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# Species Sensitivity Distribution (SSD) profiles towards λ-cyhalothrin for key ecosystem service provider (ESP) species across five European countries representing different pedoclimatic zones☆

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# HIGHLIGHTS GRAPHICAL ABSTRACT

Species Sensitivity Distribution (SSD) profiles towards lambda-<br>cyhalothrin for key ecosystem service provider (ESP) species across<br>five European countries representing different pedoclimatic zones



- Single-species assays do not ensure that entire communities are protected.
- Species Sensitivity Distributions allow estimating effects at the community level.
- SSDs for λ-cyhalothrin estimated for ecosystem service providers in 5 countries.
- On average, λ-cyhalothrin applied at recommended doses affects 98.5 % species.

#### ARTICLE INFO

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Conclusion

Extremely high risk to the ESP

**Options for assessing** ecological risks

#### ABSTRACT

Although our understanding of the dramatic worldwide loss of biodiversity in recent decades is far from adequate, one of the main factors in areas dominated by agriculture is undoubtedly the widespread use of synthetic pesticides. Unfortunately, the ecological risk assessment (EcoRA) for pesticides is based on a few single-

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*Keywords:* Ecological risk assessment Nontarget arthropods Pest enemies Pesticides Biodiversity decline

species bioassays which do not allow for the evaluation of risks to whole communities. Here we present the results of an experimental assessment of the risk to the ecosystem service provider (ESP) communities – pest control agents – from exposure to the commonly used pyrethroid insecticide, λ-cyhalothrin. The study was performed in five European countries (Germany, Poland, Portugal, Spain, United Kingdom) representing different pedoclimatic zones. Representatives of the most common species of the ESP communities in each country were exposed in a standardized insecticide-coated glass vials bioassay to five doses of λ-cyhalothrin: 0.8 %, 4 %, 20 %, 100 %, and 200 % of the recommended field dose (RFD) plus an untreated control. Based on the calculated LD50s, species sensitivity distributions (SSDs) were estimated for each country and on combined data. In all five countries, the estimated hazardous concentration for 5 % of the species (HD5) was between 0.23 % and 1.67 % RFD, with HD5 = 0.44 % RFD based on combined data. At the RFD = 7.5 g a.i./ha (active ingredient per hectare), the predicted affected fraction of the ESP communities was between 96.4 % and 99.9 % of the species (98.5 % for combined data). The results indicate an extremely high risk to ESP communities across Europe associated with the use of λ-cyhalothrin at the recommended doses when these species are exposed to insecticide treatment. We recommend that EcoRA should include multi-species approaches, such as SSD, to better protect entire ESP communities from the negative impacts of pesticides.

### **1. Introduction**

The dramatic decline in biodiversity over the last few decades, especially among insects, has been demonstrated beyond doubt ([Hallmann](#page-12-0) et al., 2017; Sánchez-Bayo and Wyckhuys, 2019; [Seibold](#page-13-0) et al., [2019](#page-13-0)). Most recent estimates show losses in the diversity and biomass of insects at a rate of 2.2–2.8 % per year [\(Hallmann](#page-12-0) et al., 2017; Lister and [Garcia,](#page-12-0) 2018; [Ziesche](#page-13-0) et al., 2023) with estimates for particular groups, such as moths, being even higher (e.g. Bell et al., [2020](#page-12-0)). Among the most endangered species are those inhabiting open areas, which in much of the world are dominated by land for crop cultivation. The nature of agricultural areas causes a specific conflict of interests: on the one hand, pest management requires the use of plant protection products (pesticides), but on the other hand, non-target arthropods (NTAs) must be protected from harm by these products, especially insecticides. Many NTAs provide important ecosystem services, such as pest control, pollination and nutrient cycling. As ecosystem service providers (ESPs) are essential for human well-being (Mori et al., [2018](#page-13-0)), the threat from insecticide use is becoming a major concern [\(Chagnon](#page-12-0) et al., [2015](#page-12-0)). Unfortunately, due to the mode of action of most synthetic insecticides and the similarity of basic biochemical processes among insects, they are not currently specific enough to affect only the target pest species. As a result, the widespread use of insecticides against phytophagous insects leads to harmful effects on beneficial organisms inhabiting agricultural areas and their surroundings. Therefore, insecticides have become one of the main drivers of biodiversity loss ([Goulson,](#page-12-0) 2013; [Chagnon](#page-12-0) et al., 2015; [Kenko](#page-12-0) et al., 2023). Determining the magnitude, probability, and relevance of potential negative effects of pesticides on NTAs is, thus, crucial for protecting biodiversity and maintaining important ecosystem services.

According to the U.S. Environmental Protection Agency (EPA), the applicant registering a pesticide must prove "*that using the pesticide according to specifications will not generally cause unreasonable adverse effects on the environment*" [\(www.epa.gov\)](http://www.epa.gov). In turn, the *EC [Regulation](#page-12-0) No 1107/ 2009 of the European [Parliament](#page-12-0) and of the Council of 21 October 2009 [Concerning](#page-12-0) the Placing of Plant Protection Products on the Market and Repealing Council Directives [79/117/EEC](#page-12-0) and 91/414/EEC*, n.d. states that "*A plant protection product [...] shall have no unacceptable effects on the environment, having particular regard to the following considerations: […] its impact on non-target species, including on the ongoing behaviour of those species; its impact on biodiversity and the ecosystem*". However, according to paragraph 3.8.3 of this regulation, "*an active substance, safener or synergist shall be approved only if […] the use under the proposed conditions of use of plant protection products […] will result in a negligible exposure of honeybees or has no unacceptable acute or chronic effects on colony survival and development*". Thus, the approval procedure in the EU is entirely based on just one species – the western honey bee (*Apis mellifera* L. 1758). Although honey bees have great environmental and economic value, at the same time they are a very specific species, bred

mainly by humans, living in man-made hives, eusocial, etc. They also represent just one group of NTAs and ESPs, namely the pollinators. However, in agricultural landscapes, other NTA guilds are no less important for ecosystem functioning and our economy, with pest control agents, such as predatory arthropods and parasitoids, among others. It is therefore essential to consider a broader spectrum of non-target arthropods and ESPs when assessing the potential environmental impact of insecticides.

