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Assessing the non-target effects of herbicides on field margin plant communities after controlling for soil, climate, local context and landscape metrics

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ABSTRACT

Pesticides are often identified as one of the major causes of biodiversity decline in farmlands. However, our knowledge about this relationship has mostly being inferred from small to landscape-scale studies, or from indirect indicators of agricultural practices at large scales. Here, we used a national network of more than 500 sites monitored yearly from 2013 to 2018 in France to assess the non-target effects of herbicides on field margin plant communities. We used hierarchical generalized linear models to investigate the effects of practices on plant species richness, plant species evenness, proportion of nature-value plants, and proportion of grasses in field margins, while controlling for a large number of possible confounding effects. The intensity of herbicide use had a negative effect on plant species richness, and on the proportion of nature-value plants. In the margin of cereal fields, there was a negative effect of dicotyledon herbicides on richness and a negative effect of grass herbicides on species evenness. We also identified, in some specific crops, a negative effect of non-herbicide treatments on margin flora richness and on the proportion of nature-value plants. The presence of surrounding grasslands had a consistent favourable effect on richness and on the proportion of nature-value plants in field margins. Finally, situations of risk for pesticides drift had a negative effect on margin flora. This study confirms that reducing herbicide use represents a robust lever to maintain the floristic diversity of field margins, which could be combined with strategies reducing the risk of pesticide drift.

1. Introduction

Since the beginning of the agricultural revolution in the mid-20th century, agricultural environments have been profoundly transformed (Stoate et al., 2001). These major changes have allowed a great gain in productivity and have improved working conditions, but also had strong environmental consequences, particularly on farmland biodiversity (Lécuyer et al., 2021). Declines of biodiversity in agroecosystems have been linked to landscape simplification, habitat loss, and increasing intensification of agricultural practices (Benton et al., 2003). Negative effects of pesticides have been shown for many non-target species: field margin's plants (e.g. Gove et al., 2007), birds (e.g. Chiron et al., 2014), non-pest arthropods including pollinators and crop auxiliaries (e.g. Gill et al., 2012; Schmidt-Jeffris et al., 2021), and soil biodiversity (e.g. Karimi et al., 2020). Pesticides also include indirect effects such as sub-lethal effects, which are well highlighted for arthropods, especially pollinators (e.g. Desneux et al., 2007), and indirect repercussions via trophic chains (e.g. decline of farmland birds due to massive decline of insect populations, Goulson, 2019, Stanton et al., 2018). Herbicides can also have indirect effects by weakening plant resistance to pathogens or by impacting pathogen communities (e.g. Johal and Huber, 2009) or competition between species (Damgaard et al., 2008). However, these studies have either remained fairly local in scale, or fairly constrained in terms of pesticide description (i.e. only considering the quantity of herbicide used). We are therefore missing an understanding of the large-scale effects of a broad range of pesticides on non-target

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biodiversity.

In very intensive agricultural landscapes, field margins represent the last refuge for many species, including plants and arthropods (Marshall and Moonen, 2002). These semi-natural habitats ensure the maintenance of various ecosystem services, in particular through the provision of habitats and resources for pollinators and arthropods involved in biological control of crop pests (Marshall, 2004). However, field margins can be strongly affected by agricultural management practices applied on adjacent fields, even if they are not their main target (Boutin and Jobin, 1998). For field margin plants, previous studies showed a preponderant effect of nitrogen fertilization, over-riding the effect of herbicides (Kleijn and Verbeek, 2000). However, this apparent small effect of herbicides may come from the diversity of practices regarding their use, which potentially masks their specific influence. It is thus a challenge to distinguish whether the effects are more a question of treatment frequency, date of application, or differences in the modes of action of these pesticides. However, margin plant communities may also be dependent on many determinants other than local agricultural practices, in particular soil types, landscape composition and structure, and field margin properties and management. Because these determinants co-variate with some practices, performing a conservative assessment of the impact of agricultural practices on non-target species require to account for these possible confounding effects.

Plant communities of field margins are affected by many factors at different scales (Concepción et al., 2008). At the regional scale, history and pedoclimatic conditions restrict the presence of species according to their ecological niche (Mahaut et al., 2020). At the landscape scale, the presence of semi-natural habitats such as grasslands (Lindborg et al., 2014), as well as landscape heterogeneity (Sirami et al., 2019), and the proportion of organic fields in the landscape, increase species richness in field margins (Bengtsson et al., 2005; Henckel et al., 2015). Sometimes these landscape effects can even exceed the impact of local management (Henckel et al., 2015), highlighting the importance of considering agricultural practices and landscape composition at larger scales. Farming practices at the field scale can also impact community assembly in the field margin as a consequence of pesticides and fertilizer drift, by eliminating sensitive species or altering species competitive relationships (de Snoo and van der Poll, 1999). Management of the field margin (mowing or herbicide treatment) also affect community composition by selecting species according to their tolerance to disturbances (Bassa et al., 2011). Still, few studies have considered the detailed impact of local agricultural practices while accounting for other determinants at multiple scales, probably, due to the difficulty of simultaneously gathering the different types of data.

