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## Persistent negative effects of pesticides on biodiversity and biological control potential on European farmland

Flavia Geiger<sup>a</sup>, Jan Bengtsson<sup>b</sup>, Frank Berendse<sup>a,\*</sup>, Wolfgang W. Weisser<sup>c</sup>, Mark Emmerson<sup>d,e</sup>, Manuel B. Morales<sup>f</sup>, Piotr Ceryngier<sup>g</sup>, Jaan Liira<sup>h</sup>, Teja Tschardt<sup>i</sup>, Camilla Winqvist<sup>b</sup>, Sönke Eggers<sup>b</sup>, Riccardo Bommarco<sup>b</sup>, Tomas Pärt<sup>b</sup>, Vincent Bretagnolle<sup>j</sup>, Manuel Plantegenest<sup>k</sup>, Lars W. Clement<sup>c</sup>, Christopher Dennis<sup>d,e</sup>, Catherine Palmer<sup>d,e</sup>, Juan J. Oñate<sup>f</sup>, Irene Guerrero<sup>f</sup>, Violetta Hawro<sup>g</sup>, Tsipe Aavik<sup>h</sup>, Carsten Thies<sup>i</sup>, Andreas Flohre<sup>i</sup>, Sebastian Hänke<sup>i</sup>, Christina Fischer<sup>i</sup>, Paul W. Goedhart<sup>l</sup>, Pablo Inchausti<sup>j</sup>

<sup>a</sup>Nature Conservation and Plant Ecology Group, Wageningen University, Wageningen, The Netherlands

<sup>b</sup>Department of Ecology, Swedish University of Agricultural Sciences, Uppsala, Sweden

<sup>c</sup>Institute of Ecology, Friedrich-Schiller-University Jena, Jena, Germany

<sup>d</sup>Department of Zoology, Ecology and Plant Sciences, University College Cork, Cork, Ireland

<sup>e</sup>Environmental Research Institute, University College Cork, Cork, Ireland

<sup>f</sup>Department of Ecology, c/ Darwin, 2, Universidad Autónoma de Madrid, Madrid, Spain

<sup>g</sup>Centre for Ecological Research, Polish Academy of Sciences, Lomianki, Poland

<sup>h</sup>Institute of Ecology and Earth Sciences, University of Tartu, Tartu, Estonia

<sup>i</sup>Agroecology, Department of Crop Science, Georg-August-University, Göttingen, Germany

<sup>j</sup>Centre for Biological Studies of Chizé CNRS, Villiers-en-Bois, France

<sup>k</sup>UMR BiO3P 1099 INRA/Agrocampus Ouest/Université Rennes 1, Rennes, France

<sup>l</sup>Biometris, Wageningen University and Research Centre, Wageningen, The Netherlands

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### Abstract

During the last 50 years, agricultural intensification has caused many wild plant and animal species to go extinct regionally or nationally and has profoundly changed the functioning of agro-ecosystems. Agricultural intensification has many components, such as loss of landscape elements, enlarged farm and field sizes and larger inputs of fertilizer and pesticides. However, very little is known about the relative contribution of these variables to the large-scale negative effects on biodiversity. In this study, we disentangled the impacts of various components of agricultural intensification on species diversity of wild plants, carabids and ground-nesting farmland birds and on the biological control of aphids.

In a Europe-wide study in eight West and East European countries, we found important negative effects of agricultural intensification on wild plant, carabid and bird species diversity and on the potential for biological pest control, as estimated from the number of aphids taken by predators. Of the 13 components of intensification we measured, use of insecticides and fungicides had consistent negative effects on biodiversity. Insecticides also reduced the biological control potential. Organic farming and other agri-environment schemes aiming to mitigate the negative

\*Corresponding author. Tel.: +31 317 484973; fax: +31 317 419000.

E-mail address: frank.berendse@wur.nl (F. Berendse)

effects of intensive farming on biodiversity did increase the diversity of wild plant and carabid species, but – contrary to our expectations – not the diversity of breeding birds.

We conclude that despite decades of European policy to ban harmful pesticides, the negative effects of pesticides on wild plant and animal species persist, at the same time reducing the opportunities for biological pest control. If biodiversity is to be restored in Europe and opportunities are to be created for crop production utilizing biodiversity-based ecosystem services such as biological pest control, there must be a Europe-wide shift towards farming with minimal use of pesticides over large areas.