Different species exhibit different sensitivities to the same chemical, and the variation among these species can be described by a statistical distribution. Such species sensitivity distribution (SSD) can be used to estimate the concentration or dose that is hazardous to a certain proportion (x%) of species in a community (HCx, HDx) and the potentially affected fraction (PAF) of species at a given concentration or dose. In ecological risk assessment (EcoRA), the commonly used benchmark is the concentration (or dose) which puts at risk no more than 5 % of species in a community. Thus, HC5 (HD5) is the concentration (dose) at which no more than 5 % of species in a community represented by the SSD show negative effects, i.e., more precisely, reach the toxicity endpoint used to establish the SSD. Therefore, the analysis of SSDs is an important technique in EcoRA, primarily used to define the Predicted No Effect Concentrations (PNEC), understood as the concentrations that do not cause significant adverse effects in non-target species communities. It is important to realise, however, that the reliability of an SSD relies on the quality of the data used to estimate the distribution while the exact meaning of the derived values (HC5/HD5, PAF) depends on the endpoint used. Hence, the uncertainty analysis and the selection of an appropriate assessment factor (AF) are crucial in estimating PNEC. When PNEC is estimated using SSD, the European Chemicals Agency (ECHA – European [Chemicals](#page-12-0) Agency, 2008) recommends dividing HC5 (HD5) by AF of 1 to 5. Assessment factors lower than 5 should be only used if fully justified by the quality of the data. According to [ECHA](#page-12-0) – European [Chemicals](#page-12-0) Agency (2008), the following criteria should be considered: quality of the endpoints used – in particular whether the dataset covers chronic studies and different life stages, the diversity and representativeness of tested taxonomic groups, statistical uncertainty around the HC5/HD5 estimate, etc.; see also [Checkai](#page-12-0) et al. (2014).

Although the SSD technique dates back to the 1980s, there have been surprisingly few studies on the effects of insecticides on terrestrial communities since then. Most SSD studies have focused on aquatic environments, with over half of them addressing problems with pesticides: the Web of Science search "(SSD OR species sensitivity distribution) AND (pesticid\* or insecticid\*)" reported 483 articles while only 23 were found with the query "(SSD OR species sensitivity distribution) AND (pesticid\* OR insecticid\*) AND (terrestrial OR soil OR epigeic) NOT (aquatic OR water\*)" (as of April 2024). The search for articles specifically on the effects of λ-cyhalothrin: "(SSD OR species sensitivity distribution) AND lambda-cyhalothrin" revealed only 19 papers, almost exclusively on aquatic environments. These results point to a large gap

<span id="page-2-0"></span>LD50s for ecosystem service providers collected in oilseed rape conventional crop in Germany and exposed for 24 h to  $\lambda$ -cyhalothrin; the LD<sub>50</sub>s are expressed both in g a.i./ha (with 95 % confidence intervals, CI) and as a percent of the recommended field dose (% RFD) assumed at 7.5 g a.i./ha.



in knowledge on the effects of pesticides on whole terrestrial communities – a rather surprising conclusion considering the widely discussed and well-proven decline in abundance and diversity of non-target terrestrial arthropods in recent decades.

While SSD is a convenient method of pesticide risk assessment for whole communities, there are several reasons to expect that distinct communities are characterized by different SSDs. For example, NTA/ ESP communities in different pedoclimatic zones differ in species composition, and individual species differ in sensitivity to toxicants. On smaller geographical scales, species assemblages differ among crops and ecosystems. Finally, even the same species may differ in sensitivity across agricultural landscapes due to different histories of exposure to pesticides – e.g., populations exposed for a long term to pesticides may exhibit elevated resistance. As a result, PNECs estimated using this technique would differ among regions and communities, meaning that EcoRA should be area/community specific. Unfortunately, there are no published studies to compare SSDs for the same terrestrial ESP guilds across different geographic regions.

In this study, we aimed to fill the information gap by collecting affected/mortality data for ESPs, namely predatory and parasitic arthropods, exposed to the widely-used pyrethroid insecticide, λ-cyhalothrin, in a highly standardized way, in five countries representing the three major pedoclimatic zones of Europe: Continental (Germany and Poland), Mediterranean (Portugal and Spain), and Atlantic (United Kingdom). In each country, the ESPs were collected from crops that are commonly grown in the area, representing the typical ESP communities for that region. SSDs for terrestrial communities across larger regions may enhance the ability to develop region-specific guidelines and management strategies that would consider the unique characteristics and vulnerabilities of local ecosystems.