In this article, we analysed a dataset from a national-scale, long-term monitoring network of 500 fields representative of the main national agricultural crops. This dataset combines annual standardized surveys of field margin plant communities with agricultural practices implemented in the field, and a large number of co-variables at multiple scales. We aimed at identifying and quantifying the non-target effects of agricultural practices, and more particularly, herbicides applications, on margin plant species after consideration of multiple possible confounding effects of local conditions, soil, landscape and field margin management. By integrating a fine description of the practices, we also intended to separate the effects of herbicide use intensity, date of application, and spectrum of action of herbicides on plant communities. We expected that the intensity of herbicide use would negatively affect plant species richness in field margins, and particularly the richness of nature-value species, while the structure of the community (percentage of grasses versus dicotyledonous plants) would depend on the spectrum of action of the herbicides used. We also hypothesized that if the date of application (e.g. time since the last treatment) influences plant communities, this would reflect the short-term, direct effects of herbicide applications

2. Material and methods

2.1. Study sites

The 500 ENI network was launched in 2012 by the French Ministry of Agriculture’s Ecophyto scheme to study the non-target effects of agricultural practices (particularly fertilization and pesticide use) on biodiversity at a national scale. This network consists of approximately 500 commercial agricultural fields spread throughout metropolitan France (Fig. S1). The fields are either arable crops, market gardening or vineyards, and 20 % of them are organic productions. The location of the fields were chosen through stratified sampling to be representative of the types of production in each of 22 French regions (Andrade et al., 2021). In each field, four taxa (margin plants, beetles, earthworms and birds) are monitored yearly, and all agricultural practices are recorded. Here we focused on margin plants, and analysed 518 fields of this network (23.7 % in organic farming) monitored yearly from 2013 to 2018. Over this period, a total of 540 fields were monitored but we had to discard 22 fields that had missing values for some agricultural practices data. All crops together, most of the field margins were monitored for 6 (64 %) or 5 (19 %) years, and few were monitored for only one year (3 %). We performed all analyses including all crops together, but also for three subsets of data corresponding respectively to either cereals, market gardening or vineyards (Table 1, and Table S1 for details per year). The subset “cereals” corresponded to a selection of margin plant surveys performed in years with effectively straw cereals grown in the field (mainly wheat, barley, triticale and spelt).

2.2. Plant surveys

Plants were surveyed on the field margins (Marshall and Moonen, 2002) defined as the first 1–6 m right outside the last row of crop, excluding crop edge (Fried et al., 2018). The field margin represents the uncultivated herbaceous vegetation area between the cultivated strip and another patch in the landscape (e.g. ditch, hedgerow, road, field or grassland). Conversely to the field core, field margins defined in this way include a large proportion of grassland species but few weeds. Plant species were identified in ten 1 m^2 (2 m x 0.5 m) quadrats located at the centre of the field margins (Fig. S1) following the protocol developed by the Vigie-Flore network (Martin et al., 2019) and adapting it to the context of a linear habitat. On each quadrat, presence-absence of each plant species was recorded, so that each species can be characterised by an abundance index ranging from 0 to 10 according to their occurrence over the ten quadrats. Plant surveys were conducted at the peak of flowering, between late April and early August depending on the region. To avoid detection bias between observers for uncommon species, within the 882 total observed species, we focused on the most abundant ones using the focal list of 186 species highlighted by Fried et al. (2018), i.e. species observed in more than 1 % of the field margins during the launch of the monitoring. Four response variables were computed: (i) species richness, (ii) species evenness (based on Pielou's evenness index; Pielou, 1966), (iii) the percentage of grasses (in terms of abundance), and (iv) the percentage of nature-value plants (in terms of number of species). Nature-value plants are defined in opposition to common agrotolerant species (Aavik and Liira, 2009), which correspond to species present with a frequency > 10 % in plots of agricultural fields. This classification was taken from a national weed survey covering the same regions as the 500 ENI network, and corresponding to species adapted to current disturbance regimes in arable fields, under modern conventional agriculture (see Fried et al., 2018 for details). Nature-value plants included both rare weeds and hemerophobic species (i.e., species that are sensitive to soil tillage and/or herbicides).

2.3. General conditions and time of observation

We included in the analysis the characteristics of the fields (altitude,

Table 1
Number of fields and fields-years according to the main type of crop and the type of management.

Type of crop	Total nb. of fields		Nb. of fields with data during:					
			1 year	2 years	3 years	4 years	5 years	6 years
All crops	518	(OF: 123)	16	25	18	28	99	332
Cereal	337	(OF: 73)	78	87	92	66	11	3
Market gardening	50	(OF: 15)	1	0	2	6	9	32
Vineyards	104	(OF: 32)	2	3	3	5	23	68

OF: number of organic farming fields. In addition to vineyards, market gardening and cereals, all crops include also other crops such as oilseed rape, sunflower, soybean, … Considering all crops leads to including a given field in the analyses for a higher number of years than when considering only cereals.

biogeographic region, exposure, area and spillover risk for pesticides and fertilizers), the date of the observation (Julian days since 1st January), and the year of observation. The biogeographic region corresponded to the "Végétal Local" delineation of biogeographic regions on the basis of climatic, topographical, geological and anthropic influence (Office Français de la Biodiversité, 2021). The exposure was noted as either "full sun" or "partially shaded". The spillover risk for inputs (index ranging from 0 to 2.5) was calculated as:

$$\textit{Spillover risk index} = \textit{wind exposure} + (\textit{field margin at the bottom of a slope} \times \textit{slope inclination})$$

with wind exposure and field margin at the bottom of a slope corresponding to two binary variables (0 or 1, notation by field observers) and slope index corresponding to 0 for a 0 % slope, 0.5 for 1–10 %, 1 for 10–20 %, 1.5 for 20 %. In addition, a quadratic term was included in the analysis for altitude and date to consider a potential non-linear effect.

