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ORIGINAL RESEARCH

Improving insect conservation management through insect monitoring and stakeholder involvement

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Abstract

In recent years, the decline of insect biodiversity and the imminent loss of provided ecosystem functions and services has received public attention and raised the demand for political action. The complex, multi-causal contributors to insect decline require a broad interdisciplinary and cross-sectoral approach that addresses ecological and social aspects to find sustainable solutions. The project Diversity of Insects in Nature protected Areas (DINA) assesses insect communities in 21 nature reserves in Germany, and considers interactions with plant diversity, pesticide exposure, spatial and climatic factors. The nature reserves border on agricultural land, to investigate impacts on insect diversity. Part of the project is to obtain scientific data from Malaise traps and their surroundings, while another part involves relevant stakeholders to identify opportunities and obstacles to insect diversity conservation. Our results indicate a positive association between insect richness and biomass. Insect richness was negatively related to the number of stationary pesticides (soil and vegetation), pesticides measured in ethanol, the amount of area in agricultural production, and precipitation. Our qualitative survey along with stakeholder interviews show that there is general support for insect conservation, while at the same time the stakeholders expressed the need for more information and data on insect biodiversity, as well as flexible policy options. We conclude that conservation management for insects in protected areas should consider a wider landscape. Local targets of conservation management will have to integrate different stakeholder perspectives. Scientifically informed stakeholder dialogues can mediate conflicts of interests, knowledge, and values to develop mutual conservation scenarios.

Keywords Insect decline · Metabarcoding · Pesticides · Vegetation · Societal dialogues · Conservation practice

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Introduction

The presently documented declines in insect diversity and biomass affect essential ecosystem services and will result in serious economic consequences (Bang et al. 2005; Cardoso et al. 2020; Filser et al. 2016; Kremen et al. 2007; Noriega et al. 2018; Ollerton et al. 2014; Schowalter et al. 2018; Slade et al. 2016). Numerous studies worldwide have documented insect diversity and biomass loss throughout different habitats and even in nature reserves (Cardoso et al. 2020; Hallmann et al. 2021a; Klink et al. 2020; Seibold et al. 2019; Wagner 2020). This results in the critical need to develop strategies for preventing further insect decline. However, the cause of insect decline is multi-faceted and can only be understood by integrating ecological complexity with patterns of anthropogenic land use. This calls for inter- and transdisciplinary approaches to develop solutions for averting insect decline.

The intensification of agriculture has been identified as one of the most important drivers of insect decline (Cardoso et al. 2020; Seibold et al. 2019). In addition to agriculture practiced within protected areas, several studies have shown that the influence of intense agricultural use on biodiversity are not limited to the cultivated areas, but also radiate into neighbouring biotopes, namely nature reserves and other sensitive areas (Brühl et al. 2021; Le Provost et al. 2020; Zaller et al. 2022). To reverse this trend, new pesticide regulations and restrictions have been issued (e.g., European Commission ban of neonicotinoid insecticides 2018, German restrictions on pesticides in protected areas 2021), which influence some farming practices and operational management. Thus, the matter of regulations of conventional agricultural practices is embedded in a broader, cross-sectoral discourse on balancing agricultural productivity against conservation targets and actions to counteract biodiversity decline (Fickel et al. 2020). This requires the development of dynamic agricultural practices that protect critical biodiversity while also respecting the economic interest of farmers (Perino et al. 2022). An inter- and transdisciplinary social-ecological approach is required to achieve transformation knowledge (Hummel et al. 2017) and to move towards an integration of insect biodiversity conservation in sustainable agricultural practices.

Science-policy-society interfaces for biodiversity conservation, such as IPBES (the Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services) on the global (Díaz et al. 2015) and EKLIPSE (European Knowledge and Learning Mechanism to Improve the Policy-Science-Society Interface on Biodiversity and Ecosystem Services) on the EU level (Watt et al. 2019), have shown that changes in political and legal frameworks can be achieved most effectively by a two-way process: first, mutual learning by integrating practitioners' knowledge via their participation in knowledge assessments (Tinch et al. 2018), and second, opening the scientific process to stakeholders, including the steps of data acquisition, data interpretation, and formulation of policy recommendations, to include multiple perspectives and resolve potential conflicts of interest (Mehring et al. 2017; Watt et al. 2019; Young et al. 2014). This co-operative process of "transdisciplinary knowledge integration" is key for developing science-based policies that are suitable for implementation and to ensure acceptance by practitioners in agriculture and conservation.

Alarming numbers of insect declines (Hallmann et al. 2017) led to the development of the inter- and transdisciplinary research project DINA (Diversity of Insects in Nature protected Areas) (Lehmann et al. 2021). The project aims to quantify insect biodiversity in and around 21 selected protected nature conservation sites in Germany ("Naturschutzgebiete": NSG) designated by the German Federal Agency for Nature Conservation (Bundesamt für Naturschutz). They meet the criteria of an International Union of Conservation of nature

(IUCN) Category IV Habitat and Species Management Area and will be referred in the following text as nature reserves in line with international use. The use of Malaise traps not only enables sampling flying insects, but also allows the identification of plant traces and pesticide residues carried into the traps by insects from the ethanol used for preservation. Geo-information, vegetation surveys, pesticide residues in the environment (soil and vegetation), and weather data collected in parallel with Malaise trapping provide insight into their possible influences on flying insects. In addition to the acquisition of environmental data, the DINA project takes a broad stakeholder involvement approach. This includes three major components: a thorough stakeholder analysis including the identification of obstacles for agricultural measures for insect conservation in and around protected areas. Surveys with farmers on institutional frameworks and individual engagement for nature conservation, the development of a local science-policy-society interface to involve local stakeholders into a mutual exchange of knowledge to develop local solutions to conservation needs ("Social Labs" in the following). Further, to develop recommendations for actions and for implementing measures in nature conservation-relevant fields, it is necessary to analyse institutional frameworks, identify relevant stakeholders, and incorporate their broad range of perspectives by obtaining insight into the plurality of concerns, drivers and needs. Depending on the scope of problem, this may include stakeholders at local and regional as well as national and international levels (Raum 2018).

By combining this unique inter- and transdisciplinary design, DINA identifies causes of insect biodiversity loss, develops ideas to counteract insect declines, and considers social factors in the decision-making of stakeholders in agriculture and conservation (Lehmann et. al. 2021). To enable an evidence-based optimisation of protected area management, we assess species composition at selected sites, unprecedented in its completeness for flying insects due to the use of DNA metabarcoding and relate it to floral (Swenson et al. 2022) and spatial characteristics of the nature reserves and potential pesticide residues. By drawing on a complete set of biodiversity monitoring data, ecotoxicology data (Brühl et al. 2021) as well as spatial data on agricultural land-use (Eichler et al. 2022), this study presents a comprehensive analysis of impacts on insect biodiversity in nature reserves.