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## Zusammenfassung

Durch die Intensivierung der Landwirtschaft in den letzten 50 Jahren sind viele Pflanzen- und Tierarten auf regionaler und nationaler Ebene ausgestorben und ist die Funktion des Agrarökosystems beeinträchtigt. Die landwirtschaftliche Intensivierung umfasst viele verschiedene Faktoren, wie zum Beispiel die Homogenisierung der Landschaft, die Vergrößerung von landwirtschaftlichen Betrieben und Äckern und den zunehmenden Gebrauch von Düngern und Pestiziden. Über den relativen Beitrag der einzelnen Faktoren zu den weitgehenden Auswirkungen der Intensivierung auf die Biodiversität ist jedoch wenig bekannt. In dieser Studie haben wir den Einfluss dieser verschiedenen Faktoren auf die Diversität von Pflanzen, Laufkäfern und bodenbrütenden Ackervögeln sowie auf die biologische Schädlingsbekämpfung von Blattläusen, entwirrt.

In einer europaweiten Studie, in acht West- und Ost-Europäischen Ländern, haben wir weitgehende, negative Effekte der landwirtschaftlichen Intensivierung auf Pflanzen, Laufkäfer, bodenbrütende Ackervögel und die biologische Schädlingsbekämpfung - die Anzahl durch natürliche Feinde gefressener Blattläuse - gefunden. Von den dreizehn Faktoren der landwirtschaftlichen Intensivierung die wir gemessen haben, hatte der Gebrauch von Insektiziden und Fungiziden konsequent negative Effekte auf die Biodiversität. Insektizide reduzierten ebenfalls die biologische Schädlingsbekämpfung. Organische Bewirtschaftung und andere Formen von Ökologischem Ausgleich, die zum Ziel haben, die negativen Effekte der Intensivierung auf Biodiversität abzuschwächen, erhöhten die Pflanzen- und Laufkäferdiversität, jedoch – entgegen unseren Erwartungen - nicht die Diversität der Brutvögel.

Wir stellen fest, dass trotz jahrzehntelanger europäischer Politik gegen schädliche Pestizide, die negativen Auswirkungen von Pestiziden auf Pflanzen- und Tierarten andauern und damit auch die Möglichkeit biologischer Schädlingsbekämpfung abnimmt. Wenn die Biodiversität in Europa erhalten werden soll und die Chance auf biodiversitätsgebundenen Ökosystemfunktionen, wie biologische Schädlingsbekämpfung, beruhenden Ackerbau geschaffen werden soll, ist eine europaweite Veränderung zu einer Bewirtschaftung mit minimalem Gebrauch von Pestiziden über eine große Fläche notwendig.

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**Keywords:** Agricultural intensification; Organic farming; Agri-environment schemes; Vascular plants; Carabids; Birds

## Introduction

Farmland is the most extensive habitat for wild plant and animal species in Europe, covering 43% of the EU members states' surface area (EU-27) and still harbouring a large share of European biodiversity, e.g., 50% of all European bird species (Pain & Pienkowski, 1997) and 20–30% of the British and German flora (Marshall et al., 2003). In recent decades, however, agricultural intensification has unquestionably contributed to the impoverishment of European farmland biodiversity (Donald, Green, & Heath, 2001; Krebs, Wilson, Bradbury, & Siriwardena, 1999; Robinson & Sutherland, 2002; Stoate et al., 2001). There is considerable concern that declines in biodiversity affect the delivery of ecosystem services (Hooper et al., 2005). In agricultural landscapes, the services considered most at risk from

agricultural intensification are biological pest control (Tschardtke, Klein, Krues, Steffan-Dewenter, & Thies, 2005), crop pollination (Biesmeijer et al., 2006) and protection of soil fertility (Brussaard et al., 1997).

Agricultural intensification takes place at various spatial scales, from increased application of herbicides, insecticides, fungicides and chemical fertilizer on local fields to loss of natural and semi-natural habitats and decreased habitat heterogeneity at the farm and landscape levels (Attwood, Maron, House, & Zammit, 2008; Benton, Vickery, & Wilson, 2003; Billeter et al., 2008; Hendrickx et al., 2007; Tschardtke et al., 2005; Weibull, Bengtsson, & Nohlgren, 2000). So far it has been difficult to disentangle the impacts of intensified management of local fields from changes in land use at the landscape level, since both occur simultaneously in most agricultural landscapes (Robinson & Sutherland, 2002).

