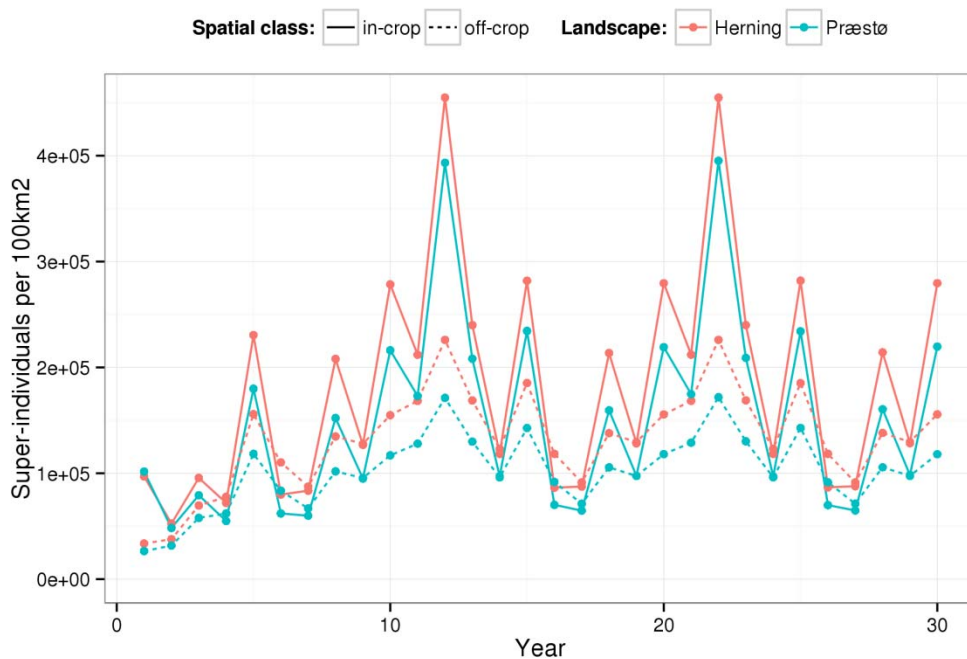


Figure 2. Thirty-year simulation, including the ‘burn-in’ period, of annual mean 12-month adult female beetle super-individuals per 100km² for Herring and Præstø landscapes. These data come from zero field boundary scenarios showing in-crop and off-crop populations when no test pesticide is applied.

To simplify comparisons and remove the direct effect of weather variation impacts, scenarios where a test pesticide was applied are shown as population size relative to the appropriate baseline. Hence, a value of 100% indicates no impact. Results were summarised as mean population impact over the final 10 years of simulation (Table 3). The ratios between in-crop and off-crop impacts clearly show a difference between the two landscapes, with impacts in off-crop populations in Herring always being higher than in Præstø. Conversely, in-crop impacts were consistently higher in Præstø (Table 3).

Increasing the width of field boundaries decreased both in-crop and off-crop impacts. Assuming comparisons with the relevant scenario with pesticide but without field boundaries, adding 1-m boundaries around fields reduced impacts by 25 and 27% in crop and 44 and 54% off-crop for Herring and Præstø landscapes respectively (Table 3). Increasing the size of field boundary to 5m decreased impacts by 34%

and 36% in-crop and 51% and 59% off-crop; increasing to 10m decreased impacts further by 45% and 46% in-crop and 59% and 65% off-crop for Herning and Præstø respectively. Hence, increasing field boundary width decreased the impact of pesticide application on the size of beetle population, but not linearly, with a 10m boundary being a less than 1.5 times as good as a 1m boundary.

The effect of adding unsprayed crop margins was similar to increasing field boundaries, but was at a lower magnitude. Adding a 10-m unsprayed crop margin to a 1m field boundary (see Table 3) decreased pesticide impacts by further 14% and 12% in-crop and 15% and 14% off-crop for Herning and Præstø respectively relative to a 1-m field boundary with no unsprayed margin.