Material and methods

Within the project DINA, the insect species diversity and insect biomass were investigated in 21 nature reserves in Germany. The sample sites were selected following a spatial analysis of all 8836 nature conservation areas. Landscape indicators were evaluated based on GIS analyses and sites were preselected that met our requirements for grassland-dominated habitat types with adjacent or integrated arable land. According to the potential cooperation with local authorities and landowners we finally assigned the 21 study sites (see Online Resource 1).

Malaise traps were used for mass sampling of insects, building on the experience and recommendations of previous studies (Sorg et al. 2019). Transects at each of the 21 sites were equipped with five Malaise traps, starting on agricultural production area (trap 1), the transition zone (trap 2) and reaching into the protected area (traps 3 to 5) with fixed distances of 25 m between each trap. With 21 areas and five traps per transect a total of 105 traps were run in parallel. The traps operated continuously throughout the months April to September 2020 and 2021 with a two-week collection interval, in order to provide phenological data and the potential to capture species with short flight times (for details on design see Lehmann et al. 2021). DNA metabarcoding used for insect species and plant traces identification, which is complemented by vegetation surveys, geospatial and pesticide residue analyses. Apart from vegetation surveys, geospatial analyses, and societal input, all data presented here, were derived from the Malaise traps, were collected in a two-week interval in the end of May (May 16 to June 2, 2020, see Online Resource 1). This interval was chosen as an example because it covers a complete data set to answer different research questions demonstrating the benefits of combined sampling and data analysis. To assess the willingness and capacity of farmers at the study sites to engage in insect conservation, a survey was conducted with a mixed method questionnaire. More detailed societal input on the individual motivation and impediments for insect conservation management in agricultural fields in an around protected areas has been retrieved from the dialogue series at two of the study sites engaging stakeholders such as local authorities, policymakers, and farmers.

Landscape analysis

With the Digital Land Cover Model Germany (LBM-DE) 2018 of the Federal Agency for Cartography and Geodesy (BKG), we extracted the amount of area in agricultural production and nature reserves in the surrounding of the 21 selected conservation areas, the German category of nature-protected areas (NSG) and the protected areas under the FFH directive of the EU (Eichler et al. 2022) (Online Resource 2, Landscape analysis). To obtain information on the proportion of the agricultural production area and nature conserved area within a radius of 500–3500 m, the transect consisting of traps 1 to 5 was buffered by 500 m, 1000 m, 1500 m, 2000 m, 2500 m, 3000 m, and 3500 m respectively.

Climate data

Data from the German Weather Service (DWD) were analysed for maximum temperature and maximum precipitation per day, including data from March 1 to May 30, 2020 (except for beginning of June in one case). For each site, the nearest weather station was identified to collect information, with distances ranging from 2.5 to 22.8 km from Malaise trap locations (Online Resource 1).

Insect monitoring

The Malaise traps used were produced in a standardized way by the Entomological Society Krefeld and installed in the field following a defined protocol (Hallmann et al. 2017; Ssymank et al. 2018) to ensure data comparability with past and future insect monitoring studies. Flying insects were collected into 1000-ml polyethylene bottles which were emptied on average every 14 days. Insect biomass was assessed following standardized protocols (Hallmann et al. 2017; Ssymank et al. 2018), with the exception that pure 96% ethanol was used to optimise DNA preservation. The ethanol was further used for plant metabarcoding and pesticide analyses.

Insect metabarcoding

To avoid underrepresentation of small-bodied insect species, each sample was fractioned into two size classes referred to as S (small, < 4 mm) or L (large, > 4 mm). Resulting subsamples were separately dried until complete evaporation of ethanol, homogenised and lysated. We combined lysates of both size fractions subsequently merged in constant proportions (90% of size class S with 10% of size class L (Elbrecht et al. 2021)). DNA was eluted and quality checked on an agarose gel. On a 96 well spin column plate, 84 tissue samples were processed and complemented with 12 negative controls (only lysis buffer (ATL, Qiagen) and 10% Proteinase K (Qiagen)). A two-step PCR protocol was applied using standard Illumina Nextera primers for dual indexing of samples, using fwhF2 forward (Vamos et al. 2017) and Fol_degen_rev reverse (Yu et al. 2012) primer respectively. Library concentration was measured with a Quantus fluorometer (Promega, Madison, USA) and on a Fragment Analyzer (Agilent Technologies, Santa Clara, CA, USA) and the pool was sent for sequencing on a Novaseq SP platform at CeGaT GmbH (Tübingen).

Data analysis was conducted on merged paired-end reads with a length of 303–323 bp as implemented in JAMP v0.78 (Elbrecht 2019). Taxonomic assignment of molecular units was conducted by comparison with an Arthropoda reference database, created by a beta version taxalogue with sequences from BOLD (Barcode of Life Data System) (Ratnasingham and Hebert 2007, 2013), NCBI GenBank (Clark et al. 2016) and GBOL (German Barcode of Life) (Geiger et al. 2016) and at least 85% sequence similarity (for full outline see Online Resource 2, Insect metabarcoding).

Vegetation surveys

Plant species diversity, composition and surface cover including mosses and lichens was recorded in 3.50 m^2 quadrats next to each applied Malaise trap (Hallmann et al. 2017; Ssymank et al. 2018). Degree of plant species coverage was scaled by the method of Braun-Blanquet (1964) modified by Barkman (Reichelt & Wilmanns 1973; Wilmanns 1998). For additional information on plant diversity in the immediate vicinity species occurring 50 m around the traps were recorded without frequency data (Hallmann et al. 2017; Ssymank et al. 2018).

These methods and their application are consistent with the standards established by the Entomological Society Krefeld for collecting metadata in biodiversity studies using Malaise traps (Hallmann et al. 2017; Ssymank et al. 2018).

Plant metabarcoding

Following the extraction of insects, 150–250 ml original collection ethanol was vacuum filtered, in a biosafety cabinet with sterile, DNA free equipment. DNA was extracted with Nucleomag 96 Plant Kit (Macherey Nagel, Oesingen, Switzerland) and transferred to a 2 ml microcentrifuge tube and then homogenised for 2.5 min on Mixer Mill MM 400 (Retsch GmbH) at 30 Hz. Following homogenisation, samples were centrifuged for 10 min at 14,600 rpm after which, 250–300 ml of lysate was transferred to clean tubes. For the elution step, 35 ml of elution buffer MC6 were added, and 25 ml were removed for further processing and 2 ml for DNA quantification with Qubit 4 fluorometer

(Thermo Fisher Scientific Inc.). DNA extraction blanks (3 replicates) and PCR negative controls (3 replicates) were included to account for contamination, and a four species (*Ambrosia artemisiifolia*, *Fagus sylvatica*, *Lilium longiflorum*, *Plantago lanceolata*) mock community positive control (3 replicates) was included to validate the efficacy of laboratory protocols.