Λ-cyhalothrin is a fast-acting contact pyrethroid, commonly used in the EU and globally, to control such agricultural pests [\(Depalo](#page-12-0) et al., [2022\)](#page-12-0) as aphids, beetles, moths, thrips, and, for human health, mosquitoes, flies and ticks [\(Leprince](#page-12-0) et al., 1992). According to the European Food Safety Authority (EFSA – [European](#page-12-0) Food Safety Au[thority,](#page-12-0) 2014), the first-tier risk assessment for  $\lambda$ -cyhalothrin indicated a high risk to NTAs in the case of all field uses. A higher-tier assessment, such as that based on SSD, is therefore highly desirable.

### **Table 2**

LD50s for ecosystem service providers collected in conventional oilseed rape and wheat crops in Poland and exposed for 24 h to  $\lambda$ -cyhalothrin; the LD<sub>50</sub>s are expressed both in g a.i./ha (with 95 % confidence intervals, CI) and as a percent of the recommended field dose (% RFD) assumed at 7.5 g a.i./ha.

Species	Taxonomic	Crop	$LD_{50}$ [g/ha]	$LD_{50}$
	group		(95 % CI)	as $%$ RFD
Abax parallelepipedus	Coleoptera:	Oilseed	0.078	1.0
(Piller $\&$	Carabidae	rape	$(0.039 - 0.157)$	
Mitterpacher, 1783)				
Amara aenea (Degeer,	Coleoptera:	Oilseed	0.061	0.8
1774)	Carabidae	rape	$(0.025 - 0.149)$	
Amara similata	Coleoptera:	Oilseed	0.111	1.5
(Gyllenhal, 1810) Anchomenus dorsalis	Carabidae Coleoptera:	rape Oilseed	$(0.052 - 0.239)$ 0.057	0.8
(Pont, 1763)	Carabidae	rape	$(0.017 - 0.192)$	
Anisodactylus sp.	Coleoptera:	Oilseed	0.07	0.9
	Carabidae	rape	$(0.042 - 0.116)$	
Asaphidion flavipes	Coleoptera:	Oilseed	0.073	1.0
(L., 1760)	Carabidae	rape Oilseed	$(0.009 - 0.559)$ 0.238	3.2
Bembidion lampros (Herbst, 1784)	Coleoptera: Carabidae	rape	$(0.105 - 0.539)$	
Carabus granulatus L.,	Coleoptera:	Oilseed	0.067	0.9
1758	Carabidae	rape	$(0.024 - 0.186)$	
Harpalus affinis	Coleoptera:	Oilseed	0.052	0.7
(Schrank, 1781)	Carabidae	rape	$(0.01 - 0.281)$	
Harpalus rufipes (Schrank, 1781)	Coleoptera: Carabidae	Oilseed rape	4.31 $(2 - 9.28)$	57.5
Harpalus tardus	Coleoptera:	Oilseed	0.085	1.1
(Panzer, 1796)	Carabidae	rape	$(0.039 - 0.184)$	
Nebria brevicollis	Coleoptera:	Oilseed	0.932	12.4
(Fabr., 1792)	Carabidae	rape	$(0.447 - 1.95)$	
Notiophilus palustris	Coleoptera:	Oilseed	0.671	8.9
(Duftschmid, 1812) Ocypus olens (O.	Carabidae Coleoptera:	rape Oilseed	$(0.304 - 1.478)$ 0.077	1.0
Müller, 1764)	Staphylinidae	rape	$(0.048 - 0.126)$	
Platynus assimilis	Coleoptera:	Oilseed	0.933	12.4
(Paykull, 1790)	Carabidae	rape	$(0.369 - 2.36)$	
Poecilus cupreus (L.,	Coleoptera:	Oilseed	0.256	3.4
1758) Pterostichus	Carabidae Coleoptera:	rape Oilseed	$(0.132 - 0.495)$ 0.59	7.9
melanarius (Illiger,	Carabidae	rape	$(0.286 - 1.22)$	
1798)				
Zabrus tenebrioides	Coleoptera:	Oilseed	0.197	2.6
(Goeze, 1777)	Staphylinidae	rape	$(0.11 - 0.355)$	
Agonum muelleri (Herbst, 1784)	Coleoptera: Carabidae	Wheat	0.342 $(0.179 - 0.654)$	4.6
Amara aenea (Degeer,	Coleoptera:	Wheat	0.135	1.8
1774)	Carabidae		$(0.057 - 0.321)$	
Amara similata	Coleoptera:	Wheat	0.077	1.0
(Gyllenhal, 1810)	Carabidae		$(0.048 - 0.126)$	
Anchomenus dorsalis	Coleoptera:	Wheat	0.208	2.8
Anisodactylus signatus	Carabidae Coleoptera:	Wheat	$(0.098 - 0.44)$ 0.083	1.1
(Panzer, 1796)	Carabidae		$(0.024 - 0.295)$	
Bembidion lampros	Coleoptera:	Wheat	0.376	5.0
	Carabidae		$(0.195 - 0.723)$	
Calathus fuscipes	Coleoptera:	Wheat	1.174	15.7
(Goeze, 1777) Carabus granulatus L.,	Carabidae Coleoptera:	Wheat	$(0.645 - 2.14)$ 0.212	2.8
1758	Carabidae		$(0.127 - 0.353)$	
Coccinella	Coleoptera:	Wheat	0.012	0.2
septempunctata	Coccinellidae		$(0 - 0.296)$	
Harmonia axyridis	Coleoptera:	Wheat	0.033	0.4
(Pallas, 1773) Harpalus affinis	Coccinellidae Coleoptera:	Wheat	$(0.019 - 0.056)$ 0.134	1.8
(Schrank, 1781)	Carabidae		$(0.084 - 0.215)$	
Harpalus rufipes	Coleoptera:	Wheat	0.382	5.1
(Schrank, 1781)	Carabidae		$(0.24 - 0.606)$	
Platynus assimilis	Coleoptera:	Wheat	0.779	10.4
(Paykull, 1790)	Carabidae		$(0.437 - 1.39)$	
Poecilus cupreus (L., 1758)	Coleoptera: Carabidae	Wheat	0.814 $(0.392 - 1.69)$	10.9
Pterostichus	Coleoptera:	Wheat	0.396	5.3
melanarius (Illiger,	Carabidae		$(0.209 - 0.748)$	
1798)				