In addition, previous assessments have generally focused on a few taxa or countries and hardly any study has simultaneously addressed the effects of agricultural intensification on key ecosystem services such as the biological control of agricultural pests.

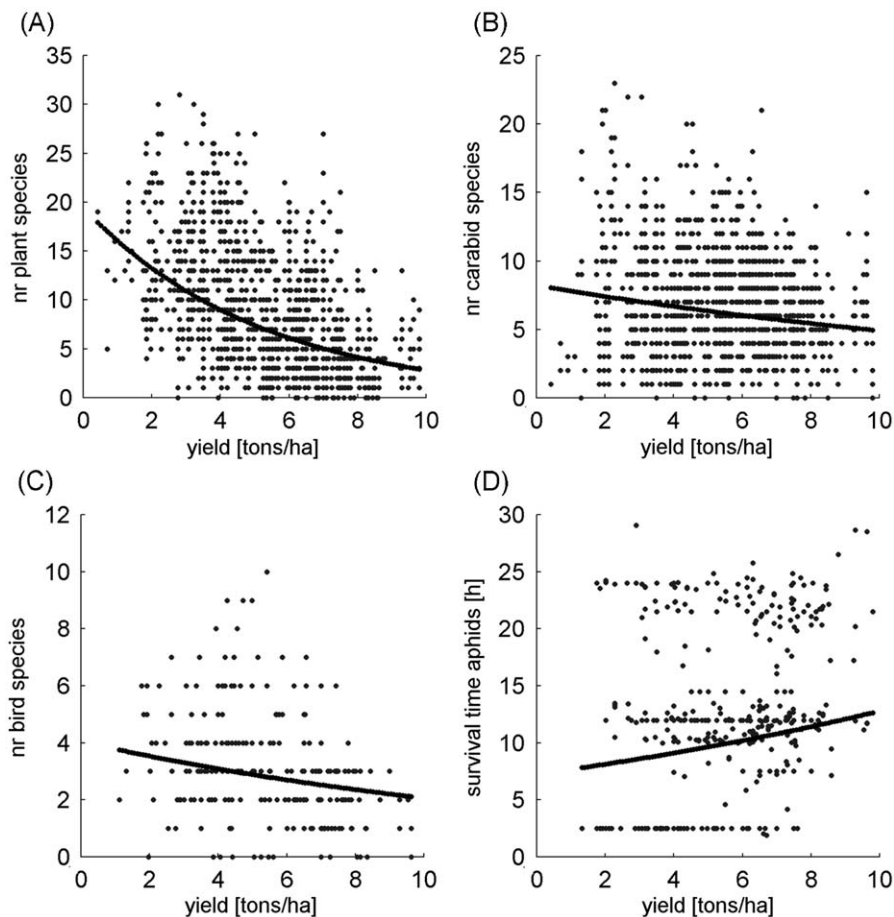
Since the early 1990s the EU has promoted initiatives to prevent and reduce the negative effects of intensive farming. In 1991, legislation limiting the use of pesticides with high risks to the environment came into force (Council Directive 91/414/EEC of 15 July 1991). The reform of the Common Agricultural Policy (CAP) in 1992 aimed to reduce the negative consequences of agricultural intensification by financially supporting agri-environment schemes and organic farming (Council Regulation 2078/92/EEC of 30 June 1992). However, several studies have shown that agri-environment schemes and organic farming do not always deliver the expected benefits (Bengtsson, Ahnstrom, & Weibull, 2005; Berendse, Chamberlain, Kleijn, & Schekkerman, 2004; Kleijn, Berendse, Smit, & Gilissen, 2001). So, an important, but yet unanswered question is whether policies have significantly reduced the adverse effects of intensive

farming on biodiversity and, closely linked to this, on the delivery of key ecosystem services such as biological pest control. In this study, we investigated in nine European areas the effects of agricultural intensification and its components on the species diversity of wild plants, carabids and ground-nesting farmland birds (thus considering three different trophic levels) and biological control potential. We measured eight landscape structure variables and 13 components of agricultural intensification at farm and field level and disentangled their different effects on biodiversity loss. Moreover, we tested the hypothesis that both organic farming and agri-environment schemes reduce the negative effects of intensive farming on biodiversity.