Amplification was performed with an adaptation of the Canadian Centre for Barcoding Platinum® Taq Protocol (Ivanova and Grainger 2007). Universal plant specific ITS2 primers were used (Kolter and Gemeinholzer 2021). Following PCR cycling, the three replicates were combined by the addition of 5 µl of each replicate for a total volume of 15 µl and purified with Thermo Scientific™ Exonuclease 1. The pooled replicates of non-indexed PCR products were sent to LGC Genomics GmbH where indices were added, and sequencing was performed with the Illumina MiSeq protocol in the 2 × 300 bp format.

Sequencing data were processed with USEARCH (Edgar 2010) and DADA2 (Callahan et al. 2016) using R (R Core Team 2019). The resulting ASVs were identified by implementing the SINTAX algorithm (Edgar 2016) using a custom Plantae reference database (Swenson et al. 2022) (for full outline see Online Resource 2, Plant metabarcoding).

Pesticide analysis

At the 21 sampling sites, soil and herbaceous vegetation was sampled in the proximity of each of the five Malaise traps. Eight individual samples of soil (sampling depth, 0–5 cm) and the aboveground vegetation were retrieved from a 3.5 × 3.5 m area and combined for a composite soil and vegetation sample, respectively. On agricultural fields, a composite weed and crop sample was taken at the Malaise trap 1, whenever available. After collection, samples were stored cooled in labelled plastic bags and shipped within 48 h to the laboratory in Landau. There, soil and vegetation samples were stored at − 20 °C until further sample preparation and LC–MS/MS analysis. Ethanol samples for pesticide analysis originated form Malaise traps, where ethanol was used to preserve trapped insects.

Pesticide analysis of ethanol was performed according to Brühl et al. (2021). Briefly, ethanol samples (50 ml) were evaporated under a gentle nitrogen stream until complete dryness for pre-concentration, then redissolved in methanol (1 ml), vortexed (60 s), filtered (0.2 µm), and analysed. Soil and vegetation samples were extracted prior to analysis according to Bakanov et al. 2022 (unpublished data). Briefly, 1 and 5 g of lyophilized soil and vegetation sample, respectively, was milled (vegetation) and sieved (soil, mesh size 2 mm), and extracted on an overhead rotator (60 min) using acidified acetonitrile with ammonium formate. Extracts were centrifuged, the recovered supernatant filtered and analysed. Vegetation samples were additionally purified after extraction using graphitized carbon black to reduce the effect of coextracted matrix.

For analytical determination we used a high-performance liquid chromatography-tandem mass spectrometer (HPLC–MS/MS; LC: Agilent Technologies LC 1260 Infinity series, MS/MS: Agilent Technologies 6495C, Waldbronn, Germany; Brühl et al. 2021). The selection of target pesticides (92 for ethanol; 98 for soil and vegetation) based upon (1) records of the JKI (Julius-Kühn-Institute) on common CUPs in Germany used in winter wheat, oilseed rape, maize, potato and wine grapes in 2016 and 2017 (JKI 2019); (2) pesticides registered recently (2015–2016); and (3) pesticides regularly detected in German small streams (Liess et al. 2021).

Statistical analysis of biodiversity and environmental data

For statistical processing, insect taxa lists were filtered to OTUs (operational taxonomic unit) and identified to species level, hereafter referred to as "insect species richness". For each site, OTUs for the same species were merged to avoid double counting. The same procedure was used for the plant lists generated by metabarcoding, hereafter referred to as "plant richness metabarcoding". The lists of all five traps per location are merged to create a list of all species found at a respective location. Insect abundance and plant cover were not calculated or analysed as these variables were not directly measured and are difficult to estimate from DNA sequence data.

Pesticide residues of the five traps were merged and traces of pesticides in vegetation and soil samples have been combined, hereafter referred to as "stationary pesticides". Only the number of different pesticides was used in this study, not their concentration. To allow for comparisons with a previous study, where the correlations between pesticides in the ethanol of Malaise traps and the area of agricultural use had been investigated, a 2000 m radius was chosen to include the area of agricultural production and the area of nature reserves (Brühl et al. 2021).

To quantify if there was a linear relationship between insect richness and biomass of insects, we calculated a Spearman correlation. Later, the two variables were used as response variables.

The nature reserves area within a radius of 2000 m, the amount of agricultural production are within a radius of 2000 m, plant richness determined by on-site screenings, species-level plant richness derived from metabarcoding, number of stationary pesticides, pesticide residues in ethanol, daily maximum temperature in °C, and hourly precipitation were checked for correlations among each other. Variables that correlated with each other and exceeded a threshold of $|r| > 0.5$ were not included in the same model to avoid potential (multi-)collinearity, the others were used as explanatory variables. To analyse the impact of the explanatory variables on the response variables, insect richness and insect biomass, generalized-linear-models (GLM) were used. Insect richness was negative binomially distributed, while insect biomass was normally distributed. We thus used negative binomial GLMs for insect richness (glm.nb function in the MASS library; Venables and Ripley 2002) and Gaussian GLMs for insect biomass. The model formula was

$$\begin{aligned} Response \sim\ & nature\ reserves\ area\ within\ 2000\,m + plant\ richness\ field\ survey \\ & + Plant\ richness\ metabarcoding + Stationary\ pesticides \\ & + Pesticides\ in\ ethanol + Precipitation \end{aligned}$$

where "Response" indicates the response variable; alternative models were constructed with agricultural production area within 2000 m replacing pesticides in ethanol.

We employed the R package sjPlot to plot the predicted marginal effects from the GLM models (Lüdecke 2022).

All statistical analyses were conducted using Rx64 4.0.1 (R Core Team 2019).

Questionnaire with participating farmers

In total, 33 farmers participated in the 21 DINA project areas who were asked for their opinion and the management approach they implement to address biodiversity conservation in a semi-structured questionnaire with a mix of free text and multiple-choice questions. The mixed method questionnaire offers the advantage of achieving comparable results to a separate nationwide survey (Becker et al. 2019) while providing open text questions that express appreciation of opinions of the respondent (Wlodarek 2019) and increases the motivation to participate (Porst 2014). The qualitative analysis of responses makes individual considerations (Baur and Blasius 2014) accessible to the research process and enables the formation of new hypotheses. The questionnaire underwent a thorough quality control before application and has been evaluated for criteria such as reliability (i.e., precision of results) and validity (i.e., correspondence of response and actual behaviour) (Möhring 2010; Moosbrugger and Kelava 2012).