LD<sub>50</sub>s for ecosystem service providers collected in different crops in Portugal and exposed for 24 h to  $\lambda$ -cyhalothrin; the LD<sub>50</sub>s are expressed both in g a.i./ha (with 95 % confidence intervals, CI) and as a percent of the recommended field dose (% RFD) assumed at 7.5 g a.i./ha.



# **2. Material and methods**

Adult individuals of NTAs – ecosystem service providers (ESPs) acting as pest-control agents (carnivorous beetles and spiders, earwigs and parasitic wasps (parasitoids) – see  $Tables 1-5$  $Tables 1-5$ ), were collected in five European countries (Germany, Poland, Portugal, Spain and the UK; [Fig.](#page-5-0) 1) from crops commonly grown in these countries (oilseed rape, wheat, barley, corn, lentils, persimmons, olives, apples and Indian figs). The ESPs were collected as live samples, using pitfall traps, sweep nets, suction samplers or directly by hand, and were transferred to the laboratory where they were maintained in climatic chambers (16:8 h light: dark regime; 20  $\pm$  2 °C; relative humidity 70  $\pm$  5 %; light intensity 600–700 lx) before use. The collection dates differed between the countries and years due to climatic differences and weather conditions which determine ESPs activity. In Germany, the ESPs were collected in the period 28.04.2022–16.06.2022, in Poland 27.04.2020–10.06.2020, 12.05.2021–30.07.2021, and 09.05.2022–08.07.2022, in Portugal 13.04.2020–25.09.2020 and 27.04.2021–30.09.2021, in Spain 14.04.2022–27.06.2022, and in the UK 27.06.2019–20.08.2019, 27.04.2020–03.11.2020, 12.05.2021–04.11.2021, and 25.03.2022–24. 11.2022. Altogether, we tested 54 species represented by 91 populations, including 10 species (10 populations) from Germany, 23 species (33 populations) from Poland, 20 species (22 populations) from

Portugal, 10 species (11 populations) from Spain and 15 species (15 populations) from the UK [\(Table](#page-6-0) 6).

Specimens that might be misidentified during the collection were identified live under a binocular microscope. After confirming the proper identification of all individuals, they were used in laboratory bioassays, usually on the day following collection. We used the IRACrecognised insecticide-coated glass vials test (IRAC – [Insecticide](#page-12-0) Resis-tance Action [Committee,](#page-12-0) 2009) using vials (ca. 60 mm h  $\times$  24 mm Ø) coated with λ-cyhalothrin applied as an active ingredient dissolved in acetone as a carrier. Individuals of each tested species were exposed to five insecticide doses, relative to the Recommended Field Dose (RFD), namely 0.8 % RFD, 4 % RFD, 20 % RFD, 100 % RFD, and 200 % RFD, plus acetone control, to cover the whole range of mortality to estimate LD50 values, which were later used to establish SSD curves. We used RFD of 7.5 g of active ingredient per hectare (g a.i./ha) as this value is most relevant to the crops covered by our study. However, it should be noted that doses of 20–25 g a.i./ha are not uncommon (EFSA – [European](#page-12-0) Food Safety [Authority,](#page-12-0) 2014) and up to 125 g a.i./ha are used in hop plantations (Scientific [Committee](#page-13-0) on Plants, 2000).

The ESPs were placed into glass vials individually to standardise between species of different sizes and avoid any potential deleterious interaction between individuals. For each species tested, 5 to 10 replicate vials per dose were used, depending on the availability of

LD<sub>50</sub>s for ecosystem service providers collected in different crops in Spain and exposed for 24 h to  $\lambda$ -cyhalothrin; the LD<sub>50</sub>s are expressed both in g a.i./ha (with 95 %) confidence intervals, CI) and as a percent of the recommended field dose (% RFD) assumed at 7.5 g a.i./ha.



#### **Table 5**

LD<sub>50</sub>s for ecosystem service providers collected in conventional oilseed rape crops in the United Kingdom and exposed for 24 h to  $\lambda$ -cyhalothrin; the LD<sub>50</sub>s are expressed both in g a.i./ha (with 95 % confidence intervals, CI) and as a percent of the recommended field dose (% RFD) assumed at 7.5 g a.i./ha.



individuals. The physical condition of the tested individuals was checked 24 h after exposure. Three categories were used:

'mobile'– normally active and fully responsive;

'affected' – uncoordinated, i.e., exhibiting a range of abnormal behaviours: from not being able to cross at least a 3 cm distance in a straight line to not being active and showing signs of paralysis of the

legs;

'dead' – not moving and not responding to mechanical stimuli in any way.

For the estimation of  $LD_{50}$  and then SSD, affected individuals were treated as dead, assuming that in ecological reality, i.e. under field conditions, such individuals would have a negligible chance of survival, as they are unlikely to recover or become prey for predators due to their reduced mobility.