## Material and methods

### Study area

The nine areas studied were located in eight countries: Sweden, Estonia, Poland, the Netherlands, Germany



**Fig. 1.** Effects of cereal yield (ton/ha) on: (A) the number of wild plant species per sampling point (in 3 plots of 4m<sup>2</sup>), (B) the number of carabid species per sampling point (per trap during 2 sampling periods), (C) the number of ground-nesting bird species per farm (one survey plot of 500 × 500m<sup>2</sup>), and (D) the median survival time of aphids (h). Trend lines were calculated using GLMM including the two surrounding landscape variables as covariates and field, farm and study area as nested random effects.

(two areas: close to Göttingen, West-Germany and Jena, East-Germany, respectively), France, Spain and Ireland (see Appendix A, Fig. 1 for the locations of the nine study areas). Each area was between  $30 \times 30$  and  $50 \times 50 \text{ km}^2$  in size to minimize differences in the regional species pools among farms within each area. In each area, 30 arable farms were selected along an intensification gradient using cereal yield as a proxy for agricultural intensification. The farms were selected so that the previous year's yield and landscape composition were uncorrelated within the study area in question.

### Sampling protocol

On each farm, five points distributed over no more than five arable fields were selected for sampling wild plants and carabids and estimating the biological control potential. Most (80%) of the sampling points were in fields of winter wheat (the major cereal crop in much of Europe). The remainder was in winter barley (9%), spring wheat (5%), winter rye (5%) or triticale (<1%). To avoid field margin effects on observations, the sampling points were positioned 10 m from the centre of one side of the field. Whenever two sampling points were located in the same field, they were placed at opposite sides of the field. On each farm, one area of  $500 \times 500 \text{ m}^2$  was selected around one of the sampled fields, for the survey of breeding birds. All sampled fields from different farms were at least 1 km apart. Sampling was performed during spring and summer 2007 and was synchronized using the phenological stages of winter wheat in each study area as a time reference.

### Wild plants

At each sampling point, vegetation relevés were made once during the flowering to the milk-ripening stage of winter wheat, using three plots of  $2 \times 2 \text{ m}^2$ . The plots were placed 5 m apart on a line parallel to the field borders. All species with at least the first two leaves (after the cotyledon) were recorded per plot. To avoid phenological effects of sampling, the sequence of farm surveys was randomized over the intensification gradient within each study area.

### Carabids

Carabids were caught with two pitfall traps per sampling point, which were opened during two periods of 7 days. The first sampling period occurred 1 week after the appearance of spikes of winter wheat (immediately after the biological control experiment, see below) and the second sampling period coincided with the milk-ripening stage of winter wheat. The two pitfall traps (90 mm diameter, filled with 50% ethylene glycol) were placed in the middle of the two outer vegetation plots. The invertebrates caught were fixed in

the lab with 70% ethanol. We identified all the species caught in one trap randomly selected from each pair of traps.

### Birds

The bird surveys were conducted according to a modified version of the British Trust for Ornithology's Common Bird Census (Bibby, Burgess, & Hill, 1992), starting according to local information on the phenology of breeding birds. They were conducted three times, at intervals of 3 weeks. Bird inventory quadrats of  $500 \times 500 \text{ m}^2$  were surveyed in such a way that each spot within the quadrat was no more than 100 m from the surveyor's route. The surveys took place between 1 h after dawn and noon, but only if it was not windy, cloudy, or raining. Breeding bird territories for ground-nesting farmland species were determined using the three survey rounds (Appendix A, Table 1). Three different criteria were used to define breeding bird territories, depending on the species' detectability and breeding behaviour (Appendix A, Table 1).

### Biological control potential

Biological control potential was estimated experimentally during the emergence of the first inflorescence of winter wheat (Östman, 2004). The experiment lasted 2 days and was repeated once within 8 days. In the morning of the first day, live pea aphids (*Acyrtosiphon pisum*) of the third or fourth instar were glued to plastic labels (three per label) by at least two of their legs and part of their abdomen. Odourless superglue was used. At noon, the labels were placed in the three vegetation plots at three of the five sampling points per farm. The labels were bent over slightly, so the aphids were on the lower surface, protected from rain. Three labels were placed along the diagonal of each plot. Hence, at each farm there were 27 labels, with 81 aphids in total. Immediately after the aphids had been placed in the field, the numbers of aphids present were recorded. Thereafter, the labels were checked four more times during 30 h: around 6 p.m. of the first day, at 8 a.m., 1 p.m. and about 6 p.m. on the following day, the exact time varying depending on the study area. After the last check, the labels with the remaining aphids were taken to the lab and checked under stereo microscopes to check whether remaining aphids could not have been removed by predators because they were covered with glue. The data used for the analyses was from one or both of the rounds, depending on what was available from each study area.