The questionnaires were sent to farmers in two parts (A and B) at different times (May and August 2020), to keep the time and effort for the participants at a minimum and maximize the response rate ("test economy"; Moosbrugger 2012) with the option of replying by telephone, post or online (Baur and Blasius 2014). Based on a thematic investigation of agricultural electronic newsletters, agricultural magazines, social media channels and personal conversations with various agricultural stakeholders, the questionnaire comprised five main topics. Part A of the questionnaire focused on (i) farm structure and (ii) opinions and recommendations, Part B contains questions focussing on (i) commitment to environmental, nature and species protection, and the extent to which the farmer participates in measures for environmental protection, (ii) everyday work practice, and (iii) regulations and political framework conditions.

Out of the 33 farmers approached, 20 responded to the questionnaire part A and 21 responded to part B. The age group within the range of 50 to 56 years comprised the largest group of respondents with 7 out of 33 farmers, reflecting the demographic change in the agricultural sector. The respondents have a very broad farm structure size range which correlates with the geographical location (northern vs. southern regions) and federal state (eastern vs. western German). However, the amount of land alone does not allow a statement about the economic competitiveness/success. The average total cultivated area of the participants is 641 ha, which is ten times higher than national average (63 ha per farm in 2020, Federal Statistical Office of Germany, Destatis). For the present study, only closed questions with a focus on appreciation of nature protection and willingness to engage in nature conservation measures have been selected for quantitative analysis of the responses.

"Social Labs" local stakeholder dialogue series

From the 21 field sites, three were selected for the Social Lab dialogue series. The site selection was based on geographical size and location, intensity of agriculture within and around protected area (such as number of farms, proximity of settlement) and political and administrative factors (such as jurisdiction of public authorities or previously existing cooperation) to maximize variability among the three sites. The selection followed exploratory talks with authorities to assess the general interest and capacity of key stakeholders to participate in and around the protected area. To allow anonymity of the participants the names of the chosen sites are not disclosed here.

The "Social Labs" were conceptualised as a series of workshops, structured in three phases: (1) setting the local context that identifies topics, obstacles, and potential fields of action; (2) facilitating two-way knowledge integration between local stakeholders in agriculture and conservation as well as scientists of the DINA project; and (3) creating a shared vision for the protected areas. For each of the three selected sites, a key stakeholder meeting (Exploratory Workshop) with local conservation authorities and representatives of local agriculture yielded a first framing of the local topics as well as a comprehensive stakeholder analysis of agricultural and conservation actors. The stakeholder analyses identified persons and institutions that are concerned with the protected area and ranked them according to their level of influence and level of interest. The primary stakeholder group (high level of interest and high level of influence) comprised, in addition to the key stakeholders of local authorities and farmers, state level authorities for agriculture and nature and landscape conservation, landowners or their representatives, local conservation professionals involved in management and planning, as well as shepherds tasked with conservation grazing in the area. Local or regional non-governmental conservation organisations, agricultural associations or people engaged in environmental education and dissemination were placed in the secondary stakeholder group (high level of interest, but low level of influence). At two sites a series of online video workshops was initiated in February 2021 and is still ongoing. At the first site, out of the 21 stakeholders that were identified with a high level of interest and a high level of influence, 16 participated in at least one of the workshop events. At the second site of 16 primary stakeholders, 9 participated. For the third site, key stakeholders rejected an online format and workshops were postponed until spring 2022. Basis for the analyses in this publication are comprehensive minutes and session notes from the workshops of phase 1 and 2 of the "Social Labs", i.e., up to the point of data integration from the monitoring activities of DINA. In phase 2 of the workshops, researchers of the project have presented and discussed site-specific data, including total numbers and bar charts for insect biomass, cumulative numbers of pesticides found within trap ethanol and in soil, as well as preliminary species numbers (total and for relevant major taxa) derived from metabarcoding. Each value shown was detailed along the local Malaise trap gradient as well as cumulated for comparison to the other DINA sites. The minutes have been analysed via qualitative content analysis in MAXQDA (VERBI Software 2021). This addressed the question of which individual motivations or impediments are perceived by stakeholders for taking action for better insect conservation in agriculture within and around the protected area. Statements have been coded into three code categories: (a) the perceived institutional frame; (b) the available knowledge and data; and (c) the interests and held values of stakeholders concerning insect conservation in an agricultural context in and around the protected area. These three main categories derive from established typologies for conflict analysis (Moore 2003; Redpath et al 2015; Sidaway 2011). Segments have been grouped into subcategories of the code tree and summarized to describe archetypical positions in the workshops. As the dialogue processes are still ongoing and the participants have been assured confidentiality, we include summaries of statements rather than verbatim quotations in an online Appendix (Online Resource 3).

Results

Insect species richness, biomass, and pesticides

The richness of insect species correlated highly with the biomass collected in the Malaise traps ($r = 0.72$, Fig. 1). Both variables will function as response variables in later analysis.

Pesticides detected in ethanol are strongly correlated with agricultural production area in the surrounding of the nature reserves (Online Resource 4). A greater coverage by agricultural production area increased the number of pesticides found in the Malaise traps

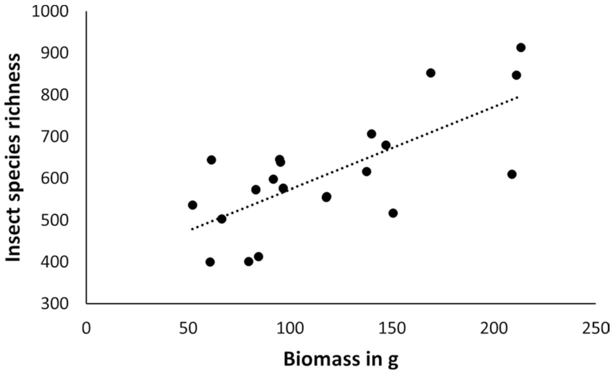

Fig. 1 Spearman correlation analysis between insect species richness and biomass of collected insects

(correlation coefficient: r = 0.65). The two variables were not included together into a GLM due to their strong correlation, but as both variables are suspected to have a strong influence on insects, both variables were further analysed separately.

Both measures of plant richness (field surveys and plant richness determined by metabarcoding of traces in the Malaise trap bottles) correlated positively with each other (r = 0.64, Online Resource 4). Both variables represent plant richness and will be included as explanatory variables, as it is not to be expected that their correlation influences their effect on the response variables. The nature reserves area within 2000 m and the number of detected stationary pesticides had no influence on any other variable analysed (Online Resource 4). The two weather variables maximum temperature and precipitation strongly correlated with each other, and only precipitation will be included for modelling (Online Resource 4).