To calculate  $LD_{50}$  values, Abbott's formula was used in those cases where control mortality was recorded. All bioassays where control mortality was above 20 % were excluded from the analyses. The doseresponse curves were fitted to Abbott-corrected mortality using generalized linear models with a binomial error distribution and a probit link function, using the glm function in base R (R Core [Team,](#page-13-0) 2022). The LD50s were estimated by applying function dose.p in the package MASS for R ([Venables](#page-13-0) and Ripley,  $2002$ ) to the fitted glms. The LD<sub>50</sub>s are reported in g a.i./ha and as per cent of the recommended field dose (% RFD). SSDs were estimated with the R package ssdtools [\(Thorley](#page-13-0) and [Schwarz,](#page-13-0) 2018) by averaging log-logistic, log-normal and gamma distributions, as the fit of the three distributions did not differ substantially and different distributions fit marginally better using different data sets, as based on Akaike's Information Criterion corrected for sample size (AICc). The average fit, with a 95 % confidence interval, was estimated using AICc-based relative weights of the distributions. The HD5 values were derived as the 5th percentile of the model-averaged SSDs and are reported both in g a.i./ha and relative to the Recommended Field Dose (% RFD). The PAF of the insect community is reported for the assumed recommended application dose of 7.5 g a.i./ha. Because SSDs were estimated based on 24-h LD<sub>50</sub>s, HD5 represents the dose at which populations of no more than 5 % of species experienced 50 % or greater mortality within 24 h after the exposure. Similarly, PAF represents the percentage of species for which the recommended field dose results in 50 % or greater mortality within 24 h. As the HD5 value, as used in EcoRA, is estimated at the tail of the SSD, for a more robust comparison of the distributions obtained for each country, we also estimated HD50s, i.e. hazardous concentrations for 50 % species, which are placed in the middle of the distribution and have, thus, better statistical properties.

# **3. Results**

The number of species tested in each country ranged from 10 in

<span id="page-5-0"></span>

**Fig. 1.** Sampling sites used in the study. For each country the number of fields from which ecosystem service providers were collected is given in brackets.

Germany to 23 in Poland and the number of populations tested – from 10 in Germany, where all species were collected from oilseed rape crops, to 33 in Poland, where oilseed rape and wheat fields were sampled. Altogether, in the five countries, 91 populations, representing 54 species, were tested ([Table](#page-6-0) 6). The majority of species were carnivorous beetles, dominated by carabids.

The tested species exhibited a broad range of sensitivity towards λ-cyhalothrin. The most sensitive case was the population of ladybird, *Coccinella septempunctata* (L. 1758), collected in a wheat field in Poland, with an  $LD_{50} = 0.012$  g a.i./ha, which is only 0.16 % of the RFD. It was also the most sensitive species in Spain and Germany: a Spanish population from a barley field had an  $LD_{50} = 0.025$  g a.i./ha (0.33 % RFD), and that from an oilseed rape crop 0.044 g a.i./ha (0.59 % RFD); the German population, collected from oilseed rape, had an  $LD_{50} = 0.206$  g a.i./ha (2.7 % RFD). In the UK, the most sensitive species was the parasitoid wasp *Diaeretiella rapae* (M'Intosh 1855) with LD<sub>50</sub> = 0.025 g a. i./ha – the second lowest  $LD_{50}$  found among all tested species in the five countries. In Portugal, the most sensitive species was the carabid *Ditomus tricuspidatus* (Fabr. 1792), with  $LD_{50} = 0.06$  g a.i./ha (0.8 % RFD). Altogether, in 10 cases (8 species), the  $LD_{50}$  was below 1 % RFD, i.e. *<*0.75 g a.i./ha [\(Tables](#page-2-0) 1–5).

On the other end of the sensitivity spectrum, the parasitoid *D. rapae* collected from a conventional oilseed rape crop in Spain had an  $LD_{50} =$ 6.75 g a.i./ha (90 % RFD). Interestingly this was the same species which was found most sensitive in the UK. Besides this species, there were four other cases with  $LD_{50}$ s above 50 % RFD, all belonging to carabids: *Harpalus rufipes* (Degeer 1774) in Poland and Portugal, *Nebria brevicollis* (Fabr. 1792) in the UK, and *Pterostichus ebenus* (Quensel, 1806) in Portugal. From the point of view of a sensitivity range, the two most interesting species are the above-mentioned *D. rapae*, with an LD<sub>50</sub> of

<span id="page-6-0"></span>Predicted Affected Fraction (PAF) of a community of beneficial arthropods (ecosystem service providers ESP) at Recommended Field Dose (RFD, 7.5 g a.i./ha) and Hazardous Doses for 50 % and 5 % ESP species (HD5) collected from different crops in five European countries and exposed for 24 h in glass vials coated with λ‑cyhalothrin pyrethroid insecticide; S – number ofspecies tested; n – number of data points per SSD (can be larger than the number of species if tested in more than one crop).





**Fig. 2.** Species Sensitivity Distribution curve for beneficial arthropods (ecosystem service providers) collected in Germany from oilseed rape fields towards lambdacyhalothrin, based on LD<sub>50</sub>s derived from laboratory-coated glass vial bioassays. The dotted line indicates the estimated HD5 value.



**Fig. 3.** Species Sensitivity Distribution curve for beneficial arthropods (ecosystem service providers) collected in Poland from oilseed rape and wheat fields towards lambda-cyhalothrin, based on LD<sub>50</sub>s derived from laboratory-coated glass vial bioassays. The dotted line indicates the estimated HD5 value.