2.4. Soil characteristics

We used data from the Soilgrids database (https://soilgrids.org/) at 250 m resolution and 5 cm depth (Hengl et al., 2014) to perform a PCA on eight variables (Fig. S2): soil nitrogen, organic carbon, sand, silt, and clay contents, bulk density, pH, and cation-exchangeable capacity. Axis 1 opposed acidic soils, with higher cation exchange capacity and more organic matter and nitrogen content (negative values), to clayey soils, more basic and with higher bulk density (positive values). Axis 2 opposed sandy soils (negative values) to soils with high silt content (positive values), see PCA in Fig. S2. These two first PCA axes were used in the analysis as a synthetic description of soil characteristics for each site.

2.5. Landscape characteristics

The landscape surrounding each field was characterized in a 1 km radius (a distance that was shown to be relevant for plant species in agricultural landscapes, e.g. Gabriel et al., 2005, Kirk et al., 2024), by the percentage of each one of 24 land cover types (Fig. S3). We then included in the analysis the percentage of winter cereal, of maize, of vineyard, and of grassland, and the Shannon diversity of crops in the landscape (using the 17 crop categories included within the 24 land use types). We also accounted for the percentage of organic farming in the landscape in a 1 km radius (CARTOBIO, Agence bio). Spatial data were treated using QGIS v. 3.14.16 and R v.4.0.1 with packages ALM (Allart et al., 2020) and sf (Pebesma et al., 2022).

2.6. Field margin characteristics and management

Field margins were described with their width, management (dummy variables: 'crushing, mowing and collecting' and 'crushing, mowing without collecting'), total number of interventions per year (summing all management events including mowing, herbicide or pasture), type of adjacent components (dummy variables: 'built area or road', 'crop', 'wood or hedgerow', 'grassland', 'grass strip', 'ditch, river or wetland') and diversity of adjacent components (number of different elements).

2.7. Agricultural practices

Information about agricultural practices was collected yearly through interviews with farmers (Andrade et al., 2021). To evaluate pesticide use intensity, we calculated the Treatment Frequency Index (TFI) (Lesueur-Jannoyer et al., 2015) at the field level as:

$$TFI = \sum\left(\frac{\textit{dose per ha}}{\textit{recommended dose per ha}}\right) \times \left(\frac{\textit{treated area}}{\textit{total field area}}\right) \times \left(\frac{\textit{treated area}}{\textit{total field area}}\right)$$

The TFI was computed separately for non-herbicides and herbicides, and averaged over the period of observation of each field. To account for the spectrum of action of herbicides, we calculated the TFI for four sub-categories: (i) broad-spectrum herbicides without crop selectivity (further referred as "non-selective herbicides", essentially glyphosate), (ii) broad-spectrum herbicides with good crop selectivity (further referred as "crop-selective broad-spectrum herbicides", corresponding mainly to sulfonylureas), (iii) dicotyledons herbicides and (iv) grass herbicides (see Table S2 for the list of herbicides included in each category and Table S3 for the list of all non-herbicides pesticides). In addition, to account for possible short-term direct effects of herbicide application on plant species, we included the number of days between the flora survey and the last treatment (further "time since last treatment", with a maximum value of 365 days), considering all herbicides together and each one of the four sub-categories of herbicides. We also included in the analysis the dose of N, P or K fertilisation (corrected by the equivalence coefficients for organic fertilization, which allows to calculate mineral quantities available to plants, see Appendix S1), the fertilisation type (dummy variables 'organic', 'mineral', 'organo-mineral', 'no fertilisation'), the dose of copper and sulphur, and the diversity of the crop sequence (number of different crops divided by the number of studied years).

2.8. Statistical analysis

To specifically analyse the effect of agricultural practices on the four margin flora response variables while accounting for all possible confounding predictors, we built a step-by-step hierarchical model selection process, using generalized linear mixed models. A summary of all explanatory variables is presented in Table S4. As already proposed in several studies in other contexts (for e.g. Braunisch et al., 2014, Alignier et al., 2013, Josso et al., 2013, Ricci et al., 2009), we used a multiple step model selection framework in which sets of predictors are sequentially included. Sets of predictors that could lead to confounding effects are included at first, while predictors of interest, namely agricultural practices, are included at the end (Fig. 1). The order in which the sets of predictors were added was from the most general to the most specific, based on existing knowledge about the drivers of plant margin communities (e.g. Poinas et al., 2023). The first model included only

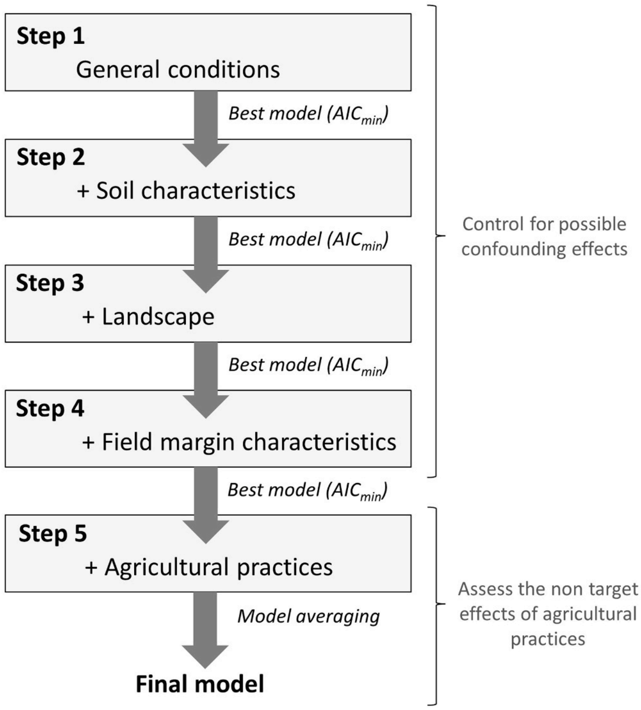

Fig. 1. Hierarchical model selection process to assess the non-target effects on agricultural practices on field margin plant communities while controlling for possible confounding effects.