Doubling the sensitivity of the beetles (see Table 3, scenarios X2) increased population impacts by 38 and 40% in-crop, and by 41 and 45% off-crop (Herning and Præstø respectively). An increase in sensitivity of x5 led to an increased impact of 133 and 172% in-crop, and 153 and 159% off-crop (Herning and Præstø respectively); increasing sensitivity x10 led to an increased impact of 176 and 204% in-crop, and by 224 and 204% off-crop (Herning and Præstø respectively).

The two components of the AOR index also showed minimal variability. The standard error for each abundance measure was less than 0.02% and for each occupancy measure less than 0.05%, hence even small differences are due to scenario factors and not noise in the data set. Relative impacts can be visualised using standard AOR-Index plots (Fig. 3). In both landscapes the impacts are much larger in zero field boundary landscapes, and are reduced maximally by having a 10-m field boundary. A reduction in impacts occurred with increasing grassy field boundary or unsprayed margin width in both landscapes following a similar pattern. There were consistent differences between the two landscapes in the responses to pesticide, with higher impacts on abundance in Herning (cf. Præstø) and higher impacts on occupancy in Præstø (cf. Herning).

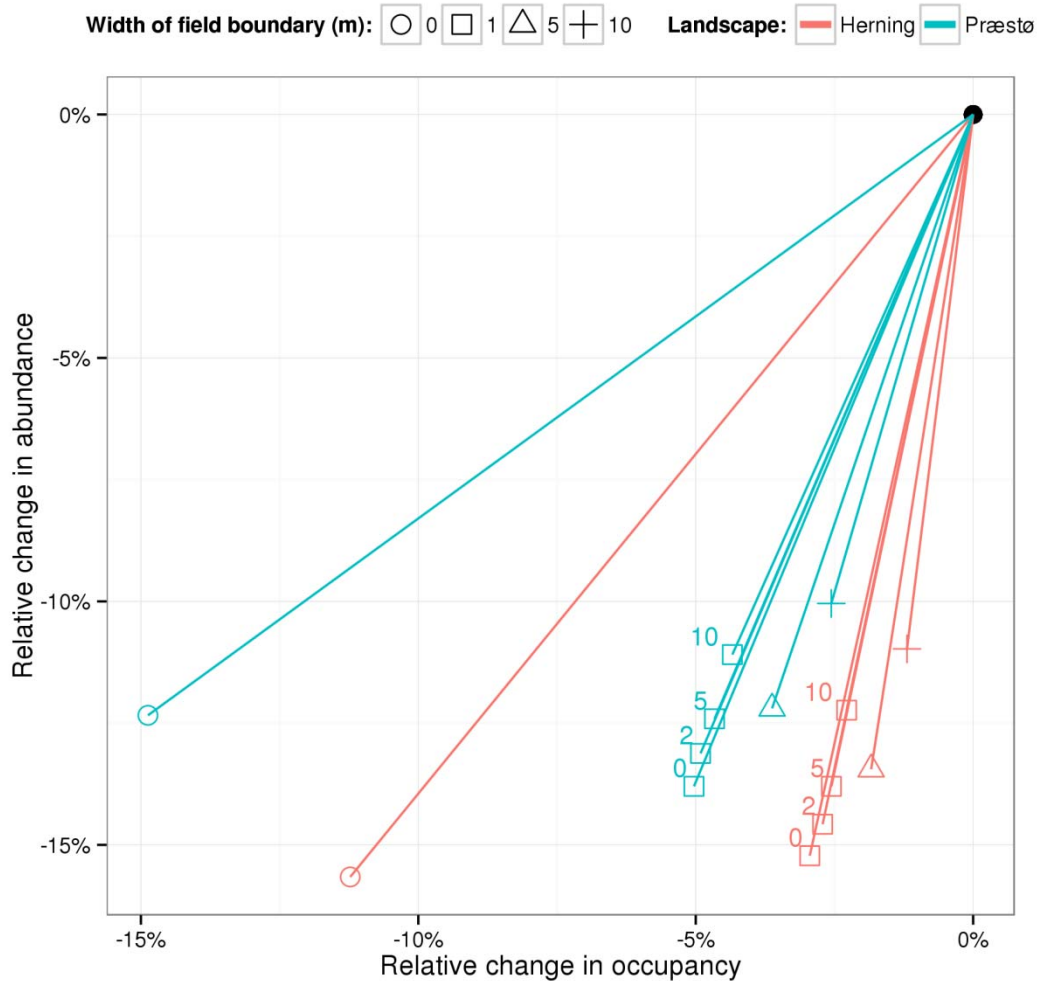


Figure 3. Changes in mean annual occupancy and abundance of *Bembidion lampros* populations (plot of Abundance to Occupancy Relationship) when pesticide is applied to different landscape scenarios. Numbers next to points with 1m field boundary indicate the width (m) of unsprayed margin if present.