0.025–6.75 g a.i./ha, and *C. septempunctata* with an LD<sub>50</sub> of 0.012–3.02 g a.i./ha [\(Tables](#page-2-0) 1–5). The *D. rapae* population from Spain was, thus, 270 times more resistant to λ-cyhalothrin than the same species collected in the UK, while for *C. septempunctata* the difference was 252-fold. Overall, the sensitivity range between the most and the least sensitive cases across Europe (*C. septempunctata* in Poland vs. *D. rapae* in Spain) was over 560-fold, demonstrating a large variation across species and populations in response to λ-cyhalothrin.

Despite the differences between the countries in the number of species and populations tested, the HD5 and PAF estimates obtained from SSD analysis [\(Figs.](#page-6-0) 2–6) appeared very consistent. The PAF of the studied ESP communities exposed to λ-cyhalothrin at the RFD 7.5 g a.i./ ha ranged from 96.4 % in Spain to 99.9 % in Germany ([Table](#page-6-0) 6). The dose at which an  $LD_{50}$  is reached for no more than 5 % of species (i.e., HD5, protecting at least 95 % of species from reaching 50 % or higher mortality) appeared extremely low: in Poland, Portugal, Spain and the UK this was below 1 % RFD, in Germany it was 1.67 % RFD. The SSD based on combined data from all five countries [\(Fig.](#page-11-0) 7) gave an HD5  $=$ 0.033 g a.i./ha, which is 0.44 % RFD ([Table](#page-6-0) 6).

The estimated 95 % confidence intervals for HD50s for all countries overlap with the confidence interval for the combined data and in most cases with one another, indicating that even for such a broad geographical coverage and using representatives of different communities, the estimated SSDs are generally similar [\(Table](#page-6-0) 6). The lowest HD50 (0.188 g a.i./ha) was found in Poland, indicating the most sensitive community among those tested, and the highest HD50 was recorded in the UK (0.603 g a.i./ha) but even in these cases the 95 % confidence intervals overlapped.

# **4. Discussion**

Our analysis highlights the potentially serious consequences of the widespread application of one of the most commonly used pyrethroids in Europe. Our findings imply that even when λ-cyhalothrin is applied at the generally recommended field dose of 7.5 g a.i./ha (which is still lower than that recommended for some non-arable crops), the predicted affected fraction of species in communities of natural enemies of crop pests exceeds 96 % in all countries studied. At just 10 % RFD (0.75 g a.i./



**Fig. 4.** Species Sensitivity Distribution curve for beneficial arthropods (ecosystem service providers) collected in Portugal from apple, Indian figs, olive, and persimmon orchards towards lambda-cyhalothrin, based on LD<sub>50</sub>s derived from laboratory-coated glass vial bioassays. The dotted line indicates the estimated HD5 value.

ha), the  $LD_{50}$  was exceeded for 74 % of species (40 of the 54 tested) and in 67 % of all bioassays (61 cases). These species represent important ecosystem service providers which are assumed to have a significant mitigating effect on pest insects [\(Diehl](#page-12-0) et al., 2013; [Symondson](#page-13-0) et al., [2002\)](#page-13-0). Reducing the populations of these natural enemies could potentially trigger a vicious cycle, resulting in increased pest outbreaks, forcing growers to use even more insecticides, which could, in turn, lead to the elimination of even the most resistant ESPs from their crops. Pest outbreaks are currently often exacerbated by the fact that pests are becoming resistant or at least less sensitive to pyrethroids and other insecticides, which is characterized by an increasing number of uncontrollable populations [\(Mota-Sanchez](#page-13-0) and Wise, 2023). Local elimination of beneficial species and biodiversity loss would not be the only effect, as the need to use more insecticides would raise production costs for growers. Our results therefore highlight the urgent need to critically evaluate current insecticide practices, not only from a conservation perspective but also from a cost-benefit point of view.

Preselection due to chronic pesticide exposure in habitats

characterized by scarce non-crop elements can lead to populations with elevated insecticide resistance. This has been confirmed in numerous pest populations (e.g., [Alyokhin](#page-12-0) et al., 2008; [Willis](#page-13-0) et al., 2020) and in several ESP species. For example, [Barbosa](#page-12-0) et al. (2016) and [Rodrigues](#page-13-0) et al. [\(2013\)](#page-13-0) showed elevated resistance to λ-cyhalothrin in the ladybird *Hippodamia convergens* (Guérin-Méneville 1842) populations from cotton fields where this insecticide was regularly used. Another ladybird species, *Propylea japonica* (Thunberg 1781), showed elevated resistance to several insecticides from different chemical groups: abamectin (avermectin family), imidacloprid (neonicotinoid), beta-cypermethrin (pyrethroid), and chlorpyriphos (organophosphate) ([Tang](#page-13-0) et al., [2015\)](#page-13-0). The laboratory selection experiment proved that the resistance of *P. japonica* to imidacloprid can increase over 39-fold in just 20 generations (Tang et al., [2015\)](#page-13-0). This phenomenon is most probably responsible for the large variation in response to λ-cyhalothrin of the three species involved in our study – *C. septempunctata*, *P. cupreus*, and *D. rapae*, which were among the most sensitive species in some countries but the least sensitive in others. Bozsik [\(2006\)](#page-12-0) found that  $\lambda$ -cyhalothrin was



**Fig. 5.** Species Sensitivity Distribution curve for beneficial arthropods (ecosystem service providers) collected in Spain from barley, lentil, and oilseed rape fields towards lambda-cyhalothrin, based on LD<sub>50</sub>s derived from laboratory-coated glass vial bioassays. The dotted line indicates the estimated HD5 value.

moderately harmful to adult *C. septempunctata*. Apparently, we need to be cautious when interpreting such results and performing risk assessments based on just one population of a species because, as our study has shown, the sensitivity of a single species to an insecticide can differ between populations over 200-fold.