predictors related to general conditions ('step 1'); then we successively added soil predictors ('step 2'), landscape predictors ('step 3'), field margin predictors ('step 4'), and finally agricultural practices ('step 5'). At each step, a model selection was performed using the pdredge function in R v.4.0.1 based on AIC. Among all equivalent models (delta AIC<2), we kept all the predictors present in the most parsimonious models and fixed these predictors for the next steps. This avoids keeping too many variables with a low explanatory power. Predictors that were highly correlated between them ($r > 0.7$) were never included together in a given model (see correlation plots in Fig. S4). In the last step ('step 5'), we performed a model averaging on all the best models (delta AIC<2 compared to the best model). In all models, the biogeographical region and the field identity were included as random effects. This whole procedure was performed for the four response variables (yearly values for species richness, species evenness, percentage of grasses, and percentage of nature-value plants) and the four datasets: (i) all crops, (ii) cereals, (iii) market gardening, and (iv) vineyards (Table 1). Finally, we evaluated in each case the quality of prediction of the averaged model by cross validation: we used k-fold validation, training the model on 90 % of the data and testing on the last 10 %, repeating this operation randomly 10 times and computing the R^2 by comparing the prediction (all repetitions) and the observed response variables (e.g. species richness).

Regarding agricultural practices, the predictors describing herbicide use that were added at 'step 5' were not the same for the four sets of crop types. In the case of all crops, we intended to identify global effects of herbicides on the margin flora, and thus included at this step the herbicides' TFI, the time since the last herbicide treatment, and the interaction between the herbicides' TFI and the time since the last herbicide treatment. Conversely, in the case of the three crop-specific datasets, specific types of herbicides may be used preferentially in different types of crops. Therefore, in these cases, we included the TFI of the four subcategories of herbicides (four predictors) and the corresponding times since the last treatment corresponding to that category (four additional predictors). It was not possible to account for the correlation between treatment frequencies and time since the last treatment when considering sub-categories of herbicides because the two predictors were always highly correlated and were thus not included together in the same model; they may nevertheless be present jointly in the final averaged models. All other predictors of agricultural practices mentioned above were always included in the procedure for each set of crops."

In order to be able to compare the relative importance of predictors, all predictors were standardized to have a mean of 0 and a standard deviation of 1. We used a negative binomial distribution (White and Bennetts, 1996) for models on species richness (glmer.nb function, lme4 package), a beta distribution for models on species evenness (glmmTMB function with beta family, glmmTMB package), a beta-binomial distribution for the percentage of grasses (glmmTMB function with betabinomial family, glmmTMB package) and a gaussian distribution to model the percentage of nature-value plants (lmer function, lme4 package). All statistical analyses were performed in R 4.0.1 (R Core Team, 2022; MuMIn, lme4 and glmmTMB packages). Because of the large number of statistical individuals, we considered a significant effect when the corresponding estimated coefficient did not include zero in its confidence interval rather than using the p-values.

3. Results

3.1. Main determinants of the margin flora characteristics

Despite all the factors considered in this study, the percentage of explained variance (EV) was always lower than 50 %: the R^2 of final averaged models ranged between 0.07 and 0.48 (Fig. 2), except in one case (evenness in market gardening, $R^2 < 0.01$). Overall, the general conditions added in the first step consistently represented more than 6 % of the EV, and up to 93 %, followed by field margin characteristics and management (between 0 % and 76 % of the EV), agricultural practices (between 6 % and 54 % of the EV) and landscape characteristics (between 0 % and 31 % of EV), while soil characteristics never contributed to more than 10 % of the explained variance. However, the type of predictors that mainly influenced the margin flora was highly variable depending on the response variable and the set of crop types (Fig. 2). Plant richness in margins was explained mostly by general conditions considering all crops together, but agricultural practices were the main determinants in cereal while field margin characteristics and management played a key role in vineyards and market gardening. Plant evenness was mainly driven by field margin characteristics in all crops, but agricultural practices were equally important in cereal (44 % for practices and to 51 % for margin characteristics) and vineyards (33 % for practices and 32 % for margin characteristics). The percentage of nature-value plants was mainly related to general conditions and field margin characteristics and management. Finally, the percentage of grasses was also determined mainly by the general conditions in all crops, and specifically in vineyard (93 % of the EV), whereas the main effects were the field margin characteristics in market gardening (76 % of the EV) and agricultural practices in cereal (53 % of the EV).

3.2. General conditions

Spillover risk had a negative effect on the percentage of nature-value plants (all crops together and cereal), the percentage of grasses (all crops together), richness and evenness (vineyard). In addition, when considering all crops together, small fields favoured plant species richness (Fig. 3).