As expected from the overall population size impacts (Table 3), increasing the toxicity of the applied pesticide had major impacts on both abundance and occupancy with similar patterns in both landscapes (Fig. 4). Impacts on beetle abundance were, however, generally greater in the Herring landscape, whereas impact on occupancy were broadly similar with a tendency for higher impacts in Præstø.

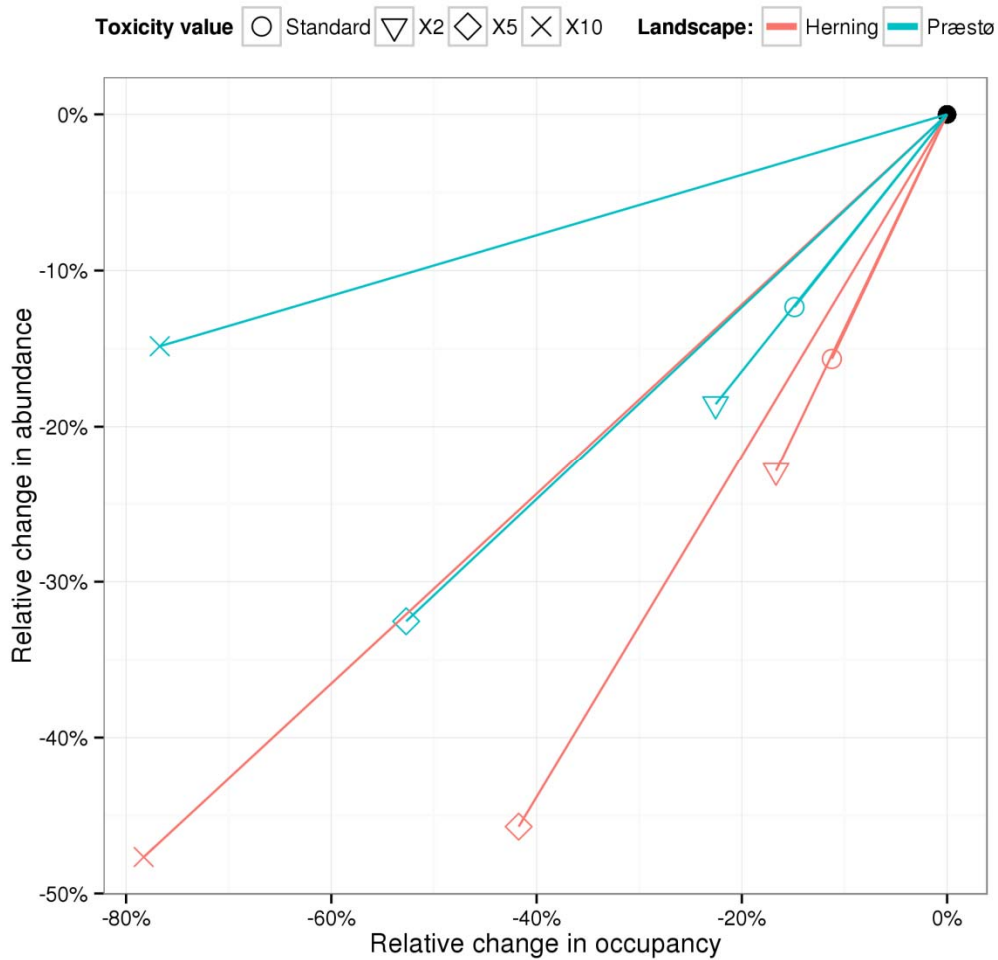


Figure 4. Changes in mean annual occupancy and abundance of *Bembidion lampros* populations (plot of Abundance to Occupancy Relationship) in a landscape scenario without field boundaries when the toxicity of the pesticide is increased. X= toxicity increase of the pesticide: 2, 5 or 10 fold the standard value.