# *4.1. Reliability of HD5 and PAF estimates and their meaning*

Using the SSD approach, with HD5 and PAF as endpoints, in environmental risk assessment and management offers clear advantages but has also several drawbacks. HD5 provides a clear quantitative measure, indicating the specific dose or concentration of a hazardous substance that can cause unacceptable harm to a community. This allows for targeted interventions and regulatory guidelines based on a well-defined threshold. Both HD5 and PAF provide straightforward means of communication with agronomists, advisors, growers and other stakeholders, facilitating a better understanding of potential risks. On the other hand, relying solely on HD5 and PAF may oversimplify the complexity of real-world insecticide exposure scenarios. When based on

short-term bioassays, like in our study, these measures do not consider the potential effects of chronic exposures or cumulative damage over time. If bioassays are performed on single toxicant exposures, as is usually the case, they cannot capture possible interactions among different agrochemicals. This may potentially be an important problem as it is known that some plant protection products may have nonadditive interactive effects, including synergism ([Siviter](#page-13-0) et al., 2021). However, there is nothing to prevent the SSD technique from being used to estimate the risk from chronic exposures or exposures to mixtures of chemicals. The only impediment is the need for more complex experimental setups, requiring the use of larger numbers of experimental organisms and the allocation of longer experimental time. The advantage of using SSDs for a better understanding of risk distribution within a community, considering the variability in susceptibility among species, is however invaluable. The results of our study, covering several crops in five countries from different pedoclimatic zones, revealed surprising consistency of the estimated endpoints: PAF at the dose of 7.5 g a.i./ha ranged between 96.4 % and 99.9 %, and HD5 was 0.23–1.67 % RFD. Interestingly, the highest PAF (i.e., the largest effect at RFD) and the



**Fig. 6.** Species Sensitivity Distribution curve for beneficial arthropods (ecosystem service providers) collected in the United Kingdom from oilseed rape and wheat fields towards lambda-cyhalothrin, based on LD<sub>50</sub>s derived from laboratory-coated glass vial bioassays. The dotted line indicates the estimated HD5 value.

highest HD5 (the highest 'safe' dose) were found for the German ESP community, indicating the steepest relationship between LD50 and the cumulative effect on the community. Such consistency of the endpoints allows for estimating Europe-wide values for PAF (98.5 %) and HD5 (0.44 % RFD or 0.033 g a.i./ha).

The large differences in sensitivity between populations of several species demonstrated in our study highlight the complexity of species' responses to pesticides and point out the pitfalls of assessing ecological risk based on bioassays on specific populations of single species. In contrast to single-species-based risk assessment, the SSD approach offers more robust results, even if individual species differ in sensitivity by orders of magnitude (cf. [Maltby](#page-12-0) et al., 2005). Our study shows that despite different community compositions and considerable differences in sensitivity to λ-cyhalothrin in several species, the endpoints estimated using SSDs, namely HD5 and PAF, were remarkably similar across the different crops and pedoclimatic zones. This allowed us to calculate 'Europe-wide endpoints' based on combined data from all five countries. We therefore conclude that ecological risk assessment for pesticides should be based on the SSD approach rather than on single-species tests.

## *4.2. Possible recovery by migration and evolution of resistance*

It has been proposed that some of the adverse effects of insecticides could be mitigated through the "recovery by migration of beneficials" from non-crop habitats. This idea relies on the movement of beneficial insects from natural habitats into agricultural fields to restore local populations (Marshall and [Moonen,](#page-12-0) 2002). However, the feasibility of this strategy is closely linked to the landscape structure. Factors such as the distribution, size and shape of habitat patches are key to the success of this restoration strategy [\(Taylor](#page-13-0) et al., 1983). Furthermore, species' dietary habits, dispersal tendencies, overwintering traits, and their interaction with landscape structure and configuration make predictions exceedingly difficult ([Martin](#page-12-0) et al., 2019). A meta-analysis by [Karp](#page-12-0) et al. [\(2018\)](#page-12-0) also highlights that although landscape composition explains a significant part of the variation of pest and enemy abundances, no consistent trend can be found: surrounding non-crop habitats do not necessarily improve pest management, highlighting the shortcomings in our understanding of the recovery by migration systems. The SSDderived endpoints – HD5/HC5 and PAF – do not take this

<span id="page-11-0"></span>

**Fig. 7.** Combined Species Sensitivity Distribution curve for ecosystem service providers collected in five European countries representing different pedoclimatic zones towards lambda-cyhalothrin, based on LD<sub>50</sub>s derived from laboratory-coated glass vial bioassays. The dotted line indicates the estimated HD5 value.

phenomenon into account, so from this point of view they can be considered 'worst-case scenarios'.