## Farmers' questionnaire

Information about yields and farming practices (pesticide and fertilizer use, ploughing and mechanical weed control regime) and farm layout (number of crops, percentage land covered by an agri-environment scheme, field size) was collected by means of a questionnaire sent out to all participating farmers. The response was 98%.

## Landscape structure

On the landscape scale, eight landscape structure variables were estimated within circles around each sampling point (with radii of 500 and 1000 m) and additionally four variables around each bird quadrat (used for the analysis of the bird data; only 500 m radius): mean field size and its standard deviation, the percentage of land planted with arable crops within the area and the Shannon habitat diversity index. The following habitat classes were used to estimate the habitat diversity (according to the definitions from the European Topic Centre on Land Use and Spatial Information (Büttner, Feranec, & Jaffrain, 2000): continuous urban fabrics, discontinuous urban fabrics, cultivated arable lands, fallow lands under rotation systems, permanent crops, pastures, forests, transitional woodland-scrub and water.

## Statistics

Generalized linear mixed models (GLMM) (Breslow & Clayton, 1993) were used to analyse the effects of agricultural intensification on biodiversity and the biological control potential in GenStat 11.1 (Payne et al., 2008). All explanatory variables were included as fixed effects. Because of the sampling structure of the data, fields nested within farms, farms within study area, and study area, were included as random effects. To identify the distribution of the species diversity data (Appendix A, Table 5), the variance to mean relationship was explored at the lowest stratum. This revealed that a Poisson distribution was appropriate for the numbers of plant, carabid and bird species. The median survival time of aphids was heavily skewed and was therefore assumed to follow a lognormal distribution.

Heavily skewed explanatory variables were log-transformed and the percentage of land planted with arable crops was logit transformed (Appendix A, Table 4). All variables were standardized according to  $(x-\mu)/\sigma$ , with  $x$  = measurement,  $\mu$  = mean and  $\sigma$  = standard deviation to enable comparison of the magnitude of their effects.

To reduce the number of landscape variables, a principal component analysis (PCA) was done with

Canoco for Windows 4.5 (Ter Braak & Šmilauer, 2002). This revealed two distinct groups of variables (see Appendix A, Fig. 2): the first group was mean field size and its standard deviation, the second was the percentage of land planted with arable crops and the Shannon habitat diversity index. Because it had the highest correlation with the plane defined by the two axes (longest PCA arrow), mean field size within a radius of 500 m was selected from the first group to be included in the analyses. In the second group, the highest correlation with the plane was the percentage land planted with arable crops. We chose the variable with the same radius as mean field size (selected from the first group), i.e. 500 m.

Only a few intensification variables showed high correlations between each other (Pearson correlation coefficient  $>0.7$ ; see Appendix A, Tables 6A and B). The number of insecticide applications correlated with the amount of fungicides applied ( $r=0.75$ ) and the number of fungicide applications ( $r=0.73$ ) at the farm level only. Number of fungicide applications and the amount of fungicides applied were correlated both at the farm level and at the sampling point level ( $r=0.87$  and  $0.83$ , respectively), as were the amount of inorganic fertilizer applied and the amount of fungicides used (for both,  $r=0.72$ ). These correlations cannot be considered problematic under the present modelling approach (Brotons, Thuiller, Araújo, & Hirzel, 2004).