To analyse the combined impact on insect richness and biomass, four generalized linear models (GLM) were used to detect variables that cause variance in the two response variables (Table 1). The models included the four factors of the previous matrix that did not exceed |r| = 0.5 when correlated with each other, precipitation, and either pesticides in ethanol or agricultural production are within 2000 m as fixed factors (Table 1). With insect species richness as the response variable, plant richness from field surveys had a positive effect on insect species richness, while plant richness from metabarcoding, stationary pesticides, pesticides contained in ethanol, precipitation (Table 1A), and agricultural production area within 2000 m (Table 1B) had negative effects (Fig. 2).

For biomass as a response variable, only precipitation had a negative effect in both models and plant richness from metabarcoding only had a negative effect in model C with pesticides in ethanol as an explanatory variable (Table 1C + D). Nonetheless, the overall effects of the explanatory variables were similar for insect species richness and biomass.

Results of the farmer questionnaire

To contextualise the farmers' situation at the DINA sites, we consider a selection of single-choice questions of the questionnaire concerned with insect conservation management (Table 2). 14 out of 20 respondents specify that the proximity to nature-protected areas is neither advantageous nor disadvantageous for their work (Q1). 18 viewed nature and insect protection as very important or important, one person quoted "neutral" (Q2). However, 13 of the 21 respondents state that they do not participate in

Table 1 Output of generalised linear models (GLM) using A + B) insect species richness and C + D) insect biomass as the response variable

p < 0.05
p < 0.01
p < 0.001

A

Insect species richness	Estimate	Estimate (beta)	Std. Error	z-value	VIF	p-value
Intercept	8.077	-	0.298	27.094	-	< 0.001
Nature reserve area in km² within 2000m	0.019	0.0003	0.011	1.609	1.252	0.108
Plant richness field survey	0.007	0.0010	0.002	3.545	2.244	< 0.001
Plant richness metabarcoding	-0.008	-0.0012	0.002	-4.186	2.520	< 0.001
Stationary pesticides	-0.007	-0.0004	0.004	-1.943	1.226	0.052
Pesticides in ethanol	-0.021	-0.0006	0.006	-3.448	1.042	< 0.001
Precipitation	-0.029	-0.0014	0.004	-6.634	1.382	< 0.001

B

Insect species richness	Estimate	Estimate (beta)	Std. Error	z-value	VIF	p-value
Intercept	7.778	-	0.243	32.060	-	< 0.001
Nature reserve area in km² within 2000m	0.014	0.0002	0.010	1.362	1.411	0.173
Plant richness field survey	0.006	0.0009	0.002	3.917	2.496	< 0.001
Plant richness metabarcoding	-0.006	-0.0009	0.002	-3.733	2.733	< 0.001
Stationary pesticides	-0.009	-0.0005	0.003	-2.578	1.301	0.009
Agricultural production area in km² within 2000 m	-0.041	-0.0008	0.009	-4.685	1.141	< 0.001
Precipitation	-0.026	-0.0013	0.004	-6.780	1.324	< 0.001

C

Insect Biomass	Estimate	Estimate (beta)	Std. Error	z-value	VIF	p-value
Intercept	458.819	-	111.538	4.114	-	0.001
Nature reserve area in km² within 2000m	4.199	0.0723	4.373	0.960	1.531	0.353
Plant richness field survey	0.492	0.0720	0.688	0.715	2.831	0.487
Plant richness metabarcoding	-1.543	-0.2469	0.670	-2.304	2.849	0.037
Stationary pesticides	-1.611	-0.0009	1.437	-1.121	1.400	0.21
Pesticides in ethanol	-0.105	-0.0033	2.234	-0.047	1.064	0.963
Precipitation	-5.779	-0.2844	1.622	-3.563	1.269	0.003

D

Insect Biomass	Estimate	Estimate (beta)	Std. Error	z-value	VIF	p-value
Intercept	7.596	-	0.758	10.018	-	< 0.001
Nature reserve area in km² within 2000m	0.019	0.0003	0.033	0.597	1.651	0.560
Plant richness field survey	0.003	0.0005	0.005	0.594	2.943	0.562
Plant richness metabarcoding	-0.010	-0.0016	0.005	-1.937	3.002	0.073
Stationary pesticides	-0.018	-0.0009	0.011	-1.568	1.402	0.139
Agricultural production area in km² within 2000 m	-0.055	-0.0010	0.029	-1.885	1.212	0.080
Precipitation	-0.045	-0.0022	0.013	-3.579	1.295	0.003

The models A + C include pesticides in ethanol as an explanatory variable, while models B + D include agricultural production area within 2000 m instead. Significant terms at different levels of significance are highlighted by colour (light orange: $p < 0.05$, medium orange: $p < 0.01$, dark orange: $p < 0.001$). Estimate (beta) = standardised estimates, *VIF* variance inflation factor

agri-environmental-climate-measures (AECM) (Q3). Although farmers are aware of the importance of nature protection, their participation in the AECM is small, according to our survey. The reasons for not participating provided by the respondents are predominately related to the premiums paid. Some farmers describe the recurring topic of bureaucratic hurdles as "catastrophic", referring to "constant change in commitments", time intensity and increasing constraints. The study, however, finds that 14 out of 19 the farmers are willing to play an active part in nature conservation, whereas 5 of them were negative in this respect (Q4). The need for cooperation between farmers for nature conservation was also surveyed and illustrates the importance of action, the feasibility of implementation, the acceptance of existing regulations in the light of the overall situation in the period of the questionnaire in 2020, May to October. The self-assessment regarding the regulations on nature conservation is positive. 11 farmers out of 17 within the survey state their willingness to collaborate with other farmers to provide ecological services (Q5).

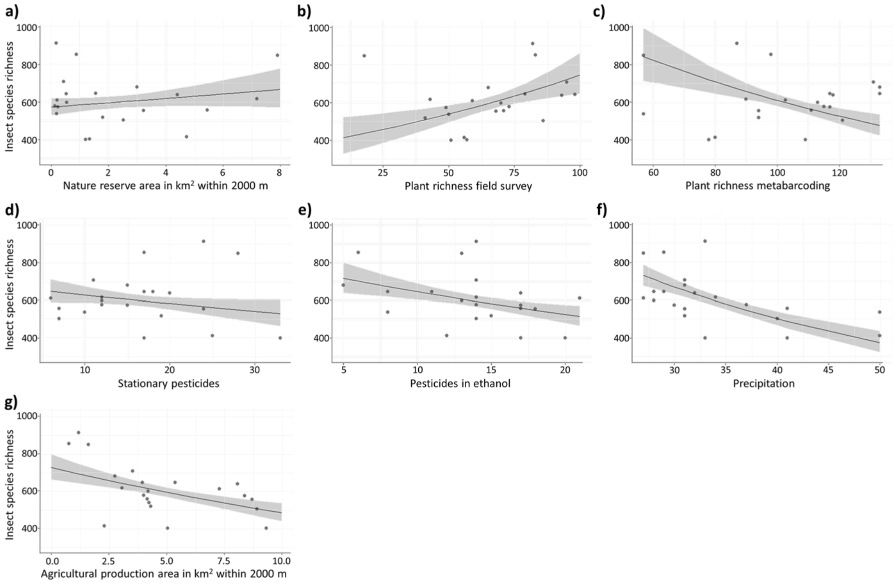

Fig. 2 Marginal effect models for GLM A with insect species richness as the response variable and **a** nature reserve are in km^2 within 2000 m, **b** plant richness from field surveys, **c** plant richness from metabarcoding, **d** stationary pesticides, **e** pesticides in ethanol, **f** precipitation, and **g** agricultural production area in km^2 within 2000 m (from GLM B) as explanatory variables

Another question (Q6) of interest within DINA and in relation to the EU goal of a 50% reduction of pesticides and nutrients until 2030 is the farmers' willingness to contribute. As a sub-question of (Q2), we received responses that 7 out of 9 farmers who find nature conservation very important can imagine reducing their use of pesticides and nutrients input. Also, 6 out 9 respondents who find nature conservation important, can imagine reducing their use. Even farmers having the opinion that nature conservation is at least important (neutral), can imagine reducing their use resulting in a total of 15 out of 17 that are potentially willing to reduce pesticides.

Results from "Social Labs" local stakeholder dialogue series

From an analysis of statements from the first and second phase of the "Social Labs" at two sites, we derived the individual perspectives on the motivations and obstacles for stakeholders in agriculture and conservation on the topic of insect conservation in an agricultural context within and around protected areas. The position subcategories are shown in Fig. 3, for detailed positions see Online Resource 3 (a data document with summaries of individual statements is provided as supplementary data).

Perceived institutional frame

Stakeholders referred to institutional frameworks when talking about obstacles or opportunities for the integration of insect conservation measures into agricultural practices (see Fig. 3, Codes 1.1 to 1.4). Existing incentives and funding schemes for implementing

Table 2 Survey results of a mixed method questionnaire with farmers at sites of the DINA project

Question	Answers	Sample size
Q1: How does the proximity to the nature reserves affect you?		20
Beneficial	3 (15%)	
No matter	14 (70%)	
Unfavourable	3 (15%)	
Q2: How important is nature protection to you?		19
Very important	9 (47%)	
Important	9 (47%)	
Neutral	1 (5%)	
Less important	0 (0%)	
Not important	0 (0%)	
Q3: Have you applied for and are you implementing agri-environmental and climate protection measures (AECM)?		21
Yes	6 (29%)	
Not anymore	2 (10%)	
No	13 (62%)	
Q4: Do you desire opportunities to participate in the development and implementation of nature conservation measures?		19
Yes	14 (74%)	
No	5 (26%)	
Q5: Can you imagine collaborating with farmers to produce ecosystem services?		17
Yes	11 (65%)	
Yes, I already do so	1 (6%)	
No	5 (29%)	
Q6: Can you imagine reducing the use of plant protection products and nutrients?		17
Yes	15 (88%)	
No	2 (12%)	

Shown are frequencies of responses to selected closed questions of the questionnaire

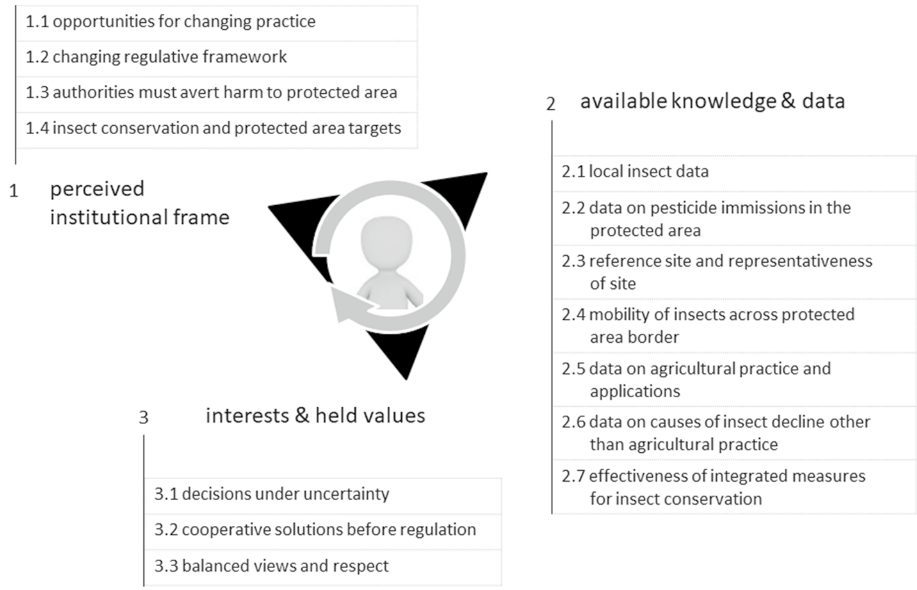

Fig. 3 Categorized arguments within three dimensions of stakeholder motivations for action as derived from stakeholder dialogue series ("Social Labs") at two study sites

short-term insect conservation measures in agricultural practices were perceived impractical, too bureaucratic, and/or financially unrewarding for farmers. For longer-term measures, landowners must be involved and convinced, which was seen as an obstacle for conservation authorities. With reference to the recent changes in national regulations for pesticide applications within protected areas and around water bodies (the so-called 'Insektenschutzgesetz', an amendment to German national laws on nature conservation and pesticide application; Bundesgesetzblatt 2021), agricultural stakeholders expressed concern about the implementation of new legal frameworks on short notice. Economically feasible adjustments in agricultural practice are only considered possible if reliable time frames are set and adhered to.

The conservation authorities and agricultural actors acknowledge the responsibility to avert harm to the protected area and its insect populations but highlight limitations to the mandate of conservation authorities to intervene in agricultural practices within and beyond the protected area borders, e.g., by establishing a buffer zone. Insect conservation had no priority in the targets of protected area management, and certain conflicts with other conservation targets, e.g., trade-offs with habitat types to support migratory birds, were highlighted by conservation stakeholders at one site. Further, the conservation authorities pointed out limited capacities in personnel and funding to strengthen efforts for insect conservation in cooperation with practitioners.

Available knowledge and data

The data basis of the research activities was discussed broadly in terms of data available, and data required for evidence-based decision-making (see Fig. 3, Codes 2.1 to 2.7, and Online Resource 3). Having data from local insect monitoring across multiple years was generally considered of high importance and urgency for decision-making in both, agriculture and conservation. Data on biomass and species lists from the DINA project were met with critical discussion, e.g., regarding the representativeness of the Malaise-trapping method for the locally relevant taxa. Evidence for a local decline of insect species richness and abundances is considered inconclusive by most stakeholders in agriculture and conservation, due to a lack of temporal resolution and existing positive assessments of the local fauna. The list of pesticides detected inside the protected area prompted questions of stakeholders regarding the concentrations and potential effects on insect populations. So far, no information on the effect of pesticide mixtures at low concentrations on insects exist, and therefore the risk cannot be estimated. Agriculture stakeholders acknowledged the lack of scientific knowledge on cross-reactions in mixtures, toxicity, and sub-lethal effects as well as decay rates in soil under field conditions.

With regard to the data on insect biomasses and species lists from the DINA project, stakeholders in agriculture and conservation reasoned that the comparison with local, ecosystem-specific data on insect biodiversity, including earlier monitoring efforts, has higher relevance for them than a nationwide comparison. Standing for themselves, the new local data are not considered to provide insights into potential hazards to the local protected area by most stakeholders. A major point in the discussion of knowledge gaps after having discussed the local data was the presumed mobility of insects in and out of the protected area. Stakeholders questioned whether pesticides found in insect samples were carried in by highly abundant generalists or even agricultural pest species from outside the protected area, while valuable rare species may be less mobile and might have lower pesticide loads.

Intersecting local data on crop rotations and pesticide applications with detected residues on insects was seen as a great value for both agricultural and conservation actors to resolve the mobility of insects across the protected area boundary and to relate detected residues with applications in the field. Agricultural stakeholders have a detailed knowledge about the types of substances and their application and willingly shared these data with researchers in the dialogue series. Other causes of insect decline beyond pesticides were discussed as a potential management subject. Agricultural stakeholders pointed to climate change as well as other local environmental changes, like light pollution or recreational use, as potential causes of insect decline to be considered. Conservation stakeholders identified a lack of knowledge regarding detrimental effects for insect biodiversity from other conservation measures, e.g. those supporting bird habitats. Stakeholders in agriculture and conservation identified knowledge gaps on the effectiveness of integrated measures for insect conservation in the agricultural practices, for instance, how buffer strips and edge habitats affect the mobility of insects and their exposure to pesticides.

Interests and held values

Stakeholders expressed their interests and held values when it comes to deriving actions or directions from the data basis and from the dialogue process (see Fig. 3, Codes 3.1 to 3.3).

Some agricultural stakeholders demanded that interventions in agricultural practices should be grounded on a clear data basis and measures should be conclusive, while others acknowledged the difficulties to obtain conclusive data. The precautionary principle was acknowledged by conservation stakeholders as well as agricultural stakeholders, but only some conservation actors urged for immediate action.

Agricultural stakeholders defended the need for pesticide applications in conventional production practices as essential for food security. They demanded cooperative solutions to develop an economically feasible integration of conservation into agricultural practice, rather than creating new dependencies on subsidy schemes for farmers. Regulative interventions were seen as an option by conservation stakeholders if cooperation fails to solve imminent problems. Agricultural stakeholders were particularly concerned about misunderstandings in public perception regarding the compliance with pesticide application regulations and the key role of agriculture in conservation practice. Agricultural and conservation actors framed the local public as a stakeholder of the dialogue process that should be informed and committed to the dialogue outcomes. As a key quality of future local dialogues, stakeholders named a balanced representation of agricultural and conservation interests, as well as mutual trust and openness.

Discussion

Within the framework of DINA, we were able to demonstrate that Malaise trap samples can be used not only for studies on insect species richness and biomass, but also to examine traces carried into the traps by insects to analyse plant species richness through metabarcoding (Swenson et al. 2022) and pesticide residues in ethanol (Brühl et al. 2021). Results from our Malaise trap transect studies for the analysed sampling intervals show a clear positive correlation between insect species richness and insect biomass, confirming previous observations in Europe among certain insect taxa such as hoverflies (Diptera: Syrphidae; Hallmann et al. 2021a). Showing a clear correlation between decline of insect biomass and reduction of insect diversity in natural habitats, our findings support the documented loss of insect biomass (Hallmann et al. 2017) can be employed as an indicator of overall insect species decline. It is therefore important to protect the remaining insect diversity and reject the assumption that insect biomass and diversity are possibly decoupled (Redlich et al. 2021; but see Hallmann et al. 2021b). Restricting analyses on insects determined to species level may risk that species richness is underestimated, while guaranteeing that species are not counted more than once.

A positive correlation was observed for the plant diversity resulting from field surveys and determined by metabarcoding from the ethanol of the Malaise traps. Interestingly, both variables have opposing effects on insect richness and biomass in our GLM models. These differences might be related to the scale-dependence, as both estimates of plant richness measure on different scales: while vegetation surveys in the immediate vicinity of a Malaise trap indicate resources directly available to insects in a given habitat, the richness derived from pollen metabarcoding indicates resources in the wider landscape. Local plant richness is exactly known from visual taxonomic identification and linked to a specific area. In contrast, plant richness from metabarcoding is based on an unknown sampling area and indicates plants with which insects had directly or indirectly interacted. These plant species may not be directly or causally linked to insect species numbers. Certainly, these differences in the slopes of the regression lines deserve further study. Decreasing numbers of insect species could have an impact on the plant species detected, as the interactions between insects and plants can be very specific and may no longer be detectable here. However, these interactions are likely to be a small percentage of all plant–insect interactions, and the common generalists will standardise the plant components in the traps and over an entire year of sampling the metabarcoding results should largely reflect what plants are present in the environment, regardless of some unrecorded specialized interactions.

A similar pattern was seen for the pesticide residues detected in ethanol, where insect richness declined with higher numbers of pesticides. The impact of land use and farming activities on flying insects in nature reserves was found in a previous study of DINA data by Brühl et al. (2021). In the present study, pesticides in ethanol and agricultural production area within 2000 m were strongly connected and had a negative impact on insect species richness. When looking at a more detailed analysis (Online Resource 5), the number of herbicides in the ethanol correlated strongly with the amount of agricultural production area within 2000 m ($r = 0.65$) while fungicides ($r = 0.28$) and insecticides ($r = 0.42$) were less associated with agricultural production area. However, due to their toxicity to insects, insecticides may have been underrepresented because exposed insects are not likely to fly into the Malaise traps. The probability that individual herbicides and fungicides are not detected in the ethanol samples due to reduced insect numbers is low, as these substances are in many cases not causing lethal effects in insects even at high concentrations. Previous studies showed that pesticide mixtures differ between spring and autumn and that fungicides are more frequent in autumn, so these results could change over the year (Brühl et al. 2021; Zaller et al. 2022). Stationary pesticides had a strongly negative influence on species richness. Although at some locations, the prevalent wind blew directly from agricultural production areas into the nature reserve, while at other locations the prevalent wind blew in the opposite direction. The orientation of the transects was determined by several factors and local conditions, which is why pesticide drift might be underestimated at half of the sites (Lehmann et al. 2021). Further studies are needed to assess the risk of stationary pesticides on insect and plant diversity.

Our questionnaire with the farmers of the DINA project sites also shows that 18 of 19 participating farmers considered nature protection important for their work. They also expressed willingness to collaborate and change their practices in favour of insect conservation measures (15 of 17). As reasons for non-participation in agri-environmental and climate protection measures (AECM) (13 of 21) they name inter alia the perceived unclear and unstable framework conditions. According to the questionnaire, farmers perceive reliable planning, mutual acceptance and trust of society, policymakers and the regulatory system as key prerequisites for the implementation of measures for insect conservation in agricultural practices. The survey addressed persons already participating in the DINA project by providing access to their land for Malaise trapping and soil sampling and were generally open to the topic. Ten out of 33 farmers addressed did not respond to the survey for unknown reasons. Persons who objected to the sampling activities were not included. The survey therefore does not allow conclusions about farmers in Germany in general. In the process of this study, it has become increasingly clear that more dialogue on all levels (local, regional, national) is demanded by the participating farmers. To enable societal actors to contribute to insect conservation within and around protected areas, an agglomeration of institutional frameworks, societal demands, as well as individual motivations of actors must be considered and addressed.

From the two in-depth Social Lab dialogue processes, it was derived that motivations of stakeholders are driven by their subjective values and held interests, perceived institutional frameworks as well as their practical knowledge and data availability (see Fig. 3 and information in Online Resource 3). This is in line with observations in participatory research processes in environmental research (Wehn and Almomani 2019) and transdisciplinary research on pesticide reduction in France (Young et al. 2022).

Our analysis of the ongoing dialogue processes indicates that the local stakeholders demanded and appreciated the discussion of new research data in the light of the local situation. This corresponds well to previously described demands for measures that fit the local context (Bartowski and Bartke 2018). However, the data basis was not deemed sufficient by some stakeholders to assume an imminent threat to the protected area and to justify action beyond the current conservation scope and geographic boundaries of the protected area resulting in differing risk perceptions (Gore et al. 2009) that need further research. The dialogues were scheduled in parallel to the data collection in the DINA project, which limited the integration of ongoing work for the dialogue series. Nonetheless, in the setting of the dialogue series, strengthening conservation efforts to reduce pesticide immissions into the protected area based on the precautionary principle seemed to be acceptable also for agricultural actors. However, farmers perceived a lack of acknowledgement of the already existing contributions of agricultural practices for conservation of the protected area (e.g. landscape conservation, sheep pasture, own initiatives for pesticide reductions) by funding schemes and by the local community, which seems to be a structural issue (Young et al. 2022; Heinze et al. 2021). In the context of dialogues, farmers were willing to provide practical knowledge (e.g., on crop rotation, pesticide applications and historic land use practices) to evaluate data on pesticide immissions. We therefore see these procedural results in line with knowledge about the positive effect of facilitation on farmers' intention to cooperate in concrete environmental measures (van Dijk et al. 2015).

The two case studies included in this analysis cannot be seen as representative for all regions of Germany, but they provide in-depth insights in arguments and positions of local stakeholders in agriculture and conservation, and their institutional constraints as well as value-driven motivations and interests. At the time of writing, the dialogue series has not been completed yet, and it remains open if the dialogue succeeds in resolving mutually agreed targets and measures for improving local insect conservation within and around the protected area. It became obvious, however, that mediating conflicts of interests, values, and knowledge requires a broad multi-perspective approach. The emphasis of conflict mediation in a transdisciplinary setting with local practitioners and biodiversity researchers can be a key to build up trust, develop practical solutions and achieve mutual agreement on conservation targets.

Conclusions

Our study provides evidence for the strong agricultural influence on insects and a correlation between insect species richness and insect biomass. Plant richness from field surveys positively affected insect species richness. Stationary pesticides, pesticides in ethanol, the amount of agricultural production area within 2000 m, and precipitation had negative effects on insects. Efficient insect conservation in nature reserves embedded within agricultural landscapes should therefore integrate management of agricultural activities in a much wider landscape context than is currently the case.

However, there is no one-fits-all solution to the multi-level, cross-sectoral and interdisciplinary challenges of insect conservation management in agricultural contexts in and around protected areas. By involving stakeholders from agriculture and conservation practices in local dialogue processes, the economic, ecological, and societal objectives of sustainable land use practices can be integrated into decision making for a better management of insect biodiversity. To achieve the integration of biodiversity data into these dialogues, we conclude that data assessed at the broader national scale need to be contextualised spatially, e.g., through knowledge integration of practitioners, and temporally, by repeated measurements or reference data, to leverage action at the local level. The dialogues at the two DINA sites showed that to reduce pesticide immissions into the protected area from the surrounding agricultural land will require institutional capacities to be improved by re-defining responsibilities with respect to conservation measures within and outside the protected area boundaries and by providing a diverse toolset and flexible funding schemes at the state level.

By combining basic research with applied transdisciplinary research, we developed a pilot scheme where data-driven dialogues as a science-policy-society interface at the local level, can facilitate cooperation for the development of tailored solutions for the management of insect biodiversity in protected areas.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s10531-022-02519-1.

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Author contributions The study was conceptualized by GUCL, CAB, BG, AL, GM, LS, MS, and WT. SK, FDS, AT, GUCL and RM wrote the original draft. Development and design of methodology as well as data collection was conducted by NB, LE, TF, TH, GUCL, RM, FDS, MS, SJS, AT, and VMAZ. SK, FDS, CS and AT analysed the data. All co-authors reviewed and edited the document and approved the final manuscript.

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Data availability The datasets generated and/or analysed as well as R code can be made available for research purposes upon request to the corresponding authors.

Declarations

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval Permission to collect insects and soil and vegetation samples in the 21 nature conservation areas was granted by the relevant authorities of the different federal states of Germany. The study design and concept for the stakeholder dialogue series were subject to a review by the ethics committee of ISOE – Institute for Social-Ecological Research. Oral consent was obtained from all participants on the recording of the workshops and written consent was obtained for the anonymous analysis of statements for research purposes.

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