However, displaying the relative impacts of pesticides on beetle population endpoints hides major baseline differences between scenarios. Figure 5 shows changes in occupancy and abundance for the standard toxicity scenarios when pesticide is applied. The baseline population conditions vary considerably between scenarios. Adding field boundaries of 1 m width (going from 0 to 1 m in Fig. 5) increased population abundance by approximately 50% whereas subsequent increased width of these field boundaries increased occupancy with a maximum range of 67 to 74% in Herning.

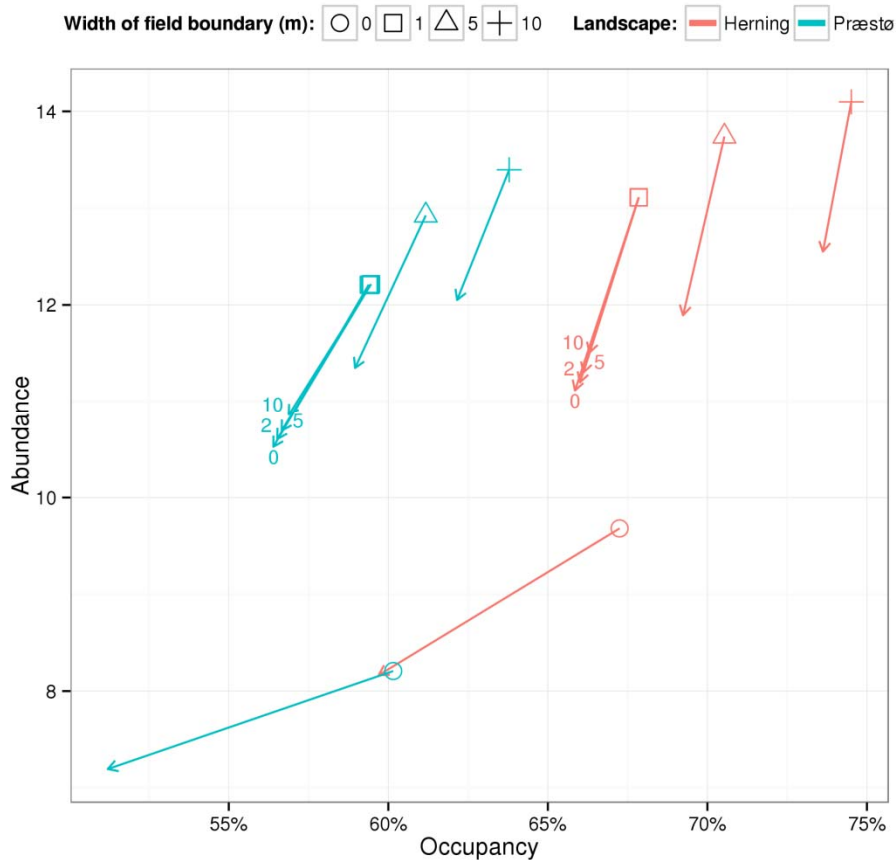


Figure 5. Changes in mean annual occupancy and abundance (mean number of super-individuals per grid cell) of *Bembidion lampros* populations when pesticide is applied to different landscape scenarios. Arrows indicate the changes in occupancy and abundance when pesticide is applied. One meter field boundary includes all such scenarios, i.e. also those with an unsprayed margin for which numbers next to arrowheads indicate the width (m) of the unsprayed margin.

Temporal effects

The ratio of impacts on the mean annual population size of *B. lampros* relative to baseline between like weather years (10 years apart) was not constant but approached stable values in all standard toxicity scenarios after 3 years. However, increasing the toxicity of the pesticide increased the time to population stabilisation from typically three years in the standard scenarios to greater than 10 years for very high toxicity scenarios (Fig. 6). Speed of relative population stabilisation was similar between the Præstø and Herring landscapes. In all cases, in-crop stabilisation was slower than off-crop stabilisation for the same scenario. Slopes of very high toxicity scenarios (value X10) were steeper than the next highest toxicity (value X5).