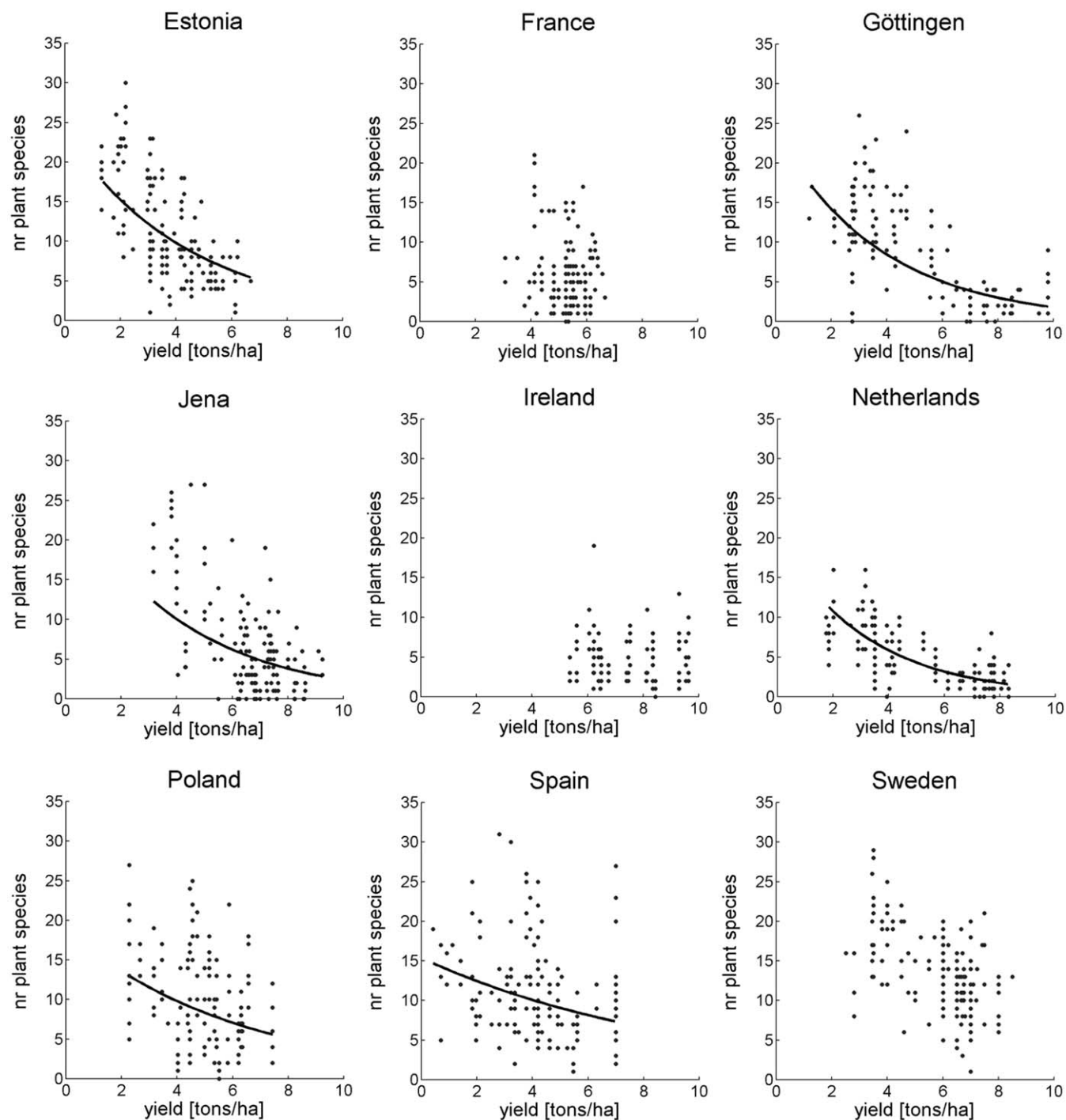
The mean values and standard deviations of all response and explanatory variables included in the analyses are given in Appendix A, Table 7.

Three separate analyses were done using different sets of explanatory variables, but always including the two landscape variables (mean field size and percentage land planted with arable crops within 500 m) as covariates. The first analysis included yield (as a summary variable for agricultural intensification, see Tilman, Cassman, Matson, Naylor, & Polasky (2002)) and its interaction with study area. The second analysis included 13 components of agricultural intensification related to farming practices: amounts of chemical N fertilizer, amounts of organic fertilizer, number of applications of herbicides, insecticides and fungicides, applied amounts of the active ingredients of herbicides, insecticides and fungicides, number of crops per farm, field size, frequency of ploughing, frequency of mechanical weed control and the percentage of arable land under agri-environment schemes (Appendix A, Table 4). Models were derived using forward and backward selection. The forward selection started with an empty model (except for the two landscape variables) and at each step the variable with the most significant effect was included, on the basis of the results of Wald tests ( $p<0.05$ ). This procedure was reiterated until variables no longer added significant effects to the model. The backward selection started with a full model and at each step the most

non-significant variable, i.e. the variable with the highest  $p$ -value, was removed. Forward, as well as backward selection, resulted in identical models in all cases. In a third analysis, we investigated the single effects of farm type (conventional or organic) and the percentage of land under agri-environment schemes. Organic farmers do not apply chemical fertilizers and use only a very

limited set of pesticides. Agri-environment schemes include commitments to lower fertilizer and pesticide applications, while in some countries, margins or entire fields are excluded from fertilizer or pesticide applications.