# *4.3. Advantages and disadvantages of estimating SSD based on LD50 and NOEC*

Although SSD can be estimated based on any test endpoint, the two most commonly used measures are acute  $LC_{50}/LD_{50}$ , as in our study, and chronic no-observed-effect concentration (NOEC), resulting in acute SSDs and chronic SSDs, respectively. Acute  $LC_{50}$  means massive mortality in a population within a short time after exposure to a pesticide (here 24 h), which can be considered an unacceptable effect. Thus, acute SSD describes the distribution of severe effects in populations and if estimated PAF reaches high values, as in our study (LD<sub>50</sub> exceeded or reached for 96.6 % – 99.9 % of species), it indicates a serious effect at the community level soon after the pesticide application. This prompts the question of the relevance of acute SSD curves and the derived HD5 and PAF values for assessing ecological risk under natural conditions. Moreover, in most cases, NTAs living in a natural environment almost certainly would be exposed less than in the coated-glass assay. Carabids, for example, spend a lot of time underground, being hidden from direct spray for at least part of the day. In turn, species living on the surface of soil or plants, like spiders or ladybirds, are exposed more than carabids but are still partially protected from direct spray by leaves above. Furthermore, in this study, for reasons explained earlier, the 'affected' individuals were classified as dead, so this also contributed to the 'worst case scenario'. Thus, the acute SSD based on a coated-glass assay would most likely overestimate the negative effect of λ-cyhalothrin on a community. Nevertheless, even if the relevance of such derived acute SSDs can be questioned due to several reasons (the specific laboratory conditions, high acute effect, i.e. 50 % mortality used to estimate the SSD, short exposure not considering prolonged effects, etc.), Van der [Brink](#page-13-0) et al. [\(2002\)](#page-13-0) showed that the SSD curves for chlorpyriphos based on

 $LC_{50}/EC_{50}$  values of aquatic arthropods collected from laboratory tests and a semi-field test were similar, with almost identical slopes and 50th percentiles. Comparing SSD profiles based on laboratory-derived  $LC_{50}$  or NOEC values with effects observed in field communities, they concluded that at the 10th percentile of the acute and chronic SSDs, no effects were observed in the field after short-term and long-term exposure, respectively. Such studies indicate that SSDs can indeed be used to protect entire communities from the adverse effects of insecticides.

The NOEC-based chronic SSDs can, in theory, reveal more about the effects of prolonged exposures and the delayed effects of short-term exposures which can be missed by short-term acute tests usually used to estimate  $LC_{50}s/LD_{50}s$ . Unfortunately, NOEC has poor statistical properties and depends on the experimental design to a much greater extent than  $LC_{50}/LD_{50}$  (cf. [Laskowski,](#page-12-0) 1995). This problem in relation to the SSD approach is discussed more in-depth by Suter II et al. [\(2002\)](#page-13-0), who listed weaknesses of using NOECs to estimate SSDs, including the fact that NOECs have no clear biological or societal meaning and do not represent any particular level of effect, hence the distributions of NOECs are distributions of no specific effects. Instead of the NOEC-based SSD, the chronic SSD can be extrapolated from the  $LD_{50}S/LC_{50}S$  using a safety factor of 10 (Van der [Brink](#page-13-0) et al., 2002) or by applying an assessment factor of 5 when the PNEC is estimated from HC5 (HD5) based on the acute SSD (ECHA – European [Chemicals](#page-12-0) Agency, 2008). This shift towards alternative methodologies offers a more robust framework for assessing the potential impacts of insecticides, ensuring greater reliability in ecological risk assessments.

## **5. Conclusion**

Our results show that the widely-used pyrethroid insecticide, λ-cyhalothrin, poses a serious threat to beneficial arthropod communities as the Hazardous Dose for 5 % of the species tested is 2–3 orders of magnitude lower than the recommended field dose for this insecticide.

<span id="page-12-0"></span>At the recommended field dose, the Predicted Affected Fraction of ESP communities exceeded 96 % in all five European countries studied, revealing an extremely high risk to ESP species of using λ-cyhalothrin. It has to be noted, however, that the coated glass vial test method represents a worst-case scenario, with test individuals constantly exposed to the insecticide-covered glass surface and not considering sink-source dynamics of populations inhabiting natural environments which act as refugia. On the other hand, if the assessment factor of 5 is used, as recommended by ECHA – European Chemicals Agency (2008), the estimated HD5 values would be five times lower than those reported here. However, given that only *<*10 % of species (5 out of 54 tested) had LD50 *>* 50 % RFD, it can be assumed that about 90 % of species would be eradicated or heavily affected in crops treated with λ-cyhalothrin at the recommended application doses. Our results indicate an urgent need to re-evaluate current risk assessment procedures by incorporating multispecies approaches, such as those based on SSDs.

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# **CRediT authorship contribution statement**

**José M. Blanco-Moreno:** Writing – review & editing, Investigation, Formal analysis. **Berta Caballero-López:** Writing – review & editing, Investigation. **Samantha M. Cook:** Writing – review & editing, Methodology, Investigation, Conceptualization. **Stephen P. Foster:** Writing – review & editing, Methodology, Investigation. **Danuta Frydryszak:** Investigation, Formal analysis. **Ryszard Laskowski:** Writing – review & editing, Writing – original draft, Project administration, Methodology, Formal analysis, Data curation, Conceptualization. **Patricia Ortega-Ramos:** Investigation. **Mykola Rasko:** Investigation. **Pauline Reichardt:** Investigation. **Jose**´ **Paulo Sousa:** Methodology, Conceptualization. **Grzegorz Sowa:** Writing – review & editing, Writing – original draft, Investigation. Renata Sliwinska-Grochot: Project administration, Investigation. **Julian Winkler:** Investigation, Formal analysis.

#### **Declaration of competing interest**

Ryszard Laskowski reports financial support was provided by European Union. If there are other authors, they declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

### **Data availability**

The  $LD_{50}$ s used to estimate the SSDs are reported in Tables 1–5. Original survival data used to calculate  $LD_{50}$ s are available on Zenodo (DOI: <https://doi.org/10.5281/zenodo.13760059>).

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