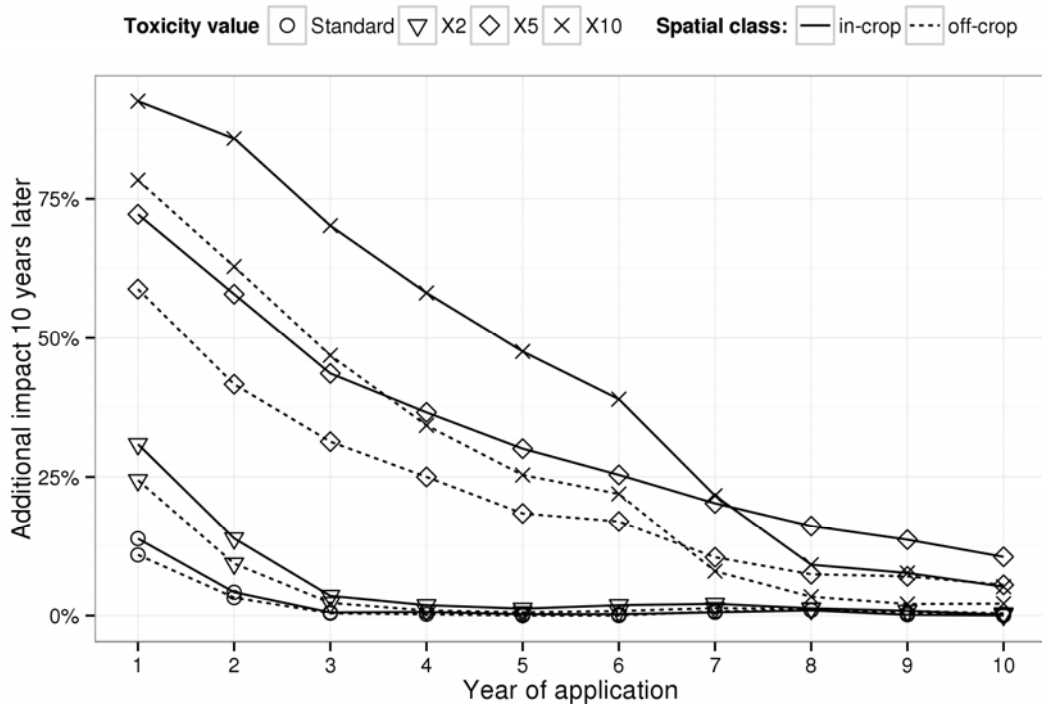


Figure 6. The additional impact on *Bembidion lampros* in years 11-20 after pesticide treatment started compared to populations 10 years earlier, for in-crop and off-crop areas in the Herning landscape scenario. Non-zero values indicate a delay in reaching a stable equilibrium population in the presence of the pesticide.

4 Discussion

4.1 Assessment of pesticide ERA impact on beetle population at landscape scale

All simulated landscape and toxicity scenarios clearly indicate the impact of source-sink dynamics on the effect of pesticide application for the population of the beetle *B. lampros*. Impacts in off-crop populations were often high, especially where there was little off-crop area. The effect of pesticide use was reduced with increasing area of suitable over-wintering and breeding habitats around fields (in this case field boundaries with permanent grass). These findings indicate no-spray zones with permanent grass to be an important consideration for the impact of pesticides on non-target populations. Buffer strips are usually intended to reduce exposure of other off-crop habitats (e.g. Broughton et al., 2014; Felsot et al., 2011; Forster and Rothert, 1998; Streloke and Brown, 2003), but here we suggest that – with proper management – they may also have an important function as source habitats for recolonization of fields, in similar way to beetle banks providing source habitats for predators (MacLeod et al., 2004). This is in line with some field studies showing the importance of field margins for colonisation of cultivated fields by non-target invertebrates (Cole et al., 2012), including species of such low migratory rates as the earthworm *Lumbricus terrestris* (Nuutinen et al. 2011).

Unsprayed crop margins reduced population impacts of the pesticide, but less so than the effect of adding an unsprayed grassy field boundary without crop. In both cases it was assumed that there was a 1-m field boundary (Table 3). The effect of adding unsprayed crop margins is, however, measured without taking into account the reduction in spray drift that these margins would also provide in the field. The lower beneficial effect of increasing unsprayed crop margin width compared to the impact of grassy field boundaries is due to the fact that the unsprayed crop margin only differs from in-crop in not receiving pesticide. The grassy field boundary does not receive pesticide, but also is not ploughed (incurring mortality), and acts as an over-wintering location for this beetle species.

Impacts of pesticide use on the population of *B. lampros* were not instantaneous but changed over a number of years, particularly in high impact scenarios. This is important because field experiments used to evaluate impact of pesticides on non-target organisms normally only consider up to a one-year time frame after first application (Candolfi et al., 2001). Year-on-year application will therefore give a greater overall population impact than would be measured from a single application if there is not 100% recovery of the population in between spray applications or spraying seasons.

In the simulated scenarios, toxicity of the pesticide was clearly a critical factor in determining impacts of pesticide application on populations at landscape scale, although landscape settings also exerted strong effects. A 10-times increase in beetle sensitivity (equivalent to a 10-times more toxic pesticide), led to long-term population declines of over 90%, or expressed as change in occupancy and abundance, a decrease of 80% in abundance and 50% occupancy. However, these results are likely to change under specific realistic scenarios. In other simulations assessing the effects of pesticides on a vole species, toxicity was only one of a number of equally important factors influencing the vole population at landscape scale (Dalkvist et al., 2009).

4.2 Methods of incorporating beetle exposure to the pesticide in the simulation

The method used to model beetle exposure in our simulations is relatively crude. We used an effect probability above a threshold to give an 80% chance of dying over a seven-day period with environmental concentration of pesticide above a trigger concentration. The disadvantage of this approach is that for long-period of exposure effects are virtually certain as probabilities combine each day, a result of multiple double jeopardy probabilities. This does not represent the case where individuals in a population have differential sensitivities to a chemical stressor; as a result, local extinction might occur in the simulations which could have important consequences for pesticide population impact and recovery. One suggestion would be to use an individual sensitivity distribution whereby individuals have different threshold levels for effects. This would prevent very high mortality with long exposure, but would equally prevent long exposure having any impact above the instant the highest dose was experienced.

In all cases the effects of multiple applications, long-term exposure and internal accumulation need to be considered, whilst avoiding 'double jeopardy' effects. It is also possible that previous exposure predisposes individuals to effects rendering them more sensitive to the same dose experienced later. One useful facet of laboratory toxicity testing is that the effect rate may change with time, typically highest in the first

period and declining with time, e.g. LC50 of cadmium (Ardestani and van Gestel, 2013). This could be considered if individuals carry a memory of past exposure and effect probabilities be reduced with time. A further complication is the need to include a dose-response relationship in whichever method is chosen and in all cases the implications of using one or other methods on the outcome of the assessment should be evaluated for future application in ERA. It should also be noted that the complications arising from linking exposure to effects in a model will be further compounded by any synergistic or antagonistic mixture effects, should mixtures or multiple stressors be considered.

4.3 Mitigation of risks deriving from pesticide application

Our results suggests an alternative way to approach the risk assessment by integrating even stronger potential mitigation strategies at landscape level as part of the authorisation conditions for pesticides. In the case of non-target arthropods (here represented by *B. lampros*) it seems that, even with pesticide applied, the condition of the population in landscapes with a minimum of 5-m properly managed field boundaries is at least as good as the landscapes without field boundaries and without pesticide application (see Figure 5). This indicates the potential to use simulation results based on more realistic scenarios to carry out an analysis of potential mitigation strategies in given landscapes. If real landscape conditions were taken into account, addition or widening of field boundaries or other non-spray areas could be considered as a way to mitigate the impact of a pesticide. The state of the population with pesticide and mitigation strategy could be compared to a baseline condition to evaluate overall impact using the model framework. A similar idea has been suggested by Kuchnicki et al. (2005) in order to link different buffer widths to environmental risk in Canada in a proposed strategy for a flexible approach to modify pesticide-specific buffer zones for agricultural applications of pesticides.

An interesting result was seen in the simulations of one landscape scenario (Herning, see Fig. 5): When adding a small 1-m field boundary under the impact of pesticides it showed similar effects on the beetle population as the same landscape but without field boundaries and without pesticides. In the landscape with the small boundaries, however, although there was a decrease in occupancy due to the pesticide application, there was still a far higher abundance of animals compared to the landscape scenario without any boundaries. This in effect means that in landscapes with small field boundaries, the range of the beetles was reduced by pesticide applications, but where the animals were still present the densities were higher than in landscape scenarios without any field boundaries.

The observed effects of adding/increasing field boundaries without crop plants will be probably even more evident for some other NTA groups, such as pollinators with herbivorous larval stages. In fields without borders or with too narrow and uniform borders, herbivorous larvae have no host plants; even without pesticide treatments, populations of such species would become extinct in many agricultural landscapes. On the other hand, adding sufficiently broad off-field habitats as e.g. grassy field boundaries or non-cropped areas in-field should help to maintain viable populations of such species in agricultural landscapes (Thomas et al., 2000), possibly even with moderate use of pesticides. This is in line with thinking on the causes of decline in biodiversity being related to a homogenisation of the landscape and its habitats (Benton et al., 2003) and to a decline in non-cropped, non-sprayed habitats. Therefore, there is real potential for improving biodiversity by increasing habitat heterogeneity and non-sprayed areas, so that, for

example, food provision for birds is supported through a highly diverse biocoenosis of non-target invertebrates (Vickery et al., 2009).

4.4 Landscape affects the impact of pesticide on invertebrate populations

Although the trends in the mitigation of risk from pesticide use associated with adding one or other boundary type to the field in different landscapes were clear and consistent, the actual size of pesticide impacts and the relative difference between in-crop and off-crop effects differed between the differing landscapes, here Herning and Præstø. This is important because it shows that real landscape configurations will have an impact on the outcome of an ERA employing simulation endpoints, even, as in this case, they appear quite similar. Pesticide impacts on population of non-target organisms are also hard to predict in advance of the simulation because overall effects depend on an interactions between stressor dynamics (spray regime and fate), landscape structure and organism dynamics.

Landscape structure resulted in differences in overall pesticide effect on population endpoints at large scales. Pesticide impacts in Herning landscape were generally higher on abundance endpoints, whereas impacts on occupancy were higher in Præstø. In baseline scenarios without pesticide application, beetle abundance was similar in both landscapes, but occupancy was much higher in Herning. As a result, in-crop pesticide impacts on beetle population in Præstø were higher than in Herning, but off-crop impacts on beetle population were higher in Herning (see relative impacts in Table 3). This suggests that the larger off-crop population in Herning was buffering the effects on the in-crop population more efficiently than in Præstø, although this larger off-crop population also suffered the largest proportional pesticide impacts. Hence, depending upon the definition of the protection goals, this could result in the populations exhibiting the best post-pesticide application health (see Fig. 4), also being designated as those most at risk. This is similar to previous simulation results with vole species, where proximity of source populations to the area of pesticide use reduced impacts at the population level (Dalkvist et al., 2013). Thus, from a population-ecological point of view, the numbers of individuals affected may not be indicative of the overall pesticide impact on populations at landscape level.

4.5 Implications of the simulation results for possible future regulatory ERA

Introducing field boundaries around crop fields resulted in significant decrease of pesticide effects on the population of the non-target invertebrate *B. lampros* both off-crop and in-crop. However, in all of the scenarios the off-crop impact was high when compared to the thresholds defined by the recent scientific opinion (EFSA 2015), i.e. a 10% decrease in population density for local direct off-crop effects and non-negligible reductions in range occupancy and wider abundance at landscape scales. Given the high toxicity of the pesticide in the simulations, even introducing 10m wide field boundaries resulted in 15% - 16% decrease of mean annual population size off-crop. However, before firm conclusions can be reached regarding the comparison of local scale, traditional ERA and the illustrated landscape approach, it would be useful to assess multiple factors potentially influencing the ERA outcome in this specific case. These would include pesticide drift, different crop rotations, weather, different landscape structures and application schedules. In developing these scenarios, the impact of other stressors, including other pesticides used on

other crops, would also need to be taken into account. Overall, this would provide essential background for selecting factors necessary for inclusion in realistic worst-case regulatory ERA scenarios (EFSA PPR, 2014).

Given suitable environmental scenarios, if we assume that comparable Abundance to Occupancy Relationship (AOR) indices equate to comparable ecological population states, then the landscape-modelling approach would also offer the possibility to test and to compare the efficiency of different risk-mitigation options using the endpoints of AOR plots as a working guide. This would offer new options for risk assessors and risk managers in the authorisation process by directly implementing mitigation options at landscape scale in the risk assessment process.

4.6 Possibilities to link regulatory ERA of pesticides to Common Agricultural Policy subsidies

When working with pesticide ERA at landscape scales, including possibly mandatory mitigation measures to reduce impacts, there are issues that overlap with other regulations (e.g. Common Agricultural Policy CAP subsidy schemes and the Sustainable Use Directive (EU Directive 2009/128/EC)). These issues cannot be solved in the context of pesticide authorisation alone. Environmental benefits under the CAP are achieved using the Cross Compliance mechanism, whereby farmers are encouraged to fulfil certain environmental conditions in return for governmental support payments (Meyer et al., 2014). With careful selection of efficacious mechanisms, there is therefore the potential to link pesticide-mitigation measures developed during the pesticide-regulation procedure to pesticide use, using the cross-compliance concept already in force. In effect this would change the focus from individual processes or products towards an integrated landscape-scale management.

Authorisation of the use of pesticides, providing that suitable landscape-scale mitigations measures are put in place, will thus potentially have two major benefits. Firstly, provision of targeted mitigation measures, developed and tested for example via landscape-specific scenario modelling, may in itself lead towards a re-biodiversification of the agricultural landscapes in Europe and could help in achieving the goal of the halt of loss of biodiversity by 2020 (Anon, 2012). Secondly, it provides a way for pesticides still to be used in agriculture in Europe with less harm to biodiversity, even if those pesticides may cause local population-level effects.

5 Conclusions and recommendations

The results of this study show that a risk assessment that is focused on the local (field) scale and on short-term studies is insufficient to predict effects on populations of non-target organisms at larger landscape scales and longer temporal scales. This is based on a number of key concepts that were demonstrated by the scenario modelling:

1. Action at a distance - We demonstrated an off-crop effect from in-crop mortality. Given highly toxic pesticides, annual effects of up to 70% or mean effects of 26% reduction in off-crop population size were predicted after 10 years (even without spray drift).
2. Long-term effects - Assessment based on a single spray application would underestimate the long-term effects. This was demonstrated by the fact that, at high toxicities, the population decline was still

ongoing after 20 years of pesticide use. Even realistic toxicity scenarios required three years for populations to stabilise.

3. Mitigation-strategy evaluation - scores of abundance and occupancy relationships (AOR) allowed comparative evaluation of pesticide impacts and consequences of implementing mitigation measures directly from the modelling outputs and provide a simple method for reducing complex spatio-temporal patterns to simple metrics.

As a consequence of these concepts, traditional higher tier approaches in pesticide risk assessment, which are conducted in small plots in treated and untreated fields, would need to be supplemented by modelling in order to take into account long-term population effects and source-sink dynamics and management effects. However, unlike the scenarios used in this study, more realistic environmental and application scenarios would be needed, e.g. carefully chosen vulnerable key driver species and implementation of drift. We therefore recommend that future research be directed towards assessing the contribution of multiple environmental, eco-toxicological and management factors to the ERA of pesticides to determine those most critical in causing variability in ERA conclusions. Evaluating both impacts and mitigation measures concurrently and taking account of these factors provides a tantalizing possibility, i.e. moving from regulation of a single pesticide, often with no explicit consideration of landscape, towards the consideration of pesticide management at landscape scales and provision of biodiversity benefits.

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