We emphasize that in the text the term ‘effect’ is used for statistical associations and relationships, and does



**Fig. 2.** Effects of cereal yield (ton/ha) on the number of wild plant species per sampling point (in 3 plots of 4m<sup>2</sup>) in each of the study areas. Trend lines were calculated using GLMM including the two surrounding landscape variables as covariates and field, farm and study area as nested random effects and are plotted, whenever the relationship was significant ( $p < 0.05$ ).

**Table 1.** Effects of different components of agricultural intensification on the diversity of plants, carabids and birds and median aphid survival time. The models selected after considering all 13 intensification variables using forward selection (backward selection produced identical models) are presented. All models included the two landscape variables (mean field size and percentage of land planted with arable crops within a radius of 500 m, in italics), even if these had no significant effects (non-significant effects are not shown). Intensification variables were only included if they had significant effects using the Wald test ( $p < 0.05$ ). AES = agri-environment scheme, amount of a.i. = amount of active ingredients.

Response variable	Explanatory variable	Standardized effect	$\chi^2_1$	p-value
Number of plant species	<i>Mean field size</i>	−0.094	6.09	0.014
	% of land under AES	0.149	12.23	<0.001
	Frequency of herbicide application	−0.1061	8.88	0.003
	Frequency of insecticide application	−0.105	6.15	0.013
	Applied amounts of a.i. of fungicides	−0.262	31.45	<0.001
Number of carabid species	% of land under AES	0.062	6.31	0.012
	Applied amounts of a.i. of insecticides	−0.061	10.87	0.001
Number of breeding bird species	Frequency of fungicide application	−0.127	5.71	0.017
Median survival time of aphids	% of land under AES	−0.144	9.43	0.002
	Applied amounts of a.i. of insecticides	0.114	11.17	0.001

not necessarily mean a causal relation between two variables.

## Data availability

For the second analysis (13 variables of farming practices as explanatory variables), sampling points with one or more missing variables were removed. Bird data were not collected in France. There were no data on aphid survival time for Spain and France.

## Results

We first investigated the relationship between cereal yield, a variable closely related to many different intensification measures (Donald et al., 2001; Tilman et al., 2002), and wild plant, carabid and breeding bird species diversity (Appendix A, Table 1) on arable fields. We found strong negative relationships between cereal yields and the species diversity of wild plants, carabids and ground-nesting farmland birds (Wald tests:  $\chi^2_1 = 141.42$ ,  $p < 0.001$ ;  $\chi^2_1 = 23.33$ ,  $p < 0.001$ ;  $\chi^2_1 = 7.33$ ,  $p = 0.007$ ; Appendix A, Table 2). On average, in the sampled area, an increase in cereal yield from 4 to 8 ton/ha results in the loss of five of the nine plant species, two of the seven carabid species and one of the three bird species (Fig. 1A–C).

Crop yield correlated positively with median aphid survival time ( $\chi^2_1 = 6.85$ ;  $p = 0.009$ ), suggesting a negative effect on the biological control potential (Fig. 1D).

The effects of wheat yield on wild plant and carabid species diversity and aphid survival time differed among study areas (yield  $\times$  study area interaction:  $\chi^2_8 = 36.87$ ,  $p < 0.001$ ;  $\chi^2_8 = 24.35$ ,  $p = 0.002$ ;  $\chi^2_6 = 17.84$ ,  $p = 0.007$ , respectively). Comparison of the yield effects among study areas revealed that in some countries, yield had negative effects on these variables, but in other countries there was no relationship (Fig. 2; Appendix A, Table 3). In two of the three study areas where we found no relationship, the variation in yield among fields and farms was much smaller than in the other countries, which probably explains the lack of significant effects. There were no consistent differences between West and East European countries.