3.3. Soil characteristics

Only few soil characteristics affected the margin flora (Fig. 3). There was a higher percentage of nature-value species in acidic soils with higher organic content and cation exchange capacity than in more basic soils with higher bulk density (all crops together, cereal and market

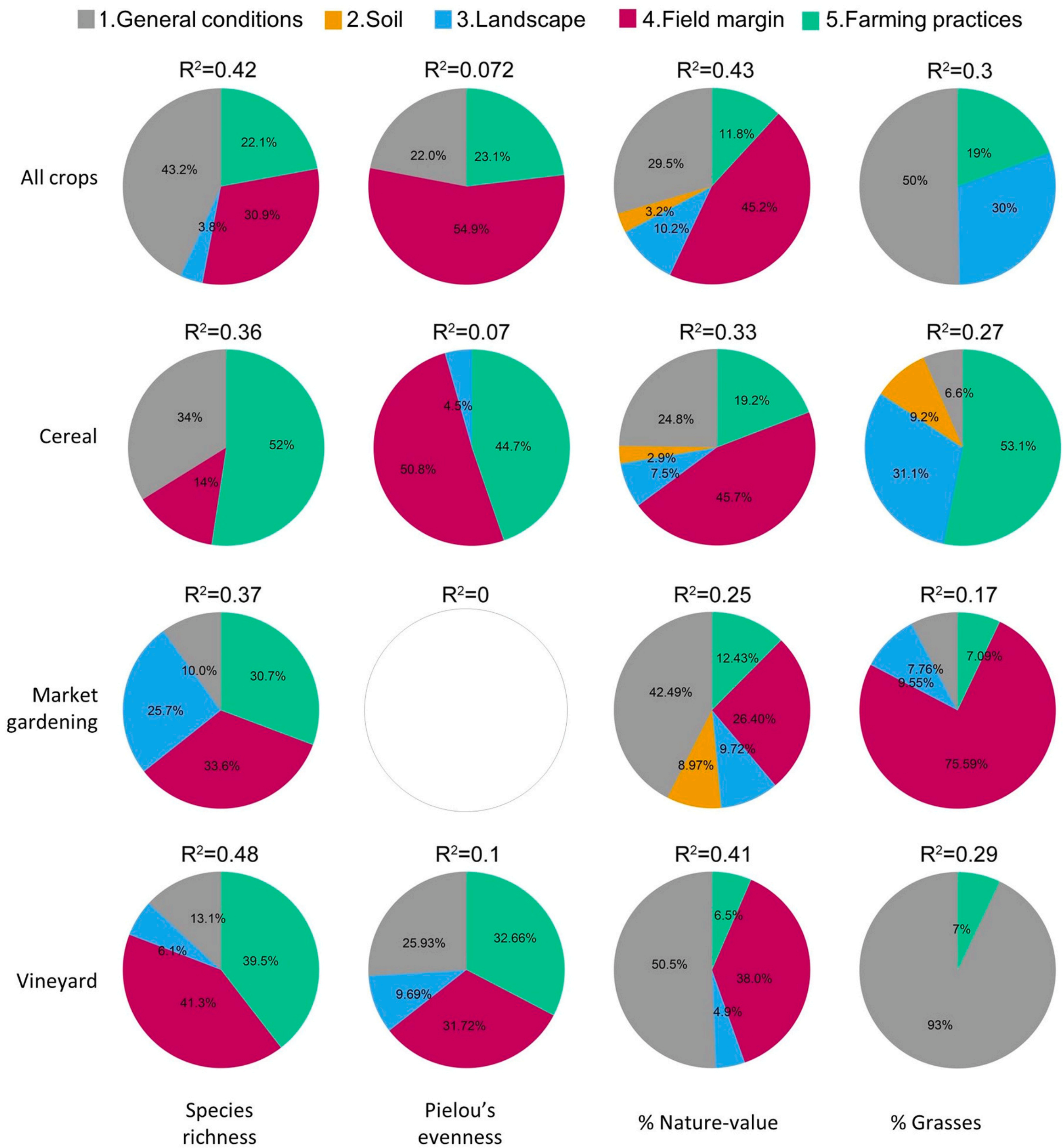

Fig. 2. Part of the variance explained by the different groups of variables. The percentages were calculated on the basis of standardized variables of the model (representation omitted in one case where the R^2 was too low). The R^2 was estimated by cross-validation, and gives an estimate of the predictive quality of the model (based on 10 randomisations with model train based on 90 % of the data and tested on 10 % remaining).

gardening; negative effect of soil PCA axis 1; Fig. S2). In cereal, there was also a higher percentage of grasses in soils with high silt content than in soils with high sand content (positive effect of soil PCA axis 2).

3.4. Landscape characteristics

The diversity of crops in the landscape increased the percentage of nature-value species in margins (all crops) and the richness (market gardening; Fig. 3). Field margins located in landscapes with a high percentage of grasslands had a higher percentage of nature-value species and a higher percentage of grasses (all crops together and cereal), and a higher evenness (vineyard). We also identified effects of specific types of crops in the landscape on the margin flora. In particular, field margins located in landscapes with a high percentage of vineyards had a lower richness (market gardening only) and a lower percentage of nature-value species (cereal only). In landscapes with a high percentage of winter cereals (that were also mostly composed of annual crops, with low perennial and woody components; Fig. S3), richness was lower (all crops) and the percentage of grasses was higher (all crops together and cereal). A high percentage of corn in the landscape was associated with a lower species richness (market gardening only) and a higher percentage of grasses (all situations except vineyard). Finally, vineyard surrounded by a high percentage of organic farming had a higher percentage of nature-value species.

3.5. Field margin characteristics and management

Larger margins had a lower species richness (all crops together and vineyard). Adjacent grasslands favoured species richness (all crops together and vineyard), and the percentage of nature-value species (vineyard) and decreased the percentage of grasses (market gardening). The presence of an adjacent crop also increased species richness (all

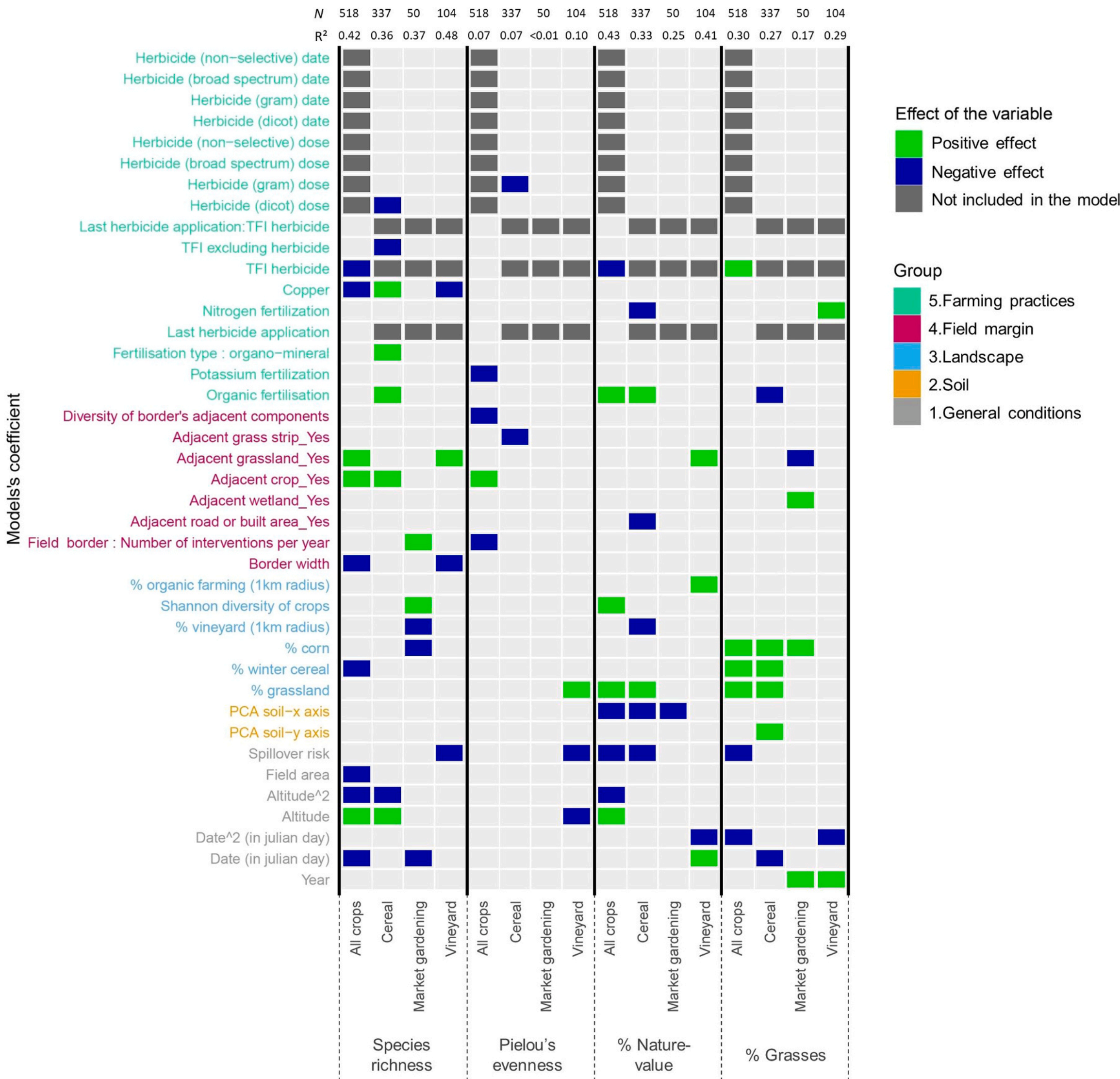

Fig. 3. Results of the averaged models: effects of the different predictors on the four margin flora response variables and for each dataset. Predictors that were not significant in any averaged model were omitted from the figure (see Fig. S5-S8 and Table S3-S6 for all estimates' values and confidence intervals). *N*: number of fields in the model; R^2: percentage of explained variance.

crops together and cereal) and evenness (all crops together). In addition, adjacent ditches, rivers or wetlands benefited grasses (market gardening only) while adjacent road and built areas disfavoured nature-value plants (cereal). A high number of adjacent elements (all crops together) or an adjacent grassy strip (cereal) decreases plant evenness (Fig. 3). Finally, a high number of management interventions decreased evenness (all crops together) and increased species richness but only in market gardening.

3.6. Herbicides and other agricultural practices

All crops confounded, there was a negative effect of herbicide's TFI on species richness, and percentage of nature-value species in field margins (Fig. 3), whereas herbicide's TFI was positively correlated with percentage of grasses. The time since the last herbicide treatment was never significant. No significant interaction between the herbicide's TFI and the date of treatment was observed. When focusing on herbicide types on specific crops, there was a negative effect of the dose of dicotyledons herbicides on richness in cereal. There was also a negative effect of the dose of grass herbicides on evenness in cereal, but the R^2 of this model was low (0.07). We did not identify any specific effects of non-selective herbicides or of crop-selective broad-spectrum herbicides. In addition, the time elapsed since the last treatment, whatever the specific type of herbicide, had no effect. Regarding the effects of pesticides other than herbicides, non-herbicide TFI had a negative effect on species richness but in cereal only. Finally, copper treatments negatively influenced species richness (all crops together and vineyard) while it was associated with higher species richness in cereal.

Regarding fertilization, the dose of nitrogen decreased the percentage of nature-value species richness (cereal) and favoured grasses (vineyard) while the dose of potassium decreased evenness (all crops together). Organic fertilization increased species richness (cereals) and the percentage of nature-value species (all crops and cereal), and decreased the percentage of grasses (cereal). Finally, organo-mineral fertilisation was positively correlated with species richness in cereals.

4. Discussion

In this study, we investigated the effects of agricultural practices on field margin flora using a large dataset covering a national extent and up to six years of monitoring. To focus specifically on the contribution of agricultural practices on field margin plant richness, evenness and type of plant species, we performed a conservative procedure that first accounts for a high number of covariables to reduce the possibility of confounding effects. Within this conservative framework, considering all crops together, agricultural practices as a whole represented about 20 % of the explained variance for field margin flora richness, and the same level for evenness. We found a negative effect of the intensity of herbicide use on species richness, with an even stronger impact on the percentage of nature-value species. When refining by different types of crops and types of herbicide, we mainly identified a negative effect of dicotyledons herbicides on richness and a negative effect of grass herbicides on evenness, both specifically in cereal field margins. The specific impact of dicotyledons herbicides is consistent with the the findings of (De Snoo & van der Poll 1999) who highlighted a decrease only in dicotyledon plants (and not monocotyledon plants) at the margin of cereals that were most frequently sprayed with MCPA, metsulfuron-methyl, fluroxypyr, mecoprop and bentazone, that is, herbicides targeting only dicotyledons. Across all crops, we also found that higher total herbicide treatment frequency (TFI) was associated with a higher relative abundance of grasses compared with dicots. This pattern does not necessarily confirm an anti-dicot effect, as it reflects the overall herbicide pressure rather than the specific action of dicot-targeting herbicides. However, if dicotyledons herbicides have more pronounced non-target effects on the adjacent flora, coupled with a general tendency of herbicide treatments to promote the prevalence of grasses, this raises the concern that herbicide pressure may ultimately impact the pollination service provided by field margins (Bohnenblust et al., 2016, Rotchés-Ribalta et al., 2023). On the other hand, the dose of grass herbicides reduces evenness, i.e. favours the dominance of certain species. This seems quite logical as grass herbicides selectively target grass species, but it suggests that repeated, high doses of grass herbicides in the field may have an unintended effect right up to the field margins. The elimination of grasses may allow certain unaffected dicotyledonous species to take their place, thus reducing the balance of species present. Alternatively, grass herbicides could also favor resistant grass weeds (e.g., *Alopecurus myosuroides*, *Lolium multiflorum*, …) that may become dominant in these field margins. In both cases, this reduces equitability by favouring the dominance of a few competing species. Evidence suggesting an impact of cultural practices on farmland floristic diversity have been mounting over the past decade. For example, Ryan et al. (2010) highlighted long-term effects of management systems on plant community assembly, which can act as a filter over relatively long-time scales. However, these studies were fairly local and our results reinforce these findings at a large extent and accounting for multiple possible confounding effects.

Interestingly, we did not identify any effect of the time since the last herbicide treatment, which indicates that the effect of herbicide use intensity predominates over the timing of the most recent application. This outcome was expected, as herbicide drift to field margins means only a fraction of the herbicide reaches the plants, and the impact of possible herbicide runoff may occur after a certain delay. Consequently, we anticipate a cumulative impact over the medium to long term, driven mainly by reduced reproductive success and a decline in the long-term persistence of perennial species, which will eventually lead to the extinction of the population, rather than causing an immediate lethal effect from the current year's application. Our results therefore confirm that reducing herbicide use intensity is a key lever to maintain margin flora richness and equitability. This can be achieved through technological improvement such as precision agriculture technics that allow targeted spraying, although achieving a significant pesticide use reduction will probably require to adapt or even redesign agricultural cropping systems (Jacquet et al., 2022).

We did not detect a specific effect of non-selective herbicides (mostly glyphosate) on the margin flora. However, the treatments carried out during the inter-cropping period (a period when numerous glyphosate treatments occurred) are poorly informed in the database used, because field observers focus mainly on the cropping season, which could have weakened our capacity to detect such effects.

In cereal crops, we also found negative effects of pesticides other than herbicides on margin flora species richness, in accordance with other studies that showed similar effect for copper (Strandberg et al., 2006) or other pesticides (Serra et al., 2020), indicating that pesticides as a whole can also impact non-targeted plant species. However, contrary to other studies that have shown a predominant effect of fertilisation on plant richness compared to the impact of herbicides (e.g. De Schrijver et al., 2011), we did not observe a strong effect of fertilisation. Nevertheless, we found a consistent effect of the nitrogen dose that was particularly negative on nature-value species (i.e. non-agricultural species that react less well to high nitrogen inputs or are subject to increased competition from better-adapted species). Similarly, the effect of the potassium dose reduces evenness, which means that here again it only benefits the abundance of some species to the detriment of others. However, the fertilisation type had larger effect than the dose of N, P and K: organic or organo-mineral fertilization promoted species richness and favoured nature-value species compared to mineral fertilization, as previously found in grasslands (e.g. Mauchamp et al., 2016). Indeed, organic fertilization release nutrients at a slower rate compared with mineral fertilizers (Shaji et al., 2021) which are often only advantageous to a limited number of ruderal species that can quickly access nutrients and gain a competitive advantage following a pulse in resource supply (Liebman and Davis, 2000).

For agricultural operations conducted inside the field to have an influence on margin flora, spatial population dynamics connecting the field core and the margin or spillover of input application need to be involved. Here, we clearly identified that margin flora was influenced by the spillover risk, calculated as a function of wind exposure and field configuration. The spillover risk reduced the proportion of nature-value species and the proportional abundance of grasses. It also negatively impacted both richness and evenness in vineyards, which is consistent with the fact that this crop is often grown on slopes and the margins located at the bottom of the slopes may more easily accumulate streams of inputs. Although these results suggest effective events of pesticides or fertilizers spillover from the field to the margin, with consecutive negative effects on flora, the precise processes underlying these effects need to be more deeply investigated. We found that smaller fields had higher margin flora richness, a result that may arise from large scale species dynamics, as configurational crop heterogeneity at the landscape scale which increases when there are numerous small fields) have been shown to favour plant diversity (but evidences for this effect mostly concerned inside-field flora, see Alignier et al. (2020). At the smallest scale, we found that larger margins had a lower species richness, which could result from the fact that the width of the survey area was fixed, and the quadrat always positioned in the middle of the field margin. Thus, the survey area was closer to the field in small margins with a possible mixing of perennial/grassland species and annual/weed species, while large margins were further from the field with a dominance of a smaller number of grassland species. The fact that a high number of management interventions increased species richness in market gardening crop margins could be explained according to the same principle. The total absence of management could lead to the most competitive perennial species being favoured, whereas an increase in the number of management interventions (up to a certain point) could reduce competition and allow the presence of annuals in the vegetation gaps, in accordance with the intermediate disturbance hypothesis (Connell, 1978), although this direction of the relationship is rather expected in ecosystems with very little disturbance (Müller et al., 2014). However, the fact that the number of interventions in general (in the all crops model) reduces evenness suggests that the main effect of increasing the number of interventions is to favour the abundance of a few species adapted (in this case to repeated mowing) to the detriment of others.

Finally, nature-value species of cereal margins were disadvantaged by the proximity of a built area or a road, while they were favoured in vineyards by the presence of grasslands, both in the immediate neighbourhood or at larger landscape scale, possibly because these perennials semi-natural habitats create particular pedo-climatic conditions favourable to these species or provide propagule sources (Lindborg et al., 2014). This contribution of grassland presence in the landscape to the structuring of field margin flora is consistent with Fried et al. (2024). Interestingly, the proportion of nature-value species also increased with the diversity of crops at the landscape scale. The diversity of crops at large scale is one component of complex landscapes that are associated with higher biodiversity levels (e.g. Estrada-Carmona et al., 2022), but we show here the specific positive effect of this metric on nature-value species of field margins. This effect held in vineyard margins, where nature-value species were also favoured by the proportion of organic farming in the surrounding. This landscape metric has already been identified as increasing species richness in field margins (Bengtsson et al., 2005; Henckel et al., 2015), although this relationship did not arise from our analysis, possibly because the percentage of organic farming is too low to detect its effects in all situations, or because this effect is masked by local practices.

As all studies based on large datasets, some possible limitations need to be underlined. As mentioned earlier, we know that inter-cropping treatments that often include glyphosate application are not always informed in the database; yet, the repeated use of this type of non-selective herbicides could significantly affect margin flora. The effects of glyphosate on margin flora therefore remains an open question. In terms of methods, although we have chosen a statistical framework capable of accurately characterizing the effects of practices on the margin flora by accounting for possible confounding effects, our approach is still correlative and our results require to be interpreted in the light of other studies. In addition, the choice of the order in which the sets of predictors are included may influence final models. This order was chosen based on the literature and on previous studies in the same system, which explored a wider range of environmental predictors at different spatial and temporal scales (e.g. Fried et al., 2018). The consistency of these results with previous ones suggests that the herbicide effects that remain after controlling for all other confounding factors represent a robust trend. Indeed, the overall effects of herbicides were either the largest among the impacts of various farming practices or ranked second, just after fertilization effects. The importance of unintended pesticide effects on field margin flora could have broader implications in the case of transfer and propagation of effects along the trophic chains in both terrestrial and aquatic ecosystems (Faburé et al., 2025).

5. Conclusion

This study confirms that reducing pesticide use, and particularly herbicides, is a robust lever to maintain the floristic diversity of field margins, and thus the whole trophic chain, which depends on it. In addition, minimizing pesticide drift from the field to the margin, for example through spray-drift limiting devices, could also be very effective. More generally, acting on management practices within agricultural fields seemed to be more efficient than modifying management of field margins in promoting plant diversity. Diversified margin potentially harbour species with diversified functional roles, and support multiple ecosystem services. Increasing the floristic diversity of field margins also limits the risk of hosting species that are highly competitive with crops and that could spread into the fields. However, determinants that favour richness or nature-value plants were not always the same, particularly when considering the characteristics and configuration of field margins. Explicit functional approaches should be applied to identify how to promote specific ecosystem services such as pollination or the provision of habitats for natural enemies of crop pests.

CRediT authorship contribution statement

Benoit Ricci: Writing – review & editing, Writing – original draft, Validation, Supervision, Project administration, Methodology, Funding acquisition, Formal analysis, Conceptualization. **Christine N. Meynard:** Writing – review & editing, Writing – original draft, Validation, Supervision, Methodology, Formal analysis, Conceptualization. **Isis Poinas:** Writing – review & editing, Methodology. **Jean-Philippe Guillemin:** Writing – review & editing, Validation, Supervision, Methodology, Data curation. **Guillaume Fried:** Writing – review & editing, Writing – original draft, Validation, Supervision, Project administration, Methodology, Funding acquisition, Formal analysis, Conceptualization. **Laura Henckel:** Writing – original draft, Methodology, Formal analysis, Data curation, Conceptualization.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.agee.2025.110190.

Data availability

The dataset is available at https://doi.org/10.57745/HORXAJ

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