As a second step, we investigated the relative importance of 13 variables we considered as relevant components of agricultural intensification (Appendix A, Table 4). The characteristics of the surrounding landscape had significant effects on wild plant species diversity only (Table 1). The number of plant species was inversely related to average size of fields within a radius of 500 m, emphasizing the importance of field margins for the establishment of wild plant species on arable land. The number of wild plant species declined as the frequency of herbicide and insecticide application and the amounts of active ingredients of fungicides increased (Table 1). The number of carabid species was negatively affected by the amounts of active ingredients of insecticide applied. Bird species diversity declined with increasing frequency of fungicide application, a variable closely correlated with the frequency of insecticide application (Pearson's correlation coefficient  $r = 0.732$ ;  $p < 0.001$ ). The predation on aphids declined

as the applied amounts of insecticides increased (Table 1).

Thirdly, we examined the effects of organic farming and the implementation of agri-environment schemes on biodiversity and the biological control potential. Organic farms comprised 22% of the total number of selected farms in our study and occurred in five of the nine study areas. These farms harboured more wild plant and carabid species (single effects of farm type:  $\chi^2_1 = 164.96$ ,  $p < 0.001$ ;  $\chi^2_1 = 3.98$ ,  $p = 0.046$ , respectively; Appendix A, Table 2), but not significantly more bird species ( $\chi^2_1 = 1.31$ ,  $p = 0.252$ ). Aphid survival was not significantly lower on organic farms as compared with conventional farms ( $\chi^2_1 = 2.93$ ,  $p = 0.087$ ). 45% of the selected farms had agri-environment schemes. These schemes had positive effects on the number of wild plant and carabid species and the predation of aphids (single effects of percentage of land with agri-environment scheme:  $\chi^2_1 = 51.97$ ,  $p < 0.001$ ;  $\chi^2_1 = 6.91$ ,  $p = 0.009$ ;  $\chi^2_1 = 13.24$ ,  $p < 0.001$ , respectively; Appendix A, Table 2), but not on bird species diversity ( $\chi^2_1 = 1.56$ ,  $p = 0.211$ ).

## Discussion

We studied the effects of agricultural intensification on biodiversity across three trophic levels and the potential for biological pest control in eight European countries. Out of the 13 studied components of agricultural intensification, use of pesticides, especially insecticides and fungicides, had the most consistent negative effects on the species diversity of plants, carabids and ground-nesting farmland birds, and on the potential for biological pest control. We conclude that despite several decades of implementing a Europe-wide policy intended to considerably reduce the amount of chemicals applied on arable land, pesticides are still having disastrous consequences for wild plant and animal species on European farmland. Importantly, this impact is also manifested as a reduction of the potential of natural enemies to control pest organisms.

It is noteworthy that both organic farms, which apply only those pesticides considered harmless to the environment, and agri-environment schemes had positive effects on plant and carabid diversity, but did not show the expected positive effects on bird species diversity. A possible explanation for the lack of such positive effects is the large spatial scale of the pollution associated with pesticide use across Europe, which inevitably leads to negative effects of pesticides – even in areas where the application of these substances has been reduced or terminated. Such large-scale effects will be especially relevant for taxa that utilize large areas, such as birds, mammals, butterflies (Rundlöf, Bengtsson, & Smith, 2008)

and bees (Clough et al., 2007; Holzschuh, Steffan-Dewenter, & Tschardtke, 2008).

We conclude that if biodiversity is to be restored in Europe and opportunities are to be created for crop production utilizing biodiversity-based ecosystem services such as biological pest control, a Europe-wide shift towards farming with minimal use of pesticides over large areas is urgently needed.

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## Author contributions

F.G., C.W., L.W.C., C.D., C.P., I.G., M.B.M., P.C., V.H., O.A., J.R., T.A., J.L., A.F., S.H., C.F., V.B. and M.P. collected the field data on plant, carabid and bird diversity and aphid survival; J.B., F.B., P.I., W.W.W., M.E., M.B.M., P.C., P.K., J.L., T.T., S.E., R.B., T.P., J.J.O. and C.T. designed the study and developed the protocol for data collection; P.W.G., F.B., J.B., F.G., P.I., W.W.W., M.E., R.B. and J.L. performed the analysis of the data sets; F.B., F.G., J.B. and P.I. wrote the manuscript. All authors discussed the results and commented on the manuscript.

## Appendix A. Supplementary material

Supplementary data associated with this article can be found in the online version at [doi:10.1016/j.baae.2009.12.001](https://doi.org/10.1016/j.baae.2009.12.